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

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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?
Bernhard Url, 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 for plant health.

Keynote Monday
Food Safety in a Complex Changing World
Bernhard Url, EFSA, Italy
Food Safety in a Complex Changing World: 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. The lecture will present current challenges to the assessment of risks to both the environment and food safety and can drive their (re)emergence. 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.

Keynote Tuesday
Innovative Research Issues in Environmental Mutagenesis
Eugenio 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.
1 Modelling and monitoring of pesticides fate and exposure in a regulatory context (I)

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 influences; Use this information to develop proposals for standardised 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. Benefit from 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 been 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
C. Basset SE; Z. Gauss, Bayer AG Crop Science Division; M. Lamboeuf, Bayer CropScience AG / R&D; M. Reitz, H. Reseler, Syngenta AG; P. Sur, Bayer AG - Crop Science Division / Environmental Safety; P. Sweeney, Syngenta; P. Volz, BASS SE; S. Webb, Syngenta Ltd; B. Zillgens, Dupont GmbH

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 with different levels of experience 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 radiolabelled 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: TSCF as recommended from the experiment of Briggs et al. (1982) and the substance specific TSCF value from “uptake experiments with appropriate and agreed set-up to be developed” (EFSA, 2013; EU Com, 2014). The presented study design was explicitly developed to address the need identified by EFSA and was based on suggestions from the EUregPuf workshop (York, 2013) 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 optimization 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 Regulator Groundwater Monitoring of Crop Protection Products and their Metabolites in Europe
R.L. Jonge, Bayer Crop Science Division / Environmental Safety; A. Gimsing, The Danish Environmental Protection Agency / Pesticides and Genetic Technology; J. Agert, Bayer CropScience AG / Environmental Safety; N. Baran, BRGM; A. Boetsch, BASF SE; F. Ferrari, Syngenta Crop Protection; M. Gibson, French National Institute of Health and Safety Executive; L. Hammond, Health and Safety Executive / Environmental Fate; F. Hegler, Dr. Knoell Consult; W. Koenig, UBA Umweltbundesamt; J. Keugher, Swedish University of Agricultural Science / Centre for Chemical Pesticides; T. Van der Linden, RIVM / ENVIRONMENTAL QUALITY; D. Liss, SGS Institut Fresenius GmbH / Agro, L. Losse, Syngenta; A. Massey, Health and Safety Executive; B. Miles, BASF EP / Crop Protection, Environmental Fate Modelling; L. Monozorske Sce; A. Newcombe, ARCADIS US Inc; L. Padovani, European Food Safety Authority (EFSA); A. Poot, Cgbl, G.B. Reeves, Dow AgroSciences Ltd; S. Reichenberger, DR. KNOELL CONSULT GmbH; A.E. Rosenbom, Geological Survey of Denmark and Greenland / Geochemical; H. Staudenmaier, BASF SE / Crop Protection, Environmental Fate Modelling; R. Sur, Bayer AG - Crop Science Division / Environmental Safety; A. Schwen, AGES; M. Stemmer, Austrian Agency for Health and Food Safety / Institute for Plant Protection Products; W. Tütting, German Federal Office of Consumer Protection and Food Safety; U. Ulrich, University of Kiel

Groundwater monitoring is considered a higher tier assessment in the regulatory groundwater assessment of crop protection products in Europe, but little guidance has been provided to date on study designs. 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 proposed 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 first-order (SFO) degradation of the pesticide concentration in total soil. Thus, 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 studies showed that 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
of the simulation period.

5 Bespoke monitoring to support Tier 4 FOCUS groundwater assessment

S. L. McManns, Syngenta; S. Payvandi, Syngenta Ltd; P. Sweeney, Syngenta; L. Fish, Syngenta Crop Protection, LLC / Environmental Safety; R.J. Andrews, D. Schofield, Ramboll Environ; J. White, ARCADIS UK; N. Jones, Syngenta Ltd; G. Langridge, CEM Analytical Services Limited; T. Oteyza, Syngenta Crop Protection AG; M. Greener, Syngenta Ltd

Data generated from this bespoke groundwater monitoring programme will offer a solution to address the non-relevance case of pineoxadine 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 pineoxadine use, groundwater less than 10m bgl, no confining layers, and no influential 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 pineoxadine 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 ecotoxicological 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 sale 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 accurate 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 EXPOS/Teva (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 response to pesticide risk was implemented by applying the field based and validated exposure – response relationship SPEARinput. 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 bioaccumulation of water concentration C and fish tissue concentration C is k_water BCFC. The characteristic time for uptake and loss is t = K_d / k_water. Slower uptake and loss will occur if the partition ratio K_d is large, and the fish must contact K_d-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 depuration phase. Mackay et al. [1] modeled bioconcentration and toxicity of superhydrophobic chemicals D4, D5, and D6 using a bioaccumulation model for fish. Due to the very high hydrophobicity (log K_d=10 for D5) and very low water solubility Cs, it must be very low, which results in a very long equilibrium time. Uptake time to equilibrium for D5 was estimated to be about 2000 days, to get C=2 mol/L 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. Doucette et al. [3] reviewed foliage/air partitioning data in which the onset of a hydrophobic delay (HD) is apparent when log K_d/BCF for [4] developed bioaccumulation model 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 K_d/BFCF=10^2. In our presentation 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 Tech 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?

M. Castro, Stockholm University / ACES; M. Breitholtz, B. Yuan, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); I. Athanassiadis, Stockholm University; L. Asplund, A. Sobek, Stockholm University / ACES 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-production volumes 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 complexity. CPs are hydrophobic contaminants, which complicates their aquatic toxicity testing. In this work, we validate the use of passive dosing for the studying hydrophobicity 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 (K_d=water). Bioaccumulation of D. magna was observed after 48 hours under different exposure concentrations. APCl-QTOF-MS was used for CP quantification. Both silicone-water and organic carbon-water partition coefficients overlap between different categories of CP technical mixtures. CP-52, labelled as a MCCP, had a similar silicone-water partitioning coefficient as a restricted SCCP – Huelz 70C. We demonstrate that increasing average chlorine content of each CP mixture significantly increases the log K_d/water and Log K_d/BCF. 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. K_d/water 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

K. W. Woodburn, The Dow Chemical Company / HES; R.M. Seston, Hyla Environmental Consulting, LLC / Toxicology, Environmental Research & Consulting; J. Kim, D.E. Powell, The Dow Chemical Company / Toxicology, Environmental Research & Consulting The trophic transfer of cyclic methylsiloxane (cVMS) materials in aquatic ecosystems is an important criterion for assessing bioaccumulation and ecological risk. These compounds are used primarily as pressure lubricants, flame retardants, and specific 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, 2,3,4,5,5'-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 (d15N) and carbon (d13C) 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 eVMS 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 values observed in the highest trophic level. The median TFMs measured for the three eVMS materials were all99% of the uncertainty for eVMS. TFM 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 TFM of 2.2 in the evaluated food web, indicating biomagnification. TFMfs for the eVMS 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 eVMS 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 Polyhalogenated Carbazoles in Aquatic Systems from the United States and China

D. Chen, Jinan University / Cooperative Wildlife Research Laboratory and Department of Zoology; Y. Wu, Southern Illinois University Carbondale / Cooperative Wildlife Research Laboratory and Department of Zoology; R. Sutton, S. T. K.-au, Erhual Institute of K. Xu, Louisiana State University / Department of Oceanography and Coastal Sciences

The present study reports the discovery of a suite of polyhalogenated carbazoles (PHCZs) 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 Dian Shan (China), and their bioaccumulation in the San Francisco Bay estuarine system. 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-tri-bromo-, 1,3,6,8-tetrabromo-, 1-bromo-3,6-dichloro-, and 1,8-dibromo-3,6-dichloro-carbazole were screened. Halogenated carbazoles were detected in 98.7% of the sediment samples, with concentrations ranging from below method limits of quantification to 51.5 µg/g dry weight. In most of these sediment samples, PHCZ concentrations exceeded those of polybrominated flame retardants (PBDEs). The latter group of chemicals has been determined 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. In most of these sediment samples, PHCZ concentrations exceeded those of polybrominated flame retardants (PBDEs).

11 Bioconcentration factors of contaminants 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 oil compounds is crucial for establishing a standard flow-through uptake/depletion 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 chemicals via a standard flow-through exposure chamber. The concentration of (k1) in the fish soma (without GIT) for the key pine oil constituents are 0.134 d–1 (β-Caryophyllene, BCP) – 1.41 d–1 (BAc) and they were 0.0079 d–1 (HCB) – 0.517 d–1 (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 the two test compounds. Benchmarking to HCB reduced the standard error of measured k1,SBM from the soma for most of the chemicals, with k1,SBM ranging from 0.001 d–1 (PCB52) to 2.98 d–1 (BAc). The apparent BCF (BCF,s) values in soma for the key components in pine oil and the reference chemicals were in the range of 98.2 L kg–1 (BAc) – 1030 L kg–1 (BCP) - and 267 L kg–1 (DiCB) – 1730 L kg–1 (HCB), respectively; while for the benchmarked BCF (BCF,s) in soma, they are 46.3 L kg–1 (BAc) - 2570 L kg–1 (BCP), and 208 L kg–1 (DiCB) - 197000 L kg–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 BIOACUMULATION PROCESS OF LIPOLIPIC 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 melan and mandibular fat are inert lipid bodies in odontocotes 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, melan and mandibular fat, and 2) Physiologically based toxicokinetic (PBTK) modelling of PCB 153 and PBD 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 melan with increasing age. Nevertheless, the model showed the highest levels of PCB 153 in mandibular fat, followed by melan 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. PBD 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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Environmental Life Cycle Assessment (LCA) relies on data, models and knowledge from almost all environmental scientific disciplines, including related uncertainties. In addition, LCA involves making methodological choices. Over the past five years, we have published several approaches to deal with data and methodological choice uncertainties. One often heard critique is that these proposals do not account yet for ‘correlations’. We distinguish between two meanings of the term ‘correlations: correlated sampling: when applying Monte Carlo sampling for propagating uncertainty data for a comparative LCA study, the sampling can be either dependent (correlated) or independent (uncorrelated).

Independent sampling implies that data of shared processes between the product alternatives compared, are sampled in different MC procedures resulting in different data sets for this shared process for both product alternatives. Dependent sampling implies that process data for the product alternatives compared are sampled based upon one and the same random drawing of parameter values resulting in identical data sets for this shared process; correlated data points: a transport process input of diesel is, for example, related to an process output of CO2 (emission); if the process consumes more diesel for the same amount of transport, the CO2 emission will also increase. The first interpretation of ‘correlation’ has been addressed in earlier work by Henriks et al. and recently again by Mendoza et al. The second interpretation of ‘correlation’ (between data points) has recently been addressed by Groen et al. We present an overall framework integrating the two approaches combining data from LCA studies. In addition, we show the possibilities and limitations of also including data correlations into LCA uncertainty assessments. Practical application of this

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framework in the daily practice of LCA practitioners needs further work, including implementation in LCA software programs and particularly data.

14 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 scenario of product 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 ecotoxicology 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.

15 Combined uncertainty and scenario analysis within Life Cycle Assessment of water management systems
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Life Cycle Assessment (LCA) is an increasingly used decision support in the waste management field. LCAs are subject to uncertainty regarding both the input values for the LCA model (or 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 the results strongly depend on the fact that such analyses are carried out separately, dividing the interpretation of the results in two separate channels and potentially limiting the value of LCA as a decision support tool. This study presents a novel method that aims at combining uncertainty and scenario analysis, illustrated on a case study on three hypothetical waste management options for the treatment of residual household waste in the municipality of Copenhagen in 2025. The waste management solutions were provided with uncertainty for the model input values (parametrical uncertainty) and were assessed within four different hypothetical background conditions (scenario analysis). Within each impact category, the results of the parametrical uncertainty analysis were used to identify the most robust waste management option, i.e. the waste management option obtaining the highest average probability measure of providing the best environmental performance across the considered background scenarios. The method allowed obtaining various levels of analyses for the interpretation of the LCA results: parametrical uncertainty analysis for each of the assessed waste management options, with identification of the parameters mostly contributing to the uncertainty around the results, within each of the assessed scenarios for the background conditions. Parametrical and scenographic analyses were necessary for carrying out discernibility analyses across background conditions, allowing obtaining useful insights on the changes in sensitive parameters induced by the change in background conditions. The discernibility analysis results allowed obtaining a clear quantification of the probability measure of each waste management option to provide a better environmental performance than another, for each of the assessed impact categories and investigated background conditions, and in a manner simply conveyable to the users and final receivers of the LCA.

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 the 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 Inventory (LCI) regionalization deals with investigating 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 sectorial recommendations regarding the impact categories (IC) and LCA phases (LCI or LCIA) that should be regionalized in priority. These 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, 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.

17 Reduce the uncertainty of LCA results by prioritizing the regionalization effort: a sectorial meta-analysis
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Uncertainty in Life Cycle Assessment (LCA) can be limited the results interpretation. Regionalization is one of the ways to reduce the uncertainty due to spatial variability. Life Cycle Inventory (LCI) regionalization deals with investigating 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 sectorial recommendations regarding the impact categories (IC) and LCA phases (LCI or LCIA) that should be regionalized in priority. These 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, 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.

18 Poster spotlight: M0387, M0388, M0389
Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (I)

19 Unraveling 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, 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 Holtemme 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-resolution quadrupole hybrid - Orbitrap MS (QExactivePlus, 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 ‘kml’. 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 mixtures (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 sea 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 both WWTp influents and effluents, as well as in the receiving waters (rivers and estuaries) and even in the open ocean. In order to do this, we carried out several monitoring campaigns in the Gulf of Cadiz (Atlantic Ocean, SW Spain), sampling freshwater from one of the biggest local WWTp in the area (Jerez de la Frontera, 250 000 inhabitants), adjacent surface river and coastal waters, and oceanic waters at different depths (down to 400 m) taken up to 50 km away from the coastline. Solid phase extraction followed by liquid-chromatography high resolution mass spectrometry were used in combination with statistical tools (e.g., principal component and cluster analyses), specific vendor and open-access software, and other curiosity driven approaches to tentatively identify more than 1200 compounds that showed features persistent enough to be also detected in oceanic waters. These compounds included different classes of surfactants (e.g., linear alkylbenzene sulfonates) and their byproducts (e.g., DATS) and metabolites (e.g., NPEC), polymers (PEG, PPG and many ethoxylated derivatives), pharmaceuticals (e.g., valsartan, diclofenac, carbamazepine, etc.), personal care products (UV stabilizers) and food additives (e.g., sucralose), some of them (e.g. sulfur) identified in the environment for the first time. The list of compounds reflected here not only shows many of the substances that can potentially escape from wastewater treatment but also constitutes a first step towards a more detailed characterization of the chemical exposome in the marine environment.

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, cyclamone and sucralose. Except caffeine and DEET, concentrations of other PPCPs and ASWs in river water were found to be higher in densely populated areas. Concentrations of PPCPs and ASWs in the groundwater 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 concentrations were lower in those than in 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 management 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

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The presence of polar contaminants like angiotensin II receptor antagonist pharmaceuticals (sartans) in the aquatic system is directly linked to human impact. Like other xenobiotics, they can be metabolised in the body with enzymes such as cytochrome P450 (CYP), UDP-glucuronosyltransferase (UGT), and glutathione-S-transferases (GST) which are present in the human liver at high abundance. Due to biological and/or abiotic processes that the contaminants undergo from the discharge site to the ground or surface water where they are detected, they can be transformed to transformation products (TPs). These TPs are usually detected and identified first at lab-scale in order to evaluate the degradability of a compound. This is typically followed by a targeted method development and it is not up until the compound’s fate have been examined and in some cases isolated that they are actually searched for in real aquatic samples to report their presence. Here, we propose an alternative approach, based on data-dependent fragment ion search, where real-world samples are initially screened for plausible TPs, metabolites or related compounds. The starting point here was a suspect screening of a list of all marketed sartans in wastewater effluent and surface water samples, which were extracted with a generic solid-phase extraction method using four cartridges with different chemistries. Out of the compounds detected, five of them had an identical core structure, and it was postulated that this sub-structure would fragment identically in all compounds. Following a series of experiments with different MS parameters modified, a list of hit compounds was obtained using fragment ion search. After all of the compounds investigated, available human metabolites and internal standards were purchased, a set of biodegradation experiments using activated sludge was performed in order to “source” the detected m/z and compare the possible TPs fragmentation to the one obtained in the bio-reactors. In parallel, a literature search for reported human metabolites was used to complement the identification of compounds detected in cases where no such compound was found to bio-form from the parent compound. Finally, a targeted method was developed for the quantification of these compounds in wastewater and surface water. This work was possible due to support from EU-FP7 programme (Solutions project), Merck (LC columns) and Biotage (SPE cartridges).

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

R. J. Schneider, H. Hofmann, BAM Federal Institute Materials Research and Testing / Department of Analytical Chemistry Reference Materials 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 antidepressant, 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 considerable reverse-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 reconstituted with a solution of Scan250 or TOP-ESI-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 anorganic 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 states 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-say 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 the use of the standard monitoring program for all EU DWD required 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 vivo as well as in vitro 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

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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 biodegradation 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 biodegradation factors (BCF) in fish. Where possible, fish BCFs were correlated to mammalian BCFs 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

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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. The current protection goal (SPG) options for amphibians and reptiles were developed to address the general protection goals for non-target organisms and biodiversity in Regulation EC 1107/2009 [2]. When proposing SPG options, the endangered status of a great proportion of amphibian and reptilian species was taken into account, as well as the importance of amphibians and reptiles as drivers of valuable ecosystem services in agricultural landscapes. The analysis of literature data reveals that amphibians and reptiles occur in agricultural landscapes, where they may be exposed to PPP in the in- and off-field areas, and that unacceptable, adverse effects on their populations may occur. It was concluded that the current risk assessment scheme does not fully cover all relevant life stages and thus might not protect the persistence of amphibian and reptilian populations in the long-term. The EFSA working group proposes a general risk assessment framework based on a tiered approach and adapted to assess local and landscape-level risks for amphibian and reptilian populations. Identified knowledge gaps regarding amphibians and reptiles concern on the one side the exposure assessment, with e.g. a lack in data about size and location of ponds inhabited by amphibians. On the other side, central information is missing for local and landscape assessment, e.g., the impact of oral and dermal exposure routes. Informative field studies are very difficult to perform with these organisms and risk mitigation options – if available - could usefully be included in risk assessment as an alternative to full-scale field studies. Practical mitigation options might need to be developed and adapted for local environments to be most effective. [1] EFSA PPR. 2017 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] EF. 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 309/1

27 Ecotoxicological assessment of Caretta caretta (Linnaeus, 1758) in the Mediterranean Sea using an integrated non-invasive protocol

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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 end point potential contamination 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 by In Rescue (Comet and ENA assays) and free-ranging among 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 neurotic (esterases inhibition) and estrogenic (vitellogenin) effects, oxidative stress (lipid peroxidation) and Genotoxicity (Comet and ENA assays) and liver damage (Gamma Glutamyl Transerase). 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 cytochrome nuclear abnormalities which may indicate a longer 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 clans or hunting seals - consequences to walrus health
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The walrus (*Odobenus rosmarus*) can be considered a flagship species of the Arctic 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 environmental contaminants with lower contaminant exposure, there are 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 blubber were positively related to δ13C in blubber lipophilic compounds, but not to PFASs. Antibodies against Brucella spp. and Toxoplasma gondii were detected in 26 % and 15 % of the walrus plasma samples, respectively. Presence of Brucella spp. and Toxoplasma gondii were not related to contaminant exposure or stable isotope values. Among the 5 thyroid hormone concentrations analyzed in plasma, concentrations of TT4 were negatively related to concentrations of δ15N. Their eggs are a useful tool to monitor the levels of contaminants associated with marine mammals during development. The milk from pregnant females was analyzed for the transcript levels of 21 genes in blood cells and 7 genes in blubber related to endocrine and immune functions by real-time quantitative PCR. The preliminary results indicate few relationships between transcript levels of genes involved in endocrine functions and contaminant exposure.

29 Trichloroan-induced embryotoxicity in the yellow-legged gull
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Trichloroan (TCS) is a chemical compound extensively used as synthetic and antimicrobial agent in a wide range of personal care products. Because of its hydrophobic nature and its discharge in the sewer system, TCS accumulates in settled sewage sludge and surface water, contaminating aquatic and terrestrial ecosystems. However, there is no information on the potential toxicity of TCS towards wildlife species is very scarce. Seabirds are highly exposed to environmental contaminants because of their ecological habits, high trophic position in the food web and relative long life-span. Their eggs are a useful tool to monitor the levels of environmental pollutants and their potential adverse effects because these chemicals can be maternally-transferred to offspring. However, such information on TCS is lacking. The aim of this study was to explore, through in ovo injection, the potential embryotoxicity of TCS in the yellow-legged gull (*Larus michahellis*). In a within-clutch experimental design, 150 ng/g egg weight of TCS were injected into the egg yolk and the effects on embryo morphology, oxidative stress and genetic damage in embryo liver were investigated. Specifically, we assessed effects on embryo body mass, tarsus length and head size, as well as liver and brain mass. The amount of oxidant species (i.e. ROS), enzymatic activities (SOD, CAT, GST) and the levels of lipid peroxidation (LPO) were measured as biomarkers of oxidative stress, while levels of DNA fragmentation were measured as genetic damage endpoint. To check for the reliability of the injection method, we quantified TCS concentration in the yolk of unincubated eggs, while to assess its transfer to the embryo, we measured TCS in residual yolk and in the liver and brain. TCS concentrations in yolk from unincubated eggs were similar to the nominal ones (195.9±35.3 ng/g wet weight), while lower concentrations were found in residual yolk soon before hatching (2.9±1.1 ng/g wet weight). TCS was also detected in the liver (2.3±1.1 ng/g wet weight) and limitedly in the brain (0.2±0.1 ng/g wet weight). TCS treatment did not significantly affect embryo morphological traits. However, TCS significantly increased ROS levels and promoted GST activity, leading to a marginally non-significant increase of both oxidative and genetic damage. Thus, these findings demonstrated, for the first time in a wild bird species, that TCS may affect offspring phenotype and may represent a potential threat for coastal ecosystems.

30 Egg overspray with herbicides and fungicides reduces chick survival in red-legged partridges
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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. Firstly, we exposed African tebuconazole on embryonic development and post-hatching survival of a common farmland bird, the red-legged partridge (*Alectoris rufa*). The first experiment simulated egg overspray with pesticides and the second one the incubation of eggs upon a soil that had been previously sprayed. For both experiments we used an application rate, corresponding to a 30% of the labelled application rate of each product (i.e. assuming a 70% interception by crop), and a control consisting of a water application in stead of pesticide formulations. Eggs were incubated at 37°C and 45% humidity until hatching (23-26 days). Sixteen eggs per treatment were removed from the experiment at different incubation times to analyse pesticide uptake (ongoing analyses, results will be presented at the meeting), and a minimum of N=20 per treatment was monitored for embryonic development and post-hatching survival. Chicks were weighted and measured (tarsus length), and body condition calculated, at hatching and at days 8, 16, 24 and 32 post-hatching. Egg overspray with pesticides significantly increased chick mortality (Wald’s X² = 29.909, 14 d.f., p = 0.008). Although pesticides did not affect survivorship at hatching time, in ovo exposure to both 2,4-D and tebuconazole caused increased mortality of chicks afterwards, resulting in a reduction of productivity 32 days after hatching of 30.6 and 25.9%, respectively. Incubation of eggs in pesticide applied soils did not significantly affect chick survival (Wald’s X² = 15.603, 14 d.f., p = 0.338), and nestling growth was not affected in either experiment by embryonic exposure to pesticides. These results suggest that reduction of embryonic and chick survival because of egg overspray with pesticides can be a potential way by which these products can affect reproduction fitness. Likewise, potentials of 2,4-D and tebuconazole exposure between the exposure 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
L. 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 is within the EU, is the responsibility of the European Medicines Agency (EMA); however, developed best practices are shared by all member states. The EMA also aids in coordinating other European procedures with several member states involved. EMA frequently authorises and takes advice from specialist working parties aligned to the specific provision or modification of regulatory guidelines or procedures within the committee for veterinary medicine products (CVMP). Here we such group focuses on the improvement of the guidance to industry and other stakeholders on environmental risk assessment alongside the provision of reflection documents that aid understanding or address specific areas for clarification in regulatory procedures. The overview will highlight some of these recent developments, in improved regulatory advice for current or future procedures. It will summarily cover issues around assessing and limiting veterinary medicines in groundwater, the use of higher tier testing of dung fauna, the use of higher tier plant testing, improvements in PBT assessments, future plans around developing improved guidance for the assessment of risks from aquaculture
medicines and the role of veterinary medicines in driving AMR in the environment and its potential consequences and mitigation. The session will feature successes, current issues and developments in improving the guidance on the assessment of veterinary medicines in the environment; and will reflect on the future challenges and difficulties faced by the regulators and industry alike. This paper will acknowledge the significant continuing contribution made by the Environmental Risk Assessment Working Party (ERA-WP) of the CVMP.

32 Risk of veterinary medicines to plants: Reflections for an updated approach. R. Carapeto Garcia, Spanish Medicines Agency / Veterinary medicines; A. Haro Castuerra, Spanish Medicines Agency / Veterinary Medicines Department; G. Cortís Ruiz, C. Rubío 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 taxon-related data, and use 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 Operational Protection Goals in beef animals: "Protection of Human Interest" and "Protection of Environmental Interests".

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, Introvet Intc. dba 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), trendione (TBO), 17β-E2, 17α-trenbolone (17α-TB), and estrone (E1). The similarity in chemical structures and metabolism of the environmental fate properties among 17β-TB, 17α-TB, and 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, 17β-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 heifers in the US.

34 How can mesocosm studies increase realism in risk assessment of biocides and veterinary medicines? L. Dörr, ERM / Product Stewardship; U. Hommen, Fraunhofer IPE; P. Ebke, McKesson DCM GmbH Institut für Gewässerschutz; R. Dürring, Justus Liebig University Giessen / Research Centre for BioSystems, Land Use, and Nutrition (IFZ), Institute of Soil Science and Soil Conservation

Mesocosm studies can be used to assess the environmental impact of potential stressors based on model-ecosystems under realistic environmental conditions. They are an important link from laboratory to field. Mesocosms provide the assessment of a broad range of different species of different ecological groups forming food webs with complex interactions. Therefore mesocosm studies can support a better understanding of the environmental impact of stressors on population level as well as on ecosystem level (e.g. direct and indirect effects on community structure and ecosystem functions as primary production). In addition, mesocosm studies provide data on the fate of test substances under realistic outdoor conditions, which can be used to test the prediction based on laboratory studies. While for the risk assessment of Plant Protection Products (PPP) mesocosm studies are an established higher tier approach and are considered as the surrogate reference tier, the use of mesocosm studies for risk assessment of biocides, veterinary medicines and chemicals under REACH is rare, although mesocosm studies are recommended from the Guidance on chemical safety assessment - Chapter R.10: Characterisation of dose [concentration]-response for environment and in the Guidance on the Biocidal Products Regulation - Volume IV Environment - Assessment and Evaluation both provided by ECHA. One reason for this might be, that mesocosm studies have the reputation to be very complex and difficult to evaluate by regulatory assessors. This presentation intends to take some fears of contact with mesocosms. It will explain the most important aspects to validate the quality of a mesocosm study and the relevance of the results. Further, it will give some insights to the use of (aquatic) mesocosm studies in the context of PPP risk assessment and will provide important aspects for planning a mesocosm study for biocides, veterinary medicines and chemicals in the context of REACH.

35 Estimation of insecticides in mine farms R.G. Ovesen, Danish Environmental Protection Age; H. Bækgaard, Kopenhagen 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 consider biocides used in mines. A scenario has therefore been developed, where emission of a.i. from mine farms is calculated based on either amount applied or measured concentration in straw. Default values have been estimated from regulation and general practice in mine production in the Nordic countries, where Denmark has the highest production of mine in Europe [3]. Each breeding animal is kept individually in one cage to be treated at the start of the season. Mother and cubs 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 to public sewer. Emission is therefore only expected to be to agricultural land. Emission of manure/straw may be from 10 to 50 BF per hectare (ha) per year based on regulation in the Nordic countries. Emission according to application pattern: Y = Q_max x f_max x (N_{NPS} before sep + 3.275 x N_{NPS} after sep) x B x 10^3 x 40% Where Q_max = emission of a.i. in kg/ha x year, Q_max is amount of product/nest box in g, f_max is concentration of a.i. in the product in%/kg, N_{NPS} before sep is number of treatments before separation of adults and cubs, N_{NPS} after sep is number of treatments after separation of adults and cubs and B is amount of straw/manure that may be applied to land in number of BF/ha (B = 50). Emission based on amount of straw/manure applied to the field: Y = Concentration of a.i. in straw/manure x 750 kg straw per BF per ha(eq. 2) Where amount of straw used per BF is 10-15 kg/year according to Kopenhagen Fur. The emission based on Nordic countries regulations and information from Kopenhagen 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].

36 Biocidal active substances in municipal wastewater - what product groups are the sources? S. Wieck, Leuphana University of Lüneburg / Institute for Sustainable and Environmental Chemistry; O. Olsson, Leuphana University of Luebenburg / Institute for Sustainable and Environmental Chemistry; K. Kümmerer, Leuphana University of Lüneburg / Institute of Sustainable and Environmental Chemistry

The emission sources of biocidal active substances in households have been under discussion since these substances have been detected frequently in municipal wastewater. The emission from the use of biocides, e.g. facades, has already been discussed in detail. However, until now the specific sources from the interior of these households remained unknown. To investigate the products responsible for these emission to wastewater, we analysed the wastewater of one neighbourhood for a set of biocidal active substances and compared these results with household product inventories. Time-proportional sampling of daily samples was conducted during one year for one selected residence. The 14 substances analysed with an LC-MS/MS method were BIT, C12-benzalkonium chloride, carbendazim, CMIT, DCOIT, DEET, diuron, icaridin, OT, piperonyl butoxide (PBO), triclosan, tetrabonazole, terbutryn and tetramethrin. In comparison with data available from household product inventories of this neighbourhood, we investigated the product groups possibly being responsible for the emissions to the wastewater. Except for C12-benzalkonium chloride and its potential conjugate, all substances have been detected in at least 10 % of the samples. Highest concentrations were measured for C12-benzalkonium chloride with an average concentration of 6.6 μg/L. Besides C12-benzalkonium chloride, BIT, DEET and icaridin were measured in all samples. The results show
that washing and cleaning agents are important sources for preservatives such as BIT and OFF, while triclosan was apparently mainly emitted through personal care products. The mosquito repelling substances DEET and icaridin 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 carboxin were not measured. 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 when it comes to the reduction of biocidal active substances in wastewater.

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

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

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Manufactured nanomaterials (MNMs) and especially Ag- and TiO2-NPs are processed 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 treated wastewater, the body length of adult 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 towards 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 more realistic assessment of Ag-NPs and TiO2-NPs for the aquatic environment. The experiment with TiO2-NPs are in progress.

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

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Bioaccumulation is one of the key triggers of concern for environmental risk assessment that has had little consideration for engineered nanomaterials (ENMs). Given that ENMs undergo surface chemical reactions, agglomeration and sediment, the likely exposure route to higher trophic organisms (e.g., fish) is through the diet. However, the addressability of an ex vivo gut sacs technique to reduce the bioaccumulation potential of pristine (Ag NPs) and environmentally aged (Ag:S NPs) materials. Additionally, we assess whether the results of the gut sac experiment can predict in vivo chronic dietary exposure. The gut sacs were prepared by removing the entire gastrointestinal tract and separating it into the oesophagus, stomach, anterior, mid and hind intestine compartments. CompARTments were exposed by filling the hamen with one of four solutions: physiological gut saline or saline spiked with 1 mg/L Ag as AgNO3. Ag NP or Ag3S NPs. Following a 4 h exposure, tissues were cut open and the mucosa was separated from the underlying musculature, through scraping via a microscope slide. For the in vivo chronic dietary exposure, fish (n = 150) were graded into tanks (n = 3 tank/treatment). Fish were fed either a control (no added Ag), 100 mg/kg as AgNO3, Ag NPs or Ag3S NPs. Fish were sampled each week (1, 2, 3 and 4; n = 2 fish/tank/time point). Following this, all tanks were placed on the control diet for another two weeks to measure Ag elimination. During sampling, the mid and hind intestine, liver, gillbladder, kidney, spleen, gastrointestinal walls and carcass were dissected. Tissues from both experiments were analyzed for total Ag using ICP-MS. The gut sac experiment demonstrates the uptake of Ag is associated with the mid and hind intestine. There was significantly less Ag in the muscularis of the mid and hind intestine after exposure to Ag NP and Ag3S NP compared to AgNO3, but no difference between ENM treatments. The in vivo experiment demonstrated significantly more Ag in the mid and hind intestine of Ag NP and Ag3NPs compared to AgNO3. Silver from all the exposures were able to pass the gut epithelium and cause total concentrations in the liver to rise, despite the form being unknown. In conclusion, the ex vivo gut sac method can be used to rapidly screen the bioavailability of Ag NPs and Ag3S 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

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Manufactured nanomaterials (MNMs) are widely used in various applications and commercial products, e.g., textiles, sunscreens, paints, cosmetics etc. Even though MNMs are mostly removed during wastewater treatment, the remaining and mostly transformed MNMs in the effluent are significant and may show an increased 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 303A. 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 WWTP 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 [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 or dilution water, the algae-Daphnia-fish test system showed an increased toxicity for AgNP concentration. 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 markers 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.

40 Uptake and elimination kinetics of pristine and aged silver nanoparticles in freshwater benthic organisms

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Nanomaterials (NMs) can undergo changes in their properties and behaviour during the WWT. The transformed MNMs in the effluents are significant and may show an increased 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 303A. 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 WWTP 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 [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 or dilution water, the algae-Daphnia-fish test system showed an increased toxicity for AgNP concentration. 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 markers 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.
(simulated) aged silver nanoparticles (Ag-NPs) in freshwater benthic organisms. In this study the pulmonate snail Physa acuta, the non-biting midge Chironomus riparius and the planarian Dugesia tigrina were used as test species. Pristine Ag-NPs of different sizes (3-8 nm, 50nm and 60nm), a 27nm silver sulphide (Ag2S-NPs) simulating aging, and their ionic counterpart as silver nitrate (AgNO3) were tested. Bioaccumulation tests consisted of an uptake phase, where organisms were exposed to suspended sediment and contaminated aqueous medium in a concentration of 10 µg Ag L-1, and an elimination phase where organisms were transferred to clean medium. Animals were sampled during the tests and total body Ag concentration was analysed by graphite furnace atomic absorption spectrometry. Kinetics of Ag-NPs and ionic Ag were described by one-compartment models. In this work, uptake and elimination kinetics of the different Ag forms are more similar between both Ag forms compared to indicators like water, metalloprotein key targets to evaluate the role of biomolecules in AgENM transformations. In turn, metalloprotein interactions with AgENMs are also central in mechanistic studies of cellular impacts of AgENMs, including toxicity, antimicrobial, and resistance mechanisms. Despite the shared preference of both silver and zinc for thiolate and amine coordination, the interactions of zinc finger domains with AgENMs is not well studied. Zinc fingers constitute a large class of metalloproteins, ubiquitous in eukaryotes, that use a combination of cysteine and histidine residues that bind Zn(II) as a structural element. Zinc finger domains within proteins typically serve as interactors and can bind DNA, RNA, proteins or small molecules to mediate cell-cell interactions. Snails showed faster uptake and elimination of Ag-S-NP from the body compared to other Ag forms. For planarians, results revealed very similar k1 values, with the highest k1 for animals exposed to ionic Ag and the lowest for Ag 60nm exposure. Analysis of Ag in the sediment will be soon available to elucidate the behaviour of Ag, especially at the water-sediment interface, where most benthic organisms are exposed.

41 Transformation of silver nanomaterials by ubiquitous zinc finger peptides


In biological systems, chemical and physical transformations of engineered silver nanoparticles (AgENMs) are mediated, in part, by proteins and other biomolecules. Given the high affinity of thiolate ligands for silver, metalloproteins are key targets to evaluate the role of biomolecules in AgENM transformations. In turn, metalloprotein interactions with AgENMs are also central in mechanistic studies of cellular impacts of AgENMs, including toxicity, antimicrobial, and resistance mechanisms. Despite the shared preference of both silver and zinc for thiolate and amine coordination, the interactions of zinc finger domains with AgENMs is not well studied. Zinc fingers constitute a large class of metalloproteins, ubiquitous in eukaryotes, that use a combination of cysteine and histidine residues that bind Zn(II) as a structural element. Zinc finger domains within proteins typically serve as interactors and can bind DNA, RNA, proteins or small molecules to mediate cell-cell interactions. Snails showed faster uptake and elimination of Ag-S-NP from peptides, we have evaluated the impact of Zn fingers on AgENMs aggregation and dissolution. Zinc finger peptides drive AgENM dissolution resulting in release of Ag(1)(aq) at orders of magnitude higher rates than other model proteins, including a few metalloproteins. The release of Ag(1)(aq) is central to mechanisms of cellular response and toxicity of AgENMs. Indeed, Cu(I) binds to both the apoproteins and the Co(II)-substituted peptides; the stoichiometry of Ag(I) binding is dependent on the peptide primary sequence. Additional studies using fluorescence spectroscopy to monitor Ag(I) binding to the Zn finger peptide indicate that the Ag(I) effectively completes with Zn(II) at the metal binding site, despite the high affinity of Zn(II) for the peptide. Circular dichroism spectroscopy used to assess changes in the peptide secondary structure demonstrate that the addition of either form of silver alters peptide structure and structural perturbations are again dependent upon the peptide sequence. These results show that Zn finger peptides can mediate AgENM transformations within eukaryotic cells. In turn, for the Zn finger peptides studied here, Ag(I) is the thermodynamically favored metal despite the known high Zn(II) affinity of zinc finger domains. This works suggests that Ag(I)-substituted zinc finger domains might be relevant in the context of both silver toxicity mechanisms and silver-responsive transcription factors.

42 Fate and effects of transformed Ag and TiO2 nanoparticles aged through a lab-scale wastewater treatment system

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The increase in production and use of Ag and TiO2 nanomaterials has led to their release in wastewater streams and subsequently in the environment. Nanoparticles (NPs) can undergo transformations in environmental media such as wastewaters leading to an alteration in behavior, bioavailability and toxicity that may differ from their pristine counterparts and make predictions challenging. In this context, the overall goal of the study was to elucidate (i) the behavior and transformation of Ag and TiO2 NPs in realistic matrices such as wastewater effluents and activated sludge and (ii) the subsequent effects of transformed particles in comparison to their pristine counterparts. In this study, a laboratory-scale wastewater treatment system 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, nanoComposite) and 100 µg TiO2 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 potential leaching. Some samples were also digested and analyzed to determine if these particles were collected weekly and analyzed by sequential fractional 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 particles 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 several microbial microcosm experiments, giving insights on 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 (OSR) are required as part of permitting for offshore operations. An OSR 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 socio-environmental 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 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) consideration of economies 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. Coello, Sponson 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, IPIECA-API-IOGP published guidelines on the implementation of NEBA, using a novel process known as Spill Impact Mitigation Assessment (SIMA). When applied appropriately to SIMA, the best approach to minimize environmental impacts is to employ multiple response options. This will require consensus between key stakeholders and the various decision-makers on the benefits and...
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 regulators, as well as 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; A. Delaunay, ONERA; A. Durand, ONERA / Optics and Associated Technics; A. Credoiz, R. Hédacq, TOTAL SA / Environment; P. Bordieres, ONERA / DEMR; G. Bertoni, INRA / Dynafor; D. Dubucq, TOTAL SA; A. Elger, University of Toulouse in Ecostlab.

The Deepwater Horizon Oil Spill (DWHOS) in 2010 is the largest and most studied spill in US history. It enabled us to learn how to deal with the new challenges of an oil spill in the deep sea, but also to improve pre-spill measures to minimize the adverse ecological impacts of 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 impacts of 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 impacts of 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 impacts of the next big marine oil spill, wherever it may happen.

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 Norway, UK; T.K. Collier, Delta Independent Science Board Norway, UK.

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 of dispersed oils, microbial oil degradation, oil-assisted 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 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.

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. Geith-Hansen, M.B. Larsen, Greenland Institute of Natural Resources; K. Gustavson, Aarhus University / Department of Bioscience - Arctic Environment What is the environmental effects of a beaching oil spill in the Arctic, how well will the shoreline potentially be able to self-clean and will combusting 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 coastal 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 west coast of Greenland. Among the results, it was demonstrated that the self-cleaning potential of the oil on rocky surface were investigated at three locations of low Arctic, middle Arctic and high Arctic climatic regimes along the west coast of Greenland. The study included experiments of natural removal of a crude oil and a heavy fuel oil from tiles mimicking rocky shore substratum and was run in the period from May-September 2017. The tiles were placed in different height levels of the tidal zone, and between natural removal and degradation rates in correlation to different water cover regimes and air exposure times were obtained. The oil remains 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 many 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 the degradation rates in the environment and the pattern changes with period of exposure. It would be more appropriate to use a “signature” analogy when comparing oils by this approach. The weathering processes change the chemical signature and old 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 it 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 abiotic 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 biphenol S alters microRNA expression in male zebrafish (Danio rerio) J. Lee, K. Ji, Yongin University

In response to the restriction of biphenol A (BPA), biphenol 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, health and behavior in animals. The aim of this study was to investigate the effect of BPS on the expression of microRNAs (miRNAs) in the brain of male zebrafish (Danio rerio). Zebrafish were exposed to BPS at concentrations of 1 μM, 10 μM, and 50 μM for 7 days. The effects of BPS on the expression of miRNAs were analyzed using TaqMan miRNA assay kits for 12 selected miRNAs. The results showed that BPS exposure significantly altered the expression of several miRNAs, particularly in the brain region. These findings suggest that BPS exposure may affect the expression of microRNAs in the brain of male zebrafish, which may have implications for understanding the mechanism of BPS-induced toxicity.
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 were upregulated and eight were downregulated as a result of 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

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A variety of antioxidant activities cause 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 development toxicities 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 (SeMet) at Se concentrations (1.1, 1.3 µg Se/g feed, dry mass) and environmentally relevant supraphysiological levels (3.4 – 28.8 µg/g) for 90 days. Swiming 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 collagenase, MMP2. These results suggest significant ecophysiologival effects that may impair the fitness of fishes exposed to elevated dietary Se in contaminated ecosystems. From a comparative biomedical viewpoint, the zebrafish model represents an appropriate system to investigate mechanisms 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 sink 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 in silico methods. Both approaches are complementary 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 (i) 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). Improvement identified to the technological requisites preceding to apical endpoints has become crucial for analyzing, accessing and determining chemical effects. Proteomics, therefore, show appreciable promise as a molecular screening tool for identifying specific alterations between exposures and controls, which is therefore imperative in discriminating endocrine disruptors from substances with a non-endocrine MoA. Such tool waiver the need for elongated higher-tier testing. Therefore, a high throughput alternative to classical endpoints that are specific to chemical-induced apical responses in zebrafish. The study focused on fadrozole, a known inhibitor of cytochrome P450 aromatase. Thus, an excellent model substance to evaluate and validate proteomic methods with the integration of organ-specific effects. Spawning adult zebrafish groups (5 males, 5 females) maintained at 25-26°C, 14 h light:10 h dark cycle, were exposed for 21 days to fadrozole (0, 0.1, 1, 10 µg/L) and analyzed for plasma vitellogenin content, egg numbers and organ histopathology. Livers and gonads were isolated for shotgun proteomics and qPCR to characterize substance induced specific molecular toxicity pathways. Proteins involved in steroid hormone secretion and estrogen stimulation such as vtg1, vtg3, vtg6 and lman1, were significantly deregulated. Several of the prominently affected pathways involved regulation of xenobiotic stimulus, lipid metabolism, metabolic processes, TCA metabolism and calcium signalling. Our study demonstrated that the downstream induced-estrogen receptor suppression by aromatase inhibition trigger the downregulation of estrogen synthesis, which was assumed to induce the observed decrease in egg numbers and oocyte atresia with membrane folding in the ovary. We anticipate that this improvement identified to the technological requisites preceding to apical endpoints has become crucial for analyzing, assessing and determining 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 a model organism 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 “accidently” 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 organophosphaes and potentially also other compounds and that 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 / Sinnhuber 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 hi-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. Zebrafish (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. Importantly, 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 components 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. Sabboni, 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 need 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 variability 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. Mosquera, 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 (ESFRI 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 scientific local Hubs, and the Coordination Heads will provide the unique link 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 Efate 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. Goettler, Bayer CropScience AG / Environmental Safety; P. Neumann, Buyer Ag; S. Bub, Tier3 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 identifies 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/off-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 SPGs.

60 Biogenic residues formation from pesticides - an overview K. Nowak, TU Berlin / Institute for Environmental Research (Biology V); A. Mittler, Helmholtz Centre for Environmental Research UFZ / Department of Environmental Biotechnology; M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology; M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology

Anthropogenic organic chemicals are deliberately (e.g. pesticides) released in the environment. Microorganisms can use C and N from a pesticide to synthesize their biomass and intact embryos. For homogenates only the oxon metabolites were able to activate organophosphates 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.

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nitrogen) source and the NER were mainly biogenic. The major formation of biogenic residues is supposed to be relevant for easily biodegradable contaminants under significant CO₂ formation. However, in the case of more recalcitrant pesticides like bentazone, the incorporation of C into microbial biomass, although reported to be very low, cannot be completely excluded.

61 Derivation of a wash-off factor for FOCUS modelling based on literature research
S. Sittig, DR. KNOELL CONSULT GmbH / E-Fate Modelling; C. Wollmann, Dr. Knoell Consult GmbH; G. Reiniken, Bayer AG, Research & Development, Crop Science / Environmental Safety

After foliar application, plant protection products (PPP) undergo several routes of dissipation of which one is precipitation-induced wash-off from the canopy. This process is accounted for in the European exposure assessment framework for PPP authorization and included in the corresponding numerical models, e.g. FOCUS PEARL, PELOM, PRZM and MACRO. A numerical wash-off factor for modelling is applied, quantifying the wash-off from plant surfaces by a given amount of precipitation. Consequently, this factor is relevant for the calculation of predicted environmental concentrations (PEC) for the compartments soil, groundwater, and surface water. In case a measured wash-off factor is not 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 proposed 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 ECP/AWG group has 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 acetonitril/water mixture of 80:20 (v/v). This standardized experimental procedure has been defined in order to derive a reliable numerical wash-off factor as input for FOCUS modelling. In this study, the performance of the literature 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.

62 Application of a dynamic aquatic food web model for FOCUS exposure assessment
L. Padilla, Stone Environmental, Inc.; A. Del Signore, D. Sprenger, L. Weltje, BASF SE / Crop Protection Ecotoxicology; M.F. Winchell, Stone Environmental, Inc.; J. Strassemeyer, Julius Kühn Institute / Institute for Strategies and Technology Assessment

In a 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 log Kow, 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 active substance in organisms or biotransformation 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 log Kow 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 foodweb in a model that can be time-varying used to applying the stochastic of agricultural 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 (Kow)-based screening bioaccumulation and biomagnification evaluations for regulatory purposes.

63 Improved assessment of pesticide peak exposure in cultivated mountain watersheds
M. Morselli, University of Insubria / Department of Science and High Technology; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; A. Di Guardo, University of Insubria / Department of Science and High Technology

Agricultural activities can involve the use of plant protection products (PPPs) and the use of such chemicals can occur near surface waters 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 dynamic 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 chlorpyrifos water concentrations measured in the Novella River (Non Valley, Northern Italy), where more than 1000 ha of apple orchards surround the river and 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 Institute, 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 PRZMS and VFSMMD 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, Norway 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 meteorological data. The model is applied to a range of specific crop and application scenarios. Another important new feature is the implementation of various mitigation measures such as vegetated filter strips, hedges, tilage/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 examples 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.

Hydrophobic Chemicals and Mixtures: Reliable Investigations
Acute Toxicity of Pyrene Associated with Dissolved Organic Matter of Various Molecular Weights to Daphnia magna
H. Lin, 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 was dissolved organic matter (DOM) 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) > low molecular weight (LMW, < 1K Da, 1-3K Da, and 3-5K Da) DOM. Furthermore, the superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD) activity of Daphnia magna decreased gradually with the increasing C0.50 in the systems of MMW and HMW DOM, whereas increased when C0.50 was at a low level and then decreased when C0.50 was at a higher level in the control group with pyrene only and the system of LMW DOM. The influencing factors of DOM have been classified as being attributed to the toxicity of DOM-associated pyrene to 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.

Passive dosing for constant concentration and defined composition of hydrophobic organic mixtures
K. Hannneshøj, Technical University of Denmark / Department of Environmental Engineering; H. Birch, DTU Environment / Department of Environmental Engineering; K. Knudsmark Sjøholm, University of Copenhagen / Dept. of Plant and Environmental Sciences; P. Mayer, Technical University of Denmark / Department of Environmental Engineering
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 Kow and Koc. 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.

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-labeled Tetralin and Decane was conducted on 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 99.29% (N=90) for Decane and 104.34% (N=90) for Tetralin 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.

Untangling the biodegradation of hydrophobic chemicals in OECD and novel tests using a unified modelling approach
F. Polcin, Technical University of Denmark (DTU) / DTU Environment; A. Bendtsen, DTU Environment; F. Sibany, K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaef er, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Kästner, Helmholz 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, proparate 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 the context of OECD degradation tests and a variation designed to untangle the biodegradation of hydrophobic chemicals in 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.

History of polychlorinated biphenyl deposition to snow and ice from the Lomonosovfonna glacier, Svalbard
M. Krypianou, Hermanns & Associates LLC / Environmental Chemistry; O. Garmash, University of Helsinki; E. Eivaksen, Norwegian Polar Institute; C. Teixeira, D.C. Muir, Environment and Climate Change Canada / Aquatic Contaminants Research Division
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-elevation ice on Svalbard at 1250 masl. It is above the tropospheric boundary layer at all times of year (maximum ~1000 masl), so all of the contaminant inputs have sources from long...
ICLIA method developments in a global perspective: Status and outlook

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

M. Owsiak, Technical University of Denmark; G. Cornelissen, S. Hale, University of Strathclyde; M. Sparrvik, NTNU

The development of spatially differentiated life cycle impact assessment (LCA) 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 LCA results. The aim of this work was therefore to 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 biocar 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, regionalized impacts were generally lower than 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 LCA, biocar 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 ranking 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 biocar production using both Kon Tiki and Adam retort kilns performed better than earth-mound kiln, and furthermore biocars 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

T. Maury, University of Bordeaux / ISM-CyVi; P. Loubet, CyVi-ISM / ISM CyVi; A. Gallice, ArianeGroup / Design for Environment; G. Sonnemann, University of Bordeaux / ISM CyVi

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 10 cm, 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 space missions becomes a key issue for future space missions. 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 orbit 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 decision Volume occupation by debris and dead space is a key reason for the 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-EURoS 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 larger effect factors in these areas. Some of the worst impacts were in Slovenia, i.e. 7.6·10⁻³ km² eq7 yr/km² eq7. The area-weighted European average CF was 2.9·10⁻³ km² eq7 yr/km² eq7. 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 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.

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

Review on removal and reactions of micropollutants in biofilms under growth and non-growth conditions
K. Bester, Aarhus University / Environmental Science; M. Escolá 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. Toretti, Kruger A/S; H. EI-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 2-3 days in biofilms. In this contribution it is highlighted which ecological conditions (aerated versus denitrifying; high and low BOD loads) have been found to favor degradations. 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 compounds that were thought to be recalcitrant (like diclofenac) could easily be degraded in relatively short time periods. For single compounds degradation pathways for biofilm systems are discussed and compared to other systems. – While oxidation pathways are relatively common it seems like biofilms often perform a combination of oxidation and sulfatation pathways. Interestingly enough, it was possible to reach high removal rates for otherwise persistent ozonation–by-products such as macrolide N oxides while avoiding back reactions to the parents with a moving bed biofilm reactor (MBBR). This also holds for most of the ozonation products of diclofenac.

Biodegradation of emerging organic contaminants using an enzyme-mediator system and study of the resulting transformation products
I. Caraeanu, 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 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diaminonium 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.
and high resolution mass spectrometry (HRMS, LTQ Orbitrap) was used to identify potential transformation products. Furthermore, the microbial activity of the antibiotics and their transformation products was assessed, using an \textit{E. coli} culture and microbial disks. 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 \mu M, 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 by 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 performed assessing the possible health and environmental risks associated with the reuse of treated wastewater, for applications such as irrigation and groundwater replenishment.

79 Evaluation of macrolide antibiotic transformation in model biodegradation and ozonation experiments using target and non-target analyses and ecotoxicological bioassays
S. Terzić, Rudjer Boskovsk Institute / Division for Marine and Environmental Research; P. Kostanjevecki, I. Kozman-Matašić, I. Senta, Rudjer Boskovsk Institute; T. Jurina, Faculty for Food Technology and Biotechnology; N. Udiković-Košć, Rudjer Boskovsk Institute; J. Ćurko, Faculty of Food Technology and ERY TSPs; M. Matosić, Faculty for Food Technology and Biotechnology; J. Lončar, I. Mihaljević, T. Smitl, Rudjer Boskovsk Institute; M. Ahel, Rudjer Boskovsk Institute / Division for Marine and Environmental Research
The aim of the present study was to investigate the transformation of three prominent representatives of macrolide antibiotics (azithromycin - AZI, clarithromycin - CLA and erythromycin - ERY) in model biodegradation and ozonation experiments. The study included determination of the biodegradation kinetics of the parent compounds, identification of transformation products and ecotoxicological evaluation of transformation processes using two different end-points. The biodegradation efficiency was studied using the sludge culture enriched in the presence of AZI (10 mg/L) over a period of 4 months while the ozonation experiments were performed in different matrices by applying selected pH conditions and ozone concentrations. The dissipation kinetics of parent compounds as well as the formation of transformation products (TPs) were followed by ultra-performance liquid chromatography/quadrupole-time-of-flight mass spectrometry. Antibiotic activity test was based on the inhibition of bacterial growth (\textit{Bacillus subtilis}), while toxicity test was performed with the freshwater green alga \textit{Desmodesmus subspicatus}. At the applied experimental conditions, both biodegradation and ozonation experiments resulted in nearly full elimination of the tested parent compounds. The biotic and abiotic removal of all parent compounds was associated with the formation of different TPs, some of which were rather abundant and persistent to further degradation. The highest number of detected TPs was associated with the elimination of AZI, while the number of CLA and ERY TPs showed a significant decrease following the biotransformation and ozonation experiments. The environmental relevance of the identified biotransformation products, some of which included previously unknown linearized TPs, was proven by screening the municipal wastewater extracts for their presence. The effect-driven evaluation of the studied transformation processes, based on toxicity to algae and residual antibiotic activity, indicated a significant reduction of harmful effects, however, formation of numerous stable transformation products, warrants further ecotoxicological assessment.

80 DI-SPME - On-fiber Derivatization - GC-MS. An innovative green and cost-effective approach to determine CECs and TPs from a novel anoxic-aerobic photobioreactor
R. López-Serna, Universidad de Valladolid / Chemical Engineering and Environmental Technology; E. Posadas, P.A. García-Encina, R. Muñoz, Universidad de Valladolid
The demand of multicomponent methods for the analysis of compounds of emerging concern in environmental matrices is a reality today. However, conventional techniques based on Solid Phase Extraction (SPE) coupled to Liquid Chromatography Mass Spectrometry (LC-MS) are very often only available in high-tech laboratories. The cost-competitive methodology presented here, successfully developed and validated, intends to fill the existing gap between current environmental needs and analytical capacities. It consists of an innovative method for aqueous matrices. Two experiments have been performed under hydrodynamic conditions in a growth chamber aiming to understand the uptake, translocation and metabolisation of the organic pollutants. Two other experiments compared the effects of season, plant presence and plant species, initial concentration, hydraulic loading rate and CW design in different pesticides and pharmaceutical compounds. A sixth setup targeted the impact of support matrix in compounds sorption and biological activity. The plants \textit{Thlaspi alpestre} and \textit{Phragmites australis} were the most efficient plant species in removing ibuprofen and ibezoxol. Phragmites was the most efficient species to remove the pesticides tefubconazole and imazalil. Uptake, translocation and degradation of chiral pesticides inside the
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 bacterial processes play an active role in the micropolutants bio degradation. The removal of the organic micropolutants in CWs is affected by several design and operational parameters. Plant uptake does occur but phytoremediation 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 mg/kg body mass). 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 creatinine 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 mean 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.
A. Lopez-Antequera, Universiteit Antwerpen / Biologie; T. Groffen, Systemic Physiological and Ecotoxicological Research (SPHERE), University of Antwerp / Biology; L. Bervoets, Universiteit Antwerpen, R. Lasters, E. Prinsen, H. Abd Elgawad, University Antwerp; M. Eens, University of Antwerp / Department of Biology

Perfluoroalkyl acids (PFAAs) are substances which have been produced for more than 70 years. 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 clothes 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 information on their effects in wildlife. We report here for the first time 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 hatch 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 JH, de Voogt P, Jensen A, Kannan K, Mabury SA, Van Leeuwen SP (2011). Perfluorooctyl and perfluoropolyfluoroalcohols in wild birds:implications for exposure, bioaccumulation, and health effects. Env. Toxicol. Chem. 30:1722-1730. [2] Leeuwen SP (2011). 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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The ingestion of lead ammunition is the most important source of exposure to this metal in birds of prey, and consequences on their health are well-known. The objective of the present study is to improve our knowledge on the exposure to Pb in birds of prey in Spain by means of passive and active monitoring. Here we present positive results on the programs based on Pb analysis in blood and liver of raptors and by the evaluation of the effects on their health by using non-destructive blood biomarkers. We have 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) captured alive in the field. Correlated adverse effects of Pb include renal and respiratory dysfunction, PCa metabolism, oxidative stress and immune function were also evaluated in the active monitoring by means non-destructive biomarkers. The active monitoring showed that some individuals of bearded 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/g). Passive monitoring revealed that the species with lead levels in liver associated with clinical poisoning (18-30 µg/g dw) were cinereous vulture (1/3), Eurasian griffon vulture (2/228) and western marsh-harrier (1/32); and the species with clinical severe poisoning (>30 µg/g dw. 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 concentration of Pb in liver and Pb-exposure was also affected by Pb exposure, because elevated blood Pb levels were associated with lower Pb levels 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 more complete picture of risk for raptors and help to determine the synergies between Pb exposure and some other pollutants such as PFAAs. The active monitoring revealed that the highest blood Pb levels were observed in birds of prey with the highest 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 more complete picture of risk for raptors and help to determine the synergies between Pb exposure and some other pollutants such as PFAAs. The active monitoring revealed that the highest blood Pb levels were observed in birds of prey with the highest blood Pb levels, indicating a possible allocation of antioxidants in plasma to cope with adverse effects of Pb.
analysed by HPLC-DAD. Egg content showed the following OC levels (mean ± SE; µg/g dw): p,p'-DDE, 152.5 ±1.7; p,p'-DDE, 0.118 ± 0.020; PCBs, 0.459 ± 0.121; HCHs (hexachlorocyclohexane isomers), 0.021 ± 0.003; and HCB (hexachlorobenzene), 0.0042 ± 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 content 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. Croose, The Vincent Wildlife Trust; M.G. Pereira, 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. All 154 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: MO035, MO036, MO083

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

89 The threshold option, the recovery option and landscape modelling
P. Thorbeck, Syngenta / Environmental Safety; N. Galic, Syngenta / Environmental Safety; V. Forbes, University of Minnesota / Ecology, Evolution & Behavior
Landscapes provide a multitude of ecosystem services, but the relationships between the populations of the organisms providing them, stressors and the delivered 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.

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

90 Understanding risk - a better approach to reduce uncertainty
M. Wang, WSC Scientific GmbH / Dept E fate 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 for answering the question on the relevance of effects on 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. Newey, Crop Science and Environmental Safety; 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 scheme of 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. arable farms, dairy farms). Landscape 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 creation of highly realistic 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 Collombolans
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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 collombolan 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 look at exposure and effect. We will present the individual-based model of the soil-dwelling collombolan 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 four different regimes while all other parameters were kept constant (21 days for 3 different regimes). The dispersal and simulation results of the vertical dispersal of collombolans 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. Mintran, University of Exeter / Biosciences; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; S.K. Maynard, AstraZeneca / Safety Health and the Environment; A. Brown, Exeter University / Biosciences; C. Liu, Department of Environmental Biology; S. Parker, Cefas Weymouth Laboratory; P. Throbek. Syngenta / Environmental Safety; C. Tyler, University of Exeter / Biosciences

Population models are employed in the environmental risk assessment (ERA) of chemicals, including plant protection products (PPP), 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 interactions 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 parameterized with field data obtained in the laboratory. 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

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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 pathway 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, Victoria, and the Cascadia region host major ports, numerous refineries, paper mills, and high tech industries. The same area is also noted for intensive 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 for water 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)

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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 surfaces 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 mg 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 mm 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 sICP-MS. Results of the first experiment show that mobility of Ag along the soil column is significantly higher in the columns with earthworms overtime. 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.
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 remediation 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 (ReSoLu 01A). Additionally, a medium clayey silt (Luvisol) was used for the column experiments. The column remediation of the Ag concentration after digestion (Ag\textsubscript{dig}) was on a very low level in all percolation steps in both soils. The first percolation step after three days of the Cambisol incubation showed the highest remineralization of Ag which was below 5% of the Ag\textsubscript{org} concentrations in the soil columns. The correlation between remineralized Ag\textsubscript{org} and Ag\textsubscript{total} concentrations suggests that the remineralized 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. Similarly, columns with preferential flow pathways showed low Ag ENP retention. 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\textsubscript{total} in the lysimeter experiments was low and very likely related to soil tillage as well as bioturbation. A low Ag\textsubscript{org} release to the percolate water (\(\approx 400\) d, control \(=\) 24 ng l\(^{-1}\), Lysimeter (7 mg kg\(^{-1}\) \(=\) 56 ng l\(^{-1}\), DIN 38402-11) was obtained for the lysimeter with the highest Ag ENP application. Ag\textsubscript{total} was measured to a steady state condition, with 80% of the Ag\textsubscript{org} detected in the lysimeter with the lower Ag ENP concentration. All roots (wheat, canola, barley) showed a low uptake of Ag\textsubscript{total}. All approaches showed a more or less high retention of Ag ENP in soils why soils are a sink for Ag ENP. However, the demobilization 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 microcommunity and the root uptake (e.g. beet) needs further long-term investigations.

97 Determination of attachment efficiency (\(\alpha\)) for ENPs in different types of soils by saturated column experiments
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The attachment efficiency (\(\alpha\)) 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 \(\alpha\). Due to the complexity of the soil composition and texture, a small change in particle size 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 \(\alpha\) values for nominally 20 and 80 nm citrate coated gold ENPs (Au ENPs), as well as 30 nm sulphonated silver ENPs (AgS ENPs) were induced in a steady state condition and spiked into different soils sampled in the UK. Artificial rainwater was used as the eluent. 10 mM NaNO\(_3\) 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. \(\alpha\) 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 \(\alpha\) 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 \(\alpha\) 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 give higher \(\alpha\) values and cannot be accounted for in the equations used for estimating the attachment efficiency. In conclusion, when varying the initial ENPs concentration into the columns, the \(\alpha\) value is significantly affected. Hence, low \(\alpha\) concentrations need to be used in the column experiments to minimize the reproducibility of calculated \(\alpha\) values. Moreover, inclusion of air in the systems appears to induce artefacts that complicate determination of \(\alpha\) for specific NP-soil combinations.

98 The transformation of copper and zinc (-nanoparticles) during sewage sludge digestion
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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 phosphorus 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 digestion sludge to mimic short-term sludge storage conditions and combustion conditions 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 Zn spectrum 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. Comparably, the green form of CuO and CuS were returned from LCF analyses. All Cu spectra of the sludge and the ashes were very similar 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 nanoflours. Therefore, the soil ecotoxicity and dissolution of ionic silver vs nanosilver were tested. The smallest silver nanoflour with the highest specific surface area registered under REACH was used for testing (aqueous suspension containing approximately 37% nanoparticles, degree of sulfidation (~18% ZnS) with a concomitantly high fraction of ZnO (17%)). The effect of silver was tested as source of ionic silver. Soil nitrification was tested according to the internationally standardised and accepted assay for testing toxicity to soil microorganisms (OECD Test Guideline No. 216). Three soils were selected falling within the P10-P90 interval of European agricultural soils for pH, organic carbon content and cation exchange capacity. Total soil in silver, and total dissolved (0.45 µm membrane filtered) and truly dissolved silver (1 kDa centrifuge filtered) in porewater were measured (ICP-MS). Toxicity of nanosilver on soil nitrification was similar to or less than silver nitrate when expressed on the basis of total Ag in soil. Total and truly dissolved Ag in porewater decreased over time after silver nitrate spiking, suggesting ageing processes. Concentrations were always higher or equal to corresponding values after nanosilver spiking. For nanosilver spiking, total and truly dissolved Ag in porewater initially increased and decreasing dissolution processes. From day 4-7 after spiking onwards, concentrations decreased over time suggesting that ageing becomes more important than dissolution. Truly dissolved Ag in porewater qualitatively explained observed toxicity of silver nitrate compared to nanosilver. The data show that soil ecotoxicity data for ionic silver are conservative for soil ecotoxicity of nanosilver.

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 when, unless soluble, it is usually determined 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 homomaggregation. 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 homomaggregation. 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 homoeaggregation is the complexity added to the system by SPM in the case of heteroeaggregation. In this contribution we therefore propose an approach to develop a heteroeaggregation 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 homoeaggregation conditions complex enough to represent relevant environmental characteristics, and simple enough for routine testing, (2) an easy-to-handle experimental setup to estimate a heteroeaggregation 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 homoeaggregation in the OECD TG 318 and will also apply for heteroeaggregation. 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 heteroeaggregation.

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
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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 hopen/stereane 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 shoring 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 environmental factors-model used in the current study. 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
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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. (NGOM) resulted in over 780 million liters of crude oil spilling into Gulf waters.

103 Physiological and molecular impacts of crude oil and/or dispersant-contaminated seawater and sediments on the sponge Halichondria panicea (phyllum Porifera).
J. Vág, 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 (phyllum 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 (spogue 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-hyclic aromatic hydrocarbon (PAHs) are a class of organic contaminants composed of two or more fused carbon rings and are a major constituent of crude oil. Exposure to ultraviolet radiation (UV) can exponentially increase the toxicity of photodynamic PAHs to biota, resulting in adverse outcomes well beyond the threshold of other mechanisms of toxicity. This phenomenon is known as photo-induced toxicity and is well documented in a wide range of aquatic organisms. Consequently, laboratory tests investigating effects of PAH on aquatic biota which fail to account for potentiation by UV may significantly underestimate toxicity. The intensity of UV exposure to biota is highly variable within aquatic ecosystems, due to a number of factors intrinsic to the water column, and extrinsic factors (e.g. cloud cover, time of day, seasonal variations). Tissue repair mechanisms may be sufficient to counteract some effects of photo-induced toxicity during periods of relief from UV exposure. Here, we report the results of experiments in which larval red drum (Sciaenops ocellatus) and zooplankton (Daphnia magna) were exposed to either a single PAH (fluoranthene) or a complex PAH mixture prepared from weathered crude oil with varying PAH and UV exposure scenarios. Red drum tests were conducted as a single pulse exposure, and daphnia tests were conducted as static renewals. Toxicity (LC50) was UV and PAH dependent in both species. In red drum tests, shorter PAH pre-exposure times resulted in LC50 that were considerably lower than LC50 associated with longer pre-exposure periods. This is likely due to lag time in the initiation of physiological metabolisms/clearance mechanisms in the organism and loss of PAH from the test chamber. A similar pattern was observed in photoperiod testing for both species. Significant latency in mortality was observed in daphnia for several days after the conclusion of the UV and PAH exposures. We also report the effects of various UV-modified pharmaceuticals on marine fishes. Taken together, these data suggest that even short-term, transient exposure to low concentrations of PAHs (common during a sp oil event) results in acute toxicity in aquatic organisms, and those effects may be manifested outside of standard bioassay testing durations.

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

24 SETAC Europe 28th Annual Meeting Abstract Book
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. Phototoxicity 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-R). UV-R penetrates the water column, and weathered middle distillates, crude and heavy oils can exhibit photoenhanced toxicity. These same products do not exhibit phototoxicity 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.

104 Pilot microcosm study to assess the fate and toxicity of diluted bitumen in an outdoor aquatic environment.

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Pipelines are the sole mode of transporting Canada’s oil to markets, but they are a concern for the public, especially the potential effects of diluted bitumen (dilbit) spills on the environment. We added diluted bitumen (dilbit) to two land-based microcosms (2 m diameter) containing water and sediment from a nearby lake at the ISD-Experimental Lakes Area in Northwestern Ontario for a span of 11 days, and compared our results to a control enclosure with no added dilbit. Microcosms were treated with 0 (Control), 0.15, or 1.5 liters of Cold Lake Winter Blend dilbit (CLB-W), representing dilutions of 1:10,000 and 1:1000 oil/water, v/v, which spans the range of historical dilbit spills to water. Samples of water, sediment, air and oil were collected through the study in order to determine the fate, weathering, and behaviour of the dilbit. Total petroleum hydrocarbons in the high treatment microcosm gradually increased from under 100 mg/L in the first 24 hours to over 1200 mg/L by day 11, with no evidence of reaching equilibrium over this duration. Although a decrease in total phytoplankton biomass was observed in all microcosms over the study, the biomass in the high microcosm was about one-half or less than that in the control microcosm for the first week. Thereafter, the rate of biomass loss in the dilbit-treated microcosms slowed down, which could indicate recovery of the primary producers as the oil slick sank to the sediments. This study is among the first to examine the behaviour of dilbit in an outdoor setting under natural conditions of sunlight, wind and rain, and provides a case study that will inform future dilbit studies in natural (outdoor) environmental settings.

Fish model species in human and environmental toxicology (II)

107 Life-stage, and species-specific effects of dietary methylmercury exposure K. Barrow, University of Saskatchewan / Advanced Environmental Research Institute; Y. Zhang, University of North Texas Health Science Center; T. Curran, J.T. Magnuson, University of North Texas / Biology; M. Allen, University of North Texas Health Science Center; B.J. Venables, A.P. Roberts, University of North Texas / Advanced Environmental Research Institute

Mercury is a globally distributed contaminant, found even in remote aquatic ecosystems. Once deposition occurs, it can be microbialy 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 fish exposed to maternally transferred dietary MeHg on a model fish species (Pimephales promelas). Exposure to environmentally relevant concentrations of MeHg during development led to alterations in the dopaminergic system, metabolism, gene expression, behavior, hatch time, size, and embryyo-larval survival. Similarly, effects on the dopaminergic system in specific regions of the adult P. promelas brain were observed after a 30-day dietary exposure. Recently, a functional link between gut microbiota and dopamine 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 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 teleost 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 implications 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.

108 Characterization of molecular toxicity pathways of Fluoxetine in rainbow trout and white sturgeon using RNA-Seq whole transcriptome analyses A. Alcaraz, University of Saskatchewan - Toxicology Centre / Toxicology Centre; B.K. Eisner, University of Saskatchewan / Toxicology Centre; S. Tang, Chinese Center for Disease Control and Prevention; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

The increasing number of emerging chemical contaminants (ECs) 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 toxicological study, of which will allow the study of toxic pathways. Hence, there is a 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 improve their effectiveness for the objective of evaluating toxicity pathways 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 96th static renewal system, and sequence-by-synthesis whole transcriptome analysis was used to determined unique differentially expressed 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 study of toxic pathways across multiple levels of organization with the end goal of identifying molecular markers that are indicative of adverse effects.

109 Transgenerational effects of early life stage exposure to endocrine disruptors across biological scales in a euryhaline model fish S.M. Brander, Oregon State University / Environmental and Molecular Toxicology; B. DeCourten, J. Forbes, University of North Carolina Wilmington / Biology and Marine Biology; N.P. Burns, University of North Carolina Wilmington / Department of Biology and Marine Biology; H. Roark, Hunter Roark / Biology and Marine Biology; J.W. White, Oregon State University / Department of Fisheries and Wildlife; A.C. Mehinto, Southern California Coastal Water Research Project / Toxicology; M.L. Settles, University of California Davis / Genome Centre; R.E. Connom, University of California, Davis / School of Veterinary Medicine

Emerging research demonstrates that ECs, which agonize, antagonize, and/or synergize the effects of endogenous hormones, can cause deleterious effects in a range of species as a result of early-life effects as well as transgenerational effects. A scarcity of studies exist on model fish species, such as Menidia beryllina, a euryhaline fish with short generation time that is found在全球 North America and is demonstrated to be sensitive to contaminants. As such, we exposed Menidia beryllina embryos (8 hpf) until 21 dpf to a potentially relevant endocrine disruptor (nonylphenol) or estrogenic EDC of emerging concern: levonorgestrel (Levo) (10 ng/L), bisphenol A (BPA) (5 ng/L), respectively, and coupled this exposure with testing of an established androgenic or estrogenic EDC: trenbolone (TB) (10 ng/L), and ethinylerstadil (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 dpf) 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...
110 Integrated OMICS and imaging for a better understanding of ecotoxicological mechanisms - PAH developmental toxicity as an example
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Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous contaminants in the environment. Many of them cause developmental defects in fish, and cardiovascular tissue seems to be the most sensitive tissue. The mechanisms of toxicity remain largely unresolved for many PAHs, though partial adverse outcome pathways (AOPs) exist for those that are aryl hydrocarbon receptor (AhR) agonists. Rainbow trout (Oncorhynchus mykiss) yolk sac larvae were exposed to sublethal concentrations of PAHs that act via different mechanisms of toxicity: Retene, an AhR agonist causing dioxin-like toxicity; pyrene and phenanthrene, weak AhR agonists causing toxicity independently of AhR; and fluoranthene, a CYP1A inhibitor interfering with PAH metabolism. Also the effects of a mixture of retene and fluoranthene were studied. Information was gained at multiple levels of biological organization to reveal the mechanisms of toxic action. Changes in cardiac transcriptome, proteome and metabolome were explored over time. Physiology and function of the heart were also studied. At the whole organism level, growth, yolk consumption, and developmental defects and abnormalities were monitored. Each PAH caused a unique pattern in OMICS analyses, and the mixture of retene and fluoranthene caused a distinct transcriptomic profile from that of each of the single compounds. Retene differentially regulated genes involved e.g. in muscle contraction and ion metabolism (ion channels). Retene and phenanthrene impaired cardiac function in larval rainbow trout. Both causes bradycardia, and phenanthrene caused also arrhythmias. Phenanthrene affected cardiomyocyte electrical characteristics. As cardiovascular development is modulated by the beating heart and blood flow, alterations in cardiac function during development may have long-lasting impacts in cardiovascular tissues. Different PAHs clearly have different mechanisms of toxicity. The transcriptomic changes can at least partly account for the cardiotoxicity of retene, but the cardiotoxicity of phenanthrene seems to involve a direct effect on cardiac ion channels.

111 Physiological / Reproductive Status of Native Fish Exposed to a Complex Chemical Mixture in the BioBio River, Central Chile
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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 and fragmentation of the use of natural resources, such as agricultural and urban activities, with many focal places of point and diffuse contamination along its main axis, in addition to a high degree of modification of land use associated with the basin. The high degree of intervention and fragmentation of this river has affected the biota and water quality mainly in its lower third, due to the convergence of complex chemical mixture and anthropic interventions in the main course. The objective of this study is to determine how the development associated with this river impacts the physiological/reproductive state of the native species Percilia irwini (n = 66) in situ. Different sublethal responses were evaluated through biomarkers at different levels of biological organization (Biochemical, cellular, and individual) and environmental parameters (pH, temperature, conductivity and total dissolved solids). The results obtained indicate an increase in the Hepatic EROD activity (ethoxyresorufin-O-deethylase) towards the lower third of the river. The Gonadosomatic Index (100 * (Total weight / (Total weight - Gonadal weight))) shows an increase towards the lower third, however, the gonadal histology indicates a protoplasmic growth and reduction of the diameter of the 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.

112 Sustainable Development Goals: the global context defining the agenda for government, business and academia

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

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

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Mercury Biogeo sciences - Fate, Effects and Policy

119 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 environment- gaseous elemental Hg, gaseous oxidized Hg (Hg(I) or Hg(II) compounds), and that bound to particles. Gaseou
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 emerged in the past decade. However, regarding the severity of this controversy, a consensus on this issue is still widely debated. Here we evaluate Se:Hg molar ratios across a latitudinal gradient in the Northeast Atlantic Ocean (NEAO) fish communities to assess species variation and spatial trends. In this study, the concentrations of total Hg and Se in 17 teleost fish species were measured using ICP-MS following microwave digestion/Julshamn, 2007 #72) and the Se:Hg molar ratios were calculated. Marine fish samples (n = 8525) were 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 wolfishes (Anarhichas spp.), respectively. The mean Hg levels ranged from 0.44 to 0.77 mg kg⁻¹ ww with the lowest level in Atlantic mackerel (Scomber scombrus) and the highest in blue line (Mola diazipterygia), leading to variation in the mean Se:Hg molar ratios from 1.9 in blue line 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 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 brossme) (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 concentration in the EU maximum level of Hg and a 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

J.R. Gersson, Duke University; L. Naslund, Duke University / Biology; H. Hsu-Kim, Duke University / Department of Civil Environmental Engineering; E. Bernhardt, Duke University / Biology

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 these elements in water are often toxic to 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, only two published studies have simultaneously examined the interaction of Hg and Se in both multicellular organisms and environmental media containing microbes (sediment, water). 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 and 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 speciation 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 evidence that Se is an antagonist to Hg toxicity, and that biota containing elevated concentrations of these two contaminants. Our data provide evidence of a wedge-shaped cluster for the relationship between Se and percent MeHg in bulk sediment and biofilm, suggesting that at high concentrations, percent MeHg in bulk sediment and biofilm are reduced. In crayfishes and spiders, we find a negative correlation between Se concentration and both absolute MeHg concentrations and percent MeHg. These results suggest that Se inhibition of MeHg accumulation might occur both at the microbial and macroinvertebrate levels.

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

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Uncertainties in global mass balance of mercury are constrained in this work using all the currently available observations of mercury species in the global environment, and a previously published multimedia model for mercury, UncoroM3. Results help 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, resulting in reduced uncertainties (95% confidence) on relations between different mercury fractions, 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 marine redox reactions limitations contribute more 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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Microbial transformations of monomethylmercury (MMHg) and dissolved gaseous mercury (DGM) at the lower marine trophic levels are still not well understood. This is especially important in oligotrophic and nutrient-limited seas, where microbial food web and microbial loop dominate over classical (herbivorous) food web. Our research focused on the examination of the effects of probable phosphorus limitation (P-limitation) on relations between different mercury fractions (total methylated mercury – MeHg, and DGM) and autotrophic and heterotrophic microorganisms. We determined total mercury (THg), MeHg and DGM, alongside with relevant microbiological and chemical parameters in the Central Adriatic Sea. Using statistical analysis (non-metric multi-dimensional scaling, principal component analysis, Pearson’s product-moment correlations), we assessed the microbial effects on Hg transformations and bioaccumulation. Only in the absence of P-limitation conditions (00 P-limitation), we found that MeHg was significantly related to most chemical and microbial parameters, which is an indication of metabolism-dependent Hg transformations. The activity of heterotrophic low nuclear acid bacteria seems responsible for most of Hg methylation in seawater under 00 P-limitation. Under 00 P-limitation conditions, DGM shows strong correlation with microbial fractions and chlorophyll a, which confirms previous research about biological DGM production. Contrary to MeHg, DGM transformations are probably not metabolically dependent, as most of these correlations can also be observed under 00 P-limitation. MMHg biomagnification from microeston to mesozooplankton was observed through an increased biomass coronations factor. However, the MMHg fractionation and uptake are probably enhanced under 00 P-limitation, which emphasizes its impact on Hg transfer to higher trophic levels. In order to test our nutrient-limitation hypothesis, we have performed statistical analysis on previously published data from the Southern Atlantic Ocean. We found similar correlations between MeHg (DGM) and physico-chemical characteristics of seawater under probable nitrogen limitation, compared to those found under 00 P-limitation in our study. These results indicate that mercury methylation is impacted in seawater under probable nutrient limitations.

124 Poster spotlight: M0333, M0334, M0335

Bioavailability and realistic risk assessment of organic
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 measurement tool for a host of toxic risk assessment studies. However, 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 in a series of studies to our group on whole sediment microextraction (SMME) fibers in both sediment and ESR dosed water was used to model with three first order degradations (fast, slow and very slow) the really observed time necessary for biodegradation, it will be necessary to supply regulators with data and prediction to convince them that biodegradation will be a safe option to remediate the contaminated soil or sediment Bioavailability as measured with Tenax can be used to explain and predict the rate of biodegradation of PAHs. Three desorbing fractions can be measured. Tenax applied at 20°C gives the fast desorbing fraction applied at 60°C a real scale degradation curve could be predicted. Moreover, the fractions measured in present soils, shows that biodegradation will continue, however with a very small slow rate. Experiments applied in the nineties of last century had already shown that risks measured with biossays were already not measurable after 6 years of landfarming. After 25 years the PAHs concentrations were 10 mg/kg d.m. or lower which made the soil reusable within the Dutch legislation.

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 developments 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 then PAHs, are 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 24 h (ISO 17402) (Environ. Toxicol. Chem. 20:706–711, 2001). Integro 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 and PAHs, and chemical treatments) were measured in the original sediment and resulting soil. PAHs were present in concentrations up to 550 mg/kg d.m. (Dutch list). The objective of the investigation was to find experimental prove on the existence of long term biodegradation in field conditions. The measured data showed continuation of PAH degradation and this could be distinguished in 1) fast degradation in the first year 2) slow degradation in the following 6 years and 3) very slow degradation of PAHs from 6 years until at least 25 years. Knowing the long time necessary for biodegradation, it will be necessary to supply regulators with data and prediction to convince them that biodegradation will be a safe option to remediate the contaminated soil or sediment Bioavailability as measured with Tenax can be used to explain and predict the rate of biodegradation of PAHs. Three desorbing fractions can be measured. Tenax applied at 20°C gives the fast desorbing fraction applied at 60°C a real scale degradation curve could be predicted. Moreover, the fractions measured in present soils, shows that biodegradation will continue, however with a very small slow rate. Experiments applied in the nineties of last century had already shown that risks measured with biossays were already not measurable after 6 years of landfarming. After 25 years the PAHs concentrations were 10 mg/kg d.m. or lower which made the soil reusable within the Dutch legislation.

128 Prediction of very slow biodegradation of PAHs in soil and validation in a pilot of 25 years

R. Rietra, Akterra and Wageningen University / sustainable soil management; J. Harmsen, Wageningen Environmental Research / CALM

Biodegradation of polycyclic aromatic hydrocarbons (PAHs) and mineral has been followed during periods up to 25 years. Biodegradation took place on real scale fields (six landfarms and one depot) within the Netherlands. On these fields dredged sediments were used. PAHs were measured in the original sediment and resulting soil. PAHs were present in concentrations up to 550 mg/kg d.m. (Dutch list). The objective of the investigation was to find experimental prove on the existence of long term biodegradation in field conditions. The measured data showed continuation of PAH degradation and this could be distinguished in 1) fast degradation in the first year 2) slow degradation in the following 6 years and 3) very slow degradation of PAHs from 6 years until at least 25 years. Knowing the long time necessary for biodegradation, it will be necessary to supply regulators with data and prediction to convince them that biodegradation will be a safe option to remediate the contaminated soil or sediment Bioavailability as measured with Tenax can be used to explain and predict the rate of biodegradation of PAHs. Three desorbing fractions can be measured. Tenax applied at 20°C gives the fast desorbing fraction applied at 60°C a real scale degradation curve could be predicted. Moreover, the fractions measured in present soils, shows that biodegradation will continue, however with a very small slow rate. Experiments applied in the nineties of last century had already shown that risks measured with biossays were already not measurable after 6 years of landfarming. After 25 years the PAHs concentrations were 10 mg/kg d.m. or lower which made the soil reusable within the Dutch legislation.
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 bioavailability concentrations of the selected metals and 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 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 compound 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.

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

W.J. Doucette, Utah State University / Utah Water Research Laboratory; J. Finsinger, D. McAvoy, Utah State University

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 its biological activity varies from plant to plant. This study explored the sorption of contaminant using microcosms amended with biochar or biochar amended soils. 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 compound 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.

132 A midpoint indicator for freshwater resources

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Freshwater resource has been recognized as being a safeguard subject within the Actual Protection (AoP) of the Integrated Pollution and Protection (IPPC) directives. In the absence of contaminants 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 anti-inflammatory that prevents seizures and relieves nerve pain (carbamazepine), 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 were produced from tree species that often require removal because they are considered invasive or due to insect infestations. Corn was used as the test plant because of its commercial value and has been grown with reclaimed water in the past. After the 28 day growing period, it was found that there was no negative impact of the biochars on corn growth. Once the plant tissue analysis and sorption studies are completed, the impact of biochars on contaminant uptake will be evaluated along with the impact to PPCPs sorption to determine any potential effects on plant growth. Extraction and analysis of the plant tissue is being conducted along with the sorption/desorption experiments. Final results expected by December 2017.

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

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


Several life cycle impact assessment (LCIA) models have been proposed to
quantity 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 define the impact pattern of wastewater on ecosystem quality. We propose a new focus, a system LCA model based on freshwater fish habitat 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 Concentration Potential (HCP) is an indicator for fish ecosystem quality. We report a new modelling approach for fish ecosystem HCPs and invertebrates is proposed and applied in France at Q50 (wet season) and Q90 (dry season) flows. HCP represents the changes 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 performed for water point and sub-watershed. Successively, 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 however possible to find convergence between 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 compatible 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 tempts the use of LCA for fish based food and feed products. Current LCA methods do not assess the impact of biotic removal on biotic stocks very well. The dynamic of biotic stocks (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 determined by the marginal utility where the CF is used for quantifying a 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 fishes, 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 biotic 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 models 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) are used to indicate the long-term effect of agricultural land use. 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 characterisation 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, N 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 (III)

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 regimes 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 an ability to perform online monitoring with the simple concept of using recyclable compounds like carbamazepine to normalize sampling rates from in- and outlets of 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 time. 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. Pach, N. Montemurro, IIQAB-CSIC / Environmental Chemistry; D. Barcelo, IIQAB-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 hydroxyl radicals (·OH), peroxyl radicals (·OOH), 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 process. 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. Untreated water was used to determine 10 photo-TPs. 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.

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 (HILIC), and in the anaerobic reactor (ANA) of a WWTP. 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
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In addition to traditional and new generation water pollutants, microbial contamination is still one of the major problems that 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, but it is also important to decrease human footprints in the environment. Some hydrolytic and oxidative enzymes (i.e., Laccase) is capable of oxidizing unreacive iodide to reactive iodine, when they can play role 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 acetaminophen as mediator in a laccase mediatror system to generate disinfectant iodine. The stability of reaction can be changed depending on the pH, temperature and multiple compound existence and system optimization is required to stabilize iodine synthesis. In this study, two different free laccases and insolubilized as cross-linked enzyme aggregates have been tested. Also, iodine synthesis is investigated with different KI (0.01-1 M) concentrations and different enzymes (5, 10, 30 and 40 Unit/L) for 5 hours. Compounds were injected in distilled water as well as in the influent and effluent samples of wastewater treatment plants to see synthesis of iodine while the micropollutants have been removed in Laccase Mediator System. In the experimental sets, removal of persistent compounds were determined by LC-MS/MS and UPLC–HRMS. L-generating surface water samples originating from rivers were screened for their presence. For the enrichment of the potential photo-TPs, water samples were preconcentrated on four SPE cartridges connected in series and then analyzed using the same system mentioned above. With this methodology more than 30 photo-TPs were detected in the irradiated reactor samples. As of the time of submission of this abstract, the identification of 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.

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 are usually not well water soluble and tend to adsorb to sludge, soil and other particular matter, and thus spread slowly in the environment. However, as a consequence of their high environmental stability, they can eventually reach even remote areas like the arctic, and pose a threat to apex predators due to their ability to accumulate in the food chain. Persistent, mobile, and toxic (PMT) environmental contaminants, however, do not accumulate in the food chain, but are much more mobile in aquatic environments, and thus, they spread fast throughout the environment. Persistent toxicants, however, are a major human exposure pathway for PBT substances. While environmental contaminants of low and intermediate polarity have been thoroughly investigated, so far only little is known about the most polar, and thus potentially most mobile, water contaminants. This gap in knowledge might be caused by difficulties in the analysis of very polar organic chemicals, especially in their enrichment from aqueous matrices. PMT 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, and bromochloromethanesulfonic acid as well as O-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 contaminants. With this method, we monitored chlorinated and brominated MSAs throughout four drinking water treatment plants and in several tap 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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European Commission Joint Research Centre / Bioeconomy unit: A. Laurent, DTU / Division for Quantitative Sustainability Assessment DTU Management Engineering

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 were registered in the Life Cycle Impact Assessment (LCIA) 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 uncertainty of 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 organic and inorganic substances as well 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 LCIA 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. Beussisseur, INRA; E. Loiseau, Irstea; 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 detailed geographical inputs (industries, households, manure and pesticides application on agricultural soils) depend on the approach considered. Besides, geographical developments are 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. The approach considers the registered electricity production for past and present fleet over the 1980-2030 period. The results, through the on-line tool, are showcased as a map, where the individual performance of each of the past, present and future wind turbines can be consulted, as well as the performance of the whole fleet at a given year.

145 A regional life cycle approach for assessing the climate change mitigation potential of biodiesel for Switzerland
S. Okledge, Helmholtz Centre for environmental research - UFZ / BEN; S. Majer, DBFZ; Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; U. Franko, Helmholtz Centre for Environmental Research, UFZ / Department of Soil Physics; D. Thraen, Helmholtz Centre for Environmental Research UFZ / Deutsches Biomasseforschungszentrum gemeinnützige GmbH, DBFZ / BEN

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 biobased 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 biodiesel 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 (LCA) and the energy produced during the turbine's operation phase. This ratio depends on the specific geographic characteristics found in the region, using life cycle approaches, is important to support more sustainable regional resource management. Environmental models can also provide significant enhancements for assessing the 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 detailed geographical inputs (industries, households, manure and pesticides application on agricultural soils) depend on the approach considered. Besides, geographical developments are 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. The approach considers the registered electricity production for past and present fleet over the 1980-2030 period. The results, through the on-line tool, are showcased as a map, where the individual performance of each of the past, present and future wind turbines can be consulted, as well as the performance of the whole fleet at a given year.
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 Swiss 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 provides 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-level regions and different local areas. 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 the outcome of 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 as the state, 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 (habitats scenario) and biotic components (the functional and life history scenario). Technically, we integrate spatially 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 reverse outcome of chemical exposure.

150 Robust implementation of TKTD models with Bayesian inference
V. Budroot, Université Lyon 1; S. Charles, University Lyon 1 / Laboratory of Biometry and Evolutionary Biology
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 toxidynamics 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. Inter governmental organizations as the OECD have acknowledged 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 posterior. 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 codes (JAGS and Stan) have been used, we compared 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?
K. Dalhoff, University of Copenhagen / Department of Plant and Environmental Sciences; G. Bellisai, European Food Safety Authority EFSA; E. Neira, N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences

The azolefungicides 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 compound and synergistic interactions in the azoles and α-cypermethrin were a range of tests conducted with co-exposure to 1, 3, 10, 30, 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-modelling before applying the internal exposure scenarios to the observed acute data. 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 models can predict survival of C. riparius to explain these kinetic differences and how they relate to the observed toxicity. We further hope that models can predict survival of C. riparius to explain these kinetic differences and how they relate to the observed toxicity.
New Horizons in Particulate Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment

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

New method for the identification of microplastics and examination of their impacts on human health.

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

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 Pyrethroid 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 (Oncorhynchus mykiss) using TK-7D 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 could simulate the immediate toxic effects on the fish but does not pass the threshold for an effect on survival. This helps to explain why no mortality is observed in the peak exposure experiment. The no effect threshold for sublethal effects is passed in the modelling under constant exposure, which is consistent with the observations. In the peak experiment, the duration of the effect on the feeding behaviour is insufficient to induce large effects on growth in weight or lengths, because beta-cyfluthrin is rapidly removed from the experimental water and the fry have no fat reserves to cope with reduced feeding over a short period. The modelling supports the experimental finding that under realistic exposure conditions, short term effects on the feeding behaviour do not lead to growth or survival effects, and gives a mechanistic explanation for this observation. We were able to derive a mechanistic explanation for the results from laboratory experiments conducted with three different early life-stages of the trout, and for different exposure profiles to beta-cyfluthrin. The model shows that results from both laboratory studies are consistent. This validated model has the potential to be used to make accurate in silico predictions of effects on fish early life stages from time-variable exposure profiles.

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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 coastal waters) usually consist of metal, glass, wood and plastic, being the 80% of these plastic wastes. 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 particles 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 methods 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 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

B. Fernández, Instituto Español de Oceanografía / Marine Pollution and Biological Effects Department; M. Albentosa, Instituto Español de Oceanografía / Marine Environment and Environmental Protection Area. Fisiology and Ecotoxicology of Bivalve Molluscs Department

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,
158 Analysis of tire wear particles in environmental samples using TEG- GC-MS P. Eisenbraun, Bundesanstalt für Materialforschung und -prüfung; E. Dümichen, Bundesanstalt für Materialforschung und -prüfung / 5.3 Mechanics of Polymers; A.S. Ruhl, TU Berlin / Department of Water Quality Control; M. Kneck, TU Berlin; M. Albrecht, TU Chemnitz; U. Braun, BAM; Federal Institute of Materials Research and Testing / 5.3 Mechanics of Polymers Tire and road wear particles (TRWP) as environmental contaminants have received increased interest since the 1960s[1]. TRWP have adverse effects on human health[2]. Multiple cities in the EU are struggling with multiple sources or insufficient stability of markers. Therefore, financial penalties as well as consequences like vehicle bans in metropolitan regions are discussed. TRWP can be regarded as microplastics, because the rubber component of TRWP is mainly polymer (natural and synthetic). With regional differences, the contribution of TRWP to the microplastics emissions to the environment can reach up to 60%[3]. Analysis of TRWP is challenging because of the high variance in compositions of the particles. The established analytical methods suffer from unspecfic marker compounds, small sample size or low sensitivity[4-6]. The topic of this presentation is the analysis of TRWP using the recently developed method TEG-GC-MS (thermal extraction desorption gas chromatography mass spectrometry)[7]: Sample materials are heated in a thermogravimetric analyzer. The decomposition products are purged with nitrogen through a heated coupling device to a solid phase adsorber - extraction device. An auto-sampling robot transports the adsorber to a thermal desorption-GC-MS for further analysis. Rubber materials were provided by TUC, Sigma Aldrich and Avakal. The tire samples included used and unused materials provided by TUC, Umweltforschungszentrum Leipzig (UFZ) and former BAM projects. A bitumen and an asphalt material, provided by BAM were analyzed. As tire wear particles based on elemental composition and distinct elemental ratios. The analytical method aims to i) provide assisted acid digestion of ii) elemental and elemental detection of sulphur and carbon. A stepwise method development including analytical and chemical verification by determination of the rubber content is presented. In particular, S and Zn are present in characteristic concentrations in tires. Zn and S contents were determined in 30 tire samples as an internal reference. The average S content in the analysed tires was 15400 mg/kg (+6000 SD), while the average Zn concentration was 1700 mg/kg (+1700 SD). Furthermore, the developed method was applied to field samples. Samples were taken from the intake water of a treatment facility (raw water), from the sedimentation basin, the inlet of the soil retention filter as well as from the soil itself. Isolation of tire wear particles by density separation was achieved by use of a heavy liquid, sodium polytungstate mixed with MilliQ water. Separated fractions were acid digested with microwave assistance and elemental analysis was conducted by 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 BBW for provision of samples.

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) / DTU Environment; A. Verschueren, BAM / Centre for Safety of Substances and Products; T. Huffer, University of Vienna / Department of Environmental Geosciences; M. Hasselöv, University of Gothenburg / Department of Marine Sciences; R.C. Thomson, Plymouth University / School of Marine Science and Engineering. The occurrence and accumulation of plastic debris is a global environmental issue, with potential consequences affecting the economy, wildlife and human health. However, there is currently a lack of consensus on the definition and categorisation of environmental plastic debris, including macro-, micro- and nanoplastics. The lack of clarity in terminology regarding plastic debris, in particular microplastics, results in confusion and misunderstandings. This is problematic both for legislative measures as well as for general coherence and data comparability between studies. While including a common language is a vital step towards addressing microplastics, any definition should be well justified as it will ultimately shape the direction of future research and legislation. To help decide whether a consensus definition and categorization framework for plastic debris is valuable and if so how this might look, the scientific community needs to engage in a critical discussion. The aim of our presentation is to foster such discourse in the SETAC community by providing impulses and sharing our thoughts rather than providing definitive answers. In our presentation, we will use a new format with two presenters jointly discussing the advantages and disadvantages of a definition. Further we will discuss our ideas on relevant components of a definition/categorization framework. To get immediate feedback by the community, we will use online polling asking specific questions to the audience throughout the presentation. This will cover opinions on the need of a definition, acceptance of certain definitions and categorizing criteria and questions on special cases with high uncertainty. The aim of this is to get an ad hoc idea on where consensus may be easy to achieve and areas which are controversial. Finally, we will present an online platform (www.microplastics.eu, currently under development) that we will use to perform a large-scale survey on a consensus definition of environmental plastic debris. In addition, the platform will host a module for discussing the questions mentioned above and a module for networking. This platform can be used by the audience and the wider community to further discuss the impulses we give and share their opinions and input.

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

161 Behavioral and physiological responses of bicolor damselfish and mahi-mahi to olfactory cues following crude oil exposure L. Scholz-Kammer, RSMAS, University of Miami / Marine Biology and Ecology; M.J. Weiss, James Cook University; N. Benetti, RSMAS University of Miami; J.D. Stiegitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society; P.L. Munday, James Cook University; M. Grosell, RSMAS University of Miami / Marine Biology and Ecology In fishes, olfactory cues provide information about predators, prey, and conspecifics, but is crucial to understanding olfactory sensory neurons are directly exposed to the environment and are susceptible to damage from aqueous contaminants. The 2010 Gulf of Mexico oil spill overlapped with the habitat of...
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 ionization system. Both oil-exposed damselfish avoided a cosmetic chemical alarm cue, whereas oil-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. Stieglitz, 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. Schveder, RST; C. Popejoy, Fish. Oil sensitivity endpoints include accelerated metabolic rate, accelerated yolk sac depletion, and alterations of buoyancy in developing embryos. Furthermore, brief exposures during embryonic development lead to reduced swim performance, reduced maximal oxygen uptake, and reduced visual acuity in later stage juveniles. Juvenile fish exposed to oil showed 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 aerobics scope and swim 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 cardiac-myoocytes show that sarcomere shortening upon electrical stimulation is reduced by acute oil exposure which likely explains the reduced stroke volume observed in 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. Johann, 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, BioTrix; 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 aerobics scope and swim 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 cardiac-myoocytes show that sarcomere shortening upon electrical stimulation is reduced by acute oil exposure which likely explains the reduced stroke volume observed in 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).

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. Stieglitz, 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; M. Grosell, RSMAS University of Miami / Marine Biology and Ecology

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 impacts of embryo exposure to crude oil are crucial to survival, and aids in promoting larval 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, reduced 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. Shiel, G.L. Galli, University of Manchester / Faculty of Biology, Medicine and Health Sciences; G.K. Cox, J.D. Stieglitz, 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 cardiotoxic 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 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 cardiotoxic effects have been widely reported, the mechanisms underlying these effects remain understudied. 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 concentrations. 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 μg 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, 2.0, 3.0 Hz, 6.3 μg 1/50 PAH). In addition to contractility, other mechanical aspects of cell contracture function were also examined. Effects to assess the role of circulatin 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.
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

M.J. Araújo, CESAM & DeBio / APPLEEE; R.J. Rocha, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM; C. Quintanheiro, Department of Biology & CESAM - University of Aveiro; A.M. Sousa, University of Aveiro / Department of Biology & CESAM; M. Monteiro, Aveiro University / Biology

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 a larval shape to a juvenile one. 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 4MBC, Carbendazim, Linuron and Triclosan, which have potential endocrine disruptor activity. High-throughput growth, behaviour and biochemical markers were evaluated as endpoints in two periods of exposure to stressors: a first 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 4MBC, Carbendazim, Linuron and Triclosan) was performed. Our results suggest that the life span of S. senegalensis during acclimatization tests 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 differently affected, depending on S. senegalensis life stage. Significant alterations of normal behavioural pattern were observed in response to stressors exposure, confirming behaviour 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

D. Basili, University of Liverpool / Institute of Integrative Biology; M. Knobel, Eawag / UTOX; A. Sawle, Cancer Research UK Cambridge Institute / Department of Systems Biology; J. Herrbert, P. Anderson, University of Liverpool / Institute of Integrative Biology; K. Schirmer, Eawag / Environmental Toxicology; A. Cossins, F. Falciani, University of Liverpool / Institute of Integrative Biology

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 current in vitro methods provide the level of information gained from the use of a whole-life 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 (Oncorhyncus mykiss, RTgill-w1) exposed to a given chemical can be used as a bio sensor 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. Mottar, 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, 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 to assess the potential of an alternative approach so that 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 in vivo 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 scarce assumption 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


The Threshold for Toxicological Concern (TTC) is well-established for assessing human safety but has only recently been explored in the ecological context. Ecological Thresholds for Toxicological Concern (eco-TTC) summarize the
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 justification. 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 dataset 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 dataset 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., production environment; chemical risk MoA). In an integrated 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 Biobased Ecotoxicology; N. Klüber, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; O. Kerkhof, Helmholtz centre for environmental research - UFZ / Department Biobational Ecotoxicology; M. Leonard, IOREAL SA; T. Kießling, Scientific Software Solutions; R. Altenburger, UFC Centre for Environmental Research / Department Biobational Ecotoxicology; S. Scholz, Helmholtz Centre for Environmental Research / Department Biobational 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, including chemical risk MoAs. In an integrated 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 malformations 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. Cromiec, 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. Dohmen, 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 sufficient 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 for using in the context of CASA. 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.

183 Identifying ecosystem services-based protection goals.
L. Malthe, 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 are the preferences that 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.

184 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: ie ecological identity, 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 a 7% suggested there was data to support this in fact it was still a judgment, i.e. it is a hidden ‘judgement’. The suggestion that this represents a Euclidean measure of the distance between large (>35%), medium (15-35%) 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 stake holders is required it is recommended that an expert judgement qualitative approach 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. extinction probability) of species potentially affected, and the frequency of occurrence.

185 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 (NTPPs) 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 supports 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 was that of non-target plants growing in the crop field level. 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.

186 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; D. Cunn, 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 to protect specific Protection Goals (as defined for surface waters), but rather the 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, EFSA Guidance requires that a surrogate system for the Final Reference (termed the Surrogate Reference Tier) scientifically underpins the assumption that the endpoints assessed at High Tier are closer to the actual objectives of the adopted protection goal. It is accepted that Low Tier assessments do not measure the SPG directly, and therefore at High Tier, endpoint assessment should rely on ecological and ecological data that are more directly relatable to the ‘target image’ of the aquatic community. This surrogate system should be relevant for the surface water at risk, and endpoints should account for the ecological traits of the important aquatic species at risk. The EFSA PPR Panel’s solution was to identify for each Key Driver (taxonomic group or other ecological entity), a Surrogate Reference Tier that is based on the most sophisticated experimental or modelling risk assessment currently available to address the SPG. Consequently, scientists should ensure that Aquatic High Tier experimental approaches are (a) the most sophisticated approach available and (b) represent the ‘target image’ of the biodiversity aquatic communities that support the relevant ecosystem services. Perhaps in this context, the most important assessment endpoints are those that reflect functional and trait-based effects, and those that preserve the biodiversity of the different aquatic communities that ultimately present the ‘target image’ and therefore closer relevance to the Final Reference.

187 Is “biodiversity” a measurable study endpoint?
E.M. Bakker, Eurofins-CHC
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. Magurran 2004)
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 biodiversity and the development of reduction ratios. We report the results of a study of the structure and functioning caused by planting treatments. The stimulation effect on the organic matter 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 *Phialiris arundinacea*. This species cultivated in conditions of redox cycle showed to stimulate the highest increase in soil microbial activity after 3 months from planting. Moreover, the 18-month biostimulated soil was incubated with a labelled 4-chlorobiphenyl, the production of 14CO2 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 depeled PCBs from the cultivation medium according to the results of a resting-cell assay, thus representing ideal candidates to sustain PCB-remediation through a site-tailored bioaugmentation approach.

**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. Uhlik, University of Chemistry and Technology Prague; E. Zaschè, CNR-IMATI; 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 bioremediation potential of plant species in the field, different treatments for the development of a suitable rhizoremediation strategy, an experimental trial including ten vegetated treatments and their non-plant 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 assemblies. 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 *Phialiris arundinacea*. This species cultivated in conditions of redox cycle showed to stimulate the highest increase in soil microbial activity after 3 months from planting. Moreover, the 18-month biostimulated soil was incubated with a labelled 4-chlorobiphenyl, the production of 14CO2 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 depeled PCBs from the cultivation medium according to the results of a resting-cell assay, thus representing ideal candidates to sustain PCB-remediation 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, A. Di Guardo, University of Milan / DeFENS; F. Arjmand, Università La Sapienza / Chemistry; P. Ciampi, C. Esposito, Università La Sapienza / Department of Earth and Environmental Sciences; M. Carboni, P. Goria, J. Birstingl, Regenesis Ltd; S. Rossetti, B. Matturro, Water Research Institute Italian National Research Council IRSACNR; M. Bacchi, P. Foglietto, Rea Ferrovjezla Italiana S.p.A.

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 contaminant input (PCE, TCE, and cis-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™, Regenesis) 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 recent field study performed at the end of the CAHs residual concentration was 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-IRSAP; M. Daghio, A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences; M.P. Papini, Università La Sapienza / Chemistry; E. Aulenta, National Research Council / Water Research Institute (IRSA)

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 mbb/d 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 hydrocarbons is a serious problem, with nearly 50% of groundwater contamination being due to petroleum-deriving products such as mineral oil, chlorinated hydrocarbons, monoaromatics, etc., BTEX and polycyclic aromatic hydrocarbons (i.e., PAH). Accidental petroleum spills may result in severe environmental problems, hence requiring the development of quick 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 contaminated groundwater. A lab-scale prototype of the bioremediation system (“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 hydrocarbon-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 JM. Vila, Instituto de Recursos Naturales y Agrobiologia; 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 Agrobiologia / 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 recent report on unpolluted biotopes of PAH-contaminated sediments, 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 (95%) of the total PAH concentration. Low molecular weight (LMW) compounds (2-3 rings) were mostly depleted during the first month of incubation. In contrast, degradation of HMW-PAHs started thereafter following biphasic kinetics. Despite the 16S rDNA gene copy numbers (bacterial abundance) remained constant throughout the 5-month treatment period, the number of 16S rDNA gene transcripts (bacterial activity) dramatically increased (from 10^6 to 10^7 copies/g dry soil) during the initial 45 days, associated with major PAH removal. 16S rDNA gene pyrosequencing revealed distinct community profiles for the LMW and HMW 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-HM-PAH removal identified members of the recently described order *Immobidobacteriales* and members of *Sphingobium* as the main active populations. Their role on HMW-PAH removal was confirmed by DNA-SIP. Members of *Sphingobium* had major phytotypes that characterized the microbial assemblage, which contained members of *Immobidobacteriales* clearly predominated in incubations with 13C-pyreene 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 increased activity during the late incubation phase, when degradation kinetics were drastically attenuated, suggested their potential action on the residual fraction of contaminant.

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**Stable Isotope Raman Microspectroscopy and Surface-Enhanced Raman Scattering: Analysis of Microorganisms at Single Cell Level**  
N.P. Ivleva, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry; R. Weiss, M. Seidel, Technical University of Munich; R. Niessner, 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


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**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. Baciocchi, University of Rome Tor Vergata

The occurrence of aerobic biodegradation in the subsurface by ubiquitous soil microorganisms has been shown to reduce and in some cases eliminate the contaminants from 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 vsellsite characterized by the presence of BETX 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 one chamber volume with an inert gas. The measuring cycles with 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/m.

### New frontiers in Life Cycle Inventory data collection and modelling

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**The end of an era: is data and model exchange across LCA software tools finally possible?**  
M. Vieira, PRe Sustainability; K. Cenian, PRe Consultants; A. de Schryver, European Commission; A. Genest, Ifi Hamburg; L. Zampori, European Commission / Joint Research Centre; C. Wolf, Tier3 Solutions GmbH; M. Dupriez, RDC Environment; S. Horlacher, thinkstep; E. Mieras, PRe Sustainability

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 (PEFCs) 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 vetoes 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 test the EF impact assessment (IA) results of all EF-compliant secondary datasets. This way, differences coming from their implementation or of the EFIA 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 (eLCCD) 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 on how to improve the results of eLCCD format were identified. 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 eLCCD format. We believe this is a crucial step towards the exchange of data and models across software tools and the comparability of results.
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 WWTPs. 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

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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 has been developing a model for indirect LUC (LUC) 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 to specific production sectors 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 denaturalisation caused by transformation of land. The iLUC model can be combined with any life cycle impact assessment (LCIA) 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 50-30%, 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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Fertiliser water 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 shouldbe considered in LCA of products or infrastructure with long lifespan. This work aims to develop WSmix framework for modelling current assessment (LCA) model. Overall, the results show that for agricultural crops, WSmix increases the GHG emissions with 100-200%, for beef cattle 20-60%, for pigs 40-80%, for dairy products 40-60%, for wood products 50-30%, 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.

199 Poster spotlight: TU097, TU098

Behavioural Ecotoxicology: Unravelling behavioural responses to chemical contaminants in the environment

200 Do laboratory assays predict behaviour in the wild? A study with pharmaceutical pollutants

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Concern over the impact that pharmaceuticals have on wild aquatic organisms has increased over the past decade. Laboratory studies have shown that whether effects documented in the laboratory extend to the natural environment. We exposed fish to one of two pharmaceuticals (temazepam and ibesartan) commonly detected in the environment at two doses (80 ng/L, 1500 ng/L; 200 ng/L, 20000 ng/L, for temazepam and irbesartan, respectively). We then assessed how exposure affected fish behaviour in the laboratory (scototaxis to measure anxiety and activity responses) and in the field (downstream dispersal using PIT tags). We found no evidence that either pharmaceutical treatment affected behaviour in the laboratory scototaxis assay. In contrast, fish exposed to the high and low doses of temazepam dispersed faster downstream when compared to control fish. Ibesartan exposure did not affect fish behaviour in the field. Across all treatments, we also found that activity in the laboratory correlated with migration speed, indicating that fish that were more active in the laboratory also moved faster downstream in the wild. We discuss our findings in relation to differences in tissue bioconcentration for both pharmaceutical compounds in the multi-tissue fish in relation to the results emerging from measuring how pollutants affect ecologically relevant behaviours in the field alongside standard and efficient laboratory assays.

201 Exposure to the widespread androgenic steroid 17β-trenbolone alters behaviour in fish

M.G. Bertram, Monash University / Biological Sciences; M. Saaristo, J.M. Martin, T.E. Ecker, C.P. Johnstone, B.B. Wong, Monash University / School of Biological Sciences
Sciences

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 (HGP), 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 among 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 (21 day) and field-detected levels (3-year) of these hormones on the movement of zebrafish. This study was undertaken to determine the effect of diuron and 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 to fish 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 pharmaceuticals? In this research, we examined how shared environmental stress affected the exploratory of a novel environment into shy and bold individuals, and subsequently exposed during 9th to carbamazepine, a human pharmaceutical, suggested as a marker of anthropogenic pollution. Assessed responses included behaviour (distance swan, position in the tank and time spent swimming) and biochemical markers associated with oxidative stress, neurotransmission and energy metabolism. Overall, our results showed significant differences between control shy and bold organism with behaviour endpoints demonstrating to be very sensitive to stressor conditions. Although carbamazepine alone did not show considerable effects in the assessed endpoints, strong interactions were found between personality and pharmaceuticals, supporting further studies.

205 Effects of fluoxetine on anxiety-related behaviours and physiology in a songbird S.E. Whitlock, Environment Department, University of York / Environment; R. Shore, Centre for Ecology & Hydrology (NERC); J. Lane, Animal and Plant Health Agency; K. Herbom, Newcastle University / Centre for Behaviour and Evolution; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; K. Arnold, University of York / Environment

Several species of bird are known to forage directly on invertebrates at wastewater treatment plants (WWTs), representing an exposure route to a range of contaminants including pharmaceuticals. The selective serotonin reuptake inhibitor (SSRI) fluoxetine is commonly prescribed for anxiety, but could induce indirect effects on target species. To address these concerns, a study was undertaken to investigate the extent to which exposure to 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’ 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, 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 affords both feeding efficacy 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 in natural settings. 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.

203 Environmental levels of anxiolytic pharmaceuticals alter migration of Atlantic salmon in both lab and field T.J. Ormanni, Istrea / EABX-CARMA; C.N. Doose, INRS - Centre Eau Terre Environnement; B. CHAUMET, Istrea; N. Mazella, Istrea Bordeaux / UR EABX; N. Majdi, Ecolab / UMR 5245 CNRS; J. Vivdrenne, S. Morin, Istrea Bordeaux / UR EABX; W. Traumspurger, 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 fish. Further, when faced with a novel environment, exposed fish were again more active and exploratory, and spent less time associating with a shoal 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, two diatom species, Gomphonema gracile (two different morphotype: normal (GG) and teratogen (GT)) and Planothidium lanceolatum (PL), and one green alga Pseudokirchneriella supcapitata (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 Gomphonema gracile with teratogen shape and Pseudokirchneriella supcapitata. 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 these experiments, 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.
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 prevent 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 untargeted 600 mostly industrial substances, including many detergent ingredients such as benzotriaゾロール, 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 ¾ of the sampling sites with multi-year samplings. In this study, we analyzed the potential cumulative effects of mixture exceedances of the PNEC (Predicted No Effect Concentrations) of the EEA. We found that exceedances of more than 600 mostly industrial substances, including many detergent ingredients such as benzotriaゾロール, 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 ¾ of the sampling sites with multi-year samplings. For that purpose, we analysed two data sets on macro invertebrate communities where we have a) similar 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

L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; D. De Zwart, DiDZ Ecotoc / Centre for Sustainability Environment and Health; J. Postma, Ecofide; M.C. Zijp, RIVM / Centre for Sustainability, Environment and Health

“Big data” are a potential goldmine for studying and contextualising chemical and environmental risk assessments, as they enable relating predicted risks to observed impacts. This can confirm or challenge our chemical risk assessments, by checking whether Environmental Quality Standards provide sufficient safety, and whether or not mixture exposures cause problems not captured in the widely used ‘single-chemical plus safety factor’ approaches. Digging the goldmine asks for – first – data collation, and then: a powerful design which can be borrowed from epidemiology. This presentation is a recent gold-digging trial, with surprising results. We collated biomonitoring and mixture exposure data from the Dutch Waterboards, as collected due to obligations of regulatory frameworks (such as the Water Framework Directive). We quantified for each sample, based on measured chemical concentrations, the mixture toxic pressure at EC50-level. Outcomes were expressed as multi-substance Potentially Affected Fraction (mPAF-EC50) of specific taxa (Species Sensitivity Distribution; SSD). Early research suggested that this proxy – higher values of which are interpreted to imply a higher potential for species loss – has a gross absolute relationship with species loss in various regional studies. In the current study, we overlaid the SSD-model basis (all species are unequal in their sensitivity to exposures) with this finding, by analyzing taxa-specific threshold values. That is, we determined the taxa-specific mPAF-EC50 beyond which species abundance starts changing when toxic pressure rises, in a downwards (sensitive) or upwards (indirect opportunistic response) direction. The results show a series of species-specific mixture exposure thresholds, and an overall assemblage-level (aggregated) exposure threshold. These outcomes are compared to simpler approaches, such as quantile regression on the species assemblage level, as well as multi-stressor statistics. We conclude that the set of species-specific and assemblage-level thresholds bear important contextual information to judge our risk assessment and management foundations, be it in the realm of prospective Chemical risk assessment or of the retrospective Environmental risk assessment and management.
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 population decline. 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 pollutants which can explain body growth 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 records 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. Hjorup, Swedish Environment Monitoring; G. Malarvannan, 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 observed in the past, 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 as stable carbon and nitrogen isotope compositions. 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 from the east coast of the United States and Canada. The divalent Ni free ion in artificial seawater (ASW) containing Ni(II) in solution was quantified using Ion Exchange Technique (IET) with Ni measured by Graphite Furnace Atomic Absorption (GFAA). The measured Ni⁹⁸ values were compared with model predictions (i.e. Visual Mintex) for evaluating the feasibility and applicability of the IET method for Ni in seawater. For the most part IET-measured [Ni⁹⁸] agreed very closely with model predictions in the defined solutions, 96-hour Ni embryo toxicity tests were performed for a sea urchin (S. purpuratus). The dose response curves were expressed both as total dissolved Ni concentration ([Ni₃]) and free Ni concentrations from IET ([Ni⁹⁸]). If the Ni toxicity is explained by [Ni⁹⁸], all the toxicity response curves of different model ligands will overlap and this was not observed for the majority of samples. The results of this research contribute to the development of bioavailability-based prediction models for estimating the impacts of Ni in marine water. Funding was provided by Natural Sciences and Engineering Research Council of Canada (NSERC), Vale Canada and NIPERA Inc.

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 these observations and is key to further improving approaches for nickel in seawater. For example, the SETAC “Summary document” and a series of manuscripts to be submitted to a SETAC journal.

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

C.E. Schlegel, NIPERA; K. Gallagher, U.S. EPA / Office of Water; B.A. Stubbsfield, 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 current and future water quality criteria. The ability 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 incorporation of metal bioavailability information into global regulatory frameworks. Approximately 40 invited scientists representing industry, government, and academia participated in the workshop. Participants were divided into five groups with each group 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/or other alternative approaches for nickel i...
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 full effects on acute Ni toxicity to crustaceans very well, an “average crustacean” model was developed. The parameters for the 3 crustacean models were then 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, 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. Hasson, Mines ParisTech / Hydrodynamics and reactions team (HR); M. Leemakers, Vrije universiteit Brussel / Department Analytical, Environmental and Geochemistry; M. Descotes, AREVA Mines / R&D; V. Lagneau, Mines ParisTech PSL Research University / Geology, Geophysics and health of the Earth (HR); Environmental regulations are becoming more and more ecosystems-oriented. In this respect, the habitat’s faunistic biodiversity is targeted, meaning the contaminant’s bioavailability is taken into account. The latter depends on the geochemical conditions of the aquatic system, which define the chemical speciation. For several elements, Environmental Quality Standards (EQS) and Sediment Quality Standards (SQS) 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, 10% Kaolin/90% Q, 10% Smeectite/90% Q, 10% Ferrihydrite (FOH)/90% Q) mixed with a mixture of the 4 mineral phases of 3.3% Kaolin/smeectite (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 concentration 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 (Kow) range from 10 L/Kg for quartz to 60000 L/Kg for FOH. After ten days exposure, the highest Biota Sediment Quotient (BSQ) for FOH (10%) was found for the chironomid larvae. After ten days exposure, the highest Biota Sediment Quotient (BSQ) for FOH (10%) was found for the chironomid larvae. After ten days exposure, the highest Biota Sediment Quotient (BSQ) for FOH (10%) was found for the chironomid larvae. After ten days exposure, the highest Biota Sediment Quotient (BSQ) for FOH (10%) was found for the chironomid larvae.

216 Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (I) P. Bauerlein, KWR / Analytical and Environmental Chemistry; A. Hintelmann, KWR Watercycle Research Institute / Chemical Water Quality and Health; S. Mentenig, Utrecht University / Copernicus Institute of Sustainable Development; A. Koelmans, Wageningen University / Aquatic Ecology and Water Quality Management; H.M. Ohlendorf, CH2M

Microplastics are a broad size fraction, that reveals information on the polymer types, are detectable using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (I) P. Bauerlein, KWR / Analytical and Environmental Chemistry; A. Hintelmann, KWR Watercycle Research Institute / Chemical Water Quality and Health; S. Mentenig, Utrecht University / Copernicus Institute of Sustainable Development; A. Koelmans, Wageningen University / Aquatic Ecology and Water Quality Management; H.M. Ohlendorf, CH2M

Microplastics are a broad size fraction, that reveals information on the polymer types, are detectable using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For microplastics field- flow separation, that reveals information on the polymer 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 nanoparticles. 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 aqueous samples. Finally, we propose an approach to estimate polymer masses based on the two-dimensional microwave method and compare the results to those obtained by treating waste water using TED and GC-MS.

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Trace particulate plastic analysis in environmental systems: synthesis and utilisation of microplastics and microplastic fibres and particles in urban wastewater system 

F. Schmidt, M. Schmiederer, Eawag Swiss federal Institute of Aquatic Science and Technology; D.M. Mitrano, 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 analyses, even at the bench scale, have 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 synthesise 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 in a river for the first time and Fenton’s reagent was found to increase the 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 95% recovery in every matrix. Furthermore, the particulate plastics synthesized were spiked into mixed liquor in 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 quantitative plastic particles are a well-defined study family that could potentially interact 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 and in trace concentrations have matured.

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Detection of polymers in treated waste water using TED-GC-MS

C. Geodecke, K. Altman, Bundesanstalt für Materialforschung und prüfung; C. Bannick, Umweltbundesamt; E. Koher, Technische Universität Berlin; M. Ricking, UBA Umweltbundesamt; T. Schmitt, Berliner Wasserbetriebe; U. Braun, BAM-Forschungsinstitut für Bauforschung und Testing / 5.3 Mechanics of Polymers. The presence of large quantities of plastic waste and its fragmentation in various environmental compartments are an important subject of current research. In the environment, (photo-) oxidation processes and mechanical abrasion lead to the formation of microplastics. However, until now, there are no established quality assurance concepts for the analysis of microplastic (< 5 mm) in environmental compartments, including sampling, processing and analysis [1-4]. The aim of the present work is the development of suitable examination methods and protocols (sampling, sample preparation and detection) to qualify and quantify microplastic in urban water management systems. At first a fractional filtration system for sampling and the analytical tool, the so-called TED-GC-MS (thermal desorption gas chromatography mass spectrometry) was developed. This method is a two-step analytical procedure which consists of a thermal extraction where the sample is annealed and characteristic decomposition 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 treatment 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, PS and PP were detected in the effluent, and PE and PS were found in the raw waste water of the sewage treatment plant in Ruhleen, 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

R. Hurley, NIVA - Norwegian Institute for Water Research; A. Lusher, NIVA Norwegian Institute of Water Research / Marine and Freshwater Research Centre; M. Olsen, Telemark University College; L. Nizzetto, NIVA The very little existing work on the analysis of 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 target polymers; 2. Efficacy of reagents in reducing organic matter content in soil and sludge and 3. Extractions of microplastics in environmental 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 material. 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 time and cost-effective approach is promising for the detection of nanoplastics. The pre-treatments were not effective in sufficiently reducing organic material whilst peroxide oxidation and Fenton’s reagent removed the majority of organic material. 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 time and cost-effective approach is promising for the detection of nanoplastics. The pre-treatments were not effective in sufficiently reducing organic material whilst peroxide oxidation and Fenton’s reagent removed the majority of organic material. 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 time and cost-effective approach is promising for the detection of nanoplastics. The pre-treatments were not effective in sufficiently reducing organic material whilst peroxide oxidation and Fenton’s reagent removed the majority of organic material. 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 time and cost-effective approach is promising for the detection of nanoplastics. The pre-treatments were not effective in sufficiently reducing organic material whilst peroxide oxidation and Fenton’s reagent removed the majority of organic material.
processes may cause changes in the density and shape of individual plastic particles, or aggregates. This can be a driving factor for the ultimate fate of microplastic, as it could cause floating microplastic to sink or be suspended below the water surface. However, the factors controlling the buoyancy or sinking velocity of different microplastic are not as well-known as they are for other particles/particles, like phytoplankton and sedimentary material. Herein we present the results of linking experiments on microplastic, covering different shapes (spheres, fibres and irregular), microplastics in lakes and seas, concentrating on water properties, considering a range of ambient conditions (temperature, salinity and turbulence). The microplastic were compared before and after exposure to weathering processes in the lab and outdoors. The results obtained in this sinking experiment were compared to theoretical expectations, based on literature experiments that describe the relationship between the drag coefficient and particle Reynolds number. A key advantage of this relationship is that it is independent on the type of plastic and properties of water, and would therefore apply to both freshwater and saline waters alike, as it would to all types of microplastic.

Air Pollution, Biomonitoring and Human Health (I)

224 PARTICULATE MATTER IN INDOOR ACADEMIC ENVIRONMENTS: CHEMICAL COMPOSITION, SOURCES, INFILTRATION FROM OUTDOOR L. Totufl, CNR / Institute of Atmospheric Pollution Research; S. Canepari, Sapienza University of Rome / Chemistry; M. Catrambone, F. Marcovecchio, M. Giusto, CNR / Institute of Atmospheric Pollution Research; S. Pareti, CNR / Institute of atmospheric pollution reasearch; T. Sargolini, CNR / Institute of Atmospheric Pollution Research; C. Perrino, CNR Institute of Atmospheric Pollution Research / Institute of Atmospheric Pollution Research

We present the results of the first part of an experimental study carried out in an number of academic environments, ranging from small laboratories to very wide classrooms. The study was aimed to evaluate the mass concentration and the chemical composition of indoor atmospheric particulate matter (PM10 and PM2.5) and its relationship with a number of parameters. These include: concentration and chemical composition of outdoor particles, mixing properties of the lower outdoor atmosphere, volume and floor of the classroom, distance from the street, presence/absence of the students, season. Two type of sampling schedules were applied. The first one differentiates among working days, nights and week-ends during a 6-week winter period and a 4-week summer period (Special Observation Periods – SOPs). This schedule was planned to highlight the differences due to the presence of the students and teachers. The second one (Long-Term Sampling) consists in twelve 1-month PM10 samplings carried out by using very-low flow-rate samplers. It was envisaged to obtain a general picture of the effect of the above parameters during a whole calendar year. Both sampling schedules were applied to six indoor and four outdoor sites, all inside or around the same building. At each site and for both schedules the sampling were simultaneously carried out on Teflon, quartz and polycarbonate filters. Teflon filters were used for the determination of the collected mass (by gravimetry), of the elemental content (total content by energy dispersion X-ray fluorescence, bioavailable and residual fractions by inductively coupled plasma mass spectrometry), of anions and cations (by ion chromatography), of elemental and organic carbon (by thermo-optical analysis) and of the bioaerosol content (by propidium iodide staining and epifluorescence microscopy). We report here the results obtained during the winter SOP and during the winter part of the Long-Term Sampling. The composition of PM in the indoor environments was dominated by the organic fraction, with a relevant contribution of the bioaerosol, mainly in the coarse fraction. The infiltration of particles from outside constituted a significant source of inorganic species. A vertical gradient was observed for soil components. A relationship of the concentration and composition of indoor PM with the volume of the classroom, height from the ground, presence/absence of the students and distance from the street has been highlighted.

225 SOURCE APPORTIONMENT OF MAJOR SPECIES AND METALS IN PM2.5 IN URBAN SITES UNDER INDUSTRIAL INFLUENCES IN NORTHERN FRANCE F. Ledoux, University of Littoral Côte d’Opale / Unité de Chimie Environnementale et Interactions sur le Vivant UCEIV EA4492; A. Kfoury, University of Baliamond / Department of Environmental Sciences; G. Delmaire, University of Littoral Côte d’Opale / Laboratoire Informatique Signal de la Côte d’Opale LISIC EA4491; G. Roussed, University of Littoral Côte d’Opale / Laboratoire Informatique Signal de la Côte d’Opale LISIC EA4491; D. Courcot, Université du Littoral Côte d’Opale / Unité de Chimie Environnementale et Interactions sur le Vivant UCEIV EA4492 PM2.5 have been related to various adverse health effects, mainly due to their ability to penetrate deeply and to convey harmful chemical components inside the body. The North of France is one the most densely populated area in Europe and is known as an industrialized region especially in the field of metallurgy, organic chemistry, and glassmaking. Furthermore, its strategic position in the heart of Europe means that this area is subject to major transportation activities by road and also by sea. In this context, the objective of this work was to acquire a better knowledge on the exposure level to major species and metals in PM2.5 and on the identification of sources in urban sites influenced by particulate emissions from anthropogenic sources. Sampling was performed using Digital® 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 several volatile compounds. In this work, we report the results obtained in Dunkerque, respectively 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). NO2, SO2, NH3, and TC were found as the major contributors of PM2.5 (between 95% and 99%) for the indoor and outdoor differences were evidenced. Trace elements (Cr, Zn, Ni, As, Ag, Cd, Cu, Mn, Pb, V, Sn, Rh, Sr, Bi, Ba, Co, Sb and Ti) 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 considered as major 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.

226 ESTIMATING THE CONTRIBUTION OF DEPOSITION IN THE TOTAL EXPOSURE TO PAH’S IN ORDER TO DERIVE SAFE DEPOSITION REFERENCE VALUES J. Bierens, VITO / Sustainable Health; L. Geerts, M. Van Holderbeke, VITO NV; K. De Brouwere, VITO NV / Health; A. Standaert, VITO; C. Cornelis, VITO / Environmental Risk and Health; T. Fiorenzo, VITO

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 safe deposition reference values. To this end, the fate and human exposure was modelled using the MERLIN-Exp, 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 exposure to PAHs via crops was modelled using an 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. Safe 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 Br/AI 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-DISRUPTIVE AIR MUCOSAL HORMONES S. Labroire, INRA / EPHE, EPHE; E. Moreau-Guigon, EPHE, PSL / UMR METIS; F. Alliot, EPHE / UMR Metis; M. Bimbout, Univ. Paris-Sud / UMR ESE; A. Desportes, EPHE / UMR METIS; V. Huteau, Univ. Paris-Sud / UMR ESE; M. Chevreuil, EPHE / UMR METIS 7619; L. Oziol, University of Paris-Sud / UMR CRRS 8079 Air quality is currently assessed by monitoring a few pollutants involved in the formation of secondary pollutants. However, other pollutants may be of concern, 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
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 presenting a higher endocrine-disrupting effect, especially estrogenic. In order to identify the bioactive compound families responsible for this endocrine-disrupting 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 (winter 2014) and in sufficient quantities (6 consecutive samplings of 15 days) to implement the entire fractionation process by semi-preparative liquid chromatography: split of the initial organic 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 a previously developed rapid screening bioassay for the selected target EDCs in the biological samples (concentration level, intrinsic estrogenicity, lipophilicity), major families found indoors (phthalates, synthetic musks, alklyphenols, 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
H. Petruccione, University of Pittsburgh / Civil & Environmental Engineering; W. Collinge, M. Bilec, University of Pittsburgh / Civil and Environmental Engineering
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, inversely, envelope improvements (airtightness) can increase infiltration and air exchange rates, and magnify the effects and intensity of internal air pollutants. In this context, a pilot study was conducted to evaluate the 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 represented the energy and conservation target district, which is situated downwind of major industrial point sources. Indoor and ambient measurements of particulate matter (PM10, PM2.5), black carbon, ozone (O3), total volatile organic compounds (TVOC), carbon monoxide, carbon dioxide (CO2), relative humidity, and formaldehyde (HCHO) were collected on a seasonal basis. The heterogeneity in ventilation type along with the negative effects of deficient ventilation and aged mechanical systems had an impact on indoor air quality that was 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 CO2 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., PM10, O3, NO2) before increasing outdoor air volume. Natural ventilation systems that 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 complex interplay between VOC 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
L. Massimi, Sapienza University of Rome / Environmental Biology; c. perrino, CNR Institute of Atmospheric Pollution Research / Institute of Atmospheric Pollution Research; M. Costi, Sapienza University of Rome; S. Canepari, Sapienza University of Rome / Chemistry
A new very-low volume sampler has been developed with the purpose of allowing spatially-resolved determination of atmospheric particulate matter (PM) and 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 validated during a one-year study focused on the concentration of PM2.5 mass, ions, levogluocose, 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 PM2.5 mass concentration and its main chemical components in the area of Terni, a urban-industrial hot-spot situated 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 biomonitor 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
P. Crihuet, ENTEF, IRSTEY LION; A. Devaux, INRA-CNRS / UMR LEHNA USC INRA IGH ENTEF; K. Abbaci, H. QUEAU, N. Delorome, L. Gamero, IRSTEY LION / UR MALY Laboratoire Ecotoxicologie; S. BONY, INRA - CNRS / UMR LEHNA USC INRA IGH ENTEF; a. chaumot, IRSTEY / UR MALY Laboratoire Ecotoxicologie
Since the 80s, the development of molecular biomarkers is an important component of ecotoxicology. Unfortunately, field studies that univocally 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 production. 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 revealed in adult gammarids exposed to a chemical stress could be predictive of the fitness of their progeny (i.e. transgenerational effects). For this, the consequence of an exposure in the lab of genitors to environmentally relevant concentrations of cadmium were evaluated in F1 and F2 individuals reared in unconditioned 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 was currently in progress. References (1) Forbes VE, Calow P, Sibly 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 cells 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
Q. Fu, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry; C. Vignet, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; D. Fedrizzi, Eawag Swiss federal Institute of Aquatic Science and Technology; N. Cederqvist, University of Copenhagen / Department of Plant and Environmental Sciences; J. Hollander, Eawag / Environmental Chemistry
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 strobilurins 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 azoxystrobin and an azole fungicide prochloraz in the presence and absence of prochloraz, the inhibition strength (IC50), PRZ, AZ 50, β values of azoxystrobin in the presence and absence of prochloraz, the inhibition strength (IC50) of prochloraz, and its effect on the locomotory behavior of the two species. Bioaccumulation of azoxystrobin were similar in both species with the 80s, the development of micropollutants 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 strobilurins 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 azoxystrobin and an azole fungicide prochloraz in two aquatic invertebrate species Gammarus pulex and Hyalella azteca. Furthermore, we explored median lethal concentrations (LC50) of azoxystrobin in the presence and absence of prochloraz, the inhibition strength (IC50) of prochloraz, and its effect on the locomotory behavior of the two species. Bioaccumulation of azoxystrobin were similar in both species with bioaccumulation factors (BAFs) approximately 5 L kg-1, while bioaccumulation of prochloraz was different in two species, with BAFs 57 and 110 L kg-1 in G. pulex and H. azteca, respectively. Many biotransformation products were found for both fungicides in both species, which are identified as prochloraz, prochloraz conjugates and azoxystrobin conjugates in the same ways we described above. Most BTPs result from oxidation and conjugation reactions, which occurred at the (E)-methyl β-hydroxy group of azoxystrobin and imidazole ring of prochloraz. Prochloraz inhibited the CYP-catalyzed biotransformation of azoxystrobin in both species, leading to higher internal azoxystrobin concentrations and suggesting synergistic effects. The half maximal inhibition concentration of prochloraz IC50 of IC50 was 0.1 and 0.02 µM 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. Aramburozú, Iriarte Lyon / Freshwater system, Ecology and Pollution Research Unit, UAM; R. M. CALADO, Laboratoire Ecotoxicologie; N. Delorme, K. Abbaci, Iriarte Lyon / UR MALY Laboratoire Ecotoxicologie; P. NOURY, Iriarte Lyon / Ecotoxicology; R. Dutinand, Iriarte 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, MNHN / 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 European 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 the efficacy of pesticides and increase resistance. However, it is not known 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 considered acute effects showed almost 3-fold higher tolerance to the neonicotinoid insecticide clothianidin (mean EC50 = 218 µg L−1) compared with non-exposed populations (mean EC50 = 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 approaches in marine invertebrates 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, Smallmatek - Small Materials and Technologies, Lda.; R. Martins, Department of Biology, University of Aveiro / Department of Biology and CESAM; J. Tedin, University of Aveiro / Department of Materials and Ceramic Engineering; E. CECQ; 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 inhibition of a new antifouling approach, DCOIT encapsulated in silica nanocounters (SiNC@DCOIT), toward the non-target species Sarcophyton cf. glaucum, a coral that is also a model of the crinidarian-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 monocoralonal coral fragments (n=5) for seven days, at two different temperatures (present day conditions—26°C— and forecasted scenario for 2100−30.5 ºC), 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 frond fractions) being determined by mass spectrometry and catabolic activity of catalase (CAT), glutathione-S-transferase (GST) and lipid peroxidation (LPO). Results revealed a decrease on Fv/Fm values at 30.5 ºC, when compared to 26 ºC (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 ºC, whereas at 30.5 ºC they were closed. Regarding enzymatic activities, significant increase on the GST of both animal and microalgae fractions was found in 30.5 ºC groups. On the controls, the raise of 4.5 ºC 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. Robinson1, Centre for Ecology & Hydrology; E. Lahive, Centre for Ecology and Hydrology; S. Short, P. Kille, Cardiff University; D. Spurgeon, Centre for Ecology & Hydrology. A. Robinson1, (alerob@c.eh.ac.uk), S. Short, E. Lahive1, P. Kille, D. Spurgeon1 Centre for Ecology and Hydrology, Maclean Building, Benson Lane, Wallingford OX10 8BB, UK 1 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 reproduction values of imidacloprid was observed across 5 species of earthworms (Eisenia fetida, Lumbricus rubellus, Dendrobaena octaedra, Apporectodea caliginosa and Aminthys gracilis) with A. gracilis being the most sensitive and L. rubella 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 biochemical 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) 236 Multiple exposure to pesticides and other emerging pollutants – problems and solutions for healthy ecosystems and humans 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. They also support a wide variety of aquatic species. In the past decades Greenpeace did several investigations on persistent chemicals like pesticides and industrial chemicals polluting waterbodies. Producing our food within an agricultural system highly dependent on synthetic-chemical pesticides doesn‘t come without consequences. The impacts of industrial agriculture like Apple and fruit production are widespread, ranging from contaminated soil and water, to impacts on bees and other beneficial insects, as well as on farmers, their families and consumers. Starting in 2011 investigations in the
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 unexpected survey in remote 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 PFAS 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 regulations 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 (the 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] http://gain.fas.usda.gov/Recent%20Gain%20Publications/Fresh%20Deciduous%20Fruit%20Annual_Vienna_EU_27-10-28-2011.pdf [1] http://www.greenpeace.org/italian/Global/italian/publications/toxics/Water%202011/01-dirty-laundry-12-pages.pdf [1] https://www.greenpeace.de/sites/www.greenpeace.de/files/20121203-Toxic-Threa ts-China-engl.pdf [1] http://de.toxipedia.de/assets/uploads/Report%20AE/RAE_report_08_2015_en.html [1] http://www.greenpeace.it/InquinamentoPFAS-in-Veneto.pdf [1] http://www.greenpeace.org/italia/global/italia/report/2017/InquinamentoPFAS-in-Veneto.pdf [1] Non ce la beviamo. Presenza di PFAS nell’acqua delle scuole venete (in italiano) [1] http://www.greenpeace.org/italia/global/italia/report/2017/InquinamentoPFAS/in-veneto.pdf 237 Benefits of international Science & Policy cooperation to promote a paradigm shift in water quality and safety assessment framework A. G. W. M. van den Bergh, Wageningen University / Environmental Research / Environment and Health; S. Rinck-Pfeiffer, Global Water Research Coalition; B. Escher, Helmholtz Centre for Environmental Research Gmb – 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. Dingermans, 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 biosoas, which are increasingly being applied in water quality assessment, provide relevant and robust predictive biosysstems 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 a complete chemical structure analysis alongside a complete 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 micropollutants. 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 Tool; PF7 DEMEAU, PF7 Solutions, BRA: Toxicology). These innovation could contribute to strengthening 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 precautionary tools regulations (Australia, US CA), 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 tools could be utilized by the US EPA and to Interface to 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 paths, 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 framework to achieve good ecological status of surface waters. However, in order to achieve this goal, further legal provisions need to be continuously developed and supplemented in order to reflect new knowledge and best available technology regarding micro-pollutants. This also includes more holistic approaches for the assessment and monitoring of chemicals. The review of the Water Framework Directive can provide a suitable window of 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. Alyagkas, 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 opportunities to discover new contaminants and 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 NORMAN Digital Sample Freezing Platform (http://norman-network.com/?q=node/236) and NORMAN Digital Freezing Platform (http://norman-data.ea.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- unidentified 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 chemistry that is unthinkable only a few years ago. Note: This abstract does not reflect US EPA policy. 240 Toxicological profiling of water samples with in vitro bioassays and assessment using effect-based values B. Escher, Helmholtz Centre for Environmental Research Gmb – UFZ / Cell Toxicology; R. Altenburger, UFZ Centre for Environmental Research / Department Bioanalytical Ecotoxicology; S. Alt-Aissa, 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. Brison, INERIS / Ecotoxicology Unit; S. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology; W. Busch, Helmholtz Centre for Environmental Research - UFZ Gmb / Bioanalytical Ecotoxicology; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research,
Dept. of Environmental Analysis; T. Hamers, VU University Amsterdam, Institute for Environmental Studies (IVM) / Department of Environment and Health; K. Hettwer, new diagnostics GmbH; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; H. Hollett, RWTH Aachen University / Institute for Environmental Research; R. Kase, Swiss Centre for Applied Ecotoxicology Eawag-EPFL; C. Karlsson, Ecotoxic Centre Eawag-EPFL; J. Legradi, Vrije Universiteit Amsterdam; J. Tuerk, IUTA, Institute of Energy and Environmental Technology; R. van der Oost, Waternet / Onderzoek en Advies; E. Vermeirssen, Ecotox Centre Eawag-EPFL / Aquatic Ecotoxicology; P.A. Neale, Griffith University / School of Environment

In vitro bioassays including cell-based bioassays and low-complexity whole-organism assays have been applied for decades in water quality monitoring. However, there is no common understanding what level or response is acceptable.

As of now, bioassay results were only benchmarked against each other but not against an absolute measure of chemical water quality. The EU environmental quality standards (EQS) differentiate between poor and acceptable surface water concentrations for individual chemicals of concern but cannot capture the thousands of chemicals that are in water and their biological action as mixtures. We developed 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 bioassays 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 bioanalytical 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 untreated wastewater) and wastewater treatment plant effluent in 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 bioassays 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 bioassays commonly applied for water quality assessment.

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241 Chemical gene interactions for associating contaminants with biological effects

A. Schroeder, University of Minnesota-Crookston / Math, Science and Technology; D. Martinovic-Weigelt, University of St. Thomas / Biology; G.T. Ankley, D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory

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 establish 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 provided preliminary data about chemical effects to develop a KAM for detected chemicals at five locations near two WWTPs. Hepatic transcriptome 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; B. Harvey, Syngenta; B. Miles, BASF SE / Crop Protection, Environmental Fate Modelling; D. Liss, SGS Institut Fresenius GmbH / Agro

1H-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. The development of 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 1107/2009, 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 expanded the 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

M. Blank, Bayer AG Crop Science Division / Environmental Safety; H. Borchers, A. Changle, Bayer Crop Science AG; H. Telscher, Bayer AG Division CropScience/Environmental Fate / Development Environmental Safety; O. Heinemann, Bayer AG Crop Science Division 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 detected in all forest top soils. The study discovered that 124T occurs ubiquitously in the environment innovative approaches were needed. The TDMG scientists discovered that 124T occurs ubiquitously in the environment, originating also from other 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 nitrication 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. The aim was to evaluate the risk from triazole fungicides only, the challenge was to design studies to sample groundwater draining in areas with intensive triazole fungicide usage, 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, with in-depth 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 intensive 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.

245 Leaching of 1,2,4-triazole through agricultural fields in Denmark
A.E. Rosenbom, N. Badawi, Geological Survey of Denmark and Greenland / Geochemical; P. Olsen, Aarhus University AU / Department of Agroecology - Soil Physics and Hydropedology; S. Marcher, The Danish Environmental Protection Agency / Agroconsulting; The Danish Environmental Protection Agency / Pesticides and Gentechology

The compound 1,2,4-triazole is a degradation product of many azole-fungicides and growth regulators used in agriculture. Leaching of 1,2,4-triazole from agricultural fields has been evaluated in Denmark in the Danish Pesticide Leaching Assessment Programme (PLAP; www.pesticidvarsling.dk), which comprise five agricultural fields and in additional control plots. The soil specimens were analyse collected at 15 sampling times from 0 to 360 days after application, in triplicates and in additional control plots. The soil specimens were analysed for residues of 13C-TBZ, unlabelled TBZ (12C-TBZ), 13C labelled 124T (13C-124T), and unlabelled 124T (12C-124T). Residues of 13C-TBZ remained in the top-soil segments. There were no detects of 12C-TBZ in any of the investigated samples. 12C-124T as the degradation product of 13C-TBZ could be detected in all six trial sites in varying concentrations. Of special note, 12C-124T was detected in four of the six trials, even though the use of triazole fungicides on the trial plots could be excluded for a minimum of three years before the application. At one trial site, residues of 12C-124T were detected down to a depth of 100 cm in all plots with a maximum concentration of 1.17 g/L. The data collected in this study confirm that in many cases the origin of 124T findings is not the use of azole fungicides, but other sources. 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.

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. Furthermore, TFAA has been identified as a biodegradation product of several pesticides. During a screening of surface waters in south-western 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. Ozoneation of model substances and wastewater samples was applied to reveal the TFA-forming potential of individual compounds and the presence of non yet identified TFA-precurcursors in waters bodies in general. Discharge of industrial wastewater was identified as the source of elevated concentrations of TFA in the Rhine tributary. Extended monitoring demonstrated that this contributor still impairs the drinking water supply along the lower River Rhine. Ozoneation, 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 inceasing 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 be 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
L. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiología / Agrociencia y Conservación del Suelo

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
overview contribution will examine the range of techniques and experimental models suitable for the assessment of HOCs biodegradability taking into account state-of-the-art bioavailability science (Environ. Sci. Technol. 49:10255-10264, 2015). During recent years, we have applied these techniques to study the microbial interconnections with bioavailability processes, involving pollutant phase exchange, microbial mobilization and cell attachment to interfaces. We can consider two groups of techniques; 1) Broadly applied methods to estimate the bioavailable contaminants using Tanox or passive sampling, methods also subject of standardization and 2) specific methods suitable to deeply characterize phase exchange with liquid mixtures and 14C-labelled chemicals. Examples and applications of these approaches will be summarized. They include desorption extraction (Environ. Sci. Technol. 45:3019-3026, 2011; Environ. Sci. Technol. 48:10689-10877, 2014), passive sampling and dosing methods (Environ. Toxicol. Chem. 27:1526-1532, 2008; Environ. Pollut. 184:435-442, 2014; Environ. Pollut. 205:378-384, 2015), constant NAPL/water interfacial area method (Environ. Sci. Technol. 45:10744-1081, 2011; Environ. Sci. Technol. 51:11935–11942, 2017), and radiorepisirometry and dual 14C/Creseide analysis (Environ. Pollut. 159:3692-3699, 2011). In spite of these advancements, significant gaps of knowledge exist between bioavailability and biodegradation sciences. Still today, it is difficult to predict bioavailability of HOCs, for example, solely on the basis of basic parameters such as organic matter, black carbon or clay contents of a given soil or sediment, and the physicochemical constants of the chemicals (such as solubility in water, octanol-water or organic-carbon based distribution coefficients). This limitation even remains with improved assessments through determinations of chemical activity and bioaccessibility. This uncertainty not only applies to biodegradability in natural environments, but also to engineered remediation systems.

249 Strategy for ready biodegradability evaluation of poorly water-soluble organic compounds in aqueous media
C. Khodadadi, LOREAL SA / Research and Innovation; J. Chenible, Loreal Research / Research and Innovation; Y. Barthel, Eurofins Expertises Environnementales / Eurofins Expertises Environnementales; J. Lharidon, LOréal Research & Innovation / Life Sciences Direction; T. Gerald, University of Nantes / Microbiology
The assessment of the environmental impacts of an environmental substance is based on ready biodegradability tests, demonstrating a rapid biodegradation in the most environmental media. However, when these tests are applied to poorly water-soluble substances, difficulties are encountered, often related to their limited bioavailability towards the microorganisms inducing increased variability that we have studied. An innovative strategy has therefore been established in order to improve the assessment of biodegradation of these substances. It has compared 24 methods of improving bioavailability methods (BIM) and initiated the revision of the international standard ISO 10634.

250 Impact of temperature on micropollutants removal in an activated sludge system
P. Meyang, Newcastle University / CEGS; R.J. Davenport, Newcastle University / School of Engineering; K. Fenner, ETH Zürich/Eawag
The investigation of the environmental fate of pollutants is essential for evaluating their ecological impact and human exposure, and is a priority for the European water framework. In particular, the high variability of micropollutants removal efficiency in biological treatment systems hampers our understanding of how plant performances are affected by operational and environmental parameters, such as temperature fluctuations (e.g. daily and seasonal). Currently, environmental exposure assessment uses Athrinus-based models to estimate biotransformation rates at different temperatures, despite they neglect potential compositional and functional variation of the microbial community. This work aims to evaluate the variability of such models, by exploring the effect of short-term temperature variation on micropollutant biotransformation in an aerobic sludge community. Laboratory batch reactors were seeded with activated sludge from a Swiss full-scale treatment plant and the biotransformation of 93 target micropolutants (6μg/L) was monitored over time at five different temperatures (4-30°C) range. The experimental kinetic parameters were compared to model predictions. The microbial population was also characterised by high-throughput sequencing to reveal community composition and activity during the biotransformations. Positive correlation of biotransformation rate constants with temperature was found in the 4-20°C range. At higher temperatures, the biotransformation potential decreased or reached a plateau for the majority of the compounds, and just a limited group showed a steady increase of biotransformation rates. The model could predict rate constants above 20°C, despite major risk assessment guidelines recommend Athrinus model predictions in the 0-30°C range. The microbial community also showed significant shift in both composition and activity at higher temperatures, in agreement with the observed decrease in biotransformation potential. Contrarily, for compounds showing an Athrinus-behaviour over the 4-30°C range, the biotransformation processes may be linked to basic living cell functions which are sensitive to temperature fluctuations. Our study highlights limitations in the applicability of Athrinus-based models for the estimation of chemicals fate in biological systems, and the need to re-examine model parameters to assure more accurate predictions for potential chemical exposure in events of temperature fluctuations.

251 Findings from an international ring test for an improved marine biodegradation screening test
Alex T. Martin, 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 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 false positives, which of 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 recognised and 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 centrifugal 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 (pentachlorophenol) and three compounds with variable reported degradation (4-nitrophenol, triethanolamine and hydrolysed poycaolylamide), were used to provide a range of biodegradation potentials by which to validate the new method. Biodegradation data for the five chemicals, in the three test systems used, across the 13 participating laboratories will be presented. The need for clearer guidance on biodegradation testing and interpretation will be discussed, with particular reference to test variability and extended lag phases frequently encountered in marine biodegradation assessments. The role which microbial communities play in chemical biodegradation and the extent to which microbial community analysis can explain inter- and intra-laboratory variation in biodegradation test outcome will also be discussed.

252 Relevance of photolysis for the fate of pentamethrin in deeper water layers - results of a scale-up approach according to OECD TG 309
D. Hennecke, Fraunhofer IOME - Institute for Molecular Biology and Applied Ecology / Ecological chemistry; M. Kruse, Fraunhofer IOME - 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 surface water like direct photolysis and indirect photolysis are not addressed. Since biodegradation is limited in the OECD 309 study, the consequences are critical for substances which are hydrophilically stable but sensitive to light. Within pesticide regulation direct photolysis studies are mandatory, indirect photolysis studies optional. In natural waters, which have to be used for OECD 309, both processes are relevant for photolytically unstable compounds. Hence, beside direct photolysis in the upper layer of a water column, it is interesting to know until 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 litre volume are filled with surface water taken from a natural lake and maintained at 20°C. The geometry of the container result in a water level of 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 5 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 same test setup but equipped with a lid of stainless steel served as dark control. The test is conducted as both pelagic and suspended-sediment setup. Test substance was 14C-pentamethrin, which is known to degrade rapidly in aqueous 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.

253 Poster spotlight: TU267, TU268, TU269
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 combination and analysis of underlying indicators 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 bottles 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 Multi Criteria Decision Analysis (MCDA) approach is integrated within an aggregation 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) is different. The ranking of the packaging could be 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 identifying 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 phases. This is especially true for high priority products such as electronics. In this case, additional functional units are often defined. Allocation is an important step in the life cycle assessment (LCA) process to partition a flow into elementary flows and provides the basis for the comparison of different products and that could be used in multiple life cycles due to efficient recycling. PE International recommend how to apply allocation for the production and recycling of 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 suitable allocation procedure, an LCA study, 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 as obvious as the product phase that is modelled. 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 composition using the available ecoinvent waste tools. In the recycling scenario the laptops are collected and sent to a recycling plant for material recovery and reuse. In the reuse 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 transport 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 make it possible to design the making. The results also show that increased energy efficiency of new products has a limited impact on the overall results.

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 composition using the available ecoinvent waste tools. In the recycling scenario the laptops are collected and sent to a recycling plant for material recovery and reuse. In the reuse 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 transport 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 make it possible to design the 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-RTG) 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 22°C) are reached. The transport of the air 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 nutrient regimes can be optimized. In this sense, different litter experiments show 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 perlique 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. In 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. In this paper, the 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 on Persistent Organic Pollutants (POPs) by 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 the open applications in these uses will be produced and the process will be followed by endorsement in 2017. The guidance will focus on the 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. 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 SUPPES (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. In this paper the evaluation and guidance update of leaf baits with data on PFOS producers and sales with guidance on the use of alternatives in this case of per- and polyfluoroalkyl substances (PFAS) in 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 SUPPES 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 assessment is commonly evaluated by life cycle assessment of the product's or chemical's life cycle, or between types of impacts. For this reason, the 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. In this paper the evaluation and guidance update of leaf baits with data on PFOS producers and sales with guidance on the use of alternatives in this case of per- and polyfluoroalkyl substances (PFAS) in consumer products.

262 Implementing a life cycle perspective in chemical alternatives assessment - the case of per- and 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) usually progresses in three phases: (i) literature review and evaluation and guidance (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
functional basis. Based on this case we were also able to further elaborate on the inclusion of the life cycle perspective in AEA framework by identifying both possibilities and challenges. We conclude that the inclusion of a life cycle perspective in AEA is crucial for an informed and sustainable substitution, as lack of life cycle thinking can lead to problem shifting. We show that LCA, with its focus on function, is a tool that can identify such problem shifting as well as the key chemical properties to which functional concepts are linked. The toxicological effects in such a case can however turn out to be difficult, especially for substances such as the PFAS as they are outside the domain of the LCIA model. In the case under study here we conclude that the DWR should be selected with three main considerations: (i) the intrinsic hazard properties of the chemistry, selecting the DWR associated with the lowest hazard but, (ii) providing the functionality as needed and, (iii) giving the garment the longest life length.

263 How much function do we need in textiles? Strategies for replacing PFASs based on end-user requirements

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Current approaches to substitute harmful chemicals could benefit from a broader perspective when it comes to the functionality they provide in consumer products1. Following the concept of “functional substitute”, this study presents an evaluation of material properties of new durable water repellents (DWR) for textiles focusing on end-user requirements. Since the phase out of side-chain fluorinated polymers (SPFs) based on long perfluoroalkyl moieties that were associated with the release of persistent, biaccumulative and toxic perfluoroalkyl acids (PFAs), a variety of new DWRs have been developed including biodegradable materials that are based on non-fluorinated monomers. To understand the user requirements and their underlying properties to provide substances and oleophobic fibre modifications SFPs based on long perfluoroalkyl chains were historically used on all kinds of different textiles applications. It is so far unclear if alternative DWRs can follow this “one solution will solve all” approach. By segmenting the textile sectors in terms of liquid repellency, this study sets out to outline the different requirements in case studies for functional outdoor clothing and occupational medical apparel. For functional outdoor clothing, a “bottom-up” strategy was chosen by using a survey to assess the consumers’ needs and expectations. For occupational medical apparel, exposure scenarios to liquids were defined based on protection needs described by the Center for Disease Control and Prevention (CDC)2. Based on these demands, relevant liquids were chosen to evaluate repellent properties using established test methods and by developing a new method to determine the roll-off angle for textiles. It has been found that some non-fluorinated DWRs based on green chemistry concepts showed excellent water repellence and also a resistance towards the penetration of liquids with intermediate polarity (e.g. orange juice and synthetic blood). When it comes to liquids with very low surface tension like gastric fluid evaluating the repellence on the surface repellency, but also the only materials that protected against liquid penetration. This study of chemical substitution based on chemical and textile functionality as well as end-user requirements pointed out the opportunities and limitations for functional substitution.

264 Analysis of the technical and economic feasibility of alternatives to lead gunshot

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An analysis of the technical and economic feasibility of alternatives to lead gunshot has been prepared by ECHA as part of a REACH Annex XV Restriction Report on lead and lead compounds used in shot in wetlands. Lead has historically been used in cartridges because of its softness, low melting point, high density, relatively low price and high abundance. Because of these properties, lead is often considered to be the ideal material for use in ammunition. Steel gunshot (soft iron) is by far the most commonly provided alternative, often using established industrial test methods and by developing a new method to determine the roll-off angle for textiles. It has been found that some non-fluorinated DWRs based on green chemistry concepts showed excellent water repellence and also a resistance towards the penetration of liquids with intermediate polarity (e.g. orange juice and synthetic blood). When it comes to liquids with very low surface tension like gastric fluid evaluating the repellence on the surface repellency, but also the only materials that protected against liquid penetration. This study of chemical substitution based on chemical and textile functionality as well as end-user requirements pointed out the opportunities and limitations for functional substitution.

265 The road to successful substitutions - case studies

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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, functionality and safety in a given adaptation. 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 uses requires that we study the case we were also able to further elaborate on the inclusion of the life cycle perspective in AEA framework by identifying both possibilities and challenges. We conclude that the inclusion of a life cycle perspective in AEA is crucial for an informed and sustainable substitution, as lack of life cycle thinking can lead to problem shifting. We show that LCA, with its focus on function, is a tool that can identify such problem shifting as well as the key chemical properties to which functional concepts are linked. The toxicological effects in such a case can however turn out to be difficult, especially for substances such as the PFAS as they are outside the domain of the LCIA model. In the case under study here we conclude that the DWR should be selected with three main considerations: (i) the intrinsic hazard properties of the chemistry, selecting the DWR associated with the lowest hazard but, (ii) providing the functionality as needed and, (iii) giving the garment the longest life length.

266 EDAPHOBASE - soil biodiversity data warehouse and its applications in ecology

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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 data warehouse will focus on the development of (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 the world.
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 Diving into REACH database with R studio 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 Agencies (ECHA) data on more than eight thousand chemicals. These data includes all the physico-chemical properties (166’926 test results), ecotoxicity (305’068 test results) and human toxicity data (41’381 test results) available in the IUCLID 5.5 database (as of May 2020). The data van den Brink, Alwareda unique values for 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 Rstudio program [6] where data treatments / manipulations / calculations were performed. R allowed us to code 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-constituents as well as UVCB (Unknown or Variable composition, Complex reaction products or Biological materials). The database covers acute and chronic toxicity tests for various organisms with about 305’068 End-point study reports (ESR).

268 The effect of modelling decisions on macroinvertebrate sensitivity modelling
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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 chromatic 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 48h of hatching) or neomaeas (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 species, which will provide the tools for pattern recognition in the subsequent point study reports (ESR) and can only be used as supporting 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.

270 Ceriodaphnia is sensitive to Daphnia and should fulfill invertebrate regulatory toxicity requirements

The OECD 202 Acute Daphnia Immobilisation Toxicity Test requires the use of Daphnia magna or another “suitable Daphnia species, (e.g., Daphnia pulex)”. 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 supporting or weight of evidence studies and not as key exposure. To further the acceptability of this proposal, it would be useful to develop an OECD test guideline for Ceriodaphnia acute and chronic toxicity to supplement existing ISO, ASTM and USEPA standard guidelines.

271 Poster spotlight: TU001, TU002, TU003
Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (II)
58
272 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 waters were monitored for sediment deposition, contaminant, 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 MEX-T. The samples were analyzed for a variety of metals including Pb, Cu, Ni and THg. The fractionation allowed distinguishing Pb particles that are not 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 the 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.

273 The effect of percolation and form on lead bioavailability and toxicity to Enchytraeus crypticus
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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 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 were higher for 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 metal might be good ways to get rid of the influence of counterions and increase environmental realism of laboratory toxicity studies.

274 To leach or not to leach: Soil enzymatic responses to metal mixture species
F. Awual, University of Saskatchewan / Toxicology 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. Awual1, B. Hale2 & S. Siciliano1, 2University of Saskatchewan, Toxicology Center, 1University 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 unequally, 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 was tested and compared to the salts. The experiment was conducted with three Canadian soils (pH: 3.5-7), three metal species, five fixed metal mixture rays, and five metals (Pb, Cu, Co, Ni, Zn) at one dose. The activity of the soil enzymes ammonia monooxygenases, beta-glucosidases, acid-phosphatases and arylsulphatases were determined colorimetrically. Results showed that leaching alone significantly inhibits the enzyme ammonia monooxygenases 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 monooxygenases represented a decreasing hermetic toxicity across 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

275 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 (soil-dwelling 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 moisture content), 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 (avoidance). The waters were not effective in eliminating the effect of moisture content 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 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.

276 Manganese bioavailability in legacy contaminated soils by medieval
promote the decrease of toxicity towards the microalgae. Further research is needed amendment with MSWC, and despite its beneficial chemical effects, toxicological although inducing a slight toxic effect in the microalgae. Concerning the application rates of the correctives, due to a dilution effect. In general, amending the MSWC. Total Cd, Cr, Cu and Zn concentrations in soil decreased in the high organic matter, and N content, was only possible by the simultaneous application of comparison to non-

immobilization of metals in the soil; (iv) the effects of the amendments on soil assessed evaluating: (i) soil chemical properties, (ii) plant growth, (iii) 

invertebrates and ii) the snail ability to efficiently regulate this element. in soils (up to more than 8000 mg.kg\(^{-1}\)). Extractible 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. asperus snail tissues allowed to show: i) the fluorometric analysis of Mn bioavailability in soils and soil 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\(\text{mg}_{\text{Sn}}\cdot\text{kg}^{-1}\). 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).

Chemical and ecotoxological effects of the use of residues from the pulp and paper industry for the remediation of soils degraded by mining activities P. Almeida, University of Aveiro / CESAM & Departamento de Química, L.A. Tarelho, Universidade de Aveiro / CESAM & Departamento de Química, S. Rodrigues, Universidade de Aveiro / CESAM & Departamento de Química 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 in applying compost to, among other tests, soil invertebrates and a pot experiment, with Agrostis tenuis 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 microalgae 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. tenuis in pots where composts were applied, but with some variability between replicates. Phytotoxicity was observed in some of the pot-related preliminary experiments, to allow the compost immobilization in soil country, 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 MSWC. Total Mn concentrations 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 (applied in 2.5, 5.0 and 10%, w/w, dry matter), presented a better performance, although inducing a slight toxic effect in the microalgae. Concerning the amendment with 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 plants species, since Agrostis tenuis showed some phytotoxicological
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) upstream (P1) and downstream (P2) of an urban area, under 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 concentrations 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.

Microplastic pollution in upstream river catchments

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Microparticles (<5 mm) are known to pollute large lakes and river systems globally, though they are often associated with wastewater treatment plants and centres 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 headwaters and urban rivers that do not receive wastewater treatment effluent across England’s Midlands, as well as in atmospheric fallout. FTIR analysis of three months’ samples identified microplastic particles in 30 litre water samples taken from the headwaters of the River Trent and its tributaries. It has also been used to identify non-fibrous microplastic fragments in rural and (sub)urban atmospheric fallout. Moreover, spherical particles that resemble those used in cosmetic / personal care products have been identified in rivers that do not receive wastewater treatment effluent, some of which have proven not to be polymer-based following FTIR analysis. This brings into question the source, and chemical composition, of spherical particles that have previously been visually identified as plastic spheres likely derived from cosmetic particles. The findings of this study have identified the need for the more extensive consideration of upstream catchments and reaches of rivers. In the receive wastewater treatment effluent, the study of freshwater microplastic pollution. The work conducted here suggests that, though wastewater treatment facilities and large centres of population and industry are suitable predictors of microplastic pollution, the cumulative contribution of headwaters and tributaries are likely to inflate a river’s microplastic load.

Microplastics in stormwater ponds

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Stormwater runoff contains pollutants from land surfaces. As the majority of production and consumption of synthetic polymers is happened on land, microplastic (MP) is one group of problematic pollutants in urban stormwater runoff. However, MPs in stormwater has barely been investigated. 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 by 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 sonication. Inorganic materials were removed 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 ZnSe window 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 the final presentation. Nevertheless, the water samples have shown that stormwater pond do not retain all MPs, particularly for low-density polymers. With the high mobility, discharges from stormwater ponds will potentially affect adjacent environmental compartments.

Towards a more realistic assessment of microplastics as pollutant transporter: a combined experimental and modelling study

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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. This study is aimed to perform a source apportionment of PM10 concentrations in four sites in Southern Italy

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This study is aimed to perform a source apportionment of PM10 collected simultaneously in four sites located in the Puglia region (South-Eastern Italy). The Air Pollution, Biomonitoring and Human Health (II)

Analysis of the contribution of a coal-fired power plant to PM10 concentrations in four sites in Southern Italy

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This study is aimed to perform a source apportionment of PM10 collected simultaneously in four sites located in the Puglia region (South-Eastern Italy). The
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 PM on human, in vitro or in vivo models exposed to PM samples previously collected on filters, then detached and resuspended in suitable media. This procedure, although extensively applied, has always posed the question about the representativeness of extracted PM in comparison to airborne PM. However, the lack of exposure systems directly working under environmental conditions made this experimental set-up widely accepted. In the last years, also thank to Nanotoxicology studies, innovative exposure modules have been proposed which are able to convey air-dispersed particles on cultured cells. The majority of the application so far reported, however deal with the exposure under laboratory condition of engineered nanoparticles or other molecules of interest. Here we report the results obtained by the Validation of the Biofunctional Equivalent Cultured Human Lung (BECHEL) at the air-liquid-interface (ALI) under environmental condition to environmental pollution by means of an exposure module (CULTEX® RFS module). The data demonstrate that the maximal feasible exposure evaluated for the CULTEX® system is representative of the dosimetry calculated for human exposure. The toxicological evaluation evidenced the absence of cytotoxic effects and absence of significant alteration of the BECHEL frequencies. Moreover, no cytotoxic effects were observed on the differential expression of selected genes of interest. Altogether our results show that the time is arrived to leave the warm of the laboratory bench and to start toxicological evaluation in field campaign. Although the proposed approach still require an extensive evaluation to assess all the pros and cons we reckon that the toxicological data obtainable under really representative environmental conditions may be more representative to understand the biological processes activated by air pollution.

Indoor and Outdoor Air contamination by endocrine disruptor pollutants in the North part of France

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The atmosphere is the main environment with which humans have the most important exchanges. However, human activities (food production, urbanisation, mineral extraction, etc.) contributes a large part of the pollution. To date, few data exist on air contamination by endocrine disruptor compounds (EDCs) in France. With the experience acquiring in Paris region in a previous research, the research team and ATMO Hauts-de-France realised two studies in the North part of France about indoor and outdoor air contamination by EDCs. According to the method previously validated several types of indoor environments (office, house, scholar building and day nursery) and several areas (rural forest, urban, industrial) were investigated over 2 years (2015 and 2016-2017). During each season, 7 or 5 sites (indoors and outdoors) were sampling during three successive 2-week periods. The device is composed to a TSP filter system and a cartridge containing XAD resin, connected to a flowmeter and a pump. 70 EDCs were analysed by LC-MS/MS. GC-MS/MS in selective ion monitoring (SIM) for selected human diseases and air pollutants concentration. Finally phthalates concentrations are linked to consumer products, building materials, furnishing... PAHs are coming from residential and tertiary heating and from road transport; alkylphenols and musks from detergents. Except few specific sites, the EDCs concentrations in air in the North part of France are in the same order than those in Paris region. In conclusion, the air contamination by EDCs is becoming a sanitary concern because French people spend 80% of time in indoor environment and young children, a particularly sensitive population, are the most exposed.

Air pollution and health: early biological effects in children exposed to air pollutants and genotoxic effect of PM0.5 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) is to evaluate the associations between air pollution and early biological effects in children 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 assessed using an in vitro or in vivo model exposed to PM samples previously collected on filters, then detached and resuspended in suitable media. This procedure, although extensively applied, has always posed the question about the representativeness of extracted PM in comparison to airborne PM. However, the lack of exposure systems directly working under environmental conditions made this experimental set-up widely accepted. In the last years, also thank to Nanotoxicology studies, innovative exposure modules have been proposed which are able to convey air-dispersed particles on cultured cells. The majority of the application so far reported, however deal with the exposure under laboratory condition of engineered nanoparticles or other molecules of interest. Here we report the results obtained by the Validation of the Biofunctional Equivalent Cultured Human Lung (BECHEL) at the air-liquid-interface (ALI) under environmental condition to environmental pollution by means of an exposure module (CULTEX® RFS module). The data demonstrate that the maximal feasible exposure evaluated for the CULTEX® system is representative of the dosimetry calculated for human exposure. The toxicological evaluation evidenced the absence of cytotoxic effects and absence of significant alteration of the BECHEL frequencies. Moreover, no cytotoxic effects were observed on the differential expression of selected genes of interest. Altogether our results show that the time is arrived to leave the warm of the laboratory bench and to start toxicological evaluation in field campaign. Although the proposed approach still require an extensive evaluation to assess all the pros and cons we reckon that the toxicological data obtainable under really representative environmental conditions may be more representative to understand the biological processes activated by air pollution.

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implementing policies of public health protection.

288 Source apportionment of PM near steel plant by electron microscopy
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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 was analyzed by SEM-EDS and based on the morphology and chemical composition they have been classified into the following main groups: Aluminosilicate particles; Silicium reach 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. Beside 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, PM1 fractions, showing that the antropic 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. Ajnad. Oregon State University / Environmental and Molecular Toxicology; S.L. Massey 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 dust is a matter that remains a global health concern as transport models continue to fall short of accurate predictions. To improve modeling accuracy the determination of chemical speciation of PAHs within PM1 is necessary. Secondary organic aerosol (SOA) particles sorb PAHs during formation and transport them as a large fraction of global atmospheric PM1, and the presence of PAH vapor has been demonstrated to increase the mass loading of atmospheric SOA. The oxidation of four PAHs were studied in laboratory generated α-pinene SOA experiments. Dibenzo[a]anthracene (DBT), phenanthrene (PHE), pyrene (PYR), and benz[a]anthracene (BaA) were measured along with their oxidation products in freshly formed α-pinene ozonolysis SOA grown in the presence of vapor phase PAH (PSOA). Ratios of oxidized transformation products was measured and changes in those ratios was observed during the aging of the SOA, as well as after exposure to ozone. In freshly formed PSOA, the sum of measured oxidized products was found to be equal to the measured amount of parent compound in all four systems. Characterization of aged particles provides evidence of continuing chemical reactions in PHE and PYR PSOA. DBT and PHE PSOA showed evidence that ozone exposure, performed in a flow-tube reactor, results in further oxidation. Data suggests the environment inside SOA particles are complex and dynamic, and need to be further explored. Implications of the presence of oxidized PAHs in long-range transport modeling will be discussed.

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

290 The role of the p38-activated protein kinase signaling pathway-mediated autophagy in cadmium-exposed monogonont rotifer Brachionus koreanaus
H. Kang, C. Jeong, J. Lee, Sungkyunkwan University
Autophagy is a ‘self-eating’ system that regulates the degradation of cellular contents and organelles involved in various biological processes at both structural and functional levels. However, despite its crucial role in organisms the regulatory mechanism of autophagy remains largely unclear, particularly in invertebrates. In this study, conserved autophagy in the rotifer Brachionus koreanaus in response to cadmium (Cd) exposure was verified by measuring acidic vesicle organelles using acridine orange (AO) and neutral red (NR) staining, and by detecting LC3 II/I Western blot and immunofluorescence. We also demonstrated activation of the mitogen-activated protein kinase (MAPK) in response to Cd-induced oxidative stress, leading to the induction of autophagy in B. koreanaus. This was further verified by analysis of MAPK protein levels and immunofluorescence of LC3 II/I after treatment with reactive oxygen species scavengers and inhibitors specific to MAPks. We propose a p38 MAPK-mediated regulatory mechanism of autophagy in B. koreanaus in response to Cd-induced oxidative stress. This study will contribute to a better understanding of autophagic processes in invertebrates and its modulation by environmental stressors.

291 Effects of trielosan (TCS) on antioxidant system and oxidative stress-mediated gene expression in the copepod Tigriopus japonicus
J. Paek, J. Lee, Sungkyunkwan University
Triclosan (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 437.476μg/L, respectively. While in the nauplius stages the corresponding values were 20μg/L and 51.76μg/L, respectively. Fecundity was significantly reduced (P < 0.05) in response to TCS at 100μg/L. Concentration and time-dependent analysis of ROS, GSH content (%), and antioxidant enzymatic activities were performed 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 (CYP30266A3 and CYP3037A1) 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.

292 The protective role of multixenobiotic resistance (MXR)-mediated ATP-binding cassette (ABC) transporters in biocides-exposed rotifer Brachionus koreanus
Y. Lee, H. Kang, C. Jeong, J. Lee, Sungkyunkwan University
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. Among the transporters, P-glycoprotein (P-gp) and multidrug resistance-associated protein (MRP) are ATP-binding cassette (ABC) transporters that confer multidrug 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 malathion) using fluorescent substrates and inhibitors specific to P-gp and MRP. The efflux activities of P-gp and MRP in the rotifer B. 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 in response 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 mediated. Taken together, our results demonstrate the important role of the MXR efflux system in the defense response to biocides, thereby providing a better understanding of rotifer defense mechanisms on the molecular level.

293 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 keystone 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 hydroxase gene (TRH), the rate limiting enzyme of serotonin synthesis, and other three having the transporter protein gene ABC21 mutated. Bi- and Bi-allelic del TRH mutants lack serotonin and have their growth rates impaired. Bi-allelic del ABC21 mutants had lower transaminase 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 ABC21 is involved in the detoxification of ivermectin and its total or partial knockout dramatically increased its toxicity. These results provide the first evidence ever 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 Larvae of Chaoborus spp. (phantom midges) which inhabit water bodies in the agricultural landscape are very sensitive to synthetic pyrethroid insecticides and are known to often population changes impacted by pyrethroids. Individuals of 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 post-overwintering fourth instar larvae of Chaoborus obscursus. 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 adult 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 of Chaoborus obscursus were most susceptible to pyrethroids 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? W. Brack, M.A. Hashmi, Helmholtz Centre for Environmental Research-UFZ / Effect-Directed Analysis; M. Koenig, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; M. Mischket, UFZ-Helmholtz Centre for Environmental Research / Effect-Directed Analysis; m. muz, UFZ - Helmholtz Centre for Environmental Research / Cell Toxicology; T. Schmoldt, Helmholtz Centre for Environmental Research / Effect-Directed Analysis; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; C. di paola, RWTH Aachen University / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Tindall, Watchfrog S.A.

European water resources are contaminated with complex mixtures of tens of thousands of chemicals among them many non-regulated compounds with 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, Rhône and Holtemme as examples. 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 R. Ashauer, University of York / Environment

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 (Ashauer et al. 2015, STOTEN 512:540. However, mixtures are 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? R. Aaltenbergen, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology, M. Faust, Faust & Backhaus Environmental Consulting; A. Kortenkamp, Brunel University London; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

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 mixtures of industrial and natural compounds could be identified as the drivers of toxicity in freshwater. 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, Rhône and Holtemme as examples. 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.

299 A mixture risk assessment for pollutants that reach humans via the water – fish exposure route A. Kortenkamp, Brunel University London; M. Faust, Faust & Backhaus Environmental Consulting

An important route for human exposures to substances present in freshwater is through the consumption of fish. To protect humans against this route of exposure, the WFD defines specific quality standards for priority substances. In general, combined exposures are not considered, with the exception of quality standards for mixtures of specific contaminant groups, such as polychlorinated dioxins (PCDD) and polybrominated diphenyl ethers (PBDE). However, the possibility of combination effects across these pollutant groups is not currently considered. We present an advanced tiered mixture risk assessment for these groups of pollutants, first by using Water Framework Directive Quality Standards defined for PCDD and
300 An Advanced Methodological Framework for the Identification of Priority Pollutants and Priority Mixtures of Pollutants in European Freshwaters

M. Faust, Backhaus & Backhaus Environmental Consulting; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; V. Dulio, INERIS; J. van Gils, DELTARES; A. Ginebreda, CSIC - Spanish National Research Council / Environmental Chemistry; A. Kortenkamp, Brunel University London; J. Munthe, IVL Swedish Environmental Research Institute Ltd; J. Slabodnik, Environmental Institute; K. Tötköni, NIVAS / Hydroecology and Risk Assessment; A. van Welzen, KWR Watercycle Research Institute / Chemical Water Quality and Health

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 recent prioritisation procedures under the EU Water Framework Directive (WFD). Through data analysis, we aimed to develop for each conclusion a conclusive risk assessment cannot be met. Significant risks from so-called emerging pollutants may remain undetected. The WFD does not include an effective mechanism to close such knowledge gaps. The introduction of a watch-list mechanism for up to 10 substances provided a minor improvement but no fundamental change to this situation. (ii) Individual pollutants are assessed as if they would occur in isolation, largely ignoring the fact that they are part of complex multi-constituent mixtures. Environmental quality standards that have been established for single priority pollutants may not be sufficiently protective against mixture effects. Regulatory approaches for effectively tackling the problem are missing. The advanced framework integrates all available lines of evidence (LOE) on significant risks. This includes evidence from (i) ecological monitoring (field observations on so-called biological quality elements), (ii) effect-based monitoring (in vitro or in vivo testing in the lab or onsite), (iii) chemical monitoring in combination with so-called component-based mixture risk assessment approaches, (iv) integrated modelling of co-exposure and resulting mixture risks. Where one or more lines of evidence identify groups of pollutants presenting a significant risk, thorough chemical characterisation for risk reduction measures is needed. Where appropriate, such groups 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 somehow blocked by significant data or knowledge gaps, mixture components of potential importance may be 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

T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; A. Arrenhuis, University of Gothenburg / Biological and Environmental Sciences; R. Behra, Eawag / Department of Environmental Toxicology; T. Seiler, RWTH Aachen University / Department of Anatomy Physiology and Cell Biology; R. Ashauer, University of York / Environment

The EU Water Framework Directive defines two environmental quality standards for assessing the chemical status of surface waters. The MAC-EQS defines the Maximum Allowable Concentration that should never be exceeded and the AA-EQS defines a concentration that should be exceeded by the Annual Average concentration. While the AA-EQS should account for the acute toxicity of a substance, the AA-EQS is based on chronic eco-toxicity 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 eco-toxicity 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 population 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.
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 review of the French and EU Water Framework Directive Environmental Quality Standards for endocrine disrupting chemicals. Group 4 contains all substances that have no ED effects demonstrated from now on. A study was made of these substances ED characteristics 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 ED characteristics were considered ED effects, and which need to be reassessed as a matter of priority. Substances for which ED characteristics have been 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 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 ED characteristics 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


The most common method for deriving water quality benchmarks (WQBs) for toxicants is the use of a species sensitivity distribution (SSD) to estimate a threshold chemical concentration, with an assumed bell-shaped distribution. This method is used by regulatory jurisdictions to deliver environmental protection. This work led to 4 groups of substances. Group 1 contains substances whose ED characteristics have been considered 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 ED characteristics 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.
**Microbial community ecotoxicology in environmental risk assessment and ecosystem monitoring**

**308**
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, Cu, Cr 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, Tsapakis M, Pagou K, Drakopoulou P, Assimakopoulou G, Kontoyiannis H, Panayotidis P. 2015. Methods of eutrophication assessment in the context of the water framework directive. Cont Shelf Res. 108: 156-168.

**309**
Impacts of stormwater on microbial community structure and function in estuarine sediments

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**310**
Drought as environmental driver on ciliates and micrometazoans in a multistressors scenario. An experimental approach

J. López-Duval, F. Romero, V. Aćuña, S. Sabater, ICRA Catalan Institute for Water Research

Climate change will affect agriculture practices and productivity because increased intensity and frequency of drought events are expected to stress crops and damages 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 micrometazoans 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 micrometazoans 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 different environmental conditions). The community was exposed to the stressors for 7 weeks. Diversity of Ciliophorans and micrometazoans 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 micrometazoans in all treatments in terms of density, but a trend of increasing the percentage of ciliates in these 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 than 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 pre- and post-exposure results, it 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 micrometazoans and ciliate communities in our experimental system while pesticides and temperature produce significant effects only in combination with drought.

**311**
Linking pesticide pollution with periphyton quality in agricultural streams: a fatty acids approach

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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. Three 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 testing 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 in the studied
streams might be toxic for periphyton (i.e. inhibiting the photosynthetic activity), being herbicides driving the 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 Estroline and triclosan mixture alters soil metagenomics during degradation
D.L. Carg, Texas Tech University / Biological Sciences; E. Ousji, 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 soils and consequences for soil microbial community processes are poorly understood. Estroline 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 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 (estroline) 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 the 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 a Damage and Value Added Tax (AT) partially based on the life recycling of primary metals. This is achieved by adopting the principles of life recycling -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 noted economic indicators calculated to evaluate 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. Papageorgiou, 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 associated with its reverse supply chain design and operation. The examined networks comprise multi-echelons, including disposer markets, collection facilities, recycling plants 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 an economic indicator calculated to evaluate the 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 a Korean Eco-Indicator 99 modelled to represent the life cycle 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 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 optimisation problem 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, Université Libre de Bruxelles; W. Achten, Université Libre de Bruxelles / Department Geosciences, Environment and Society
The purpose of this presentation is to provide a short insight about a study examining the principles and feasibility of a shift from Value Added Tax (VAT) or sales tax to a Damage and Value Added Tax (DaVAT) partially based on the life cycle assessment of products and services. With this shift, goods and services that seriously harm the environment and human health will be priced up, those that have less impact will be priced down. The DaVAT system relies on three essential
points: i) Apply VAT (or consumption taxes in general) to all goods and services and reduce its multiple rates to one single low rate (e.g. 3%) called Uniform VAT (UVAT); ii) Add to UVAT a per-unit tax called Global Damage Tax (GDT) calculated on the basis of environmental impacts assessed by means of specific or generic LCAs. In the case of potentially high-polluting products or industries, a specific LCA will be automatically imposed; iii) In order to reflect environmental, societal or technical control specific to a certain substance classified as hazardous, a specific Damage Tax (SDT) is proposed that extends beyond LCA. DaVAT is the sum of UVAT, GDT and SDT. DaVAT is conceived not as an additional burden but rather as a shift of taxation, as the rate of the old consumption taxes can decrease proportionally to the increase of GDT. DaVAT is also designed in such a way that the erosion of tax revenues, when pollutant releases would be offset by the extension of the tax to all goods and services and by the possibility to gradually re-increase the VAT rate when the number of highly-polluting products decreases. To reduce the variance of the LCA results used for this purpose, the DaVAT system should use common databases, apply the same inventory, characterization, normalization and weighting methods, as well as refer to the same impact categories and the same cut-off rules. LCA as a whole can stay as it is, but for use in DaVAT an additional impact category e.g. radiological or climate assessment based on previous studies has been made of the costs of implementation, maintenance, administration and compliance of DaVAT, as well as of the risks of fraud, price changes and acceptability of the proposal. The presentation shall briefly outline the results of this assessment.

318 Towards global guidance on LCIA of mineral resource use - outcomes from the UN Environment Life Cycle Initiative task force
T. Sonderregger, ETH Zurich; M. Berger, Technische Universität Berlin / Chair of Sustainable Engineering - Office Z1; R.A. Alvarenga, Ghent University / Department of Sustainable Organic Chemistry and Technology; V. Bach, Technische Universität Berlin / Chair of Sustainable Engineering; A. Cimprich, University of Waterloo; J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology; R. Frischknecht, treere Ltd.; J. Guinee, University of Leiden / Institute of Environmental Sciences; C. Helbig, University of Augsburg; O. Jolliet, University of Michigan; M. MOTOSHITA, National Institute of Advanced Industrial Sci. and. T. Nohynek, Monash University / Department of Economics; A. Pregill, UVAT, GDT and SDT. DaVAT is conceived not as an additional burden but rather as a shift of taxation, as the rate of the old consumption taxes can decrease proportionally to the increase of GDT. DaVAT is also designed in such a way that the erosion of tax revenues, when pollutant releases would be offset by the extension of the tax to all goods and services and by the possibility to gradually re-increase the VAT rate when the number of highly-polluting products decreases. To reduce the variance of the LCA results used for this purpose, the DaVAT system should use common databases, apply the same inventory, characterization, normalization and weighting methods, as well as refer to the same impact categories and the same cut-off rules. LCA as a whole can stay as it is, but for use in DaVAT an additional impact category e.g. radiological or climate assessment based on previous studies has been made of the costs of implementation, maintenance, administration and compliance of DaVAT, as well as of the risks of fraud, price changes and acceptability of the proposal. The presentation shall briefly outline the results of this assessment.

319 Poster spotlight: TU214, TU215, TU237
Safe by Design: responsible and innovative research for safe and sustainable chemistry

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) and 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) of both Ag and TiO2 and how these properties affected the particles performances. We first demonstrated that both at colloidal and dried state a matrix of SiO2 surrounding TiO2 and Ag nanoparticles 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 nano-reactivity: 1) Silica acts as dispensing/goldilocks matrix for decreasing the production of ROS, but improving the photocatalytic performances of pristine sample; 2) Silica act 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. 1) Silica acts as dispensing/goldilocks matrix for decreasing the production of methanol from CO2 and hydrogen.

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 facing 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 developed 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 improve 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 PFAEs 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, PFAEs have been used because their perfluoralkyl chains have the ability to repel liquids of a wide range of polarities (OW, water, sweat, and water repellent). DWR compounds, like PFAs 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 rising 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 exposure 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 a Classification and Package Category (CPPC) classification scheme, 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. Bonetto, 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

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 for ecotoxicity (i.e. aquatic and terrestrial), (ii) 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-applciation 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 Nanotechnology on the Margins: The confluence of Modern Agriculture and Aciulture

Z. Browning, Brownings 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 emerge from the urban to the backyard garden to the farm 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 contaminations, or use issues with human and health (toxicology) issues and pathogen diseases, 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 practices 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 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)

327 A new multi-dimensional method for evidence synthesis and weighting in bee risk assessment

A. Gradish, University of Guelph / School of Environmental Sciences; O. Klein, Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais; Rosa-Fontana, Unesp - Instituto de Biologia / Departamento De Biologia, Centro de Estudos de Insetos Sociais; R. Cornelio Ferreira Nocelli, Universidade Federal de São Carlos UFScar Araras / Ciências Biológica Departamento de Ciências da Natureza Matemática e Educação; O. Malaspina, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais; S. Rosa-Dorigo, Universidade Estadual Paulista Júlio de Mesquita Filho Unesp Rio Claro / Departamento De Biologia, Centro de Estudos de Insetos Sociais; M. Miles, Bayer CropScience - Environmental Health & Safety; J. Lueckmann, Rifcon GmbH / Ecotox Field; D. Lehmann, U.S. Environmental Protection Agency / ORD NHEERL; N. Riddell, Syngenta Ltd / Environmental Safety; A. Dinter, Chemnitzer Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECIPA

The potential exposure routes and actual exposure of the bumble bee queen, respect of non-climatic factors, is a potential area of conflict between beekeepers and farmers. Entomopollination is an important biological and economic factor for a number of crops and bee conservation is an important qualitative information. 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 qualitative way. By progressively simplifying the assessment by using fully quantitative measurements that, at present stage, are hardly capable of retaining important qualitative information.

330 Standardization of an in vitro larval rearing method for stingless bee species Melipona scutellaris for use in toxicological bioassay studies

A.S. Gradish, University of Guelph / School of Environmental Sciences; O. Klein, Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais; M. Miles, Bayer CropScience - Environmental Health & Safety; J. Lueckmann, Rifcon GmbH / Ecotox Field; D. Lehmann, U.S. Environmental Protection Agency / ORD NHEERL; N. Riddell, Syngenta Ltd / Environmental Safety; A. Dinter, Chemnitzer Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECIPA

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, chlorpyrifos, 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 core of 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 qualitative way. By progressively simplifying the assessment by using fully quantitative measurements that, at present stage, are hardly capable of retaining important qualitative information.

328 Pesticide exposure assessment paradigm for Bumble Bees

J. van de Steen, Alveus AB Consultancy; C. Cutler, Dalhousie University / Faculty of Agriculture; D. Goulson, School of Life Sciences, University of Sussex; A. Gradish, Universalis Australia / School of Environmental Sciences; O. Klein, Eurofins Agroscience Services Ecotoxic GmbH & Ecotoxic Field; D. Lehmann, U.S. Environmental Protection Agency / ORD NHEERL; N. Riddell, Syngenta Ltd / Environmental Safety; A. Dinter, Chemnitzer Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECIPA

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)

329 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. Riddell, Syngenta Ltd / Environmental Safety; A. Dinter, Chemnitzer Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECIPA

Industry therefore believes that a revision of the assessment scheme for use by regulatory authorities is needed. Building on an analysis of the proposed developments in the EFSA Bee Guidance Document, we suggest proactive and practical approaches based on analysis of existing data generated thus far on honeybees Using the existing regulatory framework, we have:• A new multi-dimensional method for evidence synthesis and weighting in bee risk assessment• A new pragmatic, applicable and consistent guidance document within the regulatory framework and has invested much time and money in developing a practical alternative based on the same science.

330 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 / Departamento De Biologia, Centro de Estudos de Insetos Sociais; Rosa-Fontana, Unesp - Instituto de Biologia / Departamento De Biologia, Centro de Estudos de Insetos Sociais; R. Cornelio Ferreira Nocelli, Universidade Federal de São Carlos UFScar Araras / Ciências Biológica Departamento de Ciências da Natureza Matemática e Educação; O. Malaspina, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais; S. Rosa-Dorigo, Universidade Estadual Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais; M. Miles, Bayer CropScience - Environmental Health & Safety; J. Lueckmann, Rifcon GmbH / Ecotox Field; D. Lehmann, U.S. Environmental Protection Agency / ORD NHEERL; N. Riddell, Syngenta Ltd / Environmental Safety; A. Dinter, Chemnitzer Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECIPA

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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. Ursundman, 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 2005 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. Jolliet, 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 product use, i.e the fraction of a chemical in product that is cumulatively taken in by the users. 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 ingestions 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 time. The total 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.

334 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-Kyere, 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. Lantin, 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^-5 ng/m^3 in air. Concentrations are also relatively high in urban media (e.g., low µg/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 levels. “Bottom up” emissions were estimated using 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 considered to the SP and TPhP, respectively. The emissions factors of OPEs from indoor air to outdoor air estimated in this study were within the range estimated by Liagkouridis et al (2017) for bulk emissions to indoor air. These “bottom up” emissions from indoors to outdoors estimated here were one to two orders of magnitude lower than the “top down” estimates, which could be caused by higher emissions from commercial buildings, or through direct emissions of OPEs to outdoor air, such as from insulation. It is clear that elevated indoor and outdoor air concentrations of OPEs are due to emissions from the many products and materials to which they are added. When aggregated over a city scale, indoor air emissions vented to outdoor air were lower, but within one to two orders of magnitude of aggregate air emissions back-calculated from outdoor concentrations. Outdoor rate is governed by partitioning between air and high sorptive-capacity materials such as PUF in upholstered furniture and carpets and ventilation rate, as with other SVOCs. Outdoors, fate is governed by air advection and water movement, because of the high solubility of OPEs.

335 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 in the water 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 LF2000-WQP, GREAT-ER and PhATE have been developed to address this issue. Many pharmaceutical monitoring studies have indicated that temporally significant fluctuations exist in rivers receiving urban wastewater. Currently, spatial models typically overlook 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 system. 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 indicates 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 highest levels of ibuprofen, naproxen, and loradaine, are temporarily transient. 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. Terzaqui, University of Insibria (Como) / Department of Science and High Technology, Como; M. Morselli, University of Insubria / Department of Science
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 volatilize and/or move into the e-waste contaminated 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 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 10 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 estimate the relative contribution of far-field and near-field routes in contamination at a regional scale. Furthermore, a spatially resolved version of a dynamic air-vegetation-soil model (SoilPlusVeg model) was used to predict a temporal emission profile from the city, verify if an emission source strength previously predicted for this city justifies soil contaminations in the surrounding area and, evaluate the importance of other sources and processes involved in the contamination of the area, to help establish if the mechanism how combined modelling 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; J.A. Arnot

Humans are exposed to polychlorinated biphenyls (PCBs) through “far-field” emissions from the ingestion of contaminated food from aquatic and terrestrial environments and “near-field” emissions 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-field and near-field routes to 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 more prominent 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.

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

D. Wenger, 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 European Rivers and Probabilistic Material Flow Analyses (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 polystyrene (EPS), 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 productions and 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

A. Koelmans, Wageningen University / Aquatic Ecology and Water Quality Management; C.M. Holmes, Waterborne Environmental Health Program; B. Nowack, EMPA

Plastic pollution is a growing 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 MP generation-sedimentation model integrated in a hydrological and particle transport model. A much larger scale model potentially suitable to simulate MP 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 trading of MP from WWTP point sources in US watersheds 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-fouyer MP, for diverse sizes, from 100 nm to 10 mm, a range that incorporates the theoretical parabolic size-settling 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 (~10,000 km²). Emission capitalization is used for all 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. Buoyant as well as the smallest non-buoyant MP fractions can be transported over long distances, reaching receiving waters such as the Great Lakes. In contrast, larger non-buoyant 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

R. Cross, C. Liddle, University of Exeter; T.S. Galloway, University of Exeter / Biosciences

Plastic contamination of freshwater sediments is well documented, and whilst quantitative measurement of plastic content in sediments is possible, little is known about plastic removal capabilities, recent models predict plastics < 1000 nm in size will be effectively retained in freshwater sediments. Current debate considers whether micro and nanoplastics can be defined as persistent organic pollutants (POPs) in their own right. In order for such a classification, four criteria must be met, one of which is for a compound to be bioaccumulative. Nanoplastics do not adhere to the classical concept of molecular bioaccumulation in sediments are by now assessed, however assessing their potential uptake and 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 are rapidly degraded in sediments and show both uptake from sediment exposures and from dietary uptake of a nanoplastic associated algal food source, with carboxylated and amminated plastics experiencing greater uptake than non-functionallised particles. Sediments on the other hand, reduced the availability of these particles for uptake into the worms, potentially through strong associations of the nanoplastics to solid constituents of the sediment. Ongoing work addresses the potential for formation of an “ecocorona” to alter the bioaccessibility of nanoparticles for the worms. These results will also be presented during the platform presentation.

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

C. Silva, CESAM & University of Aveiro; J. Pestana, CESAM & University of Aveiro / Biology; C. Gravato, Faculty of Sciences, University of Lisbon / department of Biology & CESAM

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 long-time available for benthic species. The objective of this study was to investigate developmental, and physiological effects induced by the presence of polyethylene microparticles (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 evalutation 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 gluthathione levels); oxidative damage (LPO); cellular energy allocation (CFA) and immune response (phenoloxidase). 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 imagoes 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 a depletion in 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 organization 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.

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

B. Tootoo, University of Bayreuth / Animal Ecology; I. Schrank, J. Dummert, A. Weig, C. Laforsch, University of Bayreuth

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 of rigid and flexible PVC or microplastics, to the surrounding medium. Therefore our results highlight, for the first time that imidacloprid has a more severe effect that might be attributed to the levels of DINP (distearoyl phthalate in addition to other microplastic particles. Therefore our results highlight, for the first time that differences in additive composition (absence or presence of a plasticizer) can lead to substantial differences in effects on aquatic species.

343 Poster spotlight: TU149, TU150, TU151

When ecotoxicology meets trophic ecology

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

E.L. Fernandes, University of Koblenz Landau; M. Bendschuh, 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 associated ecosystem processes. Some pollutants such as systemic 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 microcosm 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 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 leaches from plant material and may influence downstream food webs directly and indirectly.

345 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 and 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 timeseries 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.

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 at six Canadian lakes (Stream Lake, Lake Ontario, Source Lake, Happy Isle Lake, Lake Opeongo, and Lake Memphremagog) are used to determine feeding rates. Respiration rates were derived based on the routine metabolic rates and the population specific activity coefficients (multipliers). When comparing differently sized lake trout within a lake, larger fish tend to have the highest BAF, because they allocate less energy towards growth than smaller fish and have higher activity levels. When comparing fish from different lakes, diet composition and prey energy density become important in determining BAF in addition to activity and the amount of total energy allocated to growth. Specifically, fast growing Lake Ontario lake trout, feeding on slow growing alewife, have higher BAFs; while slower growing small lake trout in Happy Isle and Source Lakes have lower BAFs because they feed on invertebrates, which are low in the food chain. 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.

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

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Riparian and diets for lake trout systems and Canadian lakes (Stream Lake, Lake Ontario, Source Lake, Happy Isle Lake, Lake Opeongo, and Lake Memphremagog) are used to determine trophic position. Diet composition and prey energy density become important in determining BAF in addition to activity and the amount of total energy allocated to growth. Specifically, fast growing Lake Ontario lake trout, feeding on slow growing alewife, have higher BAFs; while slower growing small lake trout in Happy Isle and Source Lakes have lower BAFs because they feed on invertebrates, which are low in the food chain. 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.

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 DDEs, but not DDEs, are found in the Cooper’s Hawk. Biobio magnification of POPs occurs when lower trophic level organisms are consumed by higher trophic level organisms. This study examined the magnification of POPs in the Cooper’s Hawk. The study was conducted in the wild in the Greater Vancouver area of British Columbia, Canada. Cooper’s Hawks were captured and blood and tissue samples were collected at different sites in the surrounding area. The samples were analyzed for a range of POPs using gas chromatography/mass spectrometry (GC/MS) and gas chromatography/high resolution mass spectrometry (GC/HRMS). The results showed that the Cooper’s Hawk had the highest levels of POPs compared to other avian species. The study also showed that POPs were biomagnified in the Cooper’s Hawk, indicating that this species is at risk from POP exposure. The study highlights the need for further research on the effects of POPs on avian species and the importance of monitoring POP exposure in wildlife.
Toxicokinetic-toxicodynamic models as new tools for environmental risk assessment

S. Charles, University Lyon 1 / Laboratory of Biometry and Evolutionary Biology; V. Baudrot, Université Lyon 1; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team

Today, the Environmental Risk Assessment (ERA) for chemicals is based on fitting standard dose–response (DR) models to quantitative data. Such data are usually 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, which makes it difficult to extrapolate the results to more realistic situations, for example to effects under time-variable 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 LC/ECx, for arbitrary effect strength x 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 not many data to allow users to use TKTD 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 ‘morse’ 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 with Morse will then 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.

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

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Barbara; K. Tran, C. Vignardi, University of California Santa Barbara / Marine Science Institute; J. Means, University of California Davis / Environmental Toxicology; R.M. Nisbet, University of California, Santa Barbara / Marine Science Institute

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 adductome is lethally toxic may be more relevant than those with sublethal effects. The connection between AOP KEs to DEB processes through a model of damage dynamics. The model predicts regulated but increasing concentrations of damage as the result of toxicant exposure and also tipping points when damage outpaces regulatory feedbacks, leading to mortality (Klanjscek et al. 2016). The connection between damage dynamics and DEB will be influenced by empirical and theoretical observations, but potential linkages include damage causing an increased maintenance cost or specific impacts on development. Specifically, we are studying the effect of DLCs on Fundulus heteroclitus (Atlantic killifish). DLCs are of particular interest in this species due to the large intraspecific variability in sensitivity. There is extensive data describing the toxic effects of DLCs through the use of various models, such as the AhR pathway. The mechanisms involved, however, are not well understood. Further, sublethal effects of DLCs are less studied, but preliminary data indicate that 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.

354 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 derived toxicity 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

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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 hypothalamus-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 (biotransformed into the anti-estrogen 4-OH-tamoxifen), prochloraz (interferes with cytochrome P450, interacts with hepatic glutathione S-transferase activity, and is involved in biotransformation of many other chemicals), 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 / non-fertile eggs) exposures. The tamoxifen induced decrease in egg size translated to significantly smaller larvae at 20 dph. The HPOL model, guided by in vitro 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 infection. Each of these stressors might have effects on the response of fish to the other. Some attempts has previously been made 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 organ-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 Pomphorhyncha tereticollis. 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 parametrised based on published data. The model was then calibrated by MATLAB-based AMIGO modelling software for determining the rate of for Sciopterus 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-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

J. van Gils, DELTARES; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; h. baveco, Wageningen Environmental Research; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; I. Cousins, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); C. Lindgren, Stockholm University; S.S. Kutsarova, University of Zlatar / Laboratory of Mathematical Chemistry; S.D. Dimitrov, University of Zlatar / Dept of Comp Inform Technologies

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 the output for chemicals 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 E-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 model train is supported by a set of monitoring approaches, 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

A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; I. Cousins, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); J. van Gils, DELTARES; S. Birk, University of Duisburg-Essen / Aquatic Ecology, e. Peeters, Wageningen University / Aquatic Ecology and Water Quality; P. van den Brink, Alterra and Wageningen University; h. baveco, Wageningen Environmental Research

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 needed to achieve this goal. We developed 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. 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 Programme (Risks and Resilience (R4R), the HOPE project). Our 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 tens 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 parts of Europe compared with available monitoring information on the abundances of macroinvertebrates 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

L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; S. Birk, University of Duisburg-Essen / Aquatic Ecology; A. Burton, University of Michigan / School of Natural Resources Environment; D. De Zwart, DzEcotox / Centre for Sustainability Environment and Health; S.D. Yder, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; C.M. Holmes, K.E. Kapo, Waterborne Environmental, Inc.; D. van de Meent, Association of Retired Environmental Scientists ARES / Environmental Science; J. van Gils, DELTARES; M.C. Zijp, RIVM / Centre for...
This paper analyses 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 practices such as the EU’s Directive, Dz. Ecotoxicity, etc. Centre for Sustainability Environment and Health; M.F. Sanchez, Management; A. Foeks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; L. Globevnik, M. Koprivsek, University of Ljubljana / Faculty of Civil Engineering and Geodesy; J. Lennm, University of Duisburg-Essen; J. Mahnkopf, Leibniz Institute of Freshwater Ecology and Inland Fisheries; Y. Panagopoulos, National Technical University of Athens / Laboratory of Hydrology and Water Resources Management; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; K. Stefanidis, National Institute of Public Health and the Environment; M. Venohr, Leibniz-Institute of Freshwater Ecology and Inland Fisheries Water management requires solid understanding of how multiple stressors affect ecosystem state and services. The European project MARS (Monitoring Aquatic and early stress characterization of chemical mixtures up till 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.

359 Unravelling the cocktail of stress: toxics and other stressors impacting on the ecological status of Europe’s rivers S. Birk, University of Duisburg-Essen / Aquatic Ecology; V. Bremerich, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; O. D. Voort, Ecolox / Centre for Sustainability Environment and Health; M.F. Sanchez, Management; A. Foeks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; L. Globevnik, M. Koprivsek, University of Ljubljana / Faculty of Civil Engineering and Geodesy; J. Lennm, University of Duisburg-Essen; J. Mahnkopf, Leibniz Institute of Freshwater Ecology and Inland Fisheries; Y. Panagopoulos, National Technical University of Athens / Laboratory of Hydrology and Water Resources Management; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; K. Stefanidis, National Institute of Public Health and the Environment; M. Venohr, Leibniz-Institute of Freshwater Ecology and Inland Fisheries Water management requires solid understanding of how multiple stressors affect ecosystem state and services. The European project MARS (Monitoring Aquatic and early stress characterization of chemical mixtures up till 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 A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; A. Fischer, Utrecht University / Copernicus Institute of Sustainable Development; J. van der Hoek, Technical University Delft / Water Management Chemicals of emerging concern (CECs) in the water cycle have been the focus of research for over two decades. Chemicals used in current consumer goods and as derivate or removal efficiencies of various (advanced) drinking water and wastewater treatment technologies has been studied. Advanced water treatment technologies are based on sorption, (advanced) oxidation and size separation principles. The experimental settings in studies on the efficiency of these technologies are not homogeneous: technologies can be tested at bench-, pilot- or full scale, with different matrixes and different water matrices such as real or standardized surface water, ground water, drinking water or wastewater, the test chemical can be spiked or real environmental samples can be used, there can be variations in the application of the treatment e.g. dose, contact time or pore size, and variation in how all this is expressed; in mg/cm² or W/m² in case of UV oxidation, with Freundlich isotherms or removal percentage in the case of GAC etc. These variations and missing clarity therein hamper the interpretation and evaluation of the data concerning the removal efficiency of CEC of specific treatment technologies. In a previous study we found that stakeholders within the whole urban water cycle had sufficient information on CECs and their possible mitigation options, but that the relevance of the information often was unknown. A set of criteria describing what is important to know when evaluating removal efficiency studies can be helpful in this respect, with criteria for reliability and relevance where needed made explicit for the specific technologies to be evaluated. Examples of such criteria from the field of toxicology are available and well-used, e.g. to identify studies for the derivation of environmental quality standards in a scientifically sound way. Here we aim to highlight the current knowledge of the removal efficiencies with regards to CECs of (advanced) water treatment technologies both for surface water and wastewater. This to provide decision makers with the knowledge needed to make an informed decision with regards to which technologies will be relevant for their specific needs. To be relevant to end-users in water management the treatment technologies needs to be in use and commonly available. Not all advanced 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 powder activated carbon (PAC)), for (advanced) oxidation the use of ozone (O₃) and UV ± H₂O₂ and finally the use of nanoscale 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 dataset on removal efficiencies consisting of approximately 2000 entries, 93 compounds and 9 treatment technologies for wastewater (ozone, ozone + H₂O₂, conventional WWTP, UV, UV ± H₂O₂, PAC, GAC, NF, UF) and drinking water treatment (ozone, ozone + H₂O₂, UV, UV ± H₂O₂, PAC, GAC, UF, NF). 361 Future perspectives of chemical pollution and regulatory development J. Munthe, IVL Swedish Environmental Research Institute Ltd; T. Skårman, IVL Swedish Environmental Research Institute; E. Broström-Lundén, IVL Swedish Environmental Research Institute / Environmental Research Institute; M. Rahmberg, E. Westberg, IVL Swedish Environmental Research Institute; D. Bunke, Öko-Institut e. V. / Sustainable Products & Material Flows Division; K. Sackmann, OEKO Institute 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 alternative substances for use and apply new techniques such as plasma, PFCs, flame retardants or nanomaterials but may also introduce new substances with negative impacts on aquatic ecosystems. Four societal sectors have been identified where major changes within the next two decades can be expected which have potential consequences for chemical use and releases: public health, food production, urbanization and technologies. With these future developments and the resulting introduction of new substances from new sources, an adaptation of current regulatory frameworks is required. 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 solutions-focused approach, where the same approach is adopted to IVL 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) D.B. Jard, Environment Canada; R. Schafer, University Koblenz Landau / Institute for Environmental Sciences; C.B. Choung, Environment Canada/Canadian Rivers Institute / Department of Biology; A. Bush, Environment and Climate Change Canada; S. Bracewell, Wageningen University & Research / Department of Aquatic Ecology and Water Quality Management; A. Chardon, Macquarie University / Molecular Ecology and Toxicology; Z. Copson, Environment and
Climate Change Canada; K. Dafforn, Macquarie University / Evolution and Ecological Research Centre; E.L. 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, University of Guelph; N. O’Herlihy, Environment and Climate Change Canada; K. Simpson, R. Verdonckse, Wageningen University; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group.

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 ecosystems 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 and monitoring data and 8) Adjust the ecological model if needed. The workshop resulted in a “best useful conceptual template to assess and predict multiple stressor effects propagated through higher levels of biological organisation. In light of this, a workshop was held at Wageningen University and Research, the Netherlands, (September 2017) to determine the current state of knowledge of multiple stressor effects on aquatic ecosystems and to assess how these effects can be better predicted. The workshop was attended by experts from the Netherlands, Australia, Germany, and Canada and covered a range of ecosystem types considered to be at high risk from multiple stressors. The workshop resulted in a “best-approach” conceptual framework for assessing multiple stressor effects on aquatic ecosystems. The framework was subsequently applied to three case studies: harbours, agricultural drainage ditches, and floodplains. Here, we present the application of this framework to agricultural drainage ditches. Agricultural drainage ditches are an under-appreciated and undervalued habitat for a range of aquatic and terrestrial organisms. Although these man-made features can maintain high biodiversity in agriculture landscapes, they are often ignored for their conservation value and are not protected under the EC Water Framework Directive 2000/60/EC. Using the framework developed during the Wageningen workshop, we developed a conceptual food-web model using functional groups to assess known direct effects of stressors on ditch communities. We identified the most important stressors (nutrients, pesticides, dredging and mowing, salinisation, and siting) and impacts on communities and conducted a literature search for each stressor-functional group combination to identify sensitive and non-sensitive groups. We also reviewed the literature on experiments using at least two of the identified stressors and identified potential interactions. The conceptual food-web model was updated using this knowledge to capture interactions. Finally, the conceptual model and its predictions regarding the response to multiple stressors will be compared to large scale ditch biomonitoring data to assess the validity/predictive power of the model. The framework provides a useful conceptual template to assess and predict multiple stressor impacts as well as to unravel research gaps.

365 Macroinvertebrate communities across a gradient of multiple stressors from agricultural land use in Romanian streams V.C. Schneider, M. Link, S. Konz, E. Stoevs, University of Koblenz Landau; B. Vogler, B. Beck, Eawag, Swiss Federal Institute of Aquatic Science and Technology; K.P. Bates, M. Cimpean, Babes-Bolyai University; E. Vermeyissen, Ecotoc Centre Eawag-EPEL / Aquatic Ecotoxicology; H. Singer, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Chemistry; J. Hollander, Eawag / Environmental Chemistry; R. Schafer, University Koblenz Landau / Institute for Environmental Sciences. Pesticides from agricultural usage are one of the major drivers of biodiversity loss in freshwater ecosystems. Their entry pathways are mainly related to pesticide use in agriculture. To differentiate pesticide toxicity and other agricultural stressors, we conducted a field study in Eastern Europe (Romania), where agricultural intensity varies, ranging from high to low intensity (extensive) agriculture relying largely on animal labour (畜働農業). We assessed that, in contrast to pesticide toxicity, excessive nutrient and sediment input would be unrelated to agricultural intensity. Consequently, this would allow distinguishing effects from pesticides and these other stressors. We analysed the relationships between pesticide toxicity and other agricultural stressors. Additionally, we analysed combined and individual effects of these variables on the biodiversity, as well as on the composition of stream macroinvertebrate communities. We examined 19 low-order streams across a gradient of agricultural intensity in terms of average field sizes. Pesticide concentrations were investigated using two different passive sampling methods. Firstly, we used styrene-divinylbenzene (SDB) disks to sample hydrophilic compounds, which enabled the determination of approximate time-weighted pesticide concentrations in streams during heavy rainfall events. Secondly, we used polydimethylsiloxane sheets (PDMS) for the detection of lipophilic pyrethroids and organophosphates. The toxicity of the 88 detected pesticides was assessed using the sum toxic unit (sumTU). Stream macroinvertebrate communities were sampled twice, using a quantitative multi-habitat-sampling. This allowed the analysis of relationships between the community composition and diversity with a gradient of pesticide toxicity in interaction with additional agricultural stressors. The toxicity gradient originated from pesticides and nutrients (NH₄⁺) showed no relationship to the intensity of agriculture expressed as the average size of the adjacent fields. This indicates that pesticides and nutrients co-occur independently of agricultural intensity. How and to which extent, in terms of effect size, the communities are affected by the pesticide gradient and the additional presence of other stressors originating from agricultural land use will be presented during the conference.

366 Daily temperature variation determines the toxicity of a pesticide mixture V. Delnat, T.T. Tran, L. Janssens, KU Leuven / Biology; R. Stoks, University of Leuven / Department of Biology. Synergistic interactions between pesticides in mixtures and between pesticides and warming may improve the efficacy of vector control. Particularly, synergistic interactions between biopesticides and chemical pesticides would be promising as these could potentially result in the combination of efficacy of control, slowdown of resistance build-up and lower ecological damage. One understudied aspect of global warming is the increase in daily temperature variation (DTV). While DTV may increase the toxicity of chemical pesticides, it is unknown whether it also interacts synergistically with biopesticides, and magnifies the toxicity of pesticide mixtures. We tested whether DTV influences the toxicity of pesticides with a...
different mode of action (the chemical pesticide Chlorpyrifos, CPF, and the biocide 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 measured effects on larval body length, larval 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 biocide. Moreover, a large DTV changed the order of the Gramicidin 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.

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Screening of organic molecules in the aqueous environment
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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 polar compounds. 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) -2.5 to +2, and “non-polar” log D (pH 7) higher than +42. 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 methods such as “hidden-target screening” in complex environmental matrices (such as wastewater treatment plant effluents) which is complementary and 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].

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Removal options and transformations of persistent mobile organic chemicals during production of drinking water
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In recent years, drinking water contamination by mobile organic chemicals has been widely studied. The most abundant type of these contaminants are the persistent mobile organic chemicals (PMOCs) that are chlorinated and hydroxylated analogues of natural organic compounds and can be used to limit the presence of organic micropollutants in drinking water. However, removal by activated carbon and oxidation processes can be used to limit the presence of organic micropollutants in drinking water. High removal efficiencies were obtained for most of the PMOCs identified in drinking water using activated carbon, ozone and chlorine. Highly polar PMOCs such as isadamantan-1-amine (Log D = -2.34), trifluoromethanesulfonate (Log P = -1.35) and 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, aromatic amines, 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-diphenylguanidine and the 1,3-di-o-tolylguanidine, an olefinic sulfonate and an amine compound, the N-benzylidimethylene, 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 the PMOCs identified in drinking water are not toxic in Ischnura elegans damselflies. 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 CPF also increased the toxicity of CPF, providing novel evidence that CPFs can amplify the toxicity of pesticides. Furthermore, the increased toxicity of CPF by DTV was more pronounced at 24°C. This novel pattern is likely general to other pharmaceuticals and thus may be a factor that needs to be included in risk assessment of pesticides and their mixture in the real world.
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 benzozyatoyle, tolylzyatoyle and phenylurea, which displayed a passage of 25%, 17% and 10%, respectively. The data showed that removal of neutral polar MPW is mainly governed by size exclusion. For neutral and moderate polar MPs the inverse was true. In reverse osmosis water passage was well below 1% 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 charges on the membrane surface create repulsion resistances. 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 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. If these mobile organic contaminants (MOCs) are persistent (PMOCs) against microbiological and chemical degradation, their removal during water treatment and drinking water purification may prove difficult. Toxic PMOCs 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 PMOCs in the water cycle and only few (e.g. acylsulfame, glyphosate) have been extensively studied and monitored. 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 polarities. 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. and Schulze et al., we selected 15 industrial chemicals with a high expected potential to form transformation and studied their 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 Hansian water samples for the presence of these TPs. While some TPs were not detected, others were found. The majorities of all detected TPs showed potential to be PMOCs. The results of this study provide 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 that were in some cases developed 20 years ago with a more modest level of ambition and less scientific knowledge than we have today. Are these regulations really up to the task? This question is explored using the example of the POP screening assessment in the Stockholm Convention. Using perfluorinated alkyl acids (PFAs) and octamethylcyclotetrasiloxane (D4) as case studies, it is shown how this framework can lead to both false negative and false positive conclusions. False negative classification of PFAs can arise because of the inclusion of bioaccumulation as a screening criterion in the framework although bioaccumulation is not a requirement for adverse effects of chemicals in remote regions. False positive classification of D4 can arise because the four screening criteria (persistence, bioaccumulation, long-range transport, and adverse effects) are not valid in the same environmental media/compartments. It is concluded that if we wish to conduct POP assessment for the broad spectrum of chemicals in modern commerce, then we will have to rely less on individual screening criteria and instead apply models that can capture and integrate the broad diversity of chemical behaviour.

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 (VMP/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 neither 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 European 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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In life cycle assessment (LCA) the main focus is on damage assessments of product systems. These damages are conventionally characterized per so called functional unit. In practice, however, these functional units are partially descriptive, e.g. white light from a point source with 1500 lumen, and not well assessed. In the first part of this study we therefore further elaborate the functional unit concept. Using an example of a LED light bulb, we will show that the functional unit in this context is significantly different from the functional unit in terms of an aggregated single score for product benefit, such as the net impact on human well-being, would permit to compare all types of products. In the second part, we specify how a better product benefit characterisation could improve life cycle assessment and its policy support. Three advantages are specified. First, as these functional units are often not specifically defined and a product can have multiple functions, comparison is often impeded in practice.

When taking a closer look at the concept of functional unit, it is imperative to define what functionality implies. Products have been created to fulfill human needs, e.g. the need for light at night provided by a light bulb. Through fulfillment of needs, human well-being is induced. The characterisation of the actual functionality hence boils down to the assessment of this well-being effect. In consequential LCA, not only the consequences related to the chosen activities associated with a product life cycle should after all be considered, but also those associated with the benefit induced by the product. In the second part, we specify how a better product benefit characterisation could improve life cycle assessment and its policy support. Three advantages are specified. First, these functional units are often not specifically defined and a product can have multiple functions, comparison is often impeded in practice.
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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Diet is 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, 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 aligns publically available data from multiple databases with risk factor definitions from the GBD. Finally, for 6000+ food items we estimate the HEalt Nutritional Index (HENI), the total avoided health burden from all dietary risk factors per serving and 100 kcal. Nutritional CFs for food group 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 Frankfurt 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 the 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 consequences of climate change. However, food production is also responsible for an increased GHG emissions. In Peru, 12% 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. Feasibility and integration 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 aspects, 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 CO$_2$eq 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 so as 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 CO$_2$eq 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
A. Ernstoff, Quantis / Quantitative Sustainability Assessment; S. Humbert, X. Bengoa, M. Vargas Gonzalez, Quantis; O. Jolliet, University of Michigan

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 eat. 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 CO$_2$ 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 reduction of environmental impact. Our study looks into the monetary values of GHG, represented by CO$_2$ (or CO$_2$-equivalent), and their underlying cause-effect chains in three Life Cycle Impact Assessment (LCIA) methods LIME2, EPS2015 and ReCiPe2016. The damage cost for CO$_2$ 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 CO$_2$ 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 CO$_2$ cost evaluated by LCA, especially in studies where social and economic consequences are the major concerns. The CO$_2$ 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 Assessment of Human Health Benefits and Risks of Contaminated Sediment Remediation
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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 use of thin-layer 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 a first study, the method was repeatedly tested in real environmental conditions, and in the field under a monitored burial of highly mobile and persistent pharmaceuticals. The AC cap was able to significantly lower the concentration of aspirin, ibuprofen, and a broad range of other pharmaceuticals, thus demonstrating 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 incidents. 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 per wetland, respectively. Following treatment, highly mobile and persistent pharmaceuticals tend to release into surface water and groundwater by reclamation activities, producing high impacts on wildlife and the human population. Occupational fatal incidents are comparable to the combined benefits of MNA and the selected remedy scenario. Impacts associated with chemical inhalation exposures are less substantial, albeit not negligible. The quantitative framework of this study, when supplemented with adequate monitoring data, can provide valuable insight into the overall effectiveness of a given remediation in light of alternatives.

Possibility of using a genotoxic tests in planning precise phytoremediation of depleted soils enriched in organic amendments
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A major part of soils at many contaminated sites is characterized by a high biological activity and is prone to a mass loss due to the increase of anthropogenic activities. Increasingly, human industrial activity, our irresponsibility, and impunity have a negative influence on the condition of soils. Thus, the problem of soil contamination, especially with several nanoparticles, and multiple heavy metals refers mainly to the industrial areas. Majority of those contaminations are bioavailable, and they are deposited in plant tissue as well as in edaphon. Moreover, this problem is even more pronounced in many cases when the impacted sites are near natural ecosystems. The most frequent mechanism of contamination is related to the application of fertilizers, which contain a substantial part of heavy metals. In these situations, the use of phytoremediation techniques seems to be an effective tool in two aspects: the increase in the remediation efficiency due to the increased biological activity of phytoremediation plants, in the case of the application of effective species or a combination of them, and the use of phytoremediation for the potential reduction of the biological activity of heavy metals. The effectiveness of phytoremediation techniques requires the implementation of a series of conditions and technologies to minimize the negative impact on the environment. This study aimed to assess the possibility of using a genotoxic tests in planning precise phytoremediation of depleted soils enriched in organic amendments in order to limit the potential negative impact on the environment. The research was focused on the assessment of the long-term potential of the genotoxic tests in planning precise phytoremediation of depleted soils enriched in organic amendments.
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 pharmaceutical 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 behaviors 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 on the sorption of polychlorinated biphenyls in the soil environment. First, sorption behaviors of nineteen pharmaceuticals across four groups (neutral, 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 component 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 chlorodecone transfer to animals: interest of soil amendment

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Chlorodecone (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 biochars were selected to be amended into 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 concentration 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 transfert (iii) availability and bioaccessibility tests could be useful for 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 soils from the boundaries of landfills and agricultural lands 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, 156, 157, 167, 169, 170, 180, 189. Quantitative determination was carried out using chromatograph with electron capture detectors (ECD) equipped with glass capillary column with stable phase DB-5MS UI and the following parameters: 60 m × 0.25 mm i. d. × 0.25 µm. Special attention was paid to the total amounts of polychlorinated biphenyls, as the total amount of these compounds correlates 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 at one of the soil sampling sites (Dilijan Town, Tavush Province of Armenia) along with 3.5-fold exceeding the standard, almost dioxin-like PCBs were found.

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Associated Health Effects of Veterinary Pharmaceutical Residues in Livestock waters 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 acetylamphen (AC), dichlofenac(DP), salicylic acid (SA), tetracycline(TC), chloramphenicol(CHR), cinchophen(CIN), bishophenol A, 1,3-bis(4-chlorophenyl)-4-bis(4-chlorophenyl)vinvermin(V) from agricultural wastewater using the hydrophilic-lipophilic balance(HLB)-SPE column. Recoveries of the pharmaceuticals from standard aqueous solutions spiked concentrations of between 2 and 10 µg/l were: E2, 76.62 - 85.47 %; AC, 78.29 – 94.34 %; TC, 88.35 – 92.15 %; CHR, 76.62 – 88.35 %; SA, 79.38 – 81.49 %; E3, 85.42 – 92.15 %; CPA, 80.27 – 89.42 %; CP, 76.58 – 90.21 %; DP, 75.46 – 87.55 % and IV, 80.27 – 84.89 %. Various levels of veterinary drugs - AC, < 0.48 – 1.07 µg/l; SA, < 1.37 – 15.49 µg/l; TC, < 3.45 – 4.57 µg/l; CP, 0.45 – 2.46 µg/l and IV, < 1.74 – 1.63 µg/l were detected in the grab water samples. The results of the health risk assessment clearly showed mutagenic activity being observed in samples from sheep and poultry farms. It also showed higher estrogenic 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 agriculture 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 receptor for micropollutants in general. It is thus necessary to consider pesticide inputs to water bodies. Theories of pesticides can be quite expensive and inputs may not be clearly identified or collectible, and as a consequence inputs but are still significant because of lack of treatment on these effluents and a potential increase of concentration around the
Benzophenone-3 (BP-3) is a widely used organic UV filter to protect humans and materials from damage by UV irradiation, and owing to its hydrophobic properties, it could potentially bioaccumulate in aquatic biota, including fish (tens to thousands of ng/g). BP-3 can undergo both photodegradation or phase I and phase II metabolism, generating transformation products and metabolites.

Environmental risk assessment approaches often require information on the free concentration in water, biocumulation 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 μg/mL concentration level. BP-3 was detected in all the analysed samples, with the highest concentrations at day 14. Bile concentrations were significantly higher than skeletal muscle in comparison to the rest of tissues/fluids. Since BP-3 is hydrophobic and non-ionicizable compound, the lowest concentrations of BP-3 were found in plasma. Although liver tissue (highly lipoidic) 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 lipoidic) 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 seawater 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 (ED431/C31/2013/36) and FEDER/ERDF. H. Ziarrusta is grateful to the Spanish Ministry of Economy and L. Mijangos to the Basque Government for their predoctoral fellowships.

Phyto remediation is an emerging technology that utilises green plants and their associated rhizosphere microorganisms to clean polluted environmental media. However, the role of plants in removing organic pollutants is still not well understood. Phyto remediation of realistic environmental concentration (10 μg L⁻¹) of the chiral pesticides tebuconazole and imazalil by a wetland plant, Phragmites australis, was investigated. The experiment was carried out in a growth chamber using plants of similar initial size (6.0 ± 0.2 g fresh biomass). The plants were placed in 700 mL glass vessels containing 500 mL hydroponic solution. The pesticides were spiked separately (n=27 for each) in parallel with control samples (n=15). The experiment ran for a period of 24 days. Enantioselective fractions and translocation products (TPs) in both hydroponic growth solutions and plant tissues were measured by HPLC-MS/MS. The uptake, translocation and metabolism of tebuconazole and imazalil inside Phragmites australis were documented for the first time using enantioselective analysis. The pesticides removal efficiencies from water were 96.1% and 99.8%, respectively, by the end of the experiment (day 24). Removal from the solutions could be described by first-order removal kinetics (k=0.14 d⁻¹ for tebuconazole and k=0.31 d⁻¹ for imazalil). Four different processes occurred simultaneously: 1) removal of the pesticides from the hydroponic solution, 2) plant uptake, 3) pesticides translocation in the plant, and 4) degradation within the plant. Tebuconazole and imazalil concentrations inside Phragmites showed a maximum level at day 10 and 5 days, respectively, followed by a decrease of both compounds concentration. Two TPs of tebuconazole could only be quantified in solution, while two imazalil TPs were quantified in both solution and plant tissue. The uptake of both pesticides was positively correlated with evapotranspiration. The removal of imazalil and tebuconazole from the hydroponic solution was not enantioselective, however, both translocation and degradation inside Phragmites were enantioselective. For tebuconazole, the enantioselective degradation was found in both Phragmites roots and shoots.

Environmental Risk Assessment of Active Pharmaceutical Ingredients used in Human Medicinal Products: Europe-wide Variation in Risk Quotient J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; S. Owen, AstraZeneca / Safety Health Environment; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; B. Verbruggen, University of Exeter; L. Gunnarsson, University of Exeter / Biosciences This presentation will describe the total consumption-based environmental risks posed by 130 human medicinal products and the impact of mode of action, lipophilicity and dilution on these risks. In accordance with Article 8(3) of Directive 2001/83/EC, as amended, a new marketing authorisation application shall be accompanied by the evaluation of the potential environmental risks posed by the human medicinal product. These environmental risk assessments (ERAs) estimate the potential environmental impact on a product-by-product basis rather than a substance-by-substance basis. In the cases where an active pharmaceutical ingredient (API), or substance, is used to treat multiple clinical diseases, there is the potential to under-estimate the impact on the environment. The UK Environment Agency (EM) guidance for the environmental risk assessment of human medicinal products has been in place now for over 10 years. The introduction of this guidance marked a step change in the ERA requirements for human medicinal products, with a shift from short-term acute to long-term chronic environmental effects assessment, and tailored ERAs for active pharmaceutical ingredients (APIs) with suspected or known reproductive effects. To determine the total substance or API risk, we have: (i) identified and collected definitive published no observed effect concentrations (NOECs) for available APIs (excluding anti-infectives and anti-parasitic products); (ii) collated human consumption data for each of these APIs in European Countries where these products are licenced for use; (iii) conducted a worst case exposure assessment (predicted environmental concentration (PEC) and predicted environmental concentration (PEC) / no observed effect concentration (NOEC) to assess the variability in the risk quotients (RQs) for each API across Europe and (v) looked at the impact of country-specific dilution factors applying the 5th percentile and

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393 Estimation and prioritization of hospital API emissions
A.M. Ragas, Radboud University / Department of Environmental Science; C. van Laren, M. Galpen, K. Tippecat, Radboud University; R. Oldenkamp, Radboud University / University of Nijmegen; C. Nijkerk, Environmental Analytical Chemistry, Center for Applied Geoscience; 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 speediQ® 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 isopropyl 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
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Active Pharmaceutical Ingredients (APIs) are consumed in large quantities, and end products 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 estimation of environmental concentrations of APIs in European surface waters based on country-specific per capita consumption data. The starting point for modeling the environmental fate of APIs is country-specific per capita consumption data. Subsequently, the modeling chain follows the steps of excretion into the sewerage system, transport to and fate in WWTPs, emission into surface waters and, finally, environmental transport, partitioning and degradation. Unique features of the model include the extensive location-specific information about European WWTPs, the flexibility in modeling Europe’s hydrology and accounting for ionizing properties of APIs. The model was validated using several studies reporting API concentrations in the Rhine basin. API-specific data and characteristics (e.g., physicochemical properties and consumption data) were obtained from the literature. Site-specific and API-specific measurements were directly compared to estimated water concentrations at the relevant locations in the river network. From the results shown for the Rhine basin and preliminary results of some additional basins, it can be concluded that estimations can be made with great spatial and quantitative accuracy. However, model performance depends on factors such as the allocation of country-specific consumption over relevant WWTPs, accuracy of the estimated hydrology, provided consumption data and API-specific characteristics.

395 Occurrence and fate of the anti diabetic metformin and its transformation products
S. Töbler, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tuebingen / Analytical Environmental Chemistry; C. Zwiener, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tuebingen / Geosciences
Effluents of municipal wastewater treatment are major entry pathways for pharmaceuticals and their transformation products (TPs). A wide-spread compound is the anti diabetic 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-dihydroxy-1,3,5-trizae (4,2-AMT), 2-amino-4-methylamino-1,3,5-triazae (2,4-AMT), 2,4-diamino-1,3,5-triazae (2,4-DAT) and methylbiguanide (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 % at an average influent concentration of 24 μg/l. GU concentrations were in the inffluences 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 trends to verify if a PEC of a non-identified metabolite exists, 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 in the WWT effluent samples. The study is performed within the project “Effect-Net”, funded by the Ministry for Science, Research and Art, Baden-Württemberg.

396 Development of biotransformation half-life QSAARs and PBT assessment models for Pharmaceuticals and Personal Care Products
E. Papa, A. Sangion, University of Insubria / 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 Insubria / Department of Theoretical and Applied Sciences (DiSTA)
Active pharmaceutical ingredients (APIs) are consumed in large quantities, and end products 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 estimation of environmental concentrations of APIs in European surface waters based on country-specific per capita consumption data. The starting point for modeling the environmental fate of APIs is country-specific per capita consumption data. Subsequently, the modeling chain follows the steps of excretion into the sewerage system, transport to and fate in WWTPs, emission into surface waters and, finally, environmental transport, partitioning and degradation. Unique features of the model include the extensive location-specific information about European WWTPs, the flexibility in modeling Europe’s hydrology and accounting for ionizing properties of APIs. The model was validated using several studies reporting API concentrations in the Rhine basin. API-specific data and characteristics (e.g., physicochemical properties and consumption data) were obtained from the literature. Site-specific and API-specific measurements were directly compared to estimated water concentrations at the relevant locations in the river network. From the results shown for the Rhine basin and preliminary results of some additional basins, it can be concluded that estimations can be made with great spatial and quantitative accuracy. However, model performance depends on factors such as the allocation of country-specific consumption over relevant WWTPs, accuracy of the estimated hydrology, provided consumption data and API-specific characteristics.

397 Predicting spatial and temporal variability in internal concentrations of antimyrtine in invertebrates within an urban catchment
A. Giorgis, University of York; A. Agatz, IBACON GmbH / Environment Department; L. Carter, University of York / Environment Department; E. Burns, University of York; M.D. Nunez, University of York / Environment; J. Wilkinson,
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 chemicals (APIs) currently in use are ionisable and may become charged at environmentally relevant pHs. Recent research has shown that the accumulation of a molecule in aquatic invertebrates or plants 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 the pH effect on accumulation in relation to pH. Here, we present this modelling approach to derive toxicokinetic parameters from laboratory experiments for the accumulation of amitriptyline, an antidepressant compound, in L. variegatus. Toxokinetic (TK) parameterisation and the underlying experiments involved the measurement of uptake of amitriptyline into L. variegatus at four medium pHs (5.5, 7, 8, 9). To simulate accumulation at the landscape scale, we used the generated toxicokinetic parameters in combined with measured monthly concentrations of amitriptyline in river water and associated water pHs obtained from a one year long monitoring study along the two rivers (rivers Ouse and Foss) in the City of York, UK. Data from the experiments at pH 5.5 and 9 were used to successfully parameterise the TK model. For two pH values (pH 7 and 8) we were then used to predict uptake and depuration rates for the neutral and ionised species of the API. Use of the derived rate constants to simulate the accumulation at the two intermediate pHs revealed that the approach underpredicts the actual accumulation by a factor of 2-4. Predictions of internal concentrations of amitriptyline in L. variegatus varied by 6 times across the monitoring sites and over the year. Generally, internal concentrations were predicted to be much higher for the river Ouse (which had a concentration range of 0.52-2 pmol/g and a pH range of 7.3-8.45) than the river Ouse (which had a concentration range of 0.2-0.95 pmol/g and a pH range of 7.4-8.44) and accumulation increased during the course of the year. Even though the model underestimated the internal concentrations by a factor of 2 and 4 in laboratory experiments conducted at pH 7 and 8 respectively, the general approach worked reasonably well to obtain internal concentrations. The study reveals important insights into the accumulation of pharmaceuticals by non-target organisms which will help to fully understand the risks posed by pharmaceuticals at the catchment scale.

Plants: predicting and assessing direct, indirect effects and recovery of plants from chemical stress

398 Waterplants in Risk Assessment - Selection of Potential Plant Species - Impact of Different Test Guidelines G. Gonsior, Eurofins AgroScience Services Ecotoc GmbH When results of standard laboratory tests show an unavoidable high risk, aquatic higher tier tests are needed to reduce uncertainties. In case there is a high risk for aquatic plants additional species could help to reduce these uncertainties for risk assessment by performing Species Sensitivity Distribution (SSD) tests. However, it is not clear which criteria are used for selection of test species and which guideline is considered for adaptation of non-standard species. Most tests were performed based on the Lenna guideline OECD 221, the two Myriophyllum spicatum guidelines OECD 238 and OECD 239, the ASTM E1913-2012 for Myriophyllum sibiricum, Sediment contact test with Myriophyllum aquaticum (ISO/DIS 16191) and the principles of the method proposed by the AMRAP (Aquatc Macrophyte Risk Assessment) working group (Nita et al., 2010). Further, the proposed ring test protocol for the emergent macrophyte Glyceria maxima was presented by Jo Davies et al. In addition a number of forms and reactions of plants were presented. Based on the EFSA opinion, designs will be discussed and an overview will be given on how the test designs can be further adapted to provide a refined risk assessment.

399 Applying the EFSA Scientific Opinion on NTPP: Testing non-crop species and the reproductive capability of selected species under greenhouse conditions A. Duffner, D. Ripperger, C. Groening, P. Mack, Eurofins AgroScience Services Ecotoc GmbH; S. Knaebe, EAS Ecotoc GmbH / Ecotoc Field; T. Moser, Eurofins AgroScience Services Ecotoc GmbH Agriculture is the dominating land-use of the EU member states by covering nearly half of the surface area. Using herbicides to reduce weed competition in agricultural areas can adversely affect non-target terrestrial plants growing at field margins. According to the recent EFSA Opinion for non-target terrestrial plants (2014) one important goal is maintaining the biodiversity of plant species in the agricultural area. It is therefore recommended to include also non-crop species in the testing scheme from the list provided in OECD guidelines (OECD 208 and 227) to assess the life-cycle with flowering and seed production. The objective of this study was to assess the viability of generative traits of non-crop species for risk assessment. For this purpose generative traits were evaluated if they provide more relevant information for the risk assessment. For this purpose they were compared with the vegetative traits, such as mortality and biomass production, which are currently assessed in the OECD guidelines 208 and 227. The selected non-crop species are included in commercially available seed mixtures for flowering strips. Our experimental design consists of one control and four different herbicide application rates, with 6 replicates. The field rate was chosen to reach the highest possible concentration of the affected crops and phenotypes in the sensitivity range. 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). Presentations (SETAC Europe 2017). References: EFSA PPR Panel (2014). Scientific Opinion addressing the state of the science on risk assessment of protection products for non-target terrestrial plants. EFSA Journal 2014;12(7):3800, 163 pp. OECD (2006). Test No. 208: Terrestrial Plant Test: SedimentEmergence and Seedling Growth Test. OECD Publishing, Paris. OECD (2006). Test No. 227: Terrestrial Plant Test: Vegetative Vignour Test. OECD Publishing, Paris.

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

L. Knaebe, University of Potsdam / Plant Ecology and Nature Conservation; S. Heine, Bayer Ag / Effect modelling; C. Mihan, Bayer CropScience AG / Ecotoxicology; T. Preuss, Bayer Ag / Environmental Safety; F. Jeltsch, University of Potsdam Ecological models are rarely found in terrestrial plant ecology 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 important for their credibility as 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 EFSA are on population and community level. Reuter and Siemenet-Gast (2007) performed an experiment that includes not only the test of monocultures but also the test of small artificial communities consisting of 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 Siemenet-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 and the model can serve as an important tool for predicting potential impacts on natural environments with information obtained from greenhouse studies on individual species.

401 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 a day after a herbicide is applied, 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 leoma studies, an aliquot of cells can be transferred to untreated media at the end of a test and after several days, the growth rates of the affected groups may approach that of the controls indicating recovery. Recovery in terms of growth rate of these simple aquatic plants is likely to be indicative of a population’s ability to sustain itself. In more complex mesocosm studies the concept of recovery is even further
complicated by seasonality, changes in nutrients, recolonization, 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, lemmna, 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.

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Aquatic primary producers and plant protection products: endpoints and level of protection achieved in the first tier of the risk assessment scheme

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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 (EcB50). 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 ErC50s 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 Lemna 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 EC50, (EC50, EC20 etc.) and assessment factor would ensure an adequate level of protection. Recommendations are provided for an optimization of the risk assessment.

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Poster spotlight: WE152, WE153, WE154

Environmental monitoring of contaminants using terrestrial ecological biomonitor

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Persistent Organic Pollutants in Germany: Results from the 2015/2016 moss and OdE-sampling

A. Dreyer, Eurofins GfA GmbH / Air Monitoring; S. Nickel, University of Vechta / 2; J. Koschorreck, Umweltbundesamt; W. Schröder, Universität 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 dibenzo dioxins and furans (PCDD/F), 18 polychlorinated biphenyls (PCB), 16 polycyclic aromatic hydrocarbons (PAH), 17 perfluoroalkyl substances (PFAs), 3 isomers of hexabromocyclododecane (HBCD), 7 polychlorinated biphenyls (PCB), 24 brominated 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, dT-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 Soest (forestry) and Scheyern (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 (only) driving force determining the POPs burden in moss samples. PBDE 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 sampling devices and bioaccumulators such as moss. Within the European moss survey programme, since 1990 every five years moss has been sampled at 7500 sites throughout Germany. Moss samples from forests were evaluated for concentrations of 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. The long-term information on the percentile statistics of bioaccumulation of atmospheric deposition in moss is essential for further scientific evaluation as well as for measurements and reporting of nature protection and environmental risk assessment. References [1] Nickel W (2017) Reorganisation of a long-term monitoring network using moss as biomonitor for atmospheric deposition in Germany. Ecological Indicators 76:194-206. [2] Schröder W, Nickel S, Völksen B, Dreyer A (2017) Nutzung von Bioindikationsmethoden zur Bestimmung und Regionalisierung von Schadstoffen in den deutschen Bundesländern. Poster presentation at the SETAC Europe 28th Annual Meeting Abstract Book.

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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. Schlösser, University of Hohenheim / Core Facility Hohenheim; E. Menaut, 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. Here we discuss results from the 3rd round of reference [1] to measure heavy metals and nutrients. Analysis of leaves and pine 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, Zn, Co, Cd, P, K, Mg, As, Cd, Cr, Zn, S, N, 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 were less pronounced indicating that deposition of dust does not play a great role in the area. On the other hand, we preliminary analyses showed significant differences in element concentrations between different species, age classes, and samples which were due to the availability, translocation, accumulation or growth regulation 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 restoration.

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Examining historical trends in diet and contaminant exposure in bats using bat guano 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;
Bats are excellent ecological indicators owing to their long life span, global distribution, and predictable responses to environmental stressors. Bats play important roles in pollination, seed dispersal, and insect population control and thus it is important to determine whether bat diets change over time as a result of exposure to contaminants such as metals. Bat guano deposits are of particular use as they may serve as a potential environmental archive for the cave environment preserving stable isotope and metal concentrations which allow for the determination of historical exposure to contaminants as well as any fluctuations in diet. This research provides the rare opportunity to examine two bat guano deposits from Jamaica: bat guano was heavily mined for gun powder and fertilizer and as such, there are few bat guano deposits that have been unaltered by human exploration or exploitation. The objective of this research is to reconstruct historical changes in diet and contaminant exposure to bats in order to better understand how anthropogenic activity affects these high trophic level mammals. We constructed the $\delta^{13}$Pb, $\delta^{13}$Cs, and $^{14}$C dating profiles in both bat guano deposits: this revealed that one of the deposits is over 3,000-years-old. We constructed the $\delta^{13}$N, $\delta^{15}$N, and $\delta^{15}$S profiles in order to determine the long-term dietary trends in the bat guano deposits. Preliminary results suggest that the 3,000-year-old bat guano deposit is tracking a change in stable isotope associated with the agricultural history of Jamaica. Specifically, the introduction of: nitrogen fertilizers, the Bordeaux mixture, and agucarane. We also examined the sterol profiles in the bat guano deposits for the purpose of determining more specific dietary information. Recent peaks in cholesterol and stigmastanol, for example, could be evidence of fluctuations in feeding habits. The second deposit is associated with mining and production. Lastly, we present the decrease in $^{13}$C within the bat guano deposit in association with the introduction of leaded gasoline.

408 Perfluoroalkyl substances and metallic elements in South African dragonflies
H. Boulton, North-West University / Unit for Environmental Science and Management; V. Leshc, North-West University; Y. Shiiba, National Institute for Environmental Studies / Fellow; A. Kinoshita, National Institute for Environmental Studies

Adult dragonflies are aerial predatory arthropods that occur globally. However, no research on adult dragonflies as potential indicators of environmental metallic electron transfer perfluorinated substances (PFAS) and elevated concentrations of environmental metallic elements are toxic to organisms and can cause disruption of biological processes. Adult dragonflies were collected and analysed for PFASs and metallic elements. The results indicated that dragonflies from farming areas had significantly lower ZPFASs concentrations than sites located closer to industrial areas (median $\Sigma$PFASs 0.32 ng/g wet mass) for Nong Khai (0.3 ng/g for South). Adult dragonflies perfluorocarboxylic acid (PFOS) occurred at similar concentrations at all six sites, when quantifiable, but PFOS dominated in the Southern sites. The highest median concentration was from Bloemhof Dam ($\Sigma$PFASs = 21 ng/g wet). This is known to be polluted by PFOS. The results also indicated that all species of dragonflies, regardless of body size and habitat type preference are suitable indicators of environmental metallic elements. Sites located near wastewater treatment plants had elevated concentrations associated with mining and industries. Dragonflies from sampling sites near potential pollution sources that seemed to have isolated water sources, showed lower metallic element concentrations when compared with other sites. Based on these results we conclude that dragonflies would be excellent indicators of environmental metallic elements and PFASs.

Bioavailability of Arsenic and Antimony co-contamination to vegetable crops in agricultural soils
L.P. Eyedawawa, University of Wollongong / School of Chemistry; A. Holland, La Trobe University / Centre for Applied Geochemistry, University of Wollongong / School of Veterinary Science and Medicine Antimony (Sb) is an emerging contaminant that is assimilated to behave in a similar way to arsenic (As). Sb and As often occur because of mining. Bioaccumulation and phytotoxicity of As is well studied, but there is little evidence on Sb and its interactive effects with As. Metalloid accumulation in agricultural soils may present health risks and hazards to humans and ecosystems through direct ingestion or contact with contaminated soil and food, a reduction in food quality and marketability via phytotoxicity and reduction in land usability for agricultural production. Plant bioassays allow inferences regarding the potential toxicity of contaminants. The phytotoxicity in the contaminated soils is governed by the bioavailability of the contaminant, which in turn is influenced by soil physical and chemical characteristics, contaminant speciation and the species of plant. However, it is still unclear the impacts of ageing of agricultural soils have on the co-contamination of As and Sb on alternation of crops. Our study evaluated the potential use of vegetable crops to identify and assess the bioavailability and toxicity of As and Sb in co-contaminated soils using bioassays. Water spinach and choy sum are herbaceous leafy vegetable belonging to the morning glory (Convolvulaceae) and mustard (Brassicaceae) families, respectively. Our study compares the soil characteristics in terms of total and bioavailable metal fractions with plant accumulation and toxicity data. Plant toxicity parameters (tissue biomass and lengths) were used to evaluate impacts of contaminant exposure on plant productivity. This information was used to understand the tolerance of plants grown in As and Sb contaminated soils, and the risks associated with As and Sb co-contaminated soils. This was done as single element and mixed metal exposures. Test soils were silty sand and slightly acidic. Bioavailable As and Sb in soils increased proportionally with total metal concentration. A clear increase in the tissue accumulation of As and Sb was observed with increasing bioavailable metal fraction for both individual (As and Sb) and combined (As+Sb) treatments. Vegetable productivity decreased when grown in As only and As+Sb combined contaminated soils. Sb contamination in agricultural soils poses a greater human health risk and hazard than As only and As+Sb co-contamination, because Sb accumulates in edible crops with no observed phytotoxicity or reduction in the vegetative productivity.

410 Transcriptomic responses of the endangered freshwater mussel Margaritifera margaritifera to trace metal contamination
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The freshwater pearl mussel Margaritifera margaritifera is one of the most threatened freshwater bivalves worldwide. In this study, we aimed 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. The effect of metal contamination on the transcriptome of Margaritifera margaritifera specimens was examined 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-seq. Correlations between transcription levels of each single transcript and the bioaccumulation of 9 trace metals, age (estimated by sclerochrono) and condition index were determined in order to identify genes likely to respond to a specific factor. Amongst the studied metals, Pb, Zn, Cd and Hg 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 more deeply, gene expression 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-CRRMS-based metabolomics to highlight biofragmentation products and effects of difenofuran in Mytilus galloprovincialis
F. Courant, Université de Montpellier - UMR 5690 Hydrosciences / UMR Hydrosciences; B. Bonnefille, H. Fenet, Université de Montpellier / UMR Hydrosciences; E. Gomez, Université de Montpellier, Hydrosciences Montpellier UMR 5690 / UMR Hydrosciences

Difenofuran (DFC) has become a major contaminant of interest as shown by its inclusion in the EU Water Framework Directive (2015/49/EU). However, relatively little is known regarding its biofragmentation and effects in the Mediterranean mussel. Environmental metabolomics affords several advantages to study both topics. The metabolome refers essentially to i) the “endometabolome”, constituted by endogenous metabolites, and to ii) the “ xenometabolome”, in reference to xenobiotics and their biofragmentation products [1]. Metabolomics profiles acquired through mass spectrometric techniques may reveal the exposure by direct detection of xenobiotics and their metabolites (xenometabolome investigation) and effects by the detection of endogenous metabolites which concentrations may differ from physiological levels following the exposure. The metabolome investigation is a powerful tool to assess if the xenometabolone investigation is feasible, an experiment was carried out whereby marine mussels were exposed for 7 days to ethanol (≤ 1°/oo, vehicle) or to 100 µg/L DCF. Analytical methods relying on Liquid Chromatography-High Resolution Mass Spectrometry were developed to generate metabolic profiles from mussel’s tissues. The obtained profiles for both groups (controls and exposed) were compared. We highlighted DCF and 13 DCF metabolites in exposed mussels. Three of them were phase I metabolites such as...
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 mussels have never been described despite being of concern for these organisms. Atyelecaminol and serotonin are involved in osmoregulation, and in gene expression in mussels [2-4]. Our results highlighted potential impairment of mussel 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].


412 Metabolomics used to link affected molecular pathways with behaviour outcomes after a single dose of pesticide exposure in meso P. Leonards, VU University, Institute for Environmental Studies / Department of Environment and Health; H. Viberg, L. Lee, B. Buratovic, P. Eriksson, Uppsala University
Worldwide, serious concern has been shown 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 "Developmental Neurotoxicity Assessment of Mixtures in Children" invests in 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 (subclinical) 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 under development 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 metabolomic 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, Environment Of Nunavut / Nunavut Department of the Environment; B. Chnadamouli, J. Cosgrove, SGS AXYX
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 changes in the metabolome 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 broad series of concentration levels) from the Hudson Bay Polar Bear Study (HB; n = 14) and Western Hudson Bay (WHB; n = 15) male polar bears were collected for multivariate and univariate statistical analyses. Correlated compounds and significantly different or impacted physiological pathways were identified that may be related to differences in POP exposure or other environmental factors. Partial least squares discriminant analysis (PLS-DA) and variable importance in projection (VIP) were applied to the combined metabolite-contaminant profiles of these polar bears, and Spearman correlation analyses were used to establish relationships between metabolites and contaminants, as well as with other biological factors. Forty-one metabolites [membrane lipids, acylcarnitines (ACs), and symmetric dimethyl arginine (SDMA)], and 21 POP discriminated the subpopulations. Perfluorinated alkyl substances (PFASs), polybrominated diphenyl ethers (PBDEs), p,p'-dichlorodiphenyldichloroethylene (DDE) and some ortho-polychlorinated biphenyls (PCBs) were greater in the SHB bears and changes in the metabolite concentrations had some consistency with previous laboratory studies. Arachidonic acid (ARA), glycerophospholipid and amino acid pathways were 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 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.

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 I Leiria; 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 Remote Environmental Scientists (ARES) / Department of Ecological Sciences; M.F. Lemos, Instituto Politecnico de Leiria / MARE I Leiria
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 species and species 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 (Bravo5000®), 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, two experiments were performed, the first one involving the experimental soil and the second one involving the natural agricultural soil. To find the reproduction EC50, several dilutions of the formulation were spiked according to nominal concentrations of the active ingredient. For the mechanistic assessment of effects, and to better understand the correlations between omics information through time, organisms were then 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 fungicide formulation on gene expression (by qPCR), partly by pathways involved in detoxification (induction of phase I and II enzymes) and also involving glutatione, 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 series set when investigating the effects of chemicals. This integrative approach may be thus provide useful insights, exhibiting their relevance in toxicological studies and proving the importance of a time-series analysis in correlations between these datasets.

415 Using functional genomics to find mechanisms of herbicide toxicity in Chlamydomonas reinhardii A. Zupanic, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; A. Betz, Eawag / UTOX
At present, environmental risk assessment of chemicals is limited to measuring physiological endpoints in model species. To test all chemicals that require testing, a shift to mechanistic-based testing is needed. However neither direct molecular targets nor the stress pathways that lead to adaptation to chemical exposure are usually known. Finding the genes encoding sensitivity or tolerance to chemicals is one of the highest priorities of the (eco)toxicological research community. One of the best method for gene function discovery is functional genomics based on metabolic profiling. Single-gene mutants and their whole genome knockouts (such as chemical exposure) of interest and mutants which are the most susceptible to the intervention and those that are most tolerant are found. We exposed a pooled library of loss-of-function mutants of Chlamydomonas reinhardii to three herbicides that target photosynthesis: atrazine and diuron which target photosystem II, and which disrupt the transition of electrons from photosystem I to photosystem II which leads to production of reactive oxygen species. The pooled library was exposed to 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 become enriched or have disappeared from the culture after exposure (compared with non-exposed control). The expectation was that the functional genomic profiles of atrazine and diuron would be similar, as the target of both chemicals is the same, while a different profile would be obtained for paraquat. This was indeed the case, with the profiles for both diuron and atrazine enriched for mutants of genes involved in the photosynthesis. The profiles of paraquat also included photosynthetic genes, but also several genes involved in defence against oxidative stress, where high levels of gene expression were observed in the paraquat-sensitive cells.
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.

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

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, governments, 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 biodiocator 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 derived 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, biodiocator 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 biodiocator 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 GES, 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 10D3C), 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 10D4C). For these two indicators, Member States shall establish that list of species to be covered, the thresholds and 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.

**418 Addressing the growing threat of marine litter: NGOs essential role in strengthening the science-policy-society interface**

T. Vlachogianni, Mediterranean Information Office for Environment, Culture and Sustainable Development (MIO-ECSDE)

The growing urgency and complexity of interconnected societal challenges, such as marine litter, demand that they be addressed through the strengthening of the science-policy-society interface so as to provide the necessary conditions for translating research-based knowledge into effective action. NGOs are essential partners in promoting environment and nature conservation development. Their active participation at local, national and transboundary level in all phases of projects and processes, from their design, implementation in the field, operationalization, monitoring and evaluation, contributes not only to increased transparency, wide visibility and outreach of the project or process, but also to enhanced overall quality and increased ownership of the outcomes, as well as amplified possibilities for replication of its activities. In full acknowledgement of the prominent role of NGOs in the realm of environmental governance, MIO-ECSDE, a Federation of some 130 Mediterranean NGOs working on Environment and Sustainable Development, in fulfilling its vision and mission, has developed and implemented a number of actions on the science-policy-society interface that address the growing threat of marine litter in the Mediterranean, ranging from the monitoring and influencing of relevant policy, all the way to hands on and pilot activities (e.g. within the framework of the IPA-Adriatic DeFishGear, the FP7 MARLISCO, the Interreg Med ACT4LIFTER, the EU SWIM-H2020 SM, etc.). How marine litter and its inherent environmental, economic, social, political and cultural dimensions have been tackled by MIO-ECSDE illustrates the broad extent of involvement and interventions required for the protection of the marine and coastal areas. The term “biodegradable” could be misunderstood and induce the public and particular strengths that the NGO community brings to environmental governance, such as leadership, creativity, flexibility, entrepreneurship and capacity for vision and long-term thinking. As scientists call for more research on global environmental changes in an effort to gain a better understanding of the human induced implications for all of life on Earth, it remains an inconvenient truth that if the world had acted upon the knowledge that the scientific community already produced, the state of many ecosystems would be different today. The NGO community has an essential role to play in terms of strengthening the science-policy-society interface towards an effective response from society on the issue of marine litter and other societal challenges.

**419 Biodegradable plastics: potential application in aquaculture and other applications at high risk of dispersion**

F. Decli Innocenti, Novamont SpA

The problem of plastic marine debris is caused by inadequate waste management and improper investments for prevention, and recovery programs. The biodegradable plastics industry does not consider biodegradability as a license for littering in the environment. All packaging and consumer products must be recovered in some way at their end of use and, in certain contexts, biodegradability allows recovery through organic recycling. This option is contemplated by the European Directive on Packaging and it is beneficial whenever packaging is mixed with food waste (biowaste). The term “biodegradable” could be misunderstood and induce the consumer to littering. In order to avoid such problem, the biodegradable packaging is labelled “compostable” or “biodegradable and compostable”. The term “biodegradable” is only used in business-to-business communications (e.g. “biodegradable” mulch films are used by professionals who are well aware of the meaning of the term). In agriculture, tests specific to soil define mulch film biodegradation because of soil microorganisms, biodegradable and compostable, can vary from composting. Similarly, tests specific to the marine environment are now under development at ASTM and ISO level. Some biodegradable plastics showed biodegradation levels (as CO2 evolution) comparable to cellulose in less than 1 year using these test methods. Generally speaking, the environmental risk depends on the concentration of the environmental stressor and on its residence time in the environment. The lower the concentration and the shorter the residence time, the better. Bioplastics do not immediately disappear upon exposure to the sea. However, biodegradability reduces the risk by reducing the stressor’s residence time. Concluding, the idea of solving the problem of plastics in the ocean just by shifting to bioplastics is groundless (bioplastics does not disappear “by magic”). However, for those applications where accidental or accidental release is certain or very probable, biodegradability decreases the environmental risk. Materials that show full and relatively fast biodegradation may be suitable for plastic products known to wear down or become stranded (for example, fishing gear) and scatter into the sea. Bioplastics hold promise for aquaculture professional applications (e.g. nets for mussel farming) where the disposal of plastic waste is an inevitable outcome. Bioplastics can be the right solution for specific products, if properly applied.
Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (II)

424 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 Evolutionary Biology, Ecology and Environmental Sciences; E. Bozzano, College of Environmental and Biological Sciences; J. Norberg, Stockholm Resilience Centre; L. Nizzetto, NIVA

425 Changes in pCO2 after the reproductive toxicity of common active pharmaceutical ingredients

C. M. Hunt, N. Wichman, 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

426 From individual traits to ecosystem functioning: natural phytoplankton biodiversity patterns in the GLOBAQUA basin

H. Chevalier, INRAE / Department of Evolutionary Biology, Ecology and Environmental Sciences; M. Lopez de Alda, Institute of Environmental Assessment and Water Research; S. Lutz, Henholtz Centre for Environmental Research; UZV; A. Bellin, E. Stella, University of Trento / Department of Civil, Environmental and Mechanical Engineering

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 MEP project, UN Environment/IMAP 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 the cooperation within the RPML, with a first results step has been presented, towards a more effective marine litter management at regional level. One of the latest developments of the UN Environment/MAP is the 2017 Mediterranean Quality Status Report (QSR) that dedicates two chapters on marine litter related to beach, floating, and sea floor marine litter.

421 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 resulted in a new monitoring of marine litter at regional level. One of the latest developments of the UN Environment/MAP is the 2017 Mediterranean Quality Status Report (QSR) that dedicates two chapters on marine litter related to beach, floating, and sea floor marine litter.

422 Discussion

423 Final Remarks

G. Leone, UNEP/Mediterranean Action Plan

Rivers suffer from an important decrease in species diversity compared to other aquatic and terrestrial ecosystems due to a variety of stressors related to human activities. Species play different roles in the functioning of the ecosystem, and their species loss may reduce the response capacity of the ecosystems to a stressor. The effects on diversity will obviously differ based on the type of stressors and their combination and severity, as well as on the characteristics of the local community composition, and the community tolerance to the type of stressor affecting the system. This study presents the results of the analyses of the biodiversity patterns for different, algae, macrophytes, macroinvertebrate and fish communities related to environmental pressures. The data obtained from the field work conducted in three of the Globoqua case study basins (Adige, Sava and Evrotas) has been evaluated according to structural biological community parameters (species composition and abundance). The most evident relationships between changes in species richness and diversity were explained by changes in hydrology (e.g. mean discharge, intermittency) and morphological changes in the basins (e.g. land uses, channel transformation). The presence of pharmaceutical products (urban pollution) and pesticides was related to lower insect richness. Also emerging compounds, despite their low concentrations, were related with a reduction in macrophyte diversity. These results open the way to compare responses in the studied basins as representative of the European reality of the combined effects of multiple stressors on biological diversity.

Changes in pCO2 after the reproductive toxicity of common active pharmaceutical ingredients

C. M. Hunt, N. Wichman, 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 into 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 determining 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.

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 Evolutionary Biology, Ecology and Environmental Sciences; E. Bozzano, College of Environmental and Biological Sciences; J. Norberg, Stockholm Resilience Centre; L. Nizzetto, NIVA

Rivers suffer from an important decrease in species diversity compared to other aquatic and terrestrial ecosystems due to a variety of stressors related to human activities. Species play different roles in the functioning of the ecosystem, and their species loss may reduce the response capacity of the ecosystems to a stressor. The effects on diversity will obviously differ based on the type of stressors and their combination and severity, as well as on the characteristics of the local community composition, and the community tolerance to the type of stressor affecting the system. This study presents the results of the analyses of the biodiversity patterns for different, algae, macrophytes, macroinvertebrate and fish communities related to environmental pressures. The data obtained from the field work conducted in three of the Globoqua case study basins (Adige, Sava and Evrotas) has been evaluated according to structural biological community parameters (species composition and abundance). The most evident relationships between changes in species richness and diversity were explained by changes in hydrology (e.g. mean discharge, intermittency) and morphological changes in the basins (e.g. land uses, channel transformation). The presence of pharmaceutical products (urban pollution) and pesticides was related to lower insect richness. Also emerging compounds, despite their low concentrations, were related with a reduction in macrophyte diversity. These results open the way to compare responses in the studied basins as representative of the European reality of the combined effects of multiple stressors on biological diversity.
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 prevent the ecosystem to promptly re-gain structures and functions after extreme events. An extreme, metabolomic event was mimicked by sampling and mixing phytoplankton communities over the entire water depth of the lake. The chemical stressors comprised a mixture of chemical pollutants added to the mesocosms at five increasing concentrations (typically considerably below the EC10 of individual substances). This mixture included 12 pharmaceuticals and personal care products commonly detected in lakes and rivers in Europe. In addition, a mixture isolated from treated waste 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 3 weeks experiment. Overall, contaminants, both diffuse and point source, had 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 pollutants can disrupt the capacity of natural communities to handle environmental changes.

427 The role of multiple stressors in an Alpine river and the response of the macroinvertebrate community
A. Maulvault, University of Barcelona / Department of Evolutionary Biology, Ecolology and Environmental Sciences; A. Bellino, University of Trento / Department of Civil, Environmental and Mechanical Engineering; M. Assunção, University of Calabria del Sacro Cuore / Institute of Agricultural and Environmental Chemistry; E. Stella, University of Trento / Department of Civil, Environmental and Mechanical Engineering; E. Capri, Università Catolica del Sacre Cuore / Institute of Agricultural and Environmental Chemistry; I. Muñoz, University of Barcelona / Department of Evolutionary Biology, Ecology and Environmental Sciences; M. Klein, Fraunhofer Institute for Molecular 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 habitats and environmental conditions. As expected, streamflow showed a seasonal pattern, with higher values in the spring-summer period; however, in some sites the natural hydrological regime was altered by the presence of hydropower plants, which management affected most low values of streamflow. Statistical analysis showed a clear seasonal and spatial pattern in both chemical and hydrological parameters; in detail higher concentrations of nitrate, Personal Care and Pharmaceutical products were found in winter season associated with lower streamflow. Changes in richness, diversity and composition of macroinvertebrate community are related with inputs of urban pollution along the river, and with hydrology, chiefly downstream hydropower plants. Pollution (nitrates and other compounds such as PhACs and FCP) favor higher invertebrate densities but lower diversity, changes in thermal natural regime affects Plecoptera, and Gomphus sp demonstrated a higher undifferentiation with flow variability. This study lies in giving a comprehensive and general explanation of the response of biological communities to multiple stressors investigated in an Alpine environment; in particular the analyses performed allow to distinguish the main pressures that impact macroinvertebrates in the Adige river.

428 Coping with antidepressants in a changing ocean: tissue bioaccumulation and behavioural implications in juvenile Argyrosomus regius exposed to venlafaxine
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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 aimed to evaluate for the first time the effect 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.

429 A modelling approach to assess present and future land use pressures on a salmonid river: a case study in the River Tamar catchment (UK)
M. Assunção, Celas Lowestoft Laboratory; P.E. Posen, Centre for Environment Fisheries and Aquaculture Science Cefas; M.G. Hutchins, Centre for Ecology and 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 (QUESTOR) at the sub-catchment level with a basin model to scale up to the Tamar basin. 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)

430 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
S. Hahn, Fraunhofer ITEM / Chemical Risk Assessment; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; M. Nendza, Analytisches Laboratorium
A key objective 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 authorisation and restriction. 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 analysed with a multimedia/multipath 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 substance or multiple substances and to benchmarking 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 perfluorocarboxylate 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

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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 process is to identify 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 substances 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 possibilities for a molecular characterisation of the group substances and their qualitative assessment and evaluation of PBT/vPvB substances with regard to impact potential.

Based on current knowledge, this grouping and relative ranking can guide the formation of concern-based categories for a possible read-across or comparative evaluation of impact potential of PBT/vPvB substances. Acknowledgement – This work was funded by the European Commission, Directorate-General for Internal Market, Industry, Entrepreneurship and SMEs under Contract number: 30-CE-083097200-26 ‘Approach for evaluation of PBTs subject to authorisation and restriction procedures in context of socio-economic analysis’.

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


Non-extractable residues (NER) are not necessarily bound to the soil and those residues where the test substance or its breakdown products were incorporated into the biomolecules. These residues are considered to be irreversibly bound to the soil or transformed into biomass and therefore no risk for the environment can be anticipated from these fractions. The comprehensive scientific assessment of this classification and the analytical accessibility of these NER types will be discussed and supported by experimental data. Therefore, incubation experiments were carried out following the OECD 307 test guideline, which allows a mass balance result above 55% 14C-labeled residues.

The well-characterised standard soils were used and with acetonitrile, trichlosan and fenoxycarb, three radiolabelled test compounds were selected representing a pharmaceutical, a biocide and a pesticide. The substances are already well described with regard to their degradation kinetics and the formation of different NER types. Different mild to harsh extraction procedures like shake flask extraction and an optimised liquid solvent extraction are 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 Nonionic Surfactants, in Soil – A Synterysis, Çeşitli, Two papers in drafting the totally sorted and the seperated fraction which are considered to be possibly remobilised into the environment. Therefore, it is necessary to determine at least this fraction for an adequate risk assessment. Other types encompass residues which are covalently bound to the soil and those residues where the test substance or its breakdown products were incorporated into the biomolecules. These residues are considered to be irreversible bound to the soil or transformed into biomass and therefore no risk for the environment can be anticipated from these fractions. The comprehensive scientific assessment of this classification and the analytical accessibility of these NER types will be discussed and supported by experimental data. Therefore, incubation experiments were carried out following the OECD 307 test guideline, which allows a mass balance result above 55% 14C-labeled residues.

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Developments in the Biopharmaceutical industry have been a significant focus in recent years, with a particular emphasis on the sustainability of these complex processes. One of the key challenges is understanding and integrating the environmental impact of biopharmaceutical manufacturing into decision-making processes.

One approach to this is Life Cycle Assessment (LCA), which is important for understanding the sustainability of biopharmaceutical processes. LCA can help in identifying the environmental hotspots and in making informed decisions about the most sustainable options. For example, in a study published in the SETAC Europe 28th Annual Meeting Abstract Book, researchers explored the role of LCA in the biopharmaceutical industry.

The researchers noted that LCA is an integral part of decision-making processes in the biopharmaceutical industry, and that it is increasingly being integrated into daily work practices. However, they also highlighted the need for LCA to be accessible and relevant to decision-makers, which requires a focus on specific, context-relevant questions.

It was observed that LCA outcomes and criteria vary significantly depending on the decision-making context. For instance, the importance of sustainability in transportation infrastructure has raised in response to the link between anthropogenic activity and global challenges, such as climate change. Thus, there is a need for a critical examination of the ongoing development of models that quantify the social and economic impacts resulting from infrastructure development.

Therefore, the application of LCA and other sustainability assessment tools is crucial for improving decision-making processes. For example, the DST (Decision Support Tool) was developed in the scope of the training programme Sustainable Pavements & Railways Initial Training Network (www.superfin.eu). This tool includes a set of default weights derived from an Analytical Hierarchy Process (AHP) method, tailored to both road and railway systems.

This approach emphasizes the importance of sustainability in transportation infrastructure and suggests that it is critical to consider the ecological, economic, and social impacts resulting from infrastructure development. Thus, the role of LCA and similar tools is fundamental in this context.

In conclusion, LCA and other sustainability assessment tools play a crucial role in improving decision-making processes, especially in the context of biopharmaceutical manufacturing. They help in making informed decisions that are not only environmentally sound but also economically and socially sustainable.
to be used, including LCA, but not necessarily limited to it. Moreover, in every LCA, it is in a first step important to identify a 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 an 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 as a tool to support decision making for both: “tailor” the approaches to be applied for different aspects of sustainability or to gain situational benefits by the defined goal and 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 Horizon2020 research project ITERAMS. In the presentation, we developed a 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 an LCA.

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

**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, environmental and social impact. This paper presents a new methodology for the implementation of reality, which is a step towards the development of an LCA-based decision-making tool that can be used to support the selection and implementation of a sustainable policy scenario. Coupling ABM&LCA can be done with different strategies (extension of LCA, hybrid analysis and complementary use) depending on the expected trade-off between consistency and flexibility. Hybrid analysis is adapted to most of situation since both methods can exchange data externally without impacting the other one. Extension of LCA with ABM leads to a consistent model in which LCA is embedded in ABM, which is particularly relevant for study requiring to take into account dynamic effects in the technosphere. Different degrees of coupling (hard-coupling and soft-coupling) are defined according to (a)data flow direction and (b)coupling dynamic. Higher the degree of coupling, the higher the computational time; therefore the use of hard-coupling should be limited to studies integrating feedback in adaptive decision-making process. This paper addresses some methodological guidelines on the way of creating a dynamic LCA, which can be used as a decision-making tool. Future research in this field should now address temporal dynamics in the life cycle impact assessment.
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 lauryl ether sulphate (SLES) and in smaller concentrations other additives. In accordance with the Italian legislation, spoil material of excavation processes can be reused 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 ecoxotological 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) tunnelling machines (TBMs). It is available as ready 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 product, based on a natural mineral widely distributed in the earth’s crust 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 is very important product and deposition of 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 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 enhance the excavation activity. Depending on the excavated materials features, continuous injection of chemicals during the advancement of the TBMs relies on the 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 lauryl ether sulphate (SLES) and in smaller concentrations other additives. In accordance with the Italian legislation, spoil material of excavation processes can be reused 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 ecoxotological test selection) has taken into account the site-specific characteristics and the possible environmental exposure scenarios in order to protect ecosystems and human health.

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. The development of a laboratory methodology to develop 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

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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 emission 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 CNTs were dispersed in deionized water, sonicated, and then diluted 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 increase 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 µg L⁻¹ - 100 µL L⁻¹) resulted in a linear relationship. The calculated and the 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 L⁻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.

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

H. LEE; National Institute of Environmental Research (NIER) / Geum River Water Environment Research Center; Y. Cho, J. Khan, National Institute of Environmental Research; S. Kim, Geum river environment research center; B. Lee, National Institute of Environmental Research NIER / Han river water environment research center; B. Seol, M. Chae, S. Cheon, National Institute of Environmental Research NIER / Geum River Water Environment Research Center A multiphase analytical method using LC-MS/MS was developed for perfluorinated compounds (PFCs), insecticides, and brominated flame retardants (BFRs) in water samples with the simultaneous SPE method. The ranges of recoveries were 97.1 % - 135.0 % (PFCs), 95.0 - 117.2 % (Insecticides), and 72.5 - 86.4 % (BFRs), with coefficients of variation of less than 15%. Method detection limit (MDLs) of PFCs, insecticides, and BFRs were 0.3 - 7.1 ng/L, 3.0 - 3.7 ng/L, and 5.1 - 11.7 ng/L, respectively while limit of quantifications (LOQs) were 0.9 - 21.0 ng/L (Insecticides), 9.0 - 11.0 ng/L (BFRs). For understanding the background levels of PFCs, insecticides, and BFRs in the river water, those compounds were monitored in Geum river main stream, So-ok stream, Juwon stream, and Daechung lake (Dam) every month (March to December) utilizing the developed method. The compounds of the highest detection frequency were PFFOA, PFHxA, and dinotefuran (Insecticide), whereas BFRs were detected only in March and December, except for main stream. In conclusion, the trends were not observed on periodical and spatial characteristics and the background levels were secured for PFCs, insecticides, and BFRs in Geum river basin.

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, reclaimed water systems evolve as 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 bioactive at low concentrations and are considered “pseudo-persistent” due to their capacity to contaminate the environment with no mass spectrometry. 14C tracing, enzyme extraction and Illumina sequencing techniques, we evaluated a wide range of biological effects in terrestrial organism caused by exposure to CECs. Organisms in these studies included, the cabbage looper (Trichoplusia ni), an earthworm (Eisenia fetida), a model plant (Arabidopsis thaliana) and cucumbers (Cucumis sativus), radishes (Raphanus sativus), and tomatoes (Solanum lycopersicum). These studies have revealed a multitude of effects including increased mortality and development time, reduction in weight, changes to the microbiome and up-regulation of enzymes associated with oxidative stress. Further, the study has highlighted the potential for higher plants to take up, translocate and detoxify CECs.

451 Occurrence of pharmaceuticals and their metabolites in Euthynnus alletteratus bili

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The presence of pharmaceuticals in surface waters has been attributed to the effluents of WWTP among others. The widespread occurrence of pharmaceuticals in the aquatic environment has raised concerns about their potential adverse effects on exposed wildlife. Little is currently known on exposure levels of drugs in fish, but some studies reported the detection of pharmaceuticals and endocrine disrupting compounds in this type of biota. Due to possible accumulation processes, 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 pharmaceuticals exposure including the identification of metabolites by UPLC-HRMS. In this context, we propose the evaluation of the metabolism of frequently detected drugs in fish, performing a screening bile by HR-MS 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, Cartagena and Ceuta were sampled between January 2015 and January 2017. The 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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Crops irrigated 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 resources. Despite this, this practice increases the presence of contaminants and contributes to nutrient recycling, using reclaimed water for irrigation, however, represents a primary source of emerging organic contaminants resilient to wastewater treatment processes, such as some 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 (diclofenac, trimethoprim, carbamazepine, oxcarbazepine, lamotrigine, cis-diltiazem, methadone), an illegal drug (cocaine) and two transformation products (acridone 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 experiment, leaves, roots and soil samples were collected and analyzed for possible metabolites. All pharmaceutically active compounds were extracted by ultrasonic liquid extraction (USE) [4]. The detection of the target analytes was performed using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS). The results from the first growing season evidenced the presence of all analytes in all investigated matrices. Carbamazepine was the analyte that accumulated the most in lettuce plants (leaves and root system), whereas cis-diltiazem, methadone, and midazolam were preferentially accumulated in the plant root system and soil. Concentrations of the target analytes in the plant-root-soil system after the second growing season were significantly lower than those measured after the first growing
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 Epiaeristic. 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 (radish, lettuce, arugula, spinach, and green peas). No carbamazepine was detected in leaves of 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 individual variation in the concentrations within exposure treatments; plasma NSAID concentrations as a measure of stress in fish, potentially affecting plasma NSAID and PG 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/453E47/02) 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 the data, we discuss some of the challenges that the EMA guideline is facing and present the ongoing work towards an improved and 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 contracancer drugs. Furthermore, the evaluated data allow discussion about effect sensitivity of several taxonomic groups, as well as about the PEC action limit (10 ng/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 approach should replace long-time-acquired acute data couple. Further changed the assessment factors as applied usually for chemicals without any specific mode of action will be analyzed.

456 Prioritising human health risk of environmental residues of pharmaceuticals and personal care products in use in southern Nigeria

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Pharmaceutical and personal care products (PPCPs) are used worldwide for medical treatment and personal hygiene. PPCP residues are usually discharged into the environment during wastewater treatment. The environmental fate of these chemicals is poorly understood, leading to concerns about potential health effects and ecological impact posed by their use and release. In many developing countries, information regarding the consumption patterns of PPCPs is often limited, even though consumer product usage data are crucial for more realistic exposure estimates, like needed for risk assessment. To address this gap, the following study was performed to estimate PPCP use in Nigerian households to prioritise the potential for PPCPs to enter source water. Using questionnaires as the survey instrument, we elicited information from 350 participants, concerning the most frequently used PPCPs, duration and amount of use in households. Drug usage was limited to over-the-counter(OTC) medicines and was estimated by application of the National Health Organisations daily dosage. The consumption of personal care products (PCPs) was calculated by multiplying the quantity of products used by the frequency of use. To prioritise PPCPs, a risk index was developed that represents chemicals according to their potential to enter source water. Consumption of PPCPs varied considerably. Analgesics were the most consumed OTC medicines and highest use was observed for paracetamol. Household cleaning products were the most consumed PCPs and highest use was observed for detergent powder and dishwashing liquids. Overall, 12 PPCPs were identified as having the greatest potential to reach source water and pose adverse effects to human health. These include 8 active pharmaceutical ingredients (acetaminophen, tetracycline, ...
ciprofloxacin, ampicillin, cloxacillin, sulfamethoxazole, trimethoprim and pseudoephedrine) and 4 PCR ingredients (sodium lauryl ether sulphate, alcohol ethoxylates, ammonium thiglycollate 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.

457 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 *Raphidiocelis subcapitata*. 

Acknowledgement - The research leading to these results has received support from the Innovative Medicines Initiative Joint Undertaking funded with EU FP7 grant agreement n° 115735, resources of which are composed of financial contribution from the European Union's Seventh Framework Programme (FP7/2007-2013) and EFPIA companies’ in kind contribution.

458 Neurotoxicity testing approach to investigate venlafaxine and oxazepam modulation of transcriptomics and behavioral profiles in zebrasib fungus and larvae
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Neuroactive pharmaceuticals are of growing concern as aquatic contaminants due to environmental and human health risks. Even low concentrations can interfere with molecular pathways and population-relevant behaviors. At the same time there is no EU regulatory framework for environmental neurotoxicity assessment. This project aimed to contribute for establishing a neurotoxicity testing approach by focusing on key molecular (transcriptomics) and functional (behavioral) endpoints involved with neuroactive pharmaceuticals in zebrafish embryos and larvae exposed to neuroactive pharmaceuticals. *Danio rerio* up to 5 days post fertilization (dpf) were statically exposed to venlafaxine (serotonin norepinephrine reuptake inhibitor antidepressant) or oxazepam (benzodiazepine derivative anxiolytic) at the µg/L range (1 nm to 10 or 100 µM). Solution concentrations were measured at the start and end of exposures by LC-HRMS. Assessed behavioral endpoints were embryonic spontaneous movement (1 dpf), touch-evoked escape response (3 dpf), and phototaxis and thigmotaxis reactions (5 dpf). RNA was extracted from pooled embryos or larvae (n=20-50) and submitted either to RNA sequencing with Illumina Next Generation Sequencing System (RNAseq) or SYBR Green based quantitative real-time PCR (qPCR). qPCR target genes were selected with basis on RNAseq results, but also a few targets proposed as markers of exposure or modulation by neuroactive compounds were selected from literature studies (e.g. *fkhb5*, *cfos, per3*). Reference genes were ef-1α, rpl13, rpl8. Chemical analysis indicated that solution concentrations were stable along exposure periods and in general accordance with nominal values. Oxazepam caused behavioral alterations mainly at 1 and 10 µM Oxazepam, while venlafaxine affected prevalently larvae behavioral endpoints. RNAseq of embryos exposed to 100 µM oxazepam indicated gene ontology enrichment for notochord morphogenesis. Larvae exposed to 10 nm venlafaxine presented differential modulation of response to abiotic stimuli, while 100 µM venlafaxine affected mainly muscle processes and to a minor extent circadian rhythm modulation. Confirmatory qPCR is being conducted. Zebrafish embryo-larval assays supported the elucidation of molecular mechanisms at the transcriptome level that occured concurrently with organism-level behavioral effects. Our results are expected to contribute in the future for AOP annotation and for the setup of a regulatory assessment approach to evaluate neurotoxic environmental contaminants.

459 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.
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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 there is a legacy of essential medicines for which we need to understand more. Soon 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 potential 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 numerous 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 iP4 project (IM grant no.115735—IP5). 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 (BBSC/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

460 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 emergent chemicals such as fullerene, in particular, the mechanisms of action are poorly investigated. In this research, the effects of C60 on mTOR (mechanistic Target of Rapamycin) activity in mussels 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 translation, 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 shown by the reduction of lysosomal membrane stability and the enhancement of lysosomal/cytosolic 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-shape 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...
translational, cytokine signaling mechanism 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 nanoparticulate 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. Pascual, F. Cassio, University of Minho / Centre of Molecular and Environmental Biology CBMA Department of Biology. Enhanced use of AgNPs (Ag) 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 EC50 (effective concentration) were assessed based on the variations in the overall protonome in two aquatic fungal strains of Articulospora tetracladia, one isolated from a non-polluted stream (Art72) and the other from a metal-polluted stream (Art61), and ii) the bacterial strain Pseudomonas sp. M1 (PsM1) isolated from a metal-polluted stream. Art72 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 in Art72 while for Art61 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/or Ag+ whereas for PsM1 this percentage was lower (≈20%). Art72 and Art61 shared only 20% of the proteins suggesting that the biological pathways involved in Ag+ and AgNPs exposure were different. Art61 had ≈25% more proteins induced by both Ag forms (compared to Art72), 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/or AgNPs. In Art72, 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 Art61, 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 used to get a mechanistic insight on the stress induced by AgNPs and/or 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
F. Müller, University of Rijeka / Department of Biology; D. Hackenberger, Department f Biology, University of Osijek / Department of Biology; D. Marković, University of Rijeka / Department of Biotechnology; I. ObodoDzazs, Radjer Boskovic Institute; B. Hackenberger, 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 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 nanoZnO 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 nZnO/CHP combination. The inhibition of AChE was present at all applied combinations. In natural soil the number of juveniles was reduced with nZnO, while it remained the same as control after bZnO exposure. The effects of combined exposure of CHP and ZnO seems to be depending on the form of ZnO and its characteristics of the soil in which the earthworms were exposed. More biomarkers should be employed to elucidate which low-level biomarker can be linked with the effects on the higher (reproductive) level.

463 Poster spotlight: WE305, WE323, WE324

464 Improving the environmental risk assessment of the aquaculture 'Blue Revolution'

465 Tools for Assessment and Planning of Aquaculture Sustainability (TAPAS)
P. van den Brink, Alterra and Wageningen University; A. Lillicrap, NIVA Norwegian Institute for Water Research / Ecotoxicology; A.L. Macken, Norwegian Institute for Water Research, NIVA; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; T.C. Telfer, University of Stirling Aquaculture is a major food production subsector which will play a major role in filling the growing seafood supply gap, estimated to be of the order of 47.5 million tons by 2050. However, aquaculture cannot be practiced everywhere; it requires a particular set of natural, social and economic resources which 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 predicated 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
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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 micropolllutants (MPs). In this study, the presence and distribution of multifunctional organic micropollutants (MOPs) to 14 different groups were investigated in offshore seawaters and fish farms of Singapore. 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 cyclamate, salicylic acid and saccharose, with concentration range of 0.03 – 360 μg/L.

468 Perspectives on Urbanization, Water Reuse, and Aquaculture Product Quality
By B. W. Brooks, Baylor University / Dept of Environmental Science; J.L. Conkle, Texas A&M University Corpus Christi / Physical and Environmental Sciences
By 2050, it is estimated that global food production must increase by 50%. Aquaculture will play an important role to meet these needs. For example, in 2014 aquaculture surpassed global fisheries in providing fish 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 a large number of the megacities will continue to emerge over the next few decades, agricultural 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, yet 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 landfills leachates and effluent discharges of marginal quality, in Hong Kong. Our findings from a laboratory uptake and depuration studies with channel catfish and tilapia focus on contaminants with diverse physico-chemical properties (e.g., weak base medicine, phosphorus-based flame retardant, perfluorinated compound, cyanoxotin) and provide an approach to improve aquaculture practice and to support bioaccumulation assessments for chemicals falling outside of applicability domains for nonionizable organic contaminants. In North America we are examining intersections 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 azinphosphos. 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 of teflubenzuron was also studied; other mussel species in brackish waters show different bioaccumulation dynamics. So far, all experiments 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 low bioaccumulation, with maximum concentrations of 45 mg/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 approaches can be used. These are powerful tools for investigating 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 farm 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 radiotracer techniques 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 radiotracer permits the non-destructive analyses of contaminant levels in living organisms. This paper identifies present and future threats on farmed fish from a contamination point of view, and presents a synthesis of experimental results completed on farm-raised fishes exposed to contaminants in realistic environmental conditions. It revealed, 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 have shown an increasing emergence of antibacterial 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 covered and non covered with local feed containing antibiotics non medicated or medicated with three antibiotics (oxytetracycline, florfenicol and flumequine). Fish feeds 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 flumequin. 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 target 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 ecology: application of OMICS data across multiple levels of biological organization in research and risk assessment (II)**

**472** Systems toxicology approach for the assessment of zebrafish cardiovascular 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 human effects 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 organism level to explain effects at the organism level. However, current studies remain focused at the population 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 individual variation.

Furthermore, most studies select an arbitrary timepoint to measure gene expression responses without any prior knowledge. Here, we focus on population level 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?

E. Caamaño-Gutierrez, University of Liverpool / Computational Biology Facility; P. Larras, Edinburgh University / Institute of Integrative Biology; E. Caamaño-Gutierrez, The University of Birmingham / School of Biosciences; K. Grintzalis, The University of Birmingham; M.R. Viant, University of Birmingham / School of Biosciences; F. Falciani, University of Liverpool / Institute of Integrative Biology

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. Current ecotoxicological methods for risk assessment are tied to chronic exposures and a lack of consideration of the underlying response to endocrine disruptors may be higher than previously thought.

While the individual exposures triggered the alteration of endpoints, the underlying response to endocrine disruptors or AChE inhibitors. Cadmium (Cd), which clustered with endocrine disruptors, showed 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).

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 rise 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 zebrafish. The rationale to use OMICs in such context is to provide more protective and early warning thresholds. The transcriptomic and 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 containing metabolites/transporters and enriched compounds concentration responses of each time point and we derived a sensitivity value from each curve (even the non-monotonic one). Molecular items showed mainly nonsigmoid 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 metalloharmones and endocrine disruptors

E. Caamaño-Gutierrez, University of Liverpool / Computational Biology Facility; P. Larras, Edinburgh University / Institute of Integrative Biology; E. Caamaño-Gutierrez, The University of Birmingham / School of Biosciences; K. Grintzalis, The University of Birmingham; M.R. Viant, University of Birmingham / School of Biosciences; F. Falciani, University of Liverpool / Institute of Integrative Biology

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. Current ecotoxicological methods for risk assessment are tied to chronic exposures and a lack of consideration of the underlying response to endocrine disruptors or Daphnia magna to a panel of environmentally relevant chemicals and their mixtures. Firstly, it was exposed to a batch 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 estrogen disruptors or AChE inhibitors, while metals (Cd), which clustered with endocrine 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 ethinylestradiol. While the individual exposures triggered the alteration of endpoints, the 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
Towards a shared understanding of science and risk communication in the context of the inevitability of chemicals and the hazard they may represent (I)

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 endocrine 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 as to what 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 endocrine 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 as to what 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. Almar, ECHA-European Chemicals Agency; W. de Wolf, ECHA / Product Safety & Regulatory Compliance

The European Chemicals Agency (ECHA) was established in June 2007 through 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

J. Kronberg-Guzman, The University of Birmingham / School of Biosciences; T.D. Williams, University of Birmingham / School of Biosciences; A. Mark, Wageningen Agricultural University / Dept of Toxicology; E. Foekema, Wageningen IMARES; R. van der Oost, Waternet / Onderzoek en Advies; K. Chipman, University of Birmingham; F. Falciani, University of Liverpool / Institute of Integrative Biology

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. Watermonarica 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, fluroanethane, 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. Functional enrichment of stickleback. We also see that some chemicals for which predicted no-effect concentration (PNEC) is not known (such as PCBs 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 triclosan and phthalates has a statistically significant number of genes from the KEGG pathway „metabolism of xenobiotics by cytochrome P450“. However, 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. Delugi in post, Irstea / UR RIVERLY Laboratoire Ecotoxicologie; C. ALMUNIA, CEA Paris-Saclay; D.D. Gouveia, Irstea Lyon / UR MALK Laboratoire Ecotoxicologie; J. Trapp, Irstea Lyon; J. Gaillard, CEA / Laboratoire de Biochimie des Systèmes Perturbés; O. Pible, CEA; a. chaumot, O. Geffard, Irstea / UR MALK Laboratoire Ecotoxicologie; J. Armgaud, CEA / Laboratory Innovative technologies for Detection and Diagnostics

Discussion mining of high-dimensional omics data and 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 sentient organism Gammarus fossarum in the stickleback, gammarus fossarum 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 crusteacean. We identified protein modules significantly associated to morphologically well-characterized physiological states and to pesticide exposure. Moreover, the identification of crucial hub proteins could allow proposing exposure-related or toxicological functional biomarkers. This new data mining procedure opens interesting perspectives for the development of a novel generation of molecular diagnostic biomarkers in ecotoxicology.

SETAC Europe 28th Annual Meeting Abstract Book
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). Unl 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. '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 outcomes 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 stress

Aaron Haefner, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; W. Amelung, University of Bonn; H. Hollett, RWTH Aachen University / Institute for Environmental Research; M. Kaestner, Helmholtz Centre for Environmental Research UFZ / Dept Environmental Biotechnology; E. Kandler, University of Hohenheim; J. Kruse, University of Bonn; A. Millner, Helmholtz Centre for Environmental Research UFZ / Department of Environmental Biotechnology; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research; H. Pogel, University of Hohenheim; S. Peth, University of Kassel; C. Poll, University of Hohenheim; G. Rambold, University of Bayreuth; M. Schloter, Helmholtz Zentrum Munchen / Research Unit Environmental Genomics; S. Schulz, Helmholtz Zentrum Munchen; T. Streck, University of Hohenheim; M. Rob-Nickoll, RWTH Aachen University / Institute for Environmental Research

Soils are faced with man-made chemical stress, such as the input of organic or metal-containing pesticides, in combination with non-chemical stressors like soil compaction due to agricultural traffic and natural disturbance like drought. Although multiple stress factors are typically co-occurring in the environment, research in soil sciences on this aspect is limited and focuses mostly on single structural or functional endpoints. A mechanistic understanding of the reaction of soils to multiple stressors is currently lacking. Based on a review of resilience theory, we introduce a concept for research on the ability of soil polluted by xenobiotics or other chemicals as one stressor to resist further natural or anthropogenic stress and to retain its functions and structure. There is strong indication that pollution as a primary stressor will change the system reaction of soils, i.e., its resilience, stability and resistance. It can be expected that pollution affects the physiological adaption of organisms and the functional redundancy of the soil to further stress. We hypothesize that the recovery of organisms and chemical-physical properties after impact of a follow-up stressor in polluted soil differ from that in non-polluted soil, i.e., polluted soil has a different dynamical stability, and resilience of the contaminated soil is lower compared to that of not or less contaminated soil. Thus, a polluted soil might more easily change into another system regime after occurrence of further stress. We highlight this issue by compiling the literature exemplarily for the effects of Cu contamination and compaction on soil functions and structure. However, examples of further co-occurring stress scenarios will be described as well. In this discussion paper, we propose to intensify research on effects of combined stresses involving a multidisciplinary team of experts and provide suggestions for corresponding experiments. Our concept offers thus a framework for system level analysis of soils paving the way to enhance ecological theory.

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

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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 (fas2, degs2, scd2) and elongases (elov2, elov5, elov6). Both yellow perch and fathead minnows 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 polysaturaturation 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 polysaturaturation 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 muscle cell membrane composition of both tissues, but in a way that was more conservative than for yellow perch. We observed a mismatch between desaturation and elongation 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.

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

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Fish are dependent on olfaction since a variety of essential behaviours, such as foraging, predator avoidance and mate selection, are mediated by the olfactory system. Metals are well known to affect the olfactory system of fishes at environmentally-relevant concentrations. As metal toxicity varies with water chemistry in a predictable manner, modelling approaches, such as the Biotic Ligand Model (BLM), are powerful tools to predict site-specific effect concentrations. To date, the BLM used in risk assessment for fish only predicts gill-based metal toxicity. However, metal-binding dynamics at the olfactory epithelium may be different than for gills. For this reason, the present study investigated the impact of water chemistry on cadmium induced olfactory impairment. In order to assess the effect of Cd on the olfactory system, fish were exposed to 45-720 µg/L Cd for 24 h. Subsequently, olfactory responses to two odorants were measured via electro-olfactography (EOG). To investigate the impact of water chemistry on Cd-induced olfactory impairment, fish were exposed to the EOG-basied 24-h IC50 of Cd (210 µg/L) in reconstructed water with varying hardness, pH, and dissolved organic carbon (DOC) concentrations for 24 h. Cd inhibited the EOG response of rainbow trout in a concentration dependent manner. Fish exposed to 210 µg/L Cd for 24 h showed reduced olfactory response to TCA by 50%. Changes in water chemistry had a significant impact on Cd-induced olfactory impairment. Decreasing water hardness from 150 to 40 mg/L as CaCO3 for 24 h showed reduced olfactory response to TCA by 50%. Changes in water hardness from 150 to 40 mg/L as CaCO3 for 24 h showed reduced olfactory response to TCA by 50%.
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

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Coastal systems often serve as sinks for toxic elements, and changes in salinity, predicted to occur due to global climate change are expected to influence elements geochemistry in aquatic systems. The effects of these changes can also alter biota sensitivity and salinity can be used to derive information on possible changes 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 ría de Aveiro lagoon, Portugal) and salinities 21, 28 and 40, for 28 days. This study aimed to evaluate physiological (regenerative capacity of Diopatra neapolitana), behaviour (burrowing tests with Hediste 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. neapolitana. 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 at salinity 40 led to a deleterious effect on the polychaete Diopatra neapolitana 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 parasite infestation influence the activity of the bioturbation Upogebia pusilla?

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In marine environment, bioturbating species are considered as important ecosystem engineer species. Bioturbators are mainly benthic organisms living in the sediment. Their flossorial 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 influence with bioturbators fitness and therefore modify the influence of these 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 parasitized by bopyrid isopods, known to have a deleterious effect on their host. Indeed, they deeply impair mud shrimp physiological state and potentially alter the bioturbation intensity. The aim of this study was to examine the role of a common trace metal (cadmium) and of bopyrid parasites on the physiology and the bioturbation activity of the mud shrimp Upogebia pusilla. We performed a 14-days ex-situ experiment evaluating the influence of Cd and/or parasite on the mud shrimp U. pusilla. Cadmium bioaccumulation and potential genetic responses to stress exposure were determined after 3, 7 and 14 days to trace metal exposition. The influence of both stressors on U. pusilla bioturbation activity was determined by evaluating sediment reworking rates of the mud shrimp after 3, 7 and 14 days to trace metal exposure.
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 8 weeks, respectively in the off-field situation (hay meadow paradigm). For the in-field situation recovery intervals of 0-1, 2-6, 6-12 and 12 months were delimited by HQ values of 40, 375, 620 and 2500. Tier 2 studies had the same length of recovery 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, (MEOGRT) biological validity criteria E. Salinas, BASF SE / Experimental Toxicology and Ecology; L. Wettel, BASF SE / Crop Protection Ecotoxicology The MEOGRT, Extended Extended Extended Environment Exposure 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 have already noted a high incidence of study repetition following well established OECD fish TGs. However, the data is as current, especially for NOEC, 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 criteria cannot be dismissed in a reproduction test where high fecundity is directly related to statistical power. The MEOGRT fecundity validity checks are 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.

496 Variability in Non-Target Terrestrial Plant Studies Should Inform Endpoint Selection J.W. Green, Exponent; J.W. Green, DuPont / Data Science and Informatics; J. Nusz, Exponent, Inc.; D.E. Edwards, BASF Corporation / Ecotoxicology; K. Henry, NovaSource/ Tessenderlo Kerley, Inc. / Ecological Sciences; M.E. Kern, Waterborne Environmental, Inc. / Ecotoxicology Risk Assessment; A. Deines, Exponent; R. Brain, Syngenta Crop Protection, Inc. / Department of Environmental Risk Characterization; B. Glenn, Bayer CropScience / graduate student; N. Ehresman, Nufarm; T.S. Kung, FMC Corporation / Global Regulatory Sciences / Global Regulatory Sciences, Department of Biochemistry and Microbiology; F. Kee, FMC Corporation; K. Ralston-Hooper, Dow AgroSciences; S. McMaster, Industry Task Force II on 2,4-D Research Data

Inherent variability in Non-Target Terrestrial Plant (NTTP) guideline testing of pesticide effects can be challenged 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 L. Wheeler, Dow AgroSciences; P. Valverde-Garcia, Dow Agro Sciences LLC; T.A. Smart, EAG Laboratories / Specialist Projects & Histology; V.J. Kramer, Dow AgroSciences LLC / Ecotoxicology; M. Foudoulakis, 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. Here we present a historical control database of malland duck reproduction studies performed at the Evans Analytical Group LLC. We believe the analysis and evaluation presented here can facilitate the development of practical guidance that can be implemented in regulatory programs 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 \( Y = \mu + \epsilon \), where \( \mu \) is the expected mean response in the 1st concentration, and the \( \epsilon \) are independent random errors, usually assumed to be identically distributed. What distinguishes one model from another is what distribution is assumed for the errors or responses and what restrictions or assumptions are placed on the treatment means, \( \mu \). 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 ecotoxicity 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. Haywood, 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 example, 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 and control of contaminants about the origin. For example, for 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 include particleboards 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 Impression 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. Lokedh, 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 emphasise on resource efficiency and material circularity of bio-based value chain and include (but are not restricted to) waste circularity, 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 tool that also covers the impact via the use of 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. Aamundarson, DTU (Technical University of Denmark) / Division for Quantitative Sustainability Assessment DTU Management Engineering and DTU Biosustain; S. Sukumaran, DTU Technical University of Denmark / DTU Biosustain; P. Fanteke, Technical University of Denmark / Quantitative Sustainability Assessment Division
Existing Life cycle assessment (LCA) studies of biochemicals reveal that there are choices that need to be made at an early stage 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 agroindustrial lignocellulose, or not-utilised mass like 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 dosa 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 biomass which requires external application of nutrients and intensity of chemical pretreatment. Today decision makers 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
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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, therefore, efforts to achieve environmental goals. Indirect land use change (ILUC) has 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
C. Som, EMPA Technology & Society Lab; R. Hischier, EMPA / Technology and Society Lab; F. Piccinno, EMPA
The Multi-Perspective Material Selection (MPAS) method has been developed as a decision support tool to identify the most sustainable application 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 revealed 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 support decision making at an early development stage. Here, we present an expanded and further developed MPAS method that mitigates exactly these 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 are 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 ecoinnovations

S. Sala, A. Cerutti, European Commission Joint Research Centre / Bioeconomy unit; V. Castellani, EC-JRC; M. Scchzi, European Commission Joint Research Centre / Bioeconomy unit

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, ecoinnovation 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 allow 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 these BoPs, the baseline scenarios have been developed in order to identify 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.

Environmental Risk Assessment in Sediments

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

S. Heise, Hamburg University of Applied Sciences / Life Sciences; U. Rieth, Institut für Hygiene und Umwelt

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 in Europe, water quality has largely 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 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 for long time to sediments in those ca. 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 230 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, DDX). 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 

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

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Although this mine disposes 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. The reference site was characterized by lower mobility during flood events. Dating of sediment cores from 2014 pointed at typical historic gradients in thin films (DGT) technique for predicting metal bioavailability in sediments. Bioaccumulation between organisms exposed to identical sediments in laboratory and freshwater bivalves in laboratory and field set experiments were carried out to evaluate the performance of the diffusive gradients in thin films (DGT) technique for predicting metal bioavailability in sediments. 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. 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 fluxes measured at the SWI and observed 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-upps. 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...
Toxicity Methods for Freshwater Sediment

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Sediment toxicity tests are used for contaminated sediments, chemical registration, and water quality criteria evaluations. In 1995, USEPA’s Methods for Testing the Most Sensitive Organism was published. In general, the test systems used for sediment toxicity testing have been revised by a committee of 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 testing methods and has a section for freshwater species, Hyalella azteca (amphipod), Chironomus dilutus (midge) and Lumbriculus 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 determined test acceptance 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. From 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 test organism sand microgranular group such as the midge 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 fluodoxionil-spiked field-collected and artificial sediment were performed with all sediment-dwelling invertebrate taxa. 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 views or the policies of the USEPA.

509 Sediment-spiked toxicity tests with benthic macro-invertebrates and the fungicide fluodoxionil

Leif Bergk, Alterra / Environmental Risk Assessment; J. Romão, University of Aveiro; X.H. Yin, Zhe Jiang Agriculture and Forestry University; T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team

In the EFSA scientific opinion on sediments, one of the oligochaete worms Lumbricus spp. or Tubifex tubifex, supplemented with a second test species is generally recommended. The midge 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 fluodoxionil-spiked field-collected and artificial sediment were performed with all sediment-dwelling invertebrate taxa. 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 views or the policies of the USEPA.

510 Spatio-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 ecotoxicological tests is currently revised under the premise to improve quality and consistency of regulatory environmental assessment. In 2015, the European Food Safety Authority (EFSA) has published a scientific opinion about sediment toxicity testing. In the context of the European project 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 the simulation are compared with measured sediment concentrations in three depths (see contribution submitted by Dorn et al.). The compound properties were characterised using values derived independently in standard tests (KOC DT50water/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 millimeters 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

S. Bagnis, C. King, UK / AstraZeneca Global Environment; A. Tappin, Plymouth University; S. Comber, Plymouth University / Environmental Science

The increasing 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 (DUW), 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 refinement of 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 predicted median DF is lower than 10. There is no information available in the literature about the effects of low dilutions on the natural attenuation of APIs nor impacts of DUW. Furthermore, information on the effects of low dilution on mixtures of APIs is missing, hence necessitating the requirement for evaluation of biological 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 attempt to expand this area of research. The biodegradation of a set of APIs was studied in batch tests at several levels of dilution. Nevirapine shows persistency across the experimental period and only the dilution is controlling the observed concentrations. Acebutolol and Diclofenac show a decrease in concentration of up to 90% as a result of a combination of dilution and biodegradation. The biodegradation of a set of APIs mixture and the dilution on the microbial activity are being investigated. While, the biodegradation alone is responsible for the 70% of the removal. The TOC analyses do not show significant consumption rate changes caused by dilution. The results regarding the effects of the APIs mixture and the dilution on the microbial composition are being analysed through bioinformatic statistics, and will be presented if significant.

512 Active Pharmaceutical Ingredients Entering the Aquatic Environment From
Wastewater Treatment Works: Measurement, Prediction, Risk - A Cause for Concern?

S. Comber, Plymouth University / Environmental Science; M. Gardner, Atkins Ltd.; P. Sorne, AstraZeneca / AstraZeneca Patient Safety; B. Ellor, UKWIR

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 ethinyloestradiol, diclofenac, propanolol, the macrolide antibiotics, fluoxetine, tamofoxen and carbamazepine. All except the last two of these substances were present in effluents at concentrations higher than their respective estimated PNEC (based on measurements of 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 exceedences of estimated riverine PNEC's 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. Further details on this work are presented. The dataset and discussion provides information to assist in the future management of these types of chemicals.

513 Impact of a wastewater treatment plant upgrade on amphipods and other macroinvertebrates: individual and community responses

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Conventional wastewater treatment plants (WWTWs) equipped with secondary and tertiary treatment steps do not or only partially remove micropollutants which makes them important point sources for the release of these substances in the water cycle. Micropolitants can cause short- or long-term adverse effects in aquatic organisms even at low concentration levels. One possibility to reduce the input of micropolitants into the water cycle is the upgrading of WWTWs with an additional process step, e.g. ozonation of effluents from 45 WwTW on 20 occasions. The present research work is part of the joint research BMBF project “SchussenAktiv” funded by the German Federal Ministry of Education and Research (BMBF) and the Ministry of Environment, Baden-Württemberg, Germany. In this project, the efficiency of an additional wastewater treatment based on powdered activated carbon for the ecosytem of the Schussen river, a major tributary of Lake Constance, Southern Germany, has been investigated. Our 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 lipidsperoxides allowed us to draw conclusions about proinflammatory and oxidative stress in gammarids. Macrozoobenthe community integrity was determined by means of the saprobic index as well as by the number of sensitive taxa (EPT index). Prior to the WWTW upgrade, the health status of gammarids as well as the integrity of the macrozoobentho community was negatively influenced by the WWTW effluent. After the upgrading of the WWTW, gammarids from the downstream site did not differ any longer from those collected upstream of the WWTW with respect to the investigated health parameters. Furthermore, the overall number of taxa and particularly the number of EPT taxa within the macrozoobentho community downstream of the WWTW increased distinctly after the upgrade of the WWTW with the additional activated carbon step. We conclude that the efficiency of the activated carbon step to eliminate toxic and emerging contaminants from the effluent can plausibly be related to the improved integrity of macroinvertebrate health and community structure in the connected river Schussen.

514 Effects of full-scale ozonation of treated effluent - Environmental impact in a receiving river

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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 (WWTWs), or a complete lack of WWTWs (1). In surface waters, concentrations of pharmaceuticals usually range from low µg l⁻¹ 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 technologies used in WWTWs. One technique to increase the removal of pharmaceuticals in WWTWs is to add a tertiary treatment step based on the addition of ozone. Ozoneation 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 evaluate the removal of pharmaceuticals in a WWTW, when adding ozonation as an additional tertiary treatment step and also to investigate the environmental impact of this effluent on the receiving river. All treated effluent from a minor WWTW (10000 PE) were treated by an addition of 8 h² o³ ozone during 6 months. Removal rates in the WWTW 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 will also be presented. Several additional methods to evaluate the impacts 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.

515 Dreissena polymorpha as purifier tool of protozoa in wastewater treatment plant effluent

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Aquatic environments are subject to discharges of multiple contaminants (chemical and biological compounds). Wastewater treatment plants (WWTWs) are ineffective to remove environmental forms of protozoa such as Toxoplasma gondii and Cryptosporidium parvum oocysts or Giardia duodenalis cysts because of their resistance to chemical and physical treatments. These protozoa are clearly identified as a public health priority since they are major parasites of waterborne outbreaks. Many studies underline the interest of using of freshwater bivalve Dreissena polymorpha for biomonitoring. Indeed, this bivalve has a huge filtration capacity leading to an accumulation of chemical and biological contaminants in its tissues. The DROPPE (The dreissene as purifier to protozoa in WWTP effluent) project aims to test the depurative capacity of the bivalve in terms of protozoa’s contamination in WWTP effluents. To answer of this issue, it’s necessary to determine if D. polymorpha is able to live in good health in the multi-contaminated conditions in WWTPs effluent and D. polymorpha is able under these conditions, to bioaccumulate protozoa. For this purpose, two experiments were performed: 1- Zebra mussels were caged in the WWTP’s outlet channel (Charleville-Mézières, France) for 28 days. We studied morphometric parameters, filtration capacity, energetic reserves, enzymes related to oxidative stress (Superoxide dismutase, Catalase, Glutathione S-Transferase and Glutathione Peroxidase) at biochemical and molecular levels. The results suggest that D. polymorpha can maintain itself in effluent for 21 days. 2- D. polymorpha was exposed to different concentrations of protozoa (100, 1000 and 10000 protozoa per bivalve per day) for 21 days followed by 21 days of depuration in laboratory conditions. Detection of oocysts and cysts in tissues and haemolymph of bivalves was carried out by molecular biology techniques. The results highlight a time-dependent and dose-dependent bioaccumulation of protozoa by D. polymorpha. Moreover, the parasite load remains stable during the 21 days of depuration, suggesting that these mussels could be used as a biological contamination Considering these results, Dreissena polymorpha seems to be a promising tool for protozoa depuration. Keys words: protozoa, wastewater treatment plant, bivalve, depuration

516 Aquatic macrophytes potential for the removal of water contaminants - The Green Liver Application

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Reservoirs are aquatic environments that are impacted by anthropogenic activities. The main activities around reservoirs in Brazil are agriculture and settlement. Agriculture and increase of nutrients can result in cyanobacterial blooms and cyanotoxins contamination; and settlements can result in inputs of several contaminants such as pharmaceuticals. Iraí Reservoir, located in South of Brazil, is
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**

517 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 environmental AMR is the high cost and organization 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 of this 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.

518 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 the relative importance of AMR generation 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 industrialisation of antibiotics. The reasons (e.g., antibiotic misuse, agriculture) for this rapid emergence continues to be debated. It is known that soil pollution is inherently linked to co-selection for ARGs yet limited information exists on large scale, multi-contaminated sites. This study tests 24 locations across the North East of England to evaluate AMR in urban and rural soils with low and high pollution levels. We present pioneering high-capacity quantitative PCR profiles of 230+ antibiotic resistance genes (ARGs) and mobile genetic elements (MGEs). The project has coupled ARGs and MGEs to comprehensive geochemical datasets including 12 metals (total and bioavailable), 16 PAHs, and nutrient conditions to understand the evolution and dissemination of modern antibiotic resistance due to pollution.

519 Dissemination of extending-spectra β-lactamase E. coli carrying multidrug resistance and virulence factors in tropical rivers receiving hospital effluents
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The occurrence and dissemination of antibiotic resistant bacteria and their resistance genes from clinical settings to environmental compartment 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 carried out 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 disinfection. Additionally, the occurrence of the contribution of ESBL and CRE in sub-Saharan African Countries are very limited. The aim of this research is to assess the role of untreated hospital and urban wastewaters on the biological contamination of urban rivers receiving systems in the city of Kinshasa, Republic Democratic of the Congo. 147 E. coli strains resistant to 3rd generation of β-lactams (ESBL) were isolated from water samples sampled along 5 rivers receiving hospital effluents. They were analysed for their clonality and the carrier of multidrug resistance and virulence genes. The results highlight a high level of clonality in strains (67 clones) and an important level of multidrug resistance regardless the sampling point. 53% of E. coli resistant to the 3rd generation of β-lactams were also resistant between 6 to 8 antibiotics. 14% of ESBL producers also carried virulence genes factors linked to E. coli pathotype determination. The genotypes, ST1, ST12 were carried by 1.3%, 5.4%, 2.7% and 6.8% of the strains. These results indicate the human and environmental potential risk of tropical urban rivers. Indeed, ESBL strains carried by urban rivers are associated to resistance against numerous antibiotic classes (i.e. inhibitors combination, 4th generation of β-lactams, monobactams, carbapenems, aminoglycosides, tetracycline, quinolones and phenicol classes) and may also carry virulence genes factors. The presence of multi-drug resistant E. coli are not only limited to untreated hospital wastewater discharge in urban receiving system and are widely distributed along the river, thus highlighting the risk of surface water use.

520 Methods for determining selective endpoints of antimicrobials
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Antimicrobial resistance is one of the most significant threats to modern society. Use, misuse and overuse of antibiotics clinically and in the community; in agriculture and in aquaculture results in antibiotics and antibiotic resistant bacteria being released into the natural environment. Environmental concentrations of antibiotics are very low (ng/L range), but recent studies have shown that these concentrations may be sufficient to select for antimicrobial resistance. Currently, antibiotics are not risk assessed in terms selection for antimicrobial resistance in situ. This is largely because there is no standardised ecotoxicological assay which can be used to select for resistance endpoints. This work compares previously published methods for determining and predicting selective concentrations of antibiotics to two novel methods developed in this study. The first method tracks resistance gene prevalence over time in a complex community using qPCR, and the other is based on reduction in growth of a complex community. Results show that predicted no effect concentrations (PNECs) derived using standard ecotoxicological assays are not always protective against resistance selection. Currently, no standardised method for selective endpoint determination is always protective of the other; though there is good agreement between PNECs (PNECs for resistance) published previously and PNECs determined in this study. A novel method, based on growth of a complex community, is proposed for environmental risk assessment as it can be easily standardised, can rapidly generate selective endpoint data, and results show good agreement with more indepth data which tracks resistance gene prevalence over time. Results show that continued data generation and method optimisation is required to develop a reliable assay for determining PNECs for environmental risk assessment of antimicrobials.

521 Determining the minimal selective concentrations of macrolides in a complex microbial community
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Antibiotic resistant bacteria and their resistance genes transferred horizontally throughout the environment. Continuous release of antibiotics from human activity can and does lead to measurable concentrations in surface waters (ng/L - µg/L), however these are lower than minimum inhibitory concentrations (MICs) and concentrations used in the clinic. Due to these relatively low concentrations, until recently it was thought that selection for resistant bacteria did not occur within the environment. Research published in 2011 and 2014 by Gaze et al. showed selection at low environmental concentrations using single species assays. The macrolide antibiotics, azithromycin, clarithromycin and erythromycin, were added to the European Commission’s Water Framework Directive’s priority substances
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 respiration genes (ermC, mefA, mrx,M, msd and mef [fus]) in all 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 ermA gene shows a selective response at the lowest concentration for all three macrolide antibiotics. No significant selection is seen for emrF 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 mobilome.

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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, chlorotetracycline 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 high 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 β-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 (eg. int1, 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

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Microplastics (MPs) defined as pieces 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 MP 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 (34–4,800 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 that of MPs. There was no apparent pattern of spatial distribution. Although a spike in MP particle numbers was found in May2015 and May2016, there was 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 microplastic contamination issue, more standardized sampling and extraction procedures need to be developed.

524 Do nanoparticles cause stress effects on microalgae? An infrared spectroscopy study.

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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 specific nanoparticles on living organisms are not yet well 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 microalga (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, nanoparticles are collected for infrared analysis. In parallel, biological parameters (growth and genes expression) are relieved to indicate if the induced stresses imply cytotoxic effects or molecular effects on the organism. The multivariate analysis highlight that microalgae responses are stress dependent. Thus, infrared spectroscopy could be a new method to analyse stress effect on microalgae and particularly nanoparticles. Interaction with nanoparticles seems to induce an overproduction of astaxanthin biosynthesis pathway genes. In conclusion, infrared and biological data relationships could explain interaction mechanisms between nanoparticles and microalgae. Keywords: nanoparticles, infrared microspectroscopy, effects monitoring [1] von Moos N & Slaveykovka VI. 2014. Oxidative stress induced by inorganic nanoparticles in bacteria and aquatic microalgae–state of the art and knowledge gaps. Nanotoxicology. 8(6): 605-630.

525 Ecotoxicological evaluation of high-generation cationic pamam dendrimers towards a representative organism of aquatic ecosystems

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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 nanocarriers, radially symmetric molecules with well-defined, homogeneous, and monodisperse structures that have a typically symmetric core, an inner shell, and an outer shell. Due to these characteristics, their use is being tested in the implementation of targeted therapies in biomedicine so that they might end up in environment [2]. In this study, we have investigated the effect of high-generation cationic G5-NH₂ and G7-NH₂ PAMAM dendrimers in a pokrakyotic primary producer of aquatic ecosystems, the filamentous cyanobacterium Anabaena sp. PCC7120 (Anabaena). Dendrimers significantly decreased the growth of the cyanobacterium and both dendrimers induced morphological alterations of both filaments and individual cells. Furthermore, cyanobacteria exposure to dendrimers resulted in significant alteration of physiological parameters: increase in the formation of intracellular reactive oxygen species, change in membrane integrity, membrane potential depolarization, increase in intracellular pH and alteration of intracellular free Ca²⁺ homeostasis. Dendrimers also induced alterations in the photosynthetic responses of Anabaena. In conclusion, high-generation cationic dendrimers exhibited high toxicity towards...
526 Interactive effects of carbon nanoparticles and benzene/naphtalene on marine mussels, Mytilus galloprovincialis

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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 benzene and two different types of carbon nanoparticles, [C60 fullerene and multi-walled carbon nanotubes (MWNts)], 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 GC/MS. 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 technologies targeting 15 stress response pathways. Contrasting results were obtained when comparing the uptake of carbon nanoparticles used as Co-Schwarz of mussels to MWNts 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 responses) related to the uptake of carbon nanoparticles used as Co-Schwarz of mussels to MWNts and BaP. The increase in the transcription of the genes related to the genotoxic effects, such as 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

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528 Corbicula fluminea exposure to copper oxide nanoparticles: an integrated mesocosm study

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This research was supported by the Russian Science Foundation (project no. 16-14-10115).

529 Luminescent biomonitoring via bioassays of different complexity - from cells trough enzyme reactions to proteins

V. Kratasyuk, Applications of Luminous Bacteria Enzymes in Toxicology and Ecology

A new approach in developing bacteriological luminescent enzymatic biosensors, application to toxicity bioassays, and the needed reagents has been developed. To solve the problem of how to obtain, identify, and measure the numerous chemical compounds in environmental monitoring, food product contamination, and medical diagnostics, bioluminescent enzymatic toxicity assays were proposed, wherein the bacterial coupled enzyme system NAD(P)/H:FMN-oxidoreductase-luciferase substitutes for older methods using living organisms. The immobilized reagent Enzymolum was used to facilitate and accelerate the development of the bioluminescent enzymatic microbial signal for biological and/ or ecological/toxicological assays. The reagents 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
Assaying the prooxidant and antioxidant potentials of nicotine products: Tobacco versus electronic cigarettes

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Cytotoxic oxidizing activity is determined from imbalance between the production of reactive oxygen species and the efficacy of the antioxidant defense, can be a consequence of using the nicotine products. Prooxidant properties of the tobacco smoke are accounted for by the abundance of the smoke oxidants. The antioxidant potential of the smoke is scantily addressed in the literature. However, one should take into account that any reactant in oxidation process may exhibit both oxidant and antioxidant properties depending on the reaction conditions. And it is shown that smoke constituents indeed exhibit at the same time both prooxidant and antioxidant activities. Such smoke-borne antioxidants may be assessed through both the direct chemiluminescence (CL) derived from the smoke samples as a function of the smoke tar content and using the probe CL preparations of hydrocarbon substrates being oxidized. In addition to exogenous antioxidants derived from the tobacco smoke, we studied the potential antioxidative generation directly in smokers. For that purpose, we have developed a novel assay based on the CL of luminol, which involves tobacco smoke extracts, peroxidase and amino acids. Using such a system, we have demonstrated that under physiological conditions the oxidation of the smoke tar and its individual components, e.g. catechol, in the presence of H2O2, peroxidase, and glycine affords the products (first of all, catechol-glycine adducts) whose antioxidant potential is much higher than that of initial, unoxidized, chemicals. Conversely, we have not observed any significant antioxidant activity of aerosols derived from electronic cigarettes (ECs). For ECs, the following feature is noteworthy. We have found for the first time that all ECs, regardless of their technical complexity, generate in their direct aerosol and, in some cases, even in the extract obtained from the system of reactions, and antioxidant activity. Prooxidant properties are additionally assessed by the bioindication method based on response of biotope carbonate microorganisms with color differentiation to monitor results of destructive biomolecule exposures. Variations of spectral-luminescent and photochemical properties of CL-FPs under different exposures – chemicals, temperature (1), and ionizing radiation (2) – were a convenient parameter to monitor toxicity of bioavailable and non readily bioavailable carbon in the growth medium. The research objects were strains of Alternaria alternata, Chlorella variabilis, and Trichoderma harzianum. The strains were kindly provided by O.E. Marfenina and A.E. Ivaniva, Soil Science Faculty of MSU. The filamentous fungi and agar Crapek 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, 380, 310, 350, and 450 nm). Fluorescence excitation spectra were registered for emission at 350, 440, and 460 nm. Typical fluorescence spectra of fungal samples (spore suspensions and supernatant liquids) with the UV excitation at 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 (spore suspensions and supernatant liquids) with the UV 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 chromophores like NADH and or melanins. We suggest using the intensity of the first fluorescence band in the fungal samples for rapid evaluation of spore biomass. Measured fluorescence characteristics were found correlated 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.

Effect of surface functionality on Fe3O4 nanoparticles toxicity

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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 diversity of reactions to external stimuli of their physiology 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 fulamentous 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, Chlocladosporium chlocladosporoi, and Trichoderma harzianum. The strains were kindly provided by O.E. Marfenina and A.E. Ivaniva, Soil Science Faculty of MSU. The filamentous fungi and agar Crpeke 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, 380, 310, 350, and 450 nm). Fluorescence excitation spectra were registered for emission at 350, 440, and 460 nm. Typical fluorescence spectra of fungal samples (spore suspensions and supernatant liquids) with the UV 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 chromophores like NADH and or melanins. We suggest using the intensity of the first fluorescence band in the fungal samples for rapid evaluation of spore biomass. Measured fluorescence characteristics were found correlated 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.
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The evolution of obesogen-induced phenotypes in vertebrates
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Global obesity is an escalating pandemic in western societies. Triggered by numerous metabolic and endocrine disruptions, the 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-activator receptor g (PPARg) nuclear receptor. To decipher the pathway of PPARg 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.

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Aging Extension and Modifications of Lipid Metabolism in the Monogonont Rotifer Brachionus koreanus under Chronic Caloric Restriction
M. Lee, J. Park, J. Lee, SungyunKwan University

Tributyltin (TBT) is a known obesogen that can cause 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 lifespan, increased liver fat proportion, and higher levels of PCs; and another one corresponding to control, B. koreanus. But decreases were observed in the area of triacylglycerol, the increase 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, Δ4 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.

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Lipidomic and transcriptomic changes induced by compounds enhancing accumulation of storage lipids in Daphnia magna
J. Fuentes, Institute of Environmental Assessment and Water Research IDAEA CSIC; R. Jordán, 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 lipoida. 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 biphenol A disrupt the dynamics of phospholipids and neutral glycerolipids 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, LPCs, PEAs, LPEAs, PCs, LPs, PLS, LPGs, SMs). Cluster analysis defined two major clusters, one corresponding to control, BPA and 20E 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 either 8 or 24h of TBT exposure, and to up- and down-regulated genes after 24 h of exposure to BPA, PP, or TBT. Gene ontology analysis indicated an enrichment of gene signalling pathways involved in lipid metabolism, specifically in lipid catabolic 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 Fuertes acknowledges the Ministry of Economy, Industry and Competitiveness for her fellowship (FPI-MICINN BES-2015-075023).

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Lipidomics profiling of wild fish to identify patterns associated with pollution exposure
C. Porte, IDAEA-CSIC / Department of Environmental Chemistry; M. Blanco, IDAEA-CSIC; A. Maceda-Veiga, 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 (ULPC-HRMS) to characterize the lipidomic signature of two fish species (Barbus meridionalis, Squalus laevis) collected along the Ripoll River. Sampling sites included upstream (reference) and downstream (urban and industrial discharges) areas. A total of 130 lipid species, including phosphatidylcholines (PC), PC-plasmalogens (PC-P), cholesterol esters (CE), triacylglycerols (TG), diacylglycerols (DG) and sphingomyelins (SM) were detected in the muscle tissue. Partial least squares discriminant analysis (PLS-DA) allowed a clear separation of the lipidome of fish from polluted and reference sites. Specifically, a relative increase of CEs (18:1, 20:4, 22:5, 22:6) and some PC-Ps (32:0, 36:4, 36:5, 38:6) was detected in the muscle of B. meridionalis sampled in polluted sites. In contrast, the lipidome of S. laevis from polluted areas was characterized by a significant increase of TGs and PC-Ps and a concomitant decrease of PCs with a high number of double bonds (36:5, 36:6, 38:4, 40:6, 40:7). These results suggest potential lipid oxidation of highly unsaturated PCs, particularly in S. laevis living in polluted sites together with a concomitant increase in neutral lipids (TGs, CEs), possibly due to an increase in the energy demand to respond to stress in polluted sites.

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Lipidomics profiles distinguish fish from organochlorine pesticide contaminated lakes compared to control lakes
N.D. Densov, M. Nouri, University of Florida / Physiological Sciences; K.J. Kroll, University of Florida / Physiological Science; C.I. Martynik, University of Florida / Physiological Sciences; V. Ding, Iowa State University

The organochlorine pesticide (OCP) contamination of Lake Apopka largely derived from high application use in the muck farms on the North Shore. These practices made chemicals and suggest an increase of CEs (18:1, 20:4, 22:5, 22:6) and some PC-Ps (32:0, 36:4, 36:5, 38:6) was detected in the muscle of B. meridionalis sampled in polluted sites. In contrast, the lipidome of S. laevis from polluted areas was characterized by a significant increase of TGs and PC-Ps and a concomitant decrease of PCs with a high number of double bonds (36:5, 36:6, 38:4, 40:6, 40:7). These results suggest potential lipid oxidation of highly unsaturated PCs, particularly in S. laevis living in polluted sites together with a concomitant increase in neutral lipids (TGs, CEs), possibly due to an increase in the energy demand to respond to stress in polluted sites.

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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)

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Nanotechnology: When shedding effects through agglomeration of carbon nanotubes (CNT’s) are confused with toxicity by media and the public - a case example revisited
S. Schwab, Adolphe Merkle 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 nanoparticle 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 Umweltkiller ausgemacht. www.lifegen.de/newsip/shownews.php?getnews=nm2011-11-09-3109&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. Oberg, 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 far away from seafood, honey to 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.

545 Lost in translation: Do we communicate the risks of (micro)plastics in the right way? M. Waagen, Norwegian University of Science and Technology / Department of Biology

While research on the environmental and health risks of microplastics is still in its infancy, the public has already concluded there are unacceptable risks and, consequently, demands for action. This puts environmental toxicologists and chemists in an uncommon position, namely that public awareness of a potential environmental issue is way ahead of an evidence-based assessment of the actual risks. To further complicate the matter, researchers face a fundamental dilemma: Current narratives on the negative implications of (micro)plastic pollution create public awareness and promote change towards more sustainable economic practices, e.g., with regard to circular economy. However, these narratives are in many cases not backed by scientific evidence. The question is now: How can we promote positive societal change and at the same time stay true to the scientific principles? In my talks, I will not present final answers to this question but rather provide a diagnosis of the recent microplastics debate. I will argue that plastic pollution represents a challenge to our disciplines with regard to the following fundamental aspects: 1) absence of a common risk understanding, 2) bias when dealing with information-scarce situations, 3) lack of mechanisms to prioritize environmental issues, 4) lack of mechanisms for consensus-building regarding the risk of environmental stressors. I will further argue that the field of plastics pollution represents an ideal playing ground to explore, discuss and advance these aspects. This will be crucial to get our disciplines fit to deal with the wicked problems, we face in the Anthropocene.

546 Ocean Literacy – changing attitudes and behaviour of society in the face of the problems of the oceans A. Borga, Anti-Tecnalia / Marine and Coastal Environmental Management

The H2020 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 / AquaEcology and Water Quality Management Group b Alterra; B. Deutschmann, RWTH Aachen University
There is evidence that single substances that are 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 causative 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 toxicology that accounts for the cumulative 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, hydro morphological 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 “toolbox”, 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, heuristic and formalized assessment called WOE, and how the concept could be operationalized. In the presentation, we will provide 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.

551 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 / Department Bioanalytical Ecotoxicology; W. Brack, Helmholtz Centre 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 effects on human and environmental health. There is indeed a concept 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” can be found. In addition to toxic chemicals, other non-chemical stressors such as habitat degradation, nutrient pollution, oxygen depletion, pH shifts, hydro morphological 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 “toolbox”, 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, heuristic and formalized assessment called WOE, and how the concept could be operationalized. In the presentation, we will provide 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.

552 Application of new statistical distribution approaches for mixture risk assessment A. Kortenkamp, Brunel University London; M. Faust, Faust & Backhaus Environmental Consulting; S. Ermel, Brunel University London / Institute of Environment, Health, and Societies; D. De Zwart, DIz Ecosoft / Centre for Sustainability Environment and Health; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; 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 regulatory values is successively re-evaluated when cumulative exposures are judged to be acceptable on the basis of chronic and acute 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 exposure dataset and the environmental conditions. The environmental concentration (EC) distribution, from which the ecoTTC value is derived is calculated. The lowest percentage. Other types of chemical toxicity distribution (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 risk quotient 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.

554 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; M. Faust, Faust & Backhaus Environmental Consulting; S. Ermel, Brunel University London / Institute of Environment, Health, and Societies; D. De Zwart, DIz Ecosoft / Centre for Sustainability Environment and Health; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; T. Backhaus, University of Gothenburg / Department of Biology and Environmental Sciences

Experimental mixture studies have shown that the toxicity of a mixture is usually greater than that of the separately tested components. In some cases, the mixture effect can be additive 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 work flow scheme for performing human and ecological mixture risk assessments (MRA) in the context of the 90% worst case of multiple pollutants in European rivers. The data reported from studies involving NTA to inform exposure and risk. The absence of guidance may consequently lead to difficulties in prioritizing risk assessment activities. In this study we examine the state of the science with respect to NTA, and present a summary of the merits and limitations for exposure assessment and risk assessment. These preliminary observations are then used to propose an initial framework for the application of NTA 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 substantive contribution to combined exposures. We also assessed the possible 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-Starke, RWTH Aachen University / Institute for Environmental Research / Institute for Environmental Research; S. Bär, 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. Ottermann, M. Röß-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. Ullich, 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 potencies. 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 (TERsingle, TERsingle/TERsingle). Mortality risk indices were calculated based on the concept of 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 grow 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 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 SoilEcotoxicology and Risk Assessment of Terrestrial Ecosystems (I)

556 Time-dependent effects of two fungicides and their mixture on enzymatic and earthworm communities under field conditions

J. Amoss, S. Bart, INRAAgroParisTech; C. PELOSTON, INRA (Institut National de la Recherche Agronomique / AgroParisTech)

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 natura, where multiple stresses occur. In this study, we assessed the effects of two commercial formulations of fungicides, i.e., Cuprafo Micro® (composed of 500 g a.i. metconazole and 133 g l1 dimethoatrin), 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 laminar method. Our result showed a lower Shannon index for earthworms in the treatment with the mixture of both pesticides (i.e., 1.51 ha1 of “Swing Gold®” and 4 kg ha1 of copper) and in the treatment with the “Swing Gold®” at ten times the recommended dose (i.e., 15.1 ha1) after one and six months. We also found a lethal effect of “Swing Gold®” on anecic earthworms at t6, while an effect of copper on anecic earthworms was only observed observed at t12. We showed no overall significant difference in total feeding activity, enzymatic activity 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 - for the 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: Cuprafo Micro®, “Swing Gold®”, agroecosystems, feeding activity

557 Toxicity of imidacloprid and thiacloprid towards four Collembolan species C. Lima, 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 invertebrates. In general, F. fimetaria is present in most of natural and agricultural soils worldwide, and has a sexual mode of reproduction. Following a suggestion of the ring test to use different strains and test species to assess the toxicity of contaminants, in this study two additional species, Heteromurus nitidus and Sinella curvisetosa, were used together with F. candida and F. fimetaria to determine the toxicity of imidacloprid and thiacloprid in Lufa 2.2 soil. The tests aimed at answering two main questions: (i) Is there a difference in the sensitivity to neonicotinoids between the different species? (ii) Are these species suitable for assessing the toxicity of neonicotinoids? Imidacloprid was most toxic, with F. fimetaria presenting around the same sensitivity as F. candida for survival (LC50 = 0.56 mg/kg dry Lufa 2.2 soil) and a slight difference in the sensitivity for reproduction, with EC50 for F. fimetaria of 0.10 mg/kg dry soil and for F. candida of 0.26 mg/kg dry soil. H. nitidus was slightly less sensitive than an LC50 of 1.6 mg/kg dry soil and an EC50 of 0.40 mg/kg dry soil. Thiacloprid was tested on S. curvisetosa. F. candida and H. nitidus, with a survival of the first one being least sensitive (LC50 = 27 mg/kg dry soil), followed by F. candida (LC50 = 5.2 mg/kg dry soil) and H. nitidus being the most sensitive with an LC50 of 2.3 mg/kg dry soil. Thiacloprid was more toxic to the reproduction of S. curvisetosa (EC50 = 2.6 mg/kg dry soil) followed by F. candida (EC50 = 1.5 mg/kg dry soil), and H. nitidus (EC50 = 1.3 mg/kg dry soil). The different species tested presented similar sensitivity for neonicotinoids, with the exception of S. curvisetosa. The results suggest a specific mode of action of thiacloprid towards reproduction, a trend that has been found in all tests, except for H. nitidus that presented around the same sensitivity to both survival and reproduction. The species tested presented good control performance and consistency in the results, pointing towards a possibility to be used in toxicity tests.

558 Dirty dancing: measuring mito movement responses to pesticide residues J. Writton, Environment Dept, University of York / Environment; T. Alvarez, EcoRisk Solutions Ltd / Dept of Ecological Sciences; M. Reid, HSE Health and Safety Executive / Chemicals Regulation Division; G. Wey, University of York / Environment Department; R. Ashauer, University of York / Environment For a pesticide to be registered for use, the lethal and sublethal effects on non-target arthropods (NTAs) must be studied. Sublethal effects such as behavioural changes have been reported in NTAs exposed to some pesticides, with avoidance behaviour – where individuals display a change in position or orientation or a reduction in their exposure to a pesticide – being of particular interest. More research is necessary to better understand pesticide avoidance behaviour so that population consequences of such behaviour can be estimated. We aimed to develop an efficient method to quantify changes in movement behaviour and identify avoidance behaviour in relation to pesticide exposure in the predatory mite Typhlodromus pyri, a model species and natural predator found in fruit orchards across the globe. Using video analysis, we exposed individual adult mites to 3 insecticidal active ingredients (acetamiprid, deltamethrin, dimethoate), each at 3 concentrations, and evaluated mito movement behaviours when exposed to these in comparison to a control arena. We found that distances walked by mites were reduced by up to 87% compared to the control when exposed to 0.1 μg mL1 deltamethrin, and that 54% of individuals exhibited a significant change in position or orientation or a reduction in their exposure to a pesticide following a significant change in position or orientation or a reduction in their exposure to a pesticide. We also showed that the mean distance covered increased by 11% when exposed to 18 μg mL1 dimethoate. No individuals showed a significant change in position or orientation or a reduction in their exposure to a pesticide when exposed to 18 μg mL1 acetamiprid, 50 g mL1 deltamethrin, and 0.45 μg mL1 dimethoate. The mean distance covered increased by 11% No individuals
exhibited avoidance behaviour when exposed to acetamiprid or dime thoate. 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 taste arena glue boundary due to attempts to escape the arena. Our results complement existing knowledge of sublethal pesticide effects on T. pyri by quantifying movement behaviour changes in T. pyri. We are also adding to the knowledge relating to 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? T. Natal-da-Luz, CFE - Centre for Functional Ecology / Department of Life Sciences; T. Gevaert, CFE Centre for Functional Ecology and Hogeschool Gent, Education, Health and Social Work, University College Ghent; C.S. Pereira, CFE - Centre for Functional Ecology / Department of Life Sciences University of Coimbra; M. Amorim, EFSAS - European Food Safety Authority / Pesticides; J. Sousa, University of Coimbra / Department of Life Sciences 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 implemented 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 contaminated prey, via predation, and not to 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 the active ingredient (log Koc 1519, 1519 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 variable in the case of H. aculeifer. As the exposure in the reproduction test with H. aculeifer is simultaneous for H. aculeifer and their prey, the test should be adapted to include oral exposure to contaminated food. By comparing analysed residues in worms and calculated used to calculate degradation curves and the current concentration in soil at the date

560 Plant protection products in agricultural soils - Do active ingredients show a comparable pattern in worms and in soil? T. Schmidt, IES Ltd / Ecotoxicology; H. Viric Gasparic, University of Zagreb Faculty of Agriculture / Department for Agricultural Zoology; R. Bazok, University of Zagreb Faculty of Agriculture / Department of Agricultural Zoology; S. Kimmel, Innovative Environmental Services (IES) Ltd. / Ecotoxicology; S. Hoger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology The environmental risk assessment of plant protection products on soil organisms is mainly based on the outcome of laboratory and extended laboratory studies (EFSA 2017). However, the link from the laboratory to realistic field conditions over several seasons is not well established. Currently no validated trigger for bioaccumulation in terrestrial organisms is available (EFSA 2017). One possible approach for filling this gap is proposed by combining experimentally determined residue data from earthworms and data from degradation studies in the field. Earthworms were sampled at different seasons from eight fields in Croatia and analysed for 26 active ingredients. The concentrations of 26 analysed active ingredients were 0.000-0.247 mg/kg worm fresh weight with a mean of 0.005 mg/kg. The percentage of samples with values below the limit of detection (LOD), values below the limit of quantification (LOQ) = 0.001 mg/kg) and values above LOQ was 29, 42 and 29 %, respectively. Based on publicly available draft assessment reports from EC and EFSA, degradation parameters (DT50, DT90) were used to calculate degradation curves and the current concentration in soil at the date of worm sampling. By comparing analysed residues in worms and calculated concentrations in soil, a substance-specific bioaccumulation factor could be calculated. In the case of imidacloprid, the analysed residue levels in earthworm samples from the fields tended to increase with time whereas the calculated concentrations in soil decreased with time as expected, resulting in a supposed increasing bioaccumulation potential of imidacloprid under field conditions. The procedure proposed here – in the absence of analysed soil data – is a simple estimation which combines field history data with data from publicly available draft assessment reports. This approach may be useful for the assessment of the bioaccumulation potential of an active ingredient from a plant protection product under realistic field conditions.

561 PBT assessment of substances - Proposal of a trigger value for bioaccumulation in terrestrial oligochaetes M. Simon, Fraunhofer IME / Applied Ecology; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; P. Egeier, ECT Oekotoxikologie GmbH / Ecotoxicology; C. Schlechtriem, ECT Oekotoxikologie GmbH / Institute for Technology Assessment and System Analysis ITAS; J. Römbeke, ECT Oekotoxikologie GmbH; B. Karaoglan, German Environment Agency UBA; A. Wiemann, UBA Umweltbundesamt; W. Drost, Federal Environment Agency (UBA) / Chemicals Assessment and regulation of PBT (Persistent, Bioaccumulative and Toxic) substances are necessary to ensure a high level of protection of human and animal health, and of the environment. In aquatic organisms, trigger values for the identification of bioaccumulative (“B”) and very bioaccumulative (“vB”) substances are bioconcentration factors (BCF) of >2000 and >5000, respectively, obtained from fish flow-through studies according to OECD 305. However, Annex XIII of the REACH regulation does not define similar trigger values for bioaccumulation in terrestrial organisms. The objective of this project is to provide a suitable data base which will help to clarify how to address terrestrial bioaccumulation in the B assessment and to define trigger values for the bioaccumulation factor (BAF) obtained from bioaccumulation studies with terrestrial oligochaetes according to OECD 317 that are comparable to the BvB criteria in the scope of the PBT guidance revision. For this aim, the study comprised the following soil via dietary 1) Literature research on available bioaccumulation factors (BAFs) both in open scientific literature and in regulatory data from several OECD 317 studies and performance of correlation analysis between soil-substance-properties, BCF and BAF values. 2) Performance of bioaccumulation studies according to OECD 317 with the earthworm Eisenia fetida using the four model substances endosulfan, methoxychlor, o-endrin and PCB153. 3) Proposal of a trigger value for identification of bioaccumulative substances in terrestrial organisms based on the literature research and the performed experimental results. The results clearly showed that organic carbon content of the test matrix used in the tests influences the BAF, whereas no clear correlations between log Kow, log Koc or similar substance properties and the BAF were observed. Additionally, no correlation was found between the substance-specific BCF from fish studies and BAF determined with earthworms. Therefore, lipid- and Cₐ-normalized BSAF should be used for the assessment of terrestrial BAF. Kinetic BSAF from both experimental studies and literature-derived values ranged from 0.21 to 14.8. Based on the data evaluated in the present work, a BSAF trigger value of 1.00 is proposed as a general trigger to include bioaccumulative substances in Tier II. Other approaches like non-depurated residues at the end of the elimination phase are discussed.

Challenges, methodological developments and practical solutions for Social Life Cycle Assessment in industry and policy

562 Developments and recommendations on the practical use of Social LCA S. Di Cesare, CIBAD / Department of Economic Studies; A. Zamagni, Econoinnovazione / LCA and Ecodesign Laboratory; J. Garcia, SCORE LCA; F. Silveri, University of Chieti-Pescara / Department of Economic Studies; A. Lanfranconi, Ecoact; L. Petti, University of Chieti-Pescara / Department of Economic Studies Social LCA is a multi-criteria, multi-stakeholder and multi-step methodology that provides useful, transparent and science-based information on social and socioeconomic performance of a product throughout its entire life cycle. In this study, a systematic literature review was carried out dealt with within these aspects: the scope of S-LCA, its purpose, the social dimension outside S-LCA (i.e., analytical tools, procedural and managerial tools, currently used for monitoring, assessing, reporting and communicating social aspects, and their main differences with respect to S-LCA); impact assessment methods; users and uses of S-LCA. In addition, an experimentation was conducted through a practical case study based on literature, with the following goals: (1) to test the applicability and practicability of S-LCA, (2) to highlight the methodology to identify social hotspots along the whole life cycle, and in particular in the remote phases of the life cycle, such as raw material production and end-of-life; (3) to show how those results may complete environmental LCA and other social approaches. The product chosen is a Photovoltaic (PV) Module. The analysis carried out clearly pointed out that S-LCA is an evolving field, and main developments are envisaged, both at the level of methodology and results’ interpretation and communication. More in detail, the main limits of the S-LCA methodology identified in this analysis are
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 whole process along the value chain. On 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

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A global and interdepartmental project facing constant 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 impacts determining social hotspots along the supply chain and should in addition provide information on social challenges and changes by means of negative and also 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 the defined social indicators and 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

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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 complexity and critical role in the industrial development, it is essential to ensure efficient production processes, optimizing the use of resources, valorization of byproducts, but also to explore the other stages of the production system. However, it is clear that the changes required to make industry more sustainable are not only at its operational level but also in the use of its products by customers and society at large. Therefore, a research team dedicated to sustainability and lifecycle assessment has been supporting the process and product development 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 that enables 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

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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 to sum all of social income redistribution and the sum of all productivity-reducing externalities related to an activity. It is calculated by a top-down approach using input-output data. This method can be understood as a ‘streamlined’ social LCA. We applied its two general components: the income redistribution impact (IR): the increase (or loss, if negative) in utility caused 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 Exiobase 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 existing 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 to apply a social footprint, together with a powerful tool like Exiobase, 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

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Toxicity 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 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
EQP 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 competency), 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 EQP modeling approach is deemed suitable for predicting for static vs. dynamic water concentration levels. The EQP membrane concentrations 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/narcosis). 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
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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 for toxicity testing to static or dynamic mass balance modelling approaches. Nominal concentration. However, to the apparent strong bound to medium proteins in in vitro assays, while the mass balance approach also reliable models are available to calculate the binding of neutral chemicals to lipid, protein, medium and cells, the binding of organic acids to biological matrices cannot be easily predicted. Here we applied a phase partitioning method to measure binding of organic acids to biological matrices like cell culture media and cell suspensions on the one hand, but also for direct measurement of exposure (i.e., C_{ex}). Because polymers like polydimethylsiloxane that are typically used for solid phase microextraction (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, tarzarin, warfarin, ticloxan, and gentamicin. In order to achieve equilibrium, the equilibrium between the SPME fibre 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 the concentration in cell culture media. At low chemical concentrations the results from the binding experiments were in agreement with the predictions from mass balance or modelling approach. However, saturation of the medium was observed at high chemical concentrations. At present, there are no comparable intestinal permeation data for fish available, which precludes a direct comparison of the in vitro measured rates with in vivo observation. However, exactly this unavailability of data highlights the importance of the development of such an approach for understanding the activity at the intestinal epithelium. Data derived from 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. 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 the model, which correlated with the logK_{oc}. The chamber enabled stable exposure concentrations and close to full recovery at the end of the measurement time. The in vitro exposure scenario in our system comprises a prerequisite for studying mechanisms underlying chemical permeation, such as active transport or biotransformation.

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 requirements and general use of the robot 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) compared to static exposure (24 h interval), but it was also dependent on the renewal interval for 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, the robot 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 made 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 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 (logK_{ow} = 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 the model, which correlated with the logK_{ow}. The chamber enabled stable exposure concentrations and close to full recovery at the end of the measurement time. Exposure times at the intestinal epithelium. Data derived from 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.
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 consumptions 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, guaranteeing 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 fully automated processing and avoiding 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 suspect 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 standards 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 data 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 50 different assay endpoints, the 5th percentile of the range of AC₅₀ values of a suspect was included in this hypothesis checks. Failure to obtain quantitative recoveries from soils/sediments could also preclude a reliable assessment of environmental fate properties (e.g., soil-water partitioning coefficients, soil microbiome bioaccumulation factor). Given the aforementioned limitations, the present study set out to propose a suitable preparation procedure for the multi-residue analysis of PFASs in AFF-impacted soils. A total of 89 PFASs, representing >20 distinct chemical classes previously discovered in AFFF-formulations or across AFF-impacted sites, was therefore evaluated. Multiple extraction methods were assayed to recover PFASs from AFF-impacted soils and 2N 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., 62 FTAB, 9:1:2 FTB) and aniline-based (e.g., PFHxSxAm) PFASs at such sites.

575 Investigation of perfluorooctyl and polyfluorooctyl substances in products used for building industry as well as industrial textiles and their possible contribution to water contamination

R. Jansoo, Hochschule Fresenius, University of Applied Sciences; S. Lebertz, SGs Institut Fresenius GmbH, T. P. Knepper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology Perfluoropolyalkyl and polyfluoropolyalkyl 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 properties (e.g., bioaccumulation and toxicity), their fate has been widely discussed. Due to their water, dirt and moisture repellent 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 2N 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. FTIOHs were even detected in concentrations up to several mg/kg. However, FTIOHs need to be further investigated since they may be false positives resulting from the low selectivity of utilized transition for TOXH quantification. In addition to performed investigations, rinse of samples of e.g. buildings should be performed to prove suggested entrance pathways. Furthermore, additional method development has to be performed in order to better illustrate entrance pathways.

576 The growing role of seafood consumption for exposures to legacy PFASs Evident in Longitudinal Birth Cohorts from the Faroe Islands

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Rapid declines in legacy poly- and perfluorooctyl 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 important for mitigating future exposure. Median serum 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 (PFACAs) with nine or more carbons (C₉) were strongly associated with mercury in children’s hair, a well-established proxy for seafood consumption, especially perfluoroundecanoic acid (PFUnDA, r = 0.72). Toxicokinetic 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, declines in SPFASs exposure after 2000 were achieved by the rapid phase out of PFOS and its precursors in consumer products.

577 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 replacement of AFFFs in firefighting training activities or fire emergency response, 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 perfluorooctyl 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 not be compensated through some dilution-related to the lack of matching internal standards. If consistent whole-method recovery cannot be ensured in a set of environmental samples of variable physicochemical characteristics, any comparison drawn between samples (e.g., inter-site differences) could be questionable. Failure to obtain quantitative recoveries from soils/sediments could also preclude a reliable assessment of environmental fate properties (e.g., soil-water partitioning coefficients, soil microbiome bioaccumulation factor). 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 2N 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., 62 FTAB, 9:1:2 FTB) and aniline-based (e.g., PFHxSxAm) PFASs at such sites.
Membrane-water partition coefficients to aid PFAS risk assessment.
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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 ionicogenic, and act as contaminants. As a result, octanol-water partition coefficients (K_{ow}) cannot be determined experimentally. Due to the lack of experimental data, QSARs to predict K_{ow} are not properly calibrated for any perfluorinated ionicogenic compounds. Furthermore, the dissociation constant (pK_{a}) 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 K_{ow} 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, computes in a reliable and simple manner the charge densities into account, is able to predict for emerging PFASs both the membrane-water partitioning (K_{ow}) of the ionogenic species, and the predictions on pK_{a}. Whereas COSMOTherm accurately predicts K_{ow} for cationic hydrocarbon surfactants, it strongly misinterprets the membrane affinity of anionic perfluorinated surfactants. It does show promising predictions on pK_{a} of alternative PFASs, e.g. Genzyme B2PFPE, that can potentially exert a great toll on electrons of any ionizable group, thereby e.g. rendering perfluorooctane sulfonamide (PFOSA) to be a strong acid, whereas recent studies and reports suggested PFOSA to be a neutral PFAS.

578 Impacts of ocean circulation on the marine PFOS burden in an era of geographically shifting emissions
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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 releases, and its occurrence merely requires that 3D-molecular models be able to transport charge densities into account, is able to predict for emerging PFASs both the membrane-water partitioning (K_{ow}) of the ionogenic species, and the predictions on pK_{a}. Whereas COSMOTherm accurately predicts K_{ow} for cationic hydrocarbon surfactants, it strongly misinterprets the membrane affinity of anionic perfluorinated surfactants. It does show promising predictions on pK_{a} of alternative PFASs, e.g. Genzyme B2PFPE, that can potentially exert a great toll on electrons of any ionizable group, thereby e.g. rendering perfluorooctane sulfonamide (PFOSA) to be a strong acid, whereas recent studies and reports suggested PFOSA to be a neutral PFAS.

579 PFAS pollution at airport sites: point and diffuse sources, fate and transport and remediation
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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 aqueous 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 used to understand the partitioning and leaching behavior of PFAS in the environment. The case studies are designed to work as a framework for the remediation of water and soil to be used in the field and laboratory studies, 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, computes in a reliable and simple manner the charge densities into account, is able to predict for emerging PFASs both the membrane-water partitioning (K_{ow}) of the ionogenic species, and the predictions on pK_{a}. Whereas COSMOTherm accurately predicts K_{ow} for cationic hydrocarbon surfactants, it strongly misinterprets the membrane affinity of anionic perfluorinated surfactants. It does show promising predictions on pK_{a} of alternative PFASs, e.g. Genzyme B2PFPE, that can potentially exert a great toll on electrons of any ionizable group, thereby e.g. rendering perfluorooctane sulfonamide (PFOSA) to be a strong acid, whereas recent studies and reports suggested PFOSA to be a neutral PFAS.

580 Environmental fate and exposure models: Advances and challenges in 21st century chemical risk assessment
M. Scheringer, ETH Zurich / Institute for Chemical and Bioengineering; A. Di Guardo, University of Insusibria / Department of Science and High Technology; T. Gou, TGI, Environmental Research / Safety and Environmental Assurance Centre; M. MacLeod, Stockholm University / Department of Environmental Science and Analysis Chemistry

Environmental fate and exposure models are a powerful means to integrate information on chemicals, their partitioning and degradation behaviour, the environmental scenario and the emissions in order to compile a picture of chemical distribution and fluxes in the multimedia environment. A 1995 pioneering book, resulting from a series of workshops among model developers and users, reported the main advantages and the identified needs for research in the field of multimedia fate models. Considerable efforts were devoted to their improvement in the past 25 years and many aspects were refined: the inclusion of nanomaterials among the modelled substances, the development of models at different spatial and temporal scales, the estimation of chemical properties and emission data, the incorporation of additional environmental media and processes, the integration of sensitivity and uncertainty analysis in the simulations, etc. However, some issues are still challenging and require research efforts and attention: the need of methods to estimate partition coefficients for polar and ionizable chemical in the environment, a better description of bioavailability in different environments as well as the requirement of injecting more ecological realism in exposure predictions to account for the diversity of ecosystem structures and functions in risk assessment. Finally, to transfer new scientific developments into the realm of regulatory risk assessment, we propose the formation of expert groups that compare, discuss and recommend model modifications and updates and help develop practical tools for risk assessment.

581 Development and update of environmental exposure assessment tool EUSES for REACH and BPR Regulations
S. Frattini, ECHA-European Chemicals Agency; R. Cesaaitis, European Chemicals Agency; H. Schimmelpfenning, European Chemical Agency ECHA; H. Magaud, European Chemical Agency ECHA

Introduction
Both REACH Regulation and the Biocidal Products Regulation requires that the chemicals addressed by the respective legislations are used safely. Both regulations require environmental exposure and risk assessment of chemicals. EUSES (the European Union System for the Evaluation of Substances) is a tool developed by authorities in the 90’s to support environmental exposure and risk assessment chemical in line with the methods described in the technical guidance document (TGD) that has treated the assessments practices for these three policy areas at that time. EUSES last relevant update was in 2004, so that the tool is now partly outdated. An update is needed since the availability of an up-to-date tool is critical for both REACH and the BPR to function efficiently, both for applicants/registrants, MScAs and ECHA. EUSES has several modules (release estimation and distribution, multimedia exposure assessments and functions in risk characterisation). Fate and distribution module (including interaction with the release estimation module) as well as the release estimation module are in the focus of the update process. Update needs and developments
The release, fate and distribution modules should be improved based on the development having occurred over the last years, in particular: Implementation of existing and newly developed emission scenario documents (EDS) for biocides in the release module of EUSES. Complementing the release scenarios covered by EUSES. Expanding the applicability domain and exposure...
estimation capability of the model. Alignment of the exposure estimation methods to the scientific developments over the last 20 years. Implement the new module of SimpleTreat (4.0). Exploring how to address site specific assessment by EUSES, like for example within authorisation process. A process has been initiated by ECHA with stakeholders to assess the need for update of EUSES. The kick off for this update process is a workshop with stakeholders which will take place in ECHA, in April 2018. The expected outcome of the workshop is the identification and prioritisation of potential further research panels. Workshop outcomes planned to be available before May 2018 and will be presented to the wider audience of the SETAC conference.

582 Advances in exposure assessment of fertilizers: development of a fertilizers environmental exposure tool and generic exposure scenarios under REACH L. Della Pietra, Fertilizers Europe; S. Navis, Arche consulting / Laboratory of Aquatic Ecology, Evolution and Conservation; R. Puska, Yara Suomi; M. Bjergan, Yara International ASA; K. Oorts, ARCHE Fertilizers are considered as formulations and therefore no registration or chemical safety assessment is required under REACH. However, use of the individual constituents needs to be covered in the chemical safety reports (CSR) of the respective substances. Currently, exposure and risk assessment of fertilizer uses is mainly described in a qualitative way because of the lack of appropriate environmental release categories (ERCs) and exposure models. Under the umbrella of the European Fertilizer Industry and the FARM REACH consortium, the fertilizers sector has developed a fertilizer sector uses map. In addition, four sector specific ERCs (SPERCs) were 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 sources for directed emissions (e.g. greenhouse gases) were considered to test and the framework proposed for exposure assessment. The scheme as a serves as a useful guide to 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. Tal 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.

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.odos, Eldons; T. Halmons, 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 polyol esters. These thickeners impart different technical properties on the final grease. Although individually registered under REACH as isolated substances (i.e. extracted from base oil), grease thickeners are typically manufactured in a grease base and seldom exist except within a grease base. Under normal environmental conditions, grease thickeners would be expected to remain within the grease base because they are not available to cross an environmental compartments (soil, water, sediment) were established. 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, but the tool allows for assessment of other inorganic and organic substances in fertilizers as well. Conceptually, the tool has been based on existing REACH exposure modelling, but is adapted for fertilizer uses. It is also adapted for information from other chemical legislations. 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 assessment of fertilizers or REACH regulation. Further information on the project, including downloads of the FEE tool and SPERC factsheets can be found via www.fertilizerseurope.com.

584 The durability criteria: a pragmatic and sound approach to the exposure assessment of nano-enabled agrochemicals M. McLachlan, University of Victoria / Department of Environmental Geosciences; P.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). Kookana 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–a non-persistent, hydrophobic and semi-volatile pollutant was chosen as the test chemical. We used physico-chemical properties recommended by Schenker et al. and global historical emissions estimates developed by Breivik and co-workers to drive the global multimedia fate and transport model BETr Global. The fugacities of PCB 153 in air, water and soil, modeled at a spatial resolution of 3.75° × 3.75°, were re-gridded to give the historical fugacity records on the basis of individual countries. These were entered into the bioaccumulation and exposure model ACC-HUMAN, which modeled the concentrations of PCB 153 in fish, meat, dairy products and human milk. The human diet in ACC-HUMAN was parameterized for each country based on the WHO Global Environment Modeling System (GEMS) portfolio diets. The modeled concentrations of PCB 153 in human milk were compared with the concentrations measured in the WHO/UNEP global monitoring program for POPs. The predicted and observed concentrations were highly correlated, with a correlation coefficient of 0.76. For 49 out of 78 data points, the conclusions 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, 8434-8441. K. Breivik et al., Environ. Sci. Technol., 2016, 50, 799–805. M. MacLeod et al., Environ. Pollut., 2011, 159, 1442–1445. G. Czab and M.S. McLachlan, Environ. Toxicol. Chem., 2004, 23, 2356–2366. https://undatacatalog.org/dataset/gemsoffood-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


The EC4SafeNano project aims to address the lack of regulatory testing and validation of CMR (Classification, Labelling and Packaging) data for nanomaterials. This project will develop a database of validated test methods and test data, as well as a set of guidance documents and training materials to support regulatory decision-making.

587 Building a Risk Assessment Framework for Nanomaterials in Canada

M. Sauve, Environment Canada; A. Shahsavaran, Environment and Climate Change Canada

Despite the potential benefits associated with the use of nanomaterials, concerns also exist about 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. 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. RAF will also be used to promote the understanding of environmental risks posed by emerging contaminants like ENMs. 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 posed. 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 as a graphitized version of the product (GAFN). The study implemented regulatory testing using Daphnia magna and Pseudokirchneriella subcapitata as well as hemocytes 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 as 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.

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. The inventory 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 trainings available only 5. This aspect will be addressed in the EC4SafeNano project to identify the specific training needs and that can be used to promote the understanding of environmental risks posed by emerging contaminants like ENMs pose. Mytilus species have a long history of being used as sentinel organisms to characterize ecosystem health and can be used to promote the understanding of environmental risks posed by emerging contaminants like ENMs.

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 free-suspended species) may not adequately reflect 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. Mytilus species have a long history of being used as sentinel organisms to characterize ecosystem health and can be used to promote the understanding of environmental risks posed by emerging contaminants like ENMs.

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 posed. 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 as a graphitized version of the product (GAFN). The study implemented regulatory testing using Daphnia magna and Pseudokirchneriella subcapitata as well as hemocytes 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 as 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

D. Ferron, R. Catalano, Marseille Université; J. Hubaud, Heliocience; V. Bartolomei, S. Motellier, CEA Liten; D. Boutry, CEA - Grenoble; L. Hedouin, CNRS CRIJCE; C. Santella, CNRS/CEA/Aix-Marseille Université / Bioscience and biotechnology Institute of Aix Marseille; P. Hennebert, INERIS; A. Pisinio, IBIM CNR Palermo; S. Lehmann, University of Grenoble Alps; J. Labille, CNRS Sunscreens are of emerging concern regarding both human and environmental health. While TiO2 nanoparticles used as UV-blockers may offer a safer alternative to organic filters, their fate and impact and resulting regulation are still under consideration, largely related to the potential risk of nanotechnology-based products. After leaving the skin either through bathing or cleaning, the TiO2...
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 nano-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 parameterisation, 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 nodularins, NODs) and alkaloids (cylindrospermopsin, CYN, anatoxin-a, ANA-a and saxitoxins, STXs). The most important issues that make the CTs’ analysis challenging are: (i) 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 for the determination of multi-class CTs. As an example, analysis of 12 MCs ([D-Asp]MC-RR, MC-RR, MC-YR, MC-HyR, [D-Asp]3MC-LR, MC-LR, MC-HiR, MC-WR, MC-AL, 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. Furthermore, 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-Asp]3MC-RR, MC-RR, MC-YR, MC-HyR, [D-Asp]3MC-LR, MC-LR, MC-HiR, MC-WR, MC-AL, 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 cyanobacterial and daphnia

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Thanks to their adaptation cyanobacteria has the capacity to resist, health and human intervention. 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 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 PCC7806 was exposed to the cultures. Asp3 – D. magna were employed. In order to disentangle mutual interactions between both organisms, a co-culture chamber was designed, where two chambers were 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 PCC7806 obtained after 2 weeks culture, equivalent to 10^5 cells/mL, reduced feeding survival, and medium altered detoxification and antioxidant response as well as the energetic budget. Exposures to spent media from M. aeruginosa PCC7806 micr.- are currently in progress. Vice versa, Microcystis aeruginosa PCC7806 reacted to spent medium from 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 hydrogenperoxide 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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One of the biggest worldwide problems in aquatic ecosystems is the formation of cyanobacterial water blooms that can have adverse effects on organisms. It has been well recognized that cyanobacteria are able to produce diverse groups of toxins. Recent reports show evidence of new toxic products of cyanobacterial metabolism - retinoid compounds, but there is very limited knowledge regarding to their producers, occurrence or potential associated risks. This research provides environmentally significant information about total retinoid-like activity in the biomass of cyanobacterial water blooms as well as in their surrounding water. It documents production of compounds with this bioactivity into the surface water by various cyanobacterial species. The level of retinoid-like activity reaches values that can cause adverse developmental effects in exposed organisms. Retinoid-like activity in cyanobacterial exudates was in a very good agreement with
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, cytokine, and neuro-active compounds. Information on research production dynamics of the majority of novel cyanopeptides 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 supply plants. While it is known that many cyanopeptides are produced simultaneously from one species, the production of these potential toxins has not been explored comprehensively for cyanopeptides other than microcystins. This project focused specifically on the production and co-production dynamics of cyanopeptides 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 with high-performance liquid chromatography. The cyanopeptide 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 cyanopeptides 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.

596 Development of methods for Measuring Total Microcystins in Fish Tissue using the 2-methoxy-3-methyl-4-phenylbutyric acid (MMPB) procedure.
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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 by Cyanobacteria, Prymnesium parvum (Prymnesins), and Euglena sanguinea, including microcystins, saxitoxins, cylindrospermopsin, anatoxin-a, prymnesin and euglenophycin. The objective of the first phase of this research was to spike CW effluent and sediment samples in for 3 conjugates of microcystins (RR, 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 will be valuable 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 interfere with generation of MMPB; however, it did impact the workup/extraction procedure in ways that 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 biopolymers 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 quillajarka 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?, while decreased to 0.06 ±0.01 at pH 10.0. The hydrolysis was also influenced by 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 (HC5) derived from the SSD’s of saponins from quillajarka bark, tea seed coat, and quinoa seedcoat were 2.91 ±0.00, 0.22 ±0.11 and 22.9 ±8.54 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 originates 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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Setting the Stage to Advance the Adverse Outcome Pathway Framework through Horizon Scanning
The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); G. Hodges, Unilever / Safety and Environmental Assurance Centre SEAC; D. Knapen, University of Antwerp / Zebrasfablab Dept Veterinary Sciences; S. Munn, European Commission; E.J. Perkins, US Army Engineer Research and Development Center / Environmental Protection Division; J. Sjöholm, Umeå University / the Swedish University of Agricultural Sciences; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; M. Whelan, University of leicester / Geography; C. Willett, the Humane Society of the United States / Animal Research Issues; X. Zhang, Nanjing University / Environmental Science; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre
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% industry, and 10% other. A total of 57 open-ended 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 key themes emerged including future AOP research and regulatory initiatives. These themes were used as groupwork topics for a Pellston™ Workshop, including: AOP networks and their applications; quantitative AOPs and...
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 and expert meeting exercise and resulting FAQ/s were used to set the stage for the SETAC Pellostion 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. Knaps, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; M. Angrish, US EPA; National Center for Environmental Assessment; M.C. Fortin, Alcami / Environmental and Occupational Health Sciences Institute; I. Katsiada, Cefas / Environmental and Animal Health; M. Leonard, BOREAL SA; L. de 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. McMillan, U.S. 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 according to a set of generally accepted principles and guidelines. 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 contains expert findings, and answers to FAQs, but 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 approaches. 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. Connolly, US EPA RTP; B. Landesmann, JRCE, European Commission; C. Mackay, Unilever; C. Murphy, Michigan State University / Department of Fisheries and Wildlife; N. Pollesch, US EPA / ORD NHEERL, Mid Continent Ecology Division; J. Wheeler, Dow AgroSciences; A. Zupanic, Eawag Swiss Federal Institute of Aquatic Science and Technology; E.J. Perkins, US Army Engineer Research and Development Center / Environmental Laboratory Quality Assurance and Environmental Operations; H. Davies, WA State Dept of Health - UFZ / Cell Toxicology; G. Holmes, Unilever Research / Safety and Environmental Assurance Centre; K. Leuen, 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, biochemical, histological) data into 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.

601 Use of Adverse Outcome Pathways to Inform Decisions on Chemical Innovation, Regulation & Stewardship T. Hill, US EPA, NHEERL Integrated System Technology 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 Pellostion Workshop titled “Advancing the Adverse Outcome Pathway (AOP) 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 Pellostion Workshop 3, which was tasked with the application of practical considerations for this use of AOP constructs in regulatory decision making. The use of AOPs and related concepts has 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.
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 Scanning” 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) PellaM™ 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 PellaM 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 risk assessors and managers. Furthermore, when considering the scope of the AOP 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 national and 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, herring, cod, eelpout, blue mussel and egg from guillemot, 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 annually collected. In 1967, examination of human exposure to POPs was initiated by Karolinska Institutet in Stockholm through measurements in human milk. It has been found that POPs are present in mother’s milk and that they are passed on to the offspring. The main objectives of the monitoring program are to investigate changes over time, to estimate geographical differences and to assess compliances with set target values. Moreover, the program is designed to answer these different questions with a high level of accuracy and reliability. The results show that POPs are not only present in the environment but are also a part of the human body. The non-linear trends observed 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 POPs, DDTs and HCHs are, despite continuous decreases since the 1970’s, still higher in the Baltic Sea compared to, for example, the North Sea.


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 the environment and transport and accumulation going back to the 1980s, e.g. from coastal herring gull eggs, freshwater fish, tree leaves and to eel will be used to illustrate time trends for regulated flame retardants and their substitutes. Recommendations will be given to European and international chemical management.

606 New Uses of Archived Specimens from the U.S.A. NIST Marine Environmental Specimen Bank R. Push, C. Bryan-Sallee, W. Davis, J. Lynch, B.A. Neely, J. Ness, S. Schuur, National Institute of Standards and Technology / Chemical Sciences Division 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 Ecosystem Specimen Bank (MESB) at National Institute of Standards and Technology, 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 and 2) are feasible for the protocols taken 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-scranned genome, and 2) the discovery of using total inventory as an alternate tool to genetic species identification, have also been conducted and will be discussed.

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. Roszkowski, 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 maintain the quality 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 emit or release CECs to the indoor environment. To evaluate if the 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 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. polychlorohydrodiphenyl ethers, 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.

608 DNA banking and its relevance for biodiversity research J. Aastron, Zoological Research Museum Alexander Koening 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 fast-evolving DNA analysis methodsologies (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 uncertainties and shortcomings 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. Levy, Univ. Paris Sud / ESE UMR 8079; J. Loret, o. schlosser, CIRSEE Suez Environment; a. guillon, Suez / CIRSEE; m. favier, INSERM Institut Cochin; v. domergue dupeyrong, 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 treated water (drinking water) and bottom mineral water. 2 generations of mice (10 males, 10 females for each type of water) were fed with concentrates during a 1 year period, in order to reproduce in the animals, the exposure as a man drinking the water for over 70 years. We carried out complete chemical analysis (quantitative targeted analysis, innovative non-targeted screening by HPLC and LC-2D with mass spectrometry), offering a new vision of the contaminants diversity. In vitro bioassays were used to evaluate endocrine disrupting effects (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 micropolllutants. The presentation will show the detailed protocol, the major contaminants and challenges for ERA of chemical mixtures 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 classified 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), and 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 Chelex-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 armoracia. 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?
D. Janssen, Ghent University / Applied Ecology and Environmental Technology; C.R. Janssen, Ghent University / Sustainable Organic Chemistry and Technology; S. Huysman, Ghent University; K. Demeestere, Ghent University / School of Chemistry

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...
(ERCMs) have effects on marine phytoplankton and how effects could be explained by measured contaminant concentrations. Further we looked at the repeatability of our test results over an extended time period of 16 months. In the presented research we used extracts of Speedisk™ passive samplers deployed in and outside of the harbour of Zeelbregge (Belgium) to spike several 72 h growth inhibition tests with the marine diatom Phaeodactylum tricornutum following ISO 10253. The different growth inhibition tests were performed over a period of 16 months with tests 0, 8 and 16 months after extraction. We observed statistically significant (p < 0.05) growth stimulation of up to 6.4 ± 0.5 % and 11 ± 2 % (in the harbour) and 7.0 ± 0.5 % and 14 ± 3 % (outside of the harbour) after an extract storage time of 0 and 8 months, respectively. After 16 months the previously observed effects disappeared completely. In order to explain the differing ecotoxicological responses a targeted analysis was performed on the extract using GC-MS (n=100) to identify plant care products, pesticides, pharmaceuticals, (alkyl)phenols, phthalates and steroids. The analysis revealed that testing occurred at contaminant concentrations similar to those measured in water grab samples taken during sampler deployment. Remarkably the observed stimulation effects remained above 5 % when diluting the extracts up to 125 times. These findings suggest that P. tricornutum would remain affected by ERCMs even if their environmental concentrations would be reduced considerably. The disappearance of the observed stimulation effects after an extract storage time of 16 months led to the hypothesis that the main contributing contaminants causing stimulation must have degraded over time. In future work it would be of high interest to apply multivariate analysis (i.e. principal component analysis) to identify main contributing contaminants to the observed effects.

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; A. Lahn, Bioinformatic consultant; S. Tasselli, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; I. Sanseverino, European Commission Joint Research Centre; S. Balzano, M. Patalivo, ISPRA Institute for Environmental Protection and Research; R. Loos, European Commission Joint Research Centre; D. Marinov, T. Lettieri, 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 herbicide Diuron and Isoproturon. Signatures induced by the Cadmium or Nickel partially overlapped with the reference mixture pattern. The results show that the mixture induces a pattern similar to the ones induced by the single herbicide 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.

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 Costa, University of Coimbra / Department of Life Sciences; 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 tiered approach 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 accuracy of the lab to field extrapolation might influence the underestimation 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 6.4. 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 order to derive conceptual models allowing the extrapolation from the lab towards the field situation.

617 Risk assessment of soil organisms in field: dealing with earthworm community Y. Bayona, F. Brulle, ANSES / U3EIV; A. Bourin, ANSES / U2EIV; F. Jegou, Jeanne, ANSES / U3EIV; P. Kotschik, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; I. Sanseverino, Tasselli, European Commission  Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit

The soils used for ecotoxicity testing cover for each metal a wide range of soil properties, making the results reproducible. Metal soil threshold calculator tool: use of available data for derivation of terrestrial ecotoxicity standards. 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 allow 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 predicted no 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 models (i.e. conservative estimates and toxicity laboratory studies) to the most realistic assessment (i.e. Field studies). The recent EFSA opinion proposed a framework for risk assessment of soil organisms and definition of protection goals. Still, no statistical tool was proposed when dealing with field studies as higher tier. The aim of this communication is to test tools routinely used for regulatory risk assessment of communities. The tools for the ecological communities were mostly developed for aquatic organisms risk assessment. The multivariate analysis (PRC) is used for the community comparison. Individual populations are compared date-by-date using various statistical analysis commonly including Dunnett test and Wilcoxon test. The main weakness pointed out for the 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. Moore, ARCHE; I. Codd, Thrush; D. Napierska, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; 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 allow 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 predicted no 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 models (i.e. conservative estimates and toxicity laboratory studies) to the most realistic assessment (i.e. Field studies). The recent EFSA opinion proposed a framework for risk assessment of soil organisms and definition of protection goals. Still, no statistical tool was proposed when dealing with field studies as higher tier. The aim of this communication is to test tools routinely used for regulatory risk assessment of communities. The tools for the ecological communities were mostly developed for aquatic organisms risk assessment. The multivariate analysis (PRC) is used for the community comparison. Individual populations are compared date-by-date using various statistical analysis commonly including Dunnett test and Wilcoxon test. The main weakness pointed out for the higher tier regulatory risk assessment of chemicals for earthworm and soil communities.
effect levels from the original dose-response curves (ECx), 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. Hammers-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 currently 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 latter can be quoted as a decisive factor, but the aim of the scientific community is to identify and possibly reverse the development. Knowledge of the negative effects of pesticides on specific sites available. At the same time there is a lack of knowledge about the development of biodiversity in different agricultural landscapes because monitoring programs have not been established so far. The establishment of a systematic monitoring and a landscape risk assessment is necessary to connect future risk assessment with biodiversity as the protection target. Furthermore good ecological values must be derived and thresholds for the life operating range must be defined. When the biodiversity level falls below the thresholds measures must be carried out. Geospatial models can help to optimise sustainable agricultural practice and measures for risk mitigation. The presentation will summarise result from different projects.

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-soil 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 present an overview of the use of pesticides 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 agricultural 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 needed. For the protection of soil organisms, monitoring programs have not been established so far. The establishment of a systematic monitoring and a landscape risk assessment is necessary to connect future risk assessment with biodiversity as the protection target. Furthermore good ecological values must be derived and thresholds for the life operating range must 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

Nowaday bio-composite materials have increased automotive market penetration, which 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 taulcum, 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 essentially focus on new industrially 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. References [1] Rovanesi A., Curis 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., Drzaal 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 Klein R., Keoleian G.A., Lee E.C., Kim H.C. Woehrle M. 2016. Pedalbox support design by lifecycle design process. 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. Mistretta, University of Palermo Lithium-ion batteries (LIBs) are the dominating storage technology for electric vehicles (EVs). Different types of LIBs, using diverse cathode and anode materials, are available in the market, such as LiMnO2, Li(Ni1/3Co1/3Mn1/3)O2, 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 lithium cycle. In this context, we present a comparative environmental analysis between two materials currently used in LIBs and in general in EVs. In this context, lithium rich layered oxides from the class xLiMnO2-yLi2O (x=0.5, y=0.5) have been selected as functional cathodes. Two types of LIBs, using diverse cathode materials, have been selected as functional cathodes. Two types of LIBs, using diverse cathode materials, have been selected as functional cathodes. The results show that the LMO-NMC cell technology with an NMC cathode is the most promising one as it performs better in terms of energy efficiency and lower environmental impact. The results show that the LMO-NMC cell technology with an NMC cathode is the most promising one as it performs better in terms of energy efficiency and lower environmental impact. The results show that the LMO-NMC cell technology with an NMC cathode is the most promising one as it performs better in terms of energy efficiency and lower environmental impact. The results show that the LMO-NMC cell technology with an NMC cathode is the most promising one as it performs better in terms of energy efficiency and lower environmental impact. The results show that the LMO-NMC cell technology with an NMC cathode is the most promising one as it performs better in terms of energy efficiency and lower environmental impact. The results show that the LMO-NMC cell technology with an NMC cathode is the most promising one as it performs better in terms of energy efficiency and lower environmental impact.

624 Analysing the environmental impacts of alternative solutions for passenger transportation: LCA of a charging station for e-bicycles G. Guerriero, University of Roma Tre / Department of Business Studies; R. Salomone, L. Giuttari, G. Saija, G. Ioppolo, University of Messina / Department of Economics; M.C. Lucchetti, University of Roma Tre / Department of Business Studies

The transport sector causes 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
Developments in the use of bioassays for chemical and environmental risk assessment (II)

SIMONI: Smart integrated monitoring of the water quality

R. van der Oost, Wateren / Onderzoek en Advies; G. Sileno, Wateren / Research and Development; M. Thao Nguyen, Waterproef, L. Moria, Wateren / 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 bioanalytical 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 bioanalytical 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 applied to several ecologically generally occurring at agricultural sites. In addition, increased ecological risks were also observed at waters receiving wtp 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 chemical pollutants), at most sites with increased ecological risks due to micropollutants, similar classifications were obtained with bioanalytical and extensive chemical analyses. The tier 2 research at agricultural greenhouse areas identified eight pesticides contributing most to the increased environmental risks.

Bioassay battery responses to POCIS and Speedick passive sampler extracts

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A large portion of the toxicological 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 makeup of a water body. The effects observed in surface waters is not only but also 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.
samplers, the often used polar organic chemical integrative sampler (POCIS) and the recently introduced Spedisc, POCIS and Spedisc passive samplers were simultaneously deployed at sites likely to be contaminated with polar compounds, including agricultural greenhouse sites and wastewater treatment plant (WWTP) impacted locations. The extracts of the passive samplers were subjected to a battery of bioassays, specifically responsive to polar compounds, including ERu, anti-AR and GR chemical activated luciferase gene expression (CALUX) bioassays, as well as the RIKILT WaterSCAN for antibiotics activity. In addition, the MicroTox test for non-specific toxicity to bacteria was run. The two investigated passive sampling devices generated a different toxicity profile in the applied bioassay battery, with several unique responses per passive sampler. Nonetheless, POCIS caused bioassay responses more frequently and more intensely, leading to more frequent trigger values exceedances and thereby to the detection of ectotoxicological risks. Hence, POCIS outperformed Spedisc in most bioassays at the majority of the investigated locations. These results thus suggest that POCIS is best fit for application as passive sampling device targeting polar compounds in effect-based water quality monitoring strategies.

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 ultraviolet 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-2 (BP2), octocrylene (OC) and octyl salicylate (OS) are four 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 BP3 and OC and the molecular mechanism underlying the toxic effects. Docking analyses, 24 h and 96 h chemical exposures will be conducted on the ZFL cells to determine the potential binding affinities to the estrogen receptor (ER) and the half-lethal concentration (LC50) for the UV filter chemicals. The gene expression profiles on the ER pathway and the aryl-hydrocarbon receptor (AhR) pathway will also be measured by quantitative real-time PCR in ZFL and larvae exposed to the sunscreen chemicals. A dual-luciferase reporter gene system with AhR and ER receptors transfected in ZFL cells will be used to confirm the biological activities of these sunscreen chemicals in ZFL cells.

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, Š. Strápacová, Veterinary Research Institute, Brno / Chemistry and Toxicology; L. Svrčková, Veterinary Research Institute, Brno / Chemistry and Toxicology; J. Necka, M. Ciganek, Veterinary Research Institute, Brno / Chemistry and Toxicology; I. Bartonková, Z. Dvorák, Faculty of Science, Palacky University, Olomouc / Toxicology; J. Tomášek, Experimental Medicine, CAS, Prague; J. Vondráček, 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 number of poly cyclic aromatic compounds, including PAHs, methyl-, nitro- and oxy-PAHs, thiophenes, dinitropyurranes, benzacendines 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 assay, in order to compare the potencies in rodent and human models. Using a set of various CALUX assays, responses were selected for individual classes of organic contaminants. The general outline of these 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, chemical 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) effluents is a topic of concern to wastewater treatment companies 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-qMS 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 and anti-estrogenic activities were observed and progesterogen activities were present in similar concentrations in untreated as in treated WW. The glucocorticoid activity in influent was fully explained by prednicarbacte, trimcinolone acetonide, dexamethasone and amicronide. 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 shown to be due to compounds present in the raw wastewater and not androsterone 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 androsterone. The QToF MS data are currently interpreted to elucidate the identity of the unknown glucocorticoids. Also bioassaychromatograms 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. Pauw, only found in Bordeaux WWTP / LPTC UMR 5805 CNRS; M. Dèviter, University of Bordeaux / EPOC / LPTC UM 5805 CNRS; E. Maillot-Maréchal, INERIS / UMR SEBIO ECO; C. Geneuste, University of Bordeaux / EPOC / LPTC UM 5805 CNRS; S. Ait-Aissa, INERIS / UMR SEBIO ECOT; H. Budzinski, University of Bordeaux Waste waters represent a major pathway of introduction of EDCs into the aquatic environment. Considering the high concentration of pharmaceuticals and their possible transformation products, many EDCs are currently unknown. Therefore, 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 spectroscopy 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 glucocorticoid receptors. The non-target screening allowed detecting 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, it 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 tramacrolol) were recurrent found. This approach combined to LC-QTOF has allowed establishing a list of systematic detected non-target compounds in several wastewaters.

Indigeniety 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. Shovelton, 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 activity, with the Conservation and Status of Endangered Wildlife in Canada identifying 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 government representatives, 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 sturgeon, 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 uses for: enabling 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 had 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,

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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 insecurity important on both biosecurity and food security. For Indigenous communities 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 chemically-similar 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 Kuujjuaq-Whapmagoostui (K-W) and Kangiqsualujjuaq 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 present perspectives from the Inuit perspective and community engagement: Can we detect REEs in freshwater, marine and terrestrial plants and animals? Do REEs bioaccumulate and bioamplify in northern food webs? How to best establish a sustainable community-based environmental monitoring project? 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 questions from the scientific perspective and on community engagement: Can we detect REEs in freshwater, marine and terrestrial plants and animals? Do REEs bioaccumulate and bioamplify in northern food webs? How to best establish a sustainable community-based environmental monitoring project? 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 questions from the scientific perspective and on community engagement:

637 Te Ohu Mō Papatūānuku: A Collective Response to Healing

T. Godfrey, H. Hirere, Te Whare Whanga O Awanuiarangi / School of Undergraduate Studies

The use of pentachlorophenol (PCP) as 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 PCP and other contaminants upon both human and environmental health has led to the formation of an inter-disciplinary group known as Te Ohu Mō Papatūānuku, which is comprised 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 conservation however, is the role of indigenous knowledge – 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 synchronistic approach –
whereby science is an integral remedial component and provides a vehicle for remediation to occur, but cultural and soul connections of the indigenous people are the drivers of reciprocal remediation, and subsequent healing. Scientific knowledge applies science and indigenous knowing revitalises relationships, informing and infusing behaviour with an ethos of respect, empathy, and reciprocity.

638 Discussing the Unfamiliar but Contentious: Hydraulic Fracturing Consultation with Remote, Indigenous Communities in the Northern Territory, Australia
R. Smith, Hydrobiology Pty Ltd; R. Pepper, Land and Environment Court of New South Wales; D. Ritchie, Ninti ONE Foundation
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 in the NT, involving rounds of visits for community forums and hearings. These could not be conducted in the same manner as larger community consultation, and featured language and background knowledge barriers that are not typical of even remote non-indigenous community consultation in Australia. A number of these issues, and how they were addressed are discussed. Although the timeframe allotted to the Inquiry was particularly challenging for indigenous community consultation, and hence the extent of engagement in the process varied greatly between communities, nonetheless the Inquiry did achieve substantial engagement with most. The community feedback gained thereby was a vital input into the Inquiry’s assessment of the potential cultural impacts of shale gas development in the NT.

639 Incorporating cultural values and perspectives of First Peoples’ (Aboriginal People) into water planning, science and environmental water management
B.J. Moggridge, Institute for Applied Ecology, University of Canberra / Institute of Applied Ecology
Australia is the world’s driest 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 the 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, re-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 across both, acute and chronic exposures. The BLM approach has been demonstrated to both total and dissolved metal concentrations. Metal-specific BLMs were proposed for different biological species and both, acute and chronic exposures. The BLM approach for the site assessment of metals in aquatic systems. There is an increasing awareness by leading regulatory bodies in the USA, Europe and other countries that the bioavailable fractions of metals could be better descriptors of their risks. The principal concept of metal bioavailability is the Biocidal Ligand Model (BLM) which allows site-specific assessments of metals’ risks by considering the environmental factors which determine the bioavailability of dissolved metals in the aquatic environment. For many metals bioavailability in freshwaters is modulated by dissolved organic carbon (DOC) concentrations, water hardness, the pH of the water and other factors such as temperature, concentrations of further ions and suspended solids as well as metal speciation. Metal-specific BLMs were proposed for different biological species and both, acute and chronic exposures. The BLM approach for the site-specific evaluation of surface water monitoring data was hampered by the huge data requirements of the original BLMs (several site-specific water parameters). But the recent development of user-friendly BLM-based bioavailability tools (e.g., www.PNEC-pro.com) allows the consideration of bioavailability for the evaluation of freshwater monitoring data of relevant metals. Such tools, which only need a basic set of easily available water parameters as input (mostly pH, Ca concentration, DOC, and dissolved metal concentration), are currently available for metals such as lead, nickel, copper and zinc. The new EU Water Framework Directive environmental quality standards for lead and nickel according to Directive 2000/60/EC will consider the bioavailable fractions of these metals. In this contribution, the advantages and possible drawbacks of BLM-based bioavailability tools are presented highlighting feasibility, ranges of validity, and comparability between tools. Finally, recommendations for the regulatory implementation are given. This contribution is based on the outcome of the IUFAC-supported project “Consideration of bioavailability of metals/metal compounds in the aquatic environment” (J2011-060-1600).

640 Tap water intake of poly- and perfluoroalkyl substances (PFASs) in relation to serum concentrations in a nationwide prospective cohort of U.S. women
X. Hu, F. Laden, Q. Sun, P. Grandjean, Harvard University; L.W. Yeung, University of Oœebro / Department of Chemistry; E.M. Sunderland, Harvard University / School of Engineering and Applied Sciences
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 are 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 matched 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 highly influenced by up to 3 factors of 2-3. We will next 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 fluorides (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.

641 Consideration of the bioavailability of metals and metal compounds in freshwaters in regulatory frameworks
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Recent research has demonstrated that both total and dissolved metal concentrations are not the most appropriate parameters for the risk assessment of metals in aquatic systems. There is an increasing awareness by leading regulatory bodies in the USA, Europe and other countries that the bioavailable fractions of metals could be better descriptors of their risks. The principal concept of metal bioavailability is the Biotic Ligand Model (BLM) which allows site-specific assessments of metals’ risks by considering the environmental factors which determine the bioavailability of dissolved metals in the aquatic environment. For many metals bioavailability in freshwaters is modulated by dissolved organic carbon (DOC) concentrations, water hardness, the pH of the water and other factors such as temperature, concentrations of further ions and suspended solids as well as metal speciation. Metal-specific BLMs were proposed for different biological species and both, acute and chronic exposures. The BLM approach has been demonstrated extensively in the scientific literature, and BLMs have been applied for the risk assessment of metals and metal compounds (e.g., for copper and zinc in the EU). In the past, the broader use of the BLM approach for the site-specific evaluation of surface water monitoring data was hampered by the huge data requirements of the original BLMs (several site-specific water parameters). But the recent development of user-friendly BLM-based bioavailability tools (e.g., www.Bio-net.net, www.PNEC-pro.com) now allows the consideration of bioavailability for the evaluation of freshwater monitoring data of relevant metals. Such tools, which only need a basic set of easily available water parameters as input (mostly pH, Ca concentration, DOC, and dissolved metal concentration), are currently available for metals such as lead, nickel, copper and zinc. The new EU Water Framework Directive environmental quality standards for lead and nickel according to Directive 2000/60/EC will consider the bioavailable fractions of these metals. In this contribution, the advantages and possible drawbacks of BLM-based bioavailability tools are presented highlighting feasibility, ranges of validity, and comparability between tools. Finally, recommendations for the regulatory implementation are given. This contribution is based on the outcome of the IUFAC-supported project “Consideration of bioavailability of metals/metal compounds in the aquatic environment” (J2011-060-1600).
Age-Based and Time Trends of Exposure Chemical Biomarkers in the US Population 1999-2014

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In attempt to define and measure the effects of pollutants in the aquatic ecosystem, biomarkers have attracted a great deal of interest. The principal of the biomonitoring approach is to analyze the organism's responses to pollutant exposure. Therefore, the aim of the present study was to verify the suitability of biochemical responses of estuarine fish (Microprogonias furnieri), copepod (Acartia tonsa) and crabs (Callinectes sapidus) as biomarkers to evaluate the environmental quality assessment in tropical estuarine environments in northeastern Brazil. Thus, we evaluated biomarker of metal exposure as metallothionein-like proteins (MTLP), biomarker of organic exposure as 7-ethoxyresorufin-O-deethylase (EROD), and biomarkers of general effects like lipid peroxidation (LPO) and acetylicholinesterase (ACHE). MTLP and LPO concentration were analyzed in copepod whole-body, crabs and fish hepatopancreas, while EROD activity were analyzed in fish hepatopancreas and AChE inhibition in in copepod whole-body, crabs and fish hepatopancreas and fish muscle and brain. Samples were collected in three different sites along two estuaries (São Marcos Bay and São José Bay), in two seasons (dry season/August and rainy season/January) in three different years (August/2012; January/2013; August/2013 and January/2014). In both estuaries, a high degree of heterogeneity were observed in biomarkers response over the two years of study, with divergence spatial and temporal changes. Moreover, analyzing all biomarkers studied, regardless of estuaries and season, it is possible to observe at least two biomarkers alteration in both estuaries and season, reaching up to seven different biological responses in rainy seasons. The responses confirmed the initial expectation that both São José Bay and São Marcos Bay are subjected to the impact of the adjacent river basin drainage. In this context, biomarkers response were able to discriminate the season and seasonality of the contamination which are affecting the estuaries along different seasons and year as well as the effects on local species. Regard test organisms, estuarine fish (Microprogonias furnieri) showed to be more sensitive to environment alterations, revealing most of the results obtained. Therefore, these results show that biomarkers is a promising tool for the assessment and monitoring of macro-tidal estuaries from tropical aquatic ecosystems impacted by anthropogenic activity.

Ecotoxicology of micro and nanoplastics: Mechanistic

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SETAC Europe 28th Annual Meeting Abstract Book
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 two freshwater organisms, a water flea (C. dubia) 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 the environmentally relevant level. 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. plastics additives and side products, attracting less interest. 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), polyethylene (PE), polypropylene (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 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 unspecific toxicity, oxidative stress and antiandrogenic 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 LDPE, 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. So far, the potential risk of plastics is not considered in hazard and risk assessment and management of plastic products. 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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Polymer degradation in marine ecosystems is 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 three recently labeled 0.05, 0.5, and 6 µm polystyrene microbeads. Exposure to polystyrene microbeads led to significant size-dependent negative effects on growth rate, reproduction, and lifespan. In consistent, 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 (degradation or weathering) of microplastics (e.g. microbeads) of marine plastics in cosmetics, industries and domestic sources. Virgin plastic polymers are, in principle, biologically inert and thus non-toxic. However, plastic production includes additives such as plasticizers, colorants or fire retardants that can be toxic. In addition to the potential toxicity caused by additives, MPs offer surfaces where hydrophobic pollutants can be adsorbed, including persistent organic pollutants (POPs). In marine environments, such chemicals are found at high concentrations in the surface layer, where low-density microplastics are most abundant. Indeed, these small particles can act as vectors and carriers for a wide range of pollutants and be ingested directly by organisms, causing chronic physical and/or toxicological effects. When cumulation 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 are very complex and poorly understood. Therefore, objectives of the present study were to investigate the sorption kinetics of two model 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 FPOS, 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 (virgin or spiked). No differences were found regarding sub-lethal endpoints (genotoxicity, photomotor response (PMR) and EROD activity) between the control group and fish exposed to virgin MPs, spiked MPs or compared controls. 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 microparticles. Experiments are currently being done to test the toxicity of MPs.

650 Comparative role of microalgae and microplastics in the effects of chlorpyrifos on 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 ecosystems is the 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 three recently labeled 0.05, 0.5, and 6 µm polystyrene microbeads. Exposure to polystyrene microbeads led to significant size-dependent negative effects on growth rate, reproduction, and lifespan. In consistent, 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.
habitations is that they might act as vectors of pollutants to marine organisms, since hydrophobic organic contaminants with low water solubility tend to concentrate on the surface of these particles. In this study we have compared the role of polyethylene MPs and microalgae (MA) of similar size, as vectors of the organophosphorus insecticide chlorpyrifos (CPF) to *Mytilus galloprovincialis* marine mussels. With that aim, CPF pre-exposed MP and MA were offered to the mussels in batch exposures and biometric parameters and biotransformation were measured. AChE activity in digestive gland and gills was significantly inhibited at all CPF treatments, disregarding exposure time. Levels of GST activity in the digestive gland in the three CPF treatments (CPF, MA+CPF and MP+CPF) after 7 days exposure were significantly higher than levels in treatments without CPF. However, after 21 day exposure, GST activity in the controls significantly increased, and differences with controls disappeared. For GST in gills, a significant increase in activity was observed in the MP, CPF and MA-CPF treatments after 7 days, compared to the MA control. When the nine biomarkers recorded are combined using the Integrated Biomarker Response (IBR) index a similar response in the three CPF treatments is initially observed (7 d), but after 21 d an enhanced response in observed in the MA+CPF and MP+CPF treatments only. In conclusion, AChE inhibition was similar in all CPF treatments disregarding the presence of particles. However, both MP and MA particles in CPF-exposed mussels produced in the long term an increase in biomarker response compared to waterborne exposure. Therefore MP seem to play a similar role as natural organic particles as vectors of organics to marine organisms.

651 Poster spotlight: TH001, TH002, TH003

Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (II)

652 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 *Pteridium aquilinum* which are classified in Group 2B 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 in surface and ground water has not been studied. Under sterile conditions, dissipation (= hydrolysis) of PTA in aqueous solution follows classical first order kinetics: $k_{obs} = k_{0} [H^{+}] + k_{acid} + k_{neutral} [H^{+}].$ The rate constants are: $k_{obs} = 25.7±1.0 M^{-1} h^{-1}$; $k_{acid} = 9.5±6.0·10^{-1} M^{-1} h^{-1}$ and $k_{neutral} = 4.8±0.0·10^{-2} M^{-1} h^{-1}$. 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 dissipation of PTA under natural environmental conditions using 10 different surface and groundwater samples from Denmark and to compare the degradation kinetics with the existing model for hydrolysis. Degradation 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 a special concern for public health. Current detection methods need expensive equipment, trained personnel and complicates protocols. In particular, the determination of marine biotoxins, released as a consequence of toxic algae blooms, is performed offline on fish / shellfish homogenates rather than on-line, resulting not appropriate for monitoring programs that require real-time warnings like in specific situations relevant for public health, such as bathing sites and aquaculture plants. Thus, there is a special need for on-line, continuous, rapid and sensitive field tests. To fill this gap, in this work, it was developed and tested a portable flow-injection - Linked Immuno-Magnetic Concentration micro assay for the detection of Domoic Acid, Saxitoxin and Okadaic Acid in seawater. The assay is based on the fact, that in the presence of the target toxin, competition occurs and consequently the color production decreases proportionally to the toxin concentration. This analytical approach allows to combine antibody selectivity, convenience of a separation step through the use of magnetic beads and simplicity of the system. Neat, the main assay was integrated within a fully automated PC-controlled on-line analyzer based on the micro Loop Flow Reactor technology able to host three immunoassay sub-modules. Specific volumes of reagents were injected to a flow cell, equipped with a heater, a magnet and an optical group. Because most of the reagents needed to be kept at 4 °C, a Pelletier refrigerated compartment was designed and incorporated in the instrument. Laboratory measurements were executed to validate the prototype efficiency to detect sub-ppb concentrations of the algae toxins. The obtained calibration lines were consistent with the strict requirements limiting the presence of the toxins in environmental waters. On-line suitability was demonstrated by a field installation on a floating platform in the port of La Spezia, Italy, for daily monitoring of real marine water samples, in which the instrument was integrated together with dedicated module for real-time data analysis. At the center. In conclusion, results obtained showed that the automated measurements are repeatable and sensitive. Further work must go into developing additional specific antibodies to extend the application on other natural pollutants released by plants, algae and microorganisms, with a particular eye on freshwater cyanotoxins.

654 A decade of chemical studies on Ostreopsis. What’s left?

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Over the last decade massive blooms of the benthic dinoflagellate Ostreopsis cf. ovata, once confined to tropical and subtropical areas, have occurred in more temperate regions worldwide including the Mediterranean Sea. Concurrently, negative impacts on human health mainly due to inhalation of toxic aerosols and/or skin irritation have been observed together with death of marine invertebrates. When first toxic outbreaks related to *O. cf. ovata* occurred in 2005 along the Ligurian coasts (Italy), little was known on several aspects of the phenomenon. Although some *Ostreopsis* spp. were known to produce congeners of palytoxin (PLTX), *O. cf. ovata* was not known as a toxic species and its metabolic profile had never been investigated. Secondly, although PLTX itself was reported as one of the most potent marine toxins known, the role of PLTX itself as the causative agent of fatal food poisonings in the tropics, it had never been suspected to exert toxicity through inhalation. Last but not least, the role of the environmental conditions on *O. cf. ovata* proliferation and toxin production had been poorly studied. Therefore, the need for increase knowledge on potential risks for humans and ecosystem stimulated research in the field. An Italian interdisciplinary network was created, including scientists from the Academy and operators of the regional environmental protection agencies and food safety laboratories. This joint effort led to clarify many of the aspects related to the *Ostreopsis* phenomenon that still represents one of the major threats to humans in the Mediterranean area. This presentation is meant to summarize the results of our studies on *O. cf. ovata*, highlighting inter- and intra-specific variability, variables, structural variability of the PLTX congeners detected to date, and, in some cases, linking such differences to the risk that PLTX congeners pose to humans following inhalatory, dermal and oral exposure. Phylogenetic relationships among many isolates of Mediterranean *O. cf. ovata* were investigated as well as innovative molecular qPCR based assay was developed for monitoring activities. A general overview of the environmental conditions that favour proliferation and toxin production will be also provided based on laboratory and field observations. The methodological approach, besides addressing many of the palytoxin-related issues, may serve as template for facing in due time any emerging toxin-related threat to humans.

655 Untangling the geosmin appearance in a Mediterranean river: relationship of geosmin concentration and physicochemical parameters over a year

C. Espinosa, L. Llenas, Universitat de Vic / Universitat Central de Catalunya / BETA Technological Centre; M. Ordeix, Universitat de Vic / Universitat Central
Identification and prioritization of emerging risks for food safety: climate change as a driver A. Magriore, European Food Safety Authority (EFSA) / Risk Assessment and Scientific Assistance Department; A. Alonso, EFSA; European Food Safety Authority / Risk Assessment and Scientific Assistance Department; A. Rotaia, European Food Safety Authority; G. De Sanctis, D. Verloo, C. Gardi, S. Dohlander, Y. Van der Stede, F. Boelaert, M. Binaglia, J. Tarazona, EFSA European Food Safety Authority

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

658 Hazard identification of endocrine disrupting properties of pesticides on non-target organisms: state of the art and future perspectives E. Alh, EFSA - European Food Safety Authority / Pesticides Unit; M. Arena, EFSA - European Food Safety Authority / Pesticides; D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit

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 allow for 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 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 (more specifically in amphibians). 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.

659 Contaminants of emerging concern in the North American Great Lakes: Evidence of reproductive disruption from field and laboratory studies H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory; L. Thomas, St. Cloud State University / Biological Sciences; L. Wang, St. Cloud State University / Aquatic Toxicology Laboratory; C. Cipollitti, St. Cloud State University / Aquatic Toxicology Laboratory; J. Zorgenson, St. Cloud State University / Environmental Contaminants; S. Elliott, U.S. Geological Survey / Minnesota Water Science Center; M.E. Brigham, U.S. Geological Survey

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 the CECs can be attributed to dichothomous urban or agricultural upstream land use. Mixtures found in watersheds with urban influences commonly contained steroidal estrogens, BPA, alkylphenols, pharmaceuticals and personal care products. Agriculturally influenced sampling sites contained herbicides and pesticides in addition to BPA and alkylphenols, 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 maximal 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. Evensten, Norwegian School of Veterinary Science; L. Evensten, The Arctic University of Norway; F. Falciani, University of Liverpool; T. Iguchi, YOKohama City University / Molecular Environmental Endocrinology; C. LaLone, U.S. EPA / Mid Continent Ecology Division; Y. Li, NIVA Norwegian Institute for Water Research; M. Cronin, Liverpool John Moores University. School of Pharmacy and Biomolecular Sciences; E. Perkins, US Army Corps of Engineers ERDC; T. Runderberget, Norwegian Institute for Water Research; B. Salbu, Norwegian University of Life Sciences; I. Sylte, The Arctic University of Norway / Department of Medical Biology; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; N. Vinas, Mississippi State University / Biomarker Research and Decontamination Science; NEU NIOE Norwegian Institute for Water Research / Ecotoxicology and Risk Management

A number of exogenous compounds have the potential to interfere with the endocrine system of animals and may perturb vital endocrine processes to a degree causing an adverse effect (outcome) on ecologically relevant endpoints such as 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 focused 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 (EcR) and the Juvenile Hormone (methyl farnesoate) receptor (MfR) have been identified to be toxicologically relevant. The present paper focus 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 defined toxicity endpoints suitable for Integrated Approaches for Testing and Assessment, IATA, 5) identify potential EDCs and 6) practical implementation of AOP in routine life cycle risk assessment. The present project will be informed by ongoing 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. Kluge, 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 impairment of specific chemical pathways do not adequately detect all EDC compounds 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 mechanistic actions. 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. Holhech, University of Southern Denmark / Biology; L. Weltje, 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. Braumbeck, 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 239, 230 and 234. A reduction of VTG production (mainly in males) by 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 function and structure, 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 / ovary fish. Thus, we investigated the effects of two well-known hepatotoxicants, 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 (vtg1, vtg3 and esr1) and hepatotoxicity-related marker genes (fabpl10a, apoal, cyp2k19 and cyta) 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 hepatectomy degeneration. VTG levels in APAP-exposed females were elevated, while no effect was observed in INZ-exposed fish. Likewise, gene expression responses 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 path (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; H. Segner, University of Bern / Centre for Fish and Wildlife Health; M. Cronin, L. Haneklaus, Posthaus, University of Bern / Institute of Animal Pathology; H. Schacht, University of Heidelberg / Aquatic Ecology and Toxicology; E. Hillebrecht, M. Teiger, Fraunhofer IME / Ecotoxicology

The discussion about the regulation of endocrine disruptors (ED) is on-going between groups of scientists, authorities and stake-holders. Especially dose-dependency, low dose effects and effect thresholds are still under debate. To address parts of these questions, a static life-cycle test with zebrafish (Danio rerio) has been performed to examine if a pulse exposure of an ED might lead to distinguishable and the establishment of a dose-response relationship is possible. The results will be used for a comparison with available data originating from a flow-through study with TC in zebrafish. A water-sediment system has been set up to expose different life-stages (group A: 40 eggs, group B: 20 juveniles, group C: 16 adults) to a known ED, Tamoxifen citrate (TC). Observed endpoints included early-life-stage growth parameters, reproduction and mortality, as well as adult growth, sex ratio, vitellogenin levels and F0-generation early-life stage. Four concentrations of TC were applied as a pulse in three replicates each, ranging from 125 µg/L to 1000 µg/L (spacing factor 2). Four controls replicates were included. Mortalities occurred in all developmental stages (groups A to C), especially in high concentrations (500 µg/L, 1000 µg/L). In sexually mature fish (group C) mortality was higher in males. A decline in fertility could be observed for group C, possibly related to the higher male mortality. Total egg numbers appeared unaffected. The results were mirrored for fish introduced as juveniles (group B): While fertility rates declined. To further examine the population level mortalities occurred in all developmental stage. Four concentrations of TC were applied as a pulse in three replicates each, ranging from 125 µg/L to 1000 µg/L (spacing factor 2). Four controls replicates were included. Mortalities occurred in all developmental stages (groups A to C), especially in high concentrations (500 µg/L, 1000 µg/L). In sexually mature fish (group C) mortality was higher in males. A decline in fertility could be observed for group C, possibly related to the higher male mortality. Total egg numbers appeared unaffected. The results were mirrored for fish introduced as juveniles (group B): While fertility rates declined.
664 A Tiered Approach for Screening Chemicals for Biomagnification Potential in Humans
A. Sangion, University of Insitria / Department of Theoretical and Applied Sciences (DiSTA); J.A. Arnot, ARC Arnott Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology. P. Gramatica, E. Papa, University of Insubria / Department of Environmental Science/Department of Pharmacology and Toxicology.

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 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) mode 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 Arnott 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 Arnott Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); A. Lookey, ARC Arnott Research and Consulting Inc.; J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; M. Embry, ILSI, ILSI Europe; B. Tebben, University of Toronto, Applications of Modelling & Quantitative Methods (AMOD); C. Tebby, Trent University, Applications of Modelling & Quantitative Methods (AMOD); T. Luckenbach, Helmholtz Centre for Environmental Research / Dept.Bioanalytical Ecotoxicology / Department Bioanalytical Ecotoxicology; E. Kürster, Helmholtz Centre for Environmental Research, Dept.Bioanalytical Ecotoxicology / Department Bioanalytical Ecotoxicology; T. Lackenbach, Helmholtz Centre for Environmental Research UFZ / Department Bioanalytical Ecotoxicology.

Critical evaluation of a human in vitro biotransformation rate database is required for tiered approaches in risk assessment and for integrating in vitro and in vivo methods. Despite the fundamental value of biotransformation rate information, relatively few estimates (half lives) are available for humans compared to the thousands of chemicals for which Kow and Koa data are available. We developed and applied novel IVIVE methods to characterize the human exposure setup (tier 4) shown to be necessary for human exposure assessment with reasonable confidence. Here we present predictions of bioaccumulation and trophic transfer.

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 invertebrates 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 behaviour may add compound uncertainty to predictions of bioaccumulation and trophic transfer.
assessment 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 main factors influencing the toxicokinetics (water temperature, growth dilution, reproduction cycle, …). Default values for compound-specific parameters were estimated by QSAR models based on hydrophobicity [2, 3]. An optional interaction terms was added to the mixture PBTK models for metabolic interactions such as competitive inhibition. Two case studies were selected based on 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 chlopyrifos and permethrin was modelled. The models developed enable 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.

**660**

**Application of Aqueous and Dietary In-Vivo Bioaccumulation Tests to Determine Biotransformation Rates, Elimination Rates and other Bioaccumulation Metrics**

F. Gobas, Simon Fraser University / Resource & Environmental Management; M. Dimauro, K. Compton, Simon Fraser University; Y. Lee, Simon Fraser University / Resource and Environmental Management; V. Otton, Simon Fraser University / Resource and Enav Management; J.C. Lo, Simon Fraser University / Biological Sciences; G. Allard, Simon Fraser University / Faculty of Environment

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. Kroll, 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  

In order to make informed decisions, it is crucial to assess the potential impact of oil spills on freshwater ecosystems. The objective of this study was to develop and validate an in-situ amphibian metamorphosis assay as a tool for evaluating oil spill-related toxicity. The assay was tested in the field in a freshwater environment in the presence of diesel fuel and bitumen. The results showed that the assay was sensitive to the presence of oil and bitumen, and could be used to detect changes in amphibian metamorphosis. The assay has potential to be used as a rapid and cost-effective tool for assessing the impact of oil spills on freshwater ecosystems.

MO002  
APPLICABILITY OF RISK BASED, TIERED ASSESSMENT OF PRODUCED WATER DISCHARGE IN NIGERIAN SHALLOW OFFSHORE ENVIRONMENT  
M.G. Smit, Shell International; O. Anako, SPDC Nigeria Ltd  

The applicability of risk-based, tiered assessment tools for the classification of produced water discharges in the Nigerian offshore environment was assessed. The study identified a number of critical issues that need to be addressed in order to make the tiered assessment tools applicable to the Nigerian offshore environment. These issues include the need for more comprehensive data on the properties of produced water discharges, the need for more detailed information on the effects of produced water discharges on marine organisms, and the need for more robust modeling tools for predicting the effects of produced water discharges on marine ecosystems.

MO003  
Assessment of the biological impact of using chemical dispersants to remediate oil spills in different environmental conditions using zebrafish embryos  

The use of chemical dispersants to remediate oil spills in different environmental conditions can have significant biological impacts. Zebrafish embryos were used as a model system to assess the biological impact of dispersants. The results showed that dispersants can have different effects on zebrafish embryos depending on the environmental conditions. The study highlights the need for further research to better understand the biological impacts of dispersants in different environmental conditions.

MO004  
Behaviour and effects of a marine diesel oil in a semi-static exposure experiment using mussels (Mytilus spp.) from the Baltic Sea  

The study investigated the behaviour and effects of a marine diesel oil on mussels (Mytilus spp.) from the Baltic Sea. The results showed that the diesel oil significantly affected the growth and survival of the mussels. The study highlights the need for further research to better understand the effects of marine diesel oil on marine ecosystems.

MO005  
Biliary PAHs and enzymatic biomarkers in the teleost Eucogres brasiliensis along four tropical estuaries in the Brazilian Northeast  
J.S. Silva, R.N. Alves, UFPE Universidade Federal de Pernambuco / Zoology;
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, is commonly used 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 (Benzo(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 waterborne toxics induced higher catalase and glutathione S-transferase activities and induction of acetylcholinesterase activities. Exposure to waterborne toxics 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 (CTM2012-31359), University of the Basque Country – UPV/EHU (UFI 11/13) and Basque Government through Consolidated Research Groups fellowship (IT810-B). MOUSE6 Bio-marker and gene transcription variability in perch in reference sites used for biomonitoring studies L. Förin, N. Asker, University of Gothenburg / Department of Biological and Environmental Sciences; M. Töpel, University of Gothenburg / Department of Marine Sciences; T. Osterlund, 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 studies of reference sites offer the possibility to follow 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 plausible 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 (EROD), plasma levels of vitelloprotein, markers for oxidative stress, white blood cells count and gonad sizes. The RNA sequencing data showed that there was no significant enrichment 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 periods 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 response, 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. D. Fernández ESTAC / PiE University of the Basque Country UPV/EHU / Departamento de Zoología y Biología Animal Celular; U. Izaguirre, 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. Mollusks are common indicator organisms of marine pollution, being widely used for monitoring purposes. The current study focused on two different sites in the Northern Atlantic and compared key physiological and histological data of Mytilus edulis individuals. At the first site (2009, Lekeitio, Spain) the main pollutants were metals and organics, while at the second site (2012, Pent hemat, Spain) the main pollutants were metals. Histological analysis of malformation in mussels at both sites revealed liver alterations such as hyperaemia, melanomacrophage centres and necrosis. The presence of stress-related genes, such as metallothioneins and superoxide dismutase, was observed in the mussels from the contaminated site, whereas the mussels from the clean site did not show the same expression pattern. These results suggest that mussels can be used as indicators of pollution in the Northern Atlantic and that they can be used to detect early signs of pollution even at very low concentrations.
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 & port, WWTP dumping area) and reference sites in Tromsø (69° 40’ N) and Trondheim (63° 26’ N) were sampled in the autumn of 2016 and late summer 2017. Different tissue-level biomarkers including cell type composition (VvBAS) in digestive gland epithelium, structural changes of digestive alveoli (MLR/MET), relative proportion of digestive and connective tissue (CTD) and histopathological alterations in the digestive gland were measured. In addition, lipofuscin and neutral lipid accumulation in the digestive gland was measured. Histological response to environmental stressors was investigated in the endo-lyosomal system (LSC) of digestive cells were also determined. Higher VvBAS values were recorded in polluted sites than in mussels from reference sites in both study areas. Moreover, mussels from impacted sites exhibited enhanced atrophy of the digestive alveoli (high MLR/MET values) and retraction of digestive diverticula in apparently higher relative extent of interstitial connective tissue (high CTD values). Lower lipofuscin and neutral lipid accumulation in digestive glandular tissue (produced at 10 and 15°C) and at 50% and 100% WAF (produced at 20°C). These WAF produced with dispersant at the three different temperatures was measured. WAF was moderately cytotoxic to mussel hemocytes. In vitro testing approach using hemocytes of the marine mussel Mytilus galloprovincialis (L.) G. Nicolussi, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology, Science and Technology Faculty and Plentzia Marine Station, University of the Basque Country (UPV/EHU); Basque Country, Spain; 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; D. Bilbao, University of the Basque Country (UPV/EHU) / IMDEA Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; N. Etxebarria, University of the Basque Country UPV/EHU / Plentzia Marine Station (PielH4) & Dep Analytical Chemistry; M.P. Cajaravilla, 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. Oil pollution coming from accidental oil spills and from activities related to oil production on the seabed is a significant threat. The impact of 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 Finsal 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 (produced at 10°C) were observed. Exposure to dispersant at 50 and 20°C. These results suggest that relevance of temperature of WAF production on its cytotoxicity is limited. WAF caused a significant induction of ROS production in hemocytes, indicating occurrence of oxidative stress. When tested alone, the dispersant caused a slight but significant decrease in cell viability at the two highest concentrations. However, WAF produced with dispersant at the three different temperatures was not toxic to hemocytes. These results appear to indicate that the dispersant had efficiently reduced the toxicity of the crude oil WAF in the selected cell model. Overall, the in vitro toxicity testing approach in mussel hemocytes could be used as a rapid screening tool for environmental risk assessment of oil spills and oil response strategies in the marine environment. * Funded by EU H2020 GRACE project (679266), Spanish MINECO NACE project (CTM2016-81130-R), Basque Government (consolidated research group IT810-13) and UPV/EHU (UIF 11/37).

MO011 Determination of inorganic cations and anions in wastewater, surface water, and neutralizing amine solutions by IC with a coupled quadrupole MS


Inorganic cation and amine determinations are important to assess salt build-up in systems containing 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, alkanolamines (monoethanolamine, diethanolamine, and methyldiethanolamine) are used routinely to prevent corrosion during transportation to the refinery or to remove sour gases during the refining process. The use of dispersion plants requires accurate structural analyses in the endo-lyosomal system (LSC) of digestive cells were also determined. Higher VvBAS values were recorded in polluted sites than in mussels from reference sites in both study areas. Moreover, mussels from impacted sites exhibited enhanced atrophy of the digestive alveoli (high MLR/MET values) and retraction of digestive diverticula in apparently higher relative extent of interstitial connective tissue (high CTD values). Lower lipofuscin and neutral lipid accumulation in digestive glandular tissue (produced at 10 and 15°C) and at 50% and 100% WAF (produced at 20°C). These WAF produced with dispersant at the three different temperatures was measured. WAF was moderately cytotoxic to mussel hemocytes. In vitro testing 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 Finsal 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 (produced at 10°C) were observed. Exposure to dispersant at 50 and 20°C. These results suggest that relevance of temperature of WAF production on its cytotoxicity is limited. WAF caused a significant induction of ROS production in hemocytes, indicating occurrence of oxidative stress. When tested alone, the dispersant caused a slight but significant decrease in cell viability at the two highest concentrations. However, WAF produced with dispersant at the three different temperatures was not toxic to hemocytes. These results appear to indicate that the dispersant had efficiently reduced the toxicity of the crude oil WAF in the selected cell model. Overall, the in vitro toxicity testing approach in mussel hemocytes could be used as a rapid screening tool for environmental risk assessment of oil spills and oil response strategies in the marine environment. * Funded by EU H2020 GRACE project (679266), Spanish MINECO NACE project (CTM2016-81130-R), Basque Government (consolidated research group IT810-13) and UPV/EHU (UIF 11/37).

MO012 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

G. Zhou, R.W. Lai, R.C. Sham, C. Lam, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; K. Yeung, J.C. Astudillo, The University of Hong Kong / K.K.H. Ho, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; M.M. Jung, Y.K. Yau, The University of Hong Kong / K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

On 3 August 2017, two container vessels collided in the Pearl River Estuary, southwest of Hong Kong, leading to release over 1,000 tonnes of palm stearin into adiabatic cooling solutions, ammonia wastewater, municipal treatment plant wastewater, drinking, and surface water samples by cation-exchange separation followed by suppressed conductivity and mass spectrometry detections in a serial calibration. Cations, alkylamines, and alkanolamines were determined in full scan from m/z 18 to 250 and individual SMs as bare ions and when further sensitivity is needed, as their hydrated adducts. Unlike earlier IC-MS methods for cation determinations, the new simple method employs the MS to detect, quantify, and identify ammonium, alkylamines and alkanolamines, 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, and intertidal gastropods were collected twice (within seven days and four months after the incident) in six locations along the south coast of Hong Kong; it was toxic to microalgae species as reflected by growth inhibition. Its toxic mechanisms on the microalgae may be associated with its adsorption onto microalgal cells, and reduction of light penetration to the cells due to obstruction from the stearin and palm oil. At present, we are generating additional toxicity data on other microalgal species (Thalassiosira weissflogii and Tetraselmis suecica). Using all toxicity data generated from this study, we will ultimately determine an interim WQG for the palm stearin, and use this WQG to assess ecological risk of the palm stearin in the world, and the results will facilitate informed decision-making by the environmental authority.

MO013 Ecological impacts of larvicidal oil on the marine ecosystem: implications on its management

K. Yeung, The University of Hong Kong / K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

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
Effects of oil spill on coastal seaweed in the Arctic
S. Wegeberg, Aarhus University / Department of Bioscience; J. Fritt-Rasmussen, K. Gavstov, Aarhus University / Department of Bioscience - Arctic Environment
In case of an acute oil spill response operation, decision making regarding the operational response strategy and prioritizing biology at risk must be resolute. 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 the effects of 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 oil types were tested at concentrations less than 10 µg/L, impacting fish vision. The present study provides evidence 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 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 with a reduction in retinal layers and neuronal connections that play an important role in visual function and image processing.

MO014 Effects of a coastal oil spill on marine invertebrates and their potential to recover
M.F. Lemos, S. Silva, Instituto Politécnico de Leiria / MARE IPLeiria
There has been an increasing public concern and focus on marine contamination issues mainly due to the arising 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 costal 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 were investigated to detect the different species, the neurotoxicity > 1 from exposure to larvalicidal oil using Monte Carlo simulation, indicating that the current risk was unacceptable high. Hence, monitoring and control on the use of larvalicidal 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. Khursigara, 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, RMSA 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. The studies were conducted to investigate their toxic effects to the marine macroalgae Isochrysa galbana and Chaeotoceros gracilis (primary producers), the intertidal copepod Tigriopus japonicas (a primary consumer), the brine shrimp Artemia franciscana and fish embryos of the marine medaka Orzylas melasticina. Our results showed that although all test marine species were not very sensitive to larvalicidal oil with the ranking of their acute median lethal concentrations (LC50) that were all above the estimated hazardous concentration for 5% of species (HC5), the results of a probabilistic risk assessment showed that the local marine ecosystem had 65.7% of chance to be at risk considering the larvalicidal oil was applied at concentrations higher than 10 µg/L. Hence, monitoring and control on the use of larvalicidal oil as mosquito control pesticide would be urgently needed to mitigate its ecological risks.

MO016 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 / Marine Research Centre; J. Nuutinen, Finnish Environment Institute / Laboratory Centre; K.K. Lehtonen, K.S. Jorgensen, Finnish Environment Institute / Marine Research Centre
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 counteractive 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 biodiversity. In addition to the loss of dispersant effectiveness, the in-water conditions is not well studied. In the present study, impacts of a crude oil and the dispersant Finsol 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 acetylhlecinesterase, glutathione-S-transferase, catalase and glutathione reductase activities, lipid peroxidation, and protein carbonylation. In addition, changes in Mytilus-associated bacterial community, the communities extracted from the gills and digestive glands of the mussels were investigated by sequencing of 16S rRNA 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/L in 5.6 WAF-D compared to 44 mg/L 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/L at 5.6 and 1.82 mg/L 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 for the ratio to 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 estuaries
A.G. Torreiro-Melo, UPPE Universidade Federal de Pernambuco / Department of Zoology; J.S. Silva, UPPE Universidade Federal de Pernambuco / Zoology; E. Zanardi-Lamardio, Universidade Federal de Pernambuco / Department of Oceanography; P.S. Carvalho, UPPE - Universidade Federal de Pernambuco / Zoology
Integrated approaches using biological responses in multiple organization levels are essential for environmental monitoring of tropical estuaries 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

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 / Marine Research Centre; J. Nuutinen, Finnish Environment Institute / Laboratory Centre; K.K. Lehtonen, K.S. Jorgensen, Finnish Environment Institute / Marine Research Centre
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soil characteristics, the group composition was determined for each borehole of all pollutants from soil samples were done using the Soxhlet apparatus with the aim of our research was to determine the presence of petroleum pollutants. Heating plant “New Belgrade” is located on the left coast of the Sava River, about 1.5 km from the mouth of the river. The site is potentially contaminated by petroleum products due to both natural and anthropogenic sources (i.e. oil seeps and rivers flowing over surface bitumen, and pipeline ruptures, grounded ships, storage tank leaks and rainwater). Mass measurement and analysis via HPLC is therefore efficient and as an alternative to be highlighted for solution. In the samples containing analytes from the soil contaminated with gasoline, it is intended to validate the profitability of a similar methodology. 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.

**NEW METHODOLOGY 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

Benzo[a]anthracene, dibenz(a,h)anthracene, chrysene, and fluoranthene, commonly referred to as BTEX, are constituents of fossil fuels that cause serious 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 = 254 nm and T = 50 °C. The analysis was carried out using the Agilent 1200 HPLC system equipped with an automatic injector, a column oven, and a diode array detector. Data were processed using the OpenLAB A.01.05 software. A calibration curve for BTEX standards was constructed in 7 concentration levels: 1 to 68 ppm for benzene, 1 to 80 ppm 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.

**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 risk of the discharge. If the result is out of the regulatory constraints, a PW discharge is permitted. For each PWMP would be specific to the discharged effluent, platform and area, aiming to minimise environmental risk of each PW discharge. The RBA method is compiled of a six-step process. The steps are based on a standard method where a Predicted Environmental Concentration (PEC) and a Predicted No Effect Concentration (PNEC) of the PW or individual products are determined, and a PEC/PNEC ratio is calculated. The PEC/PNEC ratio and Environmental Impact Factor (EIF) which describes a PEC/PNEC ratio in a specified volume of water characterises the potential risk imposed to the receiving environment. With use of a distribution model to determine the fate of the PW, the PEC/PNEC ratio can be mapped specifically to the installation area providing an overall risk profile. The PW is additionally characterised at a substance level, highlighting components which contribute to the overall environmental risk, and will be assessed by the PWMP. Notably in the UK RBA methodology is the presence of PW WECA dealing with sensitivity to fish, and we therefore studied the comparative impact of the RBA approach with the conventional regulatory framework. A study provided unique and important empirical data, and information to evaluate significant considerations for implementation of regulatory PW management methodology. In addition to the potential environmental impact and comparative contribution from production chemicals & naturally occurring substances, and validity of the step-wise tiered screening approach, the investigation provided valuable assessment into adequacy and sensitivity of ecologically relevant species and the implications for regulatory monitoring regime.
MO023
Risk-based assessment of produced water discharges - need for alignment
M.G. Smijg, Shell International

Produced formation water is the main waste stream from upstream oil and gas activities. For offshore installations, next to produced water reinjection (PWI), 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 such 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 testing 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 (US-EPA). 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. Nicolaou, Cefas Lowestoft Laboratory / Environment and Ecosystems

Nine trace elements including As, Cd, Cu, Fe, Hg, Ni, Pb and Zn, and total petroleum hydrocarbons (TPH, adsorbed and dissolved) from two 23 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 of trace metals is significant for Cu and Cd. 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 L.d. Miguel, University of the Basque country (UPV/EHU) / Department of Zoology and Animal Cell Biology, U. Izaguirre, 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 PII; I. Marigomez, Euskal Herriko Unibertsitatea / Zoology & Animal Cell Biology (Sci & Tech Fac)

Maritime traffic and oil platforms in the North and Baltic Sea have been growing and the on going to identify other halogenated compounds especially those containing chlorine atoms are likely to be more environmentally persistent than their non-halogenated analogues because of the presence of the halogen atoms. In addition, the toxicity of some halogenated PAHs have been found to be similar to dibenzo-p-dioxins and dibenzofurans. Because Cl and Br ions are present in the marine environment, we hypothesize that halogenated PACs can be formed and will be bioaccumulate in biota samples. Here we present a method based on high resolution mass spectrometry combined to mass spectrometry using specific multiple reaction monitoring (MRM) ion transitions in the electron ionization mode to detect and quantify halogenated PACs in biological samples. The method was used on a NIST Standard Reference Material (SRM) of mussels (Mytilus edulis) tissue (SRM-2974a) collected from a marine environment. Preliminary results show that 1-chlorophenyle is present in this sample. In addition, we observed multiple peaks in the mass chromatogram coupled to mass spectrometry with chloro-anthracene/phenanthrene but we do not have authentic analytical standards to match retention times. Work is ongoing to identify other halogenated compounds present in biological samples from Canada.

MO028
The experience with the use of biomarkers as Risk Indicators in Environmental Risk Assessment of oil based discharges offshore S. Sanni, International Research Institute of Stavanger / Environment; E. Lyng, D.M. Pampanin, International Research Institute of Stavanger / Environment
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 on 123 oil platforms of the Norwegian Continental Shelf (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, the environmental contamination by drill cuttings was the only source of contamination. 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.

**Tissue-level biomarkers and histopathological alterations in mussels (Mytilus trossulus) from the Baltic Sea exposed to water accommodated fractions of crude oil**

J. Bir, Khulna University / FMRT department; E. Gil-Urriarte, University of the Basque country (UPV/EHU) / Zoology and Cell Biology; A. Alho, Finnmil Environment Institute / Marine Research Centre, R. Tuja, Finnish Environment Invention Institute / Marine Research Centre, K. K. Lehren, Finnish Environment Institute / Marine Research Centre; U. Izaguirre, 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 P.I.E; I. Marigome, Euskal Herriko Unibertsitatea / Zoology & Animal Cell Biology (Sci & Tech Fac); M. Soto, University of the Basque Country / Z gifts and Analytical Chemistry (Sci & Tech Fac); Research Centre for Experimental Marine Biology and Biotechnology PIEUPVEHU

The brackish Baltic Sea is a fragile ecosystem potentially sensitive to oil spills. Chemical dispersants are an effective method to mitigate coastal impacts of oil spills; however, oil treated with dispersants may have unknown toxic effects on biota. The Baltic Sea blue mussel (*Mytilus trossulus*), a particular variety of marine mussels adapted to low salinity. Early winter mussels were collected scuba diving in Tvarminne (Finland) in November 2016, taken to laboratory facilities and acclimatized at the experimental temperature of 5°C 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 (dispersed 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 area ratio (MDR) of 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 MDR did not show response. High CTD values in mussels observed at the salinity of 5.6 indicate a poorer condition of the digestive gland at low salinities than in mussels maintained at 15 psu. Pathological responses (atrophy, necrosis, vacuolization, haemocytic infiltration, granulocytomas) were assessed, being more evident in mussels exposed to WAF and WAF-D (21 d). Salinity is a major factor controlling the biology of mussels in the Baltic Sea. The results obtained here indicate that during the early winter the health of native mussels in the very low salinity central-northern part of the sea is more easily impaired than in those inhabiting the more saline southern regions. The current study is among the first ones applying tissue level biomarkers in *Mytilus trossulus* in the Baltic Sea and provides preliminary reference values for future biomonitoring programmes in the area. **Acknowledgements:** Funded GRACE project (EU H2020 grant N°769296) and a Basque Gov. fellowshipment to EGU

**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 based on an environmental quality standard (EQS) that the PW discharges shall not induce significant adverse effects on the 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 Tvarminne (Finland) in November 2016, taken to laboratory facilities and acclimatized at the experimental temperature of 5°C to two different salinity regimes, the local 5.6 and the artificially increased 15.0 representing the southern Baltic Sea. 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 total PW volatile material in these fractions. The total extracts and fractions were thoroughly characterized using GC–MS, GCxGC–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*. LC0 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, toxicity of the current fraction was mainly attributed to the polar fractions, with LC0 values ranging between 0.17–0.57 mg L⁻¹. Interestingly, toxicity was mainly attributed to the apolar fraction of the fourth PW, with an LC0 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 thereby demonstrates that PW toxicity may be apolar–rich, with cover, throughs 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 compounds that contribute to toxicity. These observed differences may be attributed to the selective loss of some chemical components upon GC separation, the occurrence of polar compounds that are currently poorly characterized. Polar fractions may contain compounds not amenable to GC, or that contribute to the GC–based quantification 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.

**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 species. The central parts of the food chain, as well as its life history strategies and Arctic adaptation, makes it a relevant and valuable test species to provide empirical data on oil component kinetics. *C. hyperboreus* of developmental stage copepodite three (CIII) and five (CV) were exposed to the water soluble fraction (WSF) of crude oil (Troll B) in continuous renewal system (4 or 8 d) followed by a recovery period (20 or 35 d). Water samples were taken 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 total PW volatile material in these fractions. The total extracts and fractions were thoroughly characterized using GC–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*. LC0 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, toxicity of the current fraction was mainly attributed to the polar fractions, with LC0 values ranging between 0.17–0.57 mg L⁻¹. Interestingly, toxicity was mainly attributed to the apolar fraction of the fourth PW, with an LC0 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 thereby demonstrates that PW toxicity may be apolar–rich, with cover, throughs 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 compounds that contribute to toxicity. These observed differences may be attributed to the selective loss of some chemical components upon GC separation, the occurrence of polar compounds that are currently poorly characterized. Polar fractions may contain compounds not amenable to GC, or that contribute to the GC–based quantification 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.
CLIs, the slope was close to unity, indicating a similarity between structural lipids and octanol. The lower slope for CV signifies that storage lipids are less well represented by octanol. Elimination rates were consistently higher in the CLIs than the CVs, resulting in a substantially longer half-time of elimination and high retention of oil components in the CVs. We discuss the role that various biological factors that may contribute to this difference.

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 identification of PACs and their alkylated homologues can be performed from muscle samples and separate whole fish samples, and the results indicated the presence of oxidized forms of CWAs.

MO034 Using the hagfish (Myxinus glutinosa) to study biological effects of a wound 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 (CWAs) dumped after the Second World War. Entire ships loaded with CWAs were subsequently sunken and 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 may have significant deleterious effects on local marine biota. Within the research programme of the EU Baltic Sea Region Interreg project DAIMON (Decision Aid for Marine Munitions, www.daimonproject.com), one of these wrecks was selected to study the leakage of CWAs and their possible biological effects. From the few fish species that inhabit the studied depth range in the region, the hagfish (Myxinus glutinosa), a sediment-dwelling chordate, was selected as target organism for chemical analyses of CWAs in tissues and biological effect studies. Samples were taken using bait traps near the wreck and from a reference area known to contain no wrecks. Liver and muscle samples were used for oxidative stress biomarkers (including lipid peroxidation, protein carbonylation, glutathione-S-transferase, glutathione reductase and catalase activity) and for histopathological biomarkers, and muscle tissue was analyzed for acetyicholinesterase 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 phencyanolic compounds in most of the 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

population assessments (P)

MO035 Seabird-derived contaminants and genotoxicity in Collembola from the Arctic S. Kristiansen, University of Oslo / Department of Biosciences; K. Hylland, Department of Biosciences, University of Oslo, Oslo, Norway / Department of Biosciences; H. Leinaas, University of Oslo / Department of Biosciences; G.W. Gabrielsen, Norwegian Polar Institute; D. Herzke, NILU / Norwegian Institute for Air Research; K. Borga, Department of Biosciences, University of Oslo / Department of Biosciences

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 nutritious and contaminant-enriched guano and is generally characterised by rich and diverse vegetation. In these areas, springtails (Collembola) contribute to a high proportion of the terrestrial biomass. They play a vital role in ecosystems 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) with 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 chlorodanes (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 micronucles 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 micronucleus frequency were associated with both Hg and contaminant concentrations. Seabirds were shown to function as important biovectors of chlorinated PCBs and CHLs. The sensitivity to induced oxidative stress was negatively correlated with higher chlorinated PCBs (6-7 chlorine substituents).

MO036 Higher contaminators and poorer condition in an Antarctic avian top predator from 2001 to 2013
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The Antarctic seabird Caracta acta maccormicki, has been shown that south polar skua (Catharacta maccormicki) has the highest levels of biomagnifying contaminants among Antarctic seabirds. The present study quantifies OHC levels in south polar skua (Catharacta maccormicki) 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 2001/2013, predominant contaminants were Mirex (8484 ng/g bw) and Hexachlorobenzene (HBC) (3561 ng/g bw). 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 lower than lower polybrominated diphenyl ethers (PBDEs). Due to low intraspecific variation in δ13C and δ15N, no significant associations were found between OHCS and isotope. 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. ∑PCB, Mirex and HCB increased with previous data from the same colony, collected during the season of 2001/2002. Ratios of Mirex/∑PCB and Mirex/HCB decreased between the two seasons, suggesting stabilizing Mirex levels and possibly declining levels in the future. Further studies should elucidate the effects of wintering grounds and diet, as well as the level and contaminant occurrence in Antarctic Petrel (Thalassia antarctica), the main prey of...
MO037 Evaluation of malformations induced by a hospital effluent of Tolua (Estado de Mexico) in Lithobates catesbeianus
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Hospital effluents are important from the ecological 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 15 times higher than those detected in synoptic 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 Tolua in the Estado de Mexico 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%, IT=3.8) and in L. catesbeianus (EC50=0.351%, LC50=1.431%, IT=4.0), the main alterations being microcephaly, cardiac and facial edema, malformations in the eye, notochord, tail, fin and intestine. However, the lightest concentration of 0.1% induced significant alterations when 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 Horloff and Usa in Central Germany. The Nidda river system is regarded as a typical river system for central Europe as it passes a very densely populated area and as it is heavily influenced by anthropogenic factors including agriculture, communal waste water and industrial discharges. To get a broad overview of the situation fish face in this river system and the resulting 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 LC10 values of 0.74 mg/l (0.63-0.89) (p<0.001), exposure periphiyton to fipronil under standard medium. However, toxicity was almost inexisten 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 kind of pesticides.

MO041 Use of organophosphorus insecticides in agriculture lands, in a small test birds says please not
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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 exposure that is made are indicators of water quality. Results show that the biodiversity of agroecosystems. The main pesticides used in agriculture are organophosphates (OP), they are highly toxic. In birds, the main route of intoxication to OP is through the consumption of contaminated food, although it can also occur by inhalation when flying over or inhabiting the crop fields and its surroundings. The OP affects the nervous system by inhibiting the function of cholinesterase, which is responsible for transforming the neurotransmitter acetylcholine into choline and acetate. A decreased level of cholinesterase (ChE) activity in the animal tissue is a strongly indicative sign that some type of exposure to an inhibitory agent of this enzyme has occurred. Our objectives were to document ChE levels in house sparrows in response to their exposure to an OP product used in agriculture and determine the extent of their negative effects on birds. In the University Campus (UDED, Durango, Mexico), we collected 15 house sparrows and serum ChE activity was determined by spectrophotometry before and after the consumption of food treated with malathion. The results show a ChE inhibition (11.58 %) after treatment (p=0.03), also we observed nonsignificant
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 sparrows 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 reduced if the non-lethal effects would be expected under realistic worst-case field conditions.

MO043 Biomonitoring and validation of non-invasive samples for the analysis of metals in farming areas
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The mobilization of metals from soils 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 (biomarker 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 preventing 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 terrapins from the most contaminated sites (100% of terrapins 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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Bacillus thuringiensis var. israelensis (Bti) exposure levels used in mosquito control
S. Allgeier, B. Frombold, University Koblenz-Landau; V. Mingo, Trier University / Biogeography; C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences

B. thuringiensis var. israelensis (Bti) is presumed to be an environmental friendly method for use in mosquito control. An evaluation of 86 publications on the effects of Bti has been carried out in terms of species, exposed species, and life stage endpoints, and the available standard test guidelines with regard to these important exposure routes to frequent 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 European common frog (Rana temporaria) larvae show subcellular responses to field-relevant Bacillus thuringiensis var. israelensis (Bti) exposure levels used in mosquito control
S. Allgeier, B. Frombold, University Koblenz-Landau; V. Mingo, Trier University / Biogeography; C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences

Bacillus thuringiensis var. israelensis (Bti) is presumed to be an environmental friendly method for use in mosquito control. An evaluation of 86 publications on the effects of Bti has been carried out in terms of species, exposed species, and life stage endpoints, and the available standard test guidelines with regard to these important exposure routes to frequent 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.

MO047 Influence of salinity and temperature on tadpoles of Xenopus laevis C. Monteiro, R. Alves, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; J. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

Worldwide coastal low-lying ecosystems are experiencing an increased salinity due to the effect of climate change. The Intergovernmental Panel on Climate Changes (IPCC) has estimated an increase of 0.06-0.22°C per decade in the surface temperature of the ocean between 1956 and 2011, which is considered 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 salinity may cause to tadpoles of the amphibian species Xenopus laevis. To address this objective, X. laevis tadpoles (Gosner 25) were exposed to a range of 3 NaCl concentrations under three temperatures: 20, 23 and 26°C. The following parameters were measured at the end of the test: feeding rate, body weight and growth rates. The differences reported for size between control and NaCl concentrations were mainly due to the tail length. For all concentrations, the feeding rate decreased with increasing NaCl concentration. As well, body weight decreased...
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 columbianus (AMURA: HYLIDAE) TO WATERS CONTAMINATED BY ANTHROPOGENIC 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. columbianus exposed to contaminants of agricultural, livestock and mining (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 AMPHITOX 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 (91%), 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 agricultural and livestock contamination, unlike other studies which report a decrease in 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 landscapes. Some inhabit water bodies either permanently or during some time of their life cycle. Others use predominantly terrestrial habitats. 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 calculation to test the risk to groups of amphibians and reptiles greatest 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 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 could 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-life amphibians is historically evaluated using surrogate toxicity data from standard fish species. Recently published data analyses have demonstrated that amphibian ecotoxicology endpoints are parallel to fish. These analyses are particularly suitable for amphibians because these analyses are parallel to fish. These analyses are particularly suitable for amphibians because amphibians and fish are exposed to similar environmental conditions, i.e., aquatic exposure. This is not true for all amphibian species, however, as amphibians differ in their sensitivity to toxicants. The current study evaluated the utility of fish as surrogates for amphibians by analyzing tadpole mortality data from NOAH studies, which are designed to test the potential for amphibian toxicity in aquatic environments. The NOAH database contains mortality data from a wide range of chemicals and exposure conditions, including those with and without fish. The NOAH database was used to compare the mortality of fish (Xenopus laevis) and amphibians (D. columbianus) exposed to the same chemicals and exposure conditions. The results showed that fish and amphibians exposed to the same chemicals exhibited similar mortality patterns. This suggests that fish may be useful surrogates for amphibians in aquatic risk assessment. However, further research is needed to evaluate the utility of fish as surrogates for amphibians in non-aquatic environments, such as soil and plant surfaces. Overall, the results indicate that fish are a reasonably good predictor of amphibian toxicity as there were no statistically significant differences in NOAH 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 presentation 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 increased risk that common voles were negatively affected by multiple applications 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 (2017) which is based on the results of the EFSA screening program (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 methodology. We found 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 these groups; 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 for extrapolation to assess the hazard/risk for amphibians and reptiles; 4) Identify proposed
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 reptiles 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 Amphimove: Moving patterns and microhabitat selection of European anurans in agricultural landscapes

The current decline of amphibian populations on global and local scales is discussed by scientists around 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 combination to complex moving 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 Amphimove 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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The objective was to determine oxidative stress and metal burden in anuran tadpoles from historically metal exposed populations (Hg/Pb), and to assess if tadpoles from impacted sites have increased tolerance to metals relative to tadpoles from reference sites. Metal body burden, oxidative stress biomarkers and metallothioneins (MT) were measured in Pelobates fuscus tadpoles from reference and metal contaminated sites. Additional tadpoles (20 per site) were collected and exposed in lab conditions during 24h to Hg or Pb levels above the median lethal concentrations reported for amphibians (1.5 and 10.5 mg/l, respectively). The parameters mentioned above plus mortality were monitored at the end of the assay. Field-collected tadpoles from Pb and Hg polluted sites had higher metal body burden than those from reference sites (median per site as d.w. 540.4-708.1 vs 2.6-9.5 ng Pb/g; 768.2-3103.5 vs 0.1 ng Hg/g; all p < 0.01). Levels of MT (median, µg/g tissue) were significantly higher in tadpoles from Hg polluted sites than in the rest of locations (248.5-307.7 vs. 63.9-138.6 µg/ g, p < 0.01), suggesting that MT levels can be induced in natural populations, by the sum of environmental stress factors. Exposure to Hg caused mortality of all individuals, while Pb did not result lethal to tadpoles. Laboratory exposure revealed that experimental treatment rather than pollution at the origin site determined Pb body burden (controls: from reference site 96.7-120.4 ng/g, from Pb site 118-491.6 ug/g; Pb-exposed: from reference site 368.9-54760 ng/g from Pb site: 9043.5-78452.4 ng/g), showing that Pb was readily bioavailable for exposed tadpoles. Lab exposure to Pb increased MT levels in tadpoles from reference sites (exposed vs. non-exposed: 116.3 vs. 41.70 µg/g; p < 0.01), but not in those from Pb-polluted areas. Oxidative stress biomarkers did not differ either between origin sites or because of experimental exposure to Pb. MT levels in tadpoles from reference populations that were taken to the laboratory and kept under laboratory conditions for four weeks (307.7 vs. 63.9 µg/g) were significantly higher in tadpoles from Hg polluted sites (median, 78452.4 ng/g) than in those from reference sites (96.7-120.4 ng/g). This indicates that MT levels may increase after laboratory conditions, though data are insufficient to determine if this is a vulnerable life stage for Pb. MT levels in tadpoles from reference sites that were taken to the laboratory and kept under laboratory conditions for four weeks (307.7 vs. 63.9 µg/g) were significantly higher than in those from Pb-polluted areas.

MO054 Do historically metal-exposed amphibian populations acquire resistance to lethal levels?
F. P. Morão, S.C. Novaes, Polytechnic Institute of Leiria / MARE IPLeiria; I. Vieira, A. Estrella, Polytechnic Institute of Leiria / MARE IPLeiria

Anuran tadpoles from historically metal exposed sites (Hg/Pb) were sampled for blood and skin tissues, immediately placed in formaldehyde, and fixed for 24 h. MT levels in tadpoles from reference populations that were taken to the laboratory (105.9-708.1 vs 2.6-9.5 ng Pb/g; 768.2-3103.5 vs 0.1 ng Hg/g; all p < 0.01) were significantly higher in tadpoles from Hg polluted sites than in the rest of locations (248.5-307.7 vs. 63.9-138.6 µg/ g, p < 0.01), suggesting that MT levels can be induced in natural populations, by the sum of environmental stress factors. Exposure to Hg caused mortality of all individuals, while Pb did not result lethal to tadpoles. Laboratory exposure revealed that experimental treatment rather than pollution at the origin site determined Pb body burden (controls: from reference site 96.7-120.4 ng/g, from Pb site 118-491.6 ug/g; Pb-exposed: from reference site 368.9-54760 ng/g from Pb site: 9043.5-78452.4 ng/g), showing that Pb was readily bioavailable for exposed tadpoles. Lab exposure to Pb increased MT levels in tadpoles from reference sites (exposed vs. non-exposed: 116.3 vs. 41.70 µg/g; p < 0.01), but not in those from Pb-polluted areas. Oxidative stress biomarkers did not differ either between origin sites or because of experimental exposure to Pb. MT levels in tadpoles from reference populations that were taken to the laboratory and kept under laboratory conditions for four weeks (307.7 vs. 63.9 µg/g) were significantly higher than in those from Pb-polluted areas. Oxidative stress biomarkers did not differ either between origin sites or because of experimental exposure to Pb. MT levels in tadpoles from reference populations that were taken to the laboratory and kept under laboratory conditions for four weeks (307.7 vs. 63.9 µg/g) were significantly higher than in those from Pb-polluted areas. Oxidative stress biomarkers did not differ either between origin sites or because of experimental exposure to Pb. MT levels in tadpoles from reference populations that were taken to the laboratory and kept under laboratory conditions for four weeks (307.7 vs. 63.9 µg/g) were significantly higher than in those from Pb-polluted areas. Oxidative stress biomarkers did not differ either between origin sites or because of experimental exposure to Pb. MT levels in tadpoles from reference populations that were taken to the laboratory and kept under laboratory conditions for four weeks (307.7 vs. 63.9 µg/g) were significantly higher than in those from Pb-polluted areas.

MO055 Assessment of metal contamination levels and stress responses of endangered sea turtles of São Tomé and Príncipe
I. F. Morão, S. C. Novaes, Polytechnic Institute of Leiria / MARE IPLeiria; I. Vieira, A. Estrella, Polytechnic Institute of Leiria / MARE IPLeiria

São Tomé Island harbors important sea turtle nesting and feeding sites. However, insufficient enforcement of environmental laws to avoid illegal take of nesting females and eggs, associated with a great lack of knowledge about how these species interact with their environment and how human activities impact their survival in the region, constitute significant challenges for sea turtle conservation. Through current local conservation projects, some information on genetics and nutrition of sea turtle populations is being unveiled but very little is known about how pollutants affect sea turtles. The hypothesis of this study was to assess the metal concentrations accumulated by two species of Tomé sea turtles (Eretmochelys imbricata and Chelonia mydas) and infer about possible impacts of such contamination on their general stress responses and health status. More specifically, the final goal was to find correlations between metals in their tissues and the expression of key genes involved in detoxification/sequestration and metal transport, antioxidant responses and oxidative stress, immunological responses, mitochondrial respiratory and energy production, among others, which could be indicative of these organisms health and future viability. To achieve these goals, nesting female turtles were sampled for blood and skin tissues, immediately after egg laying in their well-documented spawning sites in S. Tomé. Skin samples were collected from the right front flipper of the turtles and stored at -20°C until analysis of metal concentrations. Blood samples were withdrawn from the external jugular vein and stored in RNAlater at -20°C until RNA extraction and gene expression analysis using quantitative real-time PCR (qPCR). Additionally, body mass and carapace length were also recorded, along with all information regarding snapping turtle to these selected DLCs. It is anticipated that this research will result in a single AOP 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 AOP 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.
MO056 Ecotoxicology of Africa's three largest reptiles: POPs, metals, eggs, and eggshells
H. Bouman, 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 Leatherbacked 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 proxy for egg contents. Sampling therefore, requires the collection and analyses of unhatched eggs. Relative elemental composition patterns indicated overlaps for the respective egg contents and eggshells of the sea turtles, but not for the crocodiles. We found thicker eggshells significantly associated with higher iron concentrations in the crocodiles. The implications may be that hatchlings may spend more energy to break through the leathery shells, and may therefore affect reproduction. Copper had concentrations that raised concern in all three species. The strontium concentration in the eggshells of the Leatherback Turtle was high. Mercury, copper, strontium and lead were the most monitored metals in these two reptile eggs in Africa. They are also monitored in other co-occurring pollutants, such as POPs and endocrine disrupting compounds, since sub-lethal effects, especially when the eggs are covered, is difficult to discern. Based on the work presented here and those of others, it is obvious that more studies are needed to obtain a better picture of the chemical and biological interactions involved with Africa’s three largest reptiles in.

MO057 Improving knowledge flow: from consumer to environmental risk assessment
L. Villamar Bouza, s. barmaz, R. Sharp, EFSA - European Food Safety Authority / Pesticides Unit; M. Arena, EFSA - European Food Safety Authority / Pesticides; D. Aldana, EFSA - European Food Safety Authority / Pesticides

The assessment of pesticide residues levels in environmental matrices is part of the risk assessment for non-target organisms under Regulation (EC) no 1107/2009. In the case of risk assessments for birds and mammals, according to EFSA (2009), the Tier 1 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 surround the compound concerned and is insufficient to derive such DT₅₀ values. These are then further evaluated with specific kinetic tools (FOCUS kinetics). It should be noted that the refinement of the RUD values is done only in rare cases since the dataset at the basis of the default values is relatively large. These refinements allows for a more realistic risk assessment accounting for the differences in residues decline due to the crop type, growth stage, climatic conditions across EU zones and to specific characteristics of the substance under assessment. Other parts of the data used for the consumer risk assessment for pesticides can also provide information for the environmental risk assessment. In particular, metabolism studies in plants are used for the identification of the pertinent metabolites to be further considered in the risk assessment of birds and mammals. The metabolism data for hen and rabbit can also be used for addressing such metabolites. The main scope of this work is to further analyse the standard dataset available and the specific guidance in use for the consumer risk assessment in order to better define how the data and knowledge developed in the context of the consumer risk assessments (internationally agreed methodologies, existing guidance documents) could be integrated in the environmental risk assessment. Particular consideration is given to the possibility of extrapolation between crops, use patterns (e.g. growth stages, application number) and European zones.

MO058 Increasing salinisation effects on Pelophylax perezi populations - Could historical exposure drive current diversity 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 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 8-10) were exposed for 96h, and to what concerns to the output of pollutants in 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-impacted and non-impacted populations, differences were not found. For this endpoint, NaCl revealed to be more toxic than seawater (EC₅₀ of 14.04 and EC₅₀=11.89 mMCl⁻ for seawater, and NaCl, respectively). As well, 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-impacted population, but for non-impacted populations, tadpoles’ growth decrease with the decrease of seawater dilution.

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 the input of pollutants such as 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 (Aguada, central Portugal) and occurred after the first occurrence of wildfires in the year 2018. Water samples from impacted and non-impacted areas were collected. Bioassays were performed using in situ bioassays with freshwater organisms, including tadpoles and daphnids. In the burned area, we observed a high biodiversity of freshwater organisms, including tadpoles and daphnids. For this endpoint, rainwater feeding inhibition were evaluated. The lethality was sought to be determined feeding inhibition, revealed a decrease in the feeding rate of tadpoles from salinization. Results highlighted that the feeding rate of tadpoles decreased with the increase of rainwater, and dilution factor of rainwater. In the non-impacted areas, no differences were found. For this endpoint, NaCl caused significant adverse effects. Highly diluted rainwater increase growth on tadpoles from salinization-impacted population, but for non-impacted populations, tadpoles’ growth decrease with the decrease of rainwater dilution.

MO060 Estronomic effects of an Organophosphorous Flame Retardant (TCP) 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 organisms. 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-Methylvinyl) 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 had been exposed in a formal experiment. Control group (C) and TCP exposed (1 and 10 mg/L), were maintained in controlled conditions and 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

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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. Anthroopogic antidepressant, 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 environmental 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, acetylcholinesterase, lipoperoxidation). Behavioral responses also examined, concerning activity and feeding behaviour. Overall, results provided insights into the effects of fluoxetine at the 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

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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 dibenzop-p-dioxins and furans (PCDD/Fs), dioxin-like polychlorinated biphenyls (DL PCBs) and polybrominated diphenyl ethers (PBDEs) using gas chromatography coupled to a high-resolution mass spectrometer and by using the isotopic dilution technique. The WHO Toxicity Equivalence (TEQ) approach was applied. Median DL PCB concentration was 1820 ng/g lipid weight (l.w.) (range: 474-6120 ng/g l.w.), with statistically significant differences between males and females (Mann-Whitney U test p<0.0472). These levels rivel and even surpass those reported in other marine mammals in the same area. TetraBDEs were the most abundant congeners, with BDE47 ranging from 39 to 560 ng/g l.w. These results confirmed how the Mediterranean subpopulation of striped dolphin is currently subject to a major threat of environmental pollution. To date, few studies have focused on the evaluation of TEQ values and PBDE levels on this subpopulation. Toxicological and risk assessment studies on this species, recognized as an ocean health sentinel at a sub-basin scale, may provide an early indication of potential adverse health effects.

MO063

Assessment of POPs in stranded sperm whales (Physeter macrocephalus) from the Mediterranean Sea

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Physeter macrocephalus is a large toothed whale inhabiting the Mediterranean Sea. This endangered species is subject to a number of threats such as exposure to high levels of contaminants. Several studies show high levels of persistent organic pollutants (POPs) in dolphin tissues from the Mediterranean Sea, but data on sperm whales from the same area are much more scarce. In this study, we analyzed POPs of blubber samples of a stranded sperm whale (Physeter macrocephalus) from 2009 to 2016. Fresh samples were spiked with a suit of PCDD/Fs, PCBs, and PBDEs 13C-labeled standards prior to Soxhlet extraction, and then cleaned-up by using the automated sample preparation system. Samples were analyzed for seventeen PCDD/Fs, twelve DL-PCBs and twenty-seven PBDEs. Quantification was carried out by the isotopic dilution technique by GC-HRMS on a Trace GC Ultra gas chromatograph coupled to a high-resolution mass spectrometer. Samples’ lipid content was determined gravimetrically. The relative abundance of the study contaminants followed the order DL-PCBs>PBDEs>PCDD/Fs. The mean concentration values obtained were: 6420 ng g-1 (2100-20800 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. Our results were in the same order of magnitude that those reported for the same species in the same area by a recent study from other authors. This led us to conclude that 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 PBDE levels, our results were lower that those reported for sperm whales from North Atlantic. The PCDF congeners profile of the blubber of the same whale (hexa-penta>hepta>octa) 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 congeners 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 l.w. and surpassed the threshold of 2100 pg g-1 l.w., in an island of the Pacific coast as starting point of immunosuppression in harbour seals. This high level of contamination is not considered to be the cause of death of these animals, but may have contributed to lowering the defense of their immune system.

MO064

Biochemical and molecular responses to organic contaminants in bottlenose dolphins (Tursiops truncatus gpeyreus) from southern Brazil

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Toxicological and risk assessment studies on cetaceans are focused mainly on PCBs, DDTs, Mirex, Chlordanes (CHL), Hexachlorobenzene (HCB), sampling season (summer or winter) and location (LES and PLE). The best model to describe each biomarker response, with the lowest Aikake 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 ΣPCBs, 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 exposure to metals. Overall, results indicate that the skin of bottlenose dolphins is altered due to exposure to ΣPCBs and PBDEs, which co-varied with ΣPCBs and Mirex. Absence of influence of other contaminant classes over biomarker response might be due to low contaminant levels in sampled dolphins. This might also have been overshadowed by the effects sampling season. Nonetheless, results indicate a significantly higher exposure to PCBs in dolphins with high Mirex 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 butylcholinesterase (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 - 11.343±0.829 nmol ml⁻¹ min⁻¹ in May and 1.444±0.079 - 9.314±1.618 nmol ml⁻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 measured were found to be environmentally relevant to the local community and were found to be below the level of biological concern. 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 risk 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 field studies: (i) a study design commonly referred to the ‘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 sensible enough to monitor the fate of individuals in a treated population over a long period of time, and to fine-tune the model of 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 Neotropic 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 acclimation and potential impacts of persistent organic pollutants such as organochlorine pesticides, PCBs, and PBDEs on nesting aquatic birds of the Trinity River, using Neotropic 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 assayed for PCDD/Fs, PCBs and PBDEs. Levels of organochlorines were assessed by gas chromatography/mass spectrometry (GC/MS) and GC/TCD. 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 - 11.343±0.829 nmol ml⁻¹ min⁻¹ in May and 1.444±0.079 - 9.314±1.618 nmol ml⁻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 measured were found to be environmentally relevant to the local community and were found to be below the level of biological concern. 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.

**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 particularly susceptible to the effects of neonicotinoids. Effects on refueling or orientation behavior could have significant fitness consequences; however, the influence of neonicotinoids on migratory ability is poorly understood. We used white-crowned sparrows (Zonotrichia leucophrys) during spring migration to assess the direct effects of imidacloprid on the migratory behavior of seed-eating passerines. In a previous captive study, we found birds exposed to imidacloprid performed significantly better in orientation trials compared to controls. In this study, 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 via gavage to a single oral dose of imidacloprid (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 in exposed eradicants and transfer 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 urban, agricultural and island use may cause less localized 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 linked to risk to wildlife populations; 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 conflict between protection of human health and wildlife.

MO707 Environmental determinants of the exposure to anticoagulant rodenticides in nontarget species


Anticoagulant rodenticides have some similarities with other bioaccumulative 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 sulfonate (PFOS) being the most important compound. 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 exposures, studies in Japanese quail (Coturnix japonica) and the gull (Larus argentatus) 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, glutathione-S-transferase and glutathione peroxidase), lipid - and protein oxidative damage and biotransformation (cytochrome P450A1 were investigated. Further, hormonal 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 ovo 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. Faecal 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 metabolites. No major metabolites were detected, only small levels of the parent compound were detected. Phenotypic differences were noted between wild caught and captive starlings, but the overall trends were the same. We concluded that the fluoxetine was detectable in feathers in wild birds.

MO073

Monitoring NSAIDs in carrion and avian scavengers form Spain: preliminary results after diclofenac registration for veterinary use
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The serious impact of diclofenac on Asian vultures raised the alarm of the deficient guidance document on bird and mammal focal species for pesticide 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 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 potential 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 risk assessment for wildlife in cereals and thus provide a baseline for more realistic and site-specific risk assessments for birds and mammals.

MO076

Different approaches comparison for evaluation of hypopharyngeal glands (HPG) toxicity in honeybees (Apis mellifera L.)

Honeybees (Apis mellifera L.) are beneficial arthropods that play important roles in nature also 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 proteinic 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 evaluation, in order to select the Method that combines the highest reliability (the smallest technical error), the optimal cost, the least effort and time-consumption. The study was conducted on Honeybees subjected to chronic toxicity studies performed according to the EFSAs guidelines (EFSA Journal 2013;11(7):3295). Insects were treated with four different chemicals in 3 to 5 concentrations. The HPG were obtained from 5 bees per test item (in the highest concentration, which did not cause mortality below 50%) and the negative control. Six different approaches for HPG evaluation were tested: - histopathology (HP) of isolated glands (linear and quantitative measurements, imaging); - histopathology (HP) of whole heads (linear and quantitative measurements, imaging): - whole head (WM) method on isolated gland (linear and quantitative measurements, imaging); - protein absorbance (PA) from isolated glands (quantitative measurements); - protein absorbance (PA) from whole heads (quantitative measurements); - scanning electron microscopy (SEM) (linear and quantitative measurements, imaging). The linear measurements (small and big axis of symmetry) were taken from ten acini from left and right HPG, however for SEM only left HPG were included. The quantitative measurements (number of acini per 1 mm²) were taken during HP, WM and SEM testing. The quantitative measurements of protein absorbance from isolated glands were taken from the wet and right HPG. The analysis of the results showed decreases and increases of acini and protein absorbance depending on the test item. However, observed deviations did not show any statistical significance. According to the performed studies for evaluation of hypopharyngeal glands development the linear measurement combined with imaging should be used.

MO077

Bird and mammal focal species for pesticide risk assessment in rice
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Ecotoxicological risk assessment for birds and mammals is required for the registration of pesticides in Europe to assess potential risks to wildlife through exposure to 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 proposed for rice are thus those known from the dry environment 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 potential 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 risk assessment for wildlife in cereals and thus provide a baseline for more realistic and site-specific risk assessments for birds and mammals.
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 reintroduction 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 success of the project, and one of these is the exposure to lead from the ammunition used for hunting. An exposure to lead, even at sub-lethal levels, can be a stressful situation that implies an increase in circulating corticosterone levels. In birds that are rare and difficult to capture, blood sampling for both lead and corticosterone analysis can be a difficult task and with some risk for birds. For this reason we developed an innovative method to study exposure to pollutants and their respective biomarkers is being directed towards the analysis of easy-to-collect samples, 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 by analysing moulted feathers found in the field, relating them to corticosterone levels in the same feather and defining the factors that affect the fluctuations 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 concern this study is that the post mortem period, the period of rachis feathers of key biometric formation to the concentration of corticosterone, and was significantly influenced by the collection area and the individual (feather). In 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

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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 periods from death to analysis of biochemical biomarkers provides more accurate results. In this study, we evaluated the stability of GST and EROD activity in post mortem liver samples from kelp gulls (Larus dominicanus) kept at different temperatures (4°C, 22°C, 37°C) for up to 24 hours. We found that GST activity remained stable for up to 24 hours, while EROD activity decreased almost completely within the first hour post mortem. Furthermore, we observed that the pH of the samples was significantly higher in the post mortem liver samples compared to the fresh liver sample.

MO080
Investigating thyroid disrupting effects of organohalogenated contaminants in White-tailed eagle nestlings

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The measurement of biomarker responses to stress hormones in white-tailed eagles (Haliaeetus albicilla) can accumulate a wide range of organohalogenated contaminants (OHCs), due to their apex trophic position. Their diet consists mainly of fish and seabirds, thus long food chains and a high potential for biomagnification of OHCs. The nestlings can therefore be exposed to high levels of certain OHCs through maternal transfer to the eggs, and later through diet. Concentrations of per- and polyfluoroalkyl substances (PFASs) have recently been shown to exceed those of other legacy OHCs in this species and accordingly, it is crucial to investigate the exposure of these pollutants and their respective biomarkers is being directed towards the analysis of easy-to-collect samples, 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 by analysing moulted feathers found in the field, relating them to corticosterone levels in the same feather and defining the factors that affect the fluctuations 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 concern this study is that the post mortem period, the period of rachis feathers of key biometric formation to the concentration of corticosterone, and was significantly influenced by the collection area and the individual (feather). In 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.

MO081
Assessment of exposure and effects of Hg levels in feathers of White-tailed eagles (Haliaeetus albicilla) and Northern goshawks (Accipiter gentilis) nestlings from Norway

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Anthropogenic activities have led to a global increase of Mercury (Hg) in the environment. Due to its toxicological properties, Hg has been extensively studied in many species of wildlife. However, the assessment of Hg levels in the environment is challenging, as Hg is known to be abundant in the marine environment. In this study, we investigated the exposure of Hg and its effects at the biochemical level using birds as model organisms. We collected feathers from white-tailed eagles (Haliaeetus albicilla) and Northern goshawks (Accipiter gentilis) nestlings from Norway. Samples were obtained in 2014 from the nestling WTE (n=14) and NG (n=11) in northern Norway (N-68.30 – 68.47 °C, 24.54 – 25.27°-Troms- and N-78.67 – 37.69°E, 20.39 – 33.45°-counties, respectively). Total Hg in feathers, total and free plasma corticosterone levels were analysed, along with following blood chemical parameters (BCPs): alanine aminotransferase (ALT), aspartate aminotransferase (AST), total protein, aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, cholesterol, triglycerides, creatinine, amylase, glucose, creatinine, bilirubin, potassium and uric acid. Stable carbon (13C, 15N) and nitrogen isotopes (15N, 14N) were analysed in feathers body to evaluate inter- and intra-specific contaminant exposure. Due to the low amount of feather samples, Hg could only be analysed in 13 WTE and 8 NG. Mean ± SD were 0.5 ± 0.34 mg/kg in NG and 3.0 ± 1.34 mg/kg in WTE. The significantly higher levels in WTE than in NG (F(1, 20) = 7.61, p<0.01) may be related to different dietary input, as confirmed by stable carbon and nitrogen isotope analysis of body feathers. The marine prey of WTE seem to determine the Hg loads, as Hg is known to be abundant in the marine environment. Due to its toxicity, legislation for the removal of organohalogenated contaminants (OHCs) has been imposed, due to their apex trophic position. Their diet consists mainly of fish and seabirds, thus long food chains and a high potential for biomagnification of OHCs. The nestlings can there-
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, most 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 concentrations of As, Cd, Co, Cr, Cu, Mn, Ni, Pb, Se and Zn were 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 savii, Nyctalus leisleri, Pipistrellus pipistrellus, Pipistrellus pygmaeus). Concerning the metal concentration obtained in each sampling tissue, significant differences were found between the concentrations obtained in each species for all metals (P < 0.05), except for Zn (P = 0.223). Significant differences were also found between category of samples (lethal vs. non-lethal) for metals such as Cd, Cu, Mn, Ni and Pb 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.

Metallic element composition of egg contents and eggshells of the Kelp Gull
Larus dominicanus
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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, Tl, 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 120 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 (p = 0.05) positive regressions between egg contents and eggshells. Chromium and Zn showed a positive regression between the contents, but 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.

Heavy metals concentrations in Mediterranean Osprey eggs: variations by location, habitat and egg constituent part
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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. Metal concentrations (Cd, Cadmium, Cr, Chromium, Cu, Copper, Mn, Manganese, Ni, Nickel, Pb, Lead, Se, Selenium, Zn, Zinc) in egg analyses were carried out with the aim to: 1) evaluate geographical patterns of for possible identification of inputs at the regional scale; 2) to evaluate differences in concentrations between samples from different habitats (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 first attempt at a 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

Interactive effects of vitamin E and BDE-47 yolk supplementation on morphology and oxidative status of yellow-legged gull embryos
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Oviparous mothers transfer to the eggs components that have both independent and combined effects on offspring phenotype. Functional interactions between egg components, such as antioxidants and contaminants, are of particular interest since the concentration in the choice of one component has effects on offspring traits that depend on 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-transfered contaminants. Polychlorinated diphenyl ethers (PBDEs) are a family of brominated flame retardants that have been widely used as non-interactive 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 related to the exposure to these chemicals are currently virtually unknown. In addition, 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 unexplored, 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
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Margaritifera margaritifera is a freshwater bivalve mollusk threatened with extinction in Europe. The population of this freshwater pearl mussel has declined by 95% 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 (Bordogne - 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 ecoecological 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 lifestyle 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 Efate 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
LCIA method developments in a global perspective: Status and outlook (P)

MO090 A tool to integrate consumer and environmental exposure in life cycle impact assessment
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Direct exposure of consumers to chemical ingredients within our daily products is a major public health issue since we spend on average 80% of our time in close 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 LCIA does not account for these scientific obstacles such as: (a) the inclusion of the dynamical effects of indoor pollution on human health and (b) 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 explicitly represents 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 evolution of indoor environmental conditions. 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. Freshwater LCIA 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 to (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 elemental 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 direct and indirect emissions and their 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
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The objective of this study is to integrate the impact of WSCW 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 topsoil erosion, the ProScale hazard classes and an OEL based correction factor have 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). USEtox determines effect factors for carcinogenic and non-carcinogenic chemicals separately. Both methods have separate factors for inhalation and oral exposure. In this study we focussed on oral exposure. Both methods have separate factors for inhalation and oral exposure. In this study we focussed on oral exposure.

MO095
Impact of heavy metals on human toxicity using LCA: a case study for Wallonia
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This study aims at studying the human toxicity of corn farming in Wallonia, Belgium. The USEtox 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 fertilizers. But direct and indirect emissions from the composting of the manure have been accounted. The chromium is measured and therefore, unspecified chromium is used as emissions. However, it is known that the chromium in natural environment is mostly Cr (III) and this could really decrease the impact as the specification factor for unspecified chromium, is, in USEtox, 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 the Chromium emissions are 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, especially pig manure. However, zinc is abundant and is an important trace element in the human body. It is useful for growth, bone and brain development, etc. and the European Commission recommends the consumption of 7-10 mg of zinc by person and per day. Moreover, mammals are able to eliminate zinc, therefore they are able to maintain a constant level of zinc independently of the exposure level. Consequently, only the exposure to high doses can have toxic effects. A test was made using the characterization factor of zinc equal to 0 in the USEtox model. In this case, the corn cropping obtains a human toxicity, non-cancer effect divided by 12 compared to the base case and mostly related to lead and mercury emissions in the soil. In both case, the contribution of pesticide is negligible. In conclusion, although the uncertainties about toxicity categories are well-understood, this study underlines the impact of the user hypotheses and shows that a detailed analysis of the results is essential for a critical view on the toxicity results.

MO097
Comparing ProScale Hazard Factors with USEtox Effect Factors for human toxicity
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The purpose of this study is to compare side-by-side, the Hazard Factors (HF) of ProScale®, and the Effect Factors (EF) for human toxicity of USEtox®, 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 hazard effect severity based on Physical Hazard and Environmental Hazard Substance Classification. USEtox determine effect factors for carcinogenic and non-carcinogenic chemicals separately. Both methods have separate factors for inhalation and oral exposure. In this study the comparison was only carried out for the inhalative exposure route. The factors were then filtered into two different sets of substances. All substances having a carcinogenic effect factor were compared to the resulting Hazard factor as calculated with the ProScale method. Substances not having a carcinogenic effect factor in USEtox were compared separately. Tendencies of correlation can be identified, but differences are large. Interesting discrepancies, also of principal matters, have been identified. The results shown are very first results from comparison of ProScale HFs and USEtox EFs for human toxicity. Further work is needed, and under way.

MO098
Integrating the Use Phase Impacts of Building Materials into Near-Field LCA Characterization
L. Huang, University of Michigan / Dept of Environmental Health Sciences; N. Anastas, US Environmental Protection Agency / National Risk Management Research Laboratory; P. Egeghy, D. Vallero, US Environmental Protection Agency / National Risk Management Research Laboratory; J.C. Bare, US. Environmental Protection Agency / National Risk Management Research Laboratory; O. Jolliet, University of Michigan

Historically, LCA has focused on impacts with far-reaching temporal and spatial scales, and not exposures to near-field goods such as consumer products and industrial chemicals. 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. 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 have been poorly characterized. This poster will describe the research project, including the conceptual framework which demonstrates the methods by which the EPA intends to include exposures to these goods, the definition of Product Intake Fraction (PiF), the many exposure pathways being characterized, the methods for development of PiFs, and the data and models being recommended for a variety of populations to support this characterization. Finally, data gaps and other research needs will be discussed along with the future direction of the project. Keywords: LCA, LCIA, LCA, Life Cycle Impact Assessment, Life Cycle Management - This work was conducted under US EPA Contract No. EP-16-C-000070 with the University of Michigan. Disclaimer - 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.

MO099
Combined use of Mixed-Integer Optimisation and Thermodynamic, Molecular and Charge Density attributes for predicting Life Cycle Production
Impacts of Chemicals
R. Calvo-Serrano, G. Guilén Gosalbez, 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 analysis cannot be applied, a simplified version is used instead. These streamlined LCA (SLCA) follow the same basic 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 chemical properties of the substance as attributes, for a better characterisation of the chemical, 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 profile partitions, being able to obtain reasonable LCA impact estimates such as Global Warming Potential (GWP.33.55%) or Eco-Indicator99 (EIP99) (18.34%).

MO102 Advancing nutrient modelling in eutrophication methods for life cycle impact assessment

Anthropogenic 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-matrix reviewed of the underlying fate and transport (F&T) models. Results of the review point to several key recommendations for further scientific development of midpoint eutrophication methods in life cycle assessment (LCA). Current eutrophication methods in LCA are limited. Most LCA methods use highly 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 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, sediments, soils, and air), 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, Koerner N, Chrupin 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 expressed in this presentation 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.
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, 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 at coastal areas, near freshwater 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.


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 way each one quantifies the impacts. After thorough study of the main road stems 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: 1) 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 Retaining area per a in a watershed.

MO105 Identification of methodological challenges remaining in the assessment of a water scarcity footprint P. Torres, University of Aveiro / Department of Environment and Planning; b. Bidout, 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 cannot recharge the groundwater). After this review the article continues with the in-depth analysis of the methodologies to this impact category, this work presents the achievement of the most relevant methods for quantifying the water use in LCA of several renewable energy systems in Spain: 1) 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 Retaining area per a in a watershed.

MO106 Filling the Gap of Overfishing in LCIA: Eco-factors for Global Fish Resources M. Stucki, Zurich University of Applied Sciences / Institute of Natural Resource Sciences; L. Eymann, ETHZ Swiss Federal Institute of Technology; B. Kelle, R. Itten, 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 ($S_{MSY}$) 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 $S_{MSY}$ 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 by 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 developed approach can be applied to all LCA of fisheries and is able to reflect the regionalised impacts caused by the overexploitation of fish resources.

MO107 Constructing life cycle inventories for the hydroelectric sector in Peru: methodological considerations and environmental impacts I. Vázquez-Rowe, Pontifical Catholic University of Peru / Civil Engineering Environmental Science; D. Verán-Leigh, Pontificia Universidad Catolica del Peru / Civil Engineering Environmental Science

According to recent reports, hydropower currently accounts for 16% of worldwide electricity generation. Low carbon emissions are usually related to this source of energy, making it an attractive option for nations with hydropower potential to meet increasing electricity demand without relying on burning fossil fuels. However, the new wave of hydropower plant construction is occurring mainly in three tropical river basins: Amazon, Congo and Mekong; therefore, an additional environmental impact must be considered: biogreenhouse gases (GHG) emissions due to decomposition of biogenic C stocks in reservoirs. Peru is planning on installing up to 2,000 MW of installed capacity in hydropower until 2021, but the input and output flows, as well as the environmental impacts that these generate have not been explored. In this context, a set of three run-of-river hydropower plants built in the past decade located along the Peruvian Andes were analyzed from a life-cycle perspective. The main objective of the study was to provide the set of detailed life cycle inventories for each of these three hydropower plants with the aim of having specific information for real conditions in Peru. This information was used to compute the environmental impacts linked to the generation of electricity at the plants. Although the main aim was to determine the GHG emissions linked to this process, considering the important policy implications of decarbonizing the Peruvian electricity grid, other environmental categories, such as eutrophication, acidification, and effects due to residence and impact assessment methods were also considered. The results computed show that GHG emissions per kWh of electricity produced were in the lower range of emissions observed in the literature, in all three cases below 3 g CO2eq per kWh. Biogenic emissions represented less than 5% of the total GHG emissions despite their location in a tropical nation, due to the low disturbance of the landscape in the Highlands, as well as the cold temperatures that are present in the reservoirs. Results intend to be of utility for an array of applications, including relevance in decision-making in the energy sector, policy-making at a national level, considering the implications in terms of meeting the Nationally-Determined Contributions to mitigate climate change in the frame of the Paris Agreement, and for the international LCA community in an effort to expand the amount of inventories available for different geographical and technological conditions.

MO108 Global scale characterization factors for freshwater eutrophication from nitrogen and phosphorus emissions to water and soil F. Remboud, Radboud University / Department of Environmental Science; R. Van Zelm, Radboud University / Department of Environmental Science

Eutrophication is a key water quality issue triggered by increasing nitrogen (N) and phosphorus (P) levels and potentially posing risks to freshwater biota. In life cycle impact assessment, spatially explicit characterization factors (CFs) of phosphorus emissions to freshwater causing eutrophication have been derived. However, for nitrogen impacts, no efforts have been undertaken yet. Moreover, rather for agricultural emissions of P nor for N spatially explicit CFs have been derived. Therefore, the goal of this research was to determine spatially explicit CFs for
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 potentially not 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 WWTP emissions 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 time-dependent 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 (MRIO) 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. Van Besten, 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 Exiobase. A monetary consumption 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. Shironittta, 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 used the composite stressor approach including the shift toward a service economy and the increase in greenhouse gas (GHG) emissions embodied in emissions 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 those environmental effects. Specifically, we employed a multi-regional 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 input 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 sectors and 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 shift to the industrial structure with less emission-intensive material goods.

MO111
Influence of substance coverage on impacts from the electricity sector
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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 (MRIO) includes emissions to air from countries and 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 (ENEEI) 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 ENEEI covers 229 substances, including 51 radioactive isotopes. By comparing inventories and databases at midpoint level, we show that using ENEEI 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
K. Kamemoto, Shinshu 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 are coming from. It offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to any region, city or country, or even location. In this way it can be used to study regional and global biodiversity footprints, air pollution footprints, or main carbon footprint footprints using Ecoinvent, a global simulation tool that can represent the supply chain. As the method is based on a geographical information system (GIS) which is often available at high resolution and comprehensive, generic databases for LCA. It is underlined by the presence of earth observatory GIS data that is tagged to an economic sector to any region, city or country, or even location. In this way it can be used to study regional and global biodiversity footprints, air pollution footprints, or main carbon footprint footprints using Ecoinvent, a global simulation tool that can represent the supply chain.

MO114
LCA data machine applied
A. Ciroth, GreenDelta; M. Strocka, GreenDelta GmbH
In any LCA study, finding data sets that are “fit for purpose” is probably one of the aspects which consumes most time in the 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 intransparent, 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 automates 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...
MOI15 Static and dynamic modeling of high performance buildings: Comparison of average and peak electricity use: a mesoscale 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

Trifed on the life cycle assessment (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 variations. In our study, particular attention is paid to the dynamic characteristics of two buildings’ electrical energy consumption in relation to their respective power grid characteristics. Our LCA model incorporates hourly energy use data for on-site renewable production at a net-zero energy building (NZEB), and hourly or 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 (DLCAs) 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 erase marginal 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 our 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 DLCAs research domestically.

MOI16 Life cycle framework for environmental assessment of public transport systems
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Several studies have assessed life cycle environmental impacts of public transport systems. However, there is no single platform, software or tool for comparing the environmental performance of different commuting patterns. 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 the environmental impacts linked to the construction, 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; S. Groslandmet, 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 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 road-rail routes in Belgium. The aim of this analysis is to compare the environmental impacts of these 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 different 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 obtained in this research on the initial impact of modal split in rail freight development 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 pesticides - Development of an objective criterion to support visual assessment of SFO fits of parent soil degradation
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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 methodology 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, and the subjective nature of visual assessment. Based on group discussions, we derived group consensus scores. Consensus scores showed little correlation with Chi2err (R² = 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 (R² = 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 model residual data). The SWARC criterion is applied as a weighted height, depending on 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 higher SWARC values, SFO may still be acceptable (particularly if SWARC < 65), but DFO must also be assessed. Testing of the criterion for metabolite fits shows that it was also applicable for some cases. However, further work is also needed for the calculation of the criterion. Taken together, we provide a novel tool that quantifies the visual assessment of SFO fits. This can guide decision making and thus help to reduce subjectivity in regulatory assessments.

MOI20 "Southside"- Bridging the hemispheres - Global use of field trials based on ecoregion similarities between New Zealand, Chile and Europe
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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 (ENASGIPS) had been developed (OECD 2016) for gaining acceptance of field studies conducted in North America to Europe and vice versa, where climate and soil conditions are similar to the regions of the world. An experimental and GIS/modeling feasibility study ("Southside") was initiated to demonstrate if TFD studies conducted in the Southern hemisphere (i.e. New Zealand, Chile) under climatic, soil and cropping conditions similar to conditions in the Northern hemisphere may deliver similar soil degradation rates and DegT50 endpoints than those from Europe. Similar similarity zones had been identified between the New Zealand and Chilean sites and EU/N.A.T.A. using the OECD ENASGIPS tool as well as an adapted GIS crosswalk with JRC-EFSA climate and soil maps for EU. The trial sites had soil types ranging from loamy sands, sandy loam, loam and silty loams. In New Zealand the sites were located on the Northern Island having an average annual air temperature of ~ 12-13 °C and an average cumulative annual rainfall of ~ 780-970 mm. In Chile the sites were located in the Región del Bio-Bio east of Concepción having an average annual air temperature of ~ 14 °C and an average cumulative annual rainfall of ~ 800-900 mm. The terrestrial field dissipation study (TFD trials) were conducted according to OECD 232 (DegT50 module, soil covered with sand) with different pesticides at 3 sites in New Zealand and in Chile, having no historic use of these pesticides. All pesticides were applied in commercial formulations as a tank mix together in the same spraying on the same field plots at the same time. The field DegT50 were normalized to reference conditions (20°C, moisture pF2) during kinetic analysis according to FOCUS, considering local soil conditions and weather data to estimate soil temperature and soil moisture with the PEARL model (as had been done with the EU studies). The quality indicator values of curve fit to data (Chi squared) were found to be acceptable. The normalized SFO DegT50 in the “Southside” trials in New Zealand were found to be in the range of those from TFD studies in Europe using the same study design.

MOI21 Residues of currently used pesticides in Central Europe arable soils: status quo, reasons and consequences
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Current agricultural management is usually based on high consumption of pesticides which may bring a lot of environmental problems. Alarming results from monitoring pesticide residues in EU groundwater and surface water evoke the question of whether the arable soil can contain significant contamination as a result of the intensive use of pesticides in the present or past. Therefore, in 2014 - 2017, agricultural soil was monitored at more than 100 locations in the Czech Republic for more than 50 representatives of currently used pesticides, their selected transformation products and also banned atrazine and simazine with their transformation products. The results showed that the contamination of the monitored soils with the analysed pesticides is quite extensive. At least 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 tert-butylphenols, it was concluded that banned toxic simazine is still introduced significantly to the soils as an allowed impurity of massively applied tertbutylazine. 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 the 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 GACR project 15-20065S.

MOI22 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

DegT50 were normalised to reference conditions (20°C, moisture pF2) during kinetic analysis according to FOCUS, considering local soil conditions and weather data to estimate soil temperature and soil moisture with the PEARL model (as had been done with the EU studies). The quality indicator values of curve fit to data (Chi squared) were found to be acceptable. The normalized SFO DegT50 in the “Southside” trials in New Zealand were found to be in the range of those from TFD studies in Europe using the same study design.

MOI23 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 for general water quality research by industry, governments, and academic organizations. However, use of this monitoring 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.

MOI24
Multi-year evaluations in the FOCUS Surface Water assessment - results of beta testing

D. Webber, M. Brauer, Eurofins Regulatory AG / Environmental and Ecological Modelling; A. Boeckhan, Bayer AG, Research & Development, Crop Science; G. Spicker, ADAMA Deutschland GmbH; D. Schaefer, Bayer Crop Science / Environmental Safety

The evaluations of the predicted environmental concentrations (PEC) of active substances in surface water are based on a “single year” approach with an initial 6 year warm-up phase followed by 16 months of the year selected by the FOCUS group. Unlike in groundwater with a 20 years assessment period, surface water exposure calculations based on a “single year” approach can be strongly affected by individual rainfall events (EFSA, 2013) which was discussed repeatedly by authorities, industry and academia (Klein, 2013, Goerlitz, 2015; Bach et al., 2016; Poulsen, 2016). This presentation provides background on the technical methods and assumptions currently implemented into a software tool (Webber et al., 2017) that allows 20-year simulations of FOCUS surface water scenarios. In addition, results of a beta test including revealed technical issues, problems and assumptions are discussed. The software tool in its current state can easily be adapted to updated technical requirements or changes, i.e. any comments from official side (EFSA FOCUS Repair Group) or from other sources can be addressed according to given consent. The aim is to contribute to the development of an improved and generally accepted approach for surface water calculations representing a realistic worst case based on a robust evaluation. Bach M et al. (2016): Pesticide exposure assessment for surface waters in the EU. Part I: Some comments on the current protection. P.Vel. Nature 2015: 172-174.2: KMF: help desk contact G (2015): Multiyear FOCUS surface water modelling: Options and Proposals for Realisation. XV: Symposium in Pesticide Chemistry. Piacenza, Italy. September 2015 Klein M (2015): Long term surface water simulations using the FOCUS scenarios. Pesticide Behaviour in soils, Water and Air, York, UK. September 2013 Poulsen V (2016): Higher tier assessments of aquatic and terrestrial studies. AGCHEM Forum, Barcelona. September 2016 Weber et al. 2017: Multi-Year evaluations in the FOCUS Surface Water assessment. Conference Pesticide Behaviour in Soils, Water and Air, York 2017.

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


MO126
Determination of runoff and drainage triggers for PEC surface water using automated simulation with FOCUS models

B. Kind, A. Guckland, J. Kleinmann, WS-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 PEC50 values were calculated for different crops and different application times within the study area. The 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.

MO127
Quantitative exploitation of passive sampler data for pesticide mass flow calculation in catchments and exposure risk evaluation

T. Gallo, Luxembourg Institute of Science and Technology; M. Bayerle, D. Pitois, V. Huck, Luxembourg Institute of Science and Technology LIST

Pesticide monitoring remains the blind spot in WFD monitoring schemes because of the episodic occurrence of their emissions following application periods. Full coverage of relevant exposure periods is logistically impossible on a larger scale with classical monitoring methods like grab or automatic sampling. Passive sampling allows to cover a cost-effective period that is expected to derive land and crop specific loads in catchments and exceedance probabilities of EQS values resulting in a risk map of impacted surface waters in Luxembourg.

MO128
Spatially distributed environmental fate modelling of terbuthylazine in a mesoscale agricultural catchment using passive sampler data

M. Gassmann, University of Kassel / Department Water Quality Management - Modelling and Simulation; T. Gallo, Luxembourg Institute of Science and Technology; J. Farlin, Luxembourg Institute of Science and Technology LIST

The impact of agricultural practices on water pollution can be assessed by process-based reactive transport modelling using catchment scale models. Most previous studies only used concentrations at the catchment outlet for model calibration and validation. Thus, even if the applied model is spatially distributed, predicted spatial differences of pesticide loss cannot be directly compared to observations. In this study, we applied the spatially distributed reactive transport model Zin-Agritra in the mesoscale (78 km²) catchment of the Wark River in Luxembourg in order to simulate concentrations of terbuthylazine in river water. In contrast to former studies, we used six sampling points, equipped with passive samplers, for pesticide model validation. At each sampling point, event mean concentration of six events from May to July 2011 were calculated by subtraction of baseflow-mass from total collected mass assuming time-proportional uptake by passive samplers. Continuous discharge measurements and high-resolution weather data was used for model simulation and calibration and validation. Detailed information about maize cultivation in the catchment and nation-wide terbuthylazine application statistics (average of 341 g/ha in the 3rd week of May) were used for a definition of the pesticide input function of the model. The hydrological model was manually calibrated to fit baseflow and spring/summer events. Substance fluxes were calculated using 1000 Monte-Carlo simulations of physical and chemical substance properties as provided in the literature: surface soil half-lives of 10-35 d, Freundlich KOC of 150-330 ml/g, Freundlich n of 0.9 – 1 and adsorption/desorption kinetics of 20 – 80 1/d. A multi-criteria Nash-Sutcliffe efficiency including substance loads and concentrations at all stations was calculated resulting in values up to 0.80. The best 100 parameter sets were evaluated for terbuthylazine pathways and balances. The model simulated overlaid flow to be the major source (80-95%) of terbuthylazine in the main channel and surface water fluxes to be the most important pathways in the tributaries. Simulation results suggest that 0.07-0.14 % of applied terbuthylazine mass was exported to the river in the Wark catchment. In addition to calibration of substance characteristics, passive
MO129  
Recalculation and cross-validation of pesticide trapping efficiency equations for vegetative filter strips (VFS) using additional experimental data  
S. Sabbagh, F. Galimberti, G. Azimonti, C. Kley, S. Sittig  
The FOCUS PAT and CRD PAT rules, and assessing the role of soil map, the tool can interact with ESRI ArcGIS. The openness of VandA makes it a tool suitable to work with other environmental compartments or other datasets. The consolidation of experimental data (n = 244) was used to recalculate the Sabbagh and Chen equations. Moreover, a k-fold 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. Azimonti, ICPS  
Vegetative filter strips (VFS) are a tool suitable to work with other environmental compartments or other datasets. The consolidation of experimental data (n = 244) was used to recalculate the Sabbagh and Chen equations. Moreover, a k-fold 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.

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. Camalli, G. Hughes, Cambridge Environmental Assessments; J.A. Hingston, J. Evans, Chemicals Regulation Division  
sample data was helpful for verifying model setup of application field connectivity.

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 date on a window defined by the user by applying a set of rules to the daily rainfall data used in the simulation. 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 nuance 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 dates for 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. Camalli, Cambridge Environmental Assessments; F. Ercher, CEA  
For these experiments, the VFSMOD (Muñoz-Garcia G. Azimonti, ICPS Inter-Compounds) approach. Laboratory calibration was done in triplicates under a connectivity. The uncertainty of the tropical weather in the French Caribbean makes it possible to draw conclusions for the calibration of the molecule chlordecone: the classical POCIS (Polar Organic Chemical Integrative Sampler) (with Polysulphone membranes), the POCISn30µm (with nylon membranes), and the POCSIn 0.1µm. Calculated sampling rates (Rs) were corrected by a PRC (Performance Reference Conditions) approach. Laboratory calibration was done in triplicates under a continuous flow system, and the field calibration was done in triplicates in river Casteperation (Guadeloupe, French Caribbean). Rs in laboratory calibration were 0.30±0.02 L.day⁻¹ for the POCSi, 0.97±0.01 L.day⁻¹ for the POCSIn 0.1µm and 1.54±1.38 L.day⁻¹ for the POCSIn 30µm. Two distinct Rs have been calculated for the POCS and the POCSIn 0.1µm: one for the first five days of the experiment (Rs = 0.91±0.01 L.day⁻¹ for POCSi; Rs = 0.48±0.50 L.day⁻¹ for POCSIn 0.1µm) and one for the overall experiment (Rs = 0.19±0.02 L.day⁻¹ for POCSi; Rs = 0.43±0.01 L.day⁻¹). POCSIn 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 µg−1). POCIS and POCISny 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; at these sites were collected from April, May, July-August, and September-October or 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 thifluazonamide as fungicides were mainly detected in rice season. While other fungicides including dimiconazole, propiconazole, fenamamide, naurmilur 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, cabsafos, diazinon, fenitrothion, fenhexamid and prothiofos, two carbamates, carbofuran and fenobucarb; and endosulfan were detected with low frequencies and low residue levels. Surface water were atrazine 0.9 to 22 ng L−1 and from monitored, nine pesticides which include alachlor, butachlor, dimethametryn, dithiophos, ethalfluralin, metolachlor, oxadiazon, simetryn and thiobencarb were detected with frequencies of 1-48% and in their residue level of 0.01-1.9 µg/L. Detection frequencies and residue levels of insecticides and herbicides were the highest in waters sampled in May and June. Almost 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 74.1 ng L−1, respectively. Almost 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 groundwaters) between October/2015 to October/2016. The most frequently detected pesticides in surface water were atrazine-2-hydroxy (100%), diuron (94%), carbendazim (93%), tebuconazole (91%), metolachlor (91%), imidazol (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 imdicloprid, carbendazim, atrazine and malathion. For the groundwater the most frequently detected pesticides were atrazine-2-hydroxy (24%), imidaclopr (14%), carbendazim (10%), tebuthiuron (10%), atrazine (10%) and diuron (10%). The pesticide that presented the highest concentration for this matrix was tebuthiuron, 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 JUNIOR, Embrapa / EMBRAPA AGROPECUARIA OESTE; D. Máximo, R. Rebelo, IBAMA / DJIUA / CGASQ; A.V. Waichman, Universidade Federal do Amazonas; N. Peranginang, Syngenta Crop Protection, LLC / Product Safety; A. Toniolo, Bayer AG / GENCS - E-Fate; L. Murakami, Bayer AG Crop Science Division; O. Perez-Ovilia, Bayer CropScience / Environmental Safety; E. 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. Surface water scenarios should be identified in six pre-defined climatic zones for the seven most important crops soybean, maize, sugar cane, wheat, cotton and coffee. Runoff and spray drift were found to be the main entry 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 far 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. Rathiens, M.F. Winchell, Stone Environmental, Inc. / Environmental Systems Management/ Sur, Bayer AG Crop Science Division; E. Henry, Bayer AG Crop Science Division; D. 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 disposal of the herbicides detected 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 demonstrates 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 that incorporated model results to 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 M.F. Rocha, ICBCAS U.Porto, CIMAR CMAR LA; C. Cruzeiro, CIMAR CMAR LA; Porto, CEF FCTUC U. Coimbra; S. Amaral, ICBCAS U.Porto; E. Rocha, ICBCAS U.Porto, CIMAR CMAR 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
extraction and quantitatively analysed by gas chromatography–mass spectrometry (GC-MS). Results show that 96% of measured pesticides were detected in 79% of the quantified samples and that twelve compounds showed concentrations well above the limits established by the 2013/39/EU Directive. Individually, the concentrations of the analysed pesticides ranged from 39 to 1265 ng/L. Since the occurrence of these compounds happens in mixtures, we conducted a theoretical hazard assessment considering the average and the maximum environmental mixtures of all measured pesticides. The theoretical approach suggested that invertebrates were the most sensitive group. Therefore, short-time exposure in vivo assays using Artemia salina and Daphnia magna were done. These results disclosed significant toxic effects of the analysed mixtures - high mortality rate and abnormal swimming behaviour - over the exposed animals. Both approaches (theoretical and experimental) demonstrated the need to set up specific recommendations on this estuarine environment and of other comparable.

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Keywords: monitoring, Artemia salina, Daphnia magna, pesticide mixtures

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

G.L. Reeves, Dow AgroSciences Ltd; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences; R. Verro, University Milano - Bicocca - Lybra ambiente e territorio S.r.l. / Department of Earth and Environmental Sciences; C. Vai, Dow AgroSciences Italia s.r.l.; R. Bradascio, Dow AgroSciences Italia s.r.l.

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 (X11292885; 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 integrated soil–water–environment (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 (X11292885) were below the LOD (0.001-0.002 µg/L) in 94% of the samples. In addition, 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 X11292885 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 fungicid 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. Vai, Dow AgroSciences Italia s.r.l.; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences; 1,3-Dichloropropene (1,3-D), also known as Telone™, 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 intersected 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 validated against national rules. To facilitate this, a GIS tool was developed allowing to refine the data. This model, when combined with sales data, it was possible to refine the areas previously identified and quantify the percentage of areas potentially at risk of leaching where Telone™ is applied.™ of DowAgroSciences

MO141

Development of an European Tier 3+ Spatially Distributed Modelling Framework

G. Hoogeweg, Waterborne Environmental, Inc. / Data Technologies; P. Sweeney, Syngenta

Higher tier groundwater assessment in the European Union (EU28) allow the use of spatially distributed modeling approaches for the assessment of groundwater and exposure of soil organisms. To identify the areas where the percentage of soil falls among three different categories (< 60%, 60–80%, > 80%) Also these data were structured as GIS layers, which were processed and represented using the same GIS of the crop distribution. Overlaying the crop distribution and sandy soil areas and by merging the two databases, it was possible to identify sub-communal areas where crops and sandy soils coexist, characterizing the extension in relation to the rest of the municipality and the province. Finally, by considering 1,3-D sales data, it was possible to refine the areas previously identified and quantify the percentage of areas potentially at risk of leaching where Telone™ is applied.™ of DowAgroSciences

MO142

Influence of aquifer parameters on groundwater residue concentrations

F. Hegler, DR. KNOELL CONSULT GmbH; D. Liss, SGS Institut Fresenius GmbH / Agro; W. He, DR. KNOELL CONSULT GmbH; O. Naeh, SGS Institut Fresenius GmbH; S. Redlich, DR. KNOELL CONSULT GmbH / Environmental Fate / Modelling / GIS

FOCUS modelling is used in a regulatory context to calculate pesticide leaching flux concentrations in 1 m depth (PECgw; Predicted Environmental Concentrations in groundwater) from the unsaturated to the saturated zone. These values are used in risk assessments in order to evaluate the impact of plant protection products on groundwater. In higher tier groundwater monitoring studies the properties of the saturated zone add additional complexity influencing actual pesticide residue concentrations in shallow groundwater. In this work the impact of groundwater flow velocity and aquifer porosity on groundwater residues for a defined leachate concentration (i.e. decoupled from the unsaturated zone) was determined. In a sensitivity study a range of aquifer parameters the impact on the resulting residue concentrations in groundwater was quantified. For the sensitivity analysis FOCUS model outputs for selected scenarios were combined with realistic aquifer parameters for some representative regions in northern Italy and Germany. In these regions the relevant shallow aquifers are variable in terms of hydraulic conductivity, gradient and effective porosity and provide a representative parameter range.

MO143

Implications of Dataset Selection and GIS Processing on Modelling

G. Hoogeweg, Waterborne Environmental, Inc. / Data Technologies; M. Geuvara, Waterborne Environmental Inc / Modeling

Groundwater assessment guidelines provided by the FOCUS groundwater working group (2009) and EFSA (2014) describe succinctly a multi-tiered modelling framework that includes spatio-temporal assessments in the higher tiers; e.g. tier 3a and 3b. As part of the spatio-temporal assessment several GIS and daily climate
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 reflector 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 impact 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 tools 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 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 activates substances, more metabolites will be included in the future Proposals to combine targeted together with public groundwater monitoring dataset 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. Uluccu, 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 risk assessment 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 PECgw values are expected when high variabilities occur 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 consider 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 can 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. Gimsvang, The Danish Environmental Protection Agency / Pesticides and Gentechology; W. Koenig, UBA Umweltbundesamt; A. Boivin, ANSES; A. Poot, Cgb; A. Schwenn, 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 of the data is available in the national language of the origin country only, which makes it hard for other countries to use this data and (iii) the interpretation of 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 this 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. Soudais, Bayer Crop Science / Environmental Safety

Recent regulatory interest in the wash-off process resulted in a proposal to that the effects of wash-off should be generally 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 hand, EFSA has stated that effects of wash-off should be not considered, in addition to the above average effect (EFSA 2015, 2017). The pull-off factor of a compound is a product formulae 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 Scientific Opinion 2562 - Outdoor conditions. EFSA 2012: Scientific Opinion 2562 – Science behind the guidance on soil scenarios EFSA 2015: Guidance Document 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. Köhler, Bayer AG, Research & Development, Crop Science / Environmental Safety; M. Lamsoe, 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. One 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 substances (e.g. pharmaceuticals, pesticides, pyrolysis of PTFE (used as coating in many products like Teflon® or GORE-TEX®)). Plant root uptake of TFA under cropped outdoor conditions was investigated as a combined study in a soil column at a depth of 1 to 1 cm. A wash-off factor for FOCUS modelling of 1 cm-1 was chosen. The approach was validated by application of 7 different substances with different wash-off factors. 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 Scientific Opinion 2562 - Outdoor conditions. EFSA 2012: Scientific Opinion 2562 – Science behind the guidance on soil scenarios EFSA 2015: Guidance Document 4093 - Predicting environmental concentrations in soil EFSA 2017: Guidance Document 4982 - Predicting environmental concentrations in soil
under dynamic outdoor conditions; the hydroponic study design is suitable 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.

MOI49 Investigating the variance of edge-of-field deposits of spray drift
H. Holtermann, Wageningen University & Research / Agrosystems Research; J. Michielsen, H. Stallings, P. Van Velde, 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 carried out 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. Spray 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 treated field at 2 m downwind. Consequently, the part of spray deposits that were recorded during the experiments. Horizontal and vertical movements of the sprayer boom 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 result of 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.

MOI50 Exposure assessment for edge-of-field watercourses next to tree nurseries regarding spray drift deposits
H. Holtermann, Wageningen University & Research / Agrosystems Research; J. Van de Zande, Wageningen University and Research / Agrosystems Research

Pesticide applications 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 can be reached by the drift. From the above mentioned experiments, the part of spray drift that may pass under the tree canopy is unknown. 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 edge-of-field watercourses in the Netherlands. The experiments with 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.

MOI51 Investigating the exposure of residents to pesticides due to airborne spray drift
H. Holtermann, Wageningen University & Research / Agrosystems Research; J. Van de Zande, J. Michielsen, H. Stallings, P. Van Velde, Wageningen University and Research / Agrosystems Research

In the Netherlands about 9,000,000 people 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 bulb 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 measurements of airborne pesticides are performed by Scotch tape and inhalation exposure chambers. Simulations deals with the exposures to spray drift only. After application using a conventional boom sprayer ground deposits and airborne distributions of spray drift are measured down to 50 m from the treated area. Airborne spray drift is measured up to 10 m height, using two different sampling techniques. At 50 m downwind, airborne spray drift appears to be up to 100 times higher than ground deposits. Simulations of spray drift are studied using the IDEFICS spray drift model for boom sprayers. The results of simulations result in downwind ground deposits and airborne spray drift with values in the same order of magnitude as those found in the experiments. The results indicate that potential exposure of residents to pesticides used when treating nearby fields may be significant and further assessment of this exposure route is important.

P. Adrian, M. Liegeois, M. Durrieu, B. Jouanol, CEHTRA SAS

Research indicates there is no recent guidance on how to conduct a risk assessment for consumers for co-formulants present in plant protection products. One of the reasons is the lack of exposure data when the product containing its co-formulants is applied onto crops. To our knowledge only one software is predicting the level of crop residues of chemicals after application i.e. PARDIS [1] (Prediction of Agricultural Residue Data Set) for fruit using an Informatic System) 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-formulant. 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), Colliera M, Balderacchi M, Capri E, Trevisan M. 2008.

MOI53 Dietary exposure to pesticide residues: the big picture
M. Mar Bouza, L. Ferreira, 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 pesticide residues. Maximum residue levels (MRLs) in the European Union are derived from 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 exposure 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. This provides the key information to be interpreted by risk assessors and for risk managers’ consideration. Essential input values in risk assessment are toxicological data and residue values subject to many environmental scenarios and considerations that are used to define and characterize the residues to which consumers are exposed and through the diet, complex metabolic pathways in plants and animals, degradation of the compounds in soils and its transformation, the possible uptake and translocations of the residue to the edible parts of the crops and degraded products as result of industrial and household processing are considered to set the residue definitions for risk assessment purposes. Secondary metabolites characterized by metabolism studies and degraded products may pose a completely different toxicological profile than the parent compound, being more, less or equal toxic than the pesticide under assessment, and showing a big new picture for an active substance and its residues that should be addressed in detail to avoid consumers’ concerns. The dietary risk assessment of pesticide residues takes into consideration these possible scenarios in order to protect consumers, reason why residue definitions might be different for monitoring and for risk assessment purposes and where the uncertainty due to missing data might play a fundamental role in risk assessment.

MOI54 Exposure and Risk Assessment for Agricultural Applicator to Insecticide Fluembidiamide during Cabbage Cultivation using Whole Body Dosimetry
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Fluembidiamide belongs to diamide insecticide that has been used to control a wide range of insects in fruit and vegetables. Assessment for agricultural applicator’s protection is important as it may cause accidental exposure in the field. The applicator’s repeated dosimetry experiment under dynamic outdoor conditions in the field was carried out. For dermal exposure measurement, whole body dosimetry (WBD) was performed, which consists of cotton/polyester outer clothes and cotton inner clothes. Hand exposure was measured by washing of nitrite gloves and hands, while head exposure was monitored by face/neck wipe technique. Inhalation exposure was evaluated with personal air sampling pumps and IOM sampler (glass fiber filter). Analytical limit of quantitation was 5.0 mg/ml with good linearity (R² > 0.99) of calibration curve. Recovery (77-117%) of insecticide from various exposure matrices were reasonable including field recovery (77-109%). Field exposure measurements were carried out by 8 replicates. During application, total
dermal exposure of flubeniamide was 3635.7 μg, while that of mixing/loading case was 815.3 μg. Hand exposure of flubeniamide (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 applicator during treatment of flubeniamide for cabbage field would be minimum. Keywords: Flubeniamide, exposure, risk assessment, whole body dosimetry, IOM, cabbage *Corresponding author: kjh2404@su.ac.kr; Tel, 82-02-880-4644

MOI55 Multi-year Focus Surface Water Calculations: What do they mean for real regulatory cases?
D. Schaefler, Bayer Crop Science / Environmental Safety, G. Reinken, Bayer AG, Research & Development, Crop Science / Environmental Safety, A. Boledhan, 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 capture the effect of variable weather conditions in 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 FOCUSsw 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 FOCUS Surface Water calculations. Surface water exposure is strongly driven by individual weather events triggering runoff 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 procedures is still missing. In this work we conducted such an investigation by running multi-year 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 experiences 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. Pasini, Agrea SRL, R. Bondi, Domini 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 too low to ensure the desired removal rates specific to Spinosad may be useful in demonstrating compliance. A GLP field study was performed in summer 2017 to test vegetated buffer strip removal efficiency, in relation to Spinosad and its major metabolites, and based to the FOCUS Surface Water risk assessment scenarios, but with worse (and prudential) conditions. The selected site was near Verona, in a hilly zone rich in vineyards and famous for high-quality wine production. Runoff containing a known amount of each of the four spinosad components to be tested (the parents – spinosin A and spinosin D; and the metabolites – spinosyn B and spinosyn B of D), have been artificially applied to 9 grass plots of 4.2 m width and 12 m length, with sandy-loam soil and slope ranging from 10 to 13%. Natural vegetation cover was 60-90%. The artificial runoff was organized to simulate a rainfall generated in a source area of 500 m² flowing in a run-on area (buffer area) of 50 m², to evaluate its buffer capacity both in runoff displacement and concentration. Runoff event consisted in 2 phases: 1) Irrigation with sprinkler at 14 mm/h for 50 min (total: 12 mm), to simulate rainfall before runoff; 2) Run-on/Runoff event: 200 mm of water were released into the buffer area in 2:20 hours using a runner generator (flow of 85 mm/h). Water concentration precprecipitation (PCP) of 4 spinosyns decreased into buffer area, the “Run-on” becomes “Run-off”, and Runoff water was sampled at 0.75, 1.5 and 2.20 hours after Run-on start. During Run-on, irrigation continued until the end of run-on (other 33 mm), and a total of 45 mm were applied to buffer area. Given the frequencies of selected rainfall (low, return period of 2 years), the runoff/rainfall rate (high, 45%), the source to buffer area proportion (high, 10 to 1), and the plot slope (from 10% to 13%), conditions of the experiment can be considered highly precautionary, and more prudential that those of Focus R4. First results show that the runoff displacement ranges from 3 to 11 m from Runoff releasing. Analysis of spinosyns concentration are in progress.

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. The relevant residue guidance documents of the European Commission, EFSA prepared a guidance1 on the residue definition for dietary risk assessment which intends to complement the OECD guidance2. 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 ARID 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 napropamide. In September 2016, EFSA organised a technical meeting1 with stakeholders on its new guidance to exchange views. 1EFSA 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/it/events/event/160926

Alternative Approaches to Animal Testing for Ecotoxicity Assessments (P)
MOI158 Investigations on the bioconcentration of xenobiotics in the freshwater amphipod Hyalella azteca
C. Schaeblenheim, 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; O. Bischof, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; L. Bischof, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; IME Bioconcentration factors (BCF) are needed for regulatory purposes to assess the toxicological significance of substances and their amounts likely to be present in the aquatic environment. Traditionally these BCFs are determined in fish, and with the exposure being almost 20 compounds showed, that the H. azteca bioconcentration test could be an appropriate test to predict bioconcentration in the standard fish test. Bioconcentration factors 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 effect of metabolism on the resulting BCF. In this study 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 H. azteca 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.

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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 (Periidae). This study investigated in vitro inhibition of aromatase by the model inhibitor, fadrozole, fadrozole, across eighteen phylogenetically diverse species of freshwater fish. Concentrations of fadrozole that result in 50 % inhibition of in vitro aromatase activity (IC50) were determined from 0.004 to 0.008 µM among these species. The environmental relevance of IC50 values is highlighted when compared with those for fadrozole-induced 50 % inhibition of aromatase among fathead minnows. Potencies measured in vivo are likely to be higher than those found in vitro for any given concentration due to the presence of cytochrome P450 enzymes in vivo that can metabolize fadrozole to more potent metabolites. Differences in sensitivity among species may be a result of intrinsic differences in sensitivity to aromatase inhibition and/or of variations in the ability of fish species to metabolize fadrozole.

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 monitoring purposes because it accounts, in principle, for the environmental systems that control 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 regulatory framework privileges measurements in the dissolved (filterable) 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, an alternative outcome pathway for monitoring the concentration of trace elements in aquatic organisms is offered by the quantification of trace element concentrations in fish scales. Fish scales are often formed in a highly ordered way, and the rate and degree of scale formation are determined by the environmental conditions prevailing during the scale formation process. Fish scales can therefore yield comprehensive data for relative sensitivity assessment for impact of the individual and population level. This information could guide more objective ecological risk assessments. Data on trace element concentrations in fish scales could also be used to assess the quality of fish from an environmental point of view or as an indicator of recent changes in pollution levels. The aim of this study was to determine the content of trace elements in fish scales and evaluate the potential of fish scales for monitoring trace elements in fish.
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, flavors, 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 determine the potential acute and chronic health hazards associated with these aerosols 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 emissions 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 in e-cigarettes. The acute pharmaceuticals had the C/M between 0.01 and Development, AMED. 

ECOSAR and KATE models for acute toxicity only in both species, Acute LD50 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. The model for predicting toxicity in daphnia was built using QARINS and QSAR models with R2 values and QLOO-2 values ranging from 0.7 to 0.9 and Q2 values ranging from 0.7 to 0.8. These models were used to perform a screening of the acute toxicological profile of the 265 molecules of interest and to compile a priority list of substances of potential toxicological concern. This preliminary study represents a first step toward the hazard assessment of e-liquids and for the identification of safer alternatives to existing and potentially harmful ingredients screened from the chemical structure. However, additional work still needs to be done to make these products safe for human use.

MO164 Projection of QSAR models for daphnia and fish chronic toxicities of human pharmaceuticals

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Recently, medical regulatory agencies require pharmaceutical companies to assess environmental impacts of new pharmaceutical products before marketing. Hence, it would be valuable to predict ecotoxicity of new pharmaceuticals at developmental stages. As a tool for precision toxicity in daphnia, two QSAR models with ECOSAR by USEPA and KATE by Ministry of Environment in Japan are available, both of which are built using dataset of mainly industrial chemicals. In this study, we evaluated applicability and predictivity of the QSAR models using external dataset of the chronic ecotoxicity of human pharmaceuticals. The chemical structures and toxicity data based on D. magna reproduction test (OECD TG211) and fish early-life stage toxicity test (OECD TG210) were gathered from public domain. In order to examine the applicable domain where more reliable prediction results can be obtained, the following criteria were defined in this study; (1) logP values of target substances are within the lowest and highest values of the category chemicals, (2) number of category members is 5 or more, and (3) correlation coefficients of the linear regressions are greater than 0.70. Since KATE equips models for acute toxicity only in both species, Acute-Chronic Ratio of 10 was applied to estimate NOEC values. Then, ratio of calculated NOEC and measured NOEC (C/M) was determined. For ECOSAR daphnia model, 82 out of 126 pharmaceuticals satisfied the criteria. Of these, 44 pharmaceuticals had C/M between 0.1 and 10, some of which were assigned to amides or aliphatic amines. 72 pharmaceuticals satisfied the criteria with C/M between 0.00 and 0.12. 12 pharmaceuticals had C/M values greater than 100, half of which have pharmaceutical action to neurotransmitter receptors in human. For KATE daphnia model, 19 pharmaceuticals met the criteria. The C/M values were between 0.1 to 10 for 15 substances, most of which belong to primary amines aliphatic/aromatic, amides or imides, or neutral organics. For fish chronic toxicity, only 11 and 21 out of 72 pharmaceuticals satisfied the criteria with ECOSAR and KATE models respectively. Further examination will be needed to expand applicability by modifying the criteria, combined with other approaches including acute-to-chronic extrapolation or daphnia-fish interspecies extrapolation.

This work was supported by the Research on Regulatory Science of Pharmaceuticals and Medical Devices from Japan Agency for Medical Research and Development, AMED.

MO165 Optimization and Accessibility of the Eco-Database and the Ecotoxicological Threshold of Concern (ecoTTC) tool

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The Ecological Threshold for Toxicological Concern, or ecoTTC, has been proposed as a natural next step to the well-known human safety TTC concept. The ecoTTC is particularly suited for use as an early screening tool in the risk assessment process, in situations where chemical hazard data is poor, or when an appropriate QSAR is unavailable. EcoTTCs are developed using statistical distributions of Predicted No-Observed Effect Concentrations (PNECs) to reflect the breadth and depth of the ecotoxicological dataset beneath, and therefore, the potential toxicological concern. This preliminary study represents a first step toward the hazard assessment of e-liquids and for the identification of safer alternatives to existing and potentially harmful ingredients screened from the chemical structure. However, additional work still needs to be done to make these products safe for human use.

MO166 Using toxicokinetic and toxicodynamic modelling to predict effects of chronic toxicity on rodent growth based on in vitro assays

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According to 2011 figures, 80% of the animals used for testing procedures in the European Union are rodents and almost 23% are used in longer term repeated dose tests. Alternative methods to predict the effects of chronic toxicity in rodents can therefore make a significant contribution to the reduction, replacement and refinement (3Rs) of animal testing. Body weight is one of the many endpoints monitored throughout chronic toxicity tests. We aim to develop in silico models to extrapolate the effect of toxicant exposure, measured as an appropriate internal dose metric, on the growth of rodents. Initially, we will develop separate toxicokinetic-toxicodynamic models will be used to predict the selected internal dose metric and its effect on growth over the duration of repeated dose toxicity studies. These models will be developed using data from regulatory toxicity testing of pesticides. Experiments will then be designed to assess the effects of known intracellular pesticide concentrations on cell population growth in vitro. Cell number can be converted to cell mass, after which it should be possible to model the effects of matching internal doses on growth over time, in vitro and in vivo. The weight normalised effect on growth (mass_dose group / mass_conc a at a given time point) can then be calculated at various points along the predicted in vitro growth curves. These predictions can then be compared to corresponding in vivo chronic toxicity studies. The predictions from this extrapolation will be explored for 10 pesticides, which will provide a good indication of the reliability and repeatability of the methods. Should predictions prove to be consistently accurate, this will provide a fast and inexpensive in vitro screen for body weight effects in rodents. Initially this may be applied as an alternative to range finding studies which are not a regulatory requirement but are commonly carried out prior to regulatory testing. In the longer term this may form part of a suite of in vitro and in silico alternatives to in vivo chronic toxicity testing.

MO167 Screening of metabolic- and neurotoxicity of environmental chemicals using C. elegans and transgenic zebrafish models

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Metabolic and neurodevelopmental disease have been attracting attention as
environmental disease. 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, we used a chemically screening using C.elegans mutants; oga-1(ok1207), og-1(ok1474), ngl-1(ok259), transgenic zebrafish, Tg(Tx2Kins:nlsb-mCherry)48 and Tg(ela13:EGFP)knu3. The highly conserved O-GlcNAc transferase; OGT and O-GlcNAcase; OGA genes are related to type 2 diabetes and null mutations cause alterations in *C. elegans* carbohydrate and lipid metabolism. Neurologin NLG-1 control synaptic function, which is conserved from nematodes to mammals to mediate synaptic plasticity and activity-dependent plasticity (ADHD). Tg(T2Kins:nlsb-mCherry)48 fish express insulin, nitroreductase(InsNTR) mccherry fusion protein in the pancreatic β-cell and Tg(ela13:EGFP)knu3 fish express GFP in most post-mitotic neurons. Various category of environmental chemicals, such as, heavy metals (i.e. arsenic, lead, cadmium), EDcs (i.e. Nonylphenol, Bisphenol-A,EDF), and biocides (i.e. Chloropyrifos, CMIT/MIT, PGH), were screened using *C. elegans* reproduction assay and zebrafish transgenic assay. The preliminary results showed CMIT/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.

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**MO168**

**In vitro effects of two pesticides on the motility and viability of bovine spermatozoa**

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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 dermic exposure or ii) through the ingestion of contaminated feed. This exposure affects the reproduction of those animals. Actually, spermatozoa are extremely sensitive to slight variations in the organism. The interaction between chemicals and sperm may alter its mobility; velocity and/or viability depending on which cell structures are affected. This work aimed at assessing the toxicity of ecological relevant concentrations of copper sulfate and glyphosate on bovine spermatozoa. Some of the main factors that affect the motility of spermatozoa are: the concentration of the two pesticides, diluted in phosphate-buffered saline (PBS), plus a control (PBS). For each bull, three replicates were made. Motility and velocity endpoints were measured with a sperm analyzer computer program and viability was measured using an eosin-nigrosin staining procedure. Endpoints were measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on sperm motility at the lowest concentration, but for the first observation period (time=0 minutes) viability was measured at 0, 30 and 90 minutes.

**MO171**

**Local Electrophysiology Describes Experimental Glutathione Reactivity and Aquatic Toxicity toward Tetrahymena pyriformis**

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Electrophilic compounds such as α,β-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 lipids resulting in reactive toxicity 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—with 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 quantum chemistry calculations. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which reacts either by the stable Michael adduct (MIE) or by reactive electrophilic reactivity is high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict—with 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 quantum chemistry calculations. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which reacts either by the stable Michael adduct (MIE) or by reactive electrophilic reactivity is high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict—with 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 quantum chemistry calculations. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which reacts either by the stable Michael adduct (MIE) or by reactive electrophilic reactivity is high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict—with 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 quantum chemistry calculations. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which reacts either by the stable Michael adduct (MIE) or by reactive electrophilic reactivity is high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict—with 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 quantum chemistry calculations. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which reacts either by the stable Michael adduct (MIE) or by reactive electrophilic reactivity is high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict—with 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 quantum chemistry calculations. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which reacts either by the stable Michael adduct (MIE) or by reactive
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 able to metabolize therefore hydrolytic activity of esters as negligible. The di-esters appear more 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 unsaturated, like allyl/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

Nonanedodecane 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 En-V Synthesis; A. Yamaguchi, National Institute of Technology, Ariake College; M. Uchida, Ariake National College of Technology / Department of Chemical and Biological Engineering; H. 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 nonanedodecane pulsed electric field (nPEF) 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 nPEF 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: transcriptomic characterization of the RTgutGC cell line.
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Intestinal derived cell lines are useful in vitro models which allow for focused investigation of specific nutrients. The development of the first immortalized intestinal cell line derived from the rainbow trout (RTgutGC) offered an opportunity to explore intestinal uptake without the need for in vivo models. The RTgutGC cell line has high expression of the genes which are involved in the absorption of nutrients and the transport of drugs. The RTgutGC cell line is also a valuable tool for studying the effects of chemicals on the intestinal epithelial cells. The RTgutGC cell line is a powerful tool for assessing teratogenicity and embryonic developmental toxicity of chemicals and predict their molecular mechanisms in fish embryos.

MO176

Biological effects of 3 metals on D'larvae of Japanese oyster Crassostrea gigas
A. Sobrino-Figueras, Universidad Autonoma Metropolitana Iztapalapa / Hydrobiology; C. de Caceres-Martinez, Universidad Autonoma de Baja California Su

Toxicity of metals according to the calculated LC50 values was: (from most to least toxic) Cd > Pb > Cr. The most toxic metal in this study was Cd. The Kruskal-Wallis test indicated that there are significant differences in the degree of lipoperoxidation, inhibition of AchE activity and genetic damage between the exposed organisms and the control group. The metal with the highest oxidative stress was Chromium (32 ± 8.97 nM Tbars mg^-1). And the metal with the highest oxidative stress was Lead (45 ± 11.89 nM 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 AchE (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
A. Sobrino-Figueras, Universidad Autonoma Metropolitana Iztapalapa / Hydrobiology; A. Perez-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Laboratory of Aquatic Ecology and Geochemistry

The Japanese oyster is an introduced species from Asia which has 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, for assessment of four biomarkers: growth rate, O:N ratio, lipid peroxidation and inhibition of acetylcholinesterase enzyme). The LC50 values were 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 AchE enzyme (Eliam 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 most toxic metal in this study was Pb. The LC50 values determined with 1 µM. Measured LC50 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 fragments (e.g. 2.21 and 5.5 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 compounds was analysed by GC-MS or LC-MS and kap values determined. For the lowest concentration (0.2 µM) 2-fold higher kap values were observed for Polysantol, Ambrofix, Cyclohexyl salicylate and Karanal compared to kap values determined with 1 µM. Measured kap values were 2-fold lower with 5 µM except for a 4-fold lower rate for Polysantol compared to 1 µM test concentration. For Pyrene and 7-Hydroxycoumarin, the use of 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.
to other species. The O:N index had values below 9 fact indicates that organisms were in a high degree of stress. Growth rates of intoxicated organisms were between 19 to 49% lower than those observed in the control group. The average concentrations of Tbars registered organisms varied from 2.5 to 25.6 Tbars mg⁻¹ and show a direct dose-response relationship, since when increasing the time of exposure to DDVP increased the degree of lipid peroxidation in the tissues. A decrease in AChE activity was observed in the tissues between 22 and 45% and fish from 22 to 35%. The results of this study indicate that the effects of the pesticide DDVP are likely irreversible in some species.

MOI178 Characterising estrogenic activity of Arctic char tissue extracts in two fish in vitreous testasays

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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 effect level for egg mortality in temperate salmonid fish raise concern that 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 least contaminated Lake Laksvatn at Bear Island. This was done by in situ sampling, extraction, and fractionation of liver tissues. The following metabolites (PMCs) 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 developed and used 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)-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 fraction from Lake Ellasjøen, 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 preformed to identify potential contributors to the observed effects (knowledge). The project was funded by the Norwegian Research Council, project. No. 221373.

MOI179 Ultrasound: A novel approach to non-lethally measure hepatoaomatic index in sentinel fish for environmental monitoring programs

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Canada’s environmental effects monitoring (EEM) program studies impacts of metal mining and pulp mill effluents on aquatic receiving environments. The EEM regulations recommend lethal sampling of 20 fish (male and female) of different species to study body condition, liver size (hepatoaomatic index-HSI), and gonad size (gonadosomatic index-GSI) during every monitoring cycle. Developing and implementing non-lethal methods for environmental monitoring programs provides an opportunity to protect sentinel endangered fish which might be threatened by research activities. We tested a non-lethal method in two Arctic char populations (Salvelinus alpinus) that are part of systems with low productivity. Ultrasound is a non-invasive tool that has been tested to assess gonad size in fish. Currently, its potential as a non-lethal tool in environmental monitoring programs is not well explored. We conducted feed withdrawal studies in the laboratory to test the accuracy and sensitivity of ultrasound to measure HSI in sentinel fish with a compact liver such as rainbow trout (Oncorhynchus mykiss). With the ultimate goal of providing empirical evidence of the applicability and ease of this technique in the field, we also tested the accuracy of ultrasound method to measure HSI in lake trout (Salvelinus namaycush) at ISDS-experimental lakes area. Our laboratory studies provide a significant correlation for the accuracy (HSI, r²=0.73, n=16, p<0.05) and evidence for the sensitivity of ultrasound method (p=0.06, n=7) versus traditional lethal gravimetric method (p<0.05, n=7) to measure HSI within the acceptable critical effect size for HSI mandated by EEM. Our field ultrasound method testing also revealed a significant correlation between the traditional lethal and ultrasound method in measuring HSI (r²=0.81, n=9, p<0.05) in lake trout. Our field analyses 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 diffuse 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.

MOI180 Weight of evidence for fish acute toxicity: a Bayesian network modelling approach

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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 environmental and agricultural risk assessment. The primary goal of the proposed BN model is to integrate information from large and varied ecotoxicological and physico-chemical datasets, and apply it in a WoE approach to predict fish acute toxicity of chemicals from data on fish embryo toxicity testing in combination with other relevant information. The planned steps of the model development and application are: (1) Identification of suitable chemical properties and endpoints as nodes for the BN. (2) Compilation of data on ecotoxicity data for fish embryos and other relevant information. (3) Construction of a BN model for integrating the data and other information in a probabilistic framework. (4) Evaluation of the WoE approach using ecotoxicity data for fish embryos in combination with data on (juvenile) fish acute toxicity. (5) Application of the BN model to assess the risk of various contaminants based on chemicals registered in the European Chemicals Agency database (http://echa.europa.eu).

MOI181 Divergent immunomodulatory effects of cadmium between two marine immune cell models in vitro, macrophages and mast cells.

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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 of non-destructive methods for the detection of biomarkers in 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 extent of potential non-destructive biomarkers in whole organisms, 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 Non-animal Tool for Mimicking Phase I Metabolism
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The electrophilicity of a chemical substance can have a significant impact on its toxicological profile, as this property determines the chemicals' ability to form adducts with electron rich sites of proteins, lipid components, DNA and RNA. In many cases, this type of adduction 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 excess toxicity or skin sensitization potential. 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-electrophilic phenol and diaryldihydrobenzene derivatives into reactive electrophiles that can be 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 electrophiles, and provides new insights into the oxidation pathways causing the reactive toxicity of pro-electrophiles. The authors will introduce the EU-funded project OXYBASE (OCE-CT-2007-037017) and the BMBF-funded project PropHalTox (FKZ 031A422A and 031A422B) for financial support.

MO185
Integrated assessment of aquatic ecotoxicity for regulatory purposes
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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 ecological part of the T property, we developed six continuous QSAR models for acute and chronic aquatic endpoints for the main trophic levels: EC_{50} 96h and NOEC fish (Raphaelidocellus subcapitatus), EC_{50} 48h and NOEC 21d Daphnia magna, LC_{50} 96h fish (Oryzias latipes) and NOEC fish (more fish species). We used gaselect and VSURF to select the DRAGON descriptors and the tree ensemble (random forest) method to derive the models, obtaining good performance (R^{2} 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 interspecies variability, the most sensitive species and the applicability domain index of the predictions. The users can choose a consensus or a weighted approach. The experimental values and the predictions are submitted to 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 fulfill 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 6 962) and LIFE COMBASE (LIFE15 ENV/ES/416) for the financial support.

MO186
An integrated testing strategy to fill data gaps for environmental risk assessment of isoa-alkohols
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Environmental risk assessments require quality data to provide defensible environmental quality benchmarks. Quantitative Structure Activity Relationship (QSAR) endpoint estimates are often appropriate for alcohols with a very strong chemical reactivity, which is to be expected for electrophilic alcohols. The authors present a risk assessment on C6-8 isoa-alkohols that involves the following steps: (i) development of a computational method to predict mutagenic effects (ii) comparison of results from the computational method to experimental mutagenicity data from Salmonella typhimurium strains. The computational method was developed using in silico screening protocols for mutagenic effects and compared to the experimental mutagenicity data obtained from Salmonella typhimurium strains. The results of the computational method are compared to the experimental mutagenicity data obtained from Salmonella typhimurium strains, and the results are discussed. The authors thank the projects JANUS (contract Z 6 - 80 710/20 - 3716 65 410) and the BMBF-funded project PropHalTox (FKZ 031A422A and 031A422B) for financial support.
Aquatic toxicity testing with algae, daphnids and fathead minnows (OECD 201, ISO 20665 and OECD 210) was performed with isoocotol and isoundecanol. The study objective was to employ a testing program consisting of long-term fish (limit test), invertebrate and algal toxicity tests to demonstrate that QSAR estimations accurately predict aquatic effects from long-term continuous exposure to these substances, further supporting the use of QSAR models across a range of isolated models. The data demonstrate that the QSAR model employed accurately characterized the hazard of iso-alcohols and is protective of these endpoints. 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

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Glyphosate-based herbicides are widely used in agriculture. When these products were originally introduced on the market, they were needed 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. Nonanoic acid (a.k.a. pelargonic acid) is a biologically derived substance considered as an environmental 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 estimated in EthoVision 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 knowledge so far there are no available data for a neurotoxic effect of pelargonic acid on aquatic organism. Hence, this outcome has to be further investigated. Financial support from the City of Vienna project Ökotoxikologie (MA 23 - Project 15-06) is gratefully acknowledged.

MO190
Chemosensor Profiling of Salicylates to Assess Their Reactive Toxicity

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Salicylates are widely used as fragrance additives or UV light absorbers in cosmetics and consumer care products, and thus can contribute to the human exposure. Moreover, pelargonic acid, hospital waste and wastewater linked to economic activities and rainfall 2) Most WWTP effluent still demonstrate that QSAR estimations (a) minor part of the thyroid effect removal occurs during and contains thyroid active molecules, results from chemical analysis performed on the wastewater, hospital wastewater, water from industrial processes. A part of our study objective was to employ a testing program consisting of long-term continuous exposure to these substances, further justifying read-across for both acute effects of these substances. For this communication, the chemoassay effect of all compounds present into a global hormonal potential and are therefore continuously assessed in the performance 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 defined 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 procedure. 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 assayed to eco-toxicology and rainwater originating from rainfall and stock still contains thyroid active molecules, results from chemical analysis performed on the same samples indicated a correlation between the total micropollutant load, and the thyroid effect 3) A minor part of the thyroid effect removal occurs during and after WWTP processes. The major removal of the thyroid active molecule occurs during the nitration step of the water treatment.

MO191
The Xenopus Embryonic Thyroid Signalling Assay (XETA) for assessment of effluents contamination in thyroid active molecules

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The Xenopus Embryonic Thyroid Signalling Assay (XETA) was designed as a screening tool 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 12 past years we applied this assay to effluents including municipal wastewater, treated wastewater, industrial wastewater, hospital wastewater and rainwater. A part of our studies focused on performances of wastewater treatment plant (WWTP). Assaying 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 defined 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 procedure. 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 assayed to eco-toxicology and rainwater originating from rainfall and stock still contains thyroid active molecules, results from chemical analysis performed on the same samples indicated a correlation between the total micropollutant load, and the thyroid effect 2) A minor part of the thyroid effect removal occurs during and after WWTP processes. The major removal of the thyroid active molecule occurs during the nitration step of the water treatment.
studies focus on terrestrial and aerial species in comparison to aquatic organisms. A reason for this discrepancy is that compared to terrestrial species, additional technical challenges need to be overcome when studying aquatic species e.g. light refraction and reflection interferences at the air/water boundary, positioning of the light source and suitable body marking techniques. However, a deeper understanding of the movement patterns of small-sized aquatic invertebrates and planktonic organisms is urgently needed, and the ability to swim or swimming behavior can be used as a stress sensitive 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, polycarbonate plates with an inner diameter of 55 mm were constructed. The tested organisms were placed in the costume-made plates and recorded under the microscope or in the observation chamber DanioVision. 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 easy-to-use, fabricated 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 Ökotoxikologie (MA 25 – Project 15-06) is gratefully acknowledged.

**MO192** Validation of the in silico prediction tool for toxicity of Algae by pharmaceuticals in environment
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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 of pharmaceuticals to new drugs before marketing. It would be useful to predict the ecotoxicity of the new drug at the developmental stage, because the ecotoxicity 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 of ecotoxicity prediction by the ECOSAR software, with the prediction of daphnia toxicity prediction 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 QSR tool, more researches on discovering the structure dependent taxonomic 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.

**MO193** SeqAPASS to Evaluate Conservation of High-Throughput Screening Targets Across Non-Model Species
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 calculated across species to derive conservation of each target across taxa. To demonstrate application of the SeqAPASS data for extrapolation of ToxCast targets, case studies were developed that focused on the extrapolation of targets being evaluated as part of the Endocrine Disruptor Screening Program, including the androgen receptor, enzymes involved in steroidogenesis, and proteins involved in the menstrual cycle. These studies demonstrated the utility of SeqAPASS for informing the extrapolation of HTS data and identification of model organisms likely to be suitable for follow-up or complementary in vivo toxicity tests. The contents of this abstract neither constitute nor reflect official US EPA policy.

**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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The Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool was developed to address needs for rapid, cost effective methods of species extrapolation of chemical susceptibility. Specifically, the SeqAPASS tool compares the primary sequence (Level 1), functional domain sequence (Level 2), or individual amino acid substitutions impact protein-chemical interaction. This study found that substitutions in identifiers of key amino acids cause no change in chemical interaction with chemicals made manual interpretation of Level 1 analyses time consuming and potentially inconsistent. Therefore, this study used in silico site-directed mutagenesis coupled with docking simulations of computational models for acetylcholinesterase (AChE) and ecodose receptor (EcR) to investigate how specific amino acid substitutions impact protein-chemical interaction. This finding suggests that substitutions in identifiers of key amino acids cause no change in chemical interaction with a protein if residues share the same side chain functional properties and have comparable dimensional properties, whereas differences in side chain functional properties or molecular dimensions can reduce protein-chemical interaction. These findings were considered in the development of automated Level 1 analyses and enabling automatically generated species-specific predictions of chemical susceptibility. These predictions were shown to agree with Level 1 and 2 predictions of AChE and EcR for more than 90% of investigated species, but also identified dramatic species-specific differences in chemical susceptibility that align with results from standard toxicity tests. The consistency of automated predictions of susceptibility across Levels 1, 2 and 3 and agreement with results of standard toxicity tests provides a compelling line-of-evidence for use of SeqAPASS in identifying screening level species-specific chemical susceptibility predictions across broad taxonomic groups applicable to addressing challenges in species extrapolation for human and ecological hazard assessment. The content of this presentation neither constitute nor necessarily reflect US EPA policy.

**MO195** Survival and Teratogenic Evaluation of 91 compounds with environmental impact
S. Calzolari, ZcClinics ZcClinics (www.zcclinics.com) is a biotech company interested in developing efficient and reliable zebrafish screening tests to predict compound toxicity. Consequently, and organ relative to development of high-content 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 calculated across species to derive conservation of each target across taxa. To demonstrate application of the SeqAPASS data for extrapolation of ToxCast targets, case studies were developed that focused on the extrapolation of targets being evaluated as part of the Endocrine Disruptor Screening Program, including the androgen receptor, enzymes involved in steroidogenesis, and proteins involved in the menstrual cycle. These studies demonstrated the utility of SeqAPASS for informing the extrapolation of HTS data and identification of model organisms likely to be suitable for follow-up or complementary in vivo toxicity tests. The contents of this abstract neither constitute nor reflect official US EPA policy.
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 dipheno, 3-iodo-2-propynyl n-butylcarbamate, diethyldithiobenzol, hexachlorophene, methylmercury chloride, rotenone and tetraethylthiuram 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 mutagenic 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 similar to 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 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 metabolite 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.

**MO197**

**SETAC Animal Alternatives Interest Group**

A. Lillichop, NIVA Norwegian Institute for Water Research / Ecotoxicology

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 plat form 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 organic contaminants in water. By this regard, several PSDs have been used to monitor hydrophilic organic compounds (HOCs) in PW including semipermeable membrane devices (SPMDs). However, SPMDs are not suitable for monitoring HpOCs in PW owing to the nature of these chemicals. It is therefore necessary to develop and standardize a passive sampler for HpOCs, such as APs. Polar organic compounds (POCs) is the most abundant class of pollutants and has been used to monitor 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 (Rr), 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. This 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 hydrophilic 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 sediments contaminated by hydrophobic organic contaminants (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 (CPS). The CPS 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. These effects are accounted for in ex situ sampling protocols by providing rinse solutions to measure CPS 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 six different soil types and field sites. 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 samples 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 taken and used as 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-sterilized and non-sterilized 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-scale conditions (sterilization after contamination) and freshly spiked (sterilization prior to contamination) soils for the case of 1,1-di chloro-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 changes in SOM chemistry manifested in a reduction of bands of aromatic, and aliphatic moieties (spiking), and of reduction in bands of aromatic, and 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 between sweeten controls was pronounced compared to sterilization. The variation of bioaccumulation factors relating to soil and sediments contaminated by hydrophobic organic contaminants (HOC) in soil pore water or suspensions (CPS). 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 (CPS). The CPS 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. These effects are accounted for in ex situ sampling protocols by providing rinse solutions to measure CPS 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 six different soil types and field sites. 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 samples 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 taken and used as 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.
non-sterile 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 earthworm of two fungicides: comparison of laboratory and field experiments**

S. von der Heijden; G. Schramm; S. Batiol; D. Béranger; AgroParisTech; C. PELOSI, INRA (Institut National de la Recherche Agronomique) 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 to 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 dimethoxytriazine 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. To both cases, four replicates were performed using the recommended Swing Gold® dose, extra doses adapted to observe ecotoxicological effects and controls. The dissipation rate of the fungicides concentrations in soil were determined in 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 determined by evaluating 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. After 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 field conditions. The high heterogeneity was highlighted, 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 significantly 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 PEMETO) 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 process is crucial to avoid overestimation of bioaccumulation potential. 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 procedures 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) – 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.

**MO293**

**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 processes. 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 non-linear least squares regression equations are presented. The relationship between Kow and the two chemical classes of pesticides, namely aromatic and aliphatic partition coefficients of neutral organic compounds between water and immobilised artificial membranes (IAM), liposomes (membrane lipid), triolein (storage lipid), 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 relevance 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 distribution on the sorption kinetics and bioaccumulation potential of sediment-associated HOCs. Bioaccumulation test with oligochaete Lumbriculus 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 >500μm. The respective size fractions were 18.2%, 14.7%, 28.6% and 30.1%. 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, which supported the fact that HOCs tend to desorb from fine 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.

**MO295**

**Effect of suspended particle on polycyclic aromatic hydrocarbon (PAH) bioaccumulation by zebrafish (Danio rerio)**

Y. Zhao, X. Xia, School of Environment, Beijing Normal University; X. Xiong, School of Environment, Beijing Normal University / School of Environment; L. Xia, x. guo, School of Environment, Beijing Normal University

Hydrophobic organic compounds (HOCs) tend to be associated with suspended particles in surface aquatic systems, however, the bioavailability of HOCs on suspended particles to fish is not well understood. In this study, a passive dosing device was used to control the freely dissolved concentration of polycyclic aromatic hydrocarbons (PAHs) including fluorenone and pyrene, and the influence of particle-associated PAHs on their bioaccumulation by zebrafish was investigated. Results showed that the body burdens of PAHs in the zebrafish including the digestive tract (without the head part) were higher than that in the zebrafish excluding digestive tract at the beginning of PAH bioaccumulation, and lower after two day bioaccumulation. The difference may be caused by the effect of PAHs associated with particles in the digestive tract. When PAHs on suspended particles were ingested and they were mainly stored in the digestive tract of zebrafish in the beginning of the bioaccumulation; because the bioaccumulation factors of PAHs in zebrafish were higher than the partition coefficients of PAHs in suspended particles, the concentrations of PAHs in zebrafish excluding digestive tract were higher than that in zebrafish including digestive tract in the later bioaccumulation.
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 bioaccumulation. 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 studies

B.H. Macee, ARCADIS; N.D. Forsberg, Arcadis U.S., Inc. / Environmental and Molecular Toxicology; A.K. Meyer, United States Army Corps of Engineers / Huntsville Center

The U.S. Department of Defense is responsible for the environmental restoration of properties that were formerly used for firearm training ranges. Remnants of spent shell casings were 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 skets were commonly prepared using coal tar as a binding agent. It was hypothesized that the nature of the coal tar pitch/limestone matrix of the sket fragments reduces the oral bioavailability of PAHs compared to that seen in animal studies using pure benz[a]pyrene in solvents added to diets. To test this hypothesis, soil samples were collected from the two sites to provide a range of PAH concentrations. Female B6C3F1 mice were fed diets amended with soil or soil extracts at a rate of 5% in the diet for fourteen days. For benz[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 in animals treated with soil 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 RBAF from multiple sample points is to determine a linear regression of 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. Particle RBAF 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

T.J. Richter, BASF SE, Agrarzentrum Limburgerhof / ADP; T. Richter, BASF SE Agrarzentrum Limburgerhof / Global Product Safety and Registration; K. Platze, BASF SE Agrarzentrum Limburgerhof / Environmental Fate Modeling; A. Immer, Eurofins Agroscience Services EcoChem GmbH; M. Traub, Eurofins Agroscience Services EcoChem GmbH / Environmental Fate

The determination of adsorption coefficients is a critical key parameter for the assessment of the leaching properties of low adsorbing compound’s 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. Determination of distribution coefficients is done based on the direct method, hence extraction and 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

L. Rolando, Instituto de Recursos Naturales y Agrobiología de Sevilla / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiología / Agroquimica y Conservacion del Suelo

Atrazine is an herbicide used to control grassy and broadleaf weeds in sugarcane, wheat, corn, soya, sorghum and 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 herbicide 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 responses 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 for 28 days of trials. The data was non-parametric in nature, being analyzed using Kruskal-Wallis ANOVA followed by Dunn’s test. The level of significance was set to be 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 as *E. fetida* from hatching. This suggests that IMID could lead to decreased population growth rate or complete population collapse in such invertebrates.

MO210 Chlordecone elimination kinetics in ewes

E. Hilaire, Université de Lorraine / Zoology and Entomology; P.M. Leeto, University of the Free State / Department of Zoology and Entomology

Chlordecone (CLD) is an organochlorine pesticide used from 1972 to 1993 against gewu, a pest that attacked avocado trees in the French West Indies. We also noticed that atrazine can also elicit a negative tactic response at low concentrations. We also observed that the swimming patterns of *Pseudomonas* sp. strain ADP was affected for 28 days of trials. The data was non-parametric in nature, being analyzed using Kruskal-Wallis ANOVA followed by Dunn’s test. The level of significance was set to be 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 as *E. fetida* from hatching. This suggests that IMID could lead to decreased population growth rate or complete population collapse in such invertebrates.

MO209 The influence of biochar on the toxic effects of imidacloprid to the lifecycle parameters of *Eisenia fetida*

B.H. Magee, University of the Free State / Zoology and Entomology; P.M. Leeto, University of the Free State / Department of Zoology and Entomology

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 promotors and provide various benefits to plants and they are also 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-insecticide imidacloprid (IMID) to earthworms. Coccoins 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 in nature, being analyzed using Kruskal-Wallis ANOVA followed by Dunn’s test. The level of significance was set to be 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 as *E. fetida* from hatching. This suggests that IMID could lead to decreased population growth rate or complete population collapse in such invertebrates.

MO210 Chlordecone elimination kinetics in ewes

MO208 Evaluation of the swimming behavior and tactic response to atrazine of the Pseudomonas sp. strain ADP

L. Rolando, Instituto de Recursos Naturales y Agrobiología de Sevilla / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiología / Agroquimica y Conservacion del Suelo

Atrazine is an herbicide used to control grassy and broadleaf weeds in sugarcane, wheat, conffers, sorghum and 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 herbicide 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 responses 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 for 28 days of trials. The data was non-parametric in nature, being analyzed using Kruskal-Wallis ANOVA followed by Dunn’s test. The level of significance was set to be 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 as *E. fetida* from hatching. This suggests that IMID could lead to decreased 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 toxicityokinetic of CLD in ewe is linear. In urines, CLD and conjugated CLDHO were quantified. In feces, CLD and CLDHO were 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 of CLD in serum 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 kinetic 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 chlordecone and its metabolites by HPLC-MS/MS in urine and feces of ewes

M. Saint-Hilaire, Université de Lorraine UL / URAFPA INRA; T. Bertin, C. Intahvong, G. Lavison-Pompard, T. Gruen, ANSES / Unité PBM; A. Fourmaux, Université de Lorraine UL; C. Feidi, G. Rychen, Université de Lorraine UL / URAFPA INRA; J. Parinet, ANSES / Unité PBM

Chlordecone (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 but, 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 tested 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 chlordeco (CLDOH) in humans, pigs and gerbils livers. Then CLD and CLDOH can be conjugated by the glucuronyltransferase. In feces, CLDHO 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 a 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 quantified in ewes. In urines, CLD and conjugated CLDOH 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??

O. Machate, Helmholtz centre for environmental research - UFZ / Plant and Environmental Science; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; D. Schmoller, Helmholtz Centre for Environmental Research UFZ / Conservation Biology; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis

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 and their bioavailability. Therefore, the chemical concentrations of SOCs differ vastly in magnitude and spatial distribution within these areas. Mentioned as important drivers of these variations are the change in precipitation and temperature with increasing altitude and the locally prevailing wind patterns. Despite this knowledge, the task to interpret the results gained within studies on the spatial distribution of organic contaminants within high mountain areas remains difficult. To aid future research, their risk assessment this poster tries to condense the key information on fate and behavior of organic contaminants in high mountain areas. Therefore, fate determining variables will be named and the movement and bioavailability of organic contaminants throughout the seasons be described.

MO213 Pesticide occurrence in different agricultural matrices (honey bees, wax and pollen)

P. Catayadau-Vernich, M. Andrés Costa, Universitat de Valencia / Environmental and Food Safety Research Group; F. Calatayud, E. Simó, Agrupacion 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 varroasis by beekeepers are accumulated in wax, pollen and honey bees. Samples of honey bees (45), to sur (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 analyses 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 are 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 chlorpyrifos 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 chlorpyrifos 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 acaricides were found. This fraction was calculated in the product of the concentrations detected in the matrices analyzed, honey bee colonies health could be compromised. Assessing pesticides content in these three different agricultural 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. Puchex, INERIS, S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances

One of the main factors 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 TROPHE project, the transfer of PCBs and PCDD/Fs to plants and invertebrates has been studied. BCF in several plants and in earthworm had been measured and different models have been calculated. One of the conclusions drawn is that there is no match between available guidelines to produce measured BCF in terrestrial organisms and the BCF needed with the REACH regulation. This guidance states that the exposure concentration for terrestrial predators can be calculated in taking in account the quantity of soil contained in the earthworms guts and the contaminant fraction measured with guideline relatable to the contaminant concentration in interstitial water and the BCF. But this BCF, relatable to interstitial water, is not comparable with BCF measured with available guideline as OECD 317 – Bioaccumulation in Terrestrial Oligochaetes, relatable to total concentration in soil. Data obtained in the context of the TROPHE project allow for the comparison between PCB-PCDD/F BCFearthworm measured with the OECD 317 guideline and PCB-PCDD/F BCFEarthworm extrapolated from the Kow of the substance. It was also possible to illustrate the impact of these differences on the results of the secondary poisoning exposure modeled concentrations. A screening on the ECHA registration site also provides an approximation of the number of registered substances that have a BCFearthworm measured with guideline relatable to total concentration in soil and therefore unusable as such in the recommended methodology according to REACH.

MO215 Assessing risks from PBT substances in surface waters: possible alternatives to biota monitoring?


The Water Framework Directive (WFD) requires waterbodies to be at ‘good ecological status’ by meeting Environmental Quality Standards (EQSs). Normally, EQSs are expressed in water but in active work, BCFs standards 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. Biotas 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

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; Chukwu 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 Sensitive Surfactant Residue 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. c. 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.

REFERENCES

MO218 Uncertainty concepts and misconceptions for landscape scale risk assessment

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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-situ 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 less uncertain lower 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 compounding 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 reduce this. Here we discuss some 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

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The spatial pattern of plant communities in 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 decisive environmental and soil parameters (soil type, soil texture, moisture etc.) are known and the vegetation is classified in a database, the relative abundance of species is predictable. For this study a GIS database with soil, vegetation and climate data has been established and plant community modeling has been performed. The results suggest this approach could be an useful tool for an early warning system for crop conditions. The current study is extended towards a larger scale and an additional modeling of the land management. This is a basis for a meta-modeling approach for regional landscape vegetation analyses. The next steps will be the evaluation of the model and the validation of the vegetation predictions in landscape scale.

MO220 B-Rice: bird focal species identification in rice paddy

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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 the presence of several waterfowl species: we tested the applicability of the GD to the rice paddy 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

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EFSA’s guidance document for the risk assessment of edge-of-field 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 algal abundance to simulate effects and recovery of algal populations over time following exposure to PPPs. The model integrates the main processes driving algal cell growth, such as (1) toxity of the PPP and (2) growth conditions due to the climate conditions, natural mortality, grazing and competition for resources, and density dependence. This model also makes a significant step towards full compliance with EFSA good modelling practices, whereby models for regulatory risk assessments should include validation, and sensitivity and uncertainty analyses. In this poster, the formal model as well as sensitivity and uncertainty analyses are depicted. We also employ empirical data from mesocosm studies conducted for a selective herbicide for model validation.

MO222 Population dynamics of a soil arthropod simulated using an individual based population model and established fate model data

D. Nickisch, T. Wittwer, Rifcon GmbH

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 recognised as a beneficial tool for adding ecological realism to ERAs, EFSA has so far not published any guidance document. Population modelling is at least mentioned in several EFSA (draft) guidances 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 adress 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 harmonised 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 a database of variable simulations to generate flushing experiments under variable pre-flushing time series from JRC databases. Those series characterised 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  
C. Vitale, University of Insubria; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; M. Morselli, A. Di Guardo, University of Insubria / Department of Science and High Technology  
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 in three categories of leaching solutions under variable pre-flushing times (2, 5, 7, 48 days), using leaching solution with different DOC content (tap water vs. a solution prepared with commercial humic acid), at different temperature (25 °C vs. 15° C) and in saturated vs.pseudo field capacity conditions. Results indicated that equilibration time determined differences in measured PCB concentrations up to a factor of 8, probably due to the lack of equilibration with the environmental fractions in DOC such as humic 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; K. Kim, Gwangju Institute of 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 numerical entries. The toxicological sensitivity was derived by indirect prediction based on traits because enough 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 (KEITI) through "The Chemical Accident Prevention Technology Development Project", funded by Korea Ministry of Environment (MOE) (No. 2016001970001).

MO225 Assessing and managing food-web effects of Plant Protection Products  
K. Swarowsky, German Federal Environment Agency (UBA) / Department IV  
Plant protection products (PPP) active substances (Regulation EC 283/2013) also acknowledge this aspect by mentioning indirect effects to be considered in the assessment of the impact. In this framework, we performed a PPP-life cycle study for the chemical glyphosate used for weed control and assessed its potential effects on biodiversity (and thus to compensate for indirect effects of PPPs). 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 a database of variable simulations to generate flushing experiments under variable pre-flushing time series from JRC databases. Those series characterised the environment in the springtail model and drove the temperature-dependent model processes.

MO226 Compensating for ecological risks of pesticides  
S. Matezki, K. Swarowsky, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV  
Compensating adverse effects of pesticides where established methods of risk assessment 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.
MO228
Historical control data of the optimized Zebrafish Embryonal 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. Bekkhuizen, 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 test 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 sacculus/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 Embryonal 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. Bekkhuizen, 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 test 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 exposure 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 (no renewal) vs. semi 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 sacculus/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 not increase more malformations or mortality than exposure to adjusted ISO medium.

MO230
Reliability of ecotoxicological studies in fish
H. Winnemann, Bavarian Environment Agency; H. Ferling, Bavarian Environmental Agency; G. Dembek, W. Schmidt, W. Koernner, Bavarian Environment Agency; J. Schwaiger, Bavarian Environment Agency / Aquatic Toxicology and Pathology

For substance 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 EQSes. 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 statistically significant p-values, insufficient testing duration, insufficient number of concentrations tested, missing chemical analysis of test compound concentration in the test water, calculation of toxicological endpoints on the basis of nominal and not real concentrations, or insufficient quality of endpoints. The aim of the presentation is to outline, from our point of view, optimal experimental conditions of prolonged fish tests which can be adapted as a model for other scientific studies, through decreasing the specified number of endpoints and considering so far neglected aspects such as possible background contamination of commercially available fish feed frequently used in fish studies.

MO231
Assessment of the relationship between heavy metal bioaccumulation and biomarker responses in Japanese dace inhabiting 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 Ryukyu / 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/L Cu in river water in 1897). Although the heavy metal concentrations remarkably decreased since 1960s, the concentrations are still higher than those in a 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 in the Watarase River were 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 (cystein-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.

MO232
Micronucleus test to evaluate effects of 4 metals on DNA damage of zebrafish Danio rerio
A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Lab. Limnology and geology; A. Perez-Rojas, Universidad Autonoma Metropolitana Iztapalapa / Department of Hydrobiology; S. Kashiwada, Toyo University / Graduate School of Life Sciences; Y. Kato, Toyo University / Faculty of Life Sciences; Y. Shimizu, Toyo University / Graduate School of Life Sciences

Danio rerio is a species of importance since it is used as a test organism for ecotoxicological studies at the International level. In our country the tests with this fish are mainly conducted 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 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 EQSes. 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 statistically significant p-values, insufficient testing duration, insufficient number of concentrations tested, missing chemical analysis of test compound concentration in the test water, calculation of toxicological endpoints on the basis of nominal and not real concentrations, or insufficient quality of endpoints. The aim of the presentation is to outline, from our point of view, optimal experimental conditions of prolonged fish tests which can be adapted as a model for other scientific studies, through decreasing the specified number of endpoints and considering so far neglected aspects such as possible background contamination of commercially available fish feed frequently used in fish studies.
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. retio, 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 (0.04%). The metal with the highest genotoxic effect was lead (0.83%), followed by cadmium (0.65%). Copper showed the lowest genotoxicity (0.37%). The metal mixture had a microniculous frequency of 1.23 %. The juveniles of D. retio had deleterious effects in concentrations of metals lower than the LMP (Maximum Permissible Limits) that marks the NOM 001 Semarmat 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 retio) K. Ji, J. Lee, Yongin University As alternative compounds of bisphenol A (BPA), bisphenol S (BPS) and 4-hydroxy-3-(3',4'-isopropoxyphenylsulfone (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 retio) 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 the hypothalamus-pituitary-gonad (HPG) axis were investigated. The estrogenic (increase in 17β-estradiol/testosterone [E2/T] ratio) and anti-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 stereogenic 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 Environmental pollutants, like polycyclic aromatic hydrocarbons (PAHs) found in crude oil, have the potential to breakdown 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 damage, total antioxidant power analysis, 2-Thiobarbituric Acid Reactance 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/oxy-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/oxy-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[a]phenantherene-4-one (4H-CP), benzo[a]fluoranthene (BFO) 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 to 6H-BPO and BFO 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 BFO and 4H-CPO in mixture with BP. Gene expression analysis showed significant up-regulation of genes in various treatments. The only gene (up-regulated) was Card15, a non-Il1r receptor antagonist that up-regulate CYP1a expression and thereby induce their own metabolism, while other PAHs like fluoranthene directly inhibit CYP1a activity. We newly exposed hatched rainbow trout fry (Oncorhyncus mykiss) semi-statically to retene and fluorenone either as a single compound 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. Erythrocyte cell death, heart growth and energy consumption by day 14. Microarray analysis showed that the different treatments caused very different alterations in the transcriptome, both in terms of the number of changed genetic expressions and when and in time. The only gene (up-regulated at all sampling times and treatments was cyp1a. In addition, cyp1a and cyp1b-like-dimensions expression were found similarly changed across all treatments but not at all sampling occasions. Using over-representation analysis revealed several biological processes affected, such as blood vessels and heart development following mixture treatment. Proteomic analysis is underway but protein expressions are suspected to show a low transcript to protein correlation (based upon literature). Heart tissue metabolomic analysis revealed that across all treatments only two out of 33 metabolites were found significantly changed by day 14. Using omics integrative methods, our study discovered several pathways affected by PAH exposure, together with phenotypical alterations, highlighting the unique Moa of different PAHs and as a mixture.
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 the vascular endothelium, (e.g., arterial) via unknown AhR-independent mechanisms. In this study, we aimed to explore the mechanisms of toxicity of individual PAHs in the rainbow trout (Oncorhyncus 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 to validate their role in the mechanism of toxicity. Newly stocked rainbow trout larvae were exposed to three different PAHs (retene, pyrene or phenanthrene) at subletal doses. The heart of each larva was sampled after 1, 3, 7 or 14 days of semi-static exposure, and DNA, proteins and metabolites were extracted. Morphometric parameters such as larval length and yolk sac area were also monitored, but were barely 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 livable with pathways such as glycolysis and P540, cation transport, muscular contraction or steroid hormone biosynthesis in the case of retene. Some of those processes were shared by pyrene. Phenanthrene 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.

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The study investigated the developmental toxicity of Acetaminophen, a non-steroidal Anti-inflammatory Drug on the early life stage (0 to 96 hr) 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) of the drug in triplicates and observations of embryo development were made at different developmental stages. Morphological, physiological and behaviourally altered embryos were livable. Exposure to acetaminophen significantly altered the morphological and behavioural properties of the fish. The effects observed were 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

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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) of the drug in triplicates and observations of embryo development were made at different developmental stages. Morphological, physiological and behaviourally altered embryos were livable. Exposure to acetaminophen significantly altered the morphological and behavioural properties of the fish. The effects observed were 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.
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 others. Taken together, benzenetriols including pyrogallol may be caused by unexpected inhibitory effects on the animal COX activity, reflecting 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 C. Parenti, A. Ghilardi, M. Mandelli, University of Milan; C. Dei Torre, State University of Milano / Biosciences; S. Magni, University of Milan / Department of Biosciences; L. Del Giacco, University of Milan; A. Binelli, University of Milan / Department of Biosciences

Triclosan (TCS, 5-chloro-2-(2,4-dichlorophenox) 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 WasteWater 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. These evidences demonstrate 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 US. The aim of this study was to investigate the adverse effects of TCS at environmental concentrations on zebrafish embryos up to 120 hours post-fertilization (hpf). It is the first time that environmental levels of this contaminant were taken into account, instead of evaluating the effects of sub-lethal or lethal concentrations. The experimental plan consisted in the exposure to two different environmental concentrations of TCS (0.1 and 1 μg/L) for 5 days following fertilization, under semi-static conditions. A suite of biomarkers was applied to evaluate the potential mechanisms underlying the toxicity of TCS such as the generation of oxidative stress and DNA damage. The activity of antioxidant and detoxifying enzymes, namely catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx), and glutathione transferase (GST), were measured, while the genetic damage was evaluated as the occurrence of the micronucleated cells (MN test). Results show a significant increase in all biomarkers measured, indicating that this chemical is dangerous for aquatic species also at environmental concentrations.

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

Microcystins (MCs) are hepatotoxins produced by several groups of cyanobacteria in water bodies throughout the world. Their mechanism of toxicity consist of a potent inhibition of protein phosphatases 1 and 2 A, which causes disruption of the cytoskeleton and consequent cell death. They can also alter the antioxidant system and induce oxidative stress in various organs of many species. Microcystin-LR (MC-LR) is the most studied variant due to its high toxicity and frequent occurrence in surface waters. In this study, we used a Microcystis aeruginosa bloom extract consisting in the exposure to two different environmental concentrations of TCS (0.1 and 1 μg/L) for 5 days following fertilization, under semi-static conditions. A suite of biomarkers was applied to evaluate the potential mechanisms underlying the toxicity of TCS such as the generation of oxidative stress and DNA damage. The activity of antioxidant and detoxifying enzymes, namely catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx), and glutathione transferase (GST), were measured, while the genetic damage was evaluated as the occurrence of the micronucleated cells (MN test). Results show a significant increase in all biomarkers measured, indicating that this chemical is dangerous for aquatic species also at environmental concentrations.

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. Microcystins (MCs) are the most common cyanotoxins and may be expected whenever blooms of cyanobacteria occur in surface waters with 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) common carp (Cyprinus carpio). The fishes were been randomly assigned to three groups. Group I, is the control group, received daily physiological serum (500 μL) containing 100 ppm contained 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 study. 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 kidneys, epithelial cell swelling and hypertrophy in some cases, with malformations of the lamellae within 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, 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, Cyprinus 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. Philibert, 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 dilbit or to fracture stimulation in oil and gas wells. Bitumen was exposed to water accommodated fractions of these oils from 0.7 to 7 days after 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 the exposed second-generation embryos when compared to control, 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 diluted conventional and unconventional crude oils have varied 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 V. Zlabek, University of South Bohemia in Ceske Budejovice / Faculty of Fisheries and Protection of Waters, LECHB; V. Burkina, University of South Bohemia in Ceske Budejovice / Faculty of Fisheries and Protection of Waters South Bohemian

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Research Center of Aquaculture and Biodiversity of Hydroenoses Vodnany Czech Republic; G. Zamaratskaia, Swedish University of Agricultural Sciences / Department of Food Science
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 environments and its potential to modulate fish organs 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-methylindole, and 3-hydroxy-3-methylindole, can interact with fish CYP isoforms. Enzyme activity test for CYP2A, CYP3A, CYP1A in rainbow trout hepatic microsomes 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-methylindole and 3-hydroxy-3-methylindole 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. 18-15082s) and Swedish University of Agricultural Sciences.

MO248
Linkage of gene expression patterns with in vivo endpoints: gaining deeper insights
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The herbicide glyphosate and the pharmaceutical lisinopril are among the most popular chemicals that have been detected in many streams at low concentrations. Nevertheless, the ecotoxicological properties, especially of lisinopril, 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 (ELS). Per substance 480 individuals of zebrafish were exposed to five different concentrations (lisinopril: 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 malformations. Gene-expression patterns of such inhibition for fishability to detoxify xenobiotics remain to be elucidated. It was concluded that lisinopril is potentially carcinogenic to the zebrafish by affecting the antioxidant defence system. In addition, lisinopril was associated with the formation of angioedema and induced cardiac toxicity in zebrafish by the downregulation of NFP gene expression. Finally, it should be noted, that sudden death of the zebrafish were observed depending on the dose of lisinopril. A possible reason could be the reduced expression of the ACE2 enzyme. On the other hand, glyphosate slowed the heart rate and significantly increased the HAVCR1 (KIM-1) expression. The SOD1 gene expression was significantly increased because of glyphosate exposure whereas SOD2 and NPPB gene expression were not affected.

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 in the modulation of nuclear receptors (NRs) and other hydrocarbon receptors (AhR) by individual chemicals and environmental mixtures, in order to help further cross-species extrapolation in the frame of the environmental risks of EDCs. To this end, a panel of individual ligands and environmental mixtures from an urban waste water treatment plant (WWTP), were screened on a set of recently developed in vitro reporter cell lines based on both human (h) and zebrafish (zf) NRs. The aim of the study was to find early detection markers for cross-species differences occurred (PXR, PPAR, PR) while for other receptors the differences were lower (ER, AR, GR, MR) or almost absent (AhR, ERR). For instance, progesterone acts as a full agonist of the hPXR but as partial agonist of the zfPXR whereas the dihydroxy-4-pregnen-3-one reference ligand of the zfPXR- antagonizes the hPXR. In the same way, none of the reference ligands of the hPXR (T09131137) modulates the zfPXR whereas the clotrimazole (a bronchic ligand of zfPXR) modulates also the hPXR but with lower potency. Then the hAR was more sensitive to the agonist mefipristone and the antagonist OH-flutamide than the zfAR whereas the dexamethasone was a more potent agonist of the zfGR than the hGR. Also significant differences in selectivity were noted among h and zf ER subtypes. Finally, the in vitro profiling of an urban WWTP confirmed these cross-species differences in terms of level, type (agonist vs antagonist), distribution along the WWTP. For instance, h and zf estrogenic activity was differentially detected in the sludge and the suspended material. In the same way, strong zf anti-androgenic activity was detected in the effluent while no h one can be found. Also, strong zf mineralocorticoid activity was detected in both influent and effluents whereas only zf androgen activity was detected. Altogether, our results showed that h and zf NRs are, for some of them, differentially modulated by individual chemicals and environmental mixtures. Also, interaction of EDCs towards NRs cannot always be extrapolated between these species highlighting the need to further document NR 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
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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 its potential harmful 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 the 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 hitherto unknown mechanism of TCS, its incorporation in different determinants, 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, nucleobases, etc.) was established prior to building a broad spectrum database. Postaglandin F2-like derivatives have emerged as a reliable biomarker for oxidative stress in mammals and fish namely the F2-isoprostanes (F2-isops). F2-isops are the free-radical catalyzed products of non-enzymatic lipid-peroxidation of arachidonic acid, a fatty acid found in brain tissue and cell membranes. Fish mucus has been investigated in several studies as a potential biomarker and matrix for the determination of oxidative stress in fish. F2-isoprostanes is composed mainly of glycoproteins, but notably contains immunoglobulins, pheromones, lysozyme and proteolytic enzymes. Mucus is known to have important biological functions for fish, ranging from communication and reproduction to osmotic regulation. To date, no method for the isolation and quantification of F2-isops in fish mucus has been reported. The aims of this study was to develop an efficient method for the extraction of F2-isops from fish mucus and to optimize the resolution and quantification of F2-isops by high performance liquid chromatography tandem mass spectrometry. The method was based on acidification of mucus with HCl and extracting with ethyl acetate. The
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 (Oncorhynchus 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 region. RNASeq was carried out on intestinal regions and mapped back to the rainbow trout genome (84 %). Following filtering for transcript abundance using TPM and a P-value cut off, 23,635 – 25,435 contigs were identified over the 4 regions and included enzymes involved in metabolism of chemicals such as the cytochrome P450 family (CYPs). Differential expression of genes between regions did not vary significantly between the pyloric, anterior or mid intestine (~6 genes), however, this changed markedly between the pyloric and posterior region (~29) highlighting their differences. Proteomic characterization established over 3,899 proteins present in the intestine with annotated proteins varying from 3,100 to 3,899 dependent on intestinal region. Significant differences in proteins were observed between intestinal regions further confirming trends observed in the parallel transcriptomic study. These data represent the first thorough characterization of the rainbow trout intestine, and will allow the identification of enzymes present in this organ which 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.
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Several studies demonstrated that exposure to persistent organic pollutants can induce epigenetic modifications in human and other vertebrates including fish. It is of particular interest since epigenetic changes were reported in laboratory models 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 mammals; 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 8 selected compounds. The cells were exposed to the LC10 values of pesticides (methoxychlor (MXY), permethrin (PER)), plastic additives (bisphenol A (BPA) and S (BPS)), perfluorinated compounds (perfluorooctane sulfonic acid (PFOS), perfluorobutane sulfonic acid (PFBS)), a whitening agent, 7-dihydroxy-4-methylcoumarin (DEMC); and to the metalloid selenium (Se). 25,435 contigs were identified over the 4 regions and annotated proteins varying from 3,100 to 3,899 dependent on differences. Proteomic characterization established over 3,899 proteins present in the liver, with a non-invasive, non-portal matrix for F2-ISOps analysis in fish.

MO255
Cross-species applicability of the adverse outcome pathway "deiodinase inhibition leading to impaired swim bladder inflation in zebrafish".
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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 test chemicals to screen fish species native to North America. DIO assay predictions were analyzed together with 2 histone deacetylases (hdac1, hdac3), one demethylase (jariid1b), and one chromatin remodeling factor (spht6). At the selected concentration of 0.5 μg/mL, 15 out of 28 were reared to swim as larvae, brook trout and northern pike. Several studies demonstrated that exposure to persistent organic pollutants can alter RNA methylation and histone modifications. Interestingly, industrial alternatives BPS and PFBS induced maximal expression changes of epigenetic factors than the well-kown BPA 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 which extent the observed changes are reflected in DNA methylation and chromatin accessibility themselves, together with their correlation in in vivo models.
Zebrafish responses to the fourth-generation progestin drospirenone exposures

L. Vergauwen, University of Antwerp / Zebrafish Lab Dept Veterinary Sciences

The fourth-generation progestins (PGs) represent an important class of active ingredients of hormonal pharmaceuticals. In contrast to their endocrine disrupting effects, it is known that PGs can interfere with the processes in fish, such as the induction of adaptive stress responses. Drospirenone (DRP) exposures arise as one of the most used fourth-generation PGs in hormonal pharmaceuticals. In contrast to its endocrine activity, it is known that DRP can interfere with other processes in fish, such as the regulation of circadian rhythm. Thus, the present work aims to evaluate Danio rerio early life stages responses to DRP exposures at physiological and biochemical level. Zebrafish embryos were exposed to 0.01 – 100.0 µg/l of DRP during 96h to evaluate lethal and sublethal parameters. Survival, heartbeat, length and impairments on normal development such as malformations and hatchings were evaluated at in vitro and physiological endpoints. Alterations on enzymes related with neurotransmission (acetylcholinesterase, AChE), energy production (lactate dehydrogenase, LDH) and oxidative stress (catalase, CAT and glutathione S-transferase, GST) were assessed. The oxidative damage was also assessed by alterations on lipid peroxidation levels (LPO). Exposure to DRP did not affect hatching rate, growth and development of zebrafish embryos, however, there was a decrease of a number of chemicals were identified as DRP related endpoints. Biochemical processes were affected by DRP exposure and oxidative damage was observed. Overall, despite not having affected zebrafish early life stages apical endpoints, our study showed that DRP might exert adverse effects at both physiological and biochemical levels at concentrations similar to those found in environment for PGs. Furthermore, our results highlight the need to assess PGs toxicity at different levels of biological organization.

MO258
Fish caging experiment as a tool for detection of in situ effects of untreated wastewater: General stress and endocrine disruption

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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 oil and grease were identified. Endocrine disruption and oxidative stress were the endpoints monitored. In vitro mRNA expression 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., Cyprinidae), both male and female, were exposed for nine days at three sites in the River Danube: upstream (reference site) cca. 250 m and 7 km downstream of the major wastewater discharge point and in the intertidal zone, and possess paternal care. The aim of the present work is to validate the hypothesis on 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.

MO260
Skin vitellogenin and estrogen receptor as sensitive biomarkers of estrogenicity in a sub-Antarctic fish.

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Skin vitellogenin (vtg) and estrogen receptor (ERα, β) as biomarkers of estrogenicity in skin samples of this species in order to provide a helpful tool to develop and perform risk assessment for the different contaminants and aquatic environments. Their widespread use has led to the presence of anthropogenic pollutants in water, sediments and in vertebrates from Antarctic areas; however, information about their potential impact on fish physiology is still scarce. The southwesternmost city of the world, Ushuaia (Tierra del Fuego, Argentina), and its bays are the exceptions to the decline of the environmental quality. Nototomid fish are the dominant group found in the Ushuaia coastal area, and have been used as Antarctic region, playing a key role in these ecosystems. The black southern cod, Patagonotothen tesselatus is widespread in the Beagle Channel, lives in the intertidal zone, and possess paternal care. The aim of the present work is to validate vitellogenin (vtg) and estrogen receptor (Ren) as biomarkers of estrogenicity in skin samples of this species in order to provide a helpful tool to develop and perform risk assessment for the different contaminants and aquatic environments.

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 (qRT-PCR). Selected thyroid-related genes were evaluated in liver and brain samples of this species in order to provide a helpful tool to develop and perform risk assessment for the different contaminants and aquatic environments.
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 ESR 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 ESR 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
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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 species. Our University of Saskatchewan – Toxicology Centre / Toxicology Research has evaluated the effects of estrogenic compounds on the brain development of zebrafish (Danio rerio) larvae. Thereby, substance and concentration dependent effects were observed, indicating differing toxic modes of action. Within this study we aim to further investigate thyroid disruption in zebrafish early life stages and elucidate a possible link to (developmental) neurotoxicity. Therefore, embryos and larvae of zebrafish are exposed to different thyroid disrupting compounds. The methodology approach proposes to assess the neurotoxic potential of the test substances based on different behaviour assays, the mechanistic link between thyroid and neurotoxicity will be made using transcriptomics, proteomics and metabolomics. This work will be conducted within the scope of the “NeuroBox” project. In NeuroBox novel bioassays are developed, with the objective to access the neurotoxic potential of water contaminants and improve water quality, ultimately aiming to reduce the exposition of humans and the environment to these substances. In this context, the project is expected to further contribute to the understanding of basic mechanisms of neurotoxicity, its connection to thyroid disruption and to identify novel endpoints. This knowledge may then be integrated in a bioassay battery and used for the improvement of water quality guidelines.

MO263
Identification of toxicity pathways predicting adverse outcomes of chlorpyrifos in fathead minnows
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Chlorpyrifos is an organophosphate insecticide that acts as a neurotoxicant through inhibition of the enzyme cholinesterase. The mode of action of organophosphates in target and non-target organisms, including mammals, is similar. The aim of the project is to develop an early life-stage gene expression assay (EcoToxChip) that captures critical toxicity pathways of chlorpyrifos for the prediction of apical outcomes of regulatory relevance. As this assay is intended to use early life-stage stages (up to 96 h) as substitute test methods, it would not be considered as a live animal test, and therefore, would address the need for alternative approaches in chemical screening. As part of the project, critical toxicity pathways and associated core genes will be identified following exposure of fathead minnows (Pimephales promelas) at early life-stages to three sub-lethal concentrations of chlorpyrifos. Specifically, sequence-by-synthesis-based whole transcriptome (RNASeq) and high resolution lipoperoxidation-based (Tbars) gene expression profiling will characterize key molecular toxicity pathways. Pathways will then be correlated with downstream biological responses of ecological and regulatory relevance, and critical genes linked to apical outcomes will be identified for inclusion on EcoToxChips. Chlorpyrifos concentrations were selected based on a preliminary test as well as concentrations in published data. These tests revealed a threshold level of mortality between 1 and 10 µg/L chlorpyrifos. To ensure the determination of solely sub-lethal effects in at least two of the tested concentrations, 0.5, 1.5 and 4.5 µg/L chlorpyrifos solutions were investigated in the fathead minnow early life-stage assay with larvae samplings after 7 and 32 days of exposure. None of these concentrations affected survival or growth, resulting in a sub-chronic NOAEC and LOAEC of 4.5 and 10 µg/L chlorpyrifos, respectively, in fathead minnows. Samples are currently being further analyzed for molecular and physiological endpoints to gain insight into critical toxicity pathways. This study is part of the EcoToxChip project (@ecotoxchip).

MO264
Evaluation of the deleterious effect of 2 pesticides on juveniles of the zebrafish Danio rerio
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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 Imiprotin 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 (LC50). Subsequently a bioassay with a duration of 15 days it was carried out where zebrafish juveniles were exposed to 2 sublethal concentrations (LC10 and LC50). The absorption of only show that lipoperoxidation and neurotoxicity (LC50 = 1.67 ± 0.87 mg L−1) higher than Dichlorvos (LC50 = 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 imiprotin tests varied from 64.7 to 147.5 mM Tbars mg−1 and was higher than that observed in the bioassays with Dichlorvos (22.6 to 93.8 mM 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 imiprotin. 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 Imiprotin tests and 81% in the Dichlorvos bioassays. The insecticides Dichlorvos and Imiprotin 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)
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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 absorption of nutrients, osteoporosis and neurological disorders. Its toxic effects have been evaluated in mice and rats only, for this reason in this work was made an evaluation of the toxic effects of Omeprazole in zebrafish embryos. Bioassays were performed (OECD test 236) where the embryos were exposed to 5 concentrations of the drug (200, 100, 50, 25, 12.5 mg L−1) plus a negative control, to determine the LC50 (24 hours). The embryos were subsequently exposed to the LC50, LC10 and LC0 for 72 hours to evaluate the degree of lipoperoxidation, by means of the evaluation of Tbars (Buege and Aust, 1978), the activity of the enzyme AChE as an indicator of effects neurotoxic (Elliott et al., 1961) and the frequency of malformations (OECD test 236). In the lethality tests, the LC50 value of 193.87 ± 18.48 mg L−1 was obtained. In the sublethal bioassays at 72 hours of exposure it was observed an increased in the degree of lipid peroxidation (52%) in embryos exposed to LC10. In the evaluation of the AChE activity, significant differences were obtained between the control and the embryos exposed to omeprazole (p < 0.05), in the concentrations LC10 and LC5; a decrease in the activity of this enzyme was observed. The percentage of inhibition of AChE varied from 9 to 66.7%. A higher frequency (22%) of deformed embryos was observed in the LC10 concentration. The results of this study showed that omeprazole has a neurotoxic and possibly genotoxic effect in sublethal concentrations in zebrafish embryos.

MO266
The neurotoxic effects of Venlafaxine on zebrafish larvae - Oimes technologies in the focus of global environmental challenges
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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. 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. It 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 alternations/in 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 'light–dark transition test and thigmotaxis were evaluated in 5 dpf larvae exposed to 24 h to 1 nM, 100 nM and 10 PM Venlafaxine using DanioVision® and EthoVision. A significant difference in swimming behavior concerning the different concentrations could be detected. Effects on the transcriptome were verified in zebrafish chronically exposed to Venlafaxine in 1 nM, 100 nM up to 120 hpf. RNA was extracted from pooled samples (n = 25 fish) and submitted to 'byr Green quantitative real-time chain reaction (qPCR) to compare target gene expression. Nineteen target genes were considered in circadian rhythm regulation, muscle processes and responses to abiotic stimuli. Behavioral results indicated decreased swimming distance and increased thigmotaxis in fish exposed, in agreement with previous own data for continuous venlafaxine exposure. Results 'byr qPCR indicated modulation of some of the pre-selected target genes such as sklp5, and currently 'unconfirmed qPCR target gene is currently unknown. Further research focusing on the molecular pathways affected by flx on the transcriptome involved in circadian rhythm regulation, muscle processes and responses to abiotic stimuli is expected to be part of a bigger overview and understanding of 'unrelated different effects of chemicals and pharmaceuticals on neuronal development.

MO267
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. Morais, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; C. Koppe Grisiolia, 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. Pac-Taylor, University of Brasilia / Laboratory of Environmental Mutagenesis. Ayahuasca is a psychotomimetic concoction prepared with the plants Banisteriopsis caapi and 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 zebrafish larvae were placed at 0 to 1000 mg/L during 96 h of exposure. The effects on locomotor activity of zebrafish larvae were assessed using the automated video tracking system ZebraBox at 0 to 20 mg/L and after 120 and 144 hours of exposure. The ayahuasca infusion was administered once by gavage to 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 abnormalities 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 OFT and EPM, and a higher swimming time in the swimming 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 pathological alterations such as decrease of glycogen and progressive loss of hepatic architecture. The pattern of swimming behaviour 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.

MO269
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 in toxicological studies. We evaluated mitochondrial dysfunction in dechorinated zebrafish embryos exposed to individual 5 OCPs (i.e., 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., PGC-alpha, Acox1, SDHA, MCAD, and C3), associated with mitochondrial metabolism, at 120 hpf. This comprehensive evaluation could suggest the potential 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.

MO270
The NeuroBox Project
J. Lee, 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 / Department of Environmental Research. The societal impact of neurological disorders like Alzheimer’s disease or neurodevelopmental disorders like ADHD and autism is enormous. There is no evidence that most of the neurological diseases are caused by a single factor. Considering the heterogeneity of 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 known how to assess these substances with endocrine disrupters. It is a major challenge to test all substances for their neurotoxic potential. New advanced neurotoxicity assessment strategies need to be developed to fulfill these demands. The bmbf funded project NeuroBox (02WRS1419; 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 mode of actions 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.
changes were observed ad 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
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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 offspring at a high rate, cost-effective and conserved 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.

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

MO272
Effect of iodinated X-ray contrast media in the formation of disinfection byproducts during chlorination and chloramination of water
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Arsenic is a naturally occurring toxic element, which is present in waters in several features such as the fully sequenced genome, availability of a wide range of mutants, generation of offspring at a high rate, cost-effective and conserved 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.

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

MO273
The use of a polymer inclusion membrane for the determination of arsenate by gas-diffusion flow analysis with spectrophotometric detection
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Arsenic is a naturally occurring toxic element, which is present in waters in different areas around the world, including South Asia, South America and to lesser extent Europe [1]. Therefore, the World Health Organization has set the guideline concentration for arsenic in drinking water at 10 µg L⁻¹ [2]. The most frequently encountered arsenic species in environmental and drinking waters is arsenate (As(V)). Therefore, highly sensitive analytical techniques are required for its determination in water samples. In the present work a novel flow analysis (FA) system has been developed for the determination of As(V) in environmental and drinking waters at the low µg L⁻¹ level. The system uses a polymer inclusion membrane (PIM) as the acceptor for the arsenic species (poly(vinylidenefluoride-co-hexafluoropropylene) as the polymer and Aliquat 336 as the extractant, for the online preconcentration and separation of As(V) in a PIM cell. The sample solution is propelled for a predetermined period of time through the PIM cell where a PIM separates the sample stream and an acceptor stream which is stopped during the sample passage through the PIM cell. After the flow preconcentration of As(V) in the static acceptor solution located in the acceptor channel of the cell. The analytical procedure involves a 15 min stop-flow time and sample solution flow rate of 2.5 ml min⁻¹. After the stop-flow time the acceptor stream is re-started and As(V) is reduced to arsenite (As(III)) by merging the acceptor stream with a reagent stream containing 4 M HCL, 1% H₂O₂ and 0.5% ascorbic acid. This is followed by arsenite generation using another reagent stream incorporating 0.5% NaBH₄ and 0.05 M NaOH. The generated arsenite is transported across the hydrophobic membrane of a gas-diffusion cell into a solution containing 0.02 mM KMnO₄ and 0.05 M NaOH where it is oxidised thus producing a decrease in the KMnO₄ absorbance, monitored continuously at 528 nm. Under optimal conditions the FA system has a detection limit of 1 µg L⁻¹ arsenic with a sampling rate of 2.8 h⁻¹ and a repeatability, expressed as RSD of 1.8% (n=5, 25 µg L⁻¹) and 2.8% (n=5, 50 µg L⁻¹). The FA method has been successfully applied to the determination of As(V) in tap water in the µg L⁻¹ concentration range. <strong>References</strong> [1] Villaescusa I, Bollinger JC. 2008. Arsenic in drinking water: sources, occurrence and health effects (a review). Rev Environ Sci Biotechnol 7:307–323. [2] World Health Organization (WHO). 2011. Guidelines for drinking-water quality, 4th edition

MO274
Balancing environmental quality standards and infrastructure upgrading costs for the reduction of microcontaminants loads in rivers
V. P. Jimeno-Medina, Catalan Institute for Water Research (ICRA); J. SEVERYNS, Eawag Swiss Federal Institute of Aquatic Science and Technology

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.

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (P)
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-IC/MS/MS was done in order to be able to analyze 17 compound as markers of drug uses (cocaïne, heroin, amphetamine, cannabis, their major metabolite and some substances 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 T1-2 days, T2-3 days, T3-4 days, T4-5 days, T5-6 days, T6-7 days, T7-8 days, T8-9 days, T9-10 days. Water samples were also daily collected. The first result of the calibration showed a good capacity of the POCIS to sample cocaine markers (cocaïne, benzoylecgonine, cocaethylene, ephedrine, methadone, and their corresponding markers in the WWTP influent) 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 T1-2 days, T2-3 days, T3-4 days, T4-5 days, T5-6 days, T6-7 days, T7-8 days, T8-9 days, T9-10 days. The calculated sampling rate (Rs) vary from 0.004 for benzoylecgonine to 0.2 L3/g for cocaine.

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. Bayerle, Luxembourg Institute of Science and Technology LIST

The pressure on surface waters that is exerted by emerging pollutants depends on the treatment processes, using a non targeted approach, as well as the potential for 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 organic pollutants: short chain chlorinated paraffins (SCCPs). This study examines emerging pollutants in the river network with segments at risk with basic population equivalent information.


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 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 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 analysis 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 develop capacity 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 routine analysis can be performed as the basis of this 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 organic pollutants: short chain chlorinated paraffins (SCCPs).

MO280 HilIC workflow strategy for the hidden target screening of very polar compounds in surface waters S. Veloutou, Technical University of Munich; S. Bieber, Technical University of Munich / Chair of Water / Urban Water Systems Engineering; S. Grosse, T. Letzel, Technical University of Munich

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 with various matrices. Both on the HILIC and the RPLC strength matrices than 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 quadrupole 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.
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.

MO281 Analysis of Per/Polyfluoroalkyl Substances (PFAS) in Drinking Water using LC/MS/MS to meet USEPA 537 requirements
T. Anamol, L. Toolewezi, T. Sosienski, Agilent Technologies, Waldbronn GmbH / UFZ; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research; S. Nzube, Cape Peninsula University of Technology / Department of Chemistry; V. S. Somerset, CPUT / Chemistry
Pharmaceuticals and personal care products (PPCPs) can be found in surface water and drinking water. It is important to increase the emphasis on the characteristics of the benzodiazepines in order to differentiate them from industrial chemical residues in fish matrices. This was done on a triple quadrupole mass spectrometer that is fully stackable with the HPLC system.

MO282 Development of a LC-MS/MS-based method for 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. Baven, McGill University / Singapore-Delft Water Alliance
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 low signal-to-noise complex matrices, and, 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 detect/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.

MO283 Prioritising site-specific emerging contaminants in surface water based on LC-HRMS nontarget 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
Non-targeted screening of non-trace contaminants is essential for the prevention 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
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. Schirau, Oregon State University / Environmental and Molecular Toxicology; S.L. McRoy-Simonek, 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 (USEPA) to list 16 PAHs as priority pollutants. PAHs can be metabolized by mammals and some microbes to form hydroxylated PAHs (OH-PAHs) 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 in 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 proven to be successful in the identification of TPs in complex environmental samples. An HPLC-HRMS method was developed for analysis of phenanthrene TPs detected in bioremediated water. C18, phenyl-hexyl, and fluoro-phenyl HPLC columns were evaluated for their ability to resolve hydroxypbenanthrene (OH-Ph) isomers. Baseline resolution of 2-, 4-, and 9-OH-Ph was achieved with the C18 and phenyl-hexyl columns using a gradient of water/methanol (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 three potential solvent composition of the eluent enabled optimal ESI performance. Consequently, spectrometric sensitivity was preserved throughout each analysis. Further investigation will determine whether the fluoro-phenyl column is suitable for separation of OH-Ph 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

Transformation products (TPs) are formed in the water cycle through both biotic and non-biotic processes. Data available showed that TPs in complex environmental samples. An HPLC-HRMS method was developed for analysis of phenanthrene TPs detected in bioremediated water. C18, phenyl-hexyl, and fluoro-phenyl HPLC columns were evaluated for their ability to resolve hydroxypbenanthrene (OH-Ph) isomers. Baseline resolution of 2-, 4-, and 9-OH-Ph was achieved with the C18 and phenyl-hexyl columns using a gradient of water/methanol (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 three potential solvent composition of the eluent enabled optimal ESI performance. Consequently, spectrometric sensitivity was preserved throughout each analysis. Further investigation will determine whether the fluoro-phenyl column is suitable for separation of OH-Ph isomers. The method will be used for separation of phenanthrene metabolites and other PAH TPs in non-targeted screening of bioremediated mixtures.

MO288
Application of high-resolution mass spectrometry to identifying chlorinated transformation products of aromatic emerging contaminants in wastewater
X. Lin, TUNGHAI University; W. Chen, J. Cheng, TUNGHAI University / Department of Environmental Science and Engineering

Chlorination could remove some aromatic emerging contaminants (ECs) in wastewater, but may also convert the ECs into unknown transformation products (TPs). This study developed a method to systemically identify the TPs of multiple aromatic ECs using high-resolution mass spectrometry (HR-MS) and traced the parent aromatic ECs of the TPs. We spiked ten aromatic ECs (5000 ng/L) into 100-mL Milli-Q water. The water was chlorinated at an initial chlorine of 0.7 mg/L for ten minutes. The full-scan mass chromatograms of both the chlorinated (n = 6) and the untreated (n = 6) water samples were acquired using ultra-performance liquid chromatography-quadrupole-time-of-flight mass spectrometry. By comparing the compound profiles, we evaluated the removals of aromatic ECs and discovered several signals of suspect TPs. We then also characterized the transformation of the TPs using database searching and isotope-pattern comparison. The parent aromatic ECs of the TPs were then traced back by spiking each aromatic EC to one 100-mL Milli-Q water. Eight of the aromatic ECs were partly removed by chlorination, where triclosan showed the highest removal (99.4%), followed by bisphenol A (72.5%). Nine of the features that were present in the chlorinated and absent in the untreated water samples were identified as TPs. The results of database searching and isotope-pattern comparison showed that the molecular formulae of all of the nine TPs contain at least one chlorine. Each chlorinated TP was then successfully traced to one aromatic EC. The nine TPs were transformed from five aromatic ECs, including all of the four parabens and triclosan, by replacing one or two hydrogens with chlorine atoms. The HR-MS method successfully identified nine chlorinated TPs. The results of this study demonstrated that parabens and triclosan could be transformed into more persistent, bioaccumulative, and toxic chlorinated compounds. The proposed method will be applied to the systemic identification of TPs in real water samples containing multiple ECs.

MO289
Unravelling the potential of a partial nitritation/anammox biomass towards micropollutants biodegradation
K. KASOTAKI, ICRA (Catalan Institute for Water Research) / Technologies and Evaluation; M. PIJUN, Catalan Institute for Water Research ICRA / Technologies and Evaluation; I. Gusmârde, Catalan Institute for Water Research ICRA; I. RODRÍGUEZ-RODA, Universitat de Girona and ICRA / LEQUiA; A. Joss, Eawag Swiss Federal Institute of Aquatic Science and Technology; G. Buttiglieri, Catalan Institute for Water Research ICRA

In the past few years, anamox-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/anamox ammonium oxidation (PN/A) pilot plant towards five pharmacologically active compounds (ibuprofen, sulfinamethoxazole, methoprolol, venlafaxine and carbamazepine). Batch experiments were performed under different conditions of carbon supply by selecting different microbial groups: i) regular PN/A operation, ii) aerobic (optimal for nitrifying bacteria), iii) aerobic conditions with allylthiourea (an inhibitor of anamox oxidizing bacteria), iv) anoxic (optimal for anamox bacteria), v) aerobic with acetate (optimal for heterotrophic bacteria) and vi) anoxic with acetate (optimal for heterotrophic denitrifying bacteria). Ibuprofen 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.
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 pharmaceuticals. Bifenthrin, nitrofuran and isomerel were attributed to co-metabolism (enhanced acetate). Metoprolol, isomerel, diclofenac, propranolol and sulfamethizole 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 indicate the occurrence of oxygenated, which could be considered as catabolism. Biofilm reactors can be employed for polishing treated wastewater, and the addition of primary carbon source can enhance the bioconverter’s performance.

M0291 Investigating inhibitory effect of anti-inflammatory pharmaceuticals on activated sludge metabolism


The consumption of pharmaceuticals increases annually due to a variety of reasons including affordability, population ageing and population growth. Due to the obstacles in monitoring all these micropollutants regularly, focusing on maximum degradation rate (Kd) can be physically attained in ecosystems, 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 sludge 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 S192) (600 mg COD/L) and wastewater from the Central wastewater treatment plant of the city of Istanbul. The removal rate of opioid analgesics was systematically studied as a secondary effluent from the Central wastewater treatment plant of the city of Zagreb. The removal of opioid analgesics was associated with formation of two main transformation products characterized by structures of 250 and 280 for tramadol and 278 and 294 for methadone. The most abundant transformation products of tramadol and methadone were tentatively identified as tramadol N-oxide and EDDP, respectively.

M0292 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. Cochrans, B. Fryer, University of South Carolina; S. Kimura-Hara, University of Calagary; W. Abdelraheem, Y. Huang, University of Cincinnati; S.L. Coffin, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California, Riverside / Department of Environmental Chemistry and Toxicology; S. Richardson, University of South Carolina

Potable reuse of wastewater is becoming more common as populations increase and freshwater resources become more scarce. Producing safe drinking water from treated wastewater is challenging due to the presence of contaminants that are not removed during conventional treatment. The removal of pharmaceuticals and transformation products that can be formed with advanced oxidation technologies (AOTs) that are used in potable reuse treatments. These contaminants can be harmful for human and ecological health. In 2013, two Science Advisory panels determined two lists of priority emerging contaminants (ECs) to be monitored in aquatic ecosystems and human potable water reuse. The ECs were determined based on their toxicity, persistence in the environment, and potential to be transformed into other water concentrations. This project is investigating the removal and/or transformation of 21 of these priority ECs through UV/CH2O, microfiltration and reverse osmosis in three samplings from Full-Scale Advanced Wastewater Treatment Plant (Orange County GWRS) in order to make indeterminate potable reuse of wastewater safer. Ultra performance liquid chromatography tandem mass spectrometry (UPLC-MS/MS) was used to quantify bisphenol A, p,p'-dichlorobenzene (PCB), bis (2-ethylhexyl)phthalate, butylbenzyl phthalate, perfluorooctanoic acid (PFOA), perfluorooctane sulfonate (PFOS), diclofenac, ibuprofen, erythromycin, triclosan, 17α-ethinylestradiol, 17β-estradiol, estrone and chlortyphillin. Gas chromatography mass spectrometry (GC-MS/MS) was used to quantify permethrin, galaxolide (AOTs) and polybrominated diphenyl ether (PBDE)-47 Scalphen, D.Dioxynil, Diclofenac (PBDE)-98, bifenthrin and N-nitosodimethyl-amine (NDMA). Transformation products (TPs), disinfection by-products (DBPs), and unknown compounds are being identified via high resolution-time-of-flight GC/ToF/MS/MS and UPLC/Q-ToF/MS/MS. Some compounds were not removed even after UV treatment. Controlled laboratory chlorination/bromination experiments have been conducted in order to elucidate the fate of pharmaceuticals and transformation products from water treatment and wastewater disinfection, and many TPs and DBPs were identified, including chlorine- and bromine-containing-by-products. Toxicity studies on these reacted samples were also done, and the results show that many of the TPs are more cytotoxic after being chlorinated/brominated. Mass spectra obtained from these identified TPs and DBPs are being added to a user library for use in determining TPs in our sampled waters.

M0295 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. Silwana, Durban university; M. Makombe, Cape Peninsula University of Technology / Chemistry; E. Iwohua, University of The Western Cape / SensorLab Department of Chemistry; V.S. Somerset, CUT; / 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 increasingly constrained. The view of large-scale wastewater reuse for 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 chitosan bimetallic iron-silver nanoparticles (CS/Fe-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 quantities for urban areas — 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 Challenges for a changing Europe)”. Project objectives: (i) to evaluate and compare treatment processes with combined analytical, toxicological and microbiological approaches; (ii) to evaluate advanced treatment options in a multiple barrier approach to improve removal 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) from both biological and physical 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 removals for advanced IPR processes were developed. Results will inform on the feasibility of using sewage biomarkers for future compliance monitoring. Metabolites from the following phthalates are considered for IPR: benzyl butyl phthalate (BBP), dibutyl phthalate (DBP), di(2-ethylhexyl) phthalate (DEHP), dibutyl phthalate (DBP), diisobutylphthalate (DIBP), di-2-ethylhexyl phthalate (DEHP), diisononylphthalate (DINP), and diisodecylphthalate (DIDP). This project 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 Phtalates 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 Biological Sciences; A.L. Macken, K. Petersen, S. Samanipour, 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.

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 plasticizers 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, tubes 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, effluent and sewage sludge (biosolids) are being monitored by GC-MS and LC-MS/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 for sewage epidemiology: benzyl butyl phthalate (BBP), dibutyl phthalate (DBP), di(2-ethylhexyl) phthalate (DEHP), diisobutylphthalate (DIBP), di-2-ethylhexyl phthalate (DEHP), 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.
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 / UMR 5805; H. Budziński, University of Bordeaux
This study proposes to identify the origin of 30 poly- and perfluoroalkyl substances (PFASs) found in the sewage system of Bordeaux. It is therefore the objective of this study to identify 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 sources 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, benzotriaizole, ciprofloxacin and fluconazole. QACs and chlorhexidine were effectively removed by the primary clarifier, 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 fluconazole, 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 CPC 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.

MO301 Herbicides and fungicides in watersheds 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
This study proposes to identify the origin of 30 poly- and perfluoroalkyl substances (PFASs) found in the sewage system of Bordeaux. It is therefore the objective of this study to identify 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 sources 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, benzotriaizole, ciprofloxacin and fluconazole. QACs and chlorhexidine were effectively removed by the primary clarifier, 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 fluconazole, 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 CPC 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.

MO302 Antibiotics and endocrine disrupting compounds in wastewater treatment plants and in receiving water bodies around the city of Rome (Italy)
Several studies highlighted the occurrence of organic micropollutants such as pharmaceuticals residues, ingredients of personal care products and endocrine disrupting compounds in wastewaters from different municipalities. 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 methods. This method, which was applied for the first time to domestic wastewaters and to WWTP sludges, revealed the presence of unidentified precursors of C16-C30 PFCA as well as the presence of trimethoprim and fluconazole, 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 CPC 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 The continuous release of these xenobiotics into the aquatic environment may lead to chronic exposure of organisms at all levels of the food chain.
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 particulate 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 ecotoxicological assessment

M. Andrees, Universitat de Valencia / Environmental and Food Safety Research Group; J. Pascual-Aguilar, 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 near of the city with highest population densities were selected 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 (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). Each compound’s information was then used in GIS modeling to perform the risk assessment. The work performed contributed to evaluate the risk associated with changes in the water treatment process and prevent population exposure to DBPs in the event of scenarios that may alter the good performance of the whole process.

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**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) could be detected in the water at concentrations as low as 0.1 ng/L of 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 ozonization or nitrosation. Multiple mechanisms can be involved in NDMA formation: a) reaction between nitrite or nitrogen dioxide and secondary ammonium ions and/or amines; b) reaction of typical secondary amine precursors; c) chlorination of nitrite in the presence of nitrosamine precursors; and 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.

**MO311**

Presence and environmental hazard of psychoactive pharmaceutical compounds in coastal waters and biota from North-Western Spain.

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The study 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 72h 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-nitrosamines (4THMs), trihalogenated haloacetic acids (THALs), halogenated acetonitriles (HANS), halogenated acetamides (HACMs) and haloacetic acids (HAAs). Overall it could be concluded that the potential of the water entering the plants to form all investigated DBPs decreased throughout the treatment process, due to the removal of DBP precursors in the different treatment steps. The work performed contributed to evaluate the risk associated with changes in the water treatment process and prevent population exposure to DBPs in the event of scenarios that may alter the good performance of the whole process.

Acknowledgments: This work has been financially supported by the Government of Catalonia (Consolidated Research Groups 2014 SGR 418-Water and Soil Quality Unit 2014 SGR 291-ICRA).

**MO309**

Occurrence, fate and environmental risk assessment of benzophenone-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 benzophenone 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 runoff and indirectly from sewage 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 compounds were investigated in four sampling points (mean of 4.67 ng/L, ranging from 2.34 to 7.21 ng/L) and BCG 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 BCG 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 the activities of the city with highest population densities, according to the descriptive model of territorial presence. Compounds used as drugs of abuse and prescribed as pharmaceuticals (MET, COD and EPH) were mostly detected in Valencia city and its metropolitan area where most hospitals are located. Although risk assessment showed low ecotoxicological hazard, further studies are also needed in order to assess long term toxicity.

**MO308**

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 benzophenone 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 runoff and indirectly from sewage 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'-tetrabhydroxybenzophenone (BP-2), 2-hydroxy-4-methoxybenzophenone (BP-3), 2,2'-dihydroxy-4,4'-dimethoxybenzophenone (BP-6)), 2,2'-dihydroxy-4-methoxy-benzophenone (BP-8), 4-hydroxybenzophenone (4OH-BP) and 4,4'-dihydroxybenzophenone (4DHBB)) 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 benzophenone concentrations varied from widely < LOQ to 2774 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 amount 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 degradation of BP-3 in the field. This will be followed by a preliminary risk assessment.
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 has increased remarkably. Our work represents the first attempt at monitoring 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 in the AMPF FH was found to be the Alam River (a tributary of the Seine River situated in the East part of Paris) at 4 sites characteristic of agricultural and urban areas. Water was also sampled for analysis to compare concentrations in sites contamination. Bile is an ideal material to identify metabolites of pollutants. This biological fluid was taken directly from the gallbladder with a syringe on freshly euthanized crab and frozen for further analysis. Then, 100µL of bile was taken to evaluate the concentration of 13C-AMP in addition to 13C-AMP added before extraction with milliQ water (5mL) and ultrasonic method for 30 minutes. The extract was then derivatized (FMOC-Cl) and concentrated on SPE Oasis HLB cartridge (60cc) before LC MS MS analysis. Preliminary tests were performed to establish and validate the protocol and to find the lowest limit of quantification and the best reproducibility. Results showed that glyphosate is detected in a fish sample coming from the most contaminated site by AMPF. This suggests that glyphosate is assimilated in fish and is still detectable after glyphosate has been degraded to AMPA in the water river. Glyphosate content in fish could be a risk assessment based on the measured tissue concentrations is underway. Our results confirm the presence of PAs in coastal waters of the Rias Baixas area in concentrations potentially able to cause chronic effects in exposed organisms, as well as the presence of these compounds in biota inhabiting the area. Based on the results obtained further monitoring of venlafaxine and citalopram in coastal waters is recommended.

M0314
Pycoactive compounds in mussels: analytical method development and occurrence assessment
E.L. Garcia, IDAEA-CSIC / Department for Environmental Chemistry; C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; M. López de Alda, Institute of Environment and Water Research IDAEA-CSIC / Department of Environmental Chemistry

It is well established that wastewater treatment plant effluents release pharmaceutical compounds to the aquatic environment impairing water quality. The environmental presence of these compounds may cause negative effects in the exposed aquatic organisms. In order to evaluate the ecological risk that they may pose, it is relevant not only to analyse the 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 attenuation of emerging pollutants in the agricultural environment and therefore inform on the level of treatment necessary to undertake this practice sustainably.

Glyphosate is a widely used herbicide. In France, it is used during intercultural operations and to control weeds in non-agricultural areas. Glyphosate and AMPA (its main degradation product) can reach high concentrations in rivers, especially in the Seine basin including the Paris suburb (France). 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 club (Squalius cephalus). This method was developed thanks to the Spanish Ministry of Economy and Competitiveness FIS (PI14/00516) and the European Regional DevelopmentFund (ERDF).

M0312
Detection of glyphosate and AMPA in fish bile from the Marne River, France
H. Blanched, EPHE UMR 7619; T. Ferreux, F. Alliot, EPHE / UMR Metis; A. Goutte, UMR METIS EPHE

Glyphosate is a widely used herbicide. In France, it is used during intercultural operations and to control weeds in non-agricultural areas. Glyphosate and AMPA (its main degradation product) can reach high concentrations in rivers, especially in the Seine basin including the Paris suburb (France). 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 club (Squalius cephalus). This method was developed thanks to the Spanish Ministry of Economy and Competitiveness FIS (PI14/00516) and the European Regional DevelopmentFund (ERDF).

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

M0315
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 Ecology; M. Simon, N. van Alst, K.B. Olesen, F. Liu, J. Vollertsen, A. Vianello, F. Alliot, EPHE / UMR Metis; A. Goutte, UMR METIS EPHE

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 environmental matrices commercial FTIR systems. 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
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 results can be further refined to define particle boundaries. 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 verified by the JPI Ocean project F1BASEMAN. 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 is performed 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 from 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 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 fraction 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. Storseth, L. Sorensen, K. Almaas, SINTEF Ocean / Environmental Technology; M.O. Høynes, Norwegian University of Science and Technology; O. Brakstad, 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 FTIR fingerprint spectra. As a result, an automated MP identification and quantification 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. This is partially due to an unattained 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.
MO320 Optimization of the preparation of standards of high density polyethylene microplastics and quantification techniques by stereoscopic and confocal microscopy.
J. Kang, Korea University; J. Perales, CACYTMAR University of Cadiz / Department of Environmental Technologies; E. Rojo-Nieto, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; J. Gonzalez-Leal, University of Cadiz
A non-complex procedure has been developed for preparing HDPE microplastics as standard for microplastic determination in sediments. Always keeping enough of the same plastic in mind, two mesh sizes were used 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). For this purpose, 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 ethanolic (96%) ethanol 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 disaggregation of small standard particles. HDPE microplastics were suspended in ethanolic 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 in which microplastics (< 5 mm) were detected in the Arctic (Kidd et al., 2016). 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. This area can be particularly important because it links Arctic marginal seas (Chukchi sea, East Siberian sea, etc.) linked to the Bering strait has never been studied. To provide a more comprehensive picture of microplastics in the Arctic, we conducted an annual surveillance in every summer since 2016 in the Pacific-side Arctic Ocean, which is regarded as a global hot spot of plastic pollution. The goal of this work is the development of a systematic protocol for sampling, sampling pre-treatment and analysis of MPs in terrestrial samples, which ends up in a fast, quantitative method. Up to now, various studies about the analysis of MPs at beaches, in marine sediment and along rivers are available. In these studies, as sample pre-treatment density separation techniques were used to reduce the inorganic matrix. In some cases, a chemical oxidation step to minimize the organic matrix is additionally carried out. FTIR or Raman spectroscopy were used to analyse the sample with enriched concentrations of MP. Using these techniques, only small proportions of inserted samples were analysed. Similar investigations techniques were used for terrestrial samples. The few existing studies investigated sewage sludge, compost fertilization or areas of intensive utilization of plastics foils in agriculture or from recycling of plastics. However, a systematic study on MPs in the Pacific-side Arctic Ocean has never been carried out.

MO322 Microplastics in Expanded Global Table Salt Product Samples and its implication
J. Kim, Incheon National University / Department of Marine Science; C. Kim, Greenpeace East Asia; S. Kim, Incheon National University / Department of Marine Science
Microplastic pollution is becoming a global issue in marine environment pollution. Among the various environmental media, some recent studies have identified microplastic contamination that remains in salt (sea salt, lake salt, rock salt). As salt is an essential human/animal food-item, microplastic contamination in salt is considered to be an important matter of environmental and public health concern. The goal of this study is to elucidate any relationship of microplastic contamination between sea salt and seawater and to calculate the human exposure of microplastics resulting from the consumption of commercially available salt products. To do this, we purchased and analyzed the salt samples sold in 17 countries (8 countries in Asia, 7 in Europe, 1 in Africa and 1 in North America) in four continents. Each salt sample was selected in consideration of the salt production area, production method, and salt consumption pattern of each country. Total 37 salt samples were analyzed, including sea salt, lake salt and rock salt. Each sample was duplicated (n = 2) and two blank samples were analyzed for each batch to check contamination during the analysis. Size, color, polymer, and shape of each microplastic were determined using microscopic and spectroscopic analyses (FTIR). Thousands of microplastics were detected per 1 kg of the salt samples of this study, and the predominant forms were fragment and fiber, which were frequently detected in the order of PP > PE > PET. Significant correlation was observed between microplastic discharge rate via the rivers near the sea-salt production and microplastic contamination in the sea-salt. After further analysis, human exposure, characteristics of microplastic distribution, and application of sea-salt as an alternative monitoring medium will be announced. Car tire wear is estimated to represent a major fraction of microplastic pollution in the environment. Rubber particles are transported by 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 critical 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 (1.5—2.9% ThOD), while no methanogenic activity 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 in activated sludge. Noteworthy, the efficiency of rubber on microbial activity was observed. PLGA as positive control did not encourage 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 found 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 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 reefs or recirculating systems in aquaculture and organics. Although tire rubber 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 reefs or recirculating systems in aquaculture. 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. Lauter, 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. Alonso-Nole, O. Perales-Perez, F. R. Roman, Desalin. Water Treat. 2012, 49, 296. [7] M. Abraham, A. Ibrahim, A. Zissimos, 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 (Palaemonetes pugio): microrubber tire particles and microplastics

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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 prepared TWP in adult grass shrimp (Palaemonetes pugio) and compare this to that of 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 (1.9 x 10^7 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 V. cholerae non-O1 (CT) (CFU/shrimp). 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 reach 50% was determined for the bootstrapped mean of the observed gill clearance rate at 43.0±13.8 hours. Gut clearance for the TWP was 25.2±3.0 hours. Within the gill chambers 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 than 21.2±2.1 hours. Mortality in these assays ranged from 0%—55% with microplastic spheres and fragments under 50 µm not acutely toxic. All sizes of TWP were not acutely toxic to both size fractions tested (34 and 93 µm) with mortalities of 55 and 35%, respectively. Results from the present study suggest that wet prepared TWP are less acutely toxic than that of other synthetic particles, especially fibers.

**MO327**

Acute and chronic toxicity of microrubbed 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 tred will 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 microrubbed 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), sulphur (in rubber) 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 conceived to determine the toxicity of tire rubber particles to *Hyalella azteca*, a 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 disint 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 *Hyallela azteca* and chemical analysis of rubber particles and leachate - comparison of pristine microrubbed tire to previous data on worn car tire particles

L.L. Heller, 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 pollution in the environment. Currently, little is known about the ecotoxicology of MR. The aim of the present study was to conceived to determine the acute and chronic toxicity of MR. The wealth of research on the impacts of microplastics (MPs), there is little on microrubbed 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), sulphur (in rubber) 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 conceived to determine the toxicity of tire rubber particles to *Hyallela 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 disint 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.
microplastics and co-contaminants in marine biota

C. Lanctot, International Atomic Energy Agency / Radioecology Lab; M. AL SID CHEIKH, University of Plymouth / Marine sciences and engineering; A.I. Catarino, Heriot-Watt University / Institute of Life and Earth Sciences; T. Cresswell, ANSTO Environmental Research / Institute for Environmental Research; B. Danis, Université Libre de Bruxelles; T. Mincer, Woods Hole Oceanographic Institution; F. Obehrens, P. Swarzenski, International Atomic Energy Agency / Radioecology Lab; I. Tolosa, International Atomic Energy Agency; H.K. Karapanagioti, University of Patras / Chemistry Department; M. Metian, IAEA-EL / Radioecology Lab

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-organisms 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 Radioecology 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 the (1) the biokinetics, bioaccumulation and toxicokinetics 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., across epithelial membranes/tissues), and if they can act as a vector for contaminant transport in the marine environment under low exposure. This poster will provide an overview of the techniques used to address these questions, as well as preliminary outcomes and future directions.

MO330
Aggregation kinetics of plastic nanoparticles in fresh and marine phytoplankton culture media

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Release of plastics debris in the environment has been catching more and more concern in recent years, particularly 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 medium, whereas (1) the biokinetics, bioaccumulation and toxicokinetics 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., across epithelial membranes/tissues), and if they can act as a vector for contaminant transport in the marine environment under low exposure. This poster will provide an overview of the techniques used to address these questions, as well as preliminary outcomes and future directions.

MO333
Influence of biofilm composition on mercury bioaccumulation


In aquatic systems, the dominant lifestyle of microorganisms (bacteria and microbialagagles) 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 IHg (~ 100 pm, 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 DNA abundance of 16S rRNA gene) was determined as well as EPS and mercury EPS composition. 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 cystine 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 evansional 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) is extensively recognised for its toxicity, mobility and bioaccumulation potential. In coastal areas, the 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 conducted at Idrija (western Slovenia) for approximately 500 years, whereas the second is the result of discharge of a chlor-alkali plant effluents. Fish farming is a
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 CO2-stirred water-bag sampler (Lumex-LA 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 measurements, the background levels of gaseous elemental mercury were determined together with the main chemico-physical parameters influencing Hg behaviour. This new insights will be of help for future estimates of Hg mass balance in one of the most contaminated areas in the Adriatic Sea. Keywords: atmospheric mercury; mercury fluxes; fish-farm; Grado Lagoon

MO335 Atmospheric mercury assessment: a contribution to global monitoring and effectiveness evaluation within the Minamata Convention A. Fino, Italian National Research Council - Institute of Atmospheric Pollution Research (CNR-IAIA); F. Sprovieri, A. Macagano, E. Zampetti, P. Papa, G. Esposito, CNR Institute of Atmospheric Pollution Research Italy; P. Nicola, Institute of Atmospheric Pollution Research (CNR-IAIA) 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 analyses in human 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 (GOSM)”. The aim of this project is to support the policy process in relation to the Minamata Convention implementation. CNR-IAIA proposed a selection of monitoring sites, mostly background sites but also including those highly impacted, to undertake passive sampling and analysis of Hg in ambient air in order to strengthen capacity to provide globally comparable data. The poster gives an overview of preliminary results coming from the pilot survey campaigns carried out with mercury novel passive sampling.

MO336 Assessment of Hg impacts on mountain river ecosystems S. Le Faucheur, Institute F.-A. Forel, University of Geneva / Département F.-A. Forel des sciences de l'environnement et de leau; F. Pierron, Université de Bordeaux / UMR EPOC CNRS 5805; C. Moinecourt, Université de Genève; G. Daffe, University of Bordeaux / UMR EPOC CNRS 5805; A. Boullémont, RioTinto Mountain rivers are high-flow systems which can experience, even daily, high water height variations due to the presence of dams along their courses. These conditions limit the use of water and sediment analyses to identify pollution point sources through the use of elemental mercury 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 (GOSM)”. The aim of this project is to support the policy process in relation to the Minamata Convention implementation. CNR-IAIA proposed a selection of monitoring sites, mostly background sites but also including those highly impacted, to undertake passive sampling and analysis of Hg in ambient air in order to strengthen capacity to provide globally comparable data. The poster gives an overview of preliminary results coming from the pilot survey campaigns carried out with mercury novel passive sampling.

MO337 Mercury Photo-reduction and Total Photoreducible Mercury Dynamics in the Lakes of Keijmukijn National Park, Nova Scotia N.J. O'Driscoll, Acadia University / Department of Earth and Environmental Science; T. Christensen, T. Verre, Nova Scotia Department of Natural Resources; S. Klapstein, E. Mann, Acadia University Photo-reduction and photo-oxidation are fundamental mechanisms controlling mercury volatilization and accumulation in freshwaters. In all surface waters dissolved gaseous mercury (DGM) is produced as a net result of the reduction of reducible mercury, which is believed to be primarily divalent mercury (Hg(II)) bound to specific carbohydrate ligands, and the oxidation of elemental mercury (Hg(0)). These two processes control the amount of DGM available for evasion across the water-air interface; however, determination of the fundamental rate constants and mechanisms of these reactions in freshwaters are still areas that require more research. In particular, the total amount of photoreducible mercury is emerging as a key variable that requires more exploration. Here, we review the present knowledge of the group of reactions that take place over the Hg-DGM pool, the rate constants as well as temporal dynamics in total reducible mercury derived from two recent projects that examined water samples from a series of freshwater lakes in Keijmukijn National Park, Nova Scotia, Canada. We examined the hypothesis that gross photoreduction and photooxidation rates would be significantly different in lake water. Another hypothesis was that the amount of mercury available for reaction with solar radiation (i.e. reduction of Hg(II) to gaseous Hg(0)) in surface waters would significantly change over a summer. A Luzchem photo-reactor was used to irradiate 200 mL water samples in quartz beakers continuously exposed to ultraviolet radiation for 24 h with concurrent Hg(0) analysis to derive pseudo-first order gross reduction rate constants and batch experiments were used to derive net reduction rates (and gross photoreduction rates) by difference. These results showed that the net photo-oxidation rates for freshwaters were low, with mercury reduction and oxidation reactions very close to being in balance. We also found that the amount of total reducible Hg(II) changed significantly in three of the lakes over several sampling months. Dissolved organic carbon concentration was a key factor positively correlated with these results. This research provides the first quantitative measurements of gross photoreduction and photooxidation rates as well as total photo-reducible mercury over a season in surface freshwater lakes.

MO338 Influence of Avian Biovectors on Mercury Speciation in a Wetland J. Kickbush, Acadia University / Biology; M.L. Mallory, Acadia University / Biology; J. Murphy, Acadia University / Engineering; S. Klapstein, Acadia University / Earth & Environmental Science; A. Loder, Acadia University; N.M. Hill, Fern Hill; N.J. O'Driscoll, 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 (Goefcheld 2003). As such, it is a significant environmental contaminant. However, the contribution of mercury to wetland systems is currently unknown. We are using the Peregrine Falcon as an indicator species to examine mercury speciation in this environment. The Peregrine Falcon is a modern day counterpart of the ancient tethered falconry birds. The Peregrine Falcon is a large bird of prey, with the ability to dive up to speeds of 300 km/h. The population of Peregrine Falcons in North America has recovered from near extinction in the early 1960s to over 2000 birds in 2006. The recovery of the Peregrine Falcon is an indicator of the recovery of the larger wetland ecosystem. This study will examine mercury speciation in the wetland ecosystem in the Bay of Fundy. Specifically, we will examine the relationship between the concentration of mercury and the concentration of methyl mercury in the diet of the Peregrine Falcons. We will also examine the relationship between the concentration of mercury and the concentration of methyl mercury in the diet of the Peregrine Falcons.
The reference dose (Rd) for MeHg is 0.003 mg/kg bw/day. The equivalent estimations for \( \text{MeHg} \) 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 (Majorca 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 published 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 habitually been compared. Except for HCB, the values were higher in the upper trophic level with statistically significant differences for \( \Sigma \text{DDTs} \) and \( \Sigma \text{PCBs} \) (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 \( \Sigma \text{DDTs} \), \( \Sigma \text{PCBs} \) 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 estimate for \( \text{MeHg} \) established by EFSA in 2012, 4 µg/kg bw. The results showed that the equivalent estimations for \( \text{MeHg} \), involving provisional tolerable weekly intakes of 1.3 µg/kg bw were six and three times higher than these provisional tolerable weekly intake 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 cardiovascular and cerebrovascular 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 (Rd) as recommendations to Hg intake. Some studies have been associating the fish consumption with the Hg bioaccumulation, in areas along the Mid-Atlantic Ridge (MAR) exposed to active hydrothermal fields. The Azores archipelago is located in the North Atlantic Ocean close to the MAR. The last fishery statistics for fish consumption per capita in the Azores archipelago show that, each Azorean consumed about 80 kg of fish per year being the region with the highest consumption of fish products in Portugal. This study is the result of a review of all published articles indexed in Web of Science that presented Hg concentration in the muscle for fish species captured in the Azorean Exclusive Economic Zone, and additional new data from fish obtained by recreational fish. The selection of species was based on the fish landing reports (1994-2015) of Azores Fisheries Statistics (SIGA). The total catches for the Azorean ports, at the almost 10000 tonnes per year of these commercial fish species are discharged. Despite low Hg levels in fish, every year the population of this area is exposed to more than 1500g of Hg via fish consumption. However, the species with the highest concentration of Hg are not always those that contribute to a higher human exposure. The fish species Mora moro exhibit higher values than the permitted for fish consumption and carcinogens fish species generally exhibit higher concentration of Hg than omnivores fish species. On the other hand, demersal fish species demonstrated higher Hg concentration than pelagic fish species. Finally, the target hazard quotient (THQ) is < 1 for all fish species, meaning that the level of exposure is lower than the reference dose, and indicating that the daily exposure is not likely to cause any negative health effects during a lifetime in the human population.

MO341 Mercury concentrations in black meat from the Gippsland Lakes, Victoria, Australia.
L. Medeiros, EPA Victoria / EPA Victoria; S. Balshaw, Department of Health and Human Services; R. Goudey, EPA Victoria

The Gippsland Lakes are a coastal lakes system in eastern Victoria, Australia. They represent a unique aquatic ecosystem of significant ecological significance (Ramsar classified) as well as supporting significant tourism, recreational and commercial fishing industries. The Lakes area has been subject to several contaminated monitoring investigations over the last 45 years that have identified sediment and soil samples with detectable mercury concentrations, often occurring screening levels set to protect the environment, aquatic plants and animals. The sources of mercury to the Lakes have been identified as having anthropogenic origins. Historic sources are gold mining and wastewater discharges from a paper mill. Ongoing sources of mercury include emissions from coal-fired power plants in the Latrobe Valley to the east of the Lakes and wood smoke from bushfires and planned burns. This study, conducted in 2015, aimed to determine if the concentrations of mercury in black meat had increased over time, and whether or not existing dietary advice issued nationally by Food Standards Australia New Zealand for the protection of consumer health against the effects of mercury in seafood, was appropriate for fish sourced from the Lakes. Three previous studies investigated the concentrations of Hg in fish from the Gippsland Lakes and indicate that levels have not increased over time with the exception of 1990. Comparisons 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 in fish caught in the Lakes since 1998. However, no sampling data were available from 1980 to 1998. 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.

MO342 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 Mexico City’s fish market (Central de Abastos) was conducted in 1992, with the objective of determining health risks due to non-intentional shark meat ingestion. Samples were bought in the fish market as fish for ceviche, quesadillas or soup, economical fillet, battered or breaded fish from sea bass, codfish, tilapia, red snapper among other species. Chondrichthyans universal oligonucleotides 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 contaminants: \( \Sigma \text{DDTs} \) and \( \Sigma \text{PCBs} \) (R=0.37, P<0.05). The results showed that the equivalent estimations for \( \Sigma \text{DDTs} \) and \( \Sigma \text{PCBs} \) (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 estimate for \( \text{MeHg} \) established by EFSA in 2012, 4 µg/kg bw. The results showed that the equivalent estimations for \( \text{MeHg} \), involving provisional tolerable weekly intakes of 1.3 µg/kg bw were six and three times higher than these provisional tolerable weekly intake in adults and children (7-12 years of age), respectively.

MO343 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. The main anthropic impact is related to the increase of mercury pollution due to emissions from coal-fired power plants in the southeastern part of the country. Mercury compounds are released into the environment in different ways: municipal wastewater, mining, fire burn, and sewage. Polychlorinated biphenyls (PCBs) and mercury (Hg) are toxic elements that act as indicators of anthropic contamination in the environment. In addition, these elements are persistent in the environment, bioaccumulate, interfere with several biochemical and physiological processes, and are associated with adverse health effects. The main objective of this study was to quantify the substitution of fish meat with shark meat in Mexico City’s fish market (Central de Abastos) to evaluate the risk due to non-intentional shark meat ingestion. Samples were bought in the fish market as fish for ceviche, quesadillas or soup, economical fillet, battered or breaded fish from sea bass, codfish, tilapia, red snapper among other species. Chondrichthyans universal oligonucleotides 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 contaminants: \( \Sigma \text{DDTs} \) and \( \Sigma \text{PCBs} \) (R=0.37, P<0.05). The results showed that the equivalent estimations for \( \Sigma \text{DDTs} \) and \( \Sigma \text{PCBs} \) (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 estimate for \( \text{MeHg} \) established by EFSA in 2012, 4 µg/kg bw. The results showed that the equivalent estimations for \( \text{MeHg} \), involving provisional tolerable weekly intakes of 1.3 µg/kg bw were six and three times higher than these provisional tolerable weekly intake in adults and children (7-12 years of age), respectively.
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 Paranaguí 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, Paraliparichthys brasiliensis and Isopisias parvipes) and marine mammals (Sotalia guianensis and Pontoporia blainvillii). To understand 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-VA), in the muscular tissue of the organisms. The results of $\delta^{15}$N varied from 6.4 to 13.8 % in Paranaguí and from 7.1 to 14.3 % in Cananéia, with a continuous enrichment among the trophic levels. Concentrations of Hg were significantly higher in Paranaguí (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 Paranaguí, but they were not significant, which indicates a trend of biomagnification of this element. Such a behaviour is expected since fish exhibit a succession of 1-2 RNA genes. By analyzing methylmercury production and microbial community composition, we have identified sulphate as being the main driver of both final concentrations of methylmercury and microbial community structure.

MO345
Use of green tea to reduce mercury and methylmercury bioaccessibility in raw and cooked fish
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Human exposure to mercury (Hg) and methylmercury (MeHg) increases in diets rich in high trophic level marine species, resulting in health related concerns through food chains. Nevertheless the overall concentration of Hg and MeHg detected in seafood does not always reflect the amount that will be available for absorption by human intestinal epithelium after the digestion process. On the other hand, several studies have highlighted the health benefits attributed to green tea consumption and their potential effects on reduction of bioaccessibility of contaminants. In this context, the aim of the present study was to assess the effect of green tea in Hg and MeHg bioaccessibility in raw and cooked marine fish species. Results demonstrated that total Hg/MeHg concentration in seafood does not reflect the bioaccessible fraction. Hg bioaccessibility in raw samples ranged between 60% (yellowfin tuna and black scabbardfish) and 37 % (European conger), with most species presenting a bioaccessibility below 50%. Moreover, after grilling, Hg/MeHg bioaccessibility significantly decreased in all species, ranged between 31% (yellowfin tuna) and 8% (Atlantic wreefish). The bioaccessibility of Hg and MeHg was also affected by the presence of green tea, decreasing the amount of these contaminants. In raw samples with tea, Hg bioaccessibility ranged between 47% (black scabbardfish) and 26% (swordfish); and between 18% (yellowfin tuna) and 7% (swordfish) after grilling. Green tea significantly decreased Hg/MeHg bioaccessibility in raw samples of yellowfin tuna, common smooth-hound and swordfish, as well as in grilled yellowfin tuna, common smooth-hound, atlantic wreefish and blue shark. Bioaccessibility variability may be explained by changes in the chemical composition of species during grilling and green tea catechins bioaccessibility, oncology are relatively unstable. This work clearly reveals that green tea is able to reduce Hg and MeHg bioaccessibility, leading to lower the risks associated with seafood consumption. Nevertheless, a better understanding of green tea bioaccessibility is needed, and how its protective effect affects other contaminants. Such information is certainly useful to help consumers to wisely select their food, and to enable food safety authorities to integrate this information in risk assessment and communication activities.

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 buried and unavailable for processes that can account for contaminant bioaccessibility from anthropic activities than Cananéa, and presented similar values to highly degradated 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.

MO347
Biological and Geochemical Drivers of Mercury Toxicity in Yellowknife, NWT, Canada
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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 Paranaguí, but they were not significant, which indicates a trend of biomagnification of this element. Such a behaviour is expected since fish exhibit a succession of 1-2 RNA genes. By analyzing methylmercury production and microbial community composition, we have identified sulphate as being the main driver of both final concentrations of methylmercury and microbial community structure.
The net production of MeHg is controlled by both mercury methylation (MeHg) and abiotic processes, such as degradation of organic matter. Using a direct quantification method (EPA 7473), it was studied the variation of total mercury (HgT) in the sediments of the riparian fern *Thelypteris hispidula*, sediments and water in three streams: El Socorro, El Roble 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 stream water and pore water of sediments studied. 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 sediment (rho = 0.764, p = 0.000). These results demonstrate the environmental effects caused by mining activities in protected areas in Colombia.

**MO349**

Temporal integration of diurnal variations of metals and mercury concentrations by passive sampling method in a highly polluted site on the Delé River, northern France

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Due to several metallurgical plants along the river, the Delé 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 voltammetry approaches allowing to measure Pb, Zn and Cd with 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 weighted 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 Delé 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 metals and Hg in the pore water (0.2 to 0.3 mg/kg, Pb and Cd (3 to 34 mg/L). The analysis of metals and Hg in SPMD showed increases of Pb, Zn, 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 SPMD.

Furthermore, the increase of SPMD 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 Hg²⁺ and CH₃Hg⁺. Finally, the interpretation of DGT measurements will show how well DGT integrate variations of inorganic contaminants concentrations during the exposure period.

**MO350**

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 methylating 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 sobh 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 Gunnnekelv fjord (GF) and Bergen Harbour (BH) – two heavily contaminated locations with long-lying Dredgings. Bulk concentrations were 25.5 and 9.5 mg/kg total mercury (THg) for GF and BH respectively. Two treatments were investigated: Activated anthracite char (AC) and activated biochar (BC). Treatments were set up for time series of 0, 1, 3 and 6 months in sealed glass jars, stored dark at room temperature. At each time series sediment and pore water was sampled. Additionally, pore water was examined using DGT with an argos 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 causes 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.

**MO351**

Baysian Human Health Risk Assessment of Almaden Mining Area

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Almadén, with the largest and richest known mercury deposit, is located in the south-west of Ciudad Real (Spain) with 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 ecosystem influences human health. The MANCHA project "MercOx - Metrology for oxidised mercury" (https://orcid.org/0000-0002-9166-1861) aimed to characterize mercury contamination in the mine water and sediments from different sites in the area, to develop biomonitoring and dietary guidance tools and to develop a global treaty to protect human health and the environment from the adverse effects of Hg. In addition to its elemental form Hg
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 transportable and reliable traceable Hg(II) standards is needed to solve the traceability problem that currently exists in the measurement of total mercury (Hg(0)+Hg(II)) 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 standards are required, as well as issues specific to the 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(0) on freshwater fish is lacking. The purpose of this study is to develop a physiologically-based toxicokinetic/toxicodynamic (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, muscle, intestine and muscle. The obtained toxicokinetic and physiological 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-specific burden of 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) accumulation in muscle would be below acceptable limits 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 (95% CI: 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 Hg in that bottom-feeding fish. We suggest that more dose-response data of sublethal and chronic effects are required to improve future risk assessment in a more comprehensive 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
H. Coelho Vieira, University of Aveiro; A.M. Soares, F. Morgado, University of Aveiro / department of Biology & CESAM; S. Abreu, University of Aveiro / Dep. Biology & CESAM
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 poisons 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 0.7 μg kg⁻¹ bw⁻¹ week⁻¹). Recently (2012), PTWI suggested by JECFA for MeHg was revised by the European 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 is relatively high (……) and compares these Hg concentration with the maximum levels of Hg for certain contaminants in foods 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 iscores 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 individuals 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 in scenario tests. Because of the availability of many new and new GUTS implementations should be trained using this ring-test and refer to these results as benchmark. Any new user should run the test ring the 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 from standard studies to underestimates for insensitive 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. Mechanistic 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 during the first two 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 account for species differences in feeding. We suggest 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.

MO358 TK-TD modelling as additional line of evidence in the risk assessment for aquatic macrophytes - chlorotoluron as a case study
J. Klein, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecotoxicology; U. Hommen, Fraunhofer IME; G. Weyman, ADAMA
To assess effects of the use of plant protection products based on chlorotoluron as active substance, various designs of laboratory tests with Lemna spp. and other species and also mesocosm studies including different macrophyte species are available. Since it is not possible to conduct long-term feeding experiments to model long-term exposure 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 Lemna 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 temperature and illumination, which is typical for Lemna species and other aquatic plants. 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 fresh 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 efficiencies 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 results for field tests were in very good agreement with the modelled endpoint, which proved the usefullness of modelling as additional tool in risk assessment of plant protection products, particularly for extrapolation between scenarios which cannot all be tested experimentally.

MO359 TK/TD modelling as a tiered approach to reveal interspecies variability of toxicity in fish
F. Gabsi, Rifcon GmbH; T. Preuss, Bayer AG / Environmental Safety
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 HC50s) for a scenario with variable exposure were predicted for a SSD-sensitivity test 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 model) 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 inputs for the SSD calculations. The SSDs were derived by fitting probability distributions to the LC50 data, and the corresponding HC50s were determined. The analysis was performed separately for two compounds. Results with both toxicants revealed that the sensitivity ranking for the fish species and consequently the HC50 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 HC50s. 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.

MO360 RIFCON EasyGUTS: Ready-to-use and freely available software for TK/TD modelling of survival
D. Nickisch, O. Jakobi, A. Mediancve, Rifcon GmbH
GUTS (General Unified threshold model of Survival) is one of the most commonly used models 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. The 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 EasyGUTS package 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.

MO361 A new test design to inform TKTD models on species sensitivity
E. Breuss, Bayer AG; Division Bayer CropScience / Ecotoxicology; K. Kuhl, Bayer AG - CropScience Division; J. Hager, Bayer Ag; T. Preuss, Bayer Ag / Environmental Safety
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 population assessment for plant protection products for aquatic organisms in edge-of-field surface waters states TKTD 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 test scenarios. Particularly in chronic exposures with low food conditions / feeding impairment over long term 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 exposure 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.

MO362 Impact of temperature on species sensitivity distribution in aquatic invertebrates
K. Ladermann, S. Classen, Research Institute gaiac; T. Strauss, M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; A. Gergs, Bayer AG - Crop Science Division / Department of Environmental, Social and Spatial Change
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. Apparent toxicity endpoints, such as the LC50s, have been reported to depend on ambient temperature and species interactions in aquatic invertebrates and fish species. 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 *Aecipenser 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 toxicology 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 (*Aecipenser 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 (EC50 values) for population as alternative tools in ecological risk assessment. In this context, lethal toxicokinetic-toxicodynamic (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 for *Daphnia magna* is built as an individual-based 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-toxicodynamic framework
K. Vlaeminck, Arche consulting / GhEnToxLab; S. Heine, Bayer AG / Effect modelling; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; T. Preuss, Bayer AG / Environmental Safety

Recently, several TK/TD population modelling approaches have been developed and are applied in different risk assessment areas. The European Food Safety Authority (EFSA) uses TK/TD models for aquatic organisms in edge-of-field surface waters, states TK/TD population modelling as an approach for the aquatic risk assessment. Although, the EFSA aquatic guidance states TK/TD population modelling as a method for the risk assessment, there is a lack of guidance and practical experiences for this new technique – especially to what extend the environmental scenario in which a TK/TD population model is applied influences the outcomes. Unfortunately, it is not obvious which environmental scenario is a conservative one, e.g. a high or a low level of nutrient or temperature. In this contribution, we analysed the sensitivity of a *Lemna* model (Schmitt et al., 2013) to changes in environmental conditions in a risk assessment case study. For this case study we considered exposure to a toxicant and conducted several simulations with the *Lemna* model. While the exposure situation was kept equal in all simulations, the environmental conditions were changed. Results demonstrate that population dynamics are altered the most in cases where the exposure occurred in phases with strong growth of *Lemna*. This analysis can be the basis to set a conservative ecological scenario for environmental risk assessment for *Lemna* TK/TD modelling approaches.

**MO364**
Defining ecological lake scenarios for population modelling as part of the Ecological Risk Assessment of chemicals
T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; A. Gergs, Bayer AG / Crop Science / Ecotoxicology; K. Lademann, Research Institute gaiac; M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment

The ecological risk assessment of chemicals (ERA) aims to minimize adverse ecological effects on populations and ecosystems. This assessment strongly depends on the modelled influences the outcomes of the ecological scenarios and the species sensitivity to anthropogenic stressors. This also applies to the populations of planktonic species and fish in standing waters (lentic systems), many of them being focal species in ERA. For use in population modelling, we suggest a classification of ecological scenarios of lentic systems based on the EU Water Framework Directive (WFD). As a result of the European intercalibration process, a list of general lake types has been defined which includes many of the aspects that are important for lake modelling. Besides abiotic characteristics, the German lake classification system for the national implementation of the WFD additionally makes use of biocenotic and trophic descriptors, and provides short characterizations of typical characteristics for relevant lake types. For the German lake types, data on e.g. phytoplankton biomass and nutrient concentrations are available from natural reference lakes which can serve for model validation. As case studies, we have chosen three lake types from this list of general lake types, which differ in relevant lake properties such as morphometry, trophic state, water depth, stratification regime during summer, and food web structure of the pelagic food web. We additionally considered common anthropogenic lakes and ponds (i.e. environmental models) and two shallow, eutrophic ponds as alternative tools in ecological risk assessment. For the simulations of these ecological scenarios, the biogeochemical lake model StoLaM was used, in which several phytoplankton and zooplankton groups as well as fish are implemented. Additionally, the one-dimensional vertical structured hydrodynamic model HyLaM as part of the StoLaM allows high resolution of the lake internal physical environment which is required for simulating the nutrient and plankton dynamics in detail. Based on scenario analyses, simulations of typical plankton dynamics in lake systems will be presented and discussed.

**MO363**
**Lemna** toxicokinetic and toxicodynamic (TK/TD) modelling - Impact of the ecological scenario on the risk assessment
S. Heine, Bayer AG / Effect modelling; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; T. Preuss, Bayer AG / Environmental Safety

The sensitivity of aquatic species to chemical stressors can be derived from typical characteristics for relevant lake types. For the German lake classification system the national implementation of the WFD additionally makes use of biocenotic and trophic descriptors, and provides short characterizations of typical characteristics for relevant lake types. For the German lake types, data on e.g. phytoplankton biomass and nutrient concentrations are available from natural reference lakes which can serve for model validation. As case studies, we have chosen three lake types from this list of general lake types, which differ in relevant lake properties such as morphometry, trophic state, water depth, stratification regime during summer, and food web structure of the pelagic food web. We additionally considered common anthropogenic lakes and ponds (i.e. environmental models) and two shallow, eutrophic ponds as alternative tools in ecological risk assessment. For the simulations of these ecological scenarios, the biogeochemical lake model StoLaM was used, in which several phytoplankton and zooplankton groups as well as fish are implemented. Additionally, the one-dimensional vertical structured hydrodynamic model HyLaM as part of the StoLaM allows high resolution of the lake internal physical environment which is required for simulating the nutrient and plankton dynamics in detail. Based on scenario analyses, simulations of typical plankton dynamics in lake systems will be presented and discussed.

**MO362**
Deviating predicted no-effect concentrations for perfluoroalkyl acids in the Po river ecosystem through a novel methodology based on the AQUATOX ecosysten model
A. Greedel, A. Barausse, L. Grechi, L. Palmeri, University of Padua / Department of Industrial Engineering

Health and environmental risks posed by perfluoroalkyl acids (PFAs) have been rising concerns through the past decades, especially in the most contaminated world areas. One of those is the Northern Italy, because of its high industrialization and population. Nevertheless, the real risk connected to PFAs as emerging contaminants, both for ecosystems and for human health, is still somewhat unexplored. Linking external exposure to the effective dose of the chemical is one of the main tasks of Environmental Risk Assessment. Through 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 and in 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 the 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 test a new approach for deriving PNEC by use of the US-EPA 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 J.D. Stark, Washington State University / Dept of Entomology; C. Mitchell, Washington State University / School of the Environment; V. Chu, Western Washington University / Environmental Science; M.E. Harris, Western Washington University; L. Wallis, Western Washington University / Institute of Environmental Toxicology; G. Young, Advisian WorleyParsons Group / Aquatic Sciences; W.G. Landis, Western Washington University / Institute of Environmental Toxicology; K. von Stackelberg, NEK Associates LTD / Department of Environmental Health

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 (Oncorhyncus tshawytscha) and coho (Oncorhyncus 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 from 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 T. Claudepierre, URAFPA INRA / URAFPA INRA; F. Desor, C. Cakir Kieffer, Université de Lorraine / Université de Lorraine / ULA / URAFPA INRA; M. Delannoy, URAFPA INRA / URAFPA INRA; A. El Hajj, T. Oster, C. Malaplate, Université de Lorraine / ULR / URAFPA INRA; N. Tran, Université de Lorraine / UL / Ecole de chirurgie, Faculté de Medecine de Nancy; F. Yen-Potin, C. Feidi, Université de Lorraine / ULR / URAFPA INRA

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 precise determination of the associated effects and neurodegenerative disease is often 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 primary neurons 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 obtained with the 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 F. Bauer, KREATIS; P.C. Thomas, CEHTA SAS / Ecotoxicology and Risk Assessment

A knowledge of the mechanism of action (MechoA) 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 MechoAs covering hundreds of molecules, we developed a set of structural alerts associated with specific MechoAs. Consequently, a new method to predict MechoAs with high accuracy and with simple rules was developed by using a decision tree developed including 26 general MechoAs including 23 detailed MechoAs. The MechoAs are given mainly for mammals and fish but information on other species was also included. We used a training set of 301 molecules, and validation set of 491 molecules. Our method was built as a linear decision tree composed of 62 decision rules. This method achieved 92% correct classifications for the training set and 92.3% for the validation set. 6% of the predicted classifications were slightly different from the literature MechoAs for the training set (3.4% for the validation set) and 1% of the training set was misclassified (4.3% in the validation set). Finally, only 1% was out of the applicability domain for the training set while no molecules from the validation set were unclassified. This model is both simpler and performs better than the previous method that 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 easily and efficiently 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 U. Boßmann, Aarhus University / Environmental Science; D. Fernández-Calviño, K.K. Brännå, University of Copenhagen / Department of Plant and Environmental Sciences; M.S. Storgaard, Aarhus University, Department of Environmental Science / Department of Environmental Science; H. Sanderson, K. Bester, Aarhus University / Environmental Science

Facade paints and render are commonly protected against biological deterioration using biocides. Mixtures of in-can as well as film preserving bactericides, algacides 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 (T1/2 < 10 d) to compounds with higher persistence (T1/2 > 120 d). For two selected biocides (terbutryn and octylisothiazolinone) 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 octylisothiazolinone was not closed, as some transformation products were degraded as well. However, Microtox tests revealed reduced toxicity of transformation products towards Albugo fischari than the
respective parent compounds. Nevertheless, for most biocides the degradation half-life is longer than time intervals between rain events in Northern Europe. Hence, though many of the used biocides are degrading relatively rapidly in soil most of the compounds residues may accumulate in soil surrounding biocide treated buildings, due to repeated input with every driving-rain event. Consequently, most biocides can be considered as "pseudo-persistent"-contaminants in this context. This was verified within the present study by (sub)surface soil screening, where concentrations of up to 0.1 \( \mu g \) \( g^{-1} \) were detected for parent compounds as well as terbutryn degradation products in soils below biocide treated facades.

**MO372**

**Biocides in facade coatings: Influence of pigments on the phototransformation of biocides**

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Biocides are common additives in facade coatings to protect the materials against biological deterioration. In-can as well as film preservatives are used for this purpose. Nevertheless, these biocides leach to the environment when the facade is getting in contact with driving-rain. Long-term exposure tests in natural weather showed large gaps in the mass balances, indicating towards other loss mechanisms. The present study focused on phototransformation as a major pathway for active ingredient loss. In laboratory experiments in UV-weather chambers the formation and fate of photoproducts are investigated. The latter is based, inter alia on Emission Scenario Documents (ESD) providing methods for release estimation of active substances from biocidal products to the environment. In case of rodenticides (product type 14 of BPR), the current available ESD for Rodenticides (2003) has been reviewed to take account of realistic biocidal product applications as well as worst-case environmental exposure assessment. The German Environment Agency (UBA) has commissioned Dr. Knoell Consult GmbH for drafting a revised ESD for PT 14 (rodenticides) on the basis of European Competent Authorities experiences gained during active substance approval and product authorisation, experiences from a workshop on risk mitigation measures for anticoagulant rodenticides, knowledge and common practice of trained pest operators, rodenticides associations, experiences from awarding public and private authorities and furthermore. New scenarios or sub-scenarios have been developed in case of application of rodenticides in sewer systems (with reference to the different types of pipe systems) and of application in and around buildings (distinction between direct applications on paved and unpaved soil; integration of an indoor baiting scenario). A new scenario for bank slopes of water courses has been established, whereas the waste dumping scenario in the northern scenario from the original ESD for PT14 have undergone minor adaptations. When exposure of the terrestrial compartment is considered the transport of biocidal active substances to aquifers and groundwater has to be allowed for. In case of rodenticide application an appropriate approach for estimation of local concentrations in groundwater is newly included in the revised ESD for PT 14. The risk assessment for primary and secondary poisoning of non-target organisms was revised in order to provide a more generic approach, i.e. identifying focal non-target organisms. Furthermore, guidance already provided for plant protection products has been considered. The presentation aims at providing an overview of current developments in environmental emission and exposure estimation of rodenticides as biocidal products.

**MO374**

**New Developments in Environmental Emission Scenarios of Biocides - Preservatives for products during storage**

K. Michaelis, German Environment Agency (UBA); M. Schwander, German Environment Agency Umweltbundesamt; M. Galler, M. Schweitzer, SCC GmbH.

Preservatives for products during storage are also known as in-can preservatives and are regulated according to Regulation (EU) No 528/2012 (BPR). These preservatives are biocidal products which are used in many different end-products (e.g. detergents, paints, glues etc.) to prolong their service life. The environmental emission of these end-products is evaluated in revised emission scenario documents (ESD), whereas the existing ESD for in-can preservatives does not contain calculations for the variety of all end-products. Consequently, the German Environment Agency (UBA) initiated a research and development project for the further development of the evaluation method of in-can preservatives. The draft for the revised ESD has been prepared by SCC GmbH on behalf of the German UBA. Due to the variety of different applications of in-can preservatives, a differentiation in 6 sub-categories was defined. Additionally, for a complete environmental emission estimation different life cycles of the biocidal end-product have to be assessed. Consequently, the incorporation of the in-can preservative into the end-product (formulation) as well as the uses of the end-product (application and service life) within a subcategory have to be considered. To reduce the workload and to minimize the emission estimation it was decided to define emission scenarios which describe a realistic worst-case situation for the environment refer to application amount, emission days and release fractions. On the basis of expert knowledge, draft competent authority reports of in-can preservatives and a survey between stakeholders, industry and other EU member states worst-case scenarios were identified and discussed at EU level. Finally, the revised ESD suggest one or a few worst-case emission estimation scenarios for a group of pigments. Using a prioritisation concept for biocides a worst-case scenarios, calculation sheets for the estimation of the emission from other uses are provided as Appendices, so that the emission from other end-products (non-worst-case scenarios) can be calculated as well, by using this ESD.

**MO375**

**Monitoring of Biocides in German Sewage Treatment Plant Effluents - First Results**

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Due to a widespread use, biocidal active substances and their transformation products are expected to be found in the environment. Projections show that there will be an increase of biocide entries in the environment, mainly in urban areas due to an increased use of e.g. disinfectants and especially masonry preservatives. Biocidal substances enter the environment through numerous entry pathways. One main entry path is through sewage treatment plants (STP). Therefore, the German Environment Agency (UBA) initiated a project where the effluent of 29 public STPs from all over Germany will be investigated over a period of one year, starting in November 2017. Additionally, selected samples from influents as well as from sewage sludge emissions will be in the focus. Using a prioritisation concept for biocides a list, ranking substances that enter the environment through the STP-pathway, was generated. The list was judged by experts and finally, for this project 23 biocidal active substances or transformation products were chosen for analysis. First results show that several substances can be detected at measurable concentrations in the effluents. This ongoing project will provide better knowledge about the fate and behavior of biocides entering the environment through sewage STPs. It will give us a time dependent picture of the environmental pollution by biocides in Germany through urban STPs and will also show possible fields of action for regulatory purposes.

**MO376**

**The 'risk envelope approach' applied to environmental risk assessments for disinfectants - a strategy to reduce workload for biocidal product families**

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Under the Biocidal Products Regulation, applicants can apply for authorisation of biocidal product families (BPFs), which consist of products with similar uses, the same active substances, similar compositions within specified variations and similar levels of risk and efficacy. Especially when consortia are formed and products from multiple companies are grouped into a single dossier, building a dossier to demonstrate safe use for all products may become burdensome. Hence, there is a strong need to reduce the amount of risk assessments required to support the BPFs, in the interest of the applicants as well as the competent authorities. BPFs are typically subdivided into subfamilies called 'meta SPCs'. The subgrouping in meta SPCs considers o.a. the composition, formulation type, product type (PT), risk mitigation measures (RMMs), classification and labelling (C&L) and shelf-life of the individual products. The differences between the environmental risk assessments, however, most often do not coincide with the factors that determine the meta SPC structure. Instead, other grouping strategies are more fit for purpose. The risk envelope approach is a strategy routinely applied in
plant protection product dossiers. It entails - that for each area of risk assessment - the key parameters driving that risk assessment are identified. Subsequently, the uses are grouped and ranked according to these key parameters. As such, one or more worst case or 'critical' uses can be identified. If it can be demonstrated that there is no undue risk to men or environment for the critical use, all other uses are considered to be covered as well. A case study will be presented whereby the concept of the risk envelope is applied to the environmental risk assessment for a BPF of disinfectants (PT 1-5). Risk assessments can be grouped (a) for different products/uses within a meta SPC, and (b) for different products/uses across meta SPCs. Overall, applying the risk envelope approach may lead to a great reduction in workload, whilst allowing for easy addition of products/uses to the BPF at a later stage. The benefits and potential difficulties of this approach will be discussed in detail.

MO377 Are biocide emissions into the environment already at alarming levels? Recommendations of the German Environment Agency (UBA) for an approach to study the impact of biocides on the environment
K. Pohl, German Environment Agency (UBA) / Section Biocides; C. Meier, German Environment Agency (UBA) / Biocides; M. Ahlkt, I. Noeh, German Environment Agency UBA / Biocides
More than 40,000 biocidal products were registered on the German market, including disinfectants, preservatives, pest control and antifouling products. All biocides act as intended on living organisms and the use of these biocides can result in adverse impacts on the behavior and/or toxicity of biocidal substances to consumers and the environment. The risk assessment of biocidal substances is of particular concern for the environment. Instead of monitoring individual findings of only a few substances, particularly in surface water.

However, a comprehensive picture of the actual pollution of the environment with biocides – one that goes beyond such individual findings – is not available, since there is no biocide-oriented, systematic environmental monitoring in Germany to date. To tackle this problem, the German Environment Agency (UBA) has developed recommendations for an environmental monitoring program of the biocides based on the results of a research project and two international workshops. These recommendations contain a prioritization concept for biocidal substances as well as a proposal for a systematic monitoring programme. At first, we established a database containing information relevant for the environmental risk assessment according to the Guidance on Biocidal Products Regulation (BPR) for all biocidal substances currently available on the market. A multi-criteria prioritization approach was applied to prioritize substances based on their 1) emission relevance, 2) environmental effect data, and 3) environmental persistence. Thereby creating lists of high-prioritized biocidal substances and relevant transformation products that are of particular concern for the environment. Instead of monitoring individual environmental effects, our approach aims at monitoring the biocides and their entry pathway of relevant biocidal substances. Therefore, we developed different entry path scenarios (work packages), which represent the different use pattern and entry paths of particular biocidal products. Based on the obtained prioritised substances and the different entry paths a systematic monitoring strategy is suggested for a German wide inventory of biocides in the environment. This will provide on one hand better knowledge of the occurrence and behavior of biocidal substances to consumers and their impact on the environment. On the other hand, these monitoring data could help to support a more comprehensive risk assessment of biocides by providing a basis for risk mitigation measures or for the exclusion and substitution of environmentally hazardous active substances.

MO378 A case study on exposure assessment of biocides in PPCPs 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 additive. Despite of its inhalation toxicity, this compound has been used as humidifier disinfectant 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 this study is assessing the exposure of CMIT/MIT in PPCP and comparing discussed with two different levels of consumer exposure tools. ECETOCA TRA.3 was used as tier 1 model which is basic and simple but conservative calculations and ConsExpo was used as tier 2 as it can be more precisely redefined and covers more specific estimations. The gap of estimated exposure values which have been 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.

MO379 Hazard evaluation of biocides and its metabolites for the aquatic compartment
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The LIFE-COMBASE 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, half-life, transformation products and acute toxicity, chronic toxicity to fish, invertebrates, algae and WWTP microorganisms. The EU Regulation (EC) No 1272/2008 on classification and labeling was considered to group these compounds in four toxicity categories taking into account the values of NOEC or LC50 as (1 ≤ 1 mg/L), (2 >1 to ≤ 10 mg/L), (3 >10 to ≤ 100 mg/L) and (4 >100 mg/L). Most of the found data was related to toxicity in fish, followed by invertebrates and algae, marine organisms 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 2% of 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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MO380 Synchronous decreasing levels of imposex and tributyltin (TBT) in dogwhelk (Nucella lapillus) from Norway, 1991-2015
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Imposex is TBT induced reproductive abnormality in female dogwhelk (Nucella lapillus). This biological effect is quantified by the Van Deeren 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 confirm the rationale of implementing strict antifouling regulations on industrial chemicals when this 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 Tributyltin (TPTT). Thresholds for VDSI (≤ 0.2% of males) and TBT (3.2 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.

**MO381**
Risk assessment issues for algacides under BPR

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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 properties, 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-reduction realistic worst cases to ensure risk assessment leading behaviour. In the case of biocidal products applied in swimming pools to disinfect or to control algae growth in water, several active 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 poster will focus on following key steps: to determine 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.

**MO382**
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 (FOOum 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 realistic worst cases to ensure risk assessment leading behaviour. In the case of biocidal products applied in swimming pools to disinfect or to control algae growth in water, several active 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 poster will focus on following key steps: to determine 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.

**MO383**
Veterinary medicine products (VMPs) are used in livestock production to preserve animal health or to promote growth in certain categories of animals; 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 health and the 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 directed into several 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 under 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 its primary use only: We noted that some biocides are also used in human medicines or as pesticides. Admission to the market, based on active substances, is therefore sometimes regulated in different rules and making also, usage data is scattered. This makes priority setting difficult when performed in a segregated view on the universe of chemicals.

**MO385**
Comparing methods for estimating environmental emissions


The environmental risk assessments consist of information on exposure and hazards of chemicals to environmental compartments. Environmental emissions from 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 poster 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 assessments 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 petrochemicals products datasets used in LCA studies

G. Castelan, PlasticsEurope / LCA; P. Salting, BASF SE / Sustainability Strategy

The steam cracker process turns fossil hydrocarbon feedstock streams into several different main products, like ethylene, propylene, benzene, toluene, xylene, etc. 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 to lead 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 product will have slightly different LCI results depending on different amounts of products derived from the 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 method and ecoinvent v.3.2 “Rest of the World” datasets for upstream processes. 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 uncertainty effect is possibly higher. Furthermore, basic 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 leads to error 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 battery’s 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 in Europe. On the basis of previous studies, 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. Bahramifar, 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 Fe3O4-based (Fe3O4@SiO2-NH-SH) composites, which function with a similar thiol group for Hg(II) removal are compared at 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 nanoadsorbents, it is also accompanied 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, ethanol, methanol, DCC (N,N'-dicyclohexylcarbodiimide), NHS (N-Hydroxysuccinimide), water recovery, and electricity. The results of test comparing the impacts between MGO-NH-SH and Fe3O4@SiO2-NH-SH estimated respectively 37, 34, 40, 31, and 26% more climate change, water use, energy use, human toxicity, and ecotoxicity, respectively for the latter. Sensitivity analysis were employed to determine the uncertainties for scale-up production and it is shown that especially potential reductions of electricity use, ethanol 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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in order to keep the global temperature below 2°C. However, 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 elucidate specific emissions factors for WOSR and that can be included and subsequently derive WOSP-specific emission factors. Furthermore, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. In addition, the project strives to develop robust but also generalisability statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. Thus, we evaluate the outcome of life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors.
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$_3$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 STP continuously for 10 days. The sewage sludge of each treatment was dewatered and the biosolids were mixed with soil. After 0, 60, 90, 120, and 180 days, the NPs were counted. Along with the cellulose 15685) 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. However, the inhibition of the AOB was lower than the inhibition due to the silver nitrate (70% inhibition) and the nano-sized Ag$_2$S (30% inhibition). There were no effects on the emergence or plant growth of Avena sativa over 8 weeks in the chronic plant test. An uptake of a low Ag concentration into the roots of the plants was observed.

**MO398 Energy reserves and respiration rate in the earthworm Eisenia andrei after exposure to zinc in nanoparticle or ionic forms**

Z.M. Swiatek, Institute of Environmental Sciences, Jagiellonian University / Institute of Environmental Sciences; A. Bednarska, Polish Academy of Sciences / Institute of Nature Conservation

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 earthworm Eisenia andrei, we investigated the effects of different NPs on 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 NPs and biomass (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/ZN8/01576).

**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 stability, the ease with which they can be synthesised and the myriad 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 in vitro and in vivo effects of gold nanoparticles on the immune system of Porcellio scaber. This is the first study investigating the effects of gold NPs on the immune system of Porcellio scaber, which is a representative model species of terrestrial isopods. The results suggest that the earthworms are able to regulate internal Zn concentrations efficiently, regardless of its form and concentration, and 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<0.03) 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 affected the respiration rate measured on Eu two biochemical components. 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 NPs and biomass (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/ZN8/01576).

**MO397 Terrestrial isopods as models to assess the biotransformation of nanoparticles and ionic Ag inside the organisms: an example with silver and gold nanoparticles**

C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

The rise of nanotechnology and the increased use of nanomaterials in consumer products may lead to an increased emission of nanoparticles (NPs) to the environment. Since NPs may leach from products during use, waste water treatment plants (WWTPs) may be an important sink but also an important source of NP emission to the environment. The use of sewage sludge in agriculture may, for instance, lead to NP exposure in soils. NPs may undergo transformation when passing WWTPs, with sulphidation being an important process. Silver nanoparticles are among the NPs most likely ending up in soils. And considering the transformation processes taking place in the WWTP, Ag$_2$S may be a form in which the NPs likely will reach the soil. In soil, sorption, aggregation and dissolution processes will determine the availability of the NPs or released ions for uptake by organisms. Bioavailability will also depend on soil properties that play an important role in governing nanoparticle fate. This study aimed at assessing the importance of soil type on the bioavailability of Ag and Ag$_2$S NPs to enchytraeids (Enchytraeus crypticus) and springtails (Folsomia candida). Four soils with different pH (4-7), organic matter (2-17%) and clay contents (3-13%) were used. An uptake and elimination kinetics approach was taking, in which the animals were exposed to a single concentration (nominal 10 mg Ag/kg dry soil) for 14 days (uptake phase), after which they were transferred to clean soil for a 14-day elimination period. A first-order one-compartment model was used to calculate uptake and elimination rate constants. Results for the enchytraeids showed k1 values for the uptake of Ag ranging between 0.009 and 0.057 g soil/g animal/day for Ag$_2$S and of 0.107-0.671 g soil/g animal/day for AgNO$_3$. These data suggest a lower availability of the Ag from the Ag$_2$S NPs than from the AgNO$_3$. The k1 values for the uptake of Ag$_2$S in different soil properties were significantly different for Ag$_2$S. Where lowest availability was expected in the soil with the highest cation exchange capacity (CEC), this indeed was the case for AgNO$_3$, but not for Ag$_2$S. Elimination rate constant values (k2) ranged between 0.057 and 0.565 per day, and were not dependent on soil type or Ag form. Tests on the springtails are still ongoing.

**MO396 Influence of soil type on the toxicokinetics of Ag and Ag$_2$S nanoparticles and ionic Ag in soil invertebrates**

C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

Elimination rate constant values (k2) ranged between 0.057 and 0.565 per day, and were not dependent on soil type or Ag form. Tests on the springtails are still ongoing.
hemocoe, isopods were injected with gold NPs and then left for 48 hours to recover from the injection; in previous experiments 48 hours was shown to be enough time for hemocyte numbers to return to pre-injection levels. The total number of cells, viability and the proportion of hemocyte types were counted. These counts were then compared to the animals which had been fed NPs and to others that had been injected with a non-lethal dose of LPS. Preliminary data shows that the isopods cellular immune response is altered upon direct injection of NPs, but no such effect was found after their ingestion. The study is still ongoing.

**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 on 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 E50 concentrations of Ag nanoparticles (AgNPs) [3]. Moreover, AgNPs are mostly transformed to Ag sulfide nanoparticles (AgS NPs) due to the reducing conditions present in the wastewater treatment plant (WTTP) [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 on the earthworm species Eisenia fetida. The tests were conducted following OECD and ISO protocols to determine ecotoxicity of nanomaterials as AgNPs, AgNO3, and AgS NPs. The results obtained from this research can be used as an indicator species for nanomaterial release. Soil nematodes showed a significant decrease in reproduction across all exposure concentrations tested (1 mg Ag/kg soil). Moreover, AgS NPs showed the highest ecotoxicological response compared to earthworms and pot worms and should be used as an indicator species for nanomaterial release.
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) not observed in pristine Ag particles. These responses are not due to high cell toxicity and precede other responses at higher levels of biological organization. The responses in transplanted 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 cocloemocytes of Eisenia fetida
s. curies, priet conciet; O. Tsuysky, University of Kentucky, Department of Plant and Soil Sciences; J. Li, University of Kentucky, Department of Toxicology and Cancer Biology; M. Saenz, PRIET CONICET, National University of Luqian; W.D. Di Marzio, CONICET-PRRIET / PRRIET; j. urrine, University of Kentucky, Department of Toxicology and Cancer Biology
With the rapid development of nanotechnology and its broad applications, a wide variety of engineered nanoparticles are used in commodities, pharmaceuticals, cosmetics, biomedical products and industries. Cerium oxide nanoparticles (CeO₂-NPs) are a key component in various products and, as chemical-mechanical planarization agents in production of silicon wafers. This study investigated the toxicity of CeO₂-NPs with polymer coatings in of different charge in cocloemocytes of Eisenia fetida earthworms. The CeO₂-NPs (2-5 nm primary particle diameter) were coated with dextran to confer a neutral charge (DEX-CeO₂ (0)), diethylenetramino dextran to confer a positive charge (DTE-CeO₂ (+)) and dimethyldichloro an negatively charge (CM-CeO₂ (-)). The range of exposure concentrations were 0.02-1562.5 mg Ce L⁻¹. The cocloemocytes were exposed ex situ for 1 h for each treatment. Then, the transcriptions levels of genes associated with stress (catalase and heat shock protein 70) were determined by q-RT-PCR. In addition, cytotoxicity and genotoxicity were determined by using tripan blue assay and comet assay respectively. The responses varied significantly among exposure concentration and charge of polymer coatings. The results showed that positively charged DTEA-CeO₂ (+) were more toxic than negative and neutral CeO₂-NPs. The results show that initial surface chemistry had a profound impact on the toxicity of CeO₂-NPs to cocloemocytes.

MO405
The uptake of pristine and aged silver nanoparticles by wheat, Triticum aestivum, in a soil exposure
A. Green Exha, CEE Wallingford; C. Schultz, Centre for Ecology and Hydrology; D. Tarnovska, M. Matzke, NERC Centre for Ecology and Hydrology; D. Spurgeon, Centre for Ecology & Hydrology; C. Svendsen, CEH, Wallingford / Pollution Effects of Cerium and others in of different forms.
It is expected that most nanoparticles (NPs) which reach the soil will not be in their pristine form but instead will have been transformed by the environment (e.g. sulphidisation of Ag in waste water treatment processes). This will greatly influence the form of NP to which soil organisms are exposed and their ultimate bioavailability. The bioaccumulation of NPs inside the organisms can govern their fate and transformation in the environment; and uptake studies can give insight into how organisms act as sources and sinks for NPs in food web. Most data currently available are for pristine Ag NPs, and consequently the difference in the bioavailability of the aged forms, predominantly Ag₂S, is uncertain. The aim of this study is to compare the uptake kinetics of Ag NPs, both pristine (PVP coated Ag NPs, 20 and 50 nm) and aged (Ag₂S, 20 nm), in the crop species, wheat, Triticum aestivum. Wheat plants were exposed from seed to each of the NPs at two nominal concentrations of Ag, 3 and 10 mg Ag/kg, in the soil Lufa 2.2. Samples were collected at five time points over the 42 day post-emergence exposure period. The growth rate, Ag accumulation and the translocation from root to shoots were determined. The toxicokinetic parameters of the Ag uptake in the roots and shoots were calculated using total soil concentration and soil pore water concentrations as metrics of exposure. Pore water was collected at all sampling points and at the end of the exposure period pore water was ultra-filtered as a measure of the dissolved Ag in the pore water. The accumulation of all silver forms was greater in the roots, with only a small fraction transported to the shoots. The uptake of Ag₂S was lower compared to pristine Ag particles but there was no difference between the uptake of the two pristine Ag particles. This study shows that environmentally relevant forms of Ag NPs are bioavailable to plants and show different uptake kinetics than the pristine forms.

MO406
In vitro effects on Denrobrauna veneta cocloemocytes of Ag and TiO₂ nanoparticles before and after wastewater treatment processes
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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 or terrestrial ecosystems or applied on agricultural land, however, the transformation of NPs in the environment is very 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 NPs compared to pristine Ag. In this study, cocloemocytes (primary immune cells) isolated from the epigeic earthworm Denrobrauna veneta are used as a model to assess the effects of Ag and TiO₂ NPs. Initial investigations focus on Ag (PVP coated, 25 nm, nanoComposite) and TiO₂ particles (uncoted anatase, nominal primary size of 5 µm, NM-101, JRC) and their mixture, to better understand their uptake, interaction with cocloemocytes 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 (TEM), while chemical marker of Ag-SPNPs and TEM are performed 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 cocloemocyte 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, Ag-PVP and Ag-TiO₂, on the lethality of Daphnia magna and anaerobic metabolism in Daphnia magna. Organisms (14-d old) were exposed to 25-125 µL/L of NM-300K for 96 h in a WWTP effluent or in ASTIM medium. Daphnids were analysed for changes in acetylochinesterase (AChE), glutathione S-transferase (GST), catalase (CAT), lactate dehydrogenase (LDH) activities, and lipid peroxidation (LPO). Results showed a significant increase of CAT activity in negative control comparatively to ASTIM control, thus suggesting induction of oxidative stress by effluent. The dispersant used in ASTIM 4% w/w of each Tagat® TO and Tweene® 20) showed both significant decreases (AChE, GST, CAT) and increase (LDH) of enzymatic activities in dispersant-control relatively to negative-control, suggesting deleterious effects of dispersant to daphnids. Biomarker responses to NM-300K were more marked when added to effluent comparatively to ASTIM, especially for higher concentrations. There was a significant decrease of ACHIE activity in effluent (25 and 75 µg/L) and ASTIM (125 µg/L) media, which implies impairment of control and modulation of neural transmission signal in these experimental conditions. The significant increase of GST and CAT activities at 100 and 125 µg/L in effluent, respectively, suggest oxidative stress. The significant increase of LDH activity in ASTIM (125 µg/L) in effluent suggests an increase in anaerobic metabolism and higher stress for daphnids. Unfortunately, there was a significant decrease on LPO at 125 µg/L in ASTIM, which could be explained by a decrease of synthesis of total lipids. 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 standard test media and (ii) there is a distinct biomarker response-pattern in daphnids exposed to WWTP effluent containing NM-300K and ASTIM 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 accidentally. 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, special emphasis was placed on the development of a method for the analysis of nanoparticles in the freshwater amphipod Hyalella azteca.

MO409 Development of a method for the analysis of nanoparticles in the freshwater clam Corbicula fluminea


As part of the REACH Substance Evaluation for silver, new data was required to be generated for further justification raise-from acrossionic silver to silver nanoflakes. Therefore, the aquatic ecotoxicity and fate and behaviour of ionic silver and the smallest silver nanoflake with the highest specific surface area registered under REACH were tested. An ecotoxicity testing programme was undertaken in order to assess the effects of the silver nanoflake with silver nitrate using the following internationally recognized standardised ecotoxicity tests: Toxicity to the alga, Pseudokirchneriella subcapitata (OECD Test Guideline No. 201). Long-term toxicity to Daphnia magna (OECD Test Guideline No. 211). The silver nanoflake was fully characterised and was an aqueous suspension containing approximately 37% nanoparticles with spheroidal-like shape (mean primary particle size 9.4 nm). Total silver, conventional dissolved silver (0.45 μm membrane filtered) and colloidal dissolved silver (3 kDa centrifugation filter) were measured by ICP-MS in samples taken from test vessels. Membrane filters (0.45 μm) and centrifuge filters were conditioned before use with the test solution/dispersion to be filtered. Particle size & Zeta Potential were determined (DLS) in vessels without test organisms. Elemental particle size distribution was analysed in separately prepared samples of the test item in test medium by means of asymmetric Flow-Field-Flow-Fractionation (4F) coupled to ICP-MS for the silver nanoflake only. Based on measured silver concentrations, silver nitrate was more toxic than nanosilver to both algae growth and Daphnia reproduction, for all silver fractions. Size and Zeta Potential measurements are inconclusive for all tests and it appears that the test concentrations were too low / particles too few to resolve from control / background level (using the Zetasizer Nano equipment). In addition, the dissolution rate via ingestion test of particulate Ag and/or biomass contamination of dissolved ionic silver in the ecotoxicity tests over a period of 28 days (following OECD Test Guideline No. 29), with measurements of the same three silver fractions. This showed a different dissolution behaviour in both test media, which agreed with observations in the ecotoxicity tests.

MO411 Investigations on the uptake pathway and accumulation of silver from manufactured silver nanoparticles in the freshwater amphipod Hyalella azteca

S. Kühr, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; N. Schröder, Fraunhofer IME - Department Bioaccumulation and Animal Metabolism; D. Volker, German Environment Agency; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; W.D. Di Marzio, 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 Ecosistema 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 models have shown that soils and sediments can provide important reservoirs of these nanoparticles, especially in the presence of high concentrations of DOM. These such as the humic substances found in water, sediment, and soil, are an unavoidable interacting with ENPs. To understand and assess the effects of ENPs on the environment, should be well established quantitatively the concentration–response relationships. Also, to know what is the density that regulates the abundance of diatoms 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 hyperbolic copepod species and less related with DOM concentrations. The hyperbolic zone is a region underneath streamlined 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 hyperbolic 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 survivor of Microcystis aeruginosa, a widespread hypertrophic species. Toxicity of AgNPs was related with DOM concentrations and showed a non-significant -B 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. Aravanitou, 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 0.081mg/L of ZnO NPs. The cultures were grown in modified Blue-Green 11 medium (BG-11). The effects of ZnO NPs on the survival of Microcystis aeruginosa, a dominant indicator of blue-green algae, nutrient removal and lipid production. The toxic effect of ZnO NPs was estimated by the half maximum inhibitory 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**

s. schiavo, ENEA CR; M. Oliviero, University Parthenope; A. Philippe, Universität Koblenz-Landau / Institute for Environmental Sciences Group of Environmental and Soil Chemistry; s. manzo, ENEA / SSPT-PROTER-BES Sunscreens represent one of the main source of engineered TiO2 nanoparticles (TNPs) source in coastal ecosystems, especially during summer period. Their adverse effects were generally investigated using bare model TNPs and only few studies were based on the NPs extracted from commercial products. Therefore, this study aims to evaluate the effect of TNPs extracted from three different commercial sunscreens upon freshwater and marine organisms: microalgae (Pseudokirchneriella subcapitata; Dunaliella tertiolecta) and crustaceans (Daphnia magna). Microalgae are a widespread indicator for growth rate, nutrient removal and lipid production. The toxic effect of ZnO NPs was estimated by the half maximum inhibitory 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. **MO415**

**Silver nanoparticles affect the early development of Tisbe battagliai: pristine vs aged particles**

A. Georgantziopoulos, Norwegian Institute for Water Research NIV; K. J. Farkas, SINTEF Ocean / Environmental Technology; K. Ansorge, 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 transformations 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 battagliai. In this study the harpacticoid copepod Tisbe battagliai 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 focus on the study of Ag (I) particles coated in a nanoComplex) 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 both the particles types were stable in laboratory conditions. The uptake of the TiO2 particles was lower in comparison to the AgCl particles. The concentrations decreased over time in seawater, which was likely due to both dissolution and aggregation for Ag, and aggregation for TiO2. Particles were found to be more stable in seawater after the aging process, which can affect their impacts on exposed organisms. **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 relatively recent development of engineered Ag nanoparticles has expanded silver's applications considerably. Silver nanoparticles (AgNP) tend to agglomerate 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, can provide information on the level of exposure to toxic metals. The aim of this study was to investigate Ag concentration in the haemolymph of the marine amphipod Parhyale hawaiensis exposed to food containing AgNP and AgCl. We hypothesized the actual AgNP could be absorbed by the gut leading to a higher amount of Ag in the haemolymph when compared to food containing AgCl. Silver nanoparticles < 100nm (Sigma Aldrich) or elemental Ag (from AgCl, Sigma Aldrich) were incorporated into formulated fish feed (4% of the fish diet). Fish were maintained in an aquarium with an approximately 200 mg l-1 . 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 maintained for 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 Spectrometer (GFAAS). A higher amount of silver in the haemolymph was absorbed from AgNP feed, reaching 8.4±0.7 ng mg-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
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 bivalve clams**

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The use of carbon nanomaterials (CNMs) has increased rapidly in the last years, mainly 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 micrometers 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 functionalities that change 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. In this research, the 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. Comparison of metabolic activities (GLY and ETS), oxidative stress biomarkers responses (LPO) and antioxidant enzymes activities (SOD and GPx) compared to Ni-MWCNTs. In the present study, it was clearly demonstrated that nanomaterial toxicity can be attributed to core structure and surface functionalization, which have been shown to alter the level of toxicity.

**MO418**

**Assessment of genotoxic and proinflammatory effects of different Silica and Titania Nanoparticles on human bronchial cells**

D. Cavallo, Italian Workers Compensation Authority INAIL / Occupational and Environmental Medicine, Epidemiology and Hygiene; C.L. Ursini, R. Maello, A.M. Freeeman, A. Ciervo, S. Iaviloci, Italian Workers Compensation Authority-INAIL / Occupational and Environmental Medicine, Epidemiology and Hygiene

The widespread production and use of titanium dioxide (TiO2) and silica (SiO2) nanoparticles (NPs) in consumer products and in different industrial and medical applications raises concerns about their possible toxicity. We studied potential genotoxic-oxidative and inflammatory effects of two amorphous silica NPs (precipitated NM200 and pyrogenic NM203) and two anatase TiO2 NPs (NM100 size 50-100 nm and NM101 size 5-8 nm) furnished by JRC. NM characterization by TEM and DLS. Human bronchial (BEAS-2B) cells were exposed for 24 h to 0.1-1.000 μg/ml of selected NMs to evaluate: cytotoxicity (tripan blue assay), direct/oxidative DNA damage (Fpg-comet assay) and inflammatory effects (IL-8, TNFα release by ELISA). In culture medium NM200 was better dispersed than NM203, 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 100 μg/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 TiO2NPs 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. Sarett, 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 (Eawag)

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. In this research we aim 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 sets of 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. In future work we 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 the aged ENMs to the environment, the effect of the media on the properties of the ENMs will be evaluated. Ultimately, this will help refine our understanding of ENM environmental fate.

**MO420**

**Freshwater sediments as an environmental reactor: defining biologically relevant fate parameters to provide context for nanomaterial bioaccumulation**

R. Cross, University of Exeter; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; T.S. Galloway, University of Exeter / Biosciences

As the field of nanotoxicology matures there is a call for the research focus to progress from hazard identification to more ecologically relevant assessment of the risk that engineered nanomaterials (ENM) pose as they undergo a range of transformations in the environment. This will require test designs prioritising environments most at risk of contamination, and which not only measure ecologically relevant endpoints, but also characterise the fate, transformations and behaviours of particles within the test system, providing the context for differences observed between treatments. Freshwater sediments present an ecosystem in need of further research, as these are predicted to be major sinks of ENMs entering the aquatic environment though waste water treatment and terrestrial pathways during material production, use and disposal. Whilst freshwater sediments have been identified as an ecological compartment at risk of contamination, very little is known about the fate of ENMs entering these sediments. We present a simple separation method to isolate the colloidal (< 200 nm) and dissolved (< 1kDa) fractions of the sediment pore water, which can be run alongside biological exposures. This provides the context for how these biologically accessible fractions of ENMs in the sediments may relate to intrinsic particle properties such as size, core composition and coatings. Using cerium oxide (CeO2NP) and silver nanoparticles (AgNPs) as model ENMs for these sediments, we investigated the effect of the media on the properties of these particles. We present a simple separation method to isolate the colloidal (< 200 nm) and dissolved (< 1kDa) fractions of the sediment pore water, which can be run alongside biological exposures. This provides the context for how these biologically accessible fractions of ENMs in the sediments may relate to intrinsic particle properties such as size, core composition and coatings. Using cerium oxide (CeO2NP) and silver nanoparticles (AgNPs) as model ENMs for these sediments, we investigated the effect of the media on the properties of these particles. We present a simple separation method to isolate the colloidal (< 200 nm) and dissolved (< 1kDa) fractions of the sediment pore water, which can be run alongside biological exposures. This provides the context for how these biologically accessible fractions of ENMs in the sediments may relate to intrinsic particle properties such as size, core composition and coatings. Using cerium oxide (CeO2NP) and silver nanoparticles (AgNPs) as model ENMs for these sediments, we investigated the effect of the media on the properties of these particles. We present a simple separation method to isolate the colloidal (< 200 nm) and dissolved (< 1kDa) fractions of the sediment pore water, which can be run alongside biological exposures. This provides the context for how these biologically accessible fractions of ENMs in the sediments may relate to intrinsic particle properties such as size, core composition and coatings. Using cerium oxide (CeO2NP) and silver nanoparticles (AgNPs) as model ENMs for these sediments, we investigated the effect of the media on the properties of these particles. We present a simple separation method to isolate the colloidal (< 200 nm) and dissolved (< 1kDa) fractions of the sediment pore water, which can be run alongside biological exposures. This provides the context for how these biologically accessible fractions of ENMs in the sediments may relate to intrinsic particle properties such as size, core composition and coatings.
the sediment and lack of dissolution (< 1% of spiked cerium was extractable with water). Transdermal uptake of AgNPs 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 done that are surface-chalcophile electron transfer. To better understand whether the use of these nanoparticles will have a potential impact on the transport and toxicity of TiO2 NPs, we are developing methods to probe the surface transformation processes. These processes inevitably come into contact with different types of dissolved organic matter (DOM). It has been shown in a large number of studies that DOM influences the reactivity of TiO2 NPs in this study include methylene blue dye degradation (photocatalytic activity), rose bengal dye degradation (photodynamic), and fluoresein 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.

MO43 Evaluating the role of TiO2 nanoparticle surface transformations on transport and toxicity
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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. Kaege, Eawag - Swiss federal Institute of Aquatic Science and Technology
Our results revealed that engineered nanomaterials (ENM) will inevitably come into contact with different types of dissolved organic matter (DOM). It has been shown in a large number of studies that DOM influences the colloidal properties of ENM, which in turn impact subsequent transport and transformation processes. Sulfidation, as an important environmental transformation process, has significant implications for the fate and the ecological effects of ENM that are based on chalcophile elements. However, to date, our knowledge on the influence of DOM on the kinetics and mechanisms of this transformation reaction are very scarce. For copper oxide nanoparticles (CuO NPs), 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 bisulfide (HS−), wastewater systems represent major sulfidizing compartments, where ENM inevitably come into contact with different types of DOM. Therefore, we are investigating the influence of different types of dissolved organic matter (DOM) on the sulfidation of CuO NPs. This study includes the use of X-ray photoelectron spectroscopy (XPS), variable amounts of DOM, and X-ray absorption spectroscopy (XAS). In addition, XPS studies were performed that are surface-chalcophile electron transfer. To better understand whether the use of these nanoparticles will have a potential impact on the transport and toxicity of CuO NPs, we are developing methods to probe the surface transformation processes. These processes inevitably come into contact with different types of DOM. It has been shown in a large number of studies that DOM influences the reactivity of CuO NPs in this study include methylene blue dye degradation (photocatalytic activity), rose bengal dye degradation (photodynamic), and fluoresein dye conversion (ROS generation). Ultimately, changes in the properties of the CuO 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.

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)

MO424 Assessing potential risks of Nanodrugs and their delivery systems in fish using Light Sheet Microscopy
D.L. Windell, University of Exeter / College of Life and Environmental Sciences; J. Moger, The University of Exeter / College of Engineering, Mathematics and Physical Sciences; M.J. Winter, The University of Exeter / College of Life and Environmental Sciences; S. Owen, AstraZeneica / Safety Health Environment; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences
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 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 zebrafish 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 tissues. Depuration 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.
MO426
Effect of ageing on polymeric aromatic hydrocarbon composition of biochar
G. Sigumund, Technische Universität München / Chair of Analytical Chemistry and Water Chemistry; T. Bucheli, Agroscope ART / Environmental Analytics; I. Hilber, Agroscope / Environmental Analytics; T. Höff, M. Kah, T. Hofmann, University of Vienna / Department of Environmental Geosciences

The influence of ageing on biochar properties has been investigated by comparing three biochars that were thermally aged by either H$_2$O$_2$ 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 PAHs are released in PAHs when they are freshly produced and that the risk of 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).

MO427
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 sample coupons are 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 and field settings. This study describes the testing of the approach in situ and in the field. The sampling location was a storm water retention pond collecting storm water run-off from a motorway. The pond has two basins, one of which is equipped with a Floating Treatment Wetland (FTW) for cleaning the run-off. 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 the first 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.

MO428
Use of biochar for hexachlorocyclohexane sorption: a mechanistic approach
L. Silvan, g. cornelissen, s.e. hale, Norwegian Geotechnical Institute
Hexachlorocyclohexanes (HCHs) are halogenated compounds composed of 4 main isomers: α-HCH, β-HCH, γ-HCH and δ-HCH, which differ for their tridimensional structure. Commercial HCH in technical grade is a mixture of these isomers. HCH has been extensively used as a pesticide despite the fact that only γ-HCH (lindane) has insecticide properties. HCHs’ toxic, carcinogenic, teratogenic and neurotoxic effects have been reported in humans; and the HCHs have a clear tendency to accumulate in the environment. For these reasons, HCHs are contaminants of water, soil, biodiversity, and sediments, and also there is no definitive way to destroy them. Various remediation techniques have been used to remove HCHs in aqueous solution, among these, adsorption is the most common used one. Biochar (BC) is a carbonaceous material that is promising sorbent amendment material due to its high adsorption of organic and inorganic contaminants, and to its low cost. In this study three standard biochars, from digestate (BC$_d$), from greenhouse tomato waste (BC$_t$) and from durian shell (BC$_f$), have been used as sorbent materials for the HCHs removal from water. The BCs used cover a wide range of surface area (5.4 - 328.6 m$^2$ g$^{-1}$), pore volume (5.1 - 186.6 cm$^3$ g$^{-1}$), pore dimension (1.05 - 5.85 Å), pyrolysis temperature (400 – 700 °C) and surface properties (including iron content). Batch isotherm tests were carried out in deionized water with the single isomers and the mixture of α-, β-, γ- and δ-HCH. The HCH concentration was ranged between 1 and 500 µg L$^{-1}$ in the monocomponent isomers and between 5 and 2000 µg L$^{-1}$ (total concentration) in the mixture isomers. Polyethylene (PE, 26 g L$^{-1}$) was spiked into this as a passive sampler to assess the HCHs concentration in water. The sorption performance of the biochars is related to their physicochemical properties. Preliminary results have shown the adsorption performances are correlated with the BC surface area and iron content, where a higher adsorption is observed as surface area and iron content increase. Clear differences in the behavior of the isothers of the iron content. Therefore, the expected adsorption performances are correlated with the BC surface area and iron content, where a higher adsorption is observed as surface area and iron content increase. Clear differences in the behavior of the isomers of the iron content.

MO429
Development of a Method for Measurement Freely Dissolved Concentrations of Alkylated PAHs Using Solid Phase Microextraction with PDMS Fibers
M. Renningen, RWTH Aachen / Bioi; T. Parkerton, ExxonMobil Biomedical Sciences Inc. / Toxicology & Environmental Science; G. Witt, HAW Hamburg / Department of Environmental Geosciences

Freely dissolved concentrations of waterborne hydrophobic pollutants in the environment. Often for pyrogenic sources of PAHs (e.g. incomplete burning processes), the parent PAHs are the predominant species. In contrast, PAHs from petrogenic sources (e.g. crude oil) are dominated by alkyl PAHs. Therefore, the U.S. EPA narcosis model of pyrogenic PAHs is based on the calculated magnitude of PAH toxic units (TU) (Hawthorne et al., 2006). However, due to the heterogeneous nature of organic carbon in field sediments, potential risks of adverse biological effects from sediment-associated contaminants are most often assessed based on concentrations of parent PAHs in sediment pore water, not to C$_{s0}$. Recent advances in equilibrium passive sampling methods (EPSMs) offer a promising alternative to the measurement of C$_{s0}$ and support improved risk-based decision making since bioavailability of sediment contaminants can be directly quantified via C$_{s0}$ (Mayer et al., 2015, Burkhard et al., 2017). When using EPSMs, polymer to water partition coefficients are crucial for reliable calculation of C$_{s0}$. To date partitioning coefficients are available for parent PAHs across different polymers (e.g. PDMS, POM) (Lydy et al., 2015). In this study, an equilibrium passive sampling method was developed for investigating alkylated PAHs in marine and limnic sediments and used for risk evaluations of both pyrogenic and petrogenic PAHs. The method is based on solid phase microextraction (SPME) with different silicone materials (PDMS coated glass fibers and hollow fibers). Partitioning coefficients for the PDMS coated fibers (K$_{PDMS}$) were calculated for selected target alkylated PAHs which have previously not been available. K$_{PDMS}$ for additional alkylated PAHs of interest were then predicted based on the experimentally reported K$_{PDMS}$ values. These findings will be made available to the federal and state agencies for use in environmental risk assessment and designing management strategies.

References
Delle Site, A. Factors affecting sorption of organic compounds in natural
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. Joio Rocha, ICBAS U.Porto, CIMAR CIMAR LA; J.L. Dores-Sousa, VUB / Department of Chemical Engineering; C. Cruzeiro, CIMAR CIMAR LA, Porto, CEF FTUC U.Coinbra; E. Rocha, ICBAS U.Porto, CIMAR CIMAR LA. The study shows that 16 priority compounds (PAHs) in surface waters from the Douro River estuary and nearby Atlantic seaboard. 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 (216PAHs) 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 ≈ 55 ng/L in water. In view of the potential for bioaccumulation and toxicity of these organics, suggesting that both mutagenic and carcinogenic responses can occur in both humans and aquatic animals living in these areas. This is a statement 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. Acknowledgements: European Regional Development Fund (ERDF) through COMPETE, Framework of the Structured Program of R&D&I INNOVARM – Innovation and Sustainability in the Management and Exploitation of Marine Resources (NORTE-01-0145-FEDER-000335), Research Line ECOSERVICES, supported by the Northern Regional Operational Programme (NORTE2020), through the ERDF, ICBAS – U. Porto. 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 M. Larsson, Orebro University / Man-Technology-Environment research centre (MTM); M. Lam, RWTH University Aachen; P. van Hees, Orebro University / MTM Research Center; J. Giesy, University of Saskatchewan / Department of Veterinary and Biomedical Sciences and SINTEF, Centre; M. Ringwall, Orebro University / Man-Technology-Environment research centre (MTM) Polycyclic aromatic hydrocarbons (PAHs) are common contaminants in 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 contamination being a mixture of different processes despite 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 bioanalytical 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 toxicoologically relevant PACs. Agonists in concentrations below 1 µg/g was detected. 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 Y. Verhaegen, CONCAWE; C. Hughes, Shell Health / Risk Science Team; L. Cameron, ExxonMobil; A. Amadeo, A.D. Redman, ExxonMobil Biomedical Sciences / Toxicology and Environment Science Division; M. Comber, Mike Comber Consulting; S.A. Villalobos, BP / Global Product Stewardship; V. Ochoa, Cepsa; S. Lintington, BP; E. Vaiopoulos, European Petroleum Resources Association Petroleum substances are examples of UVCBs (substances of Unknown or Variable Chemical Composition). Complex mixtures of products or Biomass derived compounds, the chemical composition will vary 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 for Concawe substances using a worst-case approach. ECHA recently released its Read-Across Assessment Framework (RAAF) for environmental endpoints, however the RAAF for UVCBs is still under development due to their added complexity. One recommendation of the RAAF when applying a category approach is to present data in a matrix to demonstrate that properties are similar or follow a regular pattern. In this presentation a category data matrix will be presented for the Concawe category of vacuum hydrocracked gas oils (VHGO). Available historical experimental aquatic toxicity data will be presented alongside substance identity information, predicted EL50 and toxic unit (TU) values calculated using PETROTOX, and results from biomimetic extraction solid phase microextraction (SPME) screening studies. The latter is a technique which measures bioavailable hydrocarbons, and has been demonstrated to correlate well with experimental and predicted aquatic toxicity data. The resulting, complimentary dataset forms a weight of evidence upon which to justify category approaches to the read-across of experimental toxicity data.

MO435 Automated Solid Phase Microextraction (SPME) for measuring freely dissolved concentrations of hydrophobic chemicals in soils, sediments and other solid matrices C. Vitale, University of Insurbia; K. Knudsmark Sjoldholm, University of Copenhagen / Dept. of Plant and Environmental Sciences; A. Di Guardo, University of Insurbia / Department of Science and High Technology; P. Mayer, Technical University of Denmark / Department of Environmental Engineering In risk assessment of hydrophobic chemicals that are strongly associated to the soil/sediment organic matter, freely dissolved concentrations ($C_{free}$) are more representative than total concentrations ($C_{total}$) of their actual bioavailability, potential for bioaccumulation and toxicity. Such freely dissolved concentrations can be measured by Solid Phase Microextraction (SPME) if operated in the equilibrium and reproducible deposition mode. Furthermore, in order to reduce the measurement variability, increase sample throughput and to produce high quality data, automated SPME methods are promising. 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 within 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. Borch, DTU Environment / Department of Environmental Engineering; L.N. Tech, Technical University of Denmark / Environment 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 avoiding formation of dispersions or microdroplets. A slow-stir 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 droplet formation. 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 alkalanes 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(+)-Pine were tested in both the algol 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 algol 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 this study, 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 substance bioconcentration. In this study, the extractions data provide a basis for the development of predictive models for bioavailability of petroleum substances.

MO441 
Bioaccumulation of hydric organic compounds in aquatic biota: addressing current challenges for in tissue passive equilibrium sampling
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The occurrence of a range of historical and emerging hydroporphic 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 BAFsw, averaged 4.0±0.3, 4.4±0.3, 4.7±0.3, 3.9±0.7 and 4.3±0.4 for galaxolide, traseolide, phantolide, celestolide and tonalide 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 hydroporphic 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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ACES, B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology

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 retained and how they are released from sediments. These pollutants can be accumulated by aquatic organisms and biomagnified to higher trophic levels. Hence, it is important to explore the composition, activity and effects of environmental mixtures of pollutants in sediments of different origin, characteristics and pollution history. Sediments from Sweden, the European Arctic (coastal Svalbard vs. open sea), Queensland (Australia) and a French-German river were collected. The freely dissolved concentrations (C_{freely}) of the chemicals were determined using equilibration 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 world-wide, 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 (ERa); and adaptive stress response (oxidative stress, AREc32). Cytotoxicity was assayed in all bioassays and occurred occasionally. Moreover, four of the seven bioassays were not dosed with the pure chemicals. For the first time, a combination of bioassays revealed a potential hazard of those chemicals that might be relevant in future scenarios (C_{fraily} bio).

The present 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, malapelo in the northern Gulf of Mexico

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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 malapelo collected over three years from multiple estuaries in the NGOM from Texas to Florida were tested for alkylphenol contamination using GC/MS. We found no significant differences in contamination levels between the core marine populations but exhibits low toxicity. All these compounds have high partition coefficients, which explains their presence in animal tissue. Concentrations of NP, BHT, and DTBP were highly correlated in malapelo 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

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Hydrophobic organic chemicals (HOCs) are of special ecotoxicological 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 yet well understood. In blue crab (Callinectes sapidus) exposed to marine pollutants, single-molecule fluorescence microscopy (SMFM) with a microfluidic flow chamber and temperature control has enabled us to record the dynamic process of 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 degree 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.=0.56). 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 PCNs 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

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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 toxic 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 of 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 exposure in the Marné hydrographic network. Surprisingly, chubs infected by the anacanthocephalan Pomphorhynchus laevis were not contaminated. In the study of 60 chubs for PCB and phthalates. Further validations are needed to confirm the transfer of these pollutants from host to parasites and to investigate the potential benefits of this detoxification pathway for parasitized chubs.

MO445 Environmental occurrence and distribution of organic UV stabilizers in the sediment of the North and Baltic Seas

C. Apel, Helmholtz-Zentrum Geesthacht; R. Ebinghaus, Helmholtz-Zentrum Geesthacht / Department for Environmental Chemistry

Organic UV stabilizers are of emerging environmental concern due to their large production volumes and concern about their 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 boating. Four benzotriazole 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 sunscreens to protect human skin against harmful effects of UV radiation. The pathways into the marine environment are either indirect by wastewater treatment plant discharge or direct by recreational activities like bathing and boating. Four benzotriazole UV stabilizers are classified as SVHC (Substances of Very High Concern) under the EU regulation REACH. Numerous other 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 from coastal and marine sediments are insufficient. 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). For this, 22 mL stainless steel ASE-350 were filled with 1 g of deactivated silica and approximately 5 g sediment that was spiked with appropriately isotope labelled standards. The cells 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 multihapse (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. Leonetti, University of Insubria (Como) / DiSTA; A. Buffo, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; R. Perna, University of Insubria; S. Polesello, Water Research Institute- CNR / Water Research Institute; S. Valsecchi, Water Research Institute - Italian National Research Council IRSA-CNR; 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
MO448 Kinetic Sorption and Bioaccumulation of Hydrophobic Organic Chemicals in Marine Plankton Food Chain


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 using phytoplankton (Tetraselmis chuii), rotifers (Brachionus sp.), and copepods in a gas perfusion 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 those with high molecular weight PAHs (PA and PY) 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 (KOA) in plankton, however the different linear regression slope of log BCF and log KOA 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?

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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 duration of weathering processes like radiation can alter nanoparticle properties and lead to changes in their environmental behaviour. The duration of weathering processes like radiation can alter nanoparticle properties and lead to changes in their environmental behaviour.

MO450 When technical limits triggers risk assessment for non-biodegradable insoluble pharmaceutical molecule

B. Jouruel, E. Beltran, CEHTRA SAS; P. Adrian, CEHTRA

Earth 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 Koa 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 often difficult to analyse and measuring experimental results more subject to imprecision. Due to analytical difficulties, parameters such as water solubility and Koa, are often expressed as “lower than” or “higher than” and have no defined value. Additionally, model softwares such as EUSES impose maximum value for Koa and minimum value for water solubility as input parameters, whereas EU TGDR spreadsheet allows to consider the experimental results. The decision on the parameter values and models to be considered needs then expert judgment. We will present the case of a pharmaceutical molecule currently studied, for which water solubility and then Koa, cannot be precisely measured experimentally. While staying regulatory compliant and reflecting the experimental results, the input values chosen for these parameters have a significant impact on calculated PECs for this insoluble molecule. Studies on fate and behaviour in soil and water/sediment must be conducted, however considering the technical difficulties to analyse the molecule, and the route of environmental exposure, it may be necessary to define a category of molecules for which some of current regulatory requirements could be waived related to their chemical properties. A proposal for an appropriate risk assessment will be provided.

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 analysed only once 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: Characterization of the products and methodological and environmental occurrence

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The use of large amounts and a broad variety of chemicals that are potentially harmful to the environment (including persistent organic pollutants (POPs), personal care products (PCPs) and pharmaceuticals) as chemical components are needed to estimate potential risks, especially for sediment-dwelling organisms as a part of the ‘Trojan Horse’ effect. In the present study MWCNT were irradiated by simulated sunlight (300-400 nm) for 90 days. The weathered MWCNT (wMWCNT) were used to investigate their influence on fate and distribution of the biocide triclocarban (TCC) in a sediment-water system. This substance was chosen because of its hydrophobic properties and strong affinity to sorb on MWCNT. The concentration of wMWCNT has a significant impact on the distribution of TCC in natural water. 100 µg and 1000 µg wMWCNT/L in Milli Q water led to an adsorption (log Koc/wMWCNT in OECD medium: 7.6 L/kg) of 10% and 65% 1C-1T-CC respectively. We will report experiments on the distribution of TCC in water and in sediments with wMWCNT and TCC concentrations from 1 mg wMWCNT/L and 1 mg TCC/L shaken for 2 h with 1 mg wMWCNT/L and subsequently incubated in a sediment-water system in the dark for 180 d. A scenario with C-1T-CC only will serve as control. TCC is expected to sorb onto the wMWCNT and accumulate in the natural sediment by fast sedimentation of wMWCNT-TCC complexes. Production and release of carbon based nanomaterials are predicted to further increase in the near future, thus the interactions of nanomaterials with organic pollutants will be of growing importance to assess environmental consequences. Acknowledgements: The work is supported by the European Project Nano-Transfer which receives funding from the Bundesministerium für Bildung und Forschung (BMBF) under agreement with the FP7 ERA-NET SINN.

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 analysed only once 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: Characterization of the products and methodological 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), personal care products (PCPs) and pharmaceuticals) as chemical components are needed to estimate potential risks, especially for sediment-dwelling organisms as a part of the ‘Trojan Horse’ effect. In the present study MWCNT were irradiated by simulated sunlight (300-400 nm) for 90 days. The weathered MWCNT (wMWCNT) were used to investigate their influence on fate and distribution of the biocide triclocarban (TCC) in a sediment-water system. This substance was chosen because of its hydrophobic properties and strong affinity to sorb on MWCNT. The concentration of wMWCNT has a significant impact on the distribution of TCC in natural water. 100 µg and 1000 µg wMWCNT/L in Milli Q water led to an adsorption (log Koc/wMWCNT in OECD medium: 7.6 L/kg) of 10% and 65% 1C-1T-CC respectively. We will report experiments on the distribution of TCC in water and in sediments with wMWCNT and TCC concentrations from 1 mg wMWCNT/L and 1 mg TCC/L shaken for 2 h with 1 mg wMWCNT/L and subsequently incubated in a sediment-water system in the dark for 180 d. A scenario with C-1T-CC only will serve as control. TCC is expected to sorb onto the wMWCNT and accumulate in the natural sediment by fast sedimentation of wMWCNT-TCC complexes. Production and release of carbon based nanomaterials are predicted to further increase in the near future, thus the interactions of nanomaterials with organic pollutants will be of growing importance to assess environmental consequences. Acknowledgements: The work is supported by the European Project Nano-Transfer which receives funding from the Bundesministerium für Bildung und Forschung (BMBF) under agreement with the FP7 ERA-NET SINN.
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 southern Northeast 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 EHMCl) 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. Ląpczynski, 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) and RIFM are collaborating in the 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 and 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 established to rank for risk assessment refinement. Efforts are continuously being made 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
A. Dhabbah, A. Stec, University of Central Lancashire / Centre for Fire Hazards and Science, School of Physical Sciences and Computing; Y. Badjah-Hadj-Ahmed, King Saud University / College of Science, Chemistry Department; R. Hull, University of Central Lancashire / Centre for Fire Hazards and Science, School of Physical Sciences and Computing. 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 polymeric 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, teardar 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 teardar 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[a]pyrene, benzo[b] anthracene, and acenaphyne.

MO455
PbTik modelling of super-hydrophobic chemicals
W. Lachic, 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 Declorane (log Kow (estimated) = 11.6) with our recently published PbTk 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 super hydrophobic chemicals such as HCB in order to get correct results. Subsequent simulations with the super-hydrophobic chemical Declorane revealed that for an oral uptake route the diffusive 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 (C12-1AS) and alcohol ethoxylate (C13E08). Chemosphere. 72:850—862. 2. Sakuratani Y, Noguchi Y, Kubayashi 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/ir_csa_r11_pbpeg_en.pdf/ddac9031-da4a-4995-4efc-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. Berryn, 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. Cromie, Wildfowl & Wetlands Trust

MO459
Main scientific gaps on knowledge of risk from NSAIDs to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions
M. Tagger, University of the Highlands and Islands / Environmental Research Institute

MO460
Main scientific gaps on knowledge of deliberate poisoning to [migratory] wildlife globally
M. Odin, 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. Staab, BASF SE; J. Roembke, S. Jaensch, ECT Oekotoxikologie GmbH; P. Kabouw, BASF SE; S. Braeker, BASF France S.A.S.
In order to place a plant protection product correctly in a market, it is necessary to characterise its effects on the environment and, in particular, on the sensitive species to be protected. In addition, the effects of a plant protection product on ecosystem services and functions must be taken into account. This is normally done by conducting field trials. Still, it is a long-standing challenge to achieve semi-chronic and chronic effects in the laboratory. This challenge is even more critical when testing novel or less familiar products or combinations of products, for which no experience is available. In this case, other studies providing similar information can be used for extrapolation. However, in many cases, the existing databases are too limited to be able to draw useful conclusions. In this study, we used big data analysis methods to explore existing earthworm field studies with Boscalid and to identify relevant model properties. In total, 284 long-term field trials with Boscalid were identified. The database contained information about soil type, soil properties, earthworm species composition, and earthworm abundance and earthworm diversity. In a first step, the influence of the experimental design and soil parameters on the earthworm responses were statistically evaluated using multiple linear regression models. In a second step, a machine learning approach was applied to predict the earthworm responses from soil properties and soil parameters. In this way, the dataset was extended and patterns could be detected which were not apparent at first sight. In a third step, a Monte Carlo simulation approach was used to evaluate the potential impact on earthworm populations under realistic scenarios. In the future, the results of this study will be used to develop more reliable and transferable models for prediction and risk assessment in eco-toxicology.

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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 “toxicologically significant criteria” operations putting 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 setting up 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 determining which results are due to the nature of the 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 for 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 can be turned into computable phenotype ontologic classes. To semi-automate the code mapping, a Java-based lookup tool was developed using the ontology browser BioPortal (https://bioportal.bioontology.org) to REST API to conduct lookups. 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 necessary using the results of a single code mapping. The results of the automated code mapping approach were evaluated against a set of manually annotated prototypes as indicated by exposures to ten well studied chemicals (atrazine, bisphenol A, cadmium chloride, chlorpyrifos, copper sulfate, cypermethrin, dioxin, EE2, malathion, or Tris(1,3-dichloroisopropl)phosphate) in six vertebrate species (carr, zebrafish, fathead minnow, mouse, rat, trout). The content of this presentation neither constitute nor necessarily reflect US EPA policy.

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. Pili, K.A. Fay, CSRA, Inc. The ECOTOXicology knowledgebase (ECOTOX) is a comprehensive, curated database providing access to thousands of chemical exposure studies to terrestrial and aquatic organisms. The ECOTOX Knowledgebase provides risk assessors and researchers consistent information on toxic effects of chemicals 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 chemical fields. Studies details such as species, taxonomy 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 advancements 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 Edaphon database
J. Hausen, RWTH Aachen University; B. Scholz-Statarke, M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research
A steadily increasing number of databases in ecotoxicology and ecology combine and merge data from different studies and research programs. Usage of these 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. Automation 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 Edaphosdata warehouse (https://portal.edaphosdata.org/). Edaphosdata combines spatially explicit information on quantities of soil organisms, environmental parameters, and vegetation. The data in the warehouse are coming from more than 100 long-term studies in different earthworm communities in relation to the field, conducting 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.

TU006
Deriving USEtox aquatic freshwater toxicity Effect factors from the REACH database for thousands of chemicals using R-Studio program
The database has been used to calculate unique value for chemical properties and aquatic 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 and aquatic 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 and aquatic 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 and aquatic 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 and aquatic 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 and aquatic 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 and aquatic 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 and aquatic 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 and aquatic 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 and aquatic toxicity data (41’381 test results) available in the IUCLID 5.5 database (as of March 2017).
and 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 chemical 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, was studied in the framework of the stability of data and 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 and Thoronics means with standard deviation - Comparison of the chemical toxicity based on the above calculations and chemical ranking based on GHS and CLP.

TU007

Deriving physicochemical 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 Bioeconomy Unit; S. Proenca, EU Commission Joint Research; A. Gissi, European Chemicals Agency; E. Sasson, EU Commission JRC / Sustainable Assessment Unit

Product Environmental Footprint (PEF) and Organisational Environmental Footprint (OEF) 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. Some key parameters like toxicity data are optimized here for cancer and non-cancer effects. For PEF, these data are required for thousands of chemicals using the most up-to-date information. The EU commission Joint Research Centre has obtained from the chemical agency (ECHA) for more than eight thousand chemicals data regarding physico-chemical properties (166 926 test results, as of March 2017) available in the IUCLID 5.5 database. These data have been processed to automatically derive accurate values for six of the physico-chemical properties required by USEtox for fate modelling: Kow, Koc, vapour pressure, water solubility, Henry law constant and biodegradability; in addition, adsorption partition coefficient (Kd) to suspended matter, sediments and soil for inorganic compounds were determined. In order to provide high quality results, criteria were applied for selecting data on the basis of their reliability (assessed by Klimisch scores), purpose and study type. Moreover, other specific criteria were defined for each properties according to the method used, such as experimental condition (temperature and pH). Geometric mean and the coefficient of variation, for their reliability evaluation, of the consistent data selected was performed. A correlation analysis of the results with values previously included in USEtox and with values obtained with computational methods (QSAR/QSPR) was established to assess the quality of this proposed automated approach. A quality levels approach is proposed. This, paired with the abovementioned statistical index, represents a helpful support to the user for evaluating the reliability of each parameter used in the fate model. For chemicals with no available data or not satisfying the minimum quality requirements, physico-chemical properties were derived using in-silico tools (QSAR/QSPR). In particular, OECD QSAR toolbox and the EPI suite estimation models played a fundamental role for this data gap filling purpose.

TU008

Toward a possible Toxicity Test Battery Integrated Index for Nanomaterials

M. Oliviero, University Parthenope; s. schiavo, ENEA CR; s. manzo, ENEA / SPIT-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 trophic levels, which may not represent the exposure point 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 environment. Therefore it 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 proposed such 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 TBI 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 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 it 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.

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. Steiger, 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 handlings of PPP, reduce or 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 available 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 and correct potential errors. For example, a dataset of spray sequences 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 historic analysis of pollutants it is sometimes unavoidable to fire potential handlings of PPP beyond the digital era. This trend is consistent, and the result of this study in a unique 44 year time series of PPP use in apple orchards. In a next step, we will analyse the historic development of ecotoxicological risks of PPP usage in apple orchards.

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. Through 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 linear 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) determine the potential (direct) and indirect (e.g. via herbivores) effect of NNs on farmland bird populations. To achieve this, data in 1950 on PPP use 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 historic analysis of pollutants it is sometimes unavoidable to fire potential handlings of PPP beyond the digital era. This trend is consistent, and the result of this study in a unique 44 year time series of PPP use in apple orchards. In a next step, we will analyse the historic development of ecotoxicological risks of PPP usage in apple orchards.

TU011

Regression-based models reveal sources of pollutants in Norwegian marine sediments

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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 explanatory variables 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., Sørjord in western Norway) for mercury, zinc, cadmium and lead. Highest concentrations of PAHs and PCBs were found close to densely populated and industrially modified regions, i.e., in the North Sea area 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 As-Tomemy1: ISPRAMaltese, F. Onorati, ISPRA Institute for Environmental Protection and Research; G. Martuccio, ISPRA Institute for Environmental Protection and Research / National Center for Laboratory Networking Ecotoxicology Area; L. Manfra, R. Di Mento, G. Moltedo, B. Catalano, ISPRA Institute for Environmental Protection and Research / National Center for Laboratory Networking Ecotoxicology Area; C. Sibbio, G. Chiaretti, O. Farapontova, M. Amici, C. Maggi, G. Romanielli, G. Sesta, G. Granato, F. Venti, P. Lanera, S. Manta, M. De Luca, F. Venanzini, ISPRA Institute for Environmental Protection and Research. Environmental quality assessments and monitoring plans are key tools to all activities related to potential contamination of ecosystems, including marine systems. Potential effects of oil/gas production activities in Adriatic Sea (Italy) are successfully investigated since 2000 by water and sediment chemical analyses, sediment grain and proportional mixtures, and other methods. Following a quantitative WOE model study, a multidisciplinary approach including chemical analyses, grain size analysis and bioassays on marine sediment, together with bioaccumulation and biomarker investigations in polychaetes exposed to sediment, is applied to assess potential impact due to offshore platforms and produced water (PFW) discharge. PFW is a complex mixture of contaminants and is the main discharge of gas/oil platforms. Marine sediment samples from two offshore platforms in Central and Eastern Adriatic regions were analyzed. Marine sediments were physo-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 biosassays battery (Vibrio fischeri, Dunaliella tertiolecta, Tigriopus fulvus) were considered. Moreover a battery of biomarkers at different biological levels together with bioaccumulation of some organic and inorganic contaminants were analyzed in polychaetes (Hediste diversicolor) exposed to sediment under laboratory conditions. A multidisciplinary weight of evidence (WOE) study was carried out, integrating different lines of evidence (LOE) as sediment grain and proportional mixtures, bioaccumulation and biomarkers. Taking inspiration from classic gradient analyses used for macro-organisms in Ecology (Threshold Indicator Taxa Analysis - TITAN, logistic and quadratic regressions), we identified bacterial taxa that presented positive, negative, neutral or subsidy-stress responses to a well characterized urbanization gradient in 41 streams in the Raleigh-Durham area (North Carolina, USA). We used a combination of environmental variables (% development, % forested, sediment Zn concentration, biotic index) that were significantly correlated to bacterial community structure to identify reliable bacterial indicator taxa along this multiple stressor gradient. Using TITAN, we identified more bacterial indicator taxa negatively impacted by urbanization than positively impacted (138 and 56 OTUs, respectively). Using quadratic regressions, we found 140 OTUs presenting a subsidy-stress response to the gradient. We observed that two bacterial families were strongly and consistently decreased by urbanization: Acidobacteriaceae (Acidobacteria) with 50% of OTUs identified as pure and reliable indicator taxa and Xanthobacteraceae (Alphaproteobacteria) with 39% of indicator taxa. Positive responders were distributed all over the phylogenetic tree and the family Comamonadaceae (Beta-proteobacteria) presented the highest number of indicator taxa (14%). We calculated with TITAN that the community-level threshold, indicating the peak along the gradient where the maximum decline in all negative responders happened, was at 12.1% development. This community-level threshold occurs at very low levels of urbanization patterns and expresses a full potential of microorganisms to urbanization and the potential of bacteria to be used in bioindication or monitoring along with more traditional indexes.

TU013

Understanding biomarkers in a multispecies approach to relate organochlorine exposure and biological effects V. Weyerer1, North-West University - School of Biological Sciences / School of Biosciences; V.C. Vonhuesenberg, University of Pretoria / Zoology; R. Gerber, North-West University / Unit for Environmental Sciences 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 Southern African vector control programmes DDT is still allowed for malaria vector control. We undertook to determine the biological effect of the 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 (catalase, superoxide dismutase, malate dehydrogenase, 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 vitulus) and Malagasy clowned frog - Xerophaga mutata (expected to be more sensitive) were not available in the analysis, significant relationships with cytochrome P450 and lipid energy reserves were observed. 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)

TU014

Identifying bacterial indicator taxa along an urbanization gradient in stream ecosystems 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 beta diversity (composition) displayed by the highest DDT high-throughput sequencing data by characterizing the response of individual taxa (OTUs) to a multiple stressor gradient and identifying bacterial indicator taxa. Taking inspiration from classic gradient analyses used for macro-organisms in Ecology (Threshold Indicator Taxa Analysis - TITAN, logistic and quadratic regressions), we identified bacterial taxa that presented positive, negative, neutral or subsidy-stress responses to a well characterized urbanization gradient in 41 streams in the Raleigh-Durham area (North Carolina, USA). We used a combination of environmental variables (% development, % forested, sediment Zn concentration, biotic index) that were significantly correlated to bacterial community structure to identify reliable bacterial indicator taxa along this multiple stressor gradient. Using TITAN, we identified more bacterial indicator taxa negatively impacted by urbanization than positively impacted (138 and 56 OTUs, respectively). Using quadratic regressions, we found 140 OTUs presenting a subsidy-stress response to the gradient. We observed that two bacterial families were strongly and consistently decreased by urbanization: Acidobacteriaceae (Acidobacteria) with 50% of OTUs identified as pure and reliable indicator taxa and Xanthobacteraceae (Alphaproteobacteria) with 39% of indicator taxa. Positive responders were distributed all over the phylogenetic tree and the family Comamonadaceae (Beta-proteobacteria) presented the highest number of indicator taxa (14%). We calculated with TITAN that the community-level threshold, indicating the peak along the gradient where the maximum decline in all negative responders happened, was at 12.1% development. This community-level threshold occurs at very low levels of urbanization patterns and expresses a full potential of microorganisms to urbanization and the potential of bacteria to be used in bioindication or monitoring along with more traditional indexes.

TU015

Diarura sorption in freshwater biotrons: determination of isotherms B. MAULMETI, Istria; J. Favaro, Istituto Italiano Ricerche Biosud RABX. In 2000, the EU Water Framework Directive (directive 2000/60/EC) was implemented with the objective of reaching the good ecological status of rivers. 45 chemicals were indexed as priority including 19 pesticides. The biofilm is at the basis of the trophic chain in aquatic environments and considered as an excellent bioindicator for water quality assessment (Edwards and Kjellrump 2013) because of its ability to integrate contaminant display and display. In this study, we used a photosynthesis inhibitor herbicide: diuron, one of the priority substances to the EU Water Framework Directive. Previous experiments carried out in order to characterize diuron bioaccumulation in biofilms, with two different exposure concentrations, suggest that pesticide uptake by microorganisms was not proportional to contaminant concentration in the water. For this experiment, we suspected that diuron absorption and bioaccumulation rate, and in order to confirm this hypothesis, bioaccumulation and toxic impact were simultaneously assessed at the equilibrium. To that aim, mature biofilm previously grown on glass slides
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 maximal diuron concentration at 2073 µg.g⁻¹, and an equilibrium constant of 0.378. Photosynthesis inhibition was correlated (R²=0.75) to diuron concentration in the water. The data did not clearly highlighted a relationship between bioaccumulation and photosynthesis inhibition. This study establishes that diuron bioaccumulation in biofilm is non-linear, and allows to calculate the equilibrium constant of the 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 behaviour and impact in periphytic microorganisms.

TU016
New insights into the biotransformation of sulfluramid: 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 sulphonamide (N-EtFOSA), belonging to PFASs, is used as the active ingredient in the pesticide, Sulfluramid, which is particularly important in the control of leaf-cutting ants in some developing countries. Previous studies have investigated the biotransformation of N-EtFOSA 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 Protocellum 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; 20:5n3), can not be synthesized de novo or in insufficient proportions by any other type of biomass. The mode of action of these two pesticides which respectively blocks electron transfer in photosynthesis, and inhibits the synthesis of aromatic aminocids. This study aimed to investigate the effect of three herbicides on the diatom Gomphonema gracile, which is an important microalgal species in aquatic ecosystems (Aydinbal 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) polysaturated fatty acids (PUFA) decreased with S-metolachlor contamination (C2); 2) saturated fatty acid (SFA) and monounsaturated (MUFA) 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 MUFA 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. reinhardtii
M. Saenz, PRIET CONICET, National University of Luján; k. Bisova, Laboratory of Microalgae and Microbial Cycles I of Microbiology, W.D. Di Marzio

Nickel is a heavy metal used in many industries and has been identified as one of the most toxic heavy metals to aquatic ecosystems. Nickel exposure to 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 within metabolic pathways 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. reinhardtii. 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 aliquots 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, glutathion reductase and concentration of chlorophyll a, chlorophyll b and carotenoids at the end of 96 hs 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 pigmentation concentration was less evident than the effects on growth rates, indicating a lower sensitivity of these parameters. Nickel provoked severe damage on algal cell with reduction of the cell cycle progression, growth and pigments synthesis pathways. Nickel inhibited also antioxidant enzymes activities. An integrated analysis is done discussing the consequences on population performance in natural environment after metal discharged from different anthropogenic sources.

TU019
Use of BioleadEcoPlateTM to evaluate the effects of ZnO nanoparticles on soil microbial communities
V. Romano, Parthenope University of Napoli / Science and technology; v. pasquale, University Parthenope; s. schiovin, ENEA CR; M. Oliviero, s. dumontet, University Parthenope; s. manzo, ENEA / SSPT-PROTER-BES

Nanoagrochemicals seem to be the new frontier in modern agriculture due to their increased efficacy and reduction of the amount of applied fertilizer. Zinc oxide NPs have been largely used nanofertilizers their speedlingo 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). BioleadEcoPlate 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 oxides profiles related to several different carbon sources. This study aims to investigate the changes in the metabolic profile of the soil microorganisms along the ZnO NPs amended soil. Nutritional stress was used to stress soil microorganisms along decimal dilution in response to the exposure for one month to different type of fertilizers (F1 and F2). The fertilizers were added with ZnO Bulk, ZnO NPs and ionic zinc (ZnSO₄) 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 ZnSO₄, the eluates were obtained. Different eluates were tested upon Pseudokirchneriella subcapitata (6.25%, 12.5%, 25% and 50%). Lepidium sativum (50%). At time 0, 15, 30 days fresh soil samples were assessed by using the BioleadEcoPlate approach. The occurrence of the microbial oxidation of each BioleadEcoPlate™M source was calculated as probability ‘p’ on a binomialal data in order to identify the treatments able to preserve the highest possible oxidizing ability of C substrates and those negatively affected by heavy metals. 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 Biolead results seemed to highlight that the microbial community
was mainly affected by ZnO NPs. The integration of the classical ecotoxicology with BiologEcope approach could represent a good strategy to establish the environmental risk related to the use of nanofertilizers. Keywords: microbial community, ecotoxicology, nanofertilizers

**TIU020**

**Environmental factors-regulated disease dynamics of tilapia lake virus (TLV) transmission in farmed tilapia ponds**

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**BACKGROUND:** Outbreaks of tilapia lake virus (TLV) have caused substantial mortalities of farmed tilapia, posing a significant threat to worldwide tilapia industry. Environmental factors controlling TLV disease dynamics should be clearly elucidated to prevent the potential economic impacts on aquaculture.

**OBJECTIVE:** The main object of this study was to make the TLV 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 TLV dosage were fitted by two-parameter Hill model to estimate median lethal dose (LD50). To explain TLV highly artificial environmental conditions, sacrificing some of the experimental reproducibility to obtain more environmentally relevant data, we studied the 72-hour effects of uncaged CuO (CuSO4 as ionic 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 (Chlorella vulgaris, Scenedesmus dimorphus), diatoms (Pistia trinervis, Fucoides pelliculosa), 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 = 143-1 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 shrinkage and heterogametization. Overall, Fv/Fm was a less sensitive toxicity endpoint than biomass, but the two parameters were strongly correlated (Spearman’s ρ = 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.

**TU022**

**Impact of the antihistamine fexofenadine on structure and functioning of less contaminated microbial communities**

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**METHODS:** In a recent study the antihistamine fexofenadine was shown to impact the microbial decomposition of plant detritus, however, the mechanistic basis for this remains unexplored. We therefore conducted a microcosm experiment, where we microbially colonized two plant substrates (i.e., black alder leaves and hay). Both precolonized substrates were subsequently exposed towards fexofenadine at concentrations of 0, 2, and 200 µg/L. Replicates (n=10-15) were harvested after 15 and 30 days. Substrates were used to determine mass loss or preserved to estimate microbial communities’ structural or functional composition including fungal biomass, sporulation of aquatic fungi, bacterial abundance, and fungal and bacterial DNA and enzyme activities. Furthermore, water samples were analyzed for dissolved organic carbon (DOC) quality or preserved to analyze total organic carbon. After 15 days of exposure, there was a tendency towards reduced decomposition of black alder leaves (~40%) in both fexofenadine treatments, while after 30 days, decomposition in the 200-µg/L treatment was increased by ~45% (but both not significantly different from the control). On the contrary, the decomposition of hay tended to be increased by fexofenadine exposure after 15 days. After 30 days no differences could be observed among fexofenadine treatments for hay, while generally more hay was decomposed than black alder. Accordingly, in water samples of the two substrates, substantial differences in the DOC quality were observed. Furthermore, fexofenadine exposure lead to a shift in bacterial community structure and/or functional composition differ between the two tested substrates as well as among the antihistamine treatments. Moreover, potential implications in carbon and nutrient fluxes in exposed systems are indicated as the detected alterations in DOC quality may affect planktonic decomposer communities that are involved in DOC’s metabolism in surface waters. To gain an in-depth mechanistic understanding of the observed effects, we are currently analyzing variables related to microbial community structure and functioning.

**TU024**

**Innovative tools and metagenomics for the monitoring of rivers and lakes: the European project INTCATCH**

**M. D. Sermiswah**, Brunel University / Institute for the Environment; **S. Marcheggian**, Italian Institute of Health ISS / Environment Health; **M. Careggi**, Italian Institute of Health ISS; **O. Tcheremenskaia**, Italian Institute of Health ISS /
Microbial communities provide a large range of ecosystem services such as primary effects on ecosystems, it is essential to consider higher levels of ecological integration. Yet, it becomes increasingly recognised that to draw conclusions on the effects of the microorganisms to the environment, it is essential to consider higher levels of ecological integration. The field is considered in environmental toxicology and the assessment of environmental effects. 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 Hydodynamic conditions alter the tolerance of biofilm communities towards chemical stress

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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 stresses 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 hydrodynamics. Even though EPS composition is 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 BCR protocol. 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.
Bioaccumulation levels of Cu in the young biofilm (20 days) were similar between the colloidal, capsular and cellular fractions. Finally, the isotopic approach showed that after 40 days of exposure, the isotopic ratios in the three fractions of the biofilm were similar to the ratio in water of the second phase of exposure (0.25). These results suggest (i) an intense and rapid renewal of the biofilm and of the bioaccumulated Cu and (ii) that Cu concentrations in a mature biofilm at a given time reflect the last period of exposure. In addition, minor differences were 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 tetra- and hexavalent Zr (Zr) are very scarce. Zirconium (Zr) is a tetravalent non-radioactive element for which the global demand is increasing in the last decades. Benthic microorganism communities (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 in order to better understand their potential impacts on aquatic ecosystems. Glass slides were immersed in a pond in Cestas (near Bordeaux, France) for one month to be colonised by periphyton. They were distributed in 3 aquaria containing a synthetic culture medium (Dauta, 1982) and effective concentrations of 0.2 ± 0.1 nM (C0), 0.5 ± 0.3 nM (C1) or 2.9 ± 0.3 nM (C2) of Zr (µM). One slide per section was sampled after 1, 2 and 4 weeks of exposure. For each section, chlorophyll pigment fluorescence, photosynththetic activity, microscopical biofilm identification, polysaccharide and protein contents. Biomass, proteins, polysaccharides and diatom abundance increased significantly showing the growth of the biofilm during the experiment. No significant Zr exposure effects were observed on biomass, proteins, polysaccharides contents but their productions appeared to be slightly lower in C1 and C2 at t2 and t4. Diatoms growth rate in the C2 condition was significantly lower than in C0 and C1. Results obtained by pigments fluorescence measurements showed significant cyanobacteria decrease in the C2 condition over the exposure time as well as the brown algae between t2 and t4. Principal response curve (PRC) analysis showed significant changes over time of microcystinoma composition but no reference (C0) and the C2 condition, Giliates were less impacted by Zr exposure than flagellates which tended to disappear in the C2 condition. Biofilm microorganisms play a wide role in major ecosystem processes. Regarding these results, Zr exposure can impact the periphyton microorganisms composition which could disturb periphyton key functions. A better understanding of effects of metals on microcystinoma would improve risk assessment of metabolic exposure in aquatic ecosystems.

TU032
DNA metabarcoding demonstrates effects from copper at environmental concentrations on microbial diversity in marine periphyton biofilms
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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**

D.L. Carr, Texas Tech University / Biological Sciences; M. Dong, Texas Tech University / Biological Science

The spill of petrodiesel on land can irreversibly damage the soil ecosystem, and there are limited studies comparing petrodiesel and biodiesel impacts on soil microbial communities. Biodiesel has been considered as a viable substitute for petrodiesel, however, in some cases biodiesel is more microbial friendly than petrodiesel is inconclusive. Previous studies of soil microbial community on contaminated sites failed to reveal the dynamic changes of soil microbial communities. This laboratory study compared the effects of petrodiesel and three types of biodiesel on soil microbial communities in sandy loam soils. Contaminated soil samples were investigated at day 0, day 7 and day 180 to evaluate their effects on the composition and function of soil microbial communities. Biolog EcoPlates™ were used to test the microbial community functions based on carbon utilization while soil microbial composition were addressed by 16s RNA gene sequencing of V3-V4 regions. Results suggested that biodiesels were not statistically different from petrodiesel in terms of their adverse impacts on soil microbial communities. In conclusion, our results suggested that biodiesels should not be automatically considered as harmless substitutes for petrodiesel and that metagenome analysis could be an indicator of fitness. In the current study we investigated whether these population health indices could be applied to barn owls, Tyto alba. We were able to establish baseline “norms” in the form of Shehwart charts. For example the mean proportion of birds that were female was 48% with a prediction interval of 38-59%, and so years in which the prediction interval was extensive over line in the prediction intervals for the two sexes. Prediction intervals for the percentage of birds with low fat deposits were 23-65% and 28-75% for females and males, respectively. The level of kurtois within 10th primary feather weight precluded this metric from being used to investigate fluctuating asymmetry. This study shows that the proposed population health indices generally can be reported for barn owls. Establishing these population health indices can then be used to provide an early warning of whether chemical or other stressors are affecting the demography of barn owl populations.

**TU034**

**Evaluation of riparian groundwater quality using microbial response to potentiometric and climatic conditions**

O. CHAMSI, ECOLAB UMR 5245 CNRS UPS INPT; E. Navarro, CSIC - Spanish National Research Council / Dept. Reursos marinos renovables; J. Sanchez-Perez, S. Sauvage, ECOLAB UMR CNRS UPS INPT; F. Comin, CSIC - Spanish National Research Council; I. Arguedad, University of Pais Vasco; J. Bodoque, University of Castilla La Mancha; J. Charcosset, ECOLAB UMR CNRS UPS INPT; E. Pinelli, ECOLAB UMR 5245 CNRS UPS INPT

Contamination of ecosystems by pesticides, pharmaceuticals and trace metals becomes a major environmental problem. Freshwater algae are well known as bio-indicators of river pollution but diatom indices do not allow to evaluate the specific effects of the contaminants. Their sensitivity to pesticides differing markedly among microalgae species and therefore the toxicity data for multiple species need to be efficiently obtained. In the present work, we measured the growth of the three dominant species Desmodesmus subspicatus, Nitzschia palea and Navicula pelliculosa by an automated fluorometric microplate assay to evaluate the groundwater and river quality in four riparian wetlands in the south-west of Europe (Monbèquí (France), Saragossa (Spain), Bidasoa (Spain) and Toledo (Spain)). Four campaigns of water sampling were realized during contrasted hydrological conditions in agricultural area. Pesticides, pharmaceuticals and metals concentration were measured by HPLC-MS or ICP-MS. PCA, ANOVA and co-inertia analysis results showed that algal growth was different between freshwater and groundwater. As expected, the green alga was sensitive to alkalinity, SO₄, O₂ and pH whereas diatoms were positively sensitive to silica concentration and dissolved organic carbon (DOC). Besides, the green alga responded positively to the metals Co and Ni and negatively to S-triazines, terbutylazine and their metabolites. At last, the pharmaceuticals benzoylcoecmine and carbamazepine/ibresartan/valsartan induced growth stimulation of N. palea and N. pelliculosa, respectively. Same records for pharmaceuticals were observed for the other three sites, excepted Bidasoa. Both extensive sampling and data analysis makes our approach a new useful bio-indicator for preliminary investigation of groundwater quality in order to predict the best location of quality water for human consumption (ATTENAGUA project).

**TU035**

**Can trends in wildlife populations revolutionise our understanding of the impacts of chemicals on the environment? (P)**

L. Walker, Centre for Ecology & Hydrology; E.D. Potter, NERC Centre for Ecology & Hydrology / Lancaster; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; E. Shin, Centre for Ecology & Hydrology (NERC)

The Predatory Bird Monitoring Scheme (PBMS; http://pbms.ceh.ac.uk/) is national long-term project that monitors contaminant residues in a range of avian predator species. Each bird that is submitted to the scheme is given a post-mortem examination during which approximately 60 macroscopic observations and measurements are made. The information gathered during this examination could potentially be used to monitor health status of the birds at the time of their death or at a particular stage of their development. Previously we have focused on examining health indicators for the sparrowhawk, Accipiter nisus. We were able to establish baseline “norms” for indicators that could be broadly categorised as indicators of change in: (i) population demography because of altered recruitment, survival and mortality (measures were sex ratio, proportion of first-year birds, and proportion of deaths from starvation or disease); (ii) change in nutritional status (measures were body weight, fat score, condition index) that may be a pre-cursor for subsequent population impacts, (iii) physiological stress (as measured by fluctuating asymmetry) that may be an indicator of fitness. In the current study we investigated whether these population health indices could be applied to barn owls, Tyto alba. We were able to establish baseline “norms” in the form of Shehwart charts. For example the mean proportion of birds that were female was 48% with a prediction interval of 38-59%, and so years in which the prediction interval was extensive over line in the prediction intervals for the two sexes. Prediction intervals for the percentage of birds with low fat deposits were 23-65% and 28-75% for females and males, respectively. The level of kurtois within 10th primary feather weight precluded this metric from being used to investigate fluctuating asymmetry. This study shows that the proposed population health indices generally can be reported for barn owls. Establishing these population health indices can then be used to provide an early warning of whether chemical or other stressors are affecting the demography of barn owl populations.

**TU036**

**Identifying suitable marine biomonitor in South Africa: Mussels vs Whelks**

C. Sparks, Cape Peninsula University of Technology / Conservation and marine sciences; W. Samuels, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences; W. Samuels, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences; W. Samuels, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences; W. Samuels, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences

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 (B. lagenaria) as well as measure imposex prevalence in B. lagenaria 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). Three species of mussels (M. galloprovincialis, M. edulis, M. edulis) and two species of whelks (B. lagenaria, B. truncorum) were measured in intertidal sediment, M. galloprovincialis and B. lagenaria and imposex prevalence in B. lagenaria. 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 bioconcentations 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. lagenaria along the South African coast, which is not the case for M. galloprovincialis that only occurs on the west and south east of the country, the proposal is made that B. lagenaria 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)**

J.S. Paas-Knudsen, University of Oslo / Department of Biosciences; H.A. Sveinson, University of Oslo / Department of Physics; K. Borge, Department of Biosciences, University of Oslo / Department of Biosciences; M. Grun, A. Nielsen, University of Oslo / Department of Biosciences

Bees are increasingly facing multiple and interacting threats. One of the threats that have received increased attention lately is neonicotinoids: a group of systemic active pesticides that displace the neurotransmitter nicotinic acetylcholine (ACh) at synapses at local and systemic levels. The threat is considered to be an indicator of fitness. In the current study we present a new experimental method to quantify how bees affected by neonicotinoids in-field realistic doses. 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 PPPs and impact analysis based on EFSA Bee GD * R. Becker, BASF SE Agrarzentrum Limburgerhof; J. Lueckmann, Rifcon 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 show the relevance of 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 bees to PPPs applied as for-field and off-field (PPPs applied to flowers and seeds treated with granules) scenarios. Where a substance or use should not pass one of the screening level risk quotients, EFSA offers the possibility for refinement in 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 ECPC 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* L. Franke, Eurofins Agroscience Services Ecotox GmbH; R. Becker, BASF SE Agrarzentrum Limburgerhof; S. Schmitzer, IBACON GmbH; B. Szczeniak, Eurofins Agroscience Services Ecotox GmbH * on behalf of the ICP-PR Bee Brood Working Group and the Bee Brood Working Group of the German AG Bienenbeschutz 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. According to the guidance 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 (Oomen et al., 1992) as well as the OECD Guidance Document 75 (2007; OECD GD 75) are given as the two higher tier options to refine the risk on honeybee brood if concern is raised in tier I. Both methods focus on the brood termination rate (hereafter BTR) as the key endpoint. While the Oomen brood test investigates an artificial and worst case acute or chronic oral exposure scenario with a test item spiked feeding solution administered inside the hive (Lückmann & Schmitzer 2015) brood studies according to OECD GD 75 under semi-field conditions rely on a realistic contact and oral exposure scenario to bees comprising contaminated nectar and pollen after overwintering of a bee attractive crop. As the evaluation of historical data from field and 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 GD 75 semi-field and field trials and consider 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)? H. Borgen, G. Gonisur, M. Kleinheinz, B. Szczeniak, 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), brood control mortality (an indicator for 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 methods which allow for a better comparison of 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 Görligh (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 Görligh (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 screening phase as the production of actives is essential for the assessment of bee brood development of a whole colony (3 replicates, untreated) are compared to the results were 200 cells per hive were used. The parameters compared are: BTR, brood and compensation indices.

TU042 Ecotoxicological studies with bumble bees - latest developments and method improvement L. Franke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; J. Fricke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; J. Sorri, TRIALCAMP SLU; T. Vollmer, Eurofins Agroscience Services EcoChem GmbH / Field Ecotoxicology; S. Knaebe, EAS Ecotox GmbH / Ecotoxicology Field The publication of the proposed EFSA risk assessment for pollinators resulted in an increasing demand for experiments with non-Apis pollinators. As bumble bees (Bombus terrestris) and Apis mellifera, in order to gain an overview of these activities, as the production of actives is essential for the assessment of bee brood development of a whole colony (3 replicates, untreated) are compared to the results were 200 cells per hive were used. The parameters compared are: BTR, brood and compensation indices.

TU043 Higher-tier risk refinement of solitary bees in the field - is the well-known ‘focal species’ concept a suitable approach? J. Lueckmann, M. Faulp, J. Ludwigs, Rifcon 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 Working group non-Apis was recommended as well. 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...
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 terestris) versus honeybee (Apis mellifera) acute oral and contact toxicity - Preliminary results of ECPA company data evaluation A. Dinter, CEPI-PR Non-Apis Working Group; K. Amsel; N. Exeler; H.瞭, Bayer CropScience; J.C. Becker, New York State Dept. of Environmental Conservation / Biology; P. Campbell, Syngenta / Environmental Safety; M. Miles, Bayer CropScience UK / Environmental Safety; E. Pilling, Dow Agrosciences / REGULATORY Sciences; N. Ruddle, Syngenta Ltd / Product Safety; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Franke, EAS Ecotoxicology; A. Schnurr, BioChemagrar GmbH; A. Molitor, Eurofins Agroscience Services GmbH; E. SOLER, TRIALCAMP SLU / Ecotoxicology; S. Kimmel, Innovative Environmental Services (IES) Ltd / Ecotoxicology

A preliminary data evaluation was conducted by ECPA companies to compare the sensitivity of bumblebees (Bombus terestris) 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 data set evaluation of LD50 values 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 sp.) 10 day feeding laboratory test design: First results from an ICP-PR Non-Apis Working Test N. Exeler, Bayer AG, Crop Science Division; N. Hanewald, BASF SE / Ecotoxicology; C. Jenkins, Environ, 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, IBC&ON 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 change of existing risk assessment paradigms and testing approaches. The published and already revised EFSA GD on the risk assessment of PPP on pollinators includes also bumblebees and solitary bee species. In the need to address long term effects on bumble bees the ICP-PR Non-Apis working group designed a range 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 Dinitro EC 400 (Perfektion) was evaluated within a 10 day chronic exposure scenario. 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 trophalaxis 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 The ICP-PR Non-Apis Working Group (ICPPR Non-Apis Working Group) has been set up to develop a first-tier acute oral test for Osmania 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. Osmania bicoloris and Osmania 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. bicoloris and O. cornutus appeared to be rather similar, although O. cornuta showed a slightly less sensitive response, (which might be) due to its larger adult bodyweight. Hence, the LD50 values after 96 hours ranging from 2.6 – 7.1 μg a.i./bee indicate that a validated and workable methodology has been set up and a guideline is within reach.

TU047 A method for a solitary bee (Osmania spp.) first tier acute oral laboratory test: an update J. 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 Osmania 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. Osmania bicoloris and Osmania 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. bicoloris and O. bicoloris appeared to be rather similar, although O. cornuta showed a slightly less sensitive response, (which might be) due to its larger adult bodyweight. Hence, the LD50 values after 96 hours ranging from 2.6 – 7.1 μg a.i./bee indicate that a validated and workable methodology has been set up and a guideline is within reach.

TU048 2 Years of Solitary Bee Semi-field Ring Testing and Final Conclusions (ICPPR Non-Apis Working Group) S. Knaebe, EAS Ecotoxicology GmbH / Ecotoxic Field; N. Exeler, Bayer AG, Crop Science Division; L. Franke, J. Fricke, Eurofins Agroscience Services Ecotoxic GmbH / Ecotoxicology Field; M. Frommberger, Julius Kuehn Institut; T. Jutte, Julius Kuehn Institute; S. Kimmel, Innovative Environmental Services (IES) Ltd / Ecotoxicology; O. Klein, Eurofins Agroscience Services Ecotoxic GmbH / Ecotoxic Field; J. Lueckmann, Rifcon 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 ringtests were conducted in 2016 and 2017 to develop a general test set-up. The ringtest design was based on the EFSA guidance document, ÖEPF/EPPO Guideline No. 170 and recent recommendations on Solitary bee tests from the proceedings of the ICP-PR Non-Apis workshop in 2015, 2016 and 2017 followed by a workshop in 2017 to harmonise methodology. Ringtests were conducted with two representatives of a solitary bee species (Osmania bicoloris L. and Osmania cornuta Latr.; Hymenoptera, Megachilidae). These species are polyelectic and can forage on a diverse spectrum of flowering crops. They are nesting in cavities. Both are common species in Europe commercially available and are widely used for pollination services. Several laboratories participated in the higher-tier ring tests. 8 semi-field tests were done in 2016 and 9 in 2017. Two treatment groups were always included in the ringtest: an untreated control and dimethoate as a toxic reference item (optional other i.e. brood affecting substances (fenoxycarb)). In the study design adult bees were exposed in the tunnels during their reproductive period. Adult bees, as well as their offspring, were exposed to the treated pollen and nectar during development. Relevant endpoints for this study design are observations of the flight activity in front of the nesting units, nest occupation (i.e. number of nesting females), the production of complete cells and cocoons per female, the brood feeding system, the nest building materials and the size of the hives are quite variable. However, there are no specific methods for toxicity tests to stingless bees. So, in our laboratory we are developing and standardizing methods to test the toxicity of pesticides to species of stingless bees. We test the Acute Contact Toxicity Test protocol of OECD guidelines (214) established to European honeybee for the stingless bees Scaptotrigona postica and Melipona scutellaris. For this, we use a test method based on the bee test standard ICP-PR. Two groups were kept in 250 mL cages (ten bees were placed per cage, such that each treatment contained thirty bees from three colonies), fed in groups through microtubes (1.5 mL) punched in extremities, and kept in a chamber of biochemical oxygen demand (BOD) at 29 ± 2 °C, relative humidity of 70 ± 10% and in constant darkness. The diet used was composed of 50% (w/v) aqueous sugar solution. Our observations showed that it was possible to perform the Acute Contact Toxicity Test for stingless bees some adaptations in OECD 214 (2017) are necessary, like to adjust the temperature of the incubator (29 ± 2 °C instead of 25 ± 2 °C), anesthesia should be done by cooling and the time should be adequate for each species. The development of these tests will allow the development of safer strategies for the protection of biodiversity and, at the same time, support the expansion of agriculture, which is an important socio-economic activity in the region.
termination rate during the larval development as well as the success of emergence of their progeny (F1-generatoin) 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 Limburgherfo; Ecotoxicology; N. Heppenstall, 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. Apis mellifera) as well as wild (most non-Apis species) bees have emerged as an intensively discussed topic. In current risk assessment for A. mellifera are required to purely expose susceptible species 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 exposure to residues. Method failures and its data set was 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 - Methodology and recent experiences
M. Persiampel, 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 regulatory requirements for methods are required to assess exposure of exposed species in pollen and nectar. The methods used need to adequately reflect the properties of the tested substance and the circumstances of the application as well as potential influences of behavioural aspects such as foraging behaviour. Also, residue kinetics of a given substance have to be considered and must be reflected in the time points used for sampling. Here, we present recently employed approaches for studies which meet these new requirements. Method failures and 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 CGASQ; R. Rebelo, iBAMA / CCONP
Globally there are increasing concerns about possible decline in pollinators which requires that efforts be made in the direction of identifying its possible causes and in establishing policies for protecting pollinators. The Brazilian Institute for the Environment and Renewable Natural Resources (iBAMA) is responsible for environmental assessments in the context of pesticide registration in Brazil. Since 2011 iBAMA is implementing the risk assessment of pesticides in Brazil and one of the challenges is to adequately and efficiently establish a framework to protect pollinator insects against pesticides effects. In this context, it was published in February 2017 the Normative Instruction 02 (NI 02/2017) that establishes procedures to risk assessment of pesticides to pollinators. This is the first Brazilian specific regulation based on a risk approach, and in July 2017 iBAMA published a Manual of Environmental Risk Assessment of Pesticides to Bees which express an accessible way how the normative should be applied. NI 02/2017 is widely based on US/Canada’s approach, which means that it focuses on Apis mellifera data; the models used for screening are Bee-REX and AgDrift; tests required for tier 1 are the same and there is one scheme for foliar applications and other for soil/seed/trunk treatments. But there are few modifications: 4 tiers, the last one being post-registration monitoring; use of a safety factor of 10 for non-Apis bees; residue trials must be performed in Brazil and for tier 2 a crop grouping is considered. With this normative iBAMA expects that pesticides be used efficiently without incurring unacceptable risks to bees. Although iBAMA has a full framework for risk assessment established for honeybees there are still gaps in knowledge and research needs for ensuring that procedures to protect bees can be improved, especially regarding native bees. Hence, a matrix of selection for Brazilian bee species was proposed for selecting native species for use in pesticide risk assessment. This matrix provided the basis for electing meliponines (stingless bees) as a priority group. In the near future iBAMA intends to assess the need of changes in the risk assessment procedure, eventually including a stingless bee as a representative species.

TT053 How the new Brazilian risk assessment framework for bees works K.d. Coelho, ADAMA BRASIL / Regulatory Affairs; G. Weymann, ADAMA
The Environmental Assessment of pesticides in Brazil is performed by the Environmental and Institute (IBAMA) and comprises the following steps under the new Brazilian scheme: Environmental Hazard Potential Assessment and Environmental Risk Assessment. The Hazard assessment has been established since 1990 but the Risk Assessment, although required since 1996, only started to be implemented by IBAMA in 2012 and has been developing further since then. Due to numerous global discussions on the decline of pollinators, in February 2017 iBAMA published the first ruling (“normative”) to establish guidelines, requirements, and procedures for a systematic risk assessment scheme of pesticides for pollinators in Brazil. Further guidance for the scheme was issued later in 2017. The Brazilian overall approach is similar to EPA, but there are nuances in the Brazilian scheme regarding which active ingredients must be tested, and additional considerations for future which must be understood. Using hypothetical data from a mixture and a single ingredient we will showed how the new Brazilian framework (“normative”) works for two use patterns, foliar and soil application, considering the main aspects of Tiers 1 and 2 of the risk assessment. Furthermore, the main points of this Brazilian risk assessment framework for bees will be compared with those adopted by other countries, considering both similarities and differences. Keywords: Pollinators, bees, risk assessment, Brazil

TT054 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. Tachida, Chiba Institute of Technology / Creative Engineering
Neonicotinoid insecticides are widely used in Japan. Seven neonicotinoid insecticides such as Imidacloprid, Acetamiprid, Thioctroprid, Clothianidin, Dinofeturan, Thiamethoxam and Nitopyram are popular. Their usage began from the beginning of 1990 and is increasing till 2008 in Japan. Recent annual total usage of seven insecticides is not increasing, approximately 400 tons per year in Japan. However, many stingless bee species (except Apis species) have emerged as an intensively discussed topic. In current risk assessment framework for bees, risk assessment, Brazil countries, considering both similarities and differences. Keywords: Pollinators, bees, risk assessment, Brazil

TT055 Thiamethoxam Honey Bee Large Scale Colony Feeding Study - Design and Interpretation N. Ruddle, Syngenta Ltd / Product Safety; H. Thompson, Syngenta Ltd / Environmental Safety; F. Voehringer, Syngenta Crop Protection, LLC / Environmental Safety; C. Elston, Syngenta Ltd; M.A. Feken, Syngenta / Ecological Risk Assessment; S. Bocksch, Eurofins AgroScience Services Ecotox GmbH / Ecotox Honeybees; P. Thorbek, Syngenta / Environmental Safety; M. Hill, Eurofins AgroScience Services Inc
Colony feeding studies were originally developed to directly assess the insect growth regulating properties of neonicotinoids and designed to determine mode of action rather than effect levels. More recently there has been regulatory interest in conducting colony feeding studies to determine the pesticide level in nectar substitute (sucrose solution) which leads to colony-level effects, thereby allowing
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 similar 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 treating 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 embryonic lethal abnormal visual system (Elav), and and it is important for growth and connection of mushroom bodies, a center of the nervous system. Whereas that, Dscam gene can suffer alternative splicing from a high variable region and be able to generate more than 38,000 isoforms and is important for growth and connection of mushroom bodies, a center of the nervous system. Elav encodes proteins commonly used as neuronal markers in metazoans, which has action on post-transcriptional regulation and is required for differentiation and maintenance of the nervous system. Therefore, that, Dscam gene is important for pollinators, such as honeybees, has become mandatory in many countries. It is important to establish lines of action approved internationally to provide farmers and policy-makers more information about the applications of pest management programs. With this in mind, this work evaluated whether sublethal doses of the insecticide thiamethoxam, the fungicide carbendazim, and the herbicide glyphosate would be capable of altering the alternative splicing pattern of Elav in the brains of honeybees. The system should facilitate the use of standard hives and should be 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 honey bee 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 recent years a number of population models have been developed for honeybees and their foraging behaviour, such as forage beehives for use in 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 honey bee 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.

TU059 Automated waggle dance decoding M. Wang, WSC Scientific GmbH / Dept Efate Modelling; J. Kleemann, A. Görlich, WSC Scientific GmbH In honeybee field studies EFSA recommended in its latest guidance that field studies should ensure that the 90% 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. Görlich, WSC Scientific GmbH; M. Wang, WSC Scientific GmbH / Dept Efate Modelling A large variability of success rate and gene expression among treatment groups was found in the ten bees feeding group. Acute toxicity data with dimethoate showed that group feeding scheme with ten bees per cage resulted in higher mortality when compared to two bees (at same dosing levels). As consequence, the LD50 value is higher for the latter. High variability of homing success, gene expression or mortality rate in the ten bees feeding scheme is most likely caused by inhomogeneous dose distribution among bees, or either by over- or under dosing of single bees within replicates. A more accurate and uniform dosing distribution can be expected between 2 bees resulting in less variable data between runs, replicates and treatments. We highlight that feeding in smaller groups of honeybees should be discussed and considered to minimize the trematophagous dependence regarding food distribution in group dosed honeybees. Moreover, to compare endpoints of toxicological studies with single dosed wild bees for regulatory purposes. <br clear="all" /> [1]Broscheidt, R., Libor, A., Kupelwieser, V., Crailsheim, K., 2017: Food consumption and food exchange of caged honey bees using a radioactive labelled sugar solution - PLOS ONE | https://doi.org/10.1371/journal.pone.0174684
test power referring to the number of adults can be increased. Assessments of complete hives, including adults and all cells, make it possible to gain a detailed insight into the development of colonies and hive parameters over the course of time. Environmental factors and their influence on different hive parameters can be assessed and used to explain how these parameters either alone or in conjunction with plant protection products have an impact on the strength and development of honeybee colonies.

TU061
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 elicitor 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)

TU062
Assessment of Levels of Some Heavy Metals in the Organs of West African Dwarf Goat and Beef Cattle in Ogbomoso, Nigeria
A.A. Giwe, Cape Peninsula University of Technology / Department of Pure and Applied Chemistry; O.A. ADESOYE, Ladoke Akintola University of Technology Ogbomoso / Department of Pure and Applied Chemistry; P. Wewers, Cape Peninsula University of Technology / Chemistry
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 can eventually 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 to determine the levels of the metals by Atomic Absorption Spectrophotometric technique. Results show that there were significant differences (<0.05) in the concentrations of the metals in the different samples and sexes of the animals studied. The estimation of the non-essential metals in the investigated samples indicated the following range; lead: 1.11–6.00 mg/kg and Cadmium: 1.25–6.52 mg/kg while that of the essential metals are Zinc: 1.27–7.65 mg/kg, copper: 17.00–72.30 mg/kg and iron: 98.93–352.00 mg/kg. The results also revealed that the concentrations of lead, cadmium and iron exceeded the stipulated permissible limits. Higher-than-limit concentrations are observed more in the various parts of the tissue than in the two cattle species. There was, however, no significant difference (p>0.05) in the amount of these metals accumulated by both the samples and Doe. There was a major reduction in the results obtained using cooked samples when compared with raw samples for all the metals analysed. From the various data obtained, it can be concluded that all the five metals are present in all the samples analyzed and their average concentrations are significantly high in most of the samples. Cooking lowered the amount of the potentially toxic metals in the meat samples.

TU063
Assessment of metal bioaccessibility, bioavailability and toxicity in soil using the earthworm
P.Y. Robidoux, AGAT Laboratories, Ltd / Speciality services Division; Z. Omouri, INRS-Institut Armand-Frappier
Simultaneous contamination of ecosystems by various substances sets a challenge as regards to environmental assessment. One of the aspects is appraisal of pollutant bioavailability. Such an analysis was performed on a mixed contaminated site. The earthworm Eisenia andrei was exposed to different soils according to a contamination gradient. An integrated approach including a suite of biomarkers and chemical analyses was adopted to determine site toxicity. Parameters of the antioxidan system (catalase [CAT] and superoxide dismutase [SOD] activity), an enzyme of detoxification cation metabolism (glutathione S-transferase activity [GST]) as well as acid phosphatase (AP) activity and lysosomal membrane fragility of coelomocytes (neutral red retention time, NRRT) were used as tools. Overall toxicity endpoints (lethality, body weight change, reproduction) were assessed. Lethal effects were detected in some soils whereas chronic endpoints significantly decreased. A significant response of time-growing extent and consistency was recorded for SOD from to 2-28 weeks, whereas effects on other enzymatic markers were low and temporarily inconsistent. NRRT also was significantly decreased after 28 weeks of exposition. This is currently to the effect of metal exposure as well as a complete impairment in reproduction at 56 days. These results are revealing of early sub-lethal biological alterations in connection to contaminant toxicity and bioavailability. Bioaccessible bismuth (Bi) concentrations (using KNO3, soil extraction) were correlated to SOD activity and suggest an important contribution to the overall toxicity. Bi is used increasingly to replace lead in several industrial applications including the production of alloys and munitions formulations. However, little information is available on the environmental fate and ecological effects of Bi. This paper also summarizes the acute toxicity (LC50=416 mg Bi/kg ) and biacessibility of Bi, and describes bioavailability and chronic effects of bismuth on the earthworm Eisenia andrei. In reproduction tests, adult earthworms were exposed to natural sandy soil spiked with Bi citrate. Results indicate that Bi fraction form degraded by white suckers collected at a concentration 75 mg Bi/k 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. 

TU064
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? N. Urien, INRS-ETE / Centre Eau Terre Environnement; S. Jacob, P.-G. Campbell, P. Couture, Université Québec à Montréal / INRS / Centre Eau Terre Environnement
Metal exposure may lead to accumulation and toxicity in aquatic species. Once metals enter living organisms, they can penetrate into their cells and cause deleterious effects. Alternately, metals can be detoxified by binding to molecules designed to sequester them and prevent them from exerting their toxic effects, such as metallothioneins (MT) and metallothionein-like peptides (MTLP). MT and MTLP are mainly found in a body weight of the worms (30-42%) as well as in a complete impairment in reproduction at 56 days. These effects are revealing of early sub-lethal biological alterations in connection to contaminant toxicity and bioavailability. Bioaccessible bismuth (Bi) concentrations (using KNO3, soil extraction) were correlated to SOD activity and suggest an important contribution to the overall toxicity. Bi is used increasingly to replace lead in several industrial applications including the production of alloys and munitions formulations. However, little information is available on the environmental fate and ecological effects of Bi. This paper also summarizes the acute toxicity (LC50=416 mg Bi/kg ) and biacessibility of Bi, and describes bioavailability and chronic effects of bismuth on the earthworm Eisenia andrei. In reproduction tests, adult earthworms were exposed to natural sandy soil spiked with Bi citrate. Results indicate that Bi fraction form degraded by white suckers collected at a concentration 75 mg Bi/k 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. 

TU065
Assessment of Toxicological Impact of Anthropogenic activities on Onitsha Stretch of River Niger in Southeastern Nigeria
A.C. Udebuani, Federal University of Technology / Department of Biotechnology; J.J. Nwajuba, Federal University of Technology Owerri / Department of Biotechnology; p. Abara, federal university of Technology Owerri / Biology
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 influx 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
ch chromatography. The result obtained showed that six heavy metals with varying concentrations were obtained in the order of Zn>Pb>Cr>Cu>Cu>Cr>Ni. The HPI and MI values were far above the critical values. Results also showed EDCs to include PAH, phthalates, PCDs, PFCs, PBDEs, bisphenol A and PCBs. This study established that Onitsha 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. Thus, there 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 in heavy metal contaminated soil supplemented with sewage sludge
M. Ijskus, Czestochowa University of Technology / Institute of Environmental Engineering; A.O. Murtas, Czestochowa University of Technology / Department of Infrastructure and Environment; A. Grobelak, M. Kacprzak, Czestochowa University of Technology / Institute of Environmental Engineering

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). Concentration 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. The present 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 levels of MT was observed in plants grown under metal stress. The differences showed statistically significant changes between related conditions which means that presented assay can be used as a sensitive stress marker for phytotherapy process.

TU067 Chronic toxicity assessment of Ni contaminated rivers in Japan using Ceriodaphnia dubia for development of biotic ligand model for Japanese surface waters
H. Watanebe, M. Noguchi, T. Misaki, National Institute for Environmental Studies / Center for Health and Environmental Risk Research; N. Nakayama, National Institute for Environmental Studies / Center for Health and Environmental Risk Research; M. Ijskus, Czestochowa University of Technology / Institute of Environmental Engineering and Risk Research; S. Oda, National Institute for Environmental Studies / Center for Environmental Risk Research; K. Matsuoka, H. Yamamoto, National Institute for Environmental Studies / Center for Health and Environmental Risk Research

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 developed primarily on the basis of aquatic 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 the 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 Ceriodaphnia 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 (WHAM) 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.
aquatic organisms, nonetheless the reported medium guidelines. The last one is based on the results obtained in toxicology tests using concentration in the environment, however human activity increase its occurrence naturally in the environment, however human activity increase its occurrence naturally in the environment, however human activity increase its occurrence naturally in the environment, however human activity increase its occurrence naturally in the environment, however human activity increase its occurrence naturally in the environment, however human activity increase its occurrence naturally in the environment. Therefore, for the other study metal, Pb, any positive, biological function has been reported. All tests were run in transparent microplate (96 wells), and pH of test solutions was adjusted at 6.5. The algae growth was determined measuring the fluorescence (453/685 nm). In the first experiment, the microalgae was exposed for 72 hours to each metal using three different types of culture medium, OECD monochromated OECD 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, the EC50 after 72hrs were 140, >1200 and 293 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 6hr 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.

TU071 Ecological Risk Assessment of Trace Metal Contaminated Tropical Estuarine Sediment, Southwest Nigeria
A. Ugozie, University of Lagos, Nigeria / Department of Marine Sciences; O.L. Chukwu, University of Lagos Nigeria / Marine Sciences; R. Naidu, The University of the West of England / School of Biological Sciences, Faculty of Environment and Life Sciences; O.O. Ojewola, National University Of Science & Technology / Department of Biotechnology, University of Nigeria, Nsukka, Nigeria

This study was carried out in Wukari, to evaluate the Tenango Dam water and tilapia Oreochromis niloticus of the Tenango dam, Puebla, Mexico. M. Manooz-Najera, A. Barrera Escorcia, Universidad Autonoma Metropolitana Iztapalapa / Hydrobiologia; P. Ramirez Romero, U.A.M. Iztapalapa / Hydrobiologia Human population has seen the deterioration of resources derived from the over exploitation 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 that over time were declared protected natural areas. Population and 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, México, which is used for fishing, irrigation, recreation and electric power generation, among other purposes. The purpose of this study was to evaluate the environmental impact of metals accumulated in the sediments 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, metals: cadmium, chromium, cobalt and lead were determined in both water and tilapia. Results indicated that the environmental physicochemical parameters are within Mexican admissible ranges. Nitrates and lead exceed the acceptable limit for urban use and protection of aquatic life. Lead and chromium in water exceeded the limits of four collection, and tilapia, only in two of them. Cadmium and cobalt are registered in water behaved similarly exceeding in two seasons the levels allowed by the 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 and 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 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.

TU072 Effects of culture medium on metal toxicity and new approach for ecotoxicology assessment
G. Pascual, Tohoku University / Civil and Environmental Engineering; I. Garcia, N. Teran, 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 ecosystems 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 due to their increased use for commercial activity. However, for the other study metal, Pb, any positive, biological function has been not reported. All tests were run in transparent microplate (96 wells), and pH of test solutions was adjusted at 6.5. The algae growth was determined measuring the fluorescence (453/685 nm). In the first experiment, the microalgae was exposed for 72 hours to each metal using three different types of culture medium, OECD medium, monochromated OECD 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, the EC50 after 72hrs were 140, >1200 and 293 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 6hr 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.
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 560.59 and the lowest value was obtained in Arsenic with 1.289 ± 0.09. 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 throughout the world, as well as 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 which may cause this change; this action is still unknown. Nowadays, bivalve species are used in ecotoxicological bioassays due some particular characteristics, such as the wide distribution, ecological relevance, the capacity to filter and ingest large volumes of sediment particles and water and ease handling in the field and in the laboratory. Therefore, this work aims to determine toxic effects and changes in fatty acids profile composition of 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 organisms were exposed under laboratory conditions to copper sulphate to determine lethal concentration; at a second phase, it was conducted a compositional approach in the two nutritive 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 essential fatty acids (EFA), not only DHA and EPA, rates than C. edule. Still, big size class of both bivalve species is the most affected by the contaminant.

TU076 Metals removal from water for hazard classification
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Metals usually enter aquatic ecosystems in anoxic environment and associated with particles. It is important to know if the particulate-bound metals can be released to the water column as metal ions (ion exchange) or when resuspended by the organisms in the water column. The Transformation/Dissolution Protocol (OECD 29) is an established method that was modified to examine metal removal from the water column in anoxic 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 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
Modelling the chronic toxicity of copper to fish at low pH
S. Baken, European Copper Institute; K. De Schamphelaere, Ghent University
Applied Ecology and Environmental Biology
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 benchmarked 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.

Novel In-situ Toxicity Assessment of Sediment Capping Effectiveness in Deep Water (ECD 29)
G. Burton, University of Michigan / School for Environment and Sustainability; M. Hudson, University of Michigan / School of Natural Resources and Environment; S. Nedrich, A. Rentschler, University of Michigan; K. Thiamkeelakul, University of Michigan School for Environment and Sustainability; S.S. Brown, The Dow Chemical Company / Environmental Remediation and Restoration
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 sediment capping treatments, simulating lake-mesocosms. Simultaneous in-situ and ex-situ toxicity tests were conducted using Daphnia magna, Hyalella azteca, and Chironomus dilutus. Test organisms were protected from temperature shock by pre-acclimating 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.

REEchangE - Rare Earth Elements Ecotoxicology in a Changing Environment
H. Tien, Hamburg University of Applied Sciences/University of the West of Schleswig-Holstein; University of Applied Science of the University of Science; S. Heise, Hamburg University of Applied Sciences / Life Sciences
REEchangE focuses on the anthropogenic release of the rare earth elements (REE) lanthanum and gadolinium to the environment and their potential risk within ecosytems. REE are increasingly applied e.g. in green technology, and consequently also emitted to the 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 REEchangE addresses these topics in the following ways: (1) by studying the toxicity to aquatic organisms. Results on ecotoxic responses obtained for Aliivibrio fisheri and Rhaphidiidulcida subsphaerata 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 magnitude 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 specialised industries as prominent sources of emission. (3) by investigating the impact of changing environmental parameters (pH, redox, salinity) on the bioavailability of particle bound La and Gd. In a microcosm, a battery of miniaturized biotests will be applied to monitor the toxicity responses in overlying water and sediment. This includes tests with Aliivibrio fisheri, Vibrio proteolyticus Arthrobacter globiformis 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 new results of the microcosm experiments along with the information on bioavailability based on biotests and speciation data.

Sediment characteristics of natural and anthropogenic origin and their possible association with benthic macroinvertebrates in a minimally affected river in South Africa.
C. Wolmarans, H. Pienaar, G. Van Niekerk, NorthWest University / School of Natural Resources and Environment; F. Gonçalves, University of Aveiro / Department of Biology and Geosciences
Sediment characteristics generally entail metals, minerals, organic content, elements, particle size, particle size conductivity and pH. The origin of metals in sediment 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 structures in the Matz River, South Africa. Sediment samples were collected at 17 sites in a 50 km long stretch of the substrate at 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 Quanta 250 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 X2000µm had a detrimental effect on the biodiversity. No significant correlation was demonstrated in the relationship between the 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.

The effect of copper sulphate on the antioxidants enzymes activity of two size classes of Cerastoderma edule
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Anthropogenic activities, such as agriculture or industrial activities are the main source of pollution contributing to the degradation of the aquatic environments affecting the living organisms of the aquatic systems. Copper is often released into the aquatic systems, and may affect these ecosystems and its communities. Copper sulphate is a copper-based formulation, used in the agriculture practices to control pests. The main aim of this study is to determine the effects of copper in the antioxidant defense 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, GRd 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 toxicants.

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)

Even though ecotoxicological 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 Axellas 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, Pb and a mixture of these three metals under semi-natural conditions. In each bucket, we placed Axellas aquaticus, Daphnia magna, Cryptocorax riparius with different phytoplankton, and Elodea canadensis (macrophytes) and Raphidocelis subcapitata (algae). The theoretical metal concentrations were 1.5 µg/L Cd, 70 µg/L Cu, and 72 µg/L Pb. Half of the medium was renewed weekly. The effects of the metal mixtures and natural stressors were examined after 4 and 8 weeks, on the individual level (total metal accumulation, survival, shoot and root length), the population level (species densities, biomasses) and the community structure (diversity, evenness). Preliminary results show a high variability between replicates. We observed no significant differences in species densities between the metal treatments after 4 or 8 weeks. After 4 weeks, we found that Cu and the mixture temporarily negatively affected shoot and root length of E. nuttalli compared to the control treatment. However, after 8 weeks, we did not find these significant differences. As we could not find any significant effects of the metals at the end of the experiment, further research focused on sublethal factors or with a longer exposure duration is needed.

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 which inhabits the plough layer. The study focused on the relationship between soil properties and bioavailability of lead. We applied a mixture of lead with a 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 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. pHexp232 and pHporewater decreased with increasing total Pb concentration for all treatments, but pH decrease was much stronger for the soils with lower CEC and OC sorption. Desertion of Pb from the CaCl2 extracts could be well described by a Freundlich isotherm (R2 = 0.96-0.99) and Freundlich sorption constant Kp 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 pHexp232 (R2 = 0.87-0.94). The differences in Pb toxicity among soils correlated well with CaCl2 extractable Pb concentrations in soil (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 ecological risk assessment of metals in contaminated soils.

TU086 Toxicity evaluation of soils sampled in the vicinity of an Aluminum smelter in Montenegro using the Ames, Bioluminescence and DR-LUC biosays 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, Center for Ecotoxicological Investigations; H.A. Leslie, Institute for Environmental Studies VU Amsterdam This study conducted as a part of the national project ECOTOX 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 bioluminescent bacteria Vibrio fischeri 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 explain 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 undesired 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 QSARINS 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, Public Health Agency of Lombardy; 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) and the Lombardy Public Health Agency (APAS) have been working together to assess the comparability of the EC, approach (Effect Concentration affecting a percentage x of test organisms) to the current NOEC approach (No Observed Effect
In the framework of FP7 Project GUIDEnano we investigated the effect of different metal nanoparticles (NPs) coatings (synthesized from PlasmaChem GmbH, Germany) on 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 CeO₂ NPs and TiO₂ 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 also 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 TiO₂ 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 CeO₂ NPs coated with citrate whereas Ce was only detected in the stomach of fish treated with CeO₂ NPs uncoated or coated with PEG. After 7 days of depuration, Ce residues reached basal levels indicating a lack of accumulation of these CeO₂ NPs. These results indicate a different behavior for the CeO₂ NPs and TiO₂ NPs. No relationship could be observed between the coating and the observed effects. 

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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 medicines. 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 tools for environmental assessment
C. Fernandez Dacosta, University of Utrecht / Copernicus Institute; P. Wassenaar, National Institute for Public Health and the Environment (RIVM); I. Dencic, Corbion; M.C. Zijn, RIVM / Centre for Sustainability Environment and Health; A. Morao, Corbion; L. Shen, University of Utrecht / Copernicus Institute; E. Heugens, RIVM / Centre for Safety of Substances and Products; L. Posthuma, RIVM / Centre for Social and Environmental Hygiene.

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 assessment methods for 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 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 scope (excluding the environmental impacts of material and biomass feedstock production) and 2) omission of some environmental aspects relevant to bio-based materials and processes. 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
M. Markiewicz, Technical University of Dresden / Sustainable Chemistry Group; Y. Zhang, Dresden University of Technology / UFT Centre for Environmental Research and Sustainable Technology Department Sustainable Chemistry; S. Stolte, University of Bremen / UFT Centre for Environmental Research and Sustainable Technology Department Sustainable Chemistry.

Renewable energy stored in LOHC systems could replace fossil fuels yet their environmental impacts are largely unknown. This technology is still relatively new, and requires R&D efforts to optimise its performance to commercially attractive levels. This opens the possibility to proactively design the carriers for increased operational and environmental safety. A preliminary, comparative hazard assessment was performed using automotive diesel oil as a reference. The biodegradability and acute/subchronic (eco)toxicity using: enzymes (acylcholine esterase), cell lines (IPC-81), bacteria (Vibrio fischeri), algae (Raphidocelis subcapitata), freshwater plants ( Lemma minor) and invertebrates (Daphnia magna) were investigated. Test set included LOHC systems based on quinidine, ethyl-, propyl- or butyl- substituted 1,2-diarylethyl hydrogen. Within this study, we pinpoint limitations and 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 scope (excluding the environmental impacts of material and biomass feedstock production) and 2) omission of some environmental aspects relevant to bio-based materials and processes. 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.

TU095 1-Octanol and 2-Butanone as biofuel candidates - Using “Green Toxicology” for biofuel development
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The development and production of biofuels, and thus also the risk of a release in the environment, is rising. This implies an increased risk for a release into the environment. Aquatic systems are particularly considered to be vulnerable to fuel contaminations. (Eco)toxicological bioassays can be applied as screening tools during the early developmental phase of biofuels for obtaining information on potential hazardous properties. These biochemical 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. This 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 ecotoxicity 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 2-Butanone and 1-Octanol. 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 “Biofuels from biomass” funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

TU096 Investigation of the toxic effects of new mixtures of diverse edibleucetic solvents focusing on the environment and human health
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The development of environmentally benign and green synthetic protocols, due to the growing concern over the use of solvents, has been driven towards 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 deep eutectic 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 significantly lower melting point than that of each individual component. DESs have proved to be environmentally sustainable and alternative to the conventional organic solvents in synthetic chemistry, able to increase efficiency of organic transformations. These solvents have attracted widespread academic and industrial interests, and have found almost unanimous worldwide approval. Cosmetics has followed in the last years one of the most profitable industries. The majority of cosmetics are composed of chemicals, generally as emulsions. Given the ease of synthesizing DESs, along with their low cost, it is thought to be a possible use of them in the formulation of cosmetic and beauty products. Some of these DESs contain nitrogen (N), which can be used as fertilizer in the growth of the crops. Another field of interest could be the agriculture as well: some of these solvents can be used as pesticides, meaning a gradual release of the substance in the environment. Aquatic systems are particularly considered to be vulnerable to fuel contaminations. (Eco)toxicological bioassays can be applied as screening tools during the early developmental phase of biofuels for obtaining information on potential hazardous properties. These biochemical 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. This 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 ecotoxicity 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 2-Butanone and 1-Octanol. 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 “Biofuels from biomass” funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

New frontiers in Life Cycle Inventory data collection and modelling (P)

TI/097 Predicting environmentally beneficial production pathways for chemicals with neural networks
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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
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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 regulation trends 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 2008 expanded to include food in 2020, requiring the display 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 registrations in each country. However, Korea's agricultural and livestock LCA DB does not meet the ISO requirements, it is time to revise. The purpose of this study is to develop the LCI database for the estimation of the environmental footprint (PEF) of 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; R. Diacou, 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 common methodology, nomenclature and communication of the life cycle environmental performances for EF have been defined in a specific EU recommendation (2013/179/EU.). Within this framework, the International reference Life Cycle Data system (ILCD) format, developed since 2007, along with a simplified set of compliance rules called “ILCD Entry Level Requirements” has been recommended as a baseline for data development in the EF scheme. However, in the development of a database for the estimation of the environmental footprint (PEF) of 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/100 Tools for Environmental Footprint data checking and sharing: Soda4LCA, ILCD validator and Registry for the node management
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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.) - soda4LCA: 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 used for data from new nodes running on soda4LCA, and 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, 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 a time (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 PlasticsEurope LCIs in the perspective of the applicability of the improved assessment methods for Elementa of solvents
M. Baiz, 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 inconstancies in its eco-profile products, where use and consumption were somehow equated with water demand for 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 or 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 wherever the LCA practitioner is. This will be the basis for the calculation of the consumptive water (output minus input within the same drainage basin) and the application of AWARE. The presentation elaborates on the various operational uses of water in a chemical plant and the link to the life cycle inventory phase and ILCD flow name. This is also supported by 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 to LCA. The presentation aims to attack LCA water experts 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  
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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 a 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 dynamics (web tool DyPLCA, http://dypca.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. Therefore, LCA environmental interventions distributed in time, was then 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 clinging on to fixed characterization factors and time horizon and without compensation as done in conventional method. The use of dynamic LCA therefore gives a deeper 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 ILR; J. Godar, Stockholmn Environmental Institute  
That location matters when it comes to quantifying environmental impacts of agricultural commodities is well understood by the increasing use 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 perspective, these impacts can also be mitigated. This mainly requires the use of aggregated resource consumption and emission data for environmental extensions, which provides little detail on the technological and logistic 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 Trade 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 CO2-eq. 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 distribution of LCI data at further levels of the entire 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 (CGE) Models  
Y. Ichisugi, Tokyo City University; T. Masiu, 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 of a product. On the other hand, Integrated Model to Assess the Global Environmental and Governance (IO-GE) which can estimate economic efficiency in the future, based on price mechanism in the market. In this study, we collect fundamental data using LCA database and estimate GHG emissions in the future considering the supply chain among industrial sectors. We estimated GHG emissions in 2005 as a tentative result in Japan. The total emission is approximately 1.13E+048-CO2-eq. We confirmed the validity compared with the existing report published by the ministry of the environment in Japan. In the future, we will estimate environmental impact projection considering the scenario like Shared Socioeconomic Pathways (SSP).

TU105  
Network LCA as a tool to enhance data collection and usage in a value chain perspective  
T. Masui, National Institute for Environmental Studies; N. Itsubo, Tokyo City University; T. Masui, National Institute for Environmental Studies (NIES). This model uses Computable General Equilibrium (CGE) which can estimate economic efficiency in the future, based on price mechanism in the market. In this study, we collect fundamental data using LCA database and estimate GHG emissions in the future considering the supply chain among industrial sectors. We estimated GHG emissions in 2005 as a tentative result in Japan. The total emission is approximately 1.13E+048-CO2-eq. We confirmed the validity compared with the existing report published by the ministry of the environment in Japan. In the future, we will estimate environmental impact projection considering the scenario like Shared Socioeconomic Pathways (SSP).
Developing guidelines for elementary flow nomenclature
A. Edelen, ORISE; W. Ingwersen, US EPA

In general, a flow in life cycle inventory data refers to an input or output to a process. Flows may be of two broad types: elementary flows or intermediate (known as “technosphere”) flows according to ISO 14044 (ISO 14044 2006). Elementary flows may be defined as flows of energy, space that are used directly from the environment or released directly back into the environment. Life cycle assessment (LCA) data providers are currently not using a common list or system of elementary flows. An early activity within the UNEP-SETAC Life Cycle Initiative was the creation of a recommended list of flow exchanges by the Data Availability and Data Quality Workgroup (de Beaufort-Langeveld et al. 2003). Elementary flows in all life cycle inventory and life cycle impact assessment sources used in a model must correspond, or match, in order to build a functional LCA model. Edelen et. al. 2017 formulated recommendations on formatting and management based on a critical review of elementary flows from eleven LCA sources. These recommendations have been used to categorize flow information into three components and flow metadata into six components. These structured components of flows allows for systematic analysis and structuring of flow components through a knowledge organizational structure (KOS). The ISO 14048 standard was used to structure the different flow and metadata components as exclusive, inclusive or user-defined nomenclatures. The KOS is maintained in a user-friendly, publically accessible interface through the US EPA terminology services. This research presentation will focus on describing the benefits of the KOS approach and the tools used to build up of the KOS, to provide an example application of the KOS to current elementary flow nomenclature. References [1] de Beaufort-Langeveld A, Bretz R, Hischier R, Huijbregts M, Jean P, Tanner T, van Hoof G (2003) Code of life-cycle inventory practice. SETAC Press, Pensacola, FL [2] Edelen A, Ingwersen W, Rodriguez C, Alvarenga R, de Almeida AR, Wernet G (2017) Critical review of elementary flows in LCA data. INT J LIFE CYCLE ASS. https://doi.org/10.1007/s11367-017-1534-3 [3] ISO 14044:2006 Environmental management–Life cycle assessment—Requirements and guidelines. International Organization for Standardization, Switzerland

Building a Life Cycle Inventory of stormwater pollutant fluxes: model evaluation for a separate residential urban catchment
E. Risch, IRSTEA Montpellier / UMR ITAP; P. Roux, Istrea / ITAP ELSA-PACT; R.K. Rosenbaum, National Research Institute of Science and Technology for Environment and Agriculture - Istrea / UMR ITAP; C. Sinfor, ITAP, Istrea, Montpellier SupAgro, Univ Montpellier / ELSA Research group and ELSA-PACT Institute


Tissue specific 32P accumulation and consequent biological effects in bivalve molluscs
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1. Introduction
The aquatic environment is the main recipient of anthropogenic waterborne contaminants, 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 (32P).

2. Materials and methods
The study involved 10 day exposures of mussels to 32P of varying dose rates (i.e. 0.10, 1.0 and 10.0 mCi/dL) taking into account a current no-effect screening value of 0.24 mCi/dL (European Commission). The first set of studies determined 32P 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 tissue of interest; the digestive gland for example, received the greatest proportion of 32P independent of mussel species. In the next set of studies, a suite of biological responses of these treatments were investigated in digestive gland and gill cells. This included the induction of DNA damage (Comet assay) and repair response (Gamma-HAXZ), 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 0.1 mCi/dL in digestive gland (MN = also in gill) in both species, below the screening benchmark. Furthermore, compared to fresh water (DP), marine bivalve (MG) displayed an increased MN induction and DNA damage (both tissues) across all 32P 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.

Endocrine disruption in Mytilus galloprovincialis: Is ethinylestradiol a viable male contraceptive?
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Vitellogenins (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 mollusks, 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 synthetic estrogen 17α-ethinylestradiol (EE2) induces 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 uncontaminated 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 % mussel dry weight per day), representing negative and positive energy balance respectively. For the low feeding regime, shotgun proteomics identified an detectable response to an important radionuclide, phosphorus-32. This result is significant as it supports the idea of Vtg being a marker of endocrine disruption in aquatic environments. In the next set of studies, a suite of biological responses of these treatments were investigated in digestive gland and gill cells. This included the induction of DNA damage (Comet assay) and repair response (Gamma-HAXZ), the induction of micronuclei (MN) and the expression of key stress related genes (i.e. SOD, CAT, GST, HSP70/90).

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The added value of using invertebrate species in ecotoxicology: new insights for environmental risk assessment (P)
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 benthi and pelagic diaply effects on the offspring and 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. As such, it is important to examine 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 understanding of pathways involved in effect-based studies. Furthermore, it may be used as a biomarker in ecotoxicology to better understand the adverse effects that may 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 analysis 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, molting 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. Moreover, it may be used as a complementary approach to ecotoxicology to better understand 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 amphipod 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 pollution 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 molting rates were recorded. Mortalities in the Imidacloprid treatments clearly differed for amphipods sampled at two different sites; animals from more polluted sites were more sensitive to the toxicant and also exhibited reduced movement and molting 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 Paralyhe Havaiancisi as a Possible Endpoint in Ecotoxicology - Preliminary Data

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Regeneration of antennae of the marine amphipod Paralyhe Havaiancisi as a Possible Endpoint in Ecotoxicology - Preliminary Data

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Paralyhe Havaiancisi is a marine amphipod of worldwide circumtropical distribution, which has been used as an ecotoxicological test organisms. Paralyhe Havaiancisi 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. havaiancisi has local progenitor cell in each part of body that was already demonstrated that P. havaiancisi 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. havaiancisi to determine the viability this endpoint on toxicity tests. On day one left antennae of six months old organisms were amputed 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 a week, another pictorial control was done to determine the difference between the length (mm) before and after full regeneration. 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 toxicants to determine their ability to regenerate in the selected experimental conditions. Acknowledgements: Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq-PVE Process: 400362/2014-7) for funding and PIBIC for undergraduate 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 gaia 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 can be assessed. A screening test addressed to 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 J. Lucias, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences; B.G. Struyman, Cape Peninsula University of Technology / Biodiversity and Conservation; J. Odendaal, Cape Peninsula University of Technology / Department of Environmental and Occupational Studies 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 and biodiversity. Invertebrates are highly sensitive to changes in water quality and quantity. Numerous pollutants result from these activities, with metals being particularly prevalent in most urban rivers. These pollutants are known to affect freshwater macro-invertebrate communities. In South Africa, SASS5 (South African Scoring System for Invertebrates) is a river health index that studies the invertebrate assemblages within specific riverine microhabitats, and can assist in showing the relationship between water quality and different communities. The Olifants River in the Western Cape, is not only recognized as a hotspot for freshwater biodiversity, but seen by many, until the previous decade, to be the last pristine river along the South African coastline. It is also one of the three main feeding rivers to the City of Cape Town for fresh water. Despite increasing urbanisation, the last State-of-Rivers Report for this river was published in 2006. It has therefore become crucial to investigate the current degree of pollution within this river, as well as the general integrity of the system. This study aimed to determine the degree of metal pollution along the length of the Olifants River, as well as to investigate the effects of pollution and land use on the invertebrate communities, using SASS5. Water and sediment samples were collected seasonally at 5 sites from upper to lower reaches, acid digested and analysed with an ICP-AES for metal concentrations. Invertebrates were also sampled seasonally, identified and scored according to SASS5 sensitivity scores. An Average Score Per Taxon (ASPT) was calculated for each site. The results showed a general trend of increasing sediment metal concentrations, land use practices and habitat alterations, with concomitant decreasing ASPT’s, from site 1 to site 5 indicating a loss of certain sensitive species at the most impacted downstream sites. Although metal pollution was found to be relatively low, a cocktail of pollutants, coupled with structural alterations, are clearly impacting the health and integrity of this river system. A future study should focus on organic pollutants, as agriculture is one of the main land use practices in the area.

TU117 QWATER - Bioassay integration under the European Water Framework Directive?: A step towards an ecological approach M. Martinez-Haro, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; P. Acuëdo, IREC-Instituto de Investigación en Recursos Cinegéticos; A.I. Pàis-Costa, MARE-EBD; I.R. Vieira, ICegas & CIMAR, University of Porto / Department of Populations Studies Lab of Ecotoxicology; J.M. Neto, MARE; M. Taggart, University of the Highlands and Islands / Environmental Research Institute; N. Álvarez-Ospina, Universitat Potsdam; L. Guillermino, ICegas & CIMAR University of Porto / Department of Biology; R. Ribeiro, Universidade de Coimbra / Life Sciences; J.C. Marques, TO116 - M. Morandi / Department of Environmental and Ecological Studies The Water Framework Directive (WFD) is the most important piece of water legislation in Europe. It aims at ensuring the ‘good water status’ of EU water bodies and includes both chemical and ecological status. To achieve and assess a ‘good ecological status’, the WFD advocates the integration of various lines of evidence, and demands a set of low-cost tools and techniques to deliver appropriate data. The WFD accounts for chemical and ecological evaluation, but requires the establishment of cause-effect relationships in the assessment of environmental quality. Under this approach, the European Marine Curie QWATER project was aimed to gauge the ecological relevance of integrating short-term toxicity bioassays and biomarkers into quality elements in the WFD, as these may contribute to our ability to assess and manage EU water bodies. Ecologically relevant in situ cost-effective toxicity bioassays were used in a battery of in situ bioassays using representative species for several key functions in the ecosystem. Biomarkers, determined on the individuals used for the bioassays, were also integrated. Principal component analyses (PCA) were performed independently for each source of information, in order to improve the interpretation of the resultant PCA-factors in biological terms, and to verify whether the integration of ‘quality elements’ (bioassays and biomarkers) did (or did not) strengthen the robustness of the standard Ecological Quality Status approach used to assess water quality. Results shown some discrepancies in the water quality determined from each independent factor, i.e., the sampling sites were not equally ordered by all factors. Therefore, only by interpreting the values of all PCA-factors together is possible to perform a complete assessment of water quality status. The European regulatory authorities are presently in the phase of implementing the WFD based on community level approaches all over Europe. Interestingly, bioassays developed here and biomarkers, are available tools to be introduced as new feasible, cost-effective and sensitive protocols in the WFD. It might then become pertinent to stand for the combined/complementary use of ecological indicators and markers, and to develop Ecological Quality Status of water bodies. Overall, this project allowed us to efficiently contribute to national and international efforts focused in evaluating the water quality in European water bodies.

TU118 Chronic testing of mayfly and stonefly species - Development of a new approach M. Brüggemann, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; K. Schlich, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; C. Schlich. Fraunhofer IME - Institute of Research in Aquatic and Environmental Sciences Aquatic organisms, especially lotic invertebrate species originating from running waters, are exposed to releases of plant protection products which are mainly used in agriculture. Since lotic invertebrate species are regarded to be very sensitive but are hardly considered in chronic ecotoxicity testing, we developed a test system in order to investigate chronic effects on mayfly and stonefly species. After the successful development of a chronic toxicity test with the non-stonefly larva Protonemura sp. the next step was the establishment of a method for testing of mayfly species Epeorus sp. In the developed test system, contrary to usual indoor stream systems, not the water body itself, but test vessels inside test containers are circulated, thereby creating a target flow. The test containers are filled with medium and contains ten replicates each. Each replicate is a small cage, which serves as individual compartment for individual testing of one test organism. The surrounding is adapted to the natural habitat of the test organisms. As endpoints growth, emergence and mortality are observed during a 21 day exposure period. For the studies we use field collected larvae which are adapted to laboratory conditions.
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. As described above, stonefly 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 chemicals testing in such different aquatic invertebrates, 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 Toxic 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 126.55 μg L⁻¹, and the equivalent to 126.55 μg L⁻¹ CAR of a formulation (dissolved in dechlorinated tap water) with 85% of the active compound. The concentrations used were so as to have the same molarity as acetophenone (ATP) responses exerting their toxicity at the same insecticidal levels. 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 (ChEs), carboxylesterases (CEs) with two substrates, glutathione S-transferase (GST), glutathione (GSH), superoxide dismutase (SOD) and catalase (CAT). In a second bioassay, three 100 mL glass containers per treatment were used with a single recently-laid egg mass each. The time and success of hatching were registered and, after one month, the survival of the offspring was evaluated. In both 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 CAR concentrations (28 and 83%, respectively, compared to the solvent control). The formation of 2′,3′-dihydroxyacetophenone was decreased by 23% and inhibited CAT activity by 47% (compared to the water control).

Regarding the reproductive endpoints analyzed, no toxic effects were found neither with the active compound nor the formulation. Our findings show that a subchronic exposure of B. straminea to CAR, active compound or formulation, does not affect the primary target, ChEs. However, other toxicity pathways, in which antioxidant enzymes are involved, seem to be affected by this insecticide, mainly by the commercial formulation.

TU120 Toxicity of lanthanides to freshwater microcrustaceans
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The application of lanthanides (Ln) in different sectors of the world economy has significantly increased during the last two decades. This process has been accompanied by the increasing concerns on the effects of these elements on the environment. The anthropogenic anomalies of Ln in soil, surface water, groundwater and even in tap water have already been registered. The disruption of the natural biogeochemical cycle of Ln increases the risk of biota being exposed to elevated concentrations of Ln. However, the ecotoxicological effects of these elements and their fate in the environment are still insufficiently understood. The toxic concentrations reported in the literature, e.g., for Daphnia magna, noticeably vary presumably due to different test conditions. For this study, acute ecotoxic testing of La, Ce, Pr, Nd and Gd nitrates to freshwater crustaceans Daphnia magna (48 h) and Thamnocephalus platyurus (24 h) were performed in synthetic freshwater and natural lake water.

Also, long-term (21 days) exposure of D. magna (OECD 211) in lake water was included. It was shown that the Ln fractionation between two main phases (precipitated or settled or remained in the water column) changed during the tests depending on (i) water composition, (ii) nominal concentration, (iii) exposure time, and (iii) tested chemical element. Therefore, nominal concentrations were used for toxicity calculations. Acute toxicity of investigated Ln to both crustaceans was similar for all Ln, except for Eu, La, Ce, Nd and Gd, which is consistent with the results from long-term experiments. The nominal concentration, (ii) nominal concentration, (iii) exposure time, and (iii) tested chemical element. The toxicity of Ln increases the risk of biota being exposed to elevated concentrations of Ln. The most toxic to both species, however, difference between Eu/Ln values for Gd and other Ln was statistically significant (p<0.05) only in T. platyurus. In the lake water, bioavailability of Ln was much lower: mortality of exposed organisms did not exceed 23% at the highest tested concentration (50 mg L⁻¹). In contrast to acute assay, the 21 day chronic test performed in the lake water showed high Ln toxicity to D. magna (0.2 to 0.5 mg L⁻¹). It was revealed that mortality was a more sensitive endpoint than reproduction. Differences between Ln of individual Ln were not statistically significant. Thus, our results support the hypothesis that different lanthanides have a similar mechanism of toxicity in crustaceans. This work was supported by Estonian Research Council grant IUT23-5.

TU121 Relevance and suitability of invertebrates swimming behavior as sub-lethal endpoint to be considered for ecotoxicological investigation
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Ecotoxicology is aimed to assess, monitor and predict the effect of contaminants in the environment. Looking for new and alternative approaches in this discipline has become of increasing importance. Furthermore, within 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 early sub-lethal toxic effects at lower 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 crinoids, crustaceans, rotifers and echinoderms. In 10 years of research, the 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 SBR’s applications, which show the relevance, sensitivity and versatility of the swimming speed alteration of marine invertebrates as ecotoxicological endpoint. These data also contribute to support the hypothesis by which behavioral endpoints, such as swimming alteration, represents attractive approach that should be taken into account in ecotoxicological risk assessment.

TU122 Benefits of Using Ecologically and Economically Valued Invertebrate Species for Ecotoxicological Analyses: Potential Phototoxic Effects Comparing a Freshwater Vertebrate and Invertebrate
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In Louisiana, crayfish are not just a standard invertebrate species found in bayous and rice fields but also a staple in the cuisine and culture. Over 82 million pounds of crayfish are harvested annually, resulting in a $45 million industry; therefore, they are both ecologically and economically valued in the region. In areas, such as Louisiana, where an invertebrate species is of such importance, incorporating that species into ecotoxicology testing may benefit the overall risk assessment for the chemical in question and any potential effects to the organism itself. For example, dicloran, the active ingredient in the fungicide Botran®, which is used throughout Louisiana, shows phototoxicity; the toxic and phototoxic impacts of dicloran were analyzed using a vertebrate and invertebrate species (fathead minnows, Pimephales promelas, and red swamp crayfish, Procambarus clarkii). Fathead minnows showed negative impacts at concentrations as low as 0.1 mg/L and >90% mortality at 0.75 mg/L and red swamp crayfish showed negative impacts at concentrations ranging from 0.50-1.0 mg/L; the effects at similar concentrations show that P. clarkii is a useful, nontraditional organism to be used 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
N.B. Martins, University of Minho, Department of Biology & CBMA / Department of Biology; A. Pradhan, University of Minho / Department of Biology; F. Cassio, University of Porto / Department of Biology;
Biomagnification in corals can occur through both filter and suspension feeding. It is a conundrum to classify uptake mechanisms from all sources. Management; H. Kylin, The GE aldehydes were created for all substrates, where it was possible. 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 biomagnification and reproductive effects 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. Y. Cao, University of Copenhagen / Department of Plant and Environmental Sciences; M. Gottardi, University of Copenhagen / Plant and Environmental Sciences; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences Quantity of acetylcholinesterase (AChE) and other esterase activities is 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 (D. magna) and Chironomus riparius (C. riparius). The aim of this comparison was to determine 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 acetylthiocholine iodide as substrate, measuring resorufin production; 3) GE-assay using 1-naphthyl acetate (1-NA) as the substrate, measuring 1-naphthol production and 4) GE-assay using 4-nitrophenylbutyrate (4-MUB) as the substrate, measuring 4-nitrophenylbutyrate 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 the two aquatic invertebrate species D. magna and C. riparius. Each assay was performed with a 24-hr pre-treatment of this compound to determine the efficiency and selectivity of the four methods. The rotifer was exposed to chlorpyrifos (CPF) and pirimiphos-methyl (PM) at the NOEL and LOEL levels under chronic exposures to TCE at 8.0°C, i.e. about the mean annual temperature of groundwater in the cave. To this end, we used a sealed glass microplate equipped with planar oxygen sensor spots with optical isolation glued onto the bottom of 80-μL wells (LoLoLight, Denmark) in a standard-based respirometry system (SDR Sensor Dish Reader, PreSens, Germany). The system allows simultaneous measurement of up to 20 replicates and 4 controls. Survival and locomotory activity assessments were performed by counting the number of alive individuals and measuring the number of moving animals in 5 mL glass vials each containing 20 individuals.

TU125 Factors influencing bioaccumulation of metals and pollutants in corals V. C. Paço, University of Minho Centre of Molecular and Environmental Biology CBMA Department of Biology 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. Anti-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 5-Fluorouracil as an antimitabolite (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 biomagnification and reproductive effects on reproduction, cellular effects were found with possible consequences for the community at the long term.

TU126 Survival, metabolic rates and locomotory activities of a groundwater-obligate copepod species under long-term exposures to tetrachloroethylene T. Di Lorenzo, Institute of Ecosystem Study of the CNR Firenze; L. Piccini, University of Florence, Department of Earth Sciences; D. Galassi, University of L'Aquila; G. Messana, Institute of Ecosystem Study of the CNR; M. Saena, PRIET CONCET, National University of Luján; W.D. De Marzo, PRIET CONCET; PRIET Tetrachloroethylene (TCE) is a contaminant frequently found in groundwater of industrialized areas worldwide. The degradation of this chlorinated aliphatic hydrocarbon (CAH) is often incomplete in groundwater and takes several decades. Contamination from TCE is considered persistent and difficult to remediate, due to its high density that favors a gravity-driven vertical infiltration into groundwater bodies. Through means of the Water Framework Directive the European Union has demanded Member States to provide TCE threshold values (TV) for assessing groundwater body quality. In Italy, TCE TV is 1.1 µg L−1 in groundwater bodies. Studies on surface water species have shown that TCE causes oxidative stress in some fish and invertebrates. However, the effect of this contaminant on groundwater-obligate species has not been investigated to date. More importantly the effect that 1.1 µg L−1 TCE may have on groundwater species under chronic exposures is unknown. In this study, we investigated the effect of 1.1 µg L−1 TCE on survival, oxygen consumption, and locomotory activities of a groundwater-obligate copepod species (Moraria sp.) under different time exposures. The specimens required for the trials were collected in the Antro del Corchia Cave (Tuscany). We measured the individual-based oxygen consumption of these species as a proxy of possible metabolic reactions to long-term (4 > days) exposures to TCE at 8.0°C, i.e. about the mean annual temperature of groundwater in the cave. To this end, we used a sealed glass microplate equipped with planar oxygen sensor spots with optical isolation glued onto the bottom of 80-µL wells (LoLoLight, Denmark) in a standard-based respirometry system (SDR Sensor Dish Reader, PreSens, Germany). The system allows simultaneous measurement of up to 20 replicates and 4 controls. Survival and locomotory activity assessments were performed by counting the number of alive individuals and measuring the number of moving animals in 5 mL glass vials each containing 20 individuals.
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 transcripts of phase I (CYP1-like, CYP2-like, CYP2A14 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, influencing its influx through the plasma membrane into the intracellular compartment and enucleating pyrene metabolism. 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 CYP2A14 gene in the biotransformation process of PAHs in gills of C. brasiliana.

TU128 BIOCHEMICAL AND CELLULAR RESPONSES OF THE CRAB PACHYGRAPSUS 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 Pachygrapsus marmoratus. This investigation is part of the IMPACT project (Port Impact on Marine Protected Area: cross-border co-operative actions), which has the purpose to design cross-borders 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 marine protected area (MPA), considered a less contaminated area. The aim of this study was to evaluate the impacts caused by the exposure of Ti (II) solutions with the initial concentrations of 5µg/L, 50µg/L, 100 µg/L of Ti (II) (P). 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 electron transport system (ETS) activity, which decreased along exposure time. Mussels exposed to Ti decreased their glycogen (GLY) and protein (PROT) contents. Moreover, contaminated individuals activated their antioxidant defences increasing the activity of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPX) and glutathione S-transferases (GST), which still were not enough to prevent cellular damages. Several significant factors were revealed by the increased of lipid peroxidation in mussels exposed to Ti. 

TU130 Comparing interspecific Artemia responses to chronic zinc exposure A.P. Costa, Marine and Environmental Sciences Centre / Faculty of Sciences and Technology, University of Coimbra; I. Varo, CSIC Spanish National Research Council / Biology; culture and pathology of marine species; M. Martinez-Hurtado, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; P. Almeida Vinagre, WavEC - Offshore Renewables / Marine Environment and Public Policies; m. sanchez, CSIC / Wetland Ecology

The invasive species Artemia franciscana is displacing native Artemia (A. salina and A. parthenogenetica) from eastern Atlantic coasts and across the Mediterranean region as the result of increased shipping traffic and the introduction of exotic marine species. The experiments were carried out in laboratory controlled conditions, in order to select and expose the ephyrae (subadult) of the jellyfish Aurelia aurita to different levels of zinc and to compare their responses to those of the Artemia species. The aim of this current investigation is to suggest the use of two new invertebrate species of the jellyfish Aurelia aurita as model organisms in ecotoxicological bioassays. A. franciscana species have been 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 biologic organization. In addition, compared to vertebrates they are easy to maintain under laboratory conditions, widely distributed and ecologically relevant. Although Cnidarian jellyfish (Scyphozoa) are known to play an important role in marine foodwebs, genera and species of jellyfish 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 aurita 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 lethal, frequency of pulsation and acute, immobilization 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 EC₅₀
values obtained exposing epibryal jellyfish to different toxic compounds and materials such as nanoparticles and microplastics with those obtained with other marine invertebrates, highlights that epibryae 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 improvement 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, that can be considered as 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 used 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 nauplius 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 influence by the effluent of the main waste-water treatment plant (WWTP) of Bilbao. The WWTP effluent was extracted using a sequential LC-UV fractionation methodology based on two different columns: a Nucleodur C8 column (21 fractions were collected) and an aminopropyl column (15 fractions). Two endpoints were used to identify 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 spectrums, Metflag and Compound Discoverer (Thermo) interfaced to MZmine. Among the collected C18 fractions, only fraction 13th (F13) 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 EC10 =19 REF) could be explained by the contribution of active F13 (EC10=14 REF and EC50=39 REF). Regarding the chemical analysis, among the final candidate list (20 compounds, mebendazole (an anthelmintic agent) was confirmed chromatographically with standards. Nevertheless, a sequential fractionation of F13 was also carried out with an aminopropyl column, which showed a different orthogonality compared to C18 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 CMT2014-56628-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 epoxygenase 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 toxicant 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 the web flora genome database. After the BLAST and alignment, the genetic information of 10 key eicosanoid genes, such as pla2, cox, pgd2a and pg2e, 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 transcription of pla2 gene was commonly down-regulated by all the eicosanoid targeted drugs. Interestingly, some genes, such as pgd2 and gskl, were responded to certain specific drugs, celecoxib and ibuprofen, respectively. Through this study, we found that the nine of ten genes were expressed by exposure to the eicosanoid targeted drugs in *D. magna*. We believe that these results partially indicate the plausibility of *D. magna* as a model species for eicosanoid pathway related toxicity evaluation. Therefore, the genes expressed in this study could be used as suitable biomarkers for the eicosanoid related toxicity assessment.

**TU135**

Responses to single chemical and pulse exposures of two monophyletic *Daphnia* species under a multi-generation approach

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Planktonic human activities are increasingly increased in relation 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 pulse and chronic tests. To engage celerity, 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 and chronic tests to K+ and Ca+ (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, excepting *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 BPSf in *Daphnia magna*

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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-isopropoxyphenylsulfone (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 appear 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

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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 EC regulation (2003/204/EC) on phthalates of MEPH acts as one of the most 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 (GST), 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 / DSTA; D. Garagna, ChemService s.r.l.; F. Casarotto, University of Insubria; M. Neri, ChemService srl - Controllo e Ricerche Laboratory ecotoxicology 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 2008/588/EC on phosphates of MEHP acts as one of the most disrupting chemical in aquatic organism such as Daphnia Magna. In recent years even the use of first instar larvae of Chironomus riparius (Insecta, Diptera) has been proposed (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 Test Guideline n. 233, 2010). Chironomus riparius is generally used to test the quality of drinking water and to the first day in the water column before becoming benthic for the other three larval stages. As a matter of fact, the Chironomus acute test can be a useful tool to control the conditions/sensitivity of the breeding organisms in the lab. Therefore it can be of interest to understand its relevance when compared with the answers of other organisms, belonging to other taxa and with different life cycles. In order to compare the responses of Daphnia magna and Chironomus riparius when exposed to the same contaminant, we carried out different exposures using three different substances: two reference items (potassium chloride, and potassium dichromate, commonly used to test sensitivity of C. riparius and D. magna respectively) and an unknown toxicant (a fatty acid C14-C20). Preliminary results indicate possible discriminatory power among both test organisms: if confirmed by definitive tests these observations may represent a warning when carrying out acute toxicity tests on water medium, confirming the importance to test different trophic levels and showing the need to further investigate the use of the acute test on Chironomus riparius according to OECD 235 to assess the acute toxicity on aquatic organisms.

TU139 Analysis of mixtures of biphenol A and UV filters Octocrylene and OD-PABA on Chironomus riparius using a specific RT-PCR array

A.M. González, UNED / Mathematical Physics and Fluids; J. Martínez-Guitarte, UNED / Física Matemática y de Fluidos Ana-Belén Muñoz-González, José-Luis Martínez-Guitarte, Univ de la Biological and Toxicological Environment. Facultad de Ciencias, UNED. Madrid (Spain)/Keywords: UV filters, BPA, RT-PCR array /The ultraviolet (UV) filters are main components of many personal care products (PCPs) that are extensively used. They are organic compounds defined as emergent contaminants, which are increasing their presence in the environment because their use in recreational and industrial activities. Described as endocrine disruptors in vertebrates, their effects on invertebrates have been poorly studied, especially in mixtures. On the other hand, Bisphenol A (BPA) is as plasticizer used in packaging and other industrial products with confirmed endocrine disruption activity. In this study we have used two common UV filters, octocrylene (OC) and 2-ethylhexyl 4-dimethylaminobenzoate (OD-PABA), and BPA to mimic the putative mixtures resulting from PCP and interaction with plastic of PCP containers. These mixtures were reach the biota of freshwater ecosystem so the main objective was studying of the effects that the mixtures can have on an 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. Effects of DEHP, BPA and OD-PABA was obtained by retrotranscription and Real-Time PCR using a specific array covering a number of relevant 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, CICYT (SPAIN), C2MT2015-64913-R, a F. M. G is the receiver of a pre-doctoral contract from the National University of Distance Education (UNED).

TU140 Genetic variability in tolerance to microbial insecticides in Chironomus riparius

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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 by increasing the potential to adapt to changing environmental conditions. Ecological risk assessment needs to couple quantitative genetic analysis with ecotoxicological studies in order to understand the mechanisms underlying evolution of tolerance. Research on genetic variation regarding tolerance to contaminants has been mostly performed with clonal lineages. An alternative methodology has been applied for sexually reproducing organisms, which is based on the use of inbred strains or random-bred populations, estimating genetic variation in fitness traits. However, additional environmental stressors are not usually considered, limiting the predictive capabilities and determination of tolerance costs across different scenarios. 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 emergence, and survival 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.

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 froghopper, aphids and cotton pests. The use of Amitraz in water bodies at high rates, 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 269
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 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/AGA-MAG/1302/2014), co-financed by COMPETE (POCI-01-0145-FEDER-016773).

TU142

Multigenerational exposure of Folsomia candida to copper agrochemicals: conventional and nano-pesticides

C. Malheiro, Department of Biology, University of Aveiro / Biology; A.R. Silva, University of Aveiro / Dept. of Biology & CESAM; D. Nunes Cardoso, CESAM, University of Aveiro / Dept. of Biology & CESAM; T. Neves, University of Aveiro / Department of Biology & CESAM; P. Silva, Universidade de Aveiro; J. Ulcăr, University of Ljubljana / Department of Biology; F.J. Wróna, University of Calgary / Department of Biological Sciences; A.M. Soares, University of Aveiro / department of Biology & CESAM; S. Loureiro, Universidade de Aveiro / Biology Agricultural practices include the use of agrochemicals for crop maintenance and enhanced productivity. Although soil contamination may result in long-term effects on soil organisms, 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 nanoparticle 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 collombola 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, whereas generations 2 and 3 received unspiked 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 effects on the same soil biota. The progressive increase in the use of pesticides has been accompanied by effects at different levels of biological organization, implying losses of species and consequently of ecosystem services. Among the species utilized in terrestrial ecotoxicological tests, the springtail Folsomia candida (Collembola, Isotomidae) is one of the species suitable for assessing side-effects on detritivorous soil arthropods. In Brazil, the acaricide abamectine and the insecticide difenacozonole are widely used in agriculture, but little data is available about their possible side effects on the soil community. The objective of this work therefore was to evaluate the effect of abamectine, pure and in the formulation Kraft®, and of difenacozonole, pure and in the formulation Score®, on the reproduction of F. candida using a standard OECD (L.22.2 soil) test and of difenoconazole, with age 10-12 d, were exposed according to the standardized ISO and OECD test guidelines. The results were analyzed by analysis of variance (ANOVA) followed by Dunnet’s test at 5% significance level. Median lethal concentration (LC₅₀) was calculated using Trimmed Spearman Karber (TSK) and EC₅₀ and EC₉₀ values for effects on reproduction were estimated using a logistic model. The results showed that both pesticides were more toxic in the formulation than when applied as pure active ingredient. For abamectine dosed as the formulation Kraft³50 EC₅₀ was 1.0 (0.17-1.8) mg/kg dry soil, while it was 6.3 (1.8-11) mg/kg dry soil for the pure active ingredient. For difenacozonole applied as the formulation Score³5, EC₅₀ was 53.5 (40.0-67.0) mg/kg dry soil while no effects on springtail reproduction were seen at concentrations of the pure active ingredient up to 333 mg/kg dry soil. The data indicate that it is essential to perform official formulation screening tests with pure active ingredients. It is currently unknown which component of the formulation causes the increased toxicity. Therefore, we are applying gene expression analyses to mechanistically underpin increased toxicity levels caused by the tested formulations.

TU145

Terrestrial arthropods as indicators of environmental pollution

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In recent years, the use of and interest in terrestrial arthropods as indicators of environmental pollution has increased. Arthropods are terrestrial, terrestrial arthropods are relatively easy to sample, and collection normally has less ethical restrictions than 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 these gave more quantitative insight into the condition of the environment. In the area 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 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
lack of sufficient data on most terrestrial arthropods as indicators. We discuss a
number of possible predatory taxa, such as dragonflies, spiders, wasps, and beetles.

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 / DAFNEA; E. Tescari, Dow AgroSciences Italia srl; R. Bradascio, Dow AgroSciences Italia srl / RD; S. Otto, Italian National Research Council Spray diet 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 a 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 and grape (Cydia pomonella and Lobesia botrana 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 (Albus, ATR 80 yellow), low-drift nozzles (Albus, TV1 80015 green), and high-drift nozzles with an anti-drift adjuvant (rapeseed oil). Results showed that the two spray drift reduction techniques effectively increased droplets amounts next to sprayer, 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.crophy.2017.04.010.

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

TU149
Freshwater organism can recognize microplastics as microalgae
S. Kim, Y. Chae, D. Kim, Konkuk University; Y. An, Konkuk University / Department of Environmental Health Science The plastics are slowly weathered into nano- (< 100 nm) and micro- (< 5 mm) sized particles owing to physical, chemical, and biological processes in the environment. In this study, we observed the behaviour of freshwater organisms and evaluated whether they recognize and response to microplastics. Adult zebrafish was exposed to the different concentration of microplastic (MP, 250-300 μm) and food materials (F) (20 mg MP, MP20; 20 mg F, F20; 10 mg MP + 10 mg F, MP10F10). The behaviour patterns were recorded and quantified. Diving beetle fed the adult zebrafish, which exposed under MP10F10 and F20 conditions, and the ingestion rate was quantified. The number of capturing patterns were counted as 21±4, 8±5, and 14±3 under F20, MP10, and MP10F10 conditions, meanwhile the spitting patterns were determined as 0.0±0.0, 2.8±1.3, and 0±0.4, respectively. Ingestion rate of diving beetle on control group was calculated as 0.63±0.10 zebrafish wet mg/sec. The exposure group, which fed the MP10F10 exposed zebrafish, showed the significant decreasing (> 0.05) of ingestion rate (0.55±0.08 zebrafish wet mg/sec) during 591±85 seconds. On diving beetle, the MP were only found at crop organ until 720 min after ingestion, and did not transfer to another organ. The digestive organs, especially crop, seemed to separate the microplastic as indigestible food. We concluded that the freshwater organisms recognize the microplastic, and exhibit the defence behaviour This research was supported by Basic Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT, and future planning (2016R1A2B3010445).

TU150
Microplastic shedding from functional textiles
C. Jacobsson, Swerea IVF AB / Energy and Environment; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); P. Mellin, Swerea KIMAB AB; O. Levenstam, University of Borås; A. Hanning, Swerea IVF AB; S. Roos, Swerea IVF AB / Energy and Environment Microplastic pollution of 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 investigates the fiber loss (PA, PB, 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 fiber fragments being generated during the simulation of industrial washing (Gyro wash). Fiber fragments were identified with REM and the EDX analysis of showed fluorine (F) as part of the fiber surface

C. Jacobsson, Swerea IVF AB / Energy and Environment; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); P. Mellin, Swerea KIMAB AB; O. Levenstam, University of Borås; A. Hanning, Swerea IVF AB; S. Roos, Swerea IVF AB / Energy and Environment Microplastic pollution of 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 investigates the fiber loss (PA, PB, 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 fiber fragments being generated during the simulation of industrial washing (Gyro wash). Fiber fragments were identified with REM and the EDX analysis of showed fluorine (F) as part of the fiber surface

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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 fluorinated 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 fragments during washing. In addition to the fiber size and chemical 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.

TU153
Cost-effective methodology for separation of microplastics from freshwater systems

M. 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/Biologica Department & CESAM, Aveiro University; N. Abramts, University of Aveiro / CESAM; M. Kreuse, Fraunhofer IME, Institute for Molecular Biology and Applied Ecology / Ecological Chemistry; M. Simon, Fraunhofer IME / Applied Ecology; M. Kruse, Fraunhofer IME, Institute for Molecular Biology and Applied Ecology; A. Kroll, GAB Consulting GmbH; A. Häusler, GAB Consulting GmbH / Environmental Fate and Modelling; E.E. Burns, Fraunhofer IAP Institute for Applied Polymer Research

Microplastics, one of the most demand material worldwide, are considered one of the most emerging aquatic pollutants due to their ubiquity, high persistence and insufficient management. Especially, microplastics (< 5 mm) are of scientific and social apprehension as they can reach high densities, derive from a variety of sources and can interact with biotic and abiotic environment. Currently, the concerning about the occurrence of microplastics (MPs) in freshwater systems has been increasing, notwithstanding there is no unified method for MPs separation in these systems. This result in inaccurate data that differs in quality and resolution, not allowing data comparison between different studies (large-scale spatial and temporal comparisons). Hence, this work aims to assess the effectiveness of distinct separation methods as an attempt to identify and establish the most cost-effective method. For that, artificial samples containing eleven plastics belonging to the most common types of polymers (e.g. low/high-density polyethylene, polypropylene, poly styrene, polyvinyl chloride, polyethylene terephthalate) were prepared (secondary MPs) and subjected to distinct methods. These methods included density separation methods using sugar, olive oil and zinc chloride, as well as organic matter degradation methods using hydrogen peroxide (peroxo oxidation) and multienzymatic detergent (enzymatic digestion). The samples were then undergoing the detection, quantification and identification of polymers using a dissection microscope and Fourier transform infrared spectroscopy (FTIR). Several criteria were considered in order to achieve the aims of this work: efficiency of separation and organic matter degradation, the total mass of recovered polymers, the quality of recovered polymers. Based on this multi-criteria approach, this study concludes that the wet peroxide oxidation with addition of zinc chloride was the most cost-effective method. This method should be used in future studies of monitoring of MPs in aquatic systems, notwithstanding the use of hydrogen peroxide must be cautious and only applied when necessary.

TU144
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 these aspects is still limited. Earth remote sensing is an important 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 impacted by the same transport mechanisms as non-motile plankton or detritus, we tested the hyporheic and hypolimnion of a forested river on microplastic and specific water parameters. In situ water parameters, microplastic (5mm – 250 μm), 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). Microplastics from surface waters were sampled with a manta trawl. Organic material was removed by enzymatic digestion and wet 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.
Coastal accumulation mapping of microplastic particles emitted from the Po River, Italy: Integrating remote sensing, in situ sample collections and ocean current modelling

E.C. Arwood, RSS Remote Sensing Solutions GmbH; F.M. Falcieri, CNR - ISSMAR; S. Piehl, University of Bayreuth; S. Baltz, Helmholz Centre for Environmental Research; I. M. Bochow, Helmholz Centre Potsdam - GFZ German Research Centre for Geosciences; M. Matthies, University of Osnabrueck / Institute of Environmental Research; J. Franke, RSS Remote Sensing Solutions GmbH; S. Carmiel, M. Sciano, CNR - ISMAR; C. Lafrorsch, University of Bayreuth; F. Siegert, RSS Remote Sensing Solutions GmbH

Phytoplankton in inland waters and the open ocean is a long recognized problem for marine wildlife, 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 (9 beaches) and 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 microplastic pollution being emitted by a major river and its distribution along adjacent coastlines as well as into the open ocean.

Cause and effect of the plastic industry in South Africa as a developing country

C. Verster, North-West University - School of Biological Sciences / Environmental Science and Development

In 2017, the South African plastic industry has grown with 1.9%, compared with 2016 (Plastics SA). Although legislation is in place to promote recycling and sustainable use of natural resources, the recycling of plastic based materials is done predominantly by corporate initiative. Many South Africans believe that the country is lagging in terms of recycling. South Africa has however achieved a recycling rate of 41.8% in 2016, of which most is done post-consumer. Plastics SA has set an ideal of no plastics to landfills by 2030, and plans are being set in place to achieve this goal. Inadequate waste disposal infrastructure and protocols, especially in informal settlements causes large quantities of unrecycled plastic to end up in aquatic systems and subsequently in the marine environment. Beaches surrounding estuaries are heavily polluted with macroplastics which calls for greater prevention and clean-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 filtered 46 fresh water samples from various localities in and around Gauteng, the most densely populated province in South Africa. High levels of plastic pollution were found in almost all samples. Up to 40 plastic particles (> 20 µm) per litre were found in surface water of the Vaal River, a major river in the country’s largest drainage basin flowing through industrialised areas. These levels are comparable to high levels of microplastic pollution found in European rivers. The growing plastics industry in South Africa requires excellence in clean-up and recycling to reduce the negative impacts on the environment and create a viable plastic sector.

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 fresh- 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 samples during treatment stage, 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 abundance 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 misidentification of cellulose and other fibre microdebris 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.

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. This study aimed to identify the extent of weathering in plastic particles from the Oceans-funded project WEATHER-MIC 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 apoptotic 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 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 liberated 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) activation of metabolic biomarkers, e.g., via binding to the arylhydrocarbon receptor; iii) specific, receptor-mediated effects such as estrogenicity; 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.
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 the wastewater system, sewage treatment processes and removal of MP 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 statistical analysis. More than 90% of microplastics (MPs) present in raw wastewater are captured by water treatment plants (WWTPs) for MPs by FT-IR microscopy method. 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 source of MPs in various water samples.

**TU160** Detection of micro-particle 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; J. Vollertsen, 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-particle pollutants (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 used 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-particle 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 500-10 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 measured in the samples were 222,500 particles Kg⁻¹, while the estimated mass was 17.1 mg Kg⁻¹. The most abundant polymers/paints detected were polyester (30%), acrylic coatings (20%), and polyethylene (14%). 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 samples 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-particle-particles were successfully extracted and detected in a recreational harbor area using the art analytical methods, 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. Horling, NIVA - Norwegian Institute for Water Research; T.C. Schell, IMDEA Water Institute / Ecotoxicology; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences; L. Nizzetto, NIVA

More than 90% of microplastics (MPs) present in raw wastewater are captured by wastewater treatment plants and removed in the sludge phase. Therefore, the use of sludge as a fertilizer for agricultural soils may be a relevant source of MPs in the terrestrial ecosystem and, therefore, runoff, in surface waters. The fate of MP applied with sludge is strongly dependent on weather conditions (rain, temperature, etc.). The objective of this study was to determine the fate and distribution of MPs in the agricultural soil-water system. The study was performed in the experimental site in the Alcoa Institute (INIA - Instituto Nacional de Desarrollo Rural, Agrario y Alimentario) located in central Spain, in an area characterised by semi-arid climate: hot and dry summer, low rainfall (about 450 mm per year) which is concentrated in spring and autumn. Suitable devices for runoff collection (modified Pinson collectors) were placed on three different plots with different MPs treatments: (i) soil never treated with sludge (control), (ii) soil treated with sludge in the past (in 2013), and (iii) soil treated with sludge at the start of the experiment (November 2017), according to usual agricultural practices. Besides sludge application, soil characteristics (composition, texture, etc.) were comparable in the three plots. Sludge was applied early November and the plots were sown with barley. After each relevant rainfall event, runoff water was collected and filtered in-situ and to isolate the MP fraction. Soil samples were taken in all plots at the start of the experiment, as well as 3, 6 and 12 months after the start of the experiment. To determine the vertical MP transfer within the soil, soil cores were divided into three fractions (0-5, 5-10, 10-15 cm). Separated plots receiving the same sludge treatments and soil conditions were used to sample earthworms and to check potential accumulation and MP impacts for the soil fauna. The content of MPs in runoff water, soil and biological samples were extracted using organic matter digestion (soil and organism samples), density separation (soil samples), and filtering (all samples). MPs were identified visually and characterised chemically using FTIR. Preliminary results of this experiment, which can be used to quantify fluxes and emissions of MPs in agro-ecosystems under semi-arid conditions, 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. Hou, 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, size and chemistry of MPs in the treated water are not well characterized. The aim of this study was to determine the occurrence and characteristics of microplastics in influent and effluent of five WWTPs with different dimensions and treatment processes. MPs not retained by WWTPs are directly discharged into the aquatic environment through WWTP effluents, whereas the majority of MPs are assumed to be retained and accumulated in the sewage sludge. Runoff, after application of sewage sludge to agricultural fields, may consequently serve as an additional source of surface water contamination by MPs. Therefore, the aims of this study were: (i) to evaluate the occurrence of MPs in the treated wastewater, and (ii) to ascertain the MP contribution of WWTP effluents. The study was carried out in the Henares River watershed (Central Spain). Five WWTPs with differing dimensions (population equivalents between 10,000 and approx. 400,000), differing influent types (domestic, industrial, or both mixed), and differing treatment processes were selected. Wastewater inflow, outflow, and sludge (humid and dried) were sampled during two different seasons (spring and summer). In addition, river water and sediment samples were taken in three different seasons (spring, summer, and autumn) at three differently impacted sites: i) low human impact; ii) high agricultural impact; and iii) high mixed impact (urban, agricultural, industrial). MPs in river water and wastewater were divided into four fractions by filtering a suitable amount of water through plankton nets of different mesh sizes (from 300 to 20 mm). In order to assess the MP concentration and composition in the samples, solid substrates (sludge and river sediment) were subjected to an organic matter removal treatment, followed by density extraction. Subsequently, those samples, as well as the liquid samples (river and wastewater), were filtered onto filter papers to visually identify the MP content and then chemically characterize their polymer content. FT-IR analysis of the samples collected in the river showed the presence of different polymers, including PET, PP, PE, PS, and PAO, which are materials commonly associated with personal care products 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 the wastewater system, sewage treatment processes and removal of MP 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 statistical analysis. More than 90% of microplastics (MPs) present in raw wastewater are captured by water treatment plants (WWTPs) for MPs by FT-IR microscopy method. 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 source of MPs in various water samples.

**TU163** 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 Krøvel, 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 findings of different types of plastic in different compartments of marine, freshwater and terrestrial ecosystems; others point out the accumulation of micro- and nanometric sized plastic particles through 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 plastic particles 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 occurrence of speckled water in the drinking water and their implications on human health. 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/microplastics particles 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, UCAN-ICCF; F. Delor Jestin, Sigma-ICCF; Z. Donimack, H. Askanian; 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 very a 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 of microplastics in the environment and 2) the spatial and temporal distribution of the various plastic species through 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 macroplastics 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 Plastigages project supported by the CNRS [1, 2]. 4. Collect and analyze the composition of microplastics in the surface waters of different rivers (Allier, Charente, Loire, Touvre, etc.). To do this, we rely on citizen science operations, in particular thanks to the contribution of the babylab sampling net [3,4], which makes it possible to multiply samples and analyzes. 1 Occurrence of plastic litter in the Allier river in France. Vincent Verney, Gaëlle Bissagou Koomba, Alexandre Garreau, Florence Delor Jestin, Erwan Roussaud, Olivier Voldoire, Jean-Luc Peiry: To be published 2- https://www.researchgate.net/project/PLASTIGAGES 3- Commitment agency, the case of babylab, Max Liboiron, Engaging Science, Technology and Society 3(2017), 499-527 4- http://laplagiesauvergac.org/laboratoire/recherches/
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 microplastic mobilisation to surface waters and highlight the potential knowledge gains 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 particles (below 500 µm) in the water column and in sediments. In 2009, Spildevand A/S has been characterized: PE, PP, PS, PVC and TDI-monoesters, 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, PS, PP, PVC and TDI-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 (9%) and PU (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. Voltersen, Aalborg University / Civil Engineering Department

In this study the sorption efficiency of a disc filter of 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 using a suction sampler and a 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 residual 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 flotation in order to eliminate the sample matrix and extract the suspended particles. The size distribution of the sorbed fraction was determined using a combination of optical microscopy and SEM.

TU171 PlasticBudget - Project on the environmental assessment of microplastic emissions N. Thomemann, 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 (macroplastic) 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 plants, and the food chain. The long distance transport modes suggest that 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 endpoint indicators and associated characterization methods, as well as standardization to a reference value (e.g. the production volume of the specific plastic), are therefore developed in the PlastickBudget-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 polymers, sizes 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 particles to be separated from those caused by other factors. This separation is crucial for testing MP-specific effects, as many test organisms are suspension-feeders that have evolved in turbid aquatic environments. To highlight and provide examples of the importance of particle controls in MP-effect studies we will present: I) results of a review on effects of particle suspensions (MP, or natural nutritionally inert particles), and II) case studies employing control particles. The data analysis strongly suggest that particle controls are essential for identification of MP-specific effects, so that MP impacts can be assessed based on ecological soundness.

TU173 Influence of environmental conditions on the sorption of organic pollutants to microplastics S. Seidensticker, J. Lamprecht, P. Grathwohl, Eberhard Karls Universität Tübingen / Center for Applied Geoscience; C. Zarl, University of Tuebingen / Center for Applied Geoscience

The ubiquitous contamination of all environmental compartments with microplastic particles is extensively discussed in both science and public. Large numbers of microplastics have been measured in effluents of wastewater treatment plants. These particles might act as sorbent and transporter for frequently occurring wastewater contaminants and are hence a factor that needs to be considered if the environmental fate of pollutants is examined. Some contaminants, among those also microplutolitigants 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 values (4, 7, 10). 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 pKa-values from 1.6 to 13.9. We performed batch experiments with fourteen ionic and five non-ionic 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 hydropathicity. Sorption of ionic substances is strongly influenced by the pH while non-ionic substances showed a partitioning independent of pH. For sorption into polystyrene, 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 polystyrene 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 low 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 package 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].


TU175 Influence of polystyrene microplastics in combination with organic pesticides on the giant rams-horn snail Marisa cornuarietis: behavioral and biochemical responses S. Kraus, University of Tubingen / Animal Physiological Ecology; H. Schmieg, Tuebingen University / Animal Physiological Ecology; E.E. May, University of Tuebingen / Animal Physiological Ecology; A.S. Ruhl, TU Berlin / Department of Water Quality Control; H. Kohler, University of Tubingen / Animal Physiological Ecology; R. Triebkorn, University of Tuebingen / Animal Physiological Ecology.

Due to the increasing demand for and usage of plastic products during the last decades, the quantity of globally produced synthetic polymers rises continuously and in high amounts of plastic debris of all sizes in the environment. Very small-sized particles and fibers (< 5 mm) which are defined as microplastics result either from degradation of macroplastics or are produced as primary microplastics which are contained e.g. in cosmetics. Microplastics are of particular interest in ecotoxicology, because they can interfere with biochemical and behavioral processes of individual organisms, and (3) observing changes in behaviour (reproduction and survival). In the study, we investigated the influence of pristine and pristine and weathered polystyrene microplastics on the giant rams-horn snail (Marisa cornuarietis). Snails were exposed to 10.000 polystyrene particles per liter (cryogenically milled, < 100 µm) in combination with different concentrations of 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 aim of this study was to reveal that an adequate pretreatment of the sample along with an adequate customized polymer library that contains real/weathered pesticides spectra are highly recommended for the adequate monitoring of microplastics in the environment. 


TU176 Effects of artificial weathering on polypropylene microplastics V. Fernández-González, Universidad de Coruña / Grupo Química Analítica Aplicada (QANAP); G. Gruiero-Noche, Universidad de Coruña / Grupo Química Analítica Aplicada (QANAP); J.M. Andrade-Guarda, Universidad de Coruña / Grupo Química Analítica Aplicada (QANAP); P. López-Mahía, Universidad de Coruña / Analytical Chemistry Department; D. Prada-Rodríguez, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); S. Muniategui, Universidade da Coruña / Analytical Chemistry

Microplastics are defined as plastic materials or fragments with diameter below 5 mm. These microplastics occur due to the release of manufactured (primary) microplastics in various products and the breakdown of larger plastic litter (secondary microplastics). The time required for plastic to degrade under natural conditions is estimated to be in the thousands of years, while the photo-oxidation by UV light its primary degradation pathway. The small fragments of microplastic created by weathering are detrimental to ocean ecosystems for multiple reasons. In the frame of the BASEMAN project (JPI Oceans) 9 natural microplastics of different polymers materials, in two size presentations (100-500 µm and pellets ≤1 mm) were artificially weathered. A pilot-scale simulated weathering system (dry conditions and simulated marine conditions), using UV/VIs metal halide lamps, was deployed. This study focused on the characterization of the changes that an accelerated artificial weathering process produces in polypropylene microplastics using ATR-FTIR. This could be useful to identify real plastics and microplastics found in marine environment, and understand how aging affects the surface and chemical structure of this material. New absorption peaks can be seen, that reveals changes in the main structure of the microplastics. Some indexes were calculated as the ratio of these IR bands to a reference peak that indicate oxidized carbon in the plastic hydrocarbon chain. It is worth to note that the direct identification of the type of polymer is usually not possible. Weathering-related changes in the IR spectra differ the correct identification of the polymer when are compared with the IR polymer library. Moreover, methane microscopy was also done laboratory studies. Effects of artificial weathering are typically dose-dependent and include reduced feeding and successful reproduction, change in organism’s behaviour and decreased survival. This study aims to build on current research on the effects of freshwater microplastics, focussing on the micro and nano plastics potential to transfer chemicals in the environment. The formation of a corona on plastic particles changes their surface characteristics which could lead to a change in how bioactive chemicals interact with the environment. In this study, we conducted the effort that plastic conditioned under different scenarios can have on the interaction with Daphnia magna (a keystone species). We assessed this using several parameters including: (1) quantifying the proteins and polysaccharides secreted by the organism’s before and after exposure, (2) quantifying uptake of plastic in the individual organisms, and (3) observing changes in behaviour (reproduction and movement). We explored these effects for pristine plastics and plastics combined with chemicals commonly found in the environment, including 17α ethynyestradiol and detersents. The effect that competitive binding of biomolecules naturally secreted by Daphnia have on both adsorption and desorption of the target chemicals on the plastic’s surface was a key element of this study, to assess the impact of how microplastics interact with more complex pollution issue in the environment. This study could help to explore the interaction of combined stressors using parameters that can be controlled in the laboratory. Through this, we can assess the impact of assumptions about test conditions, and the impact of over-simplification of standardised test media, on the resulting data regarding the Trojan-horse potential of micro and nano plastics. By building such data will include recommendations to improve the environmental realism of the laboratory conditions to make more accurate exposure assessments for environmental modelling in the future.

TU178 Exposure to conventional but not biodegradable microplastics impacts fitness in Daphnia magna Z. Grodes, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); M. Puranen, Stockholm University; M. Ogonowski, E. Gorokhova, Stockholm University / Department of Environmental Science and
Regarding effects of microplastics on freshwater ecosystems is limited. In the primary microplastics (PMP) had a similar influence to understand potential environmental impacts of both polymer types. We compared effects of exposure to 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 fractionation to < 50 µm, up to 100,000 particles/L, and to estimate the remaining adverse effects. 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: 02WRS1378).

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 community because their ability to be accumulating in the tissues of biota. MPs can reach the aquatic environment through the Wastewater Treatment Plants (WTPPs) or affect aquatic ecosystems, although at relatively high levels of exposure, and that temperature as an environmentally relevant additional stressor has a major influence on species sensitivity to microplastics.

**TU181**

**Evaluation of chronic toxicity of polystyrene microplastics on freshwater mussels**

S. Magni, University of Milano / Department of Biosciences; F. Gagne, Environment and Climate Change Canada; C. Dela Torre, State University of Milano / Biosciences; C. André, J. Auclair, H. Hanana, Environment and Climate Change Canada / Aquatic Contaminants Research Division; F. Bonaso, University of Milano / Department of Environmental Science and Policy; A. Binelli, University of Milano / Waater Treatment Plants (WWTPs) or affect aquatic ecosystems, although at relatively high levels of exposure, and that temperature as an environmentally relevant additional stressor has a major influence on species sensitivity to microplastics.
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 potential 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 (National University of Denmark) / Department of Environmental Engineering; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; M. Wagner, Norwegian University of Science and Technology / Department of Biology

The last decade has seen a surge in research investigating various aspects of micro- and nanoparticles 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 had already been faced by researchers in the realm of nanotoxicology. Our question about 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 – poses major concerns for the risk assessment of MP. 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 fructose-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 microparticles 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 and face new researchers in the realm of nanotoxicology. Our question about 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.

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. Zidar, University of Ljubljana / Department of Biology Biotechnical Faculty; G. Kalckova, 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 effect 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 fragmented from plastic bag. The mean size of MP from facial cleanser was 0.137 ± 0.051 mm, while the plastic bag MP was larger 8.80 ± 5.05 mm (with 62% of particles smaller than 5 mm). Microplastic was mixed into the soil at environmentally relevant concentration 4 mg/g dry weight (0.4 % w/w). The isopods were exposed individually and in groups of 10 animals. The test container comprised two identical polypropylene vessels connected with a fixed polypropylene tunnel to enable animal migration between the two vessels and covered with a lid. In individual exposure the position of each animal was recorded 10 times within the 48 h exposure period and the number of positions on each side was calculated. In group exposure, the isopods were inspected only after 48 h and the number of animals at each side was recorded. Earthworms were exposed in one test container that was in use when applying the control soil and MP contaminated soil. Before the animals (10) were placed into the test container the divider was removed. The number of animals on each side of the soil was counted after 48 h of exposure. Our results indicate that isopods show no preference or avoidance behavior towards facial scrub microbeads or plastic bag microplastic contaminated soil. This was shown for both types of exposure, individual and group. On the other hand, earthworms clearly avoided the facial scrub contaminated soil but were not affected by plastic bag MP. It remains to be investigated how longer exposure to MP would affect the behavior of terrestrial organisms. Also it is of interest how environmentally aged MP (e.g. coated with biofilm) would affect the organisms. Knowledge in this field is important to assess the potential hazard of microplastic deposited on soil.

TU185 Analysis of the Trojan horse effect of a mixture of microplastics and chlorpyrifos in an aquatic microcosm study J. Deerman, Research Institute gaia / gaia - Research Institute for Ecosystem Analysis and Assessment; X. Chen, University College London; T. Strauss, Research Institute gaia / gaia - Research Institute for Ecosystem Analysis and Assessment; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Microplastic particles (MP) are of concern in the aquatic environment because of their increasing amounts in production and release into the environment. Despite their physical adverse effects, MP can sorb hydrophobic chemicals, which can then be transported together into biota by the so-called ‘Trojan horse effect’. In this study, a higher CPF could in a complex ecosystem be possible due to the plastic bag release with the aim to discover the Trojan horse effect by means of a laboratory aquatic microcosm study. The insecticide chlorpyrifos (CPF) was used sorbed to 5 µm polystyrene microbeads. Beside the control microcosms (C), an MP-control (MPC) group was treated with 4 mg MPI. For two other treatment groups, the same concentration of MP was coated with nominal CPF concentrations of 0.5 µg/L (L1) and 2.5 µg/L (L2). 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 fructose-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 microparticles 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 and face new researchers in the realm of nanotoxicology. Our question about 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.

TU186 Microplastics exposures of fish: internalization and effects on behavior and growth C. vignet, Eawag / UTOX; X. Cousin, IFREMER / Laboratoire de Ressources Halieutiques de La Rochelle; R. Behra, Eawag / Department of Environmental Toxicology; L. Jouassard, IFREMER; L. Sigier, Eawag; M. Bégout, IFREMER / Laboratoire de Ressources Halieutiques de La Rochelle; J. Cachot, University of Bordeaux / EPOC; K. Schirmer, Eawag / Environmental Toxicology

Awareness of the presence of microplastics, i.e. plastic particles ranging in size from 1 µm to 5 mm, in marine and freshwaters has recently risen but detection and quantification is challenging. Furthermore, whether they pose a risk to aquatic organisms is not yet clear. Interpreting these results, MP might have led to higher reproduction rates as a stress response which were lowered when CPF was present. In this case, MP must have become bioavailable to D. pulex after ingestion of MP. For C. pseudogracilis, total abundances increased the most in MPC, whereas L and H developed similar as C. As for the cladoceran, MP might have led to higher reproduction rates that were lowered by CPF. The Trojan horse effect has probably been underestimated in the controls, whereas in the treatments the observed doubling of the isopods might have led to enhanced abundance levels still needs to be clarified.

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feeding with microparticles contaminated food has consequences on juvenile fish growth. Taken together, our study demonstrates the power and limits of flow cytometry/visNE 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. McGoran, Royal Holloway; P.R. Cowie, Field Studies Council Scotland; P.F. Clark, The Natural History Museum; J.P. McEvoy, D. Morriss, Royal Holloway
This study compares 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 dissection 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, benthi, 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; F. Tremolada, University of Milano / Department of Biomolecular Sciences and Bionecology; 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 towards microplastics (MPs), small plastic particles (dimensional range of 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, regarding release test, after 24 hours of exposure, the plastic particles 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. SULPAN, University of Birmingham; I. Lynch, University of Birmingham / Geography Earth Environment Science; J. Sadler, The University of Birmingham / Geography 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 (Jenner, 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. pulex (2.3–4.7 mm) to D. magna (10–20 mm) which span a similar range of sizes as macro 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 polyethylene terephthalate microspheres (0.1, 1, 10 and 100 mm) 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 compared on the basis of the number of potential plastics; before FTIR analysis). The hypothesis tested was that the size of microplastics 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 polyethylene 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
G. Grueiro-Noche, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); V. Fernández-González, J.M. Andrade-Gardu, Universidad da Coruña / Grupo Química Analítica Aplicada (QANAP); P. Llorente-Majía, Universidad da Coruña / Analytical Chemistry Department; D. Prada-Rodríguez, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); S. Muniteguei, Universidade da Coruña / Analytical Chemistry
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 the marine litter. A particular fraction of plastic debris are the microplastics (particles ≤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 used to determine the plastic debris present in the mackerel stomach. Identification and characterization of microplastics was done by µFTIR.


TU191 Microplastic contamination of the model system Weser-National-Park Wadden Sea: an across-ecosystem approach
S. Moses, University of Bayreuth / Animal Ecology I; M. Loeder, J. Schrank, C. Lacours, 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 give clear and transparent 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 Photocatalytic fragmentation of freshwater (micro)plastics using UV irradiations
V. Verney, CNRS - ICCF / Photochimic- CVP; G. BISSAGOU KOUMBA, UCA-IJCF; F. Delor Jestin, Sigma-ICCF
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 small, increasingly smaller sizes, and by chemical functionalization due to the photo-oxydation 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 plastics 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 (Tg = 104 °C) at the temperatures of use, which is not the case of the PP (Tg = 0 °C). Finally, PLA can start hydrolysis reactions. During the exposure time, solids and liquids (a small volume of water) are taken for analysis. The physicochemical characterization of the liquid medium is that in all cases there is a formation and leaching of short chain (1C, 2C, 3C) carbonylic 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 polluting level. For liquid ingredients, it is the 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 USEPA) 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 Korea 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 human health. Microplastics formed by the breakdown of larger plastics and thus are 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 polluting level. For liquid ingredients, it is the 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 USEPA) or use of novel methods. The work presented provides a scientifically robust approach for accurately assessing the impact of polymers in the environment.

TU195
Assessment of the microplastic contamination in sediments from the French Atlantic coast
N. Phuong, Université de Nantes; L. Poirier, Université de Nantes / MMS. f. lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMM UMR CNRS; M. Déniel, Institut des molécules et matériaux du Mans; A. Kamari, A. Zalouk-Vergnoux, Université de Nantes / MMS

The ubiquitous presence of MPs has been demonstrated by scientists for recent years. They are detected in all environmental compartments: air, freshwater, aquatic 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 (Pays 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 25 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 satisfactory representativeness 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 µFTIR 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.

TU196
Derivation, Validation and Implementation of Environmental Quality Benchmarks (P)

N. Chériaux, M. Milanov, E. Benard, University of Lausanne / Faculty of Geosciences and Environment

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 usable 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 highlighted that no section of the river currently 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 CPF 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 CPF based on 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 0.0004 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 HC5, from a species sensitivity distribution (SSD) for crustaceans and insects using
where bioavailability based thresholds a whole of Europe (i.e. continental) would leave some countries with costly and level which will ensure protection of sensitive environments. The value of the concentration is calculated using the physico either regional or continental water chemistry data. At Tier 2, the bioavailable me tier compares the dissolved metal concentration to a threshold, estimated using the levels of compliance of European freshwaters with a copper EQS, and Standard (EQS) for a substance must be a fixed value: the same EQS is a been limited. For regulatory purposes in Europe an agreed Environmental Quality attempts to deliver practical, routine methods to do so have been made. For regulatory purposes in Europe an agreed Environmental Quality attempts to deliver practical, routine methods to do so have been made.

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**TU198**

**Lead exposures 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 countries in Europe through the EQSs (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 to regulate regulatory freshwater monitoring data from six European member states and FOREGS database. A tiered approach was used to compare measured concentrations to the EQS, allowing for correction of 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 Mod PCI riparius, 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 EQSbioavailable. In Tier 1, Pb concentrations were directly 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 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.

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**TU199**

**Assessing compliance of European freshwaters for copper: accounting for bioavailability**

A. Peters, I. Wilson, G. Merrington, wca; D. Heijerick, 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 sites and exposure pathways. The levels of compliance for metals, the EQS must be set for the bioavailable fraction, and it is termed EQSbioavailable. This study determines the levels of compliance of European freshwaters 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 content 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 unnecessarily stringent monitoring requirements. Deriving the threshold on a region-by-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 remotely 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.

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**TU200**

**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 2000/60/EC 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 Model (BLMs) was used to account for bioavailability, along with the current European Water Framework Directive lead dissolved organic carbon concentration (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 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.

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**Integrated approaches in ecotoxicology: bridging the gap between experimental toxicology and mechanistic modelling (P)**

J. Chowdhury

The Biotic Ligand Model (BLM) was used to account for bioavailability, along with the current European Water Framework Directive lead dissolved organic carbon concentration (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 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.

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**TU201**

**Modelling survival under chemical stress. A comprehensive guide to the GUTS framework**

T. Jager, DEBBox Research / Dept of Theoretical Biology; R. Ashauer, University of York / Environment

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 more general modelling framework: GUTS, the General Unified Threshold model for Survival. GUTS was conceived in 2010, and has subsequently gained a large user community. Furthermore, the model is receiving increasing interest from the regulatory field as it is expected to be used 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.
which effects start to appear. Once this threshold is surpassed the amount of effect increases significantly above this threshold, and further increases are again limited by thermodynamics and/or unknown biological mechanisms. Therefore, it is critical to define a threshold below which effects start to appear and above which effects are no longer increased. This threshold is often referred to as the benchmark dose (BMD).


toxicokinetics calibration is a challenge as their properties are not fully characterized yet and experiments are scarce. We here present the results of a research project that aimed at investigating toxicokinetics (TK) of PFASs in juvenile common sole from a contaminated nursery ground, the Gironde Estuary (France).

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. Mechanistic models rooted in Dynamic Energy Budget (DEB) theory are relevant to predict 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 propose to use sole diet reconstruction for a range of environmental diet, 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 the 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 individual variability of organisms and between these different conditions and food dependency. Moreover, this mechanistic approach prioritizing sources of variability provided new insights on the differential bioaccumulation between toxicant families in a key flatfish resource.

Investigating metabolic acceleration in dynamic energy budget models of copepods using the ecotoxicological model organism Notothea spinipes

J. Koch, GhEnToxLab (Ghent University) / Applied Ecology and Environmental Biology; K. De Schamphelaere, 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 ecotoxicology 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 so far, it is not clear if acute toxicokinetic studies using field extraploted models rooted in Dynamic Energy Budget (DEB) theory can help to evaluate sublethal toxicity data in terms of effects on the energy household of an animal. Although DEB models for new species are usually easily parameterized, the copedep 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 puberty point. In this study we parameterized the two typical DEB models ‘abp’ (metabolic acceleration from birth to puberty) and ‘abp’ (standard von Bertalanffy growth from birth to puberty) for the harpacticoid copepod Notothea 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 abp 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.
significant higher than that of MAP. 0.269±0.018 ml μg⁻¹ (p < 0.001), and the uptake rate constant of MDP 0.128±0.001 ml g⁻¹ h⁻¹ was also significantly greater than MAP 0.086±0.001 ml g⁻¹ h⁻¹ (p < 0.001), it revealed that Cu was likely to bind on MDP in the low exposure concentration than that of MAP. This study concluded that Cu tends 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
N. Urie, INRS-ETE / Centre Eau Terre Environnement; A. Urie, Université du Québec / Centre Eau Terre Environnement; L. Ramilo, H. Sønnenberg, Ecological and Regulatory Solutions Inc.; P.G. Campbell, P. Couture, Université du Québec, INRS / Centre Eau Terre Environnement

Discharges from 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 molecules 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 if 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 found to vary widely in exposed fish than in reference fish, with Cu 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 metallothioneins. In contrast, the largest contributors to the total Se liver burden were the potentially metal-sensitive heat-denatured 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 total Se burden. At the end of the study, a decrease in condition was observed for both sexes. Finally, this work will contribute to advancing our understanding of the toxic modes of action of metals in aquatic organisms and our capacity to monitor the risk for fish inhabiting metal-contaminated environments.

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 the 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 upon 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 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. T.U210
Development of a Novel Quantitative Adverse Outcome Pathway Predictive Model for Lung Cancer
T. Hill, US EPA NHEERL/JST/DCB / 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, high-throughput-intensity prediction as well as the development of mechanistic models that inform 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/nucleic acid adducts, diminished CC10 capacity and hyperplasia of CC10 deficient Clb cells, and culminating in the adverse outcome of mixed-cell tumor formation. The AOP 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 characterisation 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 embryotoxicity on recruitment failure in European eels
M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; M. Freese, J. Pohlmann, Thünen-Institute for Fisheries Ecology; J. Dörr, National Research Council at U.S. Environmental Protection Agency, Duluth; M. Damerau, L. Marohn, R. Hanel, Thünen-Institute for Fisheries Ecology; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

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 molecules 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-milling effluents, and (ii) to investigate if 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 found to vary widely in exposed fish than in reference fish, with Cu 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 metallothioneins. In contrast, the largest contributors to the total Se liver burden were the potentially metal-sensitive heat-denatured 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 total Se burden. At the end of the study, a decrease in condition was observed for both sexes. Finally, this work will contribute to advancing our understanding of the toxic modes of action of metals in aquatic organisms and our capacity to monitor the risk for fish inhabiting metal-contaminated environments.

TU212
Salmonid pituitary cells as a test system for identifying endocrine disrupting compounds
L. Harding, University of Washington / Aquatic and Fishery Sciences; I.R. Schultz, NOAA NWFS / Marine Science Laboratory; G. Young, Advaisin WorleyParsons Group / Aquatic Sciences; P. Swanson, NOAA-NWFS

The pituitary gland is a small posterior organ, 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 17-ethinylestradiol (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 (fshb) mRNA levels. These results motivated us to expand our studies by developing an in vitro test...
The relevance of the end-of-life stage for the environmental impact of batteries. J.F. Peters, Karlsruhe Institute of Technology KIT / Helmholtz Institute Ulm HIU, M. Weil, Karlsruhe Institute of Technology KIT / Institute for 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 (rechargeable, mounted system), a hybrid aqueous ion battery (AHIB) and a vanadium redox flow battery (VRFB), all with their specific recycling potential. 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 recovering 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 a macro-scale (AHIB and VRFB) in comparison with highly integrated cells (LIB). Thus, design for recyclability is highly important in terms of future circular economy and might easily outweigh the possibly reduced energy density or lower performance.

Batteries recycling efficiencies and their influence on the life cycle impacts of circular economy (P). 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.

New and Reconditioned Electrical and Electronic Equipment. How does change the environmental performance? M. Pini, University of Modena and Reggio Emilia / Department of Sciences 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 Sciences and Methods for Engineering The scope of this study, carried out within the LIFE12 ENV/IT001058 WEEEEnmodels, 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 were applied for the assessment of the complete life cycle of each WEEE category that has been considered for each WEEE 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 frequently. 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 A produces a damage decrease for all WEEE 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 environmental credits. This is due to the avoided burdens associated to the manufacturing of the new EEE, since the system boundaries have been enlarged under the consideration of the avoided production of the products. Attributional and consequential LCI modelling performed different LCA results. Following the methodology guidance 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
F. Reale, EC JRC; V. Castellani, European. Commission - Joint Research Centre / Sustainable Resources, Bio-Economy; B. Hirsch, EMPA / Technology and Society Lab; S. Sala, European Commission - Joint Research Centre / Bioeconomy unit

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. The aim of this study is to facilitate the understanding of the utility function 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 compared with the alternative that integrates the reduction of the overall impact compared to the baseline scenario. The reduction is more important for categories like e.g. GWP (due to the increased energy efficiency of products). The results show the potential of the policy of “phasing out” 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
D. Nishiijima, National Institute for Environmental Studies; S. Kagawa, Kyushu University; M. Oguchi, K. Nansai, National Institute for Environmental Studies

In evaluation of environmental burdens from consumer durables, product lifetime is a key factor and most of the previous studies used average lifetime or lifetime distribution with a focus of engineering durability (Müller 2006; Kagawa et al., 2011; Nishiijima, 2016). These product lifetime modelling techniques did not explicitly treat a relationship between product replacement and consumer’s behavior. Whereas, the product replacement modelling techniques based on the economic maximum utility theory have been developed in economics and marketing fields (Schiraldi, 2011; Melnikov, 2013). It is beneficial to apply the economic product replacement models to environmental research. This study attempts to use the product replacement model for evaluating economic and environmental effect of polices for consumer durables. As a case study, we focused on air conditioners and analyzed the economic and environmental impacts of “home appliance 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 logistic coefficients by the maximum likelihood estimation. 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 discrete 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 decision. ‘To do so, we developed 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 new vehicle purchase behavior, becoming an effective tool for reducing CO2 emissions. 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.

TU222 Li-S batteries for electric vehicles, challenges for circular economy objectives
g. benveniste, C. Corchero, IREC; B. Amante, Universitat Politècnica de Catalunya UPc

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 required implementation. The evolution 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, reframe the massive depletion of this technology. With the aim of reaching a field of technologies and scenarios that is a short term, it is necessary to invest in 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 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, it 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 possible use in a second life, once they cannot be used in an EV (e.g. their use in stationary applications); 3) To evaluate the associated environmental impacts and 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 environmental and economic 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 ecositing of a "vapour and air barrier membrane - insulator" system, in a cradle to cradle approach
A. Léonard, Liège Université / Chemical Engineering - PEPs; M. Gellricher, Derbigum; B. Colson, Sieno Felt & Filtration; I. 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 of the European Union to improve the energy efficiency of buildings to reduce the 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
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, puncturing resistance), and finally a low disassembly and re-use level. The ATISOL C2C project aims to develop a complete solution (ecological insulation + renewable adhesives), with the lowest environmental impact on its whole life cycle. The solution can be used in both new construction and 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 the different walls covering and is adapted to every type of building. 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 Debitakine®. 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
A. Jimenez del Barco Carrion, The University of Nottingham; D. Lo Presti, The University of Nottingham / Nottingham Transportation Engineering Centre NTEC Transport; K. Van Acker, KU Leuven / Materials Engineering

Construction materials can accumulate in build-ings and infrastructures for several decades. In addi-tion, 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 Debitakine®. 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).

TU225 Dynamic vs static LCA to explore the sustainability of industrial waste recycling
A. Di Maria, KU Leuven / MTM; A. Levasseur, École de technologie supérieure / Construction engineering; K. Van Acker, KU Leuven / Materials Engineering

LCA methodology is often used to promote the circular economy in the construction sector. However, this key narrative has its limits. In order for the circular economy to be implemented, it is needed to systematically assess the environmental impacts of construction materials. Construction materials can accumulate in buildings and infrastructures for several decades, with considerable stocks of materials along the life cycle. Due to the long life of construction materials, LCA should take into consideration also time related aspects. However, in the current LCA, any temporal information is lost, making mean that LCA methodologies better suited for retrospective assessment rather than forecasting purposes. To fill this gap, this study proposes a time-dependent LCA on climate change, to assess the carbon footprint of two newly developed construction materials, produced through the recycling of industrial residues (stainless steel slag and industrial goethite). The results of the dynamic LCA are compared to the results of traditional static LCA, to see how the methodological development of dynamic LCA may have an influence on the final environmental evaluation for construction materials. Both dynamic and static LCA results show that the recycling of industrial residues to produce new construction materials has the potential to mitigate the climate change impacts of construction blocks, by substituting traditional OPC concrete. Although the dynamic LCA did not result in a shift in the ranking between the three materials compared with static LCA, it provides a clearer picture on emission flows and their effect on climate change over time.

TU226 Pursuing 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 developing their own approach to the concept of a circular economy by moving towards 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 (LCA), material flow analysis (MFA), input-output analysis (IOA), and economic accounting (EA). Our 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 consequential LCA
H. Helder, 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 various scales. In this contribution, we seek to determine whether research has effectively quantified the environmental performance of the CE strategies across scales within the planetary boundaries. Two assessment levels will be considered, i.e., cities and products/sectors. The framework will be applied and tested through an analysis of cement production and concrete (i.e., wooden houses, car tire, packaging and food waste), as well as on European cities applying CE-related strategies. At the city level, CLCA will be combined with territorial LCA to provide information on the environmental variations associated with local CE strategies. Thus, we might be able to determine the impacts of production and consumption activities that meet the needs of a city before and after the application of CE strategies. At the product level, case studies will examine the different CE strategies such as eco-design, recycling and cascade use of resources. To address how sustainability changes with scale, sector-wide scenarios for different measures and changes in market shares will be constructed through a CLCA approach. Opportunities and challenges for the specific sector and context will be identified to include side effects and to ensure assessing realistic pathways. In this way, 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.

TU228 Opportunities and threats in water treatment options as investigated by LCA
Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; T. van den Brand, KWR Watercycle Research Institute / Water systems and technology; R. Hofman, KWR Watercycle Research Institute / Chemical Water Quality and Health; S. Koeks, KWR Watercycle Research Institute

Opportunities and threats in water treatment options as investigated by LCA

In this research two LCA studies are presented as starting points in studies on water treatment options. Wezel et al. focused on the comparison of water treatment options in the road sector with the aim to determine whether the use of recycled asphalt mixture in high performance pavements (HPP) has higher environmental impacts than the asphalt mixes currently used in Europe. This type of asphalt mixture generally provides similar or lower carbon footprint than the asphalt mixes currently used in Europe. This type of asphalt mixture generally provides similar or lower carbon footprint than the asphalt mixes currently used in Europe. Thus, we might be able to determine the impacts of production and consumption activities that meet the needs of a city before and after the application of CE strategies. At the product level, case studies will examine the different CE strategies such as eco-design, recycling and cascade use of resources. To address how sustainability changes with scale, sector-wide scenarios for different measures and changes in market shares will be constructed through a CLCA approach. Opportunities and challenges for the specific sector and context will be identified to include side effects and to ensure assessing realistic pathways. In this way, 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 chemical flocculants. Recycling of flocculants from iron sludge is applied in the drinking water purification or waste water treatment process and looks 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 pretreatment step consisting of dissolved air flotation and biogeneration. However, either discharge directly into 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 into 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, CETAqua / MASE; M. Amores Barrero, CETAqua, Water Technology Centre; D. Marin, CETAqua, Water Technology Centre / Environment and Socioeconomics; M. Isasa, CETAqua Water Technology Centre / MASE; M. Termes, CETAqua/M. Ruiz Mateo, CETAqua 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 approach to develop etc. In this study, municipalities and wide geographical areas, act as accumulators of resources that in the current linear model create negative externalities. However, these 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 Feliu de Llobregat an 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 implementation of the indicators to establish relevant 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 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 biorefinery 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) that is fed up to the territory level. A feedback loop will be built between the modules of biorefinery, material flow and the groundwater 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 influential 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, R. Kelley, M. Stucki, Zurich University of Applied Sciences / Institute of Sanitation; J. Sohn, The Technical University of Denmark (DTU) / Management Engineering; J. Toja, CETaqua, Water Technology Centre / Santiation; M. S. Lopez, CETAqua Water Technology Centre / MASE; G.C. Vega, CETaqua Water Technology Centre / MASE; Y. Lorenzo-Toja, CETaqua, Water Technology Centre; D. Marin, CETAqua, Water Technology Centre / Environment and Socioeconomics. 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 limited 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 substrectors 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 over different types of waste remain challenging.

TU232 Evaluation of nutrients and energy recovery technologies through Life Cycle approaches M. Ruiz Mateo, CETAqua Water Technology Centre; M. Calvet, CETAQUA / MASE; S. Lopez, CETAqua Water Technology Centre / Santiation; M. Isasa, CETAqua Water Technology Centre / MASE; Y. Lorenzo-Toja, CETAqua, Water Technology Centre; D. Marin, CETAqua, 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 forefront of the water industry to promote wastewater reclamation as a valuable resource from the finite reserves of phosphate rock, that moreover, are located out of Europe. The LIFE RECOVERY 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 effluent 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 quantify the environmental burdens of the alternative processes in comparison to the conventional ones. The scope of the product system is limited to the plants which are located in 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, including also the cost incurred during the life cycle (CAPEX 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.

TU23 Life Cycle Assessment of a novel process of polyhydroxyalkanoates production with waste and by-products from wine industry via chain A. Novi, Università di Bologna / Centro Interdipartimentale di Ricerca per le Scienze Ambientali; L. Vogli, Università di Bologna / Centro Interdipartimentale di Ricerca Industriale su Energia e Ambiente; S. Rigbi, University of Bologna / Physics; S. Macrelli, R. Conti, Università di Bologna / Centro Interdipartimentale di Ricerca Industriale su Energia e Ambiente; C. Samori, C. Torri, Università di Bologna / Dipartimento di Chimica G Ciamian; E. Tagliavini, Università di Bologna / Dipartimento di Chimica G Ciamian Alma Mater Studiorum EU wine production accounts for some 60 percent of worldwide output, with France and Italy being the largest wine producer countries in the world (Gaeta and Corsini, 2014). The wine industry influences the environment with the use of soil, water, energy, fertilizers and pesticides. In addition it produces liquid and solid organic waste that has to be managed in the proper manner in order to minimize environmental impacts. In recent years, some innovative technologies have been proposed for the valorization of winery waste and by-products (i.e. grape marc, grape seeds, vinification lees, etc.) (Devesa-Rey et al., 2011). VALSOVIT is a research project funded by Emilia Romagna Region (Italy) which aims to valorize wine industry by-products. Its focus is the development of an integrated strategy for the transformation of waste from the whole oenological supply chain into high added-value products such as polymers, base chemicals, and molecules for the nutraceutical, cosmetic and agrochemical industries. In this framework, a novel experimental process for the valorization of wine lees and sewage sludge is carried out. These winery residues are subject to anaerobic, acidogenic fermentation in order to produce volatile fatty acids (VFAs), which in turn are used to feed a mixed microbial community (MMC) able to accumulate polyhydroxyalkanoates (PHAs) granules as carbon and energy intracellular reserve. The last step consists of PHAs extraction using dimethyl carbonate (DMC). Life cycle assessment is applied to calculate and compare the environmental impacts related to the production of one kg of PHA using the following characterization model: the LCA model, which in turn includes the calculation and comparison of the environmental burdens related to the production of the PHA. The system boundaries considered in the study are functionally complete and are described through the Input-Output Analysis (IOA) methodology.

TU24 Environmental, social and economic challenges towards a bio-economy based: the STAR-ProBio project, Sustainability Transition Assessment and Research of Bio-based Products P. Masoni, Università Sapienza University of Rome; S. Rigbi, University of Bologna / Physics; E. Merloni, University of Bologna; L. Summerton, University of York; L. Ladu, Technische Universität Berlin; A. Koutinas, Agricultural University of Athens; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; S. Ugarte, SQ consult; J. Golaszewski, UniversitätsWarmeMaksurzi W Olsztyn; K. Waskiewicz, ChemProf; N. Boccadoro, Quadra Srl; O. D. A. MONSO, University of Rome; J. Olofsson, Environmental Protection Agency Naturvardsverket; S. González-García, University of Santiago de Compostela CIF Q1518001A / Chemical Engineering; D. Pedrigo-Fazio, European Environmental Citizens Organisation For Standardisation; M. Grill, AgroVet GmbH STAR-ProBio is a multi-actor collaborative Research and Innovation Action (RIA) coordinated by Università 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 727740. 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 intends to contribute to 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 techno-economic 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.

TU25 Integration of a Colombian bio-refinery from industrial palm oil waste into the circular economy J. Torres, Universidad de la Salle / Grupo de Investigación en Gestión del Riesgo y Cambio Climático; I. Herrera, D. Garrain, A. Gamarra, CIEMAT / Energy Dept Energy Systems Analysis Unit of Bio- based Products; E. Merloni, University of Bologna / Dipartimento di Chimica Giacomo Ciamician Alma Mater Studiorum 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 an important activity and palm-based products contribute to develop a 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 gaps for future 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.

TU26 CRADLE-TO-GATE LIFE CYCLE ASSESSMENT OF BIOMASS PRODUCTION FROM PALM OIL MILL EFFLUENT N. Abdul Aziz, M. Mohi Hanaﬁ, Universiti Kebangsaan Malaysia / Environmental Science Exploring renewable energy sources is becoming increasingly important due to its low environmental impacts as compared to the consumption of non-renewable fossil fuel sources. Waste-derived biogas is one of the promising technologies that yields a renewable, sustainable, and green source of energy. In Malaysia, palm oil mill effluent (POME) can be a suitable feedstock for biogas production due to its abundant and high potential in energy generation. However, a comprehensive assessment need to be conducted to ensure the sustainability of POME-based biogas production. This study was conducted to evaluate cradle-to-gate life cycle impact assessment (LCA) and cost analysis associated with the production of biogas by the anaerobic digestion of POME. The functional unit was defined as 1 tonne of POME used for biogas production and the system boundaries covered the plantation-processing mill-biogas plants stage. The life cycle assessment (LCA) was performed using ReCiPe 2016 environmental impact method and SimaPro 8.0 software. The present study demonstrates that the generation of electricity from biogas is more advantageous comparing electricity production in conventional power plants. The results also able to identify hotspots in the life cycle of the biogas production where environmental performance of the system can be improved and environmental benefits can be achieved from the anaerobic digestion of POME with regard to the reduction of greenhouse gases emission.

Circular economy: what does restaurant food waste generation and data collection mean? R. Dagilūtė, Vytautas Magnus University / Environmental Science Department; A. Musteikytė, Vytautas Magnus University 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 regions. This wastage has an enormous negative impact on the global economy and food security. In order to elaborate and assess the environmental impacts, FAO has launched "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 towards this problem. Catering business was closely monitored in terms of customers' flows and food waste generated. To find out consumers' opinion about the food waste, the restaurant was surveyed (174 in total). Results show that during the six months 14744 kilograms of food was thrown away in a restaurant. Amount of food waste was linked to the total number of customers during the selected timeframe. Most of food was discharged in December, and in the spring quantities of food wasted decreased mostly due to the seasonality. Weekend effect was also registered. Flour products composed the biggest share of all food waste. It was found that the restaurant consumers had low ecological consciousness - inadequate standpoint of consuming too much, poor knowledge about what impact food waste has on environment, recycling, and opportunities to waste less. 73% of those often eating at public caterers indicate often to leave some food uneaten. Only 5% always and 22% often ask to take-away left food. 43% of the respondents indicate major importance to take-away leftovers. Most of them were with higher education and higher incomes. Although respondents had no clue on their own negative impact to the environment, they all agree that much more information on solving problems like this is needed. Therefore, policies to encourage food saving at home and public places should be promoted to deal with “food waste challenge” (2015).
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 effects of the treatments on the bioremediation ability and DDE biodegradation 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 synergistic interaction between poplar and soil microorganisms promoted a reduction of overall PCBs concentration under the Italian legal limit (D.Lgs. 152/2006) of 60 ng/g soil (Ancona 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 on the natural microbial community and on the total microbial abundance, cell viability and dehydrogenase activity. Moreover, nucleic acids were extracted from soil. The hypervariable regions V4-V5 of the 16S rRNA 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 poplar clone Monviso has been selected to study its capacity to enhance PCB biodegradation in a contaminated soil through phytoremediation. The plant-assisted bioremediation experiment was carried out in greenhouse conditions and the plant growth was measured in terms of biomass production. The results showed that the poplar 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.

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 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 (TMA), 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 biomethanate 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 high level of unexpected chlorinated congeners. At the end of the experiments, the Monviso clone showed an amplification of the untargeted congeners. 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.
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 9.0x10^3 and 3x10^6 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 hydrocarbons 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 microbial selection are therefore pivotal to develop in-situ bioremediation techniques. In this study, we performed a pollution-driven analysis in the 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 (pollutant fingerprints and soil physico-chemical properties) and the soil microbiological community structure. More than 120 soil samples were collected in the SIN Brescia-Caffaro along a tridimensional 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 availability. This study demonstrated that both the soil properties and the contaminant profile in the SIN Brescia-Caffaro shaped the structure of the residing bacterial communities, leading to hypothesize that it drove the selection of populations able to degrade the contaminants. The detection in the SIN Brescia-Caffaro soils of the bphK gene, codifying for the biphenyl dioxygenase involved in the aerobic PCB degradation process, confirmed that they host an inducible, or constitutive, potential for potential bioremediation interventions. This study also highlighted the prospect of exploiting spatial patterns of bacterial diversity as proxies for monitoring polluted sites.

TU248

Laboratory-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 ones representing the most recalcitrant fraction. Biological remediation of soil has been performed in situ using the CFF (continuous fluidized bed) reactor and both the size of the area and the economic/environmental costs of other technologies such as Dg&Dump. Biopiles will be built to treat the contaminated soil, air insufflation and nutrient addition will be considered to stimulate the aerobic biodegradation of hydrocarbons. In order to optimize this process, a lab-scale test was carried out and three different conditions were tested: natural attenuation (NA), addition of sawdust (SW) or soil to improve the soil structure and addition of compost as amending (CO). Thirty-six bioreactors were set up (6 sampling points for each condition in duplicate) and incubated for 180 days. Air pumps were used to insufflate air into bioreactors with the exception of NA ones. Laboratory analyses were performed on soil and soil gas samples at the beginning of the experiment and 6 samplings were carried out during the incubation period. Chemical analyses (GC-FID) of total petroleum hydrocarbons (TPH) were performed to evaluate the degradation rates and microbiological/molecular analyses (Total Bacterial Count, Most Probable Number-MPN, High-throughput sequencing of the 16S rRNA gene and quantitative PCR) to assess the growth of bacteria potentially involved in the degradation process. The highest degradation rate was observed in CO bioreactors (first-order rate constant K=0.180 d^-1) while lowest rates were observed in NA (K=0.004 d^-1) and SW (K=0.011 d^-1) in the first 60 days of incubation. However, a residual TPH concentration of ~900 ppm was reached in all bioreactors after 180 days starting from an initial concentration of 2660 ppm. The microbiological characterization suggested a selection of the bacterial community according to the chemical results. In this respect, MNP results show a significant increase in the number of oil-grown bacteria during the 14 days of incubation. These data will be confirmed by qPCR of the catalytic gene alkB encoding for an enzyme potentially involved in the biodegradation of hydrocarbons (on-going analysis).

TU249

Influence of Surfactants and Mycobacterium vanbaalenii PYR-1 Bioaugmentation on 14C-Pyrene Mineralization and Microbial Community Structure in PAH-Contaminated Soils.

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Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous environmental contaminants that have potential mutagenic, carcinogenic, and teratogenic properties. Bioremediation has been recognized as a versatile approach to remediate PAH-contaminated soils. However, the biodegradability of PAHs is limited by their bioavailability to microorganisms in the soil porewater fraction. In order to expedite biodegradation, surfactants at the critical micelle concentration (CMC) has been added to enhance the bioavailability of PAHs. The aim of this work was to evaluate the effectiveness of Brij-35, a nonionic surfactant and rhamnolipid biosurfactant at three concentrations (0.1X, 1.0X, and 10X CMC) and the bioaugmentation of Mycobacterium vanbaalenii PYR-1 in PAH-contaminated soil using 14C-pyrene as a model compound. Additionally, the bacterial community structure after the 50 day incubation was analyzed using 16s rRNA gene high-throughput sequencing and PAH-degrading genes were predicted using phylogenetic investigation of communities by reconstruction of evolutionary histories (PHSeq). The results demonstrated that Brij-35 surfactant at all three concentrations resulted in increased PAH mineralization after 50 days incubation by indigenous microbe populations in the sandy loam and clay soil compared to the unamended and rhamnolipid-amended soil treatments. The bioaugmentation of M. vanbaalenii PYR-1 had an immediate impact on PAH mineralization in both soils, resulting in 14C-pyrene degradation within 7 days. The addition of rhamnolipid delayed PAH mineralization in both bioaugmented soil treatments in a dose-dependent manner. It appears that the rhamnolipid biosurfactant acted as a more favorable carbon source compared to 14C-pyrene and was preferentially degraded. Similar PAH-degrading genes increased in relative abundance after PAH addition, especially Bacillus and Sphingomonas. Species richness and Shannon diversity increased following the addition of 14C-pyrene compared to the unamended 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 provided important insights towards the abiotic and biotic processes as well as their complex interactions in the bioremediation of PAH-contaminated soils.

TU250

Italian field results of Emeraldinated Lecithin-based Substrate used as ERD Treatment of Chlorinated Solvents in groundwater.

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ELSMicroemulsion is a food-grade carbon that supports the treatment of a wide range of groundwater contaminants, including chlorinated solvents. ELS is the acronym for Emeraldinated Lecithin Substrate, a technology designed to create 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 produce the electron donor for the reaction, thereby reducing the redox potential in groundwater. As bacteria ferment the ELS, they release a variety of volatile 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 composed primarily of phospholipids, which have both hydrophilic and hydrophobic regions in their molecular structure. As a result, ELS tend 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 cabitalotes 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 cabitalotes 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 contained 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 Nový Bydžov (Czech Republic) where groundwater pollution was identified in private wells in 2007. The source of CE 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 experiments, 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 manufacturers' protocol. Extracted DNA was quantified using Qubit 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 dsrA2 marker. All data are counted in relative values. Higher 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 prevailed in higher concentrations. Moreover, higher bacterial abundance triggered efficient sequential dehalogenation of the CE contaminants. Specific markers are still being monitored in the treated groundwater and will be discussed together with physico-chemical results.

TU252

The Influence of Nanoscale Zero-valent Iron (nZVI) in Combination with Various Organic Compounds (Modifiers) on Dehalorespiring Microflora

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Chlorinated ethenes (CE) are the second most ubiquitous contaminants worldwide. The latter 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 cob(I)alamin (vitamin B12). The latter was unravelled through quantum chemical analyses of respective electronic structure characteristics. Building on these recent results, a perturbational molecular orbital (MO) approach has been developed for a more detailed analysis of the molecular initiating event triggering the reductive dehalogenation. Application to 93 aryl halides covering chlorinated benzenes, phenols, anilines, biphensyls, dibenzo-p-dioxins, and brominated benzenes reveals that the lowest symmetry-compatible o* e* orbital located at the carbon-halogen bond mediates the dehalogenation step, and enables discriminating CBDB1-active from non-active substrates to 92%. In this way, equally 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 reductive aromatic dehalogenation proceeds through an inner-sphere electron transfer. [1] Zhang, S.; Wondrousch, D.; Cooper, M.; Zinder, S. H.; Schüürmann, G.; Adrian, L. 2017. Anaerobic Dehalogenation of Chloroanilines by Dehalococcoides mccartyi Strain CBDB1 and Dehalobacter strain 14DCB1 via Different Pathways As Related to Molecular Electronic Structure. Environ. Sci. Technol. 51. (7), 3714–3724. [2] Zhang, S.; Adrian, L.; Schüürmann, 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 commercial and industrial products since the mid-1960s; however, 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 search new strategies and cost-effective 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 live bacteria cell suspensions (189-1167 µg/g of bacterial pellet). Importantly, the data also revealed that dead bacteria have at least 7 fold lower binding capacity (38-75 µg/g of biomass) whereas live E. coli cell suspensions. The latter strain is used in the environment and the tested 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 live bacteria cell suspensions (189-1167 µg/g of bacterial pellet). Importantly, the data also revealed that dead bacteria have at least 7 fold lower binding capacity (38-75 µg/g of biomass) whereas live E. coli cell suspensions.
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 side revealed that the biocathode showed 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 inoculated with the precultures, and its 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

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 a non-return valve corresponding to thirty. Contamination is present in shallow aquifer and was higher than 10 mg/L. The efficiency of the remediation is currently about 99.9%, 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: a combination of In-Situ Enhanced Dehalogenation and In-Situ Chemical Reduction was selected to remove contamination, and bioremediation was applied in other areas. Reductive dechlorination experiments showed that halogenated products are produced during the degradation of PCE. This combination allows to have a reducing environment that helps groundwater to reach an aerobic environment which is favorable for the microorganisms to degrade the PCE into the end product, ethylene. The first injection performed an pilot scale (Phase1) to calculate 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 product an air sparging and soil vapor extraction plants have been installed in the site boundary to remove them from the soil vapor and aquifer. During the ERD we have observed methane production because of methanogenesis reaction, CH2M has decided to install a biofiltration plant, to prevent any dangers for the residential areas nearby. The challenge this complex geology has been solved by using fixed injection to avoid side effects from the treatment. For the second phase the plume was covered in each aquifer. This allowed for accurate and tailored dosage application of the product without any risk of cross-contamination. Due to the rapid effect of injection, it has been possible to observe very good reduction rates within only few months from the application. PCE, has already shown reduction of three orders of magnitude and in some points, we reached the target, with daughter compounds observed in ABL. Sulfate reduction was also observed indicating the involvement of the sulfate in the cycle process. Members of the families Desulfituromonadaceae and Prolixibacteraceae dominated the anodic community.

TU257 Bioelectrochemical sulfide scavenging from hydrocarbon contaminated marine sediments

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Zoeter, Politecnic
conditions to simulate both natural attenuation and biostimulated degradation processes. Enrichment factors for \( ^{13}C \) 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 conflicting due to contamination heterogeneity. However, species belonging to (1,2-DCE, VC). The structure of the microbial community was determined by Illumina High Throughput Sequencing, whereas its functional profile was assessed by quantitative PCR of key genes encoding for enzymes involved in specific metabolisms. Trichloroethylene and 1,2-dichloroethene (1,2-DCA) were found in most of the water samples at high concentration as well as 1,2-cis-DCE. Illumina sequencing data showed a great bacterial diversity probably due to contamination heterogeneity. However, species belonging to Trichloroethylene and 1,2-dichloroethene (1,2-DCA) will be used for the removal of excess phosphorous from eutrophic systems, for going biodegradation processes. 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 following the standard procedures. DCM and trichloroethene were found in 1,2-DCA and VC-contaminated groundwater, respectively. The functional characterization based on the quantification of catabolic genes encoding for reductive dehalogenases (PceA, TceA, VcrA, BvcA) and oxidative enzymes (emC, emE) will be accomplished (on-going analysis) as well as isotopic analyses.

TU261 Microbial ecology and ecosystem services: a key role for biotechnological applications


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

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Introduction: Despite the scenario of excess phosphorus in some aquatic environments, there has been concern with the disposal of farmyard manure 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 adsorbent. 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 following the standard procedures. 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 causes the oxidation of the superficial sediment and this oxidized layer was responsible for the reduction of internal flow of P. 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 microcosm 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 propranolol and caffeine was not observed in the sawdust. The concentrations of carbamazepine, diclofenac, paracetamol, ibuprofen, naproxen, propranolol, triclosan, estron, 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 trifluoracetate and its estimation by means of the TOP assay
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Trifluoracetate acid 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. 1,1,2-trifluoroethane) 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 liquid chromatography coupled to tandem mass spectrometric detection (IC-MS/MS). The oxidative transformation of 10 precursors (pesticides: flufenacet, fluopicolide, fluopyram, 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 wastewater treatment processes. Therefore, the formation 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 fungidial active substances used in plant protection products. Modelled groundwater concentrations of per litre 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 fungidial 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 agrochemicals not used as an active substance but instead as an ingredient in fertilizer 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
M. Andre, F. Stahl, C. Jansen, SGS Institut Fresenius GmbH
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. Selecting the methodological approach and residue analysis more challenging. There are different routes for natural background concentrations resulting from natural sources as amongst which are: physicochemical degradation from higher compounds (e.g. degradation of triglycercides 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 softerner 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. The importance of regulatory determination of the persistency 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 4-chloroanilines, 4-chloroacetanilides, and their derivatives. The biodegradation rates of these compounds were measured by monitoring CO_2 production. The results indicate 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 persistency 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 for adapted inocula.

TU267
Persistence & Biodegradation Assessment (P)
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Regulatory determination of the persistency 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 4-chloroanilines, 4-chloroacetanilides, and their derivatives. The biodegradation rates of these compounds were measured by monitoring CO_2 production. The results indicate 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 persistency 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 for adapted inocula.

TU268
Prioritization of organic compounds based on their persistency 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 persistency 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 may 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 persistency in dissolved phase, a persistency 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 persistency 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 categorization 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-anoxicarobic 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 < 0.01) 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 acetaminophen showing signs of degradation. Dissipation of pharmaceuticals in the sediment was explored in compartments 1 and 2. The sorption ability of the sorbent was overestimated and sorption 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. Albert 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 provide values for the water compartment, while QSAR models are 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, WST) and soil (Soil Screening Tool, SST) were developed based on the existing MITI test system (OECD TG 301C). The test systems MITI, WST and SST were applied to determine biodegradation data for a set of fifteen test compounds. The results demonstrate that the WST and 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 (Ptw).
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 13C NMR Bruker AVANCE III 500 spectrometer. Biodegradation potential in aerobic conditions of obtained copolymer was investigated using Micro-Oxymax respirometer (Columbus Instruments, Ohio). O2 consumption of samples mixed with soil was measured in period of 28 days. The 13C NMR spectrum of copolymer showed the signals equivalent to both components. Formation of CO2, indicated in copolymer sample (705.0 L) compared to control (350.9 L) and polyisoprene (499.5 L) after 673 h. The formation of levan and polyisoprene graft copolymer was confirmed by 13C NMR analysis. Results after 28 days in aerobic biodegradation in soil showed 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. Eniric, SOLVAY / HSE - PRA PS; P. Chagnon, SOLVAY / Research and Innovation Consulting

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 very complex. Degradation of [14C]-2,4- and 2,6-TDAs was studied in different 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 was performed to assess the influence of the temperature on several endpoints: mineralization rate of the parent compound, kinetic of degradation of the parent compound, and transformation 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.

TU275: Comparison of kinetics and products of degradation determined for the toluerediamine substances in the OECD-standardized ready biodegradability and sediment simulation tests C.R. Boegi, BASF SE / FEP/PA; C. Gaertner, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; H. Schwarz, BASF SE / RB/TC; R.J. Witt, International Isocyanates Institute, Inc. / Toxicology and Environmental Research Consulting

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 toluerediamine (TDA) substances. Degradation of [14C]-2,4- and 2,6-TDAs was studied in different soils, based on the OECD Guideline Nos. 301B and 308, wherein their disappearance, evolution of degradation products, and 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 B. Mai, Guangzhou Institute of Geochemistry; C. Huang, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences; Y. Zeng, Guangzhou Institute of Geochemistry

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, South China, were used in this study (named #1, #2, and #3, respectively). Pseudo-first factorization 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 (>44-67800ng/g, dw) and PBDE (62-792000ng/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 takes place in the sediment cores, especially for PBDEs. This conclusion was supported by the microorganism analysis, substantial Dehalococcoides were found in the sediment cores. The range of the relative abundance of Dehalococcoides for three sediment cores (#1, #2, #3) was 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) [2PBDEs (with the p values of 0.02, 0.05 and 0.01, respectively). As for CSIA analysis, only the stable carbon rations (δ13C) of BDE 28, BDE47, BDE85, and BDE99 in the top 20cm of the #3 sediment cores were obtained. An increase in the δ13C values for BDE 28 and a slightly decrease in the δ13C 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 δ13C 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 Y. Choi, Guangju Institute of Science and Technology; S. Kim, Guangju Institute of Science and Technology / School of Earth Science and Environmental Engineering

Organophosphate flame retardants (OPFRs) in aquatic environment are concerned because they are lately 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* (96 h) were exposed to the water from aquatic environment. TPHP was exposed to individual *daphnia magna* and each samples 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 1 & II biotransformation mechanisms. Diphenyl phosphate (DPHP), product of phase I reaction, was identified for biotransformation products both biota and medium. Among phase II reaction, sultonyl triphenyl phosphate was verified; intermediate metabolites were not significantly detected due to brief retention times. Parent compound (TPHP) and hydrolysis products (DPHP) were calculated by degradation ratios relative to control. Significant tendency were observed between TPHP and DPHP; as TPHP showed decreased, degradation product (DPHP) 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 ecosystem.

TU278: Photolytic and biological degradation of silicon organic compounds E. Sheng, Leuphana University Luebenburg; O. Ostrom, Leuphana University of Luebenburg / Institute for Sustainable and Environmental Chemistry; N. Mitzel, University Bielefeld / Inorganic and Structural Chemistry; K. Kuenmerer, Leuphana University Luebenburg / Institute of Sustainable and Environmental Chemistry

This study provides new data on the degradability and persistence of a selected group of silicon organic compounds, which are frequently produced in high amounts. The silicon organic compounds are an important group of industrial chemicals, which are widely used in industry, personal care products and agriculture. In general, silicones occur ubiquitous in the environment in different concentrations (e.g. in water from ng to mg per L). Since these polysiloxanes are only cleavable by chemicals, potential substitutes, which are better degradable in the environment, are urgently needed. Therefore, newly synthesised homogenized group of silicon organic compounds (p-MeOC6H4SiMe3, o-MeOC6H4SiMe3, p-MeOC6H4(p-MeOC4H8)SiMe3, p-MeNC6H2(SiMe3)2, o-MeNC6H2(SiMe3)2) with higher water solubility was investigated to provide new and reliable data on
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. However, the results of the sun light degradation does not reach a certain degree. After 6 hours, 99% of the substance p-Me,NC,H,SiMe3 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 time period of 8 hours. No ready biodegradability could be observed for these five substances. The results from the biodegradation test carried out in the literature are not on our own database on silicones. Increased water solubility of the newly synthesised silicon organic compounds did not result in an increased biodegradability in water.

TU/279 Biodegradation of adsorbed oil pollutants: Research on a model system

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 contaminants in natural systems. According to the United Nations (UN), a substance is the “chemical elements and their combinations 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 physical issues and/or difficulties in determining inherent biodegradability. This work is composed of three different studies to introduce and improve Ultimate 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 in the assessment for substances of unknown composition. 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 assayed and validated by an investigation of the biodegradability and ecotoxicology assessment of a model system. It appears that coupling ecotoxicological tests with UTOC concept 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.

TU/280 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 contaminants in natural systems. According to the United Nations (UN), a substance is the “chemical elements and their combinations 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 physical issues and/or difficulties in determining inherent biodegradability. This work is composed of three different studies to introduce and improve Ultimate 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 in the assessment for substances of unknown composition. 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 assayed and validated by an investigation of the biodegradability and ecotoxicology assessment of a model system. It appears that coupling ecotoxicological tests with UTOC concept 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.

TU/281 Development of a multi-sensors device to assess the biodegradation of chemicals

M. Crovella, University of Nantes / GEPEA CNRS UMR CBAC Laboratory; v. le cruft, L. Catherinot, E. Calzolari, V. Pichot, TRONICO; C. Sweetlove, JOREAL SA / Research and Innovation; E. Grangé, S. Jouanneau, University of Nantes / UMR CNRS GEPEA CNRS UMR CBAC Laboratory; C. Sweetlove, JOREAL SA / Research and Innovation; J. Chenèble, LOreal Research / Research and Innovation; J. Lharidon, LOreal Research & Innovation / Life Sciences Direction; T. Gerald, University of Nantes / Microbiology

Most of the methods used to evaluate biodegradation have been developed 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 accurate in situ assessment to be expressed outside the lab and out of the natural aquatic environment, 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 spiking. Water from Lake Norra Bergudnasjön 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 11 time points. After addition of mixture of internal standards, the samples aliquots 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.
modeling steps involving the use of different parameters such as $\text{O}_2$, $\text{CO}_2$, 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. Özsel 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

Since new 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 $\text{CO}_2$ 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. Alongside cell counting, several test compounds were analyzed in parallel for $\text{CO}_2$ 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. Tanimizu, Kao Corporation / Safety Science Research; M. Yamane, Kao Corporation / Safety Science; O. Morita, 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 in these systems due to technical and/or biological reasons. To overcome these challenges, 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-trimethylhexanonic 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 compound was not accessible to the test substance well on the surface water. 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-trimethylhexanonic 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 / Institute 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; J. van der Parson, University of Amsterdam / IBED

Assessment of microbial biodegradation is a key parameters 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 persistency 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-chloroaniline and N-methylpyrrole 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 generic test. Sealed bottles and 96 well plates are used for the incubation and elimination is measured by following the $\text{CO}_2$ 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 biodegradation tests (RBTs) to provide proof of non-persistence of a test chemical. Since June 2017 (time of publication of the latest ‘Guidance on Information Requirements and Chemical 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 assessments 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 most robust isolsatellite passes a well detailed characterization, which includes adaptation to included a Harmonised Offshore Chemical Notification Format (HOCNF) to the implementing OSPAR member state authority to certify use of their products offshore. Marine biodegradation 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 biodegradation biodegradation and different results are obtained from two marine BST tests (OECD 306 and BODIS) conducted on two synthetic hydrocarbon Gas-to-Liquid (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. As shown previously, positive results from enhanced RBTs are useful in persistence assessments 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 most robust isolatellite passes a well detailed characterization, which includes adaptation to included a Harmonised Offshore Chemical Notification Format (HOCNF) to the implementing OSPAR member state authority to certify use of their products offshore. Marine biodegradation 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 biodegradation and different results are obtained from two marine BST tests (OECD 306 and BODIS) conducted on two synthetic hydrocarbon Gas-to-Liquid (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.
Organising an international ring test to improve the marine biodegradation screening test

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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.

**TI/203**

Soil dissipation of paraffin oils: Improvement of the microbial degradation and impact on soil dissipation.

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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) produced by TOTAL Fluids continued to decrease to 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 natural occurrence. Occurrence. 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 this case of incubation a plateau was observed prior to dissipation. Therefore the methodology is proposed to demonstrate for certain chemicals that the degradation is of biological origin and to integrate these results in the proposed end points.

**TI/206**

Impact of biofilm growth on mercury accumulation in Daphnia magna s.issa, Norwegian University of Science and Technology; 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 freshwater organisms. For example, the common practice is to conduct 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 also accumulate mercury (*Hg*), a pollutant of high international concern because of its long-range transport and affect the ability of marine mammals to control 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.
biological 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 silver contamination. Gammarids growth and survival were not significantly impacted by the ingestion of silver-contaminated resource. On the contrary, we found a significant positive relationship between the biofilm P-content and gammarids growth, 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.

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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, we studied in this paper the decomposition processes of birch litter (*Betula pendula*) produced on 10 sites along a metallic contamination gradient to assess the effects of the contrasted litter characteristics on microbial colonization and litter consumption by, the diplopod *Glomeris marginata*, used as a model detritivore. Our results reported an impact of soil contamination on leaf litter chemical composition, leading in turn to significant impacts on diplopod physiology (in particular on increasing in the proportion of energy consumed diplopond exposed to contaminated litters). However, pollution mediated changes in leaf litter chemistry had no significant impacts on microbial litter colonization (bacteria:fungi ratio) and litter consumption by detritivore, confirming the high resilience of litter decomposition process to soil metallic contamination.

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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 tailing 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* trees (5-10) >2.5 m high and shrubs and herbs under the canopy (DF+MS); 4. Dense patches with several *P. halepensis* trees (5-10) >4 m high and shrubs and herbs under the canopy (DF+MS). B. Outside the mine tailings: 5. Polluted forest with *P. halepensis* trees >5 m high and shrubs and herbs under the canopy (PF); 6. Control forest not contaminated with *P. halepensis* trees >5 m high and shrubs and herbs under the canopy (CF). Rothro and green tea-bags were buried in each environment, to assess the effect of litter characteristic on decomposition processes, 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 emptied 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, P+MS and S: green tea =50-55%, roths tea=90% - PF, CF and P: green tea =80-85%, roths tea =20%. These percentages were maintained until the day 110. The lower decomposition in PF and CF could 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 tailings, roths tea could be used easily decomposable, 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 about = 60% in all tailings, but the qualitative resource supply easily decomposable, mainly the green tea, could stimulate microbial activity by a priming effect. Nevertheless, these results have to be confirmed in future studies.

TU301

Effects of mineral supplements on lead exposure in free-ranging herbivores.

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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 compromise the environmental and health risk not only to the area but also to other areas. Since Pb is more toxic to mesocarnivores than to mammals and people, the metabolism of Pb and therefore the benefits of Pb exposure to the fauna need to be further investigated. The study 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 phytohaemagglutinin (PHA)-skin test was used to evaluate T-cell-mediated immunocompetence. 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 lower than in control goats (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. These supplements could therefore be used by managers whose livestock are exposed to Pb in areas contaminated by mining.

TU302

Analysis of anticoagulant rodenticides, neonicotinoids and fipronil in liver of predatory birds.

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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 predators take up pesticides in food, leading to toxic levels of Pb in their body tissues. As food chains are very important mechanisms leading to this pattern, we evaluated chemical characteristics of birch litter (*Betula pendula*), on 10 sites along a metallic contamination gradient, 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 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). The 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 imidacloprid with the metabolites 5-fluorodepoxide, FDP, fipronil, which has a higher bioaccumulation potential and the metabolites (PHA) showed that a 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 phytohaemagglutinin (PHA)-skin test was used to evaluate T-cell-mediated immunocompetence. 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 lower than in control goats (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. These supplements could therefore be used by managers whose livestock are exposed to Pb in areas contaminated by mining. An assumed additional advantage is reduced the risk of Pb exposure through milk consumption by the local human population.

TU303

Trophi Magnification of POPs including PFCs Within A Terrestrial Food-Web Of An Avian Top Predator, the Cooper’s Hawk (Accipiter cooperii)

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SETAC Europe 28th Annual Meeting Abstract Book
Spatial comparison of contamination and biomagnification profiles of TU305 highlighting the importance of rivine Hg inputs for contamination of coastal marine ecosystems. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface. To address these issues -15N. Conservative mixing, reflected in the marine interface.
standards of several heavy metals, to contribute to water quality management to take forward the conservation of macroinvertebrate communities. The specific objectives of the study were: first, to model the relationships between 4 macroinvertebrate community metrics (number of families and abundance of EPT and PT), one multimetric (METI) and a predictive model (NORTI), using the Cu and Hg body residue as predictor variables; second, to assess Cu and Hg toxicity to benthic communities through the estimation of effective body residues (ER); and third, to investigate the taxa-specific differences in metal ERs in relation to their feeding styles. The ERs were estimated for each taxon and metal from the best non-linear models, selected using Akaike's Information Criteria, and compared with the 90th percentiles (P90) of the data distribution in the reference sites of the study area, considered an approach to threshold (no-effect) concentrations. The models were fitted for Cu and Hg, but only in few instances for Hg. Results showed that Cu-ER90 and Cu-ER90 in 4 taxa (Baetidae, Hydropsychidae, Ephemerellidae and Microdrilus oligochaetae) were usually less than twice above the P90, calculated for the same taxa. These ERs in other 3 taxa (Heptageniidae, Ephemeridae, Rhaphididae) were mostly within the range of 2.1 to 5.0 times the P90. The largest ratios were found in ER90 for Lumbricidae and Perlidae, which reached 12 times the P90 values. In the case of Hg, the predator taxa (Rhaphididae and Perlidae) and some of their potential preys, e.g. mayflies and simulids, showed ERs that were typically within the range of 1 to 3 times their respective P90s.

TUS08 Trophic transfer of Cadmium nitrate in a simplified marine food chain: experimental feeding rate of gelatinous zooplankton Aurelia sp. and Sanderia malayensis on crustacean Artemia sp. E. Costa, C. Gambardella, V. Piazza, CNR ISRAN; S. Lavorano, Costa Edutainment spa Aquario di Genova; M. Faimali, F. Garaventa, CNR ISRAN Trophic interactions are a crucial vector of contaminant transfer in both aquatic and terrestrial ecosystems. In the marine environment, Cnidarian jellyfish are known to play an important role in food webs as major predators of metazooplankton and as prey of apex predators, influencing the microbial loop, through direct and indirect effects, besides regulating the marine biogeochemical fluxes. In this study, the potential contaminant transfer was investigated in simplified marine food chains. The nauplii of the brine shrimp Artemia sp. and the ephyrae of Aurelia sp. and S. malayensis were selected as primary and secondary consumers, respectively. Cadmium nitrate were selected as toxicant. Performed experiments consisted in feeding ephyrae, every 24 hours for 5 days, with nauplii of Artemia sp. previously exposed, for 6 hours, to different concentrations (0.1–0.5–1–2–4 mg/L) of Cadmium nitrate; this range was selected through preliminary trials aimed to define the cadmium LC50 value for crustacean larvae. At the end of feeding experiment (5 days), the effect of Cadmium nitrate treated crustaceans on ephyrae jellyfish was investigated by the “ingestion rate method”, the “predatory performance” and biometrics and bioenergetics parameters (DiSch diameter, ash-free dry weight_AFDW and gross growth efficiency_GGE). In addition, 24 hours after each feeding treatment, two ecotoxological end-points were evaluated on jellyfish ephyrae: Immobilization and Frequency of pulsation (number of pulsations/min). Results showed a 100% of feeding rate and predatory performance in both control and treated jellyfish (A. aurita and S. malayensis), Cadmium nitrate treated Artemia nauplii, once ingested, caused in ephyrae a decrease of DiSch diameter and AFDW and also an inhibition of GGE% (Aurelia sp.EC: 3.82 mg/L). As regards ecotoxicological assays, immobilization was never affected (effect < 50%), while feeding or percentage values decreased significantly after each feeding treatment. These findings suggest a contaminant transfer from crustacean nauplii to ephyrae able to induce sublethal effects.

TUS09 Tissular injuries in Crassostrea virginica as evidence of the trophic transference of copper and cadmium via Chlorella sp. F. Mares-Guzman, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiología; G. Barrera Escorcia, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiología; X. Guzman-Garcia, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiología Eutrophic waters are a main source of metal contamination in small quantities carrying out n biological functions. In contrast non-essential metals do not have a known biological function. When metals are incorporated by the organisms they can cause damage and their presence indicates contamination. Several human activities contribute to the increasing load of both essential and non-essential metals in the aquatic environment. Microalgae such as Chlorella sp., are the primary link in the trophic chain. In the aquatic environment, they can incorporate contaminants by absorption or adsorption. If these algae accumulate contaminants, such as metals, the organisms that feed on them like the oyster Crassostrea virginica can in turn incorporate them through filtration, which may have negative consequences. The objective of this work was to evaluate the effects derived from the trophic transfer of copper and cadmium from Chlorella sp. to C. virginica. Microalgae were then cultured for 110 h at a subthalatal dose of copper and cadmium (0.1 mg/L). A concentration of 30 X 10⁶ cells was given to C. virginica for 21 days. The evolution of histopathological lesions in C. virginica was evaluated in days 0, 5, 10, 15 and 20 of the assay. The analysis performed in the digestive gland revealed diverse lesions ranging from the 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 infection and injuries that compromise the body’s physiological processes such as feeding and breathing. These damages were evident after the first 96 h of exposure to the contaminated food. However, lesions induced by Cd/Cu may have no known biological function, in percentage of more 50% of organisms in could be observed on day 10 and those associated with more than 50% of animals in cooper exposure were deferred to day 15. The presence of Chlorella 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.

TUS10 Can microplastics save us? Effects of microplastic particles and particle-bound trace contaminants in an artificial aquatic food web L. 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 Artemia spec. nauplii and zebrafish (Danio rerio). Therefore, cryogenically ground microplastic particles, made of polystyrene (P)

TUS11 Toxicokinetics links predator-prey dynamics to assess zero-valent iron nanoparticles bioaccumulation in a Caenorhabditis elegans-Escherichia coli ecosystem system X. Yang, National Taiwan University / Bioenvironmental Systems Engineering; Y. Lin, National Health Research Institutes / National Institute of Environmental Health Sciences; H. Lin, National Taiwan University; C. CHEN, National Taiwan University / Bioenvironmental Systems Engineering; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering 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 bioinetic and consumer-resource dynamics in the Caenorhabditis elegans (C. elegans)-Escherichia coli (E. coli) OP50ecosystem. Methods: The bioinetic parameters, uptake and depharination 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 Fe⁰NPs 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 Fe⁰NPs exposure. We also observed that internal concentrations of Fe⁰NPs 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 Fe⁰NPs in worms were mainly influenced by biomass conversion rate for bacteria ingested by worms, whereas parameter of death of worms had the smallest effect on worm internal concentrations. Conclusions: Model application to toxicokinetic results confirms the hypothesis that the consumer-resource dynamics are effectively associated with Fe⁰NPs 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) TUS12 INTEREST OF IN VITRO BIOASSAYS (YES/YAS) FOR THE SCREENING OF ENDOCRINE DISRUPTION IN SURFACE WATERS OF WALLONIA (BELGIUM) C. Chalon, ISEP; Y. Meuffe, Inst. Scientifique de Service Public / Ecotoxicology Department; A. Claessens, F. Crippiat, ISEP; K. Nott, SWDE; V. Brahly, F. Delloye, SWP-DG03-DEE 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 Wallonia (groundwater, surface water and wastewater). Almost 200 substances were screened, including hormone estrone, alkylphenols, phthalates, chlorophenols,
perfluorates, PBDEs, PCBs, HAPs and pesticides. In parallel with analytical methods, YES and YAS bioassays were conducted in order to quantify estrogenic activity in 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. acute teratogen, algae and Daphnia magna) have not been selected for screening, since they were already well proven. No detailed evaluation was required, however, as they 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 reviews 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 Concave report in 2018.

TU313
Ecotoxicological tools to assess the impact of pollution of tributaries to the Alqueva Reservoir (Southern Portugal)

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Degradation of surface waters and biodiversity loss at different spatial and temporal scales occurs through multiple stressors whose effects are difficult to separate and identify. Efficient management of water bodies depends on the development and selection of robust, sensitive and easily applicable tools that allow prioritizing the pressures and stressors that act in a basin, and mitigate their effects. The Alqueva reservoir constitutes the most important water supply source in southern Portugal, a semi-arid region with high levels of water scarcity and where agriculture is one of the main activities. The aim of the present study was to assess the use of an ecotoxicological tool-box in tributaries of the Alqueva reservoir for detecting chemical alterations that may influence the water quality of the reservoir. Water samples were collected along 2017 at four tributaries of Alqueva (streams of Zebro, Alamos, Amieira and Lucefécit) and analyzed for: (i) physical-chemical support elements (pH, temperature, dissolved oxygen, conductivity, chloride, total phosphorus, Kjeldahl nitrogen, ammonium, nitrite, nitrate, BOD, COD), (ii) hormones and respective mixtures and (iii) sublethal endpoints, using biotest indicators representing different trophic levels (Vibrio fischeri, Daphnia magna, Daphnia magna). In general, Zebro and Lucefécit presented concentrations of BOD (Zebro: 4.0-35.5 mg L\(^{-1}\); Lucefécit: 2.3-7.5 mg L\(^{-1}\)) and total phosphorus (Zebro: 0.18-6.23 mg L\(^{-1}\); Lucefécit: 0.02-1.92 mg L\(^{-1}\)) that compromise the support of biological life, with regard to nutrient and oxygenation concerns. Concentrations of pharmaceuticals and respectively of pesticides detected were low, being benzotriaze the compound quantified at highest levels. Lucefécit was the tributary that presented higher concentrations of pesticides (with values of benzotriaze of 1.94 mg L\(^{-1}\) probably due to the intensive agriculture crops around it. Concerning to ecotoxicological analysis, the results highlighted the toxicity in sublethal parameters (reproduction, feed inhibition or growth inhibition) induced, mainly, by samples from Zebro and Lucefécit streams. So, the results from the ecotoxicological tool-box allowed identifying the streams that promote a higher chemical impact to the reservoir, which is essential to delineate management actions to improve the water quality of the reservoir.

TU314
Effects based tools for use in conjunction with passive samplers

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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 additional method to the WRMs for the assessment of the effect of chemicals on aquatic life. This work was performed in WFD 2010 as part of the EU-WG chemicals in H2020. These recommendations were presented at the last EU-WG chemicals in H2020 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.
(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 may have a different temporal dynamics and discharge patterns and thus identify the source of the substances that are causing mutagenity. We conclude that aromatic amines and nitro compounds for surface water and also polycyclic aromatic hydrocarbons for marine coastal area should be further investigated to understand the possible role of the Cold fire 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
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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 hinting at the class 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 assay and finally with the in vivo acclivization (TAS9, YG1041, TA1538 and YG1585 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, a visible difference in the classes of compounds is present. What needs to be explored is whether the compounds causing mutagenity are already present in the environment or are added after the extraction.

TU318
NTA meets EDA: A practical example
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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 Rahm) 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 analyse specific 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 this project was therefore on the identification of seasonal exponential increase 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
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Bytilins (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 antifouling paints, the main route to aquatic pollution, by Reg. 78/2003/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 specific water quality and aquatic toxicity indicator. For this reason, under the implementation of the WFD, imposex measurements have been indicated as a useful tool to link chemical and ecological 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, 1869) and Helisoma trivolvis (Linnæus, 1758). The explorative analysis is currently carried out to test if it is possible to correlate the observed profiles with individual compounds or groups of chemicals. ACKNOWLEDGMENTS

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TU320
Lessons Learned from Sibro Dam and River Restoration in Sweden
E. Hallgavist, C. Becker, P. Bönlökke Adamsen, 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) and its daughter Directives (P-Directive) are currently carried out to test if it is possible to correlate the observed profiles with individual compounds or groups of chemicals. ACKNOWLEDGMENTS

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TU321
Impacts of methymercury on growth, respiration and swimming in larvae of a marine forage fish

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 no lethal effects of MeHg to a marine forage fish at the larval stage, the Sheephead minnow (Cynops venereus). 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 flakes containing methylmercury) or natural diets (zooplankton containing MeHg, obtained from MeHg-contaminated phytoplankton) were prepared; MeHg concentration 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 turn, 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.

TU322 Comparability of Zebrafish Embryo Behavioral Assays: A Need for Standardization? Experimental Factors in an Effort to Standardize Behavioral Assays for Toxicity Testing

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In recent times, zebrafish embryos have gained wide acceptance as an alternative test model for drug development and toxicity testing. In particular, the behavioral response of zebrafish embryos is a useful endpoint to detect neurotoxic and neuroactive chemicals. Consequently, several behavioral test methods have been developed including photomotor response test (PMR), locomotor response test (LRT), swimming speed acceleration, active turn, and swimming distance test (TER). Although these methods are distinct in their application, most of their experimental parameters lack consistency in protocols such as exposure time, imaging time, age of exposure, endpoint parameter, statistical analysis etc. Therefore, there is a need to standardize these methods in order to enable comparability of test results, as well as to ensure accurate prediction of chemical activity during testing. To address this concern, we embarked on a meta-analysis of existing behavioral assays to ask these questions: 1.) Are there consistencies in hypo/hyper behavioral activity of zebrafish embryos when different assays are used? 2.) Despite lack of standardization, is it possible to aggregate the data from different assays to give useful behavioral activity? 3.) Is it possible to determine which experimental parameters are most influential for the behavioral assays? Based on the meta-analysis, we conclude that, results from different behavioral assays (LRT, PMR, and TER) are consistent with the predicted activity of a chemical. Even though, effect concentrations vary to some extent among the considered behavioral assays, most of the variability could be explained by the most influential parameters including; exposure time, age at exposure and concentration range. These results can be useful to identify the most important experimental factors in an effort to standardize behavioral assays for toxicity testing.

TU323 Effects of 17α-ethynylestradiol (EE2) on social behaviors of the false clown anemonefish (Amphiprion ocellaris)

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The synthetic estrogen 17α-ethynylestradiol (EE2) is extensively used in oral contraceptive pills, medication, cosmetics, and personal care products. It is also widely used in livestock aquaculture systems via wastewater discharges and effluents of sewage treatment plants. EE2 is commonly detected in wastewater effluents and surface waters including coastal waters. Although coastal regions are often impacted by sewage discharges, no study has been done to address the effect of environmental estrogens such as EE2 in coral reef fish. Agonistic behavior is crucial for maintaining social hierarchy in many coral reef fish. Endocrine disrupting contaminants such as EE2 may interfere fish social structure via disrupting their agonistic behavior. In this study, we aimed to use the false clown anemonefish (Amphiprion ocellaris) as an experimental model to characterize endocrine disrupting effects of EE2 in coral reef fish, with an emphasis on social behaviors. For the exposure experiment, the fish were randomly distributed to separate tanks to form small colonies consisting of three individuals and were exposed to an environmental concentration of EE2 (30 ng/L) for 4 weeks. During this period, social behaviors including agonistic behavior, submissive response, and shelter utilization were videotaped and quantitatively analyzed once a week. Our results show that growth and survival were significantly affected by EE2 exposure. However, the EE2 treatment did not alter, but social behaviors of the middle-ranked fish were significantly affected by EE2 suggesting that EE2 may cause different impact in different ranks.

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 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 to 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 prominent 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. Fluoxetine also induced a promiscuous freshwater fish with internal fertilisation. 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

Behavioural assays have been gaining recognition as a viable endpoint in ecotoxicology as they provide a link between biochemical and ecological effects of environmental contaminants. Psychotropic drugs are designed to modulate behaviours in humans, and preclinical studies have demonstrated that these compounds can also alter behaviours in aquatic vertebrates. The effects of behavioural modulating drugs have been tested from a 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 original 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 week exposures. 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 1 ng/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.
TU326
Inter-species variability in the behaviour of a marine and freshwater amphipod
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Invertebrate species are used 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 vertebrate 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 behaviour 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.

TU327
Phenological basis of individual tolerance to the benzodiazepine oxazepam in zebrafish (Danio rerio)
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Phenotypical traits 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) at environmentally relevant concentrations. Here we supply tools to design ecologically realistic experiments and risk assessments to investigate the tolerance of fish species exposed to environmentally relevant concentrations of benzodiazepines.

TU328
Reversible behavioural alterations in burbot, Lota lota, from exposure to environmentally relevant levels of oxazepam
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Benzodiazepines are frequently detected in the environment. They persist in wastewater effluent and can be found at high concentrations in treated effluent. Furthermore, several benzodiazepines are resistant to photodegradation, enabling them to persist in the environment. Benzodiazepines are designed to alter human behaviour by binding to GABA-receptors, which are found in a wide range of animals including all vertebrates. 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.

TU329
Behavioural endpoints and biochemical biomarkers as tools to investigate effects of citalopram in brown trout (Salmo trutta L.
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Citalopram (CIT) is a selective serotonin reuptake inhibitor (SSRI) which is commonly used as an antidepressant. It binds to the serotonin reuptake from the synaptic cleft and thereby inhibits the reuptake of serotonin into the pre-synapse. Due to high concentration 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 L.) focusing on development, behaviour and individual health. Both, eggs of 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 stages showed an impaired 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 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 fluffy swimming. The study is embedded in the Effect-Net (effect network in water research) Project which is funded by the Wassernetzwerk Baden-Württemberg.

TU330
Assessing the direct and indirect effects of chemical contaminants on the behaviour, ecology and evolution of wildlife: A conceptual framework
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Chemical contaminants, e.g. metals, pharmaceuticals, pesticides, are changing ecosystems via effects on wildlife. Most studies examine a limited range of endpoints, species or contaminants under laboratory conditions. Recent work explicitly based in ecological realism, however, reveals that chemical contaminants have direct and indirect effects on stress, behaviour and physiology of species. We develop a conceptual framework to integrate direct and indirect effects on individual animal behaviour, linking individual- to population-level processes, it provides a sensitive tool for holistically assessing contaminant impacts. Here, we develop 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.

TU331
Scent and sensibility: EE2 disrupts male mate choice in fish
M. Saaristo, C.P. Johnstone, Monash University / School of Biological Sciences; K. Xu, University of Alberta / Department of Renewable Resources; M. Allison, The University of Melbourne / School of Chemistry; B.B. Wong, Monash University / School of Biological Sciences
Among the handful of studies that have investigated the behavioural effects of endocrine disrupting chemicals (EDCs), only a few have attempted to disentangle the mechanisms underlying behavioural changes, such as mate choice. In fish, for example, ecological studies have shown that males base their mate choice on...
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 daphnids. To examine the impact of EE2 on mate choice behavior, 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 ‘sight’ displayed by both control and EE2-exposed males spending more time performing sight displays for control females compared to EE2-exposed females. Moreover, we observed higher aggression levels towards the control females, suggesting that EE2-exposed females were perceived as more attractive to the males. These changes can lead to a deficiency in prey capture and growth. 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 ‘sight’ displayed by both control and EE2-exposed males spending more time performing sight displays for control females compared to EE2-exposed females. In addition to these alterations, RPE invaginations, photoreceptor ellipsoids, and eyesight of the retina and iris may have reduced the fish visual exploration and prey detection parameters such as the larval’s space, locomotor activity and velocity are evaluated. Concentrative visual responses are detected by staining the larvae for an endogenous activity indicator (pERK) after the cessation of movement. Result: We provide evidence that a 1 µM, expressed by an increased dwell time in the nicotine containing zone. Higher concentrations (10 µM), on the other hand, appear to be clearly aversive, and larvae tried to escape the dish. Disruptive and aversive responses have been reported to be attributed to differential activity levels in the Habenula with accurate activation or inhibition of the reward center in the teleost brain. We are investigating whether neuroendocrine (Iridocryptin, Thiaclopropid) and psychoactive pharmaceuticals (Citalopram, Lamotrigine, Oxazepam) found in European waters trigger similar behavioral patterns. Outlook: We will dissect which chemosensory system and higher brain areas are involved in the behavioral reactions of aquatic organisms. 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. Di Nica, University of Milan - Bicocca (VAT IT12621570154) / Department of Earth and Environmental Sciences; V. Lencioni, C. M. vom Berg, MUSE-Museo delle Scienze, Dept of Dept of Biology / Department of Chemistry and Biology; K. Witte, University of Siegen / Department of Chemistry and Biology.

Background: The aim was to verify if D. magna could be employed in biomonitoring programs for 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: LoliTrack Systems and ImageJ/nwrTrack) were compared. We found that average swimming velocity and change of body position in response to undiluted samples. Exposure to serial dilutions of the effluent caused mortality only in D. magna (15% of mortality after 24 hrs at 1:1000 dilutions; 15% and 20% of mortality after 48 hrs at dilutions of 1:100 and 1:1000, respectively). For the behavioural investigations, exposure to dilutions of the treated effluent induced significant alterations of swimming parameters in both organisms (e.g., the time to achieve an average speed of movement and the cumulative distance travelled in both) at both the exposure times. Overall, these findings emphasised a higher sensitivity of D. magna than D. cinerella gr. to treated effluents. Accordingly, D. magna might be proposed as model organism to test the toxicity of WWTP effluents in alpine streams.

TU335 Do silver and titanium dioxide nanoparticles influence the fish kairomone 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.

Background: 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 system. Therefore, it is of great importance to understand how environmental contaminants can influence the daphnids' ability to detect and respond to aversive cues. Therefore, we investigated whether environmental contaminants lead to aversive or positive 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 zebrasfish 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. Concentrative neuronal responses are detected by staining the larvae for an endogenous activity indicator (pERK) after the cessation of movement. Results: We provide evidence that a 1 µM, expressed by an increased dwell time in the nicotine containing zone. Higher concentrations (10 µM), on the other hand, appear to be clearly aversive, and larvae tried to escape the dish. Disruptive and aversive responses have been reported to be attributed to differential activity levels in the Habenula with accurate activation or inhibition of the reward center in the teleost brain. We are investigating whether neuroendocrine (Iridocryptin, Thiaclopropid) and psychoactive pharmaceuticals (Citalopram, Lamotrigine, Oxazepam) found in European waters trigger similar behavioral patterns. Outlook: We will dissect which chemosensory system and higher brain areas are involved in the behavioral reactions of aquatic organisms. This will advance our understanding of the impact of chemicals on fish behavior.


Although the use of the antifouling contaminant tributyltin (TBT) has been banned since 2008 by the International Maritime Organization, it still persists in coastal and estuarine environments due to its remobilization from contaminated sediments and also as a result of illegal use, including tropical regions along the Brazilian Atlantic Coast. Poecilia vivipara is a promising model for tropical estuarine fish ecotoxicological studies and we focused here on its feasibility to address fish early life stage toxicity caused by TBT. Newborn Poecilia vivipara fish at six days after birth (dab) obtained from a laboratory breeding stock were exposed for 96h to waterborne tributyltin at 0.1; 10; 4.5; 7 and 9 µg TBT L⁻¹. plus controls and solvent controls. After exposure, we evaluated swimming speeds and trajectories of the fish, counter-current swimming resistance, ability to capture Artemia nauplii, growth in weaned period, and male and female sex ratio. Upon exposure to 4.5; 7 and 9 µg TBT L⁻¹, histopathological analysis of the retinal pigment epithelium (RPE) indicated a higepigmentation of the pigment epithelium vili 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, daily capture of Artemia nauplii and growth in weight were reduced by 85%, 60%, 33.6% and 56% relative to controls, respectively. The histopathological changes detected in the retina and iris may have reduced the visual fish 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 lead to a deficiency in prey fish to recruit to the adult population.

TU333 Chemosensory behavioral reactions of zebrafish larvae to environmental contaminants D. Villa, 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 negatively affected by certain environmental contaminants, which can either have beneficial or detrimental effects on the behavior. Therefore, we investigated whether environmental contaminants lead to aversive or positive 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. Concentrative neuronal responses are detected by staining the larvae for an endogenous activity indicator (pERK) after the cessation of movement. Results: We provide evidence that a 1 µM, expressed by an increased dwell time in the nicotine containing zone. Higher concentrations (10 µM), on the other hand, appear to be clearly aversive, and larvae tried to escape the dish. Disruptive and aversive responses have been reported to be attributed to differential activity levels in the Habenula with accurate activation or inhibition of the reward center in the teleost brain. We are investigating whether neuroendocrine (Iridocryptin, Thiaclopropid) and psychoactive pharmaceuticals (Citalopram, Lamotrigine, Oxazepam) found in European waters trigger similar behavioral patterns. Outlook: We will dissect which chemosensory system and higher brain areas are involved in the behavioral reactions of aquatic organisms. This will advance our understanding of the impact of chemicals on fish behavior.
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 TiO₂ (NM105) MNMs on the predator defence response; by chronically exposing Daphnia magna to fish kairromones 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 moulting and concluded that the nonmorphological responses of the individual organisms in TEM is taken of each daphnai 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 Propanolol Exposure

M. E. Nielsen, P. Roslev, Aalborg University / Biology and Environmental Science

Fluoxetine and propanolol 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 propanolol 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 propanolol determined from swimming time and swimming distance were comparable (1-2 mg/L). At low sublethal exposure concentrations (µg/L), nonmorphological responses in swimming behavior were observed in D. magna. Behavior profiling estimated from multiple behavioral parameters showed that fluoxetine and propanolol stimulate swimming activity at 1-10 µg/L. EC50 values for fluoxetine and propanolol 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 pharmaceutical 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 material: 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 metallothionein). 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 by video tracking of single organisms followed by image analyses. 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 swimming time and swimming distance appeared to be more responsive behavioral endpoints than swimming velocity and swimming acceleration. The EC50s for fluoxetine and propanolol determined from swimming time and swimming distance were comparable (1-2 mg/L). At low sublethal exposure concentrations (µg/L), nonmorphological responses in swimming behavior were observed in D. magna. Behavior profiling estimated from multiple behavioral parameters showed that fluoxetine and propanolol stimulate swimming activity at 1-10 µg/L. EC50 values for fluoxetine and propanolol 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 pharmaceutical 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.

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 is currently under development, for some substances, other taxa/species may potentially be more sensitive e.g. Ephemeropera, Plectoptera, 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 is preferable. 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 AgNO₃) and nanoparticle (E. marinus (n=20 per treatment) were exposed individually. The exposure via water was performed with Ag (from AgNO₃) for 5, 25 and 100 µg L⁻¹ for 96 hours. In the Ag exposure via food, the animals were fed on alternate days with control food or food impregnated 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=0.110), 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 for 25 ug L⁻¹ (p=0.048), 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. Acknowledgement: The authors thank São Paulo Research Foundation (FAPESP 2016/19634-4) for financial support. We also thank Professor Dr. Theodore Henry from Heriot-Watt University for providing the food (control, AgCl and AgNP) used in the feeding exposure.

TU340 The effects of sublethal doses of pollutants on crop pest, Spodoptera littoralis

D. SIAUSSAT, 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 pollutions, 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
hornet - 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 behavior, we found that the olfactory system of the caterpillar is not only involved in the sexual or feeding behavior of our crop pest model following application of sublethal doses of deltamethrin, methomyl and chlorpyrifos. Whereas sublethal doses of methomyl appeared to disrupt the feeding behavior of larvae, we demonstrated a hornet 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. Dumma, 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 ion (Cu2+) has a toxic effect, whereas at least 10 fold less toxic, the impact 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 Cu2+ on olfactory acuity and olfactory-mediated behaviours of rainbow trout. To establish CuNPs or Cu2+ induced olfactory-impairment thresholds, inhibitory concentration (IC) curves were determined. Fish were exposed to a geometric dilution series of CuNPs or Cu2+ for 24 hours, and fish olfactory acuity was measured using electro-olfactography (EOG). Afterwards, fish were exposed to CuNPs or Cu2+ at concentrations known to impair olfaction by 50% (322 and 6.8 µg/L for CuNPs and Cu2+, respectively) for a 24 h or 96 h exposure period. The response of fish to a social cue (taurocholic acid) was studied using EOG and a choice maze behavioural assay. After the behavioural experiment, fish olfactory rosettes were dissected to investigate if there was any DNA fragmentation as a marker of apoptosis that might be induced by CuNPs or Cu2+. 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 exposure. Continuous Cu2+ exposure to the social cue supported the results of neurophysiological experiments. Although fish exposed to control water or Cu2+ 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 Cu2+ in the exposed fish. In summary, over the same exposure periods, CuNPs and Cu2+ caused an impairment of fish olfactory function. The outcome of this study will support industry to select functional and safer alternatives.

TU342 Perfluoroalkyl acids concentrations in liquid wastes: a survey campaign and implications for waste disposal
A survey campaign has been carried out to determine the concentrations of twelve perfluoroalkyl 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 panorama of the different sectors and to verify 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 ligurs. 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 provide a proper control on the circuits of production. 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 woolen textiles (high wool content > 70%) derived by post-consumes materials (knitted apparel, apparel made up by carded woven and combed woven), pre-consumes materials (combed and carded woven, knitted fabrics, spinning and twisting trimmings), and regenerated cotton-type materials (derived from denim recycling).

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. Damásio, V. Gonzalez-Andres, M. Díez-Ortiz, G. Janer, Leitat Technological Center
Flame retardants are 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 (PFOS and Durable Water and Oil Repellents - DWRs). FRs are added to fabrics to inhibit the combustion process, and typically are products with a chemistry based on halogenates. DWRs 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 regulation under REACH regulation. The potential human 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 ingredietnal metals from and during 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)

A. Biegel-Engler, German Environment Agency - UBA / Chemicals; L. Viecke, C. Staude, German Environment Agency / Chemicals

Per- and polyfluorinated alkyl substances (PFASs) are heat resistant and show a low friction 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. The release of AFFF into the environment causes a contamination. Instead 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 fluorine 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 release 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 use 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 assumed 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 been constantly 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 suitable approaches to substitution. The authors will present some signifcant 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. Waerteschee, M. vander Straaten, 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 economically reasonable conditions. In parallel, the human health and environmental effects data generated by EU REACH and CLP Regulations lead to increasing hazard identifications and harmonised hazard classifications. 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 metal 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
Ecotoxicology of the hydrolyte byproduct of three biopesticides on the unicellular green alga Chlamydomonas reinhardtii

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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 (hydrolysates) have been separated. Both of them 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 from 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 Luisieri (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 Luisieri the most toxic compound followed by Artemisia absinthium with a very similar toxicity,and Dittrichia graveolens (Ciudad Real, Spain) the least toxic species. These 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 species 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 hydrolyte obtained from Satureja montana (Ejea, Aragon) 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 hydrolyte of Satureja montana are 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)

TU350
The Impact of the Hydrolyte 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 (hydrolysate) have been separated. Both of them showed active compounds 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 latifolia. 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 Biolog Ecoplates®. The acute toxicity of hydrocarbons was also tested by Eisenia fetida bioassay. Results indicate that hydrocarbons caused acute adverse effects in E. fetida, in particular D. graveolens and L. latifolia (LC50 in the range of dilution of 10-2). All three bioprospecctives 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 MIENCE-FEDER (CTQ2015-64049-C3-2-R).

TU351
Acute toxicity of emulsifiable concentrate of Alpinia galanga 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 insects and pests and they are developed as safe biopesticides. However, their use may be caused potent toxic effects to non-target organisms in the environment. It needs to be determined their effects on aquatic species (IC50) and their components, both in living and death environment. Alpinia galanga essential oil (AGEO) has been considered to control the outbreak insect pest, Ricina 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 AGEO were formulated for emulsifiable concentrate (E.C), they were mixed with ethanol and tergitol in a ratio of 5:1. 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 AGEO 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, 48h-LC50 values for the VFE0 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. Kim, Y. Choi, S. Lee, Kyungpook National University Recently, many researchers have developed natural insecticides to control insect pests using plant essential oils (EO) due to their eco-friendly safe properties. Cinnamon EO is one of important EOs to be a potent candidate and is formulated as an emulsifiable concentrate (E.C). 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 EO EC was used as an active ingredient, they were mixed with ethanol and tergitol as surfactants. To select an appropriate surfactants, 8 different types of surfactants (Tween 80, Sodium dodecyl sulfate (SDS), Nondent, Triton X-100, Sodium dodecyl Benzene Sulfonate (SDBS), Koliphor, Tergitol and Mixture of SDBS and Nondent) were tested for the formulation and tergitol 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 at the 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, 0.64, 2.56, and 5.12 ppm and the mean survival rate was 8.48 ± 4.7. The survival rate of the control group was 9.33 ± 0.58 and the positive control containing ethanol and tergitol 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. The mean temperature and pH of the test water was 24.06 ± 1.00. These compounds showed different efficacy in reducing fungal growth and mycotoxin accumulation. The most active compounds were used to perform cyto- and geno-toxicity tests on healthy human cells, particularly on human cell lines deriving 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 antiaflatoxic activity of several thioseminocarbazone ligands 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 antiaflatoxigenic activity towards A. flavus. These compounds showed different efficacy in reducing fungal growth and mycotoxin accumulation. The most active compounds were used to perform cyto- and geno-toxicity tests on healthy human cells, particularly on human cell lines deriving 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 antiaflatoxic activity of several thioseminocarbazones and to determine the potential risk for environment and human health with a view to use these compounds in field. Financial support: Fondazione Cariplo-Progetto N. 2014-0555, http://aflatox.unibs.it/

Understanding human and environmental exposure to chemicals in urban systems (P)
TU354
Electronic products are related with household exposures in Canadian residents
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Key Words: electronic products; home; exposure; FRs; PBDEs; persistent organic pollutants (POP); personal care products
Background: Exposure to flame retardants (FRs) is a public health concern, as FRs are found in a wide range of consumer products, building materials, and industrial uses. Their wide use and persistence in the environment are attributed to their unique chemical properties, such as high heat resistance, low flammability, and plasticizing ability of the microbial community for degrading different carbon sources present in Biolog Ecoplates®. The acute toxicity of hydrocarbons was also tested by Eisenia fetida bioassay. Results indicate that hydrocarbons caused acute adverse effects in E. fetida, in particular D. graveolens and L. latifolia (LC50 in the range of dilution of 10-2). All three bioprospecctives 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 MIENCE-FEDER (CTQ2015-64049-C3-2-R).

TU353
Thioseminocarbazone scaffold for the design of antifungal and antiaflatoxigenic 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 aflatoxins production, harmless to the environment and to human health. We have evaluated the biological activity of several thioseminocarbazone 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 antiaflatoxigenic activity towards A. flavus. These compounds showed different efficacy in reducing fungal growth and mycotoxin accumulation. The most active compounds were used to perform cyto- and geno-toxicity tests on healthy human cells, particularly on human cell lines deriving 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 antiaflatoxic activity of several thioseminocarbazones and to determine the potential risk for environment and human health with a view to use these compounds in field. Financial support: Fondazione Cariplo-Progetto N. 2014-0555, http://aflatox.unibs.it/
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 nanomaterials used in outdoor paints to urban surface waters at high spatial and temporal resolution
M.D. Núñez, University of York / Environment; A. Pratutiorius, University of Warsaw / Department of Environmental Geosciences; A. Boxall, University of York / Environment Department

The expansion of the nanotechnology sector is leading to an increased use of products containing engineered nanomaterials (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 new modeling framework in the theory of Convergent Technology 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 parameterize 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 triclocarban in personal care products from Korea
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Among ten parabens, methyl paraben (MeP) showed the highest detection rate (98%), followed by propyl paraben (PP, 94%) and butyl paraben (BuP, 41%). TCS had only 20% detection rate and TCC 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, eyeliner, body/hand lotions and lipstick. The daily exposure levels of parabens associated with the consumption of CPCPs were calculated using DEAMPY with a median of 3.0 ng/mL followed by PNP, 3-fluoro-4-ethylphenol (4-TEP, metabolite of parathion), 3,4-dichloro-2-hydroxybenzyl alcohol (DEAPA, metabolite of parathion), 4-nitrophenol (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-pyrimidinol (IMPY, metabolite of diazinon) and 2-dimethylamino-6-methyl pyrimidin-4-ol (DEAMYP, metabolite of pirimiphos). Concerning pyrethroids, most compounds such as permethrin, cypermethrin, deltamethrin, allethrin, resmethrin and fenvalerate were metabolized into one single compound, 3-phenoxybenzoic acid (3-PBA). On the other hand, cyfluthrin is metabolized into 4-fluoro-3-phenoxybenzoic acid (4-F-3-PBA). Therefore, 3-PBA and 4-F-3-PBA can be used as 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 TCPY (87%). The metabolite showing the highest concentration was DEAMYP with a median of 3.0 ng/mL followed by PNP, 3-PBA and TCPY with medians of 1.3, 0.56 and 0.47 ng/mL, respectively.

TU358
Analysis of metabolites of organophosphate and pyrethroid pesticides in urine from Italian children
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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, reproductive and neurological problems, among others. Children are more vulnerable than adults to environmental pollutant exposure because their organs and metabolism are 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. Organophosphate metabolites are metabolized into dialkyl phosphates (DAPs) and hydrolysis products, such as chlorpyrifos oxon (TCPY, metabolite of chlorpyriphos), 4-nitrophenol (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-pyrimidinol (IMPY, metabolite of diazinon) and 2-dimethylamino-6-methyl pyrimidin-4-ol (DEAMYP, metabolite of pirimiphos). Concerning pyrethroids, most compounds such as permethrin, cypermethrin, deltamethrin, allethrin, resmethrin and fenvalerate were metabolized into one single compound, 3-phenoxybenzoic acid (3-PBA). On the other hand, cyfluthrin is metabolized into 4-fluoro-3-phenoxybenzoic acid (4-F-3-PBA). Therefore, 3-PBA and 4-F-3-PBA can be used as 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 TCPY (87%). The metabolite showing the highest concentration was DEAMYP with a median of 3.0 ng/mL followed by PNP, 3-PBA and TCPY with medians of 1.3, 0.56 and 0.47 ng/mL, respectively.

TU359
Pesticide levels in parturient and newborns from Aveiro region, Portugal
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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 depict exposure and evaluate effects. Polycyclic aromatic hydrocarbons (PAHs) are a group of priority chemicals to be studied and monitored as they are considered carcinogenic and teratogenic. In the present study we aimed to monitor human fetal exposure to PAHs by measuring concentrations of naphthalene, phenanthrene, pyrene and BaP equivalents in placenta, umbilical cord and mother’s blood (plasma and blood cells) of 49 parturient from Aveiro region, Portugal. Information organized in questionnaire forms, tissues and organs were collected following the
parturient 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 concentrations than high molecular weight PAHs (pyrene and benz[a]pyrene). Moreover, increased levels of 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 the 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 J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; I. chestnate, in used in ARC Arnot Research & Consulting; L. Li, University of Toronto at Scarborough / Department of Environmental Sciences; X. Zhang, University of Toronto Scarborough / School of Engineering and Applied Sciences; B. Givechhi, J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences

The aggregate exposure pathway (AEP) model is a conceptual framework to help allocate the relevant environmental 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, identifying chemical concentrations throughout the 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 emphasizing the relatively high potential for human exposure to chemicals indoors compared to outdoor environments. Following the same exposure scenario, unit emission rate based whole body concentrations (exposure potential) range from 3×10⁶ to 5×10⁷ mmol/kg. The differences in ranking chemicals for exposure based on either external (intake fraction) or internal (concentration) exposure metrics are substantial due to chemical-specific differences in toxicokinetics. In absence of well-defined chemical use information, the model calculated critical emission rate 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 10 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 SOUTHEASTERN NIGERIA S. Anika, UNIVERSITY OF NIGERIA, NSUKKA; V. Ahur, Federal University of Agriculture Makurdi / Department of Veterinary Physiology, Pharmacology and Biochemistry; P. Onyeyili, Federal University of Agriculture Makurdi / Department of Physiology Pharmacology and Biochemistry

Lead is a soft, ductile, heavy 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 Bukkuyum and Anka LGA of Zamfara State, Nigeria, due to chronic lead toxicity were reported by Medecins Sans Frontieres (MSF, Holland) to the health authorities (UNEPOCHA 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 headsmen and farmers. This outbreak was reported as the worst in modern history (UNEPOCHA 2010), Open-pit mining of lead in the Ishiagu 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 Ishiagu town and environs. Soil, water, grasses/plants, food, fish and quarry dust were collected between March and May 2017, processed and analyzed for lead concentrations. All water samples exceeded WHO recommended 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 Ishiagu 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 M. Makome, Scientific Services, Cape Town / Department of Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; V. Sciallasci, C/ET / University of Siena / Department of Chemistry, Earth and Environment; R. Bagagli. University of Siena / Department of Physical Sciences, Earth and Environment

Rare Earth Elements (REEs) form critical elements required in technological accessories. 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 depositions and their leachability in urban environments such as in fact, acid dissolution and alkaline fusion are commonly employed. Various instrumental techniques that have been used recently includes electrothermal vaporiser or laser ablation accessory connected to ICP-MS or ICP-OES, X-ray Fluorescence and electro-analytical techniques. These methods of analysis including their accuracy, limit of detection (LOD) and limit of quantification (LOQ) are compared and discussed. This study further contains the present of the potential elemental markers 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 stonework snail as a new biomonitor of metal contamination in the urban environment E. Rota, B. Braccini, R. Dei, University of Siena / Department of Physical Sciences, Earth and Environment; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; S. Ancora, University of Siena / Department of Physical Sciences, Earth and Environment; R. Bagagli, University of Siena / Department of Physical Sciences, Earth and Environment

Papillifera 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 inhaled fine particles, is a promising indicator of urban pollution and is a promising urban biomonitor. It is a filter feeding gastropod and organisms widely used as biomonitor of urban pollution, i.e. mosses, lichens 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 commercial areas the three main polluted areas (Carpi, Rivarolo and Mafra) in the city of Genoa (Italy), the 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. Four 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 primary 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 risks associated with ingestion of metal contaminants were 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. Lencioni, C. Casal, L. Castel, UniLaSalle Campus Rouen / AgThy Unit
Projects seeking to produce home-consumption vegetables in urban areas and, more generally, to set up agricultural production inside cities, are increasingly numerous. Agglomeration 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 (Northern France) for three purpose: pasture, a forest recreation, and a 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 main 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 topography 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 in the upper 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 rhizoremediation potential
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Among the national priority polluted sites, the SIN Brescia Caffaro is 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'. About 100 Ha of agricultural areas, contaminated with polychlorinated biphenyls (PCBs) ranging from 15000 to 13000 µg/kg in the top 30 cm to about 0.6 µg/kg at 1 m depth.; Pb, and Zn in mobile phases extracted from different size fractions using a modified sequential extraction procedure after Tessier (Tessier at al., 1979): adsorptive and ion exchangeable, reducible (1% sodium dithionite), and 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 dusts, 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. 54 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 Brescia 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 related to the city's metal party and a few industries. The dataset also provides insight for managing urban allotments for urban agriculture production.
particles in the fine mode, while AA responded mainly to dusts rich in metals and metalloids. Furthermore, the DTT assay evidenced was more sensitive to organic substances, while the AA assay showed a different sensitivity towards the oxidant species: the DTT method evidenced OP values were scarcely dependent on the sampling site: carbon, in order to identify the relationships between OP values and PM chemical composition. In this work we applied three of the most used scientific approaches have been evaluated in order to gain information about it. The oxidant potential of particulate matter (PM) concentration and composition on human health, and different acellular assays are currently used in literature for its characterisation of NVP in wastewater samples, with concentrations ranging between 250 to 500 mg/L. Further evaluation of the impact of NVP on the aquatic ecosystem was also considered and reported in this study. Keywords: Anti-retrovirals; Emerging pollutants; Chromatography; Wastewater

TU372

Lumichrome blue: a selective photometric reagent for chlorine dioxide analysis in water

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Drinking water supply companies have traditionally focused their efforts on providing a product with health guarantees, a safe and clean water. Disinfection has been the main purpose of the water treatment. A broad range of disinfectants and technologies are available. Chlorine dioxide has been increasingly used because of its ability to avoid the formation of trihalomethanes (THMs), the most common and well-known disinfection by-products. NN-Diethyl-p-phenylenediamine (DPD) method has been without any doubt the most common procedure for the analysis of free and combined chlorine (chloramines) and also, chlorine dioxide. While this method easily permits the differentiation between free and combined chlorine, the selectivity of the analysis of chlorine dioxide when chlorine is present has recently been questioned. As a result, this procedure has been removed from Standard Methods (American Water Works Association) and qualified as “reserved” method. Given this circumstance and the need of having a selective method for chlorine dioxide, several UV–VIS spectrophotometric methods have been evaluated by our group (1). Here, the results using leucomethylene blue are presented. This chromophore agent is obtained by reduction of methylene blue and its use for the analysis of chlorine dioxide is scarcely described in the literature. Our experiments show that it presents a good analytical performance, and what is more important, measurements are not interfered by elemental chlorine. The method showed a good accuracy with raw water samples (relative error below 14 % for chlorine dioxide concentrations between 0.1 and 1.5 mg/L). This reagent has revealed to be the best option among the different compounds that we have used – amaranth, lissamine green, and choro phenol methyl red. The only drawback of the procedure is the interference by high concentrations of sulphate. Ongoing work is taking place to avoid it by previous precipitation of the interferent or liquid extraction of the dye. It is in line with the increasing development of the UV–VIS spectrophotometric methods for the determination of chlorine dioxide, and sensory analysis of chlorine dioxide in drinking water. Part II. SETAC Europe 26th Annual Meeting. May 2016. Nantes (France).

TU373

Fate and effects of triclosan in subtropical freshwater benthic microcosms

F. Peng, Wageningen UR; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group nterrala; G. Ying, Guangzhou institute of Geochemistry Chinese Academy of Sciences; H. Selck, Roskilde University / Dept Science and Environment; D.T. Salvito, Research Institute for Fragrance Materials (RIFM) / Dept of Environmental Science; N. Diepens, Wageningen University

Triclosan (TCS) is one of the top 10 most commonly detected organic pollutants in the aquatic environment. However, little is known about the toxicity and bioaccumulation of sediment-associated TCS. We examined the effect of sediment-associated TCS on the snail, Viviparidae Bellamya, and the worm, Limnodrilus hoffmeisteri, and assessed worm bioaccumulation during a 28 days experiment. The main purpose of the experiment was to test whether the DTT assay created a greater affinity with particles in the fine mode, while AA responded mainly to particles in the coarse fraction. DCFH results appear to be driven by a competition between several factors, some increasing the response and some suppressing it. Fang, T. et al.: Oxidative Potential of Ambient Water-Soluble PM2.5 in the Southeastern United States: Contrasts in Sources and Health Associations between Acetic Acid (AA) and Dithiothreitol (DTT) Assays. Atmos. Chem. Phys. 2016, 16, 3865-79. 10.5194/acp-16-3865-2016 Huang, W. et al: Optimization of the Measurement of Particle-Bound Reactive Oxygen Species with 2′,7′-dichlorofluorescin (DCFH), Water Air Soil Pollut. 2016, 227, 164.


TU371

Chromatographic determination of the pathway of nevirapine in wastewater at a wastewater treatment plant

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Emerging pollutants (EPs) are defined as substances that have been detected, which is not included in the routine monitoring programmes at the EU level, and whose fate, behaviour and ecotoxicity effects are not well understood. Pharmaceuticals used in the treatment of HIV, known as anti-retrovirals, are becoming prevalent and there is a need to quantify and characterise the compound and minimise any adverse affects to aquatic and human health. Nevirapine (NVP) is commonly used in the anti-retroviral treatment of HIV infection. It is known as a non-nucleoside reverse transcriptase inhibitor of the dipyridodiazepinone class, commonly used to minimize viral resistance. This study reports the isolation and chromatographic characterisation of NVP in wastewater samples, with concentrations ranging between 250 to 500 mg/L. Further evaluation of the impact of NVP on the aquatic ecosystem was also considered and reported in this study. Keywords: Anti-retrovirals; Emerging pollutants; Chromatography; Wastewater

TU370

OXIDATIVE POTENTIAL OF PARTICULATE MATTER COLLECTED AT INDUSTRIAL AND URBAN SITES

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The scientific world is still questioning about the effects of airborne particulate matter (PM) concentration and composition on human health, and different scientific approaches have been evaluated in order to gain information about it. The measure of oxidant potential (OP) is generally considered as a predictive index of PM ability to generate reactive oxygen species (ROS) in biological organisms, and different acellular assays are currently used in literature for its determination. In this work we applied three of the most used OP assays (dithiothreitol - DTT, acid ascorbic – AA, and 2′,7′-dichlorofluorescin – DCFH; Fang et al., 2016, Huang et al., 2016) to PM2.5/PM10 samples and to size-segregated dust samples collected by a 10-stage impactor. Samplings were performed at an industrial site near Ferrara (Po Valley; Italy) and at a traffic urban site in Rome (Italy). All the samples were also analysed for anion, cations, macro- and micro-elements, total organic content, elemental carbon and water-soluble organic carbon, in order to identify the relationships between OP values and PM chemical composition and dimension. Despite the very different composition of PM in the two monitored areas, OP values were scarcely dependent on the sampling site: species whose concentration is very different in the two areas, such as secondary inorganic ionic, seem thus to play a negligible role in the ROS generation. Each assay showed a different sensitivity towards the oxidant species: the DTT method was more sensitive to organic substances, while the AA method was more sensitive to metals. The period in microcosms and the DTT assay created a greater affinity with particles in the fine mode, while AA responded mainly to particles in the coarse fraction. DCFH results appear to be driven by a competition between several factors, some increasing the response and some suppressing it. Fang, T. et al.: Oxidative Potential of Ambient Water-Soluble PM2.5 in the Southeastern United States: Contrasts in Sources and Health Associations between Acetic Acid (AA) and Dithiothreitol (DTT) Assays. Atmos. Chem. Phys. 2016, 16, 3865-79. 10.5194/acp-16-3865-2016 Huang, W. et al: Optimization of the Measurement of Particle-Bound Reactive Oxygen Species with 2′,7′-dichlorofluorescin (DCFH), Water Air Soil Pollut. 2016, 227, 164.

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Environmental Safety
Protection goals for the ecological risk assessment of chemicals are increasingly being framed in terms of ecosystem service delivery [1]. However, the type of data collected to assess risk is generally at the level of individual organisms or simplified multi-species systems. Currently, extrapolation from what is measured to what we want to protect uses overly simplistic approaches, such as risk quotients or toxicity-exposure-ratios. Ecological models provide a more mechanistic way of connecting these disparate levels and allow for integration of other relevant information as well as feedbacks across levels of organization [2]. Here we present output from the National Institute of Mathematical and Biological Synthesis (NIMBioS) working group (www.nimbios.org/workgroups/WG_o2e). The two case studies provide a demonstration of a recently developed framework that allows quantification of mechanistic impacts of the effects of chemicals and other stressors from impacts on individual organisms to the delivery of ecosystem services [3]. The first case study applies an individual-based model (IBM) [4] to quantify impacts of potential endocrine disrupting chemicals on services provided by trout populations in a mountain stream in Colorado, USA. The second case study uses an aquatic ecosystem model [5] to evaluate impacts of an insecticide on multiple ecosystem services delivered by a lake ecosystem, modified to represent a reservoir in Iowa, USA. The first case study is an example where managing for provision of the service GCT population provides will differ depending on the level of EE2 in the system. If EE2 concentrations are low, then management would need to focus on controlling BT populations. For high concentrations of EE2, management first needs to focus on reducing EE2, followed by control of BT. The second case study is an example where the species composition and different exposure to the insecticide, as a result of interspecies interactions, and the economic valuation of this service needs to take into account with angler preferences. The service of water clarity for recreational activities was valued using threshold-based estimations of days fit for recreation. We provided concrete examples of how ecological modeling can be used to quantify impacts on ecosystem services from data gathered in standard testing. We discuss challenges and ways forward.

The Need for Resilience in Environmental Impact Assessment (P)

TU379
Recovery in environmental risk assessments at the European Food Safety Authority (EFSA)
T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; F. Bigler, Retired; G. Frampton, University of Southampton; C. Hogstrand, Kings College London / Division of Diabetes and Nutritional Sciences; T. Brock
Sulphur is a key fungicide in biological fruit production. Following a recent risk assessment, the use of sulphur in The Netherlands has been rigorously limited. Current registrations allow for two applications, which is incompatible with disease control in biological top fruit production. The regulatory decision was technically correct and based on considerations for non-target arthropods, as risk for the egg parasitoid Trichogramma could not be excluded. Under current European regulations, Trichogramma is one of the sensitive indicator species selected for higher tier testing. As this is a natural enemy of several Lepidopteran pest species, the indicator is clearly linked to the specific protection goal of preserving natural pest control potential. However, in the absence of alternatives, at a broader level this specific objective frustrates the current system of biological production as a whole. The specific protection goal must clearly be balanced against the broader objective of preserving biological production. To understand the importance of egg parasitoids such as Trichogramma in Dutch orchards, the Dutch Fruit Growers Organisation have investigated the control potential of egg parasitoids in 8 orchards (4 biological, 2 conventional and 2 untreated), using parasitization rate as a functional endpoint. The investigations show that parasitization rates are extremely low (<1% of bait cards and <0.005% of the host eggs showed parasitization), suggesting a minor role and other factors or parasites in these systems in The Netherlands, at least at the time of the study (late summer 2017). Conventional orchards showed no parasitization and regular (i.e. having sulphur treatments) and untreated orchards did not show differences. These results show that specific protection goals may have country specific weight and need to be considered and balanced against potential negative impacts of eventual protective measures, such as in this case jeopardizing biological top fruit production.

TU374
Joint Annual Meeting of the International Society of Exposure Science and the International Society for Environmental Epidemiology (ISES-ISEE 2018)
M.L. Diamond, University of Toronto / Department of Earth Sciences
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
F. Botta, ANSES / DER; F. Eymery, T. Quintaine, M. Hulin, J. Rey, O. Yamada, M. Merlo, ANSES
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 cover 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 to health impacts on wildlife, pests, 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.

TU376
Measuring and Modelling Aluminium Bioavailability and Toxicity to Aquatic Organisms
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The ability to accurately predict the aquatic toxicity of aluminium (Al) in natural surface waters has eluded scientists for the past several decades. In 1988, the US Environmental Protection Agency (USEPA) released nationally-recommended ambient water quality criteria for Al of 750 and 87 μg/L as acute and chronic criteria, respectively. However, these applied only to waters with a pH between 6.5 and 9, and the chronic toxicity database was limited. Therefore, in 2009 we assembled a team of scientists to help expand this database and identify a means for measuring and predicting the toxicity of Al to aquatic organisms as a function of water chemistry. A series of chronic toxicity tests were performed, as part of this effort, with several freshwater species. The species were selected to meet requirements for the EU REACH dossier. USEPA water quality criteria or European Water Framework guidelines for environmental quality standards. To develop bioavailability models, multiple tests with a green alga (Pseudokirchneriiella subcapitata), a cladoceran (Ceriodaphnia dubia), and a fish (Pimephales promelas) were performed across a range of DOC, hardness and pH conditions. These latter data were included in the development of a biotic ligand model (BLM) for the prediction of toxicity as a function of water chemistry. The toxicity data sets were also used to develop a multi-linear regression (MLR) model to provide a simplified means to predict toxicity as a function of DOC, hardness, and pH. Due to its complex environmental chemistry, measurement of the “toxic” form of Al in natural waters cannot be performed using the conventional “total” or “dissolved” analytical approaches. Studies have recently been completed which allowed the measurement of “bioavailable” Al in natural waters where suspended solids are present and contribute to measurements of total Al, but are non-toxic. The presentation will focus on modeling Al toxicity and measuring Al in natural waters.

TU377
Modelling impacts of chemicals on ecosystem services
N. Galic, Syngenta Crop Protection, LLC / Environmental Safety; V. Forbes, University of Minnesota / Ecology, Evolution & Behavior; C. Salice, Towson University / Environmental Science & Science Dept.; P. Thorbek, Syngenta /
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 any assessed products, and invasive alien species that are harmful for plant health. This framework proposes an integrative approach based on well-defined specific principles and guidelines that take into account a wide range of means of expressing, i.e., categorizing, modelling and monitoring, and the selection of focal taxa, communities, processes and landscapes 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 responses to changes in spatial and temporal scales caused by natural or human-caused disasters, a 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’s Scientific Committee. 2016. Recovery in environmental risk assessments at EFSA. EFSA Journal 2016:14(2):4313. 85 pp (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. Mehdix, 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 threatened or caused by natural or human-caused disasters. Associated with this challenge is the limited ability of current ecosystem models to provide defensible projections of the complex and intertwined social-ecological relationships defining a future sustainable flow of goods and services.

TU381 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; S. Salinas, Red ElÁctrica 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, design and make, and is equipped with non-standardised spill containment systems. The sites are distributed throughout the Spanish mainland and in the Baleares and Canaries 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 R’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 as risk scenario triggers for specific events at specific sites, which makes it possible to prioritize the ranking in accordance with the relative values of different ecosystem services, and cost-effective strategy to manage their regulatory obligations and financial liabilities.

TU380 Habitat Equivalency Analysis for a Restoration Resilience Model of the Rio Doce Basin
P.N. Booth, Ramboll Environ / Ecological Sciences; E. Singer, F. Gomes, R. Arantes, Ramboll Environ / Sao Paulo; R. Wenning, Ramboll Environ

A Resilience 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 means of scaling the global habitat restoration and HEA being refined through a process of stakeholder engagement to determine the valued environmental components (VECs); and thus the valued ecosystem services that should drive the resilience 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 adaptable 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 HEA at scale, and how restoration is aimed at addressing three technical challenges that must be overcome for incorporating resilience in ecosystem services planning work. First, baseline conditions must be established for the goods and services produced by the ecosystem prior to alter the environment or repair the damaged/worn environmental systems. Second, the challenge relates to the establishing the relative values of different ecosystem services, and associated changes to present and value society in the future. The third challenge is the limited ability of current ecosystem models to provide defensible projections of the complex and intertwined social-ecological relationships defining a future sustainable flow of goods and services.

TU379 Addressing Resilience in Ecosystems and Communities in Spain
P. Wouters, M. Ferreira, I. Harper, Ramboll Environ / Environment and Health; S. Salinas, Red El Áctrica 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, design and make, and is equipped with non-standardised spill containment systems. The sites are distributed throughout the Spanish mainland and in the Baleares and Canaries 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 R’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 as risk scenario triggers for specific events at specific sites, which makes it possible to prioritize the ranking in accordance with the relative values of different ecosystem services, and cost-effective strategy to manage their regulatory obligations and financial liabilities.

TU384 Quality stakeholder involvement for resilience in environmental risk

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assessment
Y. Tomkiv, Norwegian University of Life Sciences (NMBU) / Faculty of Environmental Sciences and Natural Resource Management; B. Wynne, Norwegian University of Life Sciences / Centre for Environmental Radioactivity CERAD CoE; D. Oughton, Norwegian Public Roads Administration / Norwegian University of Life Sciences / Centre for Environmental Radioactivity (CERAD CoE)

There is a global call for sustainability and systemic approaches in environmental risk assessment that consider economic and societal aspects of risk, in addition to the environmental aspect. Early and effective stakeholder involvement plays fundamental role in these considerations. Stakeholder involvement is actively utilized in both policy and research, moreover, it is widely recognized that stakeholders will increasingly influence future consideration of environmental risk and assessment, including decision-making. However, it is important to ensure the quality of the stakeholder involvement activities in order to support democratically legitimate and robust processes. However, the existing systems of criteria tend to be focused on a rather narrow evaluation of the method applied. We argue that there is need for evaluation that goes beyond a simple assessment of the methodology and addresses the wider context that the stakeholder involvement activity is held in. This paper uses two stakeholder engagement events: one from the field of emergency preparedness and one from nanomedia, to explore the applicability of existing evaluation criteria for a quality assessment of stakeholder or broader public involvement. We use criteria developed by Rowe and Frewer (2000), namely, representativeness, independence, influence, transparency and early involvement criteria tend to be focused on a rather narrow evaluation of the method applied. We argue that there is need for evaluation that goes beyond a simple assessment of the methodology and addresses the wider context that the stakeholder involvement activity is held in.

TU385 Assessment and Management of Radiation Risks following a Nuclear Accident: The Shamsien Project Recommendations
D. Oughton, Norwegian Public Roads Administration / Norwegian University of Life Sciences / Centre for Environmental Radioactivity (CERAD CoE); E. Cardis, ISIGlobal; T. Schneider, CEPEP; Y. Tomkiv, Norwegian University of Life Sciences / Centre for Environmental Radioactivity CERAD CoE

The Fukushima Daiichi accident in 2011 represents a poignant reminder of the complex interplay between environment, society and economics. Contamination of both terrestrial and marine ecosystems had wide reaching impacts for the affected populations. While the strict control of foodstuffs ensured that the radiological impacts on human health were minimal, the economic and societal consequences have been enormous. The loss of livelihood from bans on fishing and farming have hit farming and fishing communities, exacerbating the already existing concern for recruitment of younger generations to family businesses. The return of evacuees to their former homes has been low, particularly for families with young children, leading to demographic changes in societies. Other social and cultural impacts arise from lack of access to beaches, places of heritage and festivals. The economic consequences from food bans go beyond the loss of sales, market value decreased in all products from the area due to loss in consumer trust (20% decrease compared to the rest of Japan). Strategies for radiation risk management are often at odds with the actual needs of the affected populations, and if not carried out properly can cause more harm than good. Recognising this, the EU SHAMISEN project has published a set of recommendations to improve radiation risk management after a nuclear accident. Experience suggested that existing recommendations had a technical focus, with less attention paid to social, ethical, psychological issues and that the information tended to be directed towards the decisions made by experts rather than for support of affected populations. This paper presents the main conclusions and recommendations of the SHAMISEN project. The 28 recommendations propose a management strategy that targets the overall well-being of populations, that addresses not only radiation effects, but also aims to alleviate psychosocial impacts and strengthen stakeholder engagement. In

TU386 SETAC Ecosystem Services Interest Group
S.E. Apitz, SEA Environmental Decisions Ltd

Air Pollution, Biomonitoring and Human Health (P)

TU387 Assessment of Indoor Radon Concentration and Trace Metals Composition in University Building Microenvironments
M. Mohamed Ali, Universiti Kebangsaan Malaysia; M. Mohd Hanafiah, Universiti Kebangsaan Malaysia / Environmental Science; M. Khan, Universiti Kebangsaan Malaysia / Centre for Tropical Climate Change System

This study measures the indoor particulate matter (PM10) composition 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 PM10 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 DoseMan 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 indoor stations. The results showed PM10 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 Building 1 and 2, the radon concentrations were recorded as 2.33 and 1.74 Bq m⁻³, respectively. The average equilibrium equivalent radon concentration measured in Building 1 and Building 2 was 2.33 ± 0.09 and 3.17 ± 1.74 Bq m⁻³, 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 research aimed to examine the public health impact from household fine particulate matter (PM2.5). The sampling was conducted in Kheri, India, a town with extremely high levels of PM2.5. While higher PM2.5 levels are connected to increased lung health outcomes, our study took a step further by sampling at a microscale level from both personal and home environments. This approach allows for a more comprehensive understanding of exposure pathways and the associated health impacts. In this study, paired samples of PM2.5 were collected from both the home and personal environments of participants, allowing for a comparison of exposure levels. The analysis focused on chemical constituents and biological markers to assess the potential health impacts. The results showed significant differences in the composition of PM2.5 between personal and home environments, indicating varying exposure levels and potential health effects. The study underscores the importance of considering both personal and home environments for a complete evaluation of exposure and health outcomes. Overall, this research provides valuable insights into the health implications of PM2.5 exposure, highlighting the need for targeted public health interventions and further studies to better understand the associated health risks.
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 the present study. On it, we collected two fractions of fine PM (PM$_{2.5}$,10 and PM$_{2.5}$,20) 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 released 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. Differences were 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 school managers and parents.

TU390 Acute Impacts of Extreme Hot Temperature Exposure on Emergency Room Visits
Y. Lan, C. Chang, C. Chung, China Medical University
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 hot temperatures. Increases (RR 1.02, 95% CI 1.01–1.04) in 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
Li J, Linjun University J. Zhou, C. Wu, L. Bao, L. Shi, E.Y. Zeng, Jinan University / School of Environment
Abstract Priming 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 substances (brominated diphenyl ethers and the up- to-3,4,7,8-tetra-bromodiphenyl ether, RL–9.14 × 10$^{-4}$ g m$^{-2}$) 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 $\Sigma$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 vaporization and mechanical fractionation, respectively. The difference between the size distributions of particulate PBDEs in emission sources and adjacent air implicated a noteworthy redisposition process during atmospheric dispersal.

TU392 How risky is the schoolyard? An approach from chemical composition of particulate matter
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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. To understand the current level of cement dust exposure among workers in cement factories, a pilot study of 60 workers in three cement factories was conducted. The level 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=0.001, P=0.001 and 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=0.0001, P=0.0004) 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=0.0003), glutathione peroxidase (P=0.0256) and catalase (P=0.0013) respectively. There was a slight non-significant positive correlation between vitamin C, vitamin E, catalase, glutathione peroxidase and Vitamin C (r=0.526, r=0.315, r=0.04598 and r=0.2018 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 (P=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.

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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.

Because 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秸秆 and pig waste. In the EU, Available Technologies Reference document for the Intensive Rearing of Poultry and Pigs. Different strategies can be applied to determine the effectiveness of mitigation options: the SHERPA model or other approaches like Life Cycle Assessment can indicate the environmental benefits achievable with the different scenarios analyzed. Although techniques may be implemented and managed separately, they produce synergistic effect on the farm’s environment. The software is integrated into the agri-food chain 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 NH3 and their derivatives, nitro- and oxo-NH3. This research was supported by project PAGR P503 16-115378.

TU396

Development of an In Vitro Method to Evaluate the Inhalation Bioaccessibility of Particle-Bound Hydrophobic Organic Chemicals and its Effects of Particle Size

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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 hydrophobic organic contaminants using lung fluids, i.e., artificial lysosomal fluid (ALF) and Gamble’s solution amended by dipalmitoyl-sn-glycero-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 increase with particle size, but decrease with the increasing hydrophobicity. It is noteworthy that via the human lung in the in vitro method, the particle-bound PAHs 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

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Air pollution remains to play a significant role in society even in regions worldwide. It 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 impact of the fractionation of the investigated pollutants, background, coarse particulate phase, and six PM10 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 inhalloweable fine and ultrashine 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-polyenic aromatic hydrocarbons (oxy-PAHs). This study confirms that certain molecular mechanisms of toxicity vary in urban regions worldwide. This research was supported by project GACR P503 16-115378.

TU398

Human health assessment of air pollution exposure to tuberculosis risk in regions of Taiwan

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BACKGROUND: Ambient air pollutants from anthropogenic sources play critical roles in the prevalence of tuberculosis (TB). Association between ambient air pollution and TB disease burdens should be clearly elucidated. Several studies found that air pollutants were highly likely to pose adverse effects on antimycobacterial immunity mechanisms, increasing risk of TB development. OBJECTIVE: The main objective of this study was to assess the contribution risks to TB incidences resulted from ambient air pollutant exposure. METHODS: A population-based probabilistic risk assessment framework was incorporated with air pollution concentration database and epidemiologic dose-response data to assess contribution risks of air pollutant-associated TB incidences in Taiwan regions. The contribution concept was quantified by using the population-attributable fraction (PAF). The air pollution-PAF relationships were assessed by employing a three-parameter Hill model based on hazard ratio data of TB exposed to air pollutants. The contribution of air pollution exposure to TB was evaluated by applying multiplications of PAF likelihood and TB incidence rates. RESULTS: Both under severe and moderate scenarios, the results revealed that the most likely air pollutants significantly contributing to TB incidences were carbon monoxide (CO) and nitrogen dioxide (NO2) in regions of Taiwan. Additionally, the particulate matter (PM10) 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 NO3) 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 assessment; Air pollution; Tuberculosis; Population attributable fraction; Probabilistic risk assessment
TU309
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 policy at national, regional 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 contributes 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 pads currently used on the market, replacing the frictional bond 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-tumor bronchial epithelium BEAS-2B cells as model of the respiratory tract. Effects on cell proliferation and cytotoxicity were assessed daily by 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 eco-toxicological 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
A. Kramer, Oregon State University / Environmental and Molecular Toxicology; S.L. Massey Simonich, C. Roper, Oregon State University / Department of Environmental and Molecular Toxicology; A. Zelenyuk, Pacific Northwest National Laboratory; K. Suski, Pacific Northwest Laboratories Oxidized transformation products (OTP) of polycyclic aromatic hydrocarbons (PAHs) have been known in some cases, been demonstrated to be more toxic than their parent PAHs. This is especially true in the case of secondary organic aerosol (SOA) particles. Recent laboratory studies indicate that SOA particles formed in the presence of gas-phase PAHs contain both parent PAHs and their OTP. In laboratory experiments of α-pinene SOA particles, OTP of phenanthrene, a model three-ring PAH, were observed in varying ratios. Developmental toxicity testing with zebrafish (Danio rerio) will be conducted using embryos (n=32/treatment) that will be dechorionated and placed into 96-well plates containing OTP of phenanthrene in observed ratios at 6 hours post fertilization. Developmental toxicity will be assessed by evaluating morphological changes, embryonic/larval photometer behavior, and mortality at 24 and 120 hours post fertilization. Evaluation of the oxidative potential of the SOA samples as well as individual compounds and observed ratios of compounds is underway using the dichothiolthreitol (DTT) consumption assay. The results from both assays will be discussed.

TU401
Chemical analysis and risk assessment for toxic compounds in PM2.5 in Gwangju, Korea
J. Kim, Gwangju Institute of Science and Technology; S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering Particulate matter (PM), a mixture of solid or liquid matter found in the air, has gained considerable attention as a major air pollutant in recent years. Recently, it is revealed particulate matter, especially PM2.5 (aerodynamic size < 2.5μm) cause numerous diseases such as respiratory, cardiovascular diseases, asthma and so on. The prime criteria for preventing the adverse effects of PM2.5 are based on the mass concentration, but recent research has shown that the chemical composition of PM2.5 can be a more important factor for determining PM2.5 toxicity than mass concentration. Many researchers reported the diverse chemicals in PM2.5 such as inorganic sulfates and nitrates, black carbon, metal, organic compounds, and others. In addition, a few studied detected organochlorine pesticides (OCPs) and polychlorinated biphenyls (PCBs) in PM2.5 of China. However, there is no research on OCPs and PCBs within PM2.5 in Korea. In this study, we developed the rapid analysis method for toxic compounds (PAHs, OCPs and PCBs) in PM2.5 using an accelerated solvent extractor (ASE) and solid-phase microextraction (SPME). The chemical data in Korea was collected from Oct. 2016 to Apr. 2017 (for 21 weeks). We determined the toxic compounds in the collected PM2.5 using the developed method and metal concentration in PM2.5 was also analyzed using microwave extraction. Cr, As and Cd showed high concentration in PM2.5 of all sites and several OCP and PCBs also detected. Lastly, we did the risk assessment on metals, PAHs, OCPs and PCBs in PM2.5 of Korea to determine the risk of OCPs and PCBs among the total risk of PM2.5. Our research is a valuable report on OCPs and PCBs in PM2.5 of Korea and suggests the practical method for screening trace toxicants in PM2.5.

TU402
Source apportionment study of PM10 and PM2.5 using selective wind direction sampling technique in the area of Civitavecchia (Italy)
C. de Giardis, Istituto Superiore di Sanità / Environment and Health; G. Settimo, m. Inglessi, Istituto Superiore di Sanità / Department of Environment and Health; g. marsili, osservatorio ambientale; m. soggiu, Istituto Superiore di Sanità / Department of Environment and Health

The concentrations of airborne particles generated by traditional and PM2.5 main sources located in the Civitavecchia harbour-industrial area (Central Italy), namely a large coal-fired power plant, a natural gas power plant, the harbour area, the vehicular traffic (due to both the local traffic and the highway crossing the area) and small industrial activities located in the town. 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 airflows 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 and explorative 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 allow to highlight that the applied methodology represents a valid support in source apportionment studies.

Keywords: source apportionment, wind selecting sampling device, PM10, PM2.5

TU403
Forecasting global atmospheric visibility based on air quality and meteorological data
H. Xiao, NUEORS, Chinese Academy of Sciences / NUEORS; J. Zhang, L. Tong, H. Yi, M. He, J. Zheng, IUE, Chinese Academy of Sciences

Simultaneous and continuous measurements of visibility, meteorological parameters, including relative humidity, wind direction, and the concentrations of six atmospheric pollutants emitted by local sources and small industrial activities located in the town. 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 airflows 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 and explorative 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 allow to highlight that the applied methodology represents a valid support in source apportionment studies.

Keywords: source apportionment, wind selecting sampling device, PM10, PM2.5
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$_{2.5}$) have caused some serious environmental and public health problems. In this research, we aim at investigating the health impacts associated with the PM$_{2.5}$ 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$_{2.5}$, almost of these results doesn’t include the effects of “secondary” PM$_{2.5}$. This study developed the secondary PM$_{2.5}$ concentrations emitted on every industrial point source for the Emissions Database for Global Atmospheric Research (EDGAR) emission inventory data, Weather Research and Forecasting (WRF) model and CMAQ modeling system, and then estimated the induced mortality data in Asia. We further applied structural path analysis (SPA) to the Asian International Input-Output Table (AIIOIT) to clarify the critical supply chains for the reduction of health impacts in Asia. The result shows that the Japan’s consumption-contributes to primary PM$_{2.5}$ emissions in Asia are estimated at 5.0% and we revealed top ranking supply-chain paths 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.

TU407 Non-targeted screening of DNA adducts as biomarkers for human exposure to PAHs in the environment with liquid chromatography tandem mass spectrometry Y. Feng, C. Yao, Health Canada; W. Foster, McMaster University Humans are constantly exposed to thousands of contaminants in the environment. Polycyclic aromatic hydrocarbons (PAHs) are a group of organic compounds containing two or more aromatic rings. They are released into the environment from both natural and anthropogenic sources such as combustion of organic substances and incomplete burning of coal, oil, gasoline, tobacco products and wood. PAHs are known to be bio-transformed by phase I metabolic enzymes to chromatically reactive intermediates that may bind covalently to DNA to form DNA adducts that interfere with DNA synthesis and transcription, leading to DNA mutations and/or toxicity. Furthermore, binding of electrophilic PAH metabolites to DNA is thought to be a key step in the initiation of cancer. Therefore, measurement of those DNA adducts could be an indicator or biomarker of human exposure to PAHs in the environment and of the dose of the ultimate reactive metabolite. Rapid non-targeted approaches are desired to explore a broader scope of new biomarkers associated with the contaminants in the environment. In this study, we report the non-targeted screening analysis of the full scan data to identify DNA adducts time consuming. In this presentation, we will report a non-targeted screening method for identification of covalent DNA adducts using a combination of neural loss scan and product ion scan in a Q-ToF system. The method was applied to non-targeted screening of DNA adducts in follicular cells from isolated ovarian follicles that were exposed to cigarette smoke condensate (CSC). Four DNA adducts, benzo[a]pyrene-7,8-dihydrodiol-9,10-epoxide-df(BPDE-dG), phenanthrene 1,2-quinone-dG (PheQ-dG), B[a]P-7,8-quinone-dG (BPQ-dG), and 4-aminobiphenyl-dG, were identified in the follicular cells. The results also revealed that two oxidative biomarkers, 8-hydroxy-2-deoxyguanosine (8-OH-dG) and 8-isoprostane (8-IsoP), had strong correlations with the three DNA adducts, BPDE-dG, BPQ-dG, and PheQ-dG, suggesting a strong link between the formation of covalent DNA adducts and DNA damaging oxidative stress. The method has also been successfully applied to investigate the selectivity of chemists to modify the nitrogenous bases on DNA sequence. The results showed that each chemical had a different selectivity when it modified the DNA bases. The method has been demonstrated to be a potential tool to provide screening of unknown DNA adducts as biomarkers of human exposure to the parent contaminants in the environment.
TU409

Microplastic Indoor Air Pollution Using a Simulated Breathing Mannequin - µFT-IR Imaginary Quantities

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Extensive research has been performed on indoor air quality (IAQ) over the last decades. This includes the investigation of microplastics in indoor air as a result of natural breathing. Samples have been taken in actively lived in apartments, as well as determining passive air samplers (PAS) in order 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. In addition, this effect should be carefully considered when evaluating and comparing global monitoring data.

TU411

Determination of Cross Compartment Concentration Gradients of Polycyclic Aromatic Hydrocarbons using PE Passive Samplers

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. Microplastics in indoor air and atmosphere have been used to detect PAHs and other organic contaminants. The aim of the research is to establish a method for measuring PAH concentration gradients at the interface between soil and atmosphere. This illustrates the main objective of the study: The determination of the actual flux direction of PAHs across the soil-atmosphere interface. 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. Microplastics in indoor air and atmosphere have been used to detect PAHs and other organic contaminants. The aim of the research is to establish a method for measuring PAH concentration gradients at the interface between soil and atmosphere. This illustrates the main objective of the study: The determination of the actual flux direction of PAHs across the soil-atmosphere interface.

TU412

Evaluating Computational and Structural Approaches to Predict Transformation Products of Atmospheric Polycyclic Aromatic Hydrocarbons

L. Titarev, J. L. Parkhurst, University of Colorado / Department of Chemistry; D.M. Walden, M. Ogba, Oregon State University / Chemistry; P.H. Cheong, Oregon State University / Department of Chemistry, Oregon State University / Depts of Chemistry and Environmental Molecular Toxicology Polyaromatic hydrocarbons (PAHs) under transformation reactions with atmospheric photochemical oxidants, such as hydroxyl radicals (OH•), nitrogen oxides (NOx), and ozone (O₃). The most common PAH-transformation products (PAH-TPs) are nitro-, oxygenated-, and hydroxylated-PAHs (NPAHs, OPAHs, and HPAHs, respectively) which are considered toxic to human health. This study assessed the structural and computational methods for predicting the transformation products of PAHs using a combination of computational methods. The study evaluated the structural and computational methods for predicting the transformation products of PAHs using a combination of computational methods. The study evaluated the structural and computational methods for predicting the transformation products of PAHs using a combination of computational methods.

SETAC Europe 28th Annual Meeting Abstract Book
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 available rings on 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 indicate 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. Azevedo, Instituto Federal do Rio Grande do Sul; K.S. Miglioranza, University of Wisconsin-Madison; Del Plata/La Edmundo 5680 GC-MS). The following PAHs were analyzed: naphthalene, 2-methyl-naphthalene, 1-methyl-naphthalene, acenaphthene, acenaphthylene, fluoranthene, 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)chrysenes. Results, reported a concentration of TBARS in horse hair could serve as a simple and inexpensive method for monitoring air pollution by oil industry.
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 T. Otitoju, University of Nigeria Nsuka / human nutrition and dietetics; O. Otitoju, federal University Wukari / Department of Biochemistry; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; S. Baiyeri, Federal University OyeEkiti / Agronomy Polyethylene residues are chemical components that are left over as monomers and end products after the thermal degradation of polythene. 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, Propane-1-ol, difluoromine, Hexanoic acid, Amyl nitrite, Toluene, Butenenitriile, 2, 2-Butenal, Thiraine, Nonanoic acid, Ethylendiamined, Furfural, Hydrogen azide, 2-pentene, Formic acid, and acetic 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-mannophetolose, 45% hexanoic acid, 25% propane-1- ethylthio 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 cooking. 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 residues.

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. Vasickova, M. Hvezdova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); P. Kousbova, Central Institute for Supervising and Testing in Agriculture; K. Brandstatter-Scherr, University of Natural Resources and Life Sciences / Institute for Environmental Biotechnology, Department for Agrobiotechnology (IFA-Tulln); Z. Simek, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX Application of pesticides, including conazole fungicides (CFs), is an indispensable part of modern agricultural management, contributing to food security and safety. Conazoles are a class of azole-based fungicides, commonly used to prevent fungal growth on turf grass and agricultural crops. CFs are still widely used despite their non-target ecotoxicity in water, chronic toxicity to manannimals with hepatoxicity, carcinogenicity, reproductive toxicity and endocrine disruption. For example, in the EU classification, epoxiconazole and flusilazole are suspected carcinogens [1]. Presence of such compounds in arable soils represents potential short- or long-term environmental threat with a wide range of possible negative impacts on ecosystem services and health. Hence, risk assessment considering the presence of other co-occurring non-target taxonomic groups of vertebrates, invertebrate and plant species, should be performed. The objective of the present work was to assess the ecological risks of conazole fungicides based on the data from the comprehensive survey of pesticide residues in 75 agricultural topsoil floodplain locations in 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 10% of soils, respectively). Other frequently CFs were epoxiconazole (48% of soils) and tebuconazole (36%), followed by flusilazole (23%), prochloron (21%), propiconazole (13%), cyproconazole (8%) and difenoconazole (7%). Overall, the CFs 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/ec-pesticidesdatabase [2] M. Hvezdova, et al., Sci. Total. Environ., vol. 613–614, pp. 361–370, 2018.
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 by G. DUPOURTE, 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 Universite Bordeaux / EPOC UMR 5805; I. Baldi, University of Bordeaux / ISPED, EPIFENC; F. Macary, Irstea Bordeaux; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; H. Budzinski, University of Bordeaux.

European Union countries have about 12 million agricultural holding, 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, the data of Korean victims who were exposed with CMIT/MIT were used in several apple holding, situated in south-west of France. Dislodgeable foliar residues (DFR) and pesticide residues on equipments or apples (wape sampling) were studied during the different activities of apple growing (treatments, re-entry tasks, harvests). Atmospheric levels of pesticides (outdoor and indoor) were also determined using passive samplers (Polyurethane Foams, PUF) and low-volume samplers. The data were used in this study was mainly focused on two fungicides (captan 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, chloromethylsulfathiazoloxide/methylisothiazoloxide (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 GC/GC-TOF-MS and HPLC-ESI-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.

TU426 Toxicological effects of commercial fungicides on the earthworm Eisenia fetida (Savigny, 1826): laboratory and field investigations T. Campani, I. Caliani, C. Pozzuoli, L. Poggioni, University of Siena / Department of Physical, Earth and Environmental Sciences; S. Casini, University of Siena / Science E. The cells were used as a minimal essential medium supplemented with 10% fetal bovine serum (v/v), penicillin (100 mg/ml), streptomycin (100 mg/ml), amphotericin B (2.5 mg/ml) in a humid environment. with 5% CO2 (v/v), at 37 °C. For the cytotoxicity assays, the cells were seeded in 96-well plates, for enzymatic determinations and protein damage in Petri dishes (7.5×105 cells) and for genotoxicity parameters in Petri dishes. From the MTT assay, the LC50 was determined (29.88 (25.95-34.37) mg/ml). The activity of SOD decreased significantly 40% (p = 0.05) to 25 mg/ml of Iprodione, while no effect on the activity levels of CAT and GST was observed. The content of protein carbonyls increased 30% (p = 0.001) at the highest concentration of Iprodione tested. In addition, it was observed that Iprodione induces triopar and micronuclear divisions at 17.5 and 25 µg/ml and bridges with all concentrations tested. Both the index of division and the index of replication indicate that the cells maintain their proliferation capacity, which allows to study the biomarkers of genotoxicity in this system. These results confirm that Iprodione produces genotoxicity and an alteration in the redox equilibrium at the concentrations tested, which indicates the potential risk of exposure to this xenobiotic.
Dichloron and chlorothalonil are two active ingredients in fungicides commonly used in the United States that readily undergo photolysis in the presence of sunlight. Both compounds have reported half-lives in seawater and freshwater. While the rate of degradation and C12 lifetime of dichloron is improved by the seawater (5.7 hours), the distribution of intermediate products is altered significantly; 2-chloro-1,4-benzoquinone forms at nearly double the concentration in seawater as opposed to freshwater. Chlorothalonil quickly degrades to 2-hydroxychlorothalonil via soil degradation and hydroxochlorothalonil can desorb back into the water column where it can be photochemically degraded. The degradation rate and half-life of hydroxychlorothalonil is very short, but differs significantly between freshwater (32.5 min.) and seawater (301 min.). Both dichloron and chlorothalonil have similar proposed photodegradation pathways, therefore the potential for enhanced phototoxicity due to salinity variation is possible. Dichloron has shown to be phototoxic to inland silversides at concentrations as low as 0.10 mg/L, with >90% mortality at 0.75 mg/L. Adverse sub-lethal impacts have also been observed, such as an upregulation in the CCL28 and PTGS2 genes. The effects of salinity on chemical toxicity may warrant changes to future chemical assessments.

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 for aquatic ecosystems. The use of carbendazim as a fungicide to control fungi on agricultural plants, caused the loss of D. 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 isolocal population of *Daphnia magna* (clone k6) was exposed to an environmentally relevant concentration (5 µg/L) of carbendazim during the lifetime of the generation. The effects of carbendazim on survivability, reproduction, parental length, DNA damage (determined by comet assay), biochemical biomarkers (cholesterinase, 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.

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. Bovall, 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 published 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 anionic pharmaceuticals still performed poorly (r² < 0.5). Sorption coefficients for organic cations were typically within an order of magnitude of experimental values where partition was considered as the sum of sorption to 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/FP7 Innovative Medicines Initiative Joint Undertaking (i3E grant n° 115735) for the financial support.
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 public data available.

**WE004**
The environmental concentration and evaluation of active ingredients in pharmaceuticals in rivers flowing through urban area in Japan  
T. Nishimura, Teikyo Heisei University / Faculty of Pharmaceutical Sciences; T. Suzuki, Y. Kosugi, K. Watanabe, Tokyo Metropolitan Institute of Public Health / Division of Environmental Health; A. Hirose, National Institute of Health Sciences / Division of Environmental Health and Toxicology

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 (51ng/L), valsartan (49ng/L), indapamide (88ng/L), candesartan (11ng/L), losartan (17ng/L) for antihypertensive agent, and sulpiride (546ng/L) for antipsychotic agent, citalopram (445ng/L) for antibiotic agent, ketoprofen (150ng/L) for analgesic antipyreric agent, bezafibrate (200ng/L) for hyperlipidemia treatment drug, crotonatum (845ng/L) for antipruritic agent. Among target ingredients, the detect concentration of active ingredient contains 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 depend 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 concentrations of sucralose 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 for country to consider the dilution rate 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 cloribodic 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 alternative means for optimally and cost-effectively applied API quantification. Predictive models are not without limitations and their assumptions and defaults are, at times, not representative of actual environmental conditions. Here we evaluate the accuracy of simple exposure models used to generate predicted environmental API concentrations (PECs) and their suitability for prioritisation of APIs in the aquatic environment. Water samples (n=60) were collected in triplicate at river basis for six-months both upstream and downstream from four wastewater treatment plants (WWTPs) discharging into four rivers in the UK. Measured environmental concentrations (MEC-values) of 33 APIs were determined by HPLC-MS/MS. PEC-values were determined using pharmaceutical use data from the National Health Service, the fractions of chemical excreted from the body and degraded during sewage treatment, the population equivalents of each WWTP, mean regional per-capita water use and the dilution ratio of treated sewage effluent in receiving rivers. API-specific PEC ranges were compared to complementary MEC ranges observed over the 6-month sampling campaign and PEC:MEC ratios were determined. PEC:MEC ratios were generally low (< 0.5), indicating that predicted API concentrations were lower than measured. Between rivers, PEC:MEC ratios were generally closest to measured values in the lowest flow (smallest) rivers and in stretches near the headwaters indicating that locations with minimal upstream contributions of sewage effluent produced the most accurate PECs. In terms of prioritisation, predicted concentrations successfully identified eight of the ten APIs measured at highest concentrations across both upstream and downstream study locations (metformin, gabapentin, atenolol, desvenlafaxine, fexofenadine, fluticasone propionate and paracetamol). PECs may be used for prioritisation over use in more sensitive applications, such as risk assessment, as PECs were consistently shown to underestimate API concentrations.

**WE006**
The role of the water-sediment simulation test and its outcome in the environmental risk assessment (ERA) of pharmaceuticals  
D. Gildemeister, Umweltbundesamt / German Environment Agency / IV2.2 Pharmaceuticals; S. Schmitz, S. Zahorski, German Environment Agency / UBA / IV Pharmaceuticals; A. Hein, I. Rönnefahrt, German Environment Agency / UBA / IV Section 2.2 Pharmaceuticals

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 into 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 persistency. 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 time frame of 2006-2016. 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 databases on chemical characteristics 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 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  
A. Agaz, IBACON GmbH / Environment Department; L. Carter, University of York / Environment Department; P. Andrews, A. Nellis, SimOmics; S. Owen, AstraZeneca / Safety Health Environment; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; J. Timmis, SimOmics; A. Boxall, University of York / Environment Department

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 models and/or software tools. The aim of our project was to develop an artificial intelligence tool to support the BCF assessment of pharmaceutical ingredients by interpreting European regulatory needs and considering existing guidelines and the wider literature. The system generates transparent and evidence-based compound specific BCF assessment reports and BCF testing strategies if testing is required. In our strategy, the P and T Assessments are conducted before the B assessment because P and T is currently only required to be conducted to categorise the compound as PBT or vPvB. Thus empirical BCF values are not always required as decisions are made according to specific trigger values which are either exceeded by a compound or not. This means that in many cases the use of appropriate BCF prediction models prevents the need for experimentation. If a fish BCF test is required, our tool suggests an experimental design with the ultimate aim of reducing the number of test organisms needed without sacrificing the test validity criteria. The novelty of our system is that it illustrates, in a transparent manner, how the system made its conclusions by incorporation of the argumentation tool ArtooPro. This tool visualises the system’s decision incorporating what regulatory and guideline...
development of a quantitative Adverse Outcome Pathway-informed model to predict the risk posed by mixtures of non-steroidal anti-inflammatory drugs to fish

WE008

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 cytochrome P450 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 approach to the toxicology of diclofenac and other NSAIDs in fish. From a mechanistic perspective, we propose a novel approach to the toxicology of NSAIDs for which we have the ability to measure, the ability to predict toxicity endpoints, and the ability to predict toxic potential from measured concentrations. We propose that this mechanistic approach may provide a useful predictive tool to support the implementation of effective NSAIDs ecopharmacovigilance 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. Oehlmann, Johann Wolfgang Goethe-Universität Frankfurt / Aquatic Ecotoxicology

Long-term exposure of non-target organisms is highly covariant to 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 wastewater effluents as well as drinking water. 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 strength of the model is the ability to express the toxic potential of NSAIDs mixtures, expressed as diclofenac-equivalents. We propose that this mechanistic approach may provide a useful predictive tool to support the implementation of effective NSAIDs ecopharmacovigilance strategies and facilitate the regulatory interpretation of past and future toxicity data.

WE010

Application of newly developed in vitro assay to detect physiological activities of antidepressants in wastewater

M. Ibha, M.O. Ibha, D. Kato, H. ZHANG, Kyoto University

Over recent years, growing numbers of human pharmaceuticals have been detected in wastewater effluents of WWTPs. Concern about their potential impact on development and reproduction of aquatic organisms, and long-term accumulation of pharmaceuticals in 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 wastewater effluents as well as drinking water. 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 strength of the model is the ability to express the toxic potential of NSAIDs mixtures, expressed as diclofenac-equivalents. We propose that this mechanistic approach may provide a useful predictive tool to support the implementation of effective NSAIDs ecopharmacovigilance 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 query and assessment is key to improve current understanding of the ecological risks of pharmaceutical compounds in the non-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 objectives. 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 biological endpoints highlighted the sensitivity of molecular responses, followed by individual level-responses (e.g. behavior and growth), yet variable sensitivity scores among taxa were observed when considering different pharmaceutical classes. Data availability and comparability limited quantitative 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.

WE013 Leveraging Pharmacological Data for Prioritization of the Ecological Risks of Chiral Pharmaceuticals
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A number of monitoring studies have shown that benzoylecgonine (BE), a metabolite of cocaine, is the most illicit drug residue measured in both wastewater and surface waters worldwide. 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. Enantioselective 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 impossible 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, fluoroxenine 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 for predicting stereoselective toxicity of chiral pharmaceuticals.

WE014 Effects of benzoylecgonine exposure at different levels of the biological hierarchy on Daphnia magna
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Recent studies have demonstrated that the type 2 diabetic drug metformin and its only known metabolite, guanylurea, 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 its metabolite guanylurea 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.

The last decades, the number of patients suffering from diabetes type 2 is increasing. Consequently, a rising consumption of antidiabetic drugs as metformin (MF) has become obvious. In wastewater treatment plants, metformin is only partially retained and also transformed to guanylurea (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, 10, 1000 µg/L) and GU (0, 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 European Commission Framework Program.

WE015 Impact of the antibiotic drug metformin and its transformation product guanylurea on brown trout (Salmo trutta f. fario)
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The aim of this study was to investigate the effects of chronic exposure to metformin and its metabolite guanylurea on the type 2 diabetic drug metformin. Metformin has been measured in the ng-µg/L concentration range in both German and North American surface waters and wastewater effluent. As the majority (>90%) of metformin is metabolized into
Bio-Optical probing of Bezafibrate toxicity in model marine diatom Phaeodactylum tricornutum
B. De Felice, F. Hoffmann-La Roche Ltd / Roche Group Safety, Health & Environmental Protection; T. Davidson, Bristol-Myers Squibb / EHS; K. Kappler, Johnson & Johnson; S. Owen, Merck & Company, Inc. / Global Safety & the Environment

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 fibrac acid widely used to reduce plasma triglycerides and raise the level of high-density lipoprotein cholesterol. Specifically, Bezafibrate 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, bezafibrate has been detected in surface and drinking waters as well as in wastewater effluents. This can have serious effects on marine life, including on marine primary producers and thus impacting the whole system productivity and functioning. Exposing the model diatom Phaeodactylum tricornutum to a range of environmentally relevant concentrations of bezafibrate (0–60 mg/L) revealed no serious impacts on cell growth. Nevertheless, after 48h of exposure damages in the photosynthetic apparatus were detected using bio-optical probing. The reduction of TAGs could lead to burnout of the photosystems and thus its chemical energy production (ATP). This may result from a direct effect of bezafibrate in membrane fatty acids from the chloroplast, since both photosystems are anchored in a lipidic membrane system. Moreover triglycerides (TAGs) are known to protect the photosystems against photoinhibition. The reduction of TAGs could lead to burnout of the photosystems due to excessive energy being absorbed, as observed by the high incoming photonic energy flux, this way reducing the number of active reaction centers in the algae and thus its photosynthetic ability. All these bio-optical parameters show a clear dose-effect relationship, indicating that P. tricornutum is a good candidate organism for fibre toxicity testing in marine systems, screened by non-invasive high-throughput bio-optical probing tools.

Environmental Risk Assessment for the Active Pharmaceutical Ingredient Mycofenolic Acid in European Surface Waters
B. De Felice, 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 Syntex, 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 Syntex and on new tests, all performed under GLP quality assurance, for physico-chemical characteristics, partitioning, environmental fate, biodegradability and (sub)chronic/acute toxicity and on sales amounts for the products containing MPA in Europe. A predicted environmental concentration (PEC) in Europe from all products containing 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 assessment factor of 10 to derive the environmental concentration (EPC). 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 assessment factor of 10 to derive the environmental concentration (EPC). Potential risk for surface waters was then quantified 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 assessment factor of 10 to derive the environmental concentration (EPC). Potential risk for surface waters was then quantified.
After use, pharmaceuticals and their residues eventually end up in the sewage system. Sewage treatment plants reduce the nutrient load of waste water, and while organic micropollutant removal occurs concomitantly by bacterial activity and sorption, many contaminants, including pharmaceuticals, are not entirely removed. Consequently, effluent containing pharmaceuticals and their residues is discharged into surface waters. A recent study showed that 29 of 80 monitored pharmaceuticals were regularly detected in Dutch surface water, and that five of these substances, i.e. the pain killer diclofenac, the antibiotics azithromycin, clarithromycin and sulfamethoxazole, and the antiepileptic drug carbamazepine, pose a risk to the aquatic ecosystem (Moermond et al., 2016). This raises concern, perhaps even more so when considering that for many of the around 2000 pharmaceuticals that were authorized for the Dutch market in 2016, it remains unknown to what extent they are present in surface waters, how they behave in the environment, and to what extent they exert toxicity to aquatic species individually and jointly. A class of pharmaceuticals that has received increased attention in the Netherlands, but also in the EU, e.g. PHARMAS project and Cytothre, are cytostatics. These potent substances are used to inhibit cell division in cancer patients, but the fraction released unchanged to surface water could affect aquatic species in a similar manner. This project aimed to provide an overview of the use of cytostatics in the Netherlands and to determine if cytostatics pose a potential risk to the aquatic ecosystem. For this study, an inventory of cytostatics use in the Netherlands was compiled, while available environmental fate and effect data were gathered and the available monitoring data in Dutch surface waters. For these 10 cytostatics, available environmental fate and effect data were gathered and safe environmental concentrations were derived. Comparison to predicted and measured environmental concentrations will allow to conclude if the selected cytostatics pose a potential risk to the aquatic environment.

**WE023 Environmental risk assessment of human pharmaceuticals - what can we learn from regulatory effect data so far?**

S. Schwarz, German Environment Agency UBA / Section IV 2.2

Pharmaceuticals; J. Bachmann, German Environment Agency (UBA) / Section IV 2.2

Environmental Risk Assessment of Pharmaceuticals; U. Brandt, German Environment Agency UBA / Section IV Environmental Risk Assessment of Pharmaceuticals

Since the coming into force of the guideline on the environmental risk assessment of medicinal products for human use (EMEA/CHMP/SWP/44470/00 corr 2), the German Environment Agency (UBA) is tasked with environmental risk assessment of human pharmaceuticals. Applicants seeking approval of medicinal products need to submit fate and effect data, in case predicted environmental concentrations exceed 10 ng/L in surfacewaters, or the substance is of specific concern through its mode of action. For an inventory of marketed cytostatics in the Netherlands, the top 10 of cytostatics was compiled by taking into account the metabolic transformation of cytostatics in patients, the removal efficiency in sewage treatment plants, and the available monitoring data in Dutch surface waters. For these 10 cytostatics, available environmental fate and effect data were gathered and safe environmental concentrations were derived. Comparison to predicted and measured environmental concentrations will allow to conclude if the selected cytostatics pose a potential risk to the aquatic environment.

**WE026 What makes a chemical substance a ‘natural substance’? A case study in the context of the EU veterinary medicines marketing authorisation procedure**

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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, allows to perform an initial risk assessment. However, it may 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, metabolic and transcriptomic analyses**


Exposure to PFOS (perfluorinated octyl sulfonate) 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 48 to 120 hpf. 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 analyses 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 (Oncorhyncus mykiss).**  
G. Figueroa, University of Ontario Institute of Technology / Advanced Environmental Research Institute; Z.P. Pandelides, University of Ontario Institute of Technology; D.A. Holdway, University of Ontario Inst. of Tech / Science

The environmental causes of obesity: Novel human in vitro models of obesogen effects are found in other animal groups beyond mammals. Collectively, we show that some environmental chemicals are able to alter lipid homeostasis, up in the aquatic environment, potentially affecting its ecosystems. In fact, reports of obesogen effects in aquatic settings are lacking, and we only suspect the existence of such chemicals due to our observations in terrestrial systems. In this project, we aimed to fill this gap by establishing a novel human in vitro model of adipose tissue differentiation from trout preadipocytes.  

**WE029**  
**Obesogens in the aquatic environment**  
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The obesogenic potential of the aquatic environment 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 aquatic organisms. Such a potential impact on aquatic organisms may give rise to physiological disorders and disease. Although largely unexplored from a biological perspective, the key molecular components implicated in lipid homeostasis have likely appeared early in animal evolution. Therefore, it is not surprising that the obesogenic effects are found in other animal groups beyond mammals. Collectively, data indicates that suspected obesogens impact lipid metabolism across phyla that have diverged over 600 million years ago. Here we identify the knowledge gaps in this field and we set future research priorities.

**WE030**  
**The Environmental Causes of Obesity: Novel human in vitro models of adipocyte differentiation for studying the effects of chemical exposure**  
S. Ermler, Brunel University London / Institute of Environmental, Health and Societies; B. Blumberg, University of California, Irvine / Development of Developmental and Cell Biology; J. Leegel, Utrecht University / Institute for Environmental Studies; S. Jobling, Brunel University / Institute of Environment, Health and Societies.

Obesity has become a worldwide challenge, with obesity rates not only increasing in adults, but also in children. Obesity is caused by an imbalance between caloric intake and energy expenditure. However, increased caloric intake due to changes in diet and lack of physical activity cannot solely explain the observed rise in obesity. Other factors, such as genetics or environmental stressors, also play a role. Exposure to endocrine disrupting chemicals (EDCs), which act as so-called obesogens during development, may impact on adipogenesis and susceptibility to obesity, and several such compounds have been found to stimulate adipocyte differentiation in vitro and in vivo. Recent systematic reviews in our group have shown that prenatal exposure to EDCs such as BPA and DEHP is related to increases in adiposity later in life in rodent models. To identify potential obesogens and understand the effects of EDCs, such as endocrine disruption, is required. Standard assay systems to screen for obesogens in vitro are either using reporter gene assays e.g. for activation of the peroxisome proliferator-activated receptor gamma (PPARgamma), a key regulator of adipogenesis; or differentiation assays using established cell lines such as 3T3-L1 preadipocytes. However, whereas these assay systems are good screening tools, they only assess effects of obesogens on specific receptor activation or differentiation of already committed, white mature adipocytes. Our research aims at unravelling the effects of obesogenic EDCs on human adipocyte development. The most critical effects of exposure to obesogens are elicited in utero and in early life. Therefore, in vitro models that mimic the earliest stage of adipogenesis are best suited to investigate how EDCs can disrupt normal cell differentiation during development. One promising cell culture model involves the use of murine preadipocyte lines as enriched sources of macrophagic/mesenchymal stem cells (MSCs) to mimic bone mesenchyme and brown adipocytes.

**WE031**  
**Comparing metabolic responses in Oryzias latipes to environmentally relevant concentrations of metformin and its metabolite, guanylurea**  
E. Ussery, University of Ontario Institute of Technology / Biological Sciences; K. Bridges, B.J. Venables, University of North Texas / Advanced Environmental Research Institute; Z.P. Pandelides, University of Ontario Institute of Technology; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; A. Kirkwood, University of Ontario Institute of Technology; D.A. Holdway, University of Ontario Inst. of Tech / Science

In recent years, the occurrence and fate of pharmaceuticals in the aquatic environment has become an increasing concern. Metformin, a commonly used antidiabetic drug, is also present 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 metabolite on aquatic organisms, we carried out a study to investigate whether metformin could be taken up in both embryo and larval medaka after exposure windows ranging from 24 hours to 7 days. We also conducted a metabolomics 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.

**WE032**  
**Levels of proteins, carbohydrates, lipids and cholesterol in the digestive gland of juvenile catarina clam Arcogpecten ventricosus (Sowerby, 1842), exposed to toxic metals**  
A. Sebrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Instituto de Biologia; C. Cáceres, Universidad Autónoma del Estado de México / Universidad de 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 lipids, 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)

EW033 Environmental assessment of foaming agent persistence in conditioned soil for EPB-TBM tunnelling


The Polyfoamer ECO line of products, 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 products (P1, P2). For this purpose, a set of microcosms was set up using two different soils (S1: silty-clay; S2: gravel in a clay-silty-sand matrix soils) 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 un-treated 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 DT_{50} 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.

EW034 Application of the Vibrio fischeri acute toxicity test to assess the environmental impact of spoil materials containing foaming agents


The rapid development of TBM’s 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 TBM’s relies on the use of appropriate soil stabilizing agents, containing 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 in European 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 (for 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 performed to evaluate the potential ecological impact of spoil materials the bacterium Vibrio fischeri should 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 pathway 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.
geological formation with alternance of limestone and marlstone) and the material called “S” (a cohesive clay). The amount of the surfactants inside the conditioned samples “S” and “M” have been measured with the instrument HPLC-MS (“High-Performance Liquid Chromatography Mass”) at different stages from the addition of the foam to the soil: at time 0, at 3 and at 7 days. Toxicity tests with the bio-luminescent bacteria Vibrio Fisherii (ISO 13148:2.2:2007) and the fish embryo Diploidophrys novacula have been carried out and have shown that the environmental formulations with the two new foaming agents Polyfoamer ECO confirm that these new formulations allow to reduce the impact on the soil and therefore to facilitate its re-use in short periods as a by-products. The main results obtained with the Polyfoamer ECO/100 and Polyfoamer ECO/100 Plus are: lower toxicity and lower content of organic material when compared to traditional foaming agents, faster degradation of the surfactants inside the conditioned soil, low toxicity of the conditioned soils and tendency of toxicity decrease along the time. Values comparable to the natural soil toxicity are achieved in a short period.

**WE037**

**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 for urban planning, including a huge development of the mechanized tunnelling industry by the use of Tunnel Boring Machines (TBMs). Polyfoamers of TBM are utilized in the construction 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 sulphonate (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.

**WE038**

**Distribution and persistence of anionic surfactants in leachate and conditioned soil: mesocosm study for EPB**


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 in particular sodium lauryl ether sulphate (SLES) are the main 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 two 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 (DT₀) of SLES in two soils (S1: silty-clay soil; S2: gravel in a clay-silty-sand 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, 527,200 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.

**WE039**

**Preliminary environmental risk assessment of sodium lauryl ether sulphonate contained in foaming agents used in mechanized tunnelling**

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Anionic surfactants (ANS) are a heterogeneous group of amphiphatic compounds characterized by linear aliphatic chains (ranging from C8 to C18) with a polar group (sulphate or sulfonate) neutralized with a counter ion. Given the variability of their molecular composition ANS are considered mixtures. They are utilized in several applications (i.e. detergents, cleaning products, fracking or soil conditioning in the excavation industry). Among ANS, the sodium lauryl ether sulphate (SLES) is commonly utilized as a foaming agent to facilitate the excavation procedures in mechanized tunnelling. However, its use raises concern for the environment considering the presence of SLES residues in soil debris produced during the excavation. In addition, the absence of soil threshold limit for SLES in the EU legislation does not facilitate the re-use of soil debris as by products (e.g. land covering) and, consequently, a huge amount of such detritus can be discharged as a waste with high economic costs. In absence of a threshold limit, performing an environmental risk assessment (ERA) of foaming agents containing SLES can be a possible alternative. However, the ERA is hampered by both the rather scarce data on the effects of SLES and the site specific condition of use which lead to different levels of exposure. Indeed, the selection of the type and quantity of foaming agents depends on soil, geological conditions, and characteristics of the tunnel boring machines. Furthermore, several commercial formulations are available on the market with different percentages of SLES and several other components. This study is part of a wider project aiming to develop a methodology to be applied to identify environmental acceptable levels of SLES residues in soil debris produced during the tunnelling operations in Italy. Particularly, we report the results regarding the preliminary ERA which has been used to select, among all the available commercial formulations, the one leading the lowest level of risk for the environment in a specific condition of use. The risk has been characterized based on PEC/PNEC ratios. PECs were calculated by predictive models and considering the percentage of SLES in the commercial formulations as well as the required treatment ratios for tunnelling operations. PNECs (soil and surface water) for SLES were derived from ecotoxicological data (terrestrial and aquatic organisms) which were obtained from laboratory tests on several test organisms.

**WE040**

**Ecotoxicological assessment of spoil material produced in mechanized tunnelling**


Mechanized excavations using Tunnel Boring Machines (TBM) has consolidated in recent years. In order to facilitate the overall process, specific foaming agents and polymers are added to the soil. The main component of many commercial foaming
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, two soils were prepared, containing two soils with different geopedological characteristics, conditioned with two foaming agents at the same treatment ratios (TR, L/m²) used for mechanized drills. 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 elutriate 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. Commonly accepted standards have been developed to measure the level of pollution through expeditious tests directly on site. A joint 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.

**WE041**

**Expeditious test for on-site monitoring activity in mechanized tunneling applications**


In the vast majority of tunnel projects performed with TBM 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 industrial byproducts conditioning the process. To this purpose the planned strategy of spoils disposal management in a virtuous cycle of reuse of the resources leads to relevant economical 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 evaluate the effect of specific concentration values of these chemicals on terrestrial and aquatic environments, as well as to measure the reduction of the concentration of these compounds in each environment due to the action of the microorganisms inhabiting them. All these experimental procedures must be carried out in specialized laboratories equipped with sophisticated apparatuses, in which controlled environments are predisposed, so that at present it’s not possible 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 tunneling. The results of preliminary laboratory tests convinced that the expeditious 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 this test seems to be properly suited for monitoring large volumes of spoil 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 tunneling: effects on daphnids, algae and cress.**

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Surfactants and polymers are used in mechanized tunneling 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, polycrylates 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 effect deriving 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 analyses 8 surfactants and 4 commercial polymers, using a multidisciplinary approach to determine their reference thresholds for both water and soils, and test with the worm Eisenia fetida. In parallel, sub-samples of soil and elutriate 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. Commonly accepted standards have been developed to measure the level of pollution through expeditious tests directly on site. A joint 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.

**PBt/pVb & Pmt/PvM substances and Non-extractable residues (NER): Scientific strategies, Analytical challenges and Regulatory Issues (P)**

**WE043**

**Bioaccumulation, tissue distribution, and trophic magnification of organic ultraviolet absorbents 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-ethylhexyl-4-trimethoxycinnamate were detected in eggs than in muscle, probably indicating maternal transfer of the compound. The calculated bioaccumulation factors (LogBAF)were usually > 3.3, suggesting potential of biomagnification 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 benzotriazolone 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 considerably large number of fish. 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 fish through bioaccumulation studies with Hyalella are similar to those obtained from fish (Schlechtien, 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
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, post-screening using statistical models for BCF prediction is becoming increasingly popular. While dietary bioaccumulation of neutral chemicals has been successfully associated to lipophilicity 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 5 bases. For each fish species, the predominant KOW was determined at pH 3 to 9. A parallel dataset was compiled with bioconcentration factors (BCFs) of the same chemicals derived in water exposure studies with fish (OECD 305-1 guideline). Bivariant correlation analysis (Pearson and Spearman) revealed that a logKOW was not a sufficient predictor of BMF, although with significant positive correlation (R=0.40), and b) that significant correlation was shown only with logKOW at 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., logKOW) 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 (Morgan equation and lipophilic amino acid). 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 radio-labelled test chemicals.

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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 partition 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 methods to predict the BCF values and compared those outcomes to the results from a BCF test in order to validate this alternative approach. Several fragrances from the tetranorlabdane diterpenoids family, either composed of a single or a mixture of 5,41,4,41- tetranorlabdane diterpenoids family, either composed of a single or a mixture of isomers were tested. The logKOW predicted by QSAR ranges from 4.75-5.41 and from 4.2 to 5.2 when determined by HPLC (OEC 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 (≥20), 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 the in vitro 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 nonbioaccumulative, 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.

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The 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 the food web, for example via aquatic or terrestrial food chains. This approach provides the framework for a new TMF testing scheme integrated in the German Environmental Specimen Bank (ESB) including cryo-milling, homogenization, sub-sampling, and long-term storage. During all processing steps samples will be kept constantly at a temperature < -150°C. The sample material obtained will be analyzed to derive TMF estimates for different constituents in a food web relevant for a concrete chemical. Different sources of information will be used to answer different questions in regulatory and monitoring affairs. The TMF can be used in the evaluation of the biomagnification 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 "good ecological status". So far, TMF data are only available for aquatic fish species but species specific TMF data have not 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 proof field study. A water body will be selected under consideration of several aspects such as 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 stable isotope patterns of amino acids. Sample handling will follow the protocols applied by the German Environmental Specimen Bank (ESB) including cryo-milling, homogenization, sub-sampling, and long-term storage. During all processing steps samples will be kept constantly at a temperature ≤ -150°C. The sample material obtained will be analyzed to derive TMF estimates for different constituents in a food web relevant for a concrete chemical. Different sources of information will be used to answer different questions in regulatory and monitoring affairs. The validated concept may provide the framework for a new TMF testing scheme integrated in the German Environmental Specimen Bank (ESB).

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consequences on the PBT/vPvB-identification.

**WE049**

**PBT/vPvBs: All equally bad or some worse than others? - How to inform risk management**

K. Thele, WUR; S. Gabbert, Wageningen University / Social Sciences

In the context of 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 different 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 therefore be helpful in guiding 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, industry and consumers. 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 (POMCs). We modified the Multimedia Urban Model (MUM) of Diamond and co-workers by using polyelectrolyte parameter 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**

A. Carras, Kau USA / R&D; T. Suzuki, Kao Corporation, S.A. / Product Safety & Regulations; I. Davies, Personal Care Products Council / Science; J. Coleman Il, Kao Corporation

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. However, the use of new and novel polymers in cosmetics is increasing. Nonetheless, there is a suspicion 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 database. 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, chemical molecule, 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 process 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 less clear how to adapt it for REACH substances described in different regulatory frameworks and therefore a definitive PBT/vPvB guidance for pharmaceuticals was established 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 if it is not a PBT. Furthermore, there is currently no detailed guidance 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/vPvB assessment should be performed for pharmaceutical products and the consequences for products which fulfil the PBT/vPvB criteria. This presentation will describe our experiences and the challenges we have encountered in performing PBT/vPvB 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/vPvB assessment for pharmaceuticals.

WE054 Evaluation of new assessment methods and enhancement of PBT/vPvB 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 to assign toxicological consequences. The objective of the project is to refine the P assessment of ionised 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 substances we have encountered in performing PBT/vPvB 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/vPvB assessment for pharmaceuticals.

WE055 Assessment of the persistence of ionic or ionisable organic chemicals under REACH
D. Claassen, RWTH Aachen University / Institute for Environmental Research; J. Ackermann, Federal Environment Agency Umweltbundesamt / Chemicals; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics
For the protection 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 biotical degradation. In order to improve the evaluation of the persistence of ionic and ionisable substances in the PBT-assessment, sorption and degradation patterns of charged substances are modelled by carrying either a positive, negative or non-charged functional group will be investigated. The sorption behaviour of 14C-labelled 4-n-Dodecylphenol, 4-n-Dodecylbenzenesulfsalicylic acid sodium salt and 4-n-Dodecylbenzytrimethylammonium 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 behaviour 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: ESIR investigations with nitroxide spin label
A. Riche, E. Bondarenko, H. Steinhoff, University of Osnabrueck / Physics; G. Ür, K. Hideg, T. Kälai, University of Pécs / Organic and Medicinal Chemistry; M. Mathies, University of Osnabrueck / Institute of Environmental Research
The restriction of chemical substances in the REACH was developed to assess neutral organic compounds. For this purpose, simulation tests according to the currently valid concept under the REACH regulation does not provide a sufficient basis to assign toxicological consequences. The objective of the project is to refine the P assessment of ionised 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 substances we have encountered in performing PBT/vPvB 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/vPvB 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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Non-extractable residues (NER) are defined differently in various regulatory frameworks. In some cases NER are integrated in the calculation of predicted environmental concentrations (PEC) or considered. However, distinguishing between these types of NER presents a challenge for the authorisation decision. The significance of NER in the assessment of persistence (e.g. PBT, vB/vP, 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 show that NER cannot 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 reutilised 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 evidence and research fund 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 assessment of NER. The irreversibly bound fractions have to be considered in assessment of the persistence. An UBA evaluation of regulatory data demonstrates 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 references for ion sorption phases and compared their sorption properties against those of various environmental and biological materials. Retention times on commercial ion exchange chromatography columns were measured in full aqueous eluent and were converted to retention factors (k'), which are proportional to the ion-exchanger-water partition coefficients. In the end, we established a data set for retention factors of 61 cations on a strong cation exchange column (SCX), 24 cations on a weak cation exchange column (WCX), and 66 anions on a weak anion exchange column (WAX) measured in consistent experimental conditions (i.e., pH, T, co-existing ions, injected amount). The obtained retention factors were compared to soil organic carbon (Koc), clay minerals-water (KCM/w), bovine serum albumin-water (KBSA/w), and muscle protein-water partition coefficients (KMP/w) from the literature. Relatively good correlations (R2 = 0.5-0.6) were found for some cases such as log Koc, log KMP/w, and log KBSA/w against log k' for WAX. For comparison, similar correlation analyses were performed using experimental and predicted log Kow instead of log k'. In most cases, the correlation with log Kow were lower than the correlation with log k'. Notably, log k' has a clearly larger applicability domain than log Kow, because log Kow is unavailable for ionic chemicals derived from strong acids/bases (e.g., sulfonates, quaternary ammoniums), whereas log k' can be measured for such ions too. This study offers a step forward to the development of accurate prediction models for sorption coefficients of ionic chemicals in the environment.

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 13C3-15N-glyphosate in an Oxic Sediment 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 saccharine-pathway with complete mineralization, and the incomplete pathway with AAU, non-stable bound fraction. The input parameters were 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 not living, non-amino acids), which is equal to the biogenic non-extractable residues (NER). Biogenic NER consist of assimilated 13C/N and are thus considered to be


WE060
Determination of persistent organic pollutants (POPs) in soil from sites adjacent to landfills: different provinces of the Republic of Armenia A. Akhopian, Hayazarian, Hazardous Substances & Waste Policy Division / Head of Division; Y. Buniyayyan, Environmental Monitoring and Information Center / Division of waste inventory, classification and technology investigation; V. Khachatryan, National Institute of Oncology / Thoracic Surgery Department; F. Petrosyan, UNIDO BAT/BEP Project(Armenia)
Sources of environmental pollution by persistent organic pollutants (POPs), either used or not curiously applied 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), Aragatsotn (Aragatsotn Marz). Soil samples taken from this marzes were analyzed for determination of the following POPs: − Hexachlorocyclohexane (HCH) isomers: α-, β-, γ-, and HCH; − DDT isomers: 2,4'-DDT, 4,4'-DDT; − DDT metabolites: 2,4'-DDE, 4,4'-DDE, 2,4'-DDD, 4,4'-DDD; − Hexachlorobenzene, - Heptachlor, - Aldrin - Dieldrin - Heptachlor epoxide A and Heptachlor epoxide B, - Endosulfan I and Endosulfan 2, - Endrin, - Mirex - 14 Dioxin-like polychlorinated biphenyls: congeners No. 77, 81, 105, 114, 123, 126, 156, 157, 167, 169, 180, 180, 189. Quantification of POPs was done using chromatograph with electron capture detectors (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 DDT 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 (summarized concentrations) as obvious indicator of soil pollution by the studied POPs. In the investigated soil samples HCH isomers, DDT isomers and metabolites, certain Dioxins-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 no environmental concern or as bioavailable and non-degraded residues. This may be changed if clear indications for ultimate degradation or irreversible immobilisation are available. Several research are available, such as, as well as the total amount of NER of chemicals in environmental matrices can be experimentally discriminated, sequestered (strongly sorbed and entrapped) residues (type I), containing the parent compound or transformation products or both and having the potential of release. Type II NER are residues that are covalently bound to organic matter in soils or sediments or to biological tissue in organisms. Such residues are considered very stable bound, with very low immobilisation rates and like that of humic matter degradation. However, providing the proof for type II NER is a critical issue in NER assessment. Harsh extraction conditions may release both types of NER but for type II this will rarely happen under physiological conditions. Type III NER comprises biogenic NER (bioNER) after complete metabolization of the xenobiotic and anaerobic formation of natural compounds like amino acids etc. The formation potential of bioNER can be predicted by using the theoretical microbial yield, which can be estimated using the Microbial Turnover to Biomass (MTB) method. In addition the amount of bioNER can be experimentally quantified by labelling with stable or radioactive isotopes. bioNER are of no environmental concern. Type
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 mobilizable 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 mobilizable.

**WE062** Photodegradation of Atrazine in the Presence of Indole-3-acetic Acid and Natural Montmorillonite Clay Minerals

C. Gao, Nanjing University / School of the Environment; L. Zhang, Nanjing University

In this study, a new natural degradation pathway of atrazine and the potential mechanism are proposed. Atrazine was oxidized under simulated solar irradiation by indole-3-acetic acid at the environmentally relevant concentration under aerobic condition. The reaction was initiated by the production of hydrated electrons generated from the photoreduction of indole-3-acetic acid, and then this species transformed into hydroxyl radical after a series of radical reactions with proton and dissolved oxygen. During this process, the presence of montmorillonite greatly enhanced the yield of hydrated electron and promoted the further degradation of atrazine by hydroxyl radical. The novel reaction is to some extent affected by pH and the type of exchangeable cation present on montmorillonite. Based on our results, the knowledge to extrapulate from laboratory study to the complex environment. As the first step of our undertaking to develop methodologies to extrapolate laboratory data to the aquatic environment, we chose a fragrance ingredient, Myrrhone, as an example, and used laboratory study results to calculate its photodegradation half-lives at depths in natural waters. Direct photodegradation was revealed to be the dominant photodegradation process of Myrrhone and the quantum yield was determined to be 0.51 in the laboratory. Four main volatile photodegradation products, which continued to photodegrade, were observed, isolated by preparative multidimensional GC and identified using NMR. Direct photodegradation half-lives of Myrrhone in natural waters at different depths were calculated based on its molar absorption spectrum, quantum yield, and the irradiance at depth of natural waters. The irradiance values at depth were obtained by applying diffuse attenuation coefficients (Kd) to the irradiance at the surface of water, generated using a solar irradiance calculator as a function of time, date and location. Kd are empirical values determined by the interaction of a number of factors, including absorbance by dissolved organic matter and particulate matter, scattering, and the angular distribution of the light field. A correction factor for cloud cover was also applied. In this step of the study, which gives the half-lives that ranged from 1.3 to 9.1 days for small size lakes and 6.3 to 45 days for large size lakes under realistic conditions in Australia. The calculation of half-lives at the surface of water was validated by two outdoor photolysis experiments. The calculated half-lives of 0.38 h and 1.14 h were in agreement with the measured half-lives of 0.40 h and 1.15 h, respectively. This agreement indicates that mathematical models can be developed to define complex environmental conditions for the extrapolation of laboratory data to the environment. The next step is to design experiments to measure half-lives at depth in natural water to refine and validate the calculation of half-lives. Thoroughly validated models can be valuable tools for the persistence assessment of chemicals based on photodegradation information.

**WE064** The Photolytic Fate of Fungicides

J. Apell, MIT / Civil & Environmental Engineering; K. McNeill, ETH Zurich

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’ photolysis-based environmental fate assessment is to identify photolytic degradation pathways in the environment. For example, only direct photolysis 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 radical, 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 commercially used fungicides investigated, the results show that both direct and indirect photodegradation are important to quantify in order to accurately characterize the environmental fate of fungicides.

**WE065** Study Design Considerations for E-Fate Testing of UVCB Substances

C. Lowrie, Charles River / Environmental Fate and Metabolism

Substances of unknown or variable composition, complex reaction products or biocides. In this case UVCD data is used for the exemplar which can not be identified 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 goal is to characterize the environmental 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 UVCD 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 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 UVCD data is used 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 UVCD substance was persistent and further testing implemented accordingly.

**WE066** In silico investigation of the triplet-sensitised phototransformation of phenoins induced by chromophoric dissolved organic matter

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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 and indirect phototransformation pathways. Direct phototransformation involves molecules that absorb sunlight and are transformed as a consequence. Indirect phototransformation involves reactive transients such as ·OH, CO₂, ·O₂, and the triplet states of chromophoric dissolved organic matter (CDOM*). They are generated by irradiation of photosensitizers such as CDOM (producing ·CDOM*, ·O₂ and ·OH), nitrate and nitrite (producing ·OH). Among these transient species, ·CDOM* is certainly the most abundant one in natural waters (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 phototransformation of 49 xenobiotics with standard organic matter (OM) (which is used as CDOM) proxies (1-nitronaphthalene (1-NN), riboflavin (Rf), 4-carboxybenzophenone (4CBP), and antraquinone-2-sulphonate (AQS)) have been used to derive Quantitative Structure-Activity Relationships on the basis of theoretical molecular
In this paper solubilization of persistent organic pollutants, PAHs, PCBs, pesticides and emerging pollutants, as PBDEs or PCN, was investigated in water samples, using some anionic, cationic and non-ionic surfactants. These pollutants are well known for their considerable toxicity, persistence and bioaccumulation toward both human health and environment in addition to their low aqueous solubility. However, the use of surfactants to water solution enhances solubilization of hydrophobic organic compounds. Above the critical micelle concentration (CMC), surfactants exist as aggregates in solution and hydrophobic compounds move to hydrophobic micellar core region. This technique is considered as a groundbreaking method to traditional liquid-liquid extraction with hexane. The physical and chemical properties as size, shape, ionic strength and hydrophobicity are important to identify the appropriate surfactant depending on the type of compound to be removed. The method consists in two steps: a) removal of analytes from aqueous solution by the micelles; and b) recovery of analytes from the organic solvent followed by micellar deformation with addition of NaCl. Both steps are effective, easy and with high recovery of pollutants. Furthermore, the samples are already in the solvent for quantitative analysis. The removal of analytes from aqueous solution was verified by comparing UV-Vis spectra in the range 240-360 nm before and after extraction in isoctane and then, has been quantified with capillary gas chromatography-triple quadrupole mass spectrometer. Results reveal better extraction by micelles than traditional method, mostly more interesting are binary system of surfactants, i.e. cationic-nonionic, anionic-nonionic. It was found that solubilization of analytes increases with increasing hydrophobicity of surfactant. Therefore, the aim of this study is to improve this method comparing solubilization capability of these surfactants based on different number of ethylene oxide units in nonionic surfactants, tail length and the number of chlorine or benzyl groups in cationic and anionic surfactants in order to optimize recovery of pollutants and minimize the quantity to be used in environmental monitoring programs.

WE070 Water Treatment - A Regulatory Challenge under Regulation (EC) No 1107/2009

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Regulation (EC) No 1107/2009 aims to protect humans and the environment and lays down rules for the authorisation of plant protection products in commercial form and for their placement on the market, use and control. Plant protection products consist of or contain active substances, i.e. the molecules or materials responsible for the action against the target pest, weed or fungal. Ozonation and chlorination are primary disinfection processes for central water treatment. Metabolites of certain active substances were found to react during ozonation of drinking water and to form by-products with toxic, carcinogenic and genotoxic characteristics. During the application process of active substances, data gaps have recently been identified by EFSA regarding Article 4, 3(b) of Regulation (EC) No 1107/2009: A plant protection product ... shall have no immediate or delayed harmful effect on human health ... directly or through drinking water (taking into account substances resulting from water treatment).... In contradiction, water treatment processes are not implemented in the data requirements (Reg. 283/2013 or 284/2013) and no guidance documents for environmental testing are available. Published EFSA conclusions of active substances submitted under the 3rd group of the Annex I Renewal Programme (AIR3) are evaluated for different applied ways of addressing ozonation and chlorination during the registration process. A statistical overview of the results shows whether water treatment processes have been addressed successfully. An overview of these regulatory challenges is provided with intermediate recommendations to address water treatment processes for active substance approval.

WE071 The identification of persistent, mobile, toxic (PMT) chemicals as SVHC based on their equivalent level of concern to persistent, bioaccumulative, toxic (PBT) chemicals defined in Article 7(1) of REACH

S. Hale, Norwegian Geotechnical Institute; H. Arp, NGI / Environmental Technology; L. Vierke, German Environment Agency / Chemicals; M. Neumann, German Environment Agency (UBA) / Section IV 2.3 Chemicals

The identification of polychlorinated biphenyls in top predators nearly 50 years ago led to the establishment of many environmental chemistry regulations and chemical regulatory frameworks directed towards persistent, bioaccumulative, toxic (PBT) or a very persistent, very bioaccumulative (vPvB) chemicals. In order to protect human and the environment, additional physico-chemical properties to PBT and vPvB may be relevant to consider. Mobility is one of these additional properties and persistent, mobile, toxic or very persistent very mobile compounds are commonly problematic and the current method has limitations. Only mobile compounds have entered the environment they have the potential to impact remote pristine water bodies and raw water used for drinking water owing to their ability to spread spatially. Mobility has been qualitatively defined in REACH as "the potential of the substance, if released to the environment, to transport to groundwater or far from the site of release" (Annex II of REACH). However, a commonly accepted quantitative definition of mobility is not given within REACH and registrants/manufacturers are not obligated to carry out an assessment of mobility. Here we present a case for the consideration of PMT and vPvM as substances of very high concern (SVHC) based on their identification through
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 shown that this substance has the potential to accumulate on the candidate list is the most effective 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 accumulative 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 very Persistent and mobile PMT substances have been registered under REACH? - vP/vM/PMT screening by using the Danish QSAR database

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U. Germany, has initiated work to develop criteria to identify substances which are very persistent and mobile (vPvM), and very persistent, mobile and toxic (vPvMvT). The exposure poses and uptake carcinogenicity of surface water compartment and for ground water used 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 mobility (M) and very mobile (vM) substances 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 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 only 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 able to correctly identify and exclude substances registered under REACH. 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

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The chemicals that have the greatest chance 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 great numbers when screening the screening algorithms regarding these market, there was 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. In this 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., Pscreen) was performed according to REACH guidelines. For mobility, a criteria of a measured (or estimated) log Koc 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 specific additional functional and critical properties on high-end performance garments, workwear, first responder gear and other key applications. Within the non-polymeric PFAS category, fluorotelomer-based surface active agents (e.g., "fluorosurfactants") are used in complex multi-component formulations such as Cleaning Products, Paints, Coatings and Aqueous Film Forming Foams (AFFF). The non-polymeric fluorotelomer-based products provide superior surface wetting and leveling properties, while the accumulative 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.
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 ecosystemic 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 Subregion II, which consists of sandy and silt deposits of sable 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. Aelianic sediments, which belong to the Junín Formation (Aeolian Platense), normally do not exceed 5 m in thickness and usually 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 phyllosilicates and chlorophyres, TOC, arsenic and fluoride. Also, cyto and genotoxicity of concentrated waters were studied by comet assay using coelomocytes of Eucera fletida. Water quality was analyzed in combination with the dominant taxon of invertebrates founded. They were mainly pray Copepods, Acrasi, Collembola, Insecta, Oligochaeta, Nematoidea. 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
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Very polar organic compounds are of special interest for drinking water utilities, since they are subsamples that can 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 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 non-target 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, uretropin, metformin and guanylurea and newly detected compounds cytoxan, cyanuric acid and chlorpyrifos. Despite of 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
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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 plasticizers 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 or nothing has been given to the presence of other phthalates and non-phthalate plasticizers. In consequence, this work focuses on the occurrence, fate and effects of these alternative plasticizers in the environment. 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 collected 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 assessment for DEHP and its non-regulated substitutes. Today, the high-molecular-weight-plasticizer Diisononil phthalate (DINP) is the most abundant plasticizer detected in the SPM-samples. Our results indicate a fast appearance of new plasticizers like Dioxonil monoyl cyclohexane-1,2-dicarboxylate (DINCH) in freshwater environments and we identified several compounds, for instance Di(2-propylheptyl) phthalate (DPPH), 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
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Aquatic 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 substantial concentrations of residual dye. 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 μmol m⁻²s⁻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, 300, 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, 10 × 10⁶ cells mL⁻¹ of P. subcapitata as food). P. subcapitata was more sensitive to DB15 (LC₅₀ 13.30 mg L⁻¹) than C. dubia (LC₅₀ 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 DB15 concentrations, total progeny, number of released clutches, and reproduction were significantly decreased in C. regaudii, 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. Triebskorn, 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 WWTPs were significantly modified with powdered activated carbon; the third one was 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 lipids) were assessed. The acute toxic effects of DB15 dye in cladocerans were determined at 48 h; tested concentrations were 100, 200, 300, 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, 10 × 10⁶ cells mL⁻¹ of P. subcapitata as food)). P. subcapitata was more sensitive to DB15 (LC₅₀ 13.30 mg L⁻¹) than C. dubia (LC₅₀ 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 DB15 concentrations, total progeny, number of released clutches, and reproduction were significantly decreased in C. regaudii, 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.

WE081 Application of eco-genotoxicological 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 to investigate 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 O_3/L). We exposed zebras to dechlorinated tap water (n=3), STP effluent (n=3) and ozonated STP effluent (n=3). 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 mRNA extraction 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 liver VTG-2 gene expression, a marker of estrogenic endocrine disruption, was induced by the ozonated effluent. This indicates that ozonation possibly created estrogenic OBP. Furthermore, the ozonated effluent contained with organic and a nitrogen component in lower amounts. A second oxanay-related behavioral phenotype was recorded in the adult fish exposed to the ozonated effluent. No adverse effects on the fish progeny was noted. Whether these biological effects would have an adverse impact on the population level remains speculative. Ozonation is a capable method for removing pharmaceutical residues from sewage effluents. Yet its implementation should be carefully monitored in order to minimize undesirable side effects.

**WE084** Toxicity evaluation during secondary effluents treatment by UV/H2O2 using Eruca sativa and Artemia salina

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When advanced oxidation processes are applied there is the concern of not forming more toxic compounds as a result of the oxidation and transformation of organic compounds. Therefore, the presence of contaminants of industrial origin may affect disinfected and form more toxic by-products. Thus, a detailed study of by-products formation and toxicity assessment during the oxidation process contributes to a better understanding of the characteristics of the region, time of year, etc. In the specific case of the city of Limeira SP, there is a high concentration of compounds of industrial origin and metals (Al, Fe, Zn, Cr, Ni, Cu and Pb) above that allowed by the Brazilian Legislation (CONAMA) in sewage due to the presence of many jewelry semi-jewelry industries. This work evaluated the toxicity of a secondary effluent, formed with organic and a nitrogen component in lower amounts. A second oxygen treatment (UV/H2O2), through tests with anguila seeds (Eruca sativa) and Artemia salina. Samples of secondary effluent from a pilot plant located at the School of Technology campus (UNICAMP, São Paulo, Brazil) were collected immediately after the secondary treatment and then they were treated and analyzed in triplicate. The biological reactor was a hybrid: septic tank – anaerobic filter. After collection, 250 mL of samples were transported to the laboratory and stored at 4°C. Based on the preliminary tests it can be concluded that the secondary effluent treatment by UV/H2O2 in presence of high concentration of metals, nitrate, carbonate and industrial contaminants has no significant increase on toxicity.

**WE085** Hospital effluent induced oxidative stress on Xenopus laevis larvae

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Hospitals are one of the main sources of emerging pollutants to wastewater treatment plants (WWTP) that usually are fairly equipped to treat this kind of compounds. Activities performed in hospitals require the use of several compounds, which are potentially toxic, they can reach municipal wastewater, and if they get into municipal waste water treatment plants, in some cases, the procedure carried out in this WWTP is not able to remove all the contaminants, when they are not properly disposed, exposure to them can generate harmful effects on aquatic organisms. Physiochemical and pharmaceutical (11 pharmacological) characterization of the hospital effluent were made, results shown a high concentration of mercury, and pharmaceuticals on concentrations of μg/L. Also oxidative stress was evaluated on Xenopus laevis larvae exposed to this hospital effluent; twenty oocytes were selected for each exposed group (control, 0.1, 0.3, 0.5, 0.7, 0.9 and 1%) in the middle blast stage, they were maintained at constant temperature 23 ± 2°C, for 96 hours until they reached the larval stage. They were...
weighted, homogenized and centrifugated for the determination of hydroperoxides, liperoxides, 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

K. Schröder, RWTH Aachen University; A. Shuliakevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; Y. Müller, RWTH Aachen University / Institute of Environmental Research; S. Holz, RWTH Aachen University / Institute for Environmental Research; S. Schiwy, RWTH Aachen University / Department of Ecotoxicological Analysis; H. Hollett, RWTH Aachen University / Institute for Environmental Research

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 sludges in sewage treatment plants. In order to elucidate potential-androgen-disrupting MPs is the implementation of an additional treatment step like ozonation. Within the DemO/AC 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 activity in rainwater due to 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-)AR-CALUX® assay. These studies will be conducted associated to the DemO/AC Project as part of an exploratory study. First results revealed an antiandrogenic 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 widespread 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 facilities. The Nairobi River basin (Kenya) is an example of such contaminated areas. The wastewater generated from the city’s informal settlements and the insufficient WWTT 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 additional tests in 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.

**WE089**

Occurrence of pharmaceuticals, metabolites and transformation products from combined sewer overflows in London measured by high resolution targeted, suspect screening and untargeted chemical analysis

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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, CSOs 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 the observation of high concentration in influent and effluent wastewater; (b) determination of CSO markers in receiving river water over a six-week period; (c) suspect screening to identify metabolites/transformation products; and (d) classification of samples using untargeted data analysis. By differentiating influent and effluent, CSO markers were identified including caffeine, bezafibrate, benzoylegone and furosemide which were present in influent 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 events. Furthermore, four CSO markers were previously identified as antiestrogens, androgenic steroids, and antidepressants. These observations, along with the 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 along with statistical methods is necessary to achieve a fully 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

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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)) we established that some compounds were identified as wastewater effluents treated using secondary treatment or ozonation was investigated. It was found that of the 12 pesticides showed either antioestrogenic 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 importance 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 novel 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

Paradigm of fast inorganic nutrient uptake by aquatic ecosystems and their impact on primary production: a review


The paradigm of fast inorganic nutrient uptake by aquatic ecosystems and its role on primary production remains controversial. This review presents the most recent findings and discusses the potential impacts of fast uptake on the functioning of aquatic ecosystems. The review focuses on the mechanisms of fast uptake, the distribution and importance of fast-uptake-competent phytoplankton, and the potential impacts of fast uptake on primary production and marine biogeochemistry.

WE092

Photocatalytic degradation of perfluoroalkyl substances in an aquatic food web


The study aims to investigate the fate of perfluoroalkyl substances (PFASs) in an aquatic food web affected by sewage effluent. The research was conducted in a stream where sewage effluent is discharged into the water body. The study involved the measurement of PFAS concentrations in water samples and in different organisms of the food web, including fish, macroinvertebrates, and plants. The results showed that PFAS concentrations were significantly higher in organisms closer to the sewage effluent discharge point, indicating bioaccumulation of PFASs in indicator organisms and their ability to adapt to the polluted environment. The study contributes to the understanding of the impact of PFASs on aquatic ecosystems and underscores the importance of monitoring PFAS levels in aquatic environments.
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 exploitation of energy from domestic waters from 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 medium. The removal of crystal violet from aqueous solution by sulphuric acid treated sawdust, a local biomass, and 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 and the analysis of variance (ANOVA) statistics showed a good fit for the Langmuir adsorption model. 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 decolorisation of 68.39 mg/L CV (97.2%). A linear model was used for the decolorization 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/L, depending on the composition of the adsorbate matrix. The adsorption kinetics was well described by the pseudo-second order model (R² > 0.95). All the eight adsorbate systems investigated were endothermic (ΔH positive; 35.30 to 43.66 KJ/mol), thermodynamically feasible (ΔG < 2.30 to -6.13 KJ/mol) and had increased entropy.

WE089 Rapid detection of E. coli in wastewater effluent and impact of effluent discharge on riparian invertebrate diversity
P.M. Mosoloiou, University of the Free State / Zoology and Entomology

Effluent samples from wastewater treatment plants are expected, due to the treatment processes, to have lower concentrations of toxiancients 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 Phuthaditjhaba'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 method/technique 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 ecotoxicological investigations of the wastewater treatment plant Aachen
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Microplellants (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 influence 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 compared with 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 influent 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. antispadorum 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 genotoxic and endocrine effects originate by dilution of the WWTP effluent 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 dredgings
M.H. Wagelmans, Bioclear earth

In a densely populated country like the Netherlands, with a dense sewage system,
The demand for food products is pushing aquaculture to increase its production. 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 macrolide in human diets not only in Asia but also in other regions, we can ultimately ingest high levels of these contaminants, which are not degraded the same way by other fish products. Exposure tests were performed with the macrolide Ula 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 should be designed 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 domestic bacteria might transfer to human- or animal-associated bacteria including zoonotic pathogens. In the present study we followed two swine and two dairy farms and quantified ARGs and MGEs with 363 primer pairs using a high-throughput qPCR array. Samples were taken from fresh manure, stored manure, unfertilized soil, soil fertilized with manure, and tile drainage water from ditch, which was sampled before and after land application. We aimed to answer the following questions: do ARGs disseminate to the environment (field soil and surface waters), are ARGs enriched in stored manure, and are ARG abundances elevated in soils at crop harvesting time. For evaluating the ARG mobility potential, genes related to MGEs were also quantified. DNA was isolated from the soil and manure samples with the PowerSoil DNA Isolation Kit (MO BIO Laboratories). Quantitative PCR reactions were conducted using TaqMan Genotyping Master Real-time PCR system. The ΔCt values, ΔΔCt values, relative gene abundances (R), and fold changes (FC) were calculated with R version 3.2.3 and RStudio Version 0.98. In total, 182 out of 363 ARG and MGE qPCR assays were positive in one or more samples. Out of the positive assays, 161 targeted ARGs and 21 MGEs. Fresh manure had the highest diversity of ARGs and MGEs with 130 positive assays, followed by stored manure and manured soils. The number of positive assays decreased in fertilized soil between the 2 and 6 week sampling points. Only 29 assays were positive in 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 samples 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, hence a consequence of fertilization. However, the abundance and number of these ARGs and MGEs clearly decreased from fertilized soil to fertilized soil samples taken 2 and 6 weeks after manure application. During manure storage the relative abundance increased more than 4-fold for 41 genes and more than 2-fold for 62 genes. The highest increase (up to 65-fold) was observed in tetracycline-resistance genes, followed by sulphonamide and aminoglycoside resistance genes with up to 45-fold and 41-fold increases.

**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 (ng L⁻¹ to low μg L⁻¹), they have been specifically designed to be biologically active at low concentrations in human systems 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 flumequine (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 of these species in order to evaluate whether organisms are at risk of post-exposure degradation. 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 scientists. Especially when 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 Regulation of Dispensing and Prescribing of Drug, or Restriction of Adding Antibiotics in Animal Feed, have been enforced in view of public health management. The reduction of antibiotic usage in Korea has been decreasing trend 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 uses 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 emiss

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 on microorganisms 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 (ASRT) (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 antibiotic inhibition 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.

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 diet quality, and 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 a control water or a waterborne CIP exposure was given to two different types of leaves that were microbially colonized in the presence of CIP, or a combination of the latter two effect pathways. During the feeding activity assay, G. fossarum was rather tolerant towards waterborne antibiotic exposure with LC50 and EC50 values of 13.6 and 6.4 mg CIP/L, respectively. Furthermore, the shredders did not show any significant differences in food intake. However, the fungal biomass (an important food quality parameter) was significantly reduced in the highest CIP-treatments (0.5 and 2.5 mg/L), which indicates that antibiotics might affect the quality of the food for shredders. This assumption will be supported by the results of the long term bioassay at 0.5 mg CIP/L: likely due to an alteration in fugal biomass, the shredders’ leaf consumption and growth were significantly affected when subjected to the diet-related pathway. Our data indicate that indirect effects of antibiotics on shredders via the diet-related effect pathway could be more relevant than waterborne exposure. Since shredders play a key role in the leaf litter breakdown of heterotrophic stream ecosystems, diet-related effects might result in implications for the energy dynamics of these systems.

Efficacy of removal antimicrobial resistance genes during avian manure composting process.

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Antimicrobial resistance (AR) is an emerging and global problem. Therefore, there is currently a remarkable effort to understand the mechanisms of resistance, to promote the responsible use of antimicrobials and to seek effective therapeutic alternatives. While most livestock studies are focused along the food chain, there are few available studies about the role of livestock manure in the spread of AR. The direct application of animal waste (or slurry) to crops may favor the transmission of AR from cattle to vegetables. The objective of this work is to evaluate the impact of the composting process on the persistence of AR genes. For this, a composting of 10 weeks of duration has been carried out from straw and avian manure, from a laying hen production. Composting samples were taken in triplicate at the end of each week, and total DNA was extracted from each. 22 genes coding for resistance to tetracyclines, sulfonamides, phenicols, aminoglycosides, quinolones, beta lactams, vancomycin and colistin were detected and quantified by real-time PCR. 16 of the 22 genes were detected in at least one sample. Analysis of the temporal evolution of the resistances shows that there is a marked reduction (> 97%) in the genes coding for tetracycline, b-lactam, quinolone and macrolide resistances, while an increase in aminoglycoside and sulfonamide resistance genes is observed. These genes usually form part of integrons, which have more persistence into the environment. Besides, we have found positive correlations among most all ribosomal protection genes and with the deactivation genes, whereas efflux 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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Environmental pharmaceuticals contamination is now a recognized threat in coastal and estuarine ecosystems representing unknown and potentially harmful effects to non-target organisms. It is therefore paramount to monitor their presence in the environment and to evaluate the extent of their influence. The Tejo estuary, located on the Atlantic coast of Portugal was established as an environmental monitoring site because of its proximity to the highly urbanized areas in the Great Lisbon Region, with sewage disposal and discharges from industries, hospitals, agricultural and fish farms in upstream areas as well as urban effluents, anticipating the presence of contaminants. A monitoring campaign was conducted during summer where water, sediment, macroalgae, invertebrates and fish were sampled for pharmaceutical determination. Thirty sampling stations were selected according to their proximity to discharge points where effluents were contaminated to be expected in pharmaceuticals but also including reference sites where contamination was expected to be very low. Multi-residue multi-class analytical methods developed for each matrix are being applied for the detection and quantification of 67 compounds. The compounds monitored included anti-infective and anti-inflammatory drugs, angiotensin receptor blockers, b-blockers and antibiotics (42 compounds) in a total of 67 drugs. Multi-residue multi-class analytical UHPLC-ToF MS methods developed for each matrix are being applied for the detection and quantification. The knowledge gathered will then be applied to exposure assays and 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 antimicrobial 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 case in the case of antibiotics that can accumulate in soil, such as fluoroquinolones, which have a high adsorption capacity for, hence bioaccumulate and, along persistence, and of this antibiotic in soil 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.

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 water bodies 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 antibiotic active substance as part of a repeated test. As test organisms Synechococcus leopoldinis (limnic cyanobacteria) and Synechocystis 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 (A. glutinosa) was exposed to environmentally realistic concentrations of antibiotics during the natural leaf conditioning process. The 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 feeding assays 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 andweighed. 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=0.126) did not significantly alter 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 procedures, 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 analysis for antibiotic resistance genes (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 *int I* 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 fungal infections as well as prophylactic agent to prevent fungal infections in HIV patients. The aim of the study was to determine yeast pollution, susceptibility of these to fluconazole, 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 selected using selective media and incubation at 37°C and identified using biochemical methods. Resistance to fluconazole was assessed 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 yeast isolates have been associated with polluted waters. Some isolates in the present study are pathogenic and of clinical importance. The potential impact of resistance to fluconazole is mainly associated with the use of this antifungal agent as prophylactic agent to prevent infections in HIV patients. The results demonstrated that the isolated yeast species are associated with faecal pollution and that these isolates are pathogenic and could cause superficial and life-threatening infections. Fluconazole is considered as a selective agent in the treatment of fungal infections and a potential prophylactic agent to prevent fungal infections in HIV patients. The 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 *int I* gene was found in the soil where the digestate was added, showing its introduction through this agricultural practice.

**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 Feucherosel (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 complements 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 found in agricultural soil, suggesting that 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 silt 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, su1 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 taxa, resistant to the antibiotic. These results permitted to characterize the global fate and impact of SMX in an agricultural soil. SMX appeared in mobile DiSSt which enhanced mobility 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, Misep, ARG, biodegradation

**WE115**
**Risk assessment of antibiotic resistance and related genes in human exposed 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 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. We have used the combination of different methods for obtaining that information: Parallel quantitative PCR array for high throughput quantification (1), epicPCR(2) for host information and Inverse-PCR(3) for analysis of the genetic environment. Inverse-PCR and epicPCR combined with DNA sequencing resolve also the sequence of the resistance gene. Samples were collected from different locations in Finland, mure from cattle and pig farms, soil that received the manure as fertilizer, sediments from aquaculture farms and effluent, influent and activated sludge from waste water treatment plant. Our results demonstrate that human activities results to the increase to the abundance of antibiotic resistance genes. In many cases the genes are located in mobile genetic elements with increases the probability of transfer of the them between bacterial species. The host range information obtained by epicPCR revealed wide diversity on the host range of the antibiotic resistance genes in different environments. Our results can be used for the development ecotoxicological risk analysis for antibiotic resistance. (1) Karkman, A., Johnson, T.A., Lyra, C., Stedtfeld, R.D., Tamminen, M., Tiedje, J.M. and Virta, M. (2016) FEBS Microbial Ecology 92 (3): fw014 (2) Spencer, J.S., Tamminen, M., Tiedje, J.M., Tiedje, J.M. and Virta, M. (2016) ISME Journal 10: 427–436 (3) Parnäinen, K., Karkman, A., Tamminen, M., Lyra, C., Paulin, L., Hultman J. and Virta, M. (2016) Scientific Reports 6: 35790

**WE116**
**Risk of antibiotics in the environment**

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For decades, pharmaceuticals have been fundamental tools against disease and infection 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 spurring 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 persistency 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 antibiotics 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 included environmental matrices such as surface water, ground water, soils, sludge, sediments and biotic systems (e.g. via urban effluents) and 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 only partially metabolized and a large amount is excreted unaltered or as active metabolites, reaching wastewater treatment plants (WWTPs). Most WWTPs 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 sulfonamide antibiotics to treat urinary tract infections used both in human and veterinary practices. The widespread use of SMX implies its antibiotic-induced selection for the occurrence of both biodegradable and resistant to hydrolysis. The high frequency of detection and relative persistence of SMX in environment cause a potential risk of antibiotic 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 antibiotics on the natural microbial community were evaluated in terms of cell vitality and abundance. Moreover, the spread of sulfonamides resistance genes was evaluated by quantifying the sul I gene. The antibiotic SMX was biodegraded with a DT50 of about 20 days. The microbial abundance not only was not affected by the antibiotic addition (t=0 days), but at the end of was significantly higher in treated microcosms than in control conditions. The abundance of sul I increased after addition of SMX, suggesting that ARG spread is a physiological adaptation of natural microbial community to its presence.

WE118 The effect of antibiotics on representatives of aquatic algal and plant species B. Zellini, University of the Marche, Department of Bioscience and Biotechnology and Engineering in environmental protection; R. Beisnöva, L.N. Gumilov Eurasian National University / Department of Management and Engineering in environmental protection
Nowadays, pharmaceuticals are pollutants of increasing interest. The volume of the production of pharmaceuticals has been increasing rapidly in the last decade in Kazakhstan. Antibiotics are one 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, clarithromycin, azithromycin, sulfamethoxazole, oxytetracycline) 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 ingredients (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_ species were cultured in Tarnita medium and algae numbers were counted in Goryaev chamber under a microscope. The macroide substances azithromycin and clarithromycin 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 APIs in Asia 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. Hershay, 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 antibiotic resistance in bacteria. The presence of antibiotics 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 direct to 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 veterinarian antibiotics in both urban and rural streams. Antibiotics detected include sulfamethoxazole, sulfamethazine, trimethoprim, dantofloxacin, sulfapirazine, sulprofloxacin, 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

When researching antibiotic resistance (AR) in an environmental framework there will be a number of factors, antibiotics, metals and other selective agents, that are constantly in flux, which may facilitate or inhibit the selection and transfer of ARGs between microorganisms within a matrix. This article will outline the ways in which water quality analysis (WQA) can be used as a tool for understanding key components of systems under study outside the scope of microbiology. Specifically, how WQA can contribute additional understanding with regards to environmental variation of organic and inorganic compounds and metals, alongside the complexity of a given matrix. Samples drawn from the 3000m3 capacity slurry tank of a high input/high output dairy farm in the East Midlands were tested for 16 variables. These included Zinc and Copper, Dissolved Oxygen, Chemical Oxygen Demand (COD), pH and alkalinity, and various nutrients such as Ammonium, Nitrate, Nitrite and Phosphate. In addition, WQA was used to understand matrix variation within the slurry storage tank over different time periods, as a result of different management practices such as mixing and variation between different aspects of the slurry management system on the farm. This is supplemented with data from additional external influences; rainfall, temperature and farm practices, to further understand how the system as a whole can be considered when researching AR.

WE121 Safety and efficiency assessment of antibiotic administration by magnetic nanoparticles in Zebrafish
G. Chemello, C. Piccinetti, B. Randazzo, O. Camerlani, M. Maradonna, Università Politecnica delle Marche; M. Magro, E. Bonaiuto, F. Vianello, G. Chemello, Università degli Studi di Padova; A. Fifi, Biotecnologie B.T. Srl / Ecotoxicological and chemical; F. Gigliotti, CRO BioTecnologie BT; I. Olivotto, Università Politecnica delle Marche

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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 control 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 zebrafish (Danio rerio). Fish were divided into 4 experimental groups: control; group A exposed to 4mg/L OTC (through water); group B exposed to 100mg/L iron oxide to 4mg/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. Viitala, 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 qualitative and quantitative 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 from blank contamination, especially during development of 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 2010 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 could 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 results of implementation of several important corrective actions.

**WE123 Comprehensive Analysis of Elemental Contamination in Environmental Samples Using a Triple Quadrupole Mass Spectrometer (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 soils or sediments. Targeted elements comprise the "big four" arsenic, cadmium, mercury and lead, but also many other elements. Particular 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 quadrupole 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 chromatography or Mass Spectrometry Imaging, will be presented to show the broadness of accessible applications using modern ICP-MS instrumentation.

**WE125 ANALYTICAL METHOD FOR DETERMINATION OF FULLERENE (C60) NANO PARTICLES IN SEAWATER SAMPLES**

L.R. Diniz, Universidade Estadual do Maranhão / Agroecology; L. Capellini, Universidade Federal de São Paulo UNIFESP / Departamento de Química Fullerenes are allotropes of carbon produced in highly energetic processes of organic 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: dispersive 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 fluoride 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 recently reported. 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 using liquid chromatography electrospray ionization tandem mass spectrometry (LC-ESI-MS/MS), ultra-performance convergence chromatography (UPC2) 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 polytetrafluoroethylene (PTFE) membranes**

Y. Jeong, H. Kwon, KIST Europe / Environmental Safety Group; H. Jeon, KIST Europe; A. 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

Increasing occurrence of organic contaminants in the aquatic environment has heightened the need for reliable and efficient monitoring technology. 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 the polar organic chemical integrative sampler (POCIS). POCIS consists of Oasis HLB® sorbent sandwiched between two polyethersulfone (PES) 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 Kow from -0.03 to 6.26). Less membrane sorption was found in
PTFE membrane than PES membrane for all target compounds. Two types of POCISs were then deployed in a small river and the outflow of wastewater treatment plant for two weeks. Both POCISs showed similar chemical profile and 22 contaminants were detected including 6 priority substances enlisted in EU Water Framework Directive. Although PTFE membrane showed better permeation performance than PES membrane in laboratory experiment, the lag effect was still found from the field application of POCIS-PTFE.

WE128 Occurrence and Ecological Risk Assessment of Several Endocrine Disrupting Chemicals in Urban River Water and Sediment of South China

C. Huang, Jinian University; L. Wu, Y. Guo, Jinian 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 65600 ng/L and from not detected to 492 ng/mg 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 in both water and sediment (p < 0.01), indicating that these chemicals were in common sources and widely usage. After calculation, our study found that discharge from sewage treatment plant effluent discharges the dominant contamination pathway. 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 WWTPs, lagoons, irrigation channels and coastal areas. Water samples were analyzed using liquid chromatography-tandem mass spectrometry, using a hybrid triple quadrupole linear ion trap instrument (UPLC-QqLT-MS/MS). In order to assess seasonal variations, distribution and fate of pharmaceuticals, three 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/anti-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 antiadipic (glibenclamide) and the diuretic (furosemide). Salicylic acid was the most ubiquitous quantified compound in sediments, with a maximum concentration of 18.2 ng/g dw. These results pointed out that pharmaceuticals are widespread pollutants in coastal environments and that WWTP effluent discharges are the main source of contamination by these chemicals 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 µg/kg in sediment samples. Four MCs were prominent, with mean concentrations of 0.97–29.92 ng/L in pore water samples. The total concentrations of the different classes of PPCPs followed the order: N-APs (53.26%) > MCs (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 (3.88%) 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.

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. Celic, 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 pathway. 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 WWTPs, lagoons, irrigation channels and coastal areas. Water samples were analyzed using solid phase extraction (SPE) and liquid chromatography-mass spectrometry. Other contaminants were additionally analyzed using solid phase extraction followed by pressure liquid extraction extraction. The results showed that concentrations of ∑PFCs were ND–19.6 pg m⁻³ (for ∑PFCs) in air, ND–447.8 ng L⁻¹ (for ∑PFCs) in water, ND–9.7 ng g⁻¹ dry weight (dw) (for ∑PFCs) in sediments, ND–7.7 ng g⁻¹ dw (for ∑PFCs) in soil, and ND–35.0 ng g⁻¹ dw (for ∑PFCs) in the fish, respectively. The predominant species among the PFCs were perfluorooctanoic acid (PFOA) in air, perfluorooctanoic acid (PFOA) in water, perfluorooctanesulfonic acid (PFOS) in sediment, soil, and fish, respectively. PFOS and PFOA 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 PPCPs 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.

WE130 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, Euil University; M. Kim, Seoul National University / Department of Health Science, Y. Kho, Euil 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 PFCs in samples were measured. The results showed that the concentrations of PFCs in 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 m⁻³ (for ∑PFCs) in sediment samples. Carbamazepine was detected with 100% frequency in surface water and sediment samples. Caffeine 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 µg/kg in sediment samples. Four MCs were prominent, with mean concentrations of 0.97–29.92 ng/L in pore water samples. The total concentrations of the different classes of PPCPs followed the order: N-APs (53.26%) > MCs (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 (3.88%) 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.

WE132 Seasonal changes in water and sediments’ microplastics in a Mexican estuary (Tecolutla).


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 corflute (Whatman #40) filter, which was later dried at 50 °C for 24 h. Sediment samples 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 = 1.5–2) 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 microscope (Dissecting followed by digital Celestron) and photographs of the particles were obtained with imaging software. Vivialia of 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
understand the biological significance of their presence.

WE133 Simultaneous biodegradation of water treatment additives: Transformation and byproduct formation, impact of biocide shock dosing and salinity T. Wagner, University of Amsterdam / IBED; J. Parsons, University of Amsterdam / IBED-ELD; A. Langenhoff, H. Rijnaarts, 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 uptake is used in cooling towers. Several treatment technologies such as reverse osmosis, electrodialysis and membrane distillation may facilitate the reuse of discharged brackish cooling tower water. However, with 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 removal of pharmaceuticals from such construction- 
ed wide concentrations (9-150 microg L⁻¹). The mean concentrations for sulfamethoxazole, trimethoprim, cimetidine, paracetamol, lidocaine, metformin, carbamazepine and atenolol were 55.90 microg L⁻¹, 38.89 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. Venlafaxine has the lowest mean of 4.231 ng L⁻¹ other than α-haloalkylquinones compounds. A total of 35 pharmaceuticals were detected. We compare our 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 endogenous changes in the demand of pharmaceuticals in field samples. Method validation and sensitivity analysis were performed on published data. The results provide an overview on the occurrence and fate of pharmaceuticals in the aquatic environment, which may facilitate the reuse of discharged brackish cooling tower water and 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 44,000 ha and a shoreline of 125 km, is dominated by the quality-generating population (>160000 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 (WWTP), 7 irrigation channels and the Lake of L’Albufera de Valencia (Valencia, Spain). The influence of urbanization conditions and the Lake L’Albufera effects on the occurrence and fate of these pollutants in the environment is assessed. In order to evaluate the impact that these contaminants may have on biota, the isolation and concentration were carried out by solid-phase extraction (SPE) and the contaminants were determined by ultra-high pressure liquid chromatography-tandem mass spectrometer (UHPLC-MS/MS). Cocaine as its major metabolite (benzoylcegonine), followed by cannabis as its Δ9-tetrahydrocannabinolic acid, were the main drugs detected in water samples. Regard to pharmaceuticals, caffeine and ibuprofen were the main compounds obtained in these samples. Nevertheless, other pharmaceuticals were detected at high concentrations in all samples. In spite of this, and its non-complete removal in WWTPs, nowadays there are not enough knowledge about how the presence of these pollutants can affect to aquatic ecosystems, and specially living beings.

WE137 EFFECTS OF URBANIZATION PROCESS ON WATER QUALITY OF RIVERS ON THE SANTA CATARINA ISLAND, BRAZIL M. Barbosa Xavier, Universidade Federal de Santa Catarina / Biochemistry; C.H. Soares, Universidade Federal de Santa Catarina / Biochemistry Department The metropolitan region of Florianópolis has undergone an intense urbanization process in recent decades, having increased its population from about 500,000 people in 1970 to over 1 million and 1.4 million people in 2017 and 2020 respectively. Urbanization is a complex process that generates a series of changes in the landscape, in particular, the quality of water, which is of great concern in the region. The objective of the present study was to evaluate the water quality of the Itacuruí river in its estuarine region, in order to evaluate the anthetic 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 phosphon, 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/dichloromethane). The results obtained showed high concentrations of ammonia, total phosphon, fecal coliforms and sulfide, besides high total 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 prominent in GC / TOF/MS chromatographic analyzes. The results confirmed the high contamination of the waters of the Itacuruí River by the discharge of domestic sewage. Ecotoxicological tests using fish are being conducted, including assay to assess genotoxicity.
The wastewater generated by the human activity contains several anthropogenic contaminants, including emerging pollutants. Sewage sludge is generated during the depuration 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 is used in the agricultural sector as fertilizer, and the presence of pollutants could affect to the surrounding ecosystems. The sludge samples came 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 analytes were eluted with methanol at gravity flow. Once extracted, the analytes were identified by liquid chromatography-mass quadrupole-time-of-flight mass spectrometry (LC-QqTOF-MS). As a result 50 compounds were identified, being the pharmaceuticals the most relevant, present in all samples, including nucleotides (adenosine triphosphate), amino acids (phenylalanine) or organochlorine pesticides (lindane–phenthoate). 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 diminishing, giving a reasonable safety. 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 schistosomiasis and has the most severe schistosomiasis epidemic situation in China. Na-PCP has been sprayed as molluscicide in Dongting Lake from 1960s to 1990s, it was estimated that over 9.8x106 kg of Na-PCP had been devoted into the lake; CBs were also carried into the lake with using of Na-PCP. The aims of this study were to investigate/encounter contamination status, distribution of CBs in fish from Dongting Lake.

**WE139** chlorinated benzenes in fishes from dongting lake

Li, C., Institute of Water Resources and Hydropower Research; F. Zhang, China; Institute of Water Resources and Hydropower Research

Chlorobenzenes (CBs) are of worldwide concern due to their persistence, toxicity, bioaccumulation and long-range transport. Hexachlorobenzene (HCB) and pentachlorobenzene (PeCB) are listed as persistent organic pollutants (POPs) by the United Nations Environment Program (UNEP). CBs production in China accounts for more than 50% of global wide CBs production. On the other hand, several 1,2-dichlorobenzene (1,2-DCB), 1,4-dichlorobenzene (1,4-DCB) and 1,2,4-trichlorobenzene (1,2,4-TCB) in China was 12000, 30000 and 1000 tons in 2003, respectively. HCB has never been used as pesticide in China, 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 termed as Persistent Organic Pollutants (POPs) by the United Nations Environment Program (UNEP) because they are produced in large quantities worldwide and bioaccumulate in the environment. Overall, CBs are known to cause an array of health effects on non-target species and humans. The purpose of this study is to determine the CBs occurrence in fish from Dongting Lake (China) and the potential risk for aquatic organisms. Dongting Lake is China’s second largest freshwater lake (17,800 km²) and has been globally recognized as a Ramsar site since 2002. The lake is located in the central part of the central plain of China between the Yangtze and Han rivers. The lake is a mix of freshwater and brackish water (salinity ~ 10%) and has a surface area of 3,610 km² and a mean depth of 1.9 m. Dongting Lake is an important and unique ecosystem in China, where a large number of endangered and rare species live. The lake is a key water body for the survival and reproduction of many aquatic species and is a significant habitat for migratory birds. The objective of this study was to determine the occurrence of chlorinated benzenes in aquatic organisms from Dongting Lake. Fish samples were collected from different areas of Dongting Lake and were analyzed for CBs using high-resolution mass spectrometry (HR-MS/MS) and liquid chromatography-mass spectrometry (LC-MS/MS). The results showed that the highest levels of CBs were found in the liver of fish species, with concentrations ranging from 0.1 to 10 µg/kg wet weight. The levels of CBs in fish from Dongting Lake were comparable to those found in other Chinese freshwater lakes, indicating that CBs are widespread in China. The results of this study contribute to a better understanding of the occurrence and distribution of CBs in aquatic organisms from Dongting Lake and highlight the potential risks posed by these pollutants to the local ecosystem and human health. Future studies should focus on the evaluation of the health effects of CBs on aquatic organisms and the assessment of the potential risks to human health. This study is important for the protection of the ecosystem and public health in the Dongting Lake area. The obtained data can also be used to develop and implement effective pollution control strategies to mitigate the adverse effects of CBs on the environment and human health. **WE140** Occurrence of bisphenol A in Mediterranean mussels (Mytilus galloprovincialis) sampled from the north Adriatic coastal waters (Slovenia)

V. Čerkvenik Plais, University of Ljubljana; Veterinary Faculty / Veterinary Faculty; I. Fonda, Fonda d.o.o.; M. Gombač, University of Ljubljana / Veterinary Faculty

From January to October 2015 in total 27 samples of Mediterranean mussels (Mytilus galloprovincialis) and 10 samples of sea water were collected along the Slovenian coast in the north Adriatic sea to be tested for the presence of bisphenol A. Samples were collected at three shellfish farms, at the open sea and also from the harbour of Koper. In total 31 compounds were identified and the concentration of bisphenol A was detected at the level of 0.27 µg/kg w.w. in 3 samples of mussels and 0.18 µg/kg w.w. in 2 samples of sea water. The observed concentrations indicate a relatively low contamination of the Slovenian coastal waters as a part of the north Adriatic sea, with bisphenol A, compared to available publications about Mediterranean mussels.

**WE141** Toxicity of non-steroidal anti-inflammatory drug and the behavioural response in Juvenile Catfish

I.O. Ehruhnwuese, University of Benin, Benin City, Nigeria; Animal and Environmental Biology; I. Tong, University of Benin / Laboratory of Ecotoxicology and Environmental Forensics, Faculty of Life Sciences, Department of Animal and Environmental Biology, University of Benin, Nigeria; A. Muhib, University of Benin Benin City / Department of Environmental Management and Toxicology

The increasing levels of Pharmaceutical products in surface and underground water in third world countries is on the increase. We examined the toxicity of one phase-Tamiflu and personal care products (TCS) widely used in health care sector as fertilizer, and the presence of pollutants due to the high content in organic matter resulting in its introduction into environment and it has already been detected in rice crops. Samples were extracted by liquid phase extraction (SPE) with Strata-X cartridges and analytes were eluted with methanol at gravity flow. Once extracted, the analytes were identified by liquid chromatography-mass quadrupole-time-of-flight mass spectrometry (LC-QqTOF-MS). As a result 50 compounds were identified, being the pharmaceuticals the most relevant, present in all samples, including nucleotides (adenosine triphosphate), amino acids (phenylalanine) or organochlorine pesticides (lindane–phenthoate). 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 diminishing, giving a reasonable safety. 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 schistosomiasis and has the most severe schistosomiasis epidemic situation in China. Na-PCP has been sprayed as molluscicide in Dongting Lake from 1960s to 1990s, it was estimated that over 9.8x106 kg of Na-PCP had been devoted into the lake; CBs were also carried into the lake with using of Na-PCP. The aims of this study were to investigate/encounter contamination status, distribution of CBs in fish from Dongting Lake.

**WE142** Reproductive and maternal effects of Tamiflu metabolites in medaka (Oryzias latipes)

Lindsey Heng, Y. Wu, I. Meng Ian, W. Chen, Department of Biomedical Science & Environmental Biology, Kaohsiung Medical University, Kaohsiung

Tamiflu is the most commonly used anti-influenza drug. Human intake Tamiflu and excrete the Tamiflu metabolites into the aquatic environment. The Tamiflu metabolites might pose a potential risk to aquatic organisms. The purpose of this study was to assess the reproductive effect of medaka (Oryzias latipes) under long-term Tamiflu metabolite exposure. This study aimed to observe the survival, growth, and egg production of the adult medaka, and hatchability of embryo, and larvae body length of F1 medaka under the Tamiflu metabolite exposure concentration was 0.3, 0.9 and µg/L, respectively. Results showed that the survival and growth rates of adult medaka were no significant difference between the control and exposure groups. However, the egg production and F1 hatching rate of 90 µg/L exposure group had a downward trend compared with control group, but there were no significant decrease. This study found that larvae body length of exposure groups were significantly shorter than that of control group. This study concluded that Tamiflu metabolite could have a significant impact on larval growth development.

**WE143** Earthworms (Eisenia fetida) response to chronic exposure to triclosan

J. Zalasaukaitė, Vytautas Magnus University / Department of Environmental Sciences; D. Mitkelytė, Vytautas Magnus University

Triclosan (TCS) is a broad-spectrum and antifungal agent extensively used in industrial, household and personal care products. TCS widespread use has resulted in its introduction into environment and it has already been detected in surface waters, sediments, soil, living organisms and humans as well. The aim of the present study was to determine the effect of the preservative TCS on the earthworm Eisenia fetida. Earthworms E. fetida were exposed to 10-750 mg kg of triclosan in soil for 56 days. The impact of triclosan exposure on survival, growth rate and reproduction and antioxidative system was evaluated. TCS severely reduced the growth rate of E. fetida and reproduction. Chronic exposure to TCS in the soil induced a significant increase in the activity of antioxidative enzymes and malondialdehyde concentration.

**WE144** Predicting the fate of pharmaceuticals during wastewater treatment and crop irrigation with reclaimed wastewater

M. González García, C. Fernández-López, UCAM; F. Polesel, Technical
University of Denmark (DTU) / DTU Environment; S. Trapp, Technical University of Denmark DTU / DTU Environment

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 are predicted to take up considerable amounts of reclaimed wastewater contaminants. In this study, we investigated the fate of pharmaceuticals compounds in a wastewater treatment plant (WWTP) equipped for the elimination of carbon and nutrients. The primary treatment consists of a screen, an aerated bean extraction tank and a primary clarifier. The biologically treated wastewater from the conventional activated sludge is filtered through a layer of a continuously operating sand filter prior to being disinfected by ultraviolet radiation. Effluents are used for irrigation in agriculture. We used the simulation tool “Activity SimpleTreat - fate model for ionics in wastewater treatment plants” [1] to predict the fate of pharmaceuticals compounds in the municipal WWTP. Model parameters were adapted to the situation at site. Chemical data were estimated using ACD/Lab. 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), sappe, and potassium and fruit. By the inclusion of ion trapping in the alkaline phloem fluid (pH8). The preliminary results with the new steady-state model, showed the uptake capacity of pharmaceuticals in different tissues of lettuce. The assimilation and distribution of pharmaceuticals compounds in the edible part of the lettuce leaves and the subsequent passage to the harvested plant parts is investigated. [1] Franco A. 2011. Activity SimpleTreat - fate model for ionics in wastewater treatment plants. [2] Trapp S. 2017. New release dynamic (numeric) coupled soil-plant uptake model for monovalent ionics.

WE145 Exposure Assessment of Residual Organochlorine Pesticides (OCPs) in Orchard Soils and Fruits in Korea

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Residual organochlorine pesticides (OCP) are chemical substances that are resistant to environmental degradation chemical, biological and photolytic process, and are bioaccumulated with potential significant impacts on human health and the environment. OCPs were designated as persistent organic pollutants (POPs) by the international community at the Stockholm Convention on Persistent Organic Pollutant. This study was conducted to investigate the OCPs residue in orchard soils and fruits, and to determine the possible influence of OCPs on the edibility of fruits and vegetable. The potential risk of OCPs to human and environment was evaluated by the ingestion risk assessment. In the study, we monitored six OCPs to assess the levels of OCPs in orchard soils and their edible parts. The results showed that pesticide residues in orchard soils were not higher than the maximum residue levels (MRL) set by the European Union. However, the results of the study indicate that the use of selected pesticides in orchard soils could pose a potential risk to human health and environment.

WE146 PhytoCOTE project: Assessment of organic and inorganic contamination in vineyard soils

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Viticulture is one of the agricultural crops that uses the most important quantities of pesticides in France, in particular fungicides. These regular inputs may lead to a long-term contamination of ecosystems and thereby affect fauna and flora. Demands for more process-oriented role in precision agriculture. In order to improve the knowledge about the evolution in time and scale of different chemical contaminants within different soil types, a state of contamination level in soil surfaces and a characterisation of trace element availability were assessed. 53 plots with important pedological diversity were sampled over the 0-15 cm horizon. The soils were characterised (organic matter, Fe and Al oxohydrides, CEC, granulometry, pH) and total copper, cadmium, lead, zinc and 205 organic molecules were measured. The characterisation of trace element availability was performed using passive samplers (Diffusive Gradients in Thin films). A copper contamination due to past and current uses of Bordeaux mixture (copper sulphate)

has been put in evidence on the experimental site (until 197 mg/kg of dry soil). Concerning organic pesticides, a high diversity of molecules at different levels of concentration were found depending on crops. The results of the analyses will allow to show if: (1) the copper contamination level plays a role on the molecule degradation and contamination level; (2) the soil physical and chemical parameters play a role on the molecule degradation and on the copper and molecule retention; (3) the past and current soil uses impact the contamination levels.

WE171 Analysis and Assessment of Organic Contaminants in Materials Spread on Land in Scotland

E. Stuart, WCA Environment Limited; I. Wilson, G. Merrington, we.

Due to improvements in analytical capability increasing numbers of synthetic chemicals are being found in organic materials that may be recycled to agricultural land such as sewage sludge, animal manures, compost and digestate. Commonly occurring contaminants include pharmaceuticals, veterinary medicines, personal care products and persistent organic pollutants. Application of suitable organic materials to land is an attractive and apparently sustainable option that offers a range of agronomic and environmental benefits. However, there is a balance to be struck between the benefits of application to land and potential risks, such as the possibility of human and environmental health effects from trace constituents. It is critical though that consideration of this exposure pathway and any resulting regulatory decisions are risk-based and made using robust evidence and science. Previous assessments of risks posed by contaminants in materials applied to land generally have involved several assumptions and approximations for priority organic contaminants in a range of different organic materials that are frequently spread on land in Scotland. As a large number of organic compounds (in excess of 200) have now been identified in materials that are applied to agricultural land the first phase of this project has been to undertake a risk screening exercise. The purpose of this has been to identify organic chemicals likely to persist and/or bioaccumulate and to prioritise substances that are considered to pose a risk to human health or the terrestrial environment under reasonable worst-case assumptions for spreading. 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.

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WE148 Microplastics in Agriculture Soil

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Microplastic is an environmental pollutant of worldwide concern. However, neither microplastic concentrations nor their sources or sinks are completely known. This study was conducted to investigate the microplastic concentrations in agricultural soils and the risk of microplastic uptake by crops following wastewater irrigation. In this study, we used a state-of-the-art AFIT-IR Imaging system (128x128 pixel Focal Plane Array (FPA) microscope detector). >500µm 10 kg of soil was sieved through an 8mm, 6mm, 4mm, 2mm, 1mm and 500µm sieve. After the soil was dried it floated in a

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Will spent mushroom substrate application affect the dissipation and plant uptake of phthalate esters? J. Gao, Institute of Soil Science, CAS / Key Laboratory of Soil Environment and Pollution Remediation, CAS; F. Zhu, Institute of Soil Science CAS
To investigate whether spent mushroom substrate (SMS) amendment was an appropriate way to reduce di(2-ethylhexyl) phthalate (DEHP) and di-n-butyl phthalate (DnBP) contents in soil and whether SMS could reduce DnBP accumulation in bok choy, a mesocosm experiment was conducted. In the experiment, polypropylene and soil were spiked with di(2-ethylhexyl) phthalate (DEHP) and di-n-butyl phthalate (DnBP) to prepare a monocontaminated soil system. The effects of substrate application on DnBP and DEHP dissipation in soils and plant uptake of DnBP. Variations in soil pH and enzyme activities were determined. The concentrations of phthalate esters (PAEs) in soils, bok choy and atmosphere were examined with gas chromatography or gas chromatography–mass spectrometry. The results showed that the dissipation of DEHP is accelerated after incubation with SMS for 25 d, however little effect can be found with continuing 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 can 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 and 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)
WE152 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. Mesocosm studies allow the effects on both individual species and communities to be assessed simultaneously. Using an indoor laboratory studies, where test item concentrations are artificially maintained, mesocosm studies allow for a more realistic application and dissipation of test item. Therefore, mesocosm 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.

WE153 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 Vign E. Paterson, A. Thompson, Dow AgroSciences; G. Meregalli, Dow AgroSciences Italia s.r.l.; E. Hooper, Dow AgroSciences; G. Karaiskou, 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 Vignettes studies, recommends 1-2 large plants per 15 cm. There are two 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 vignette studies conducted to this guideline, regularly exceed 750 pots. These studies require a large amount of greenhouse 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. The study must be completed in a given time frame; and increasing plant variability within the study making data interpretation more difficult. OECD 227 is a guideline and different planting densities, which are considered adequate to generate robust data, may be used. However it needs to be assessed whether these different planting densities in the pots can impact the final endpoints determined in the Vegetative Vignette studies and ultimately the risk assessment. Data will be presented for test species planted at three densities to evaluate any impact on the Vegetative Vignette Study endpoints (expressed as ER50 values) used in the risk assessment.

SETAC Europe 28th Annual Meeting Abstract Book
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 implication 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 varied isoproturon tolerance and competition ability) to 6 isoproturon concentrations (0 to 1.75 mg/L), 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 concentration and replicate there were, e.g. by degree exposure, 10 different traits corresponding to morphological, biomass and physiological responses, as well as the response profile of 50 metabolites were quantified for aerial and underground plant parts, then representing respectively soft (easy to acquire) and hard traits. The subsequent dataset generated was used to model plant responses depending on isoproturon concentration and competitor presence/absence. For soft traits, dose-responses curves were built for each species, in presence and absence of competitor for each endpoint to define (1) the sensitivity, (2) their relevance to assess toxicity, (3) how competition modify points (1) and (2). In parallel, metabolic data were treated the same way, using a workflow created to handle high-throughput dose-response datasets from omics experiments. We then calculated toxicity values and built SSDs with and without competitor presence in an attempt to quantify competition effects compared to competitor-free ecotoxicological data. A final experiment, involving a complex assembly of the same 6 species under various isoproturon exposures was carried out to assess our model validity using community scale data.

How to consider recovery of aquatic plants in risk assessment?

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Exposure of non-target plants to plant protection products or other stressors can be restricted in time. For each endpoint, several replicates were, e.g. by degree exposure, 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 some cases. Thus, we decided to study the effect side and plant recovery by comparing the recovery potential in the risk assessment of methylparaben in soil to aquatic ecosystems. Methylparaben was mostly removed in the sewage treatment process, to have low toxicity to mammals, but there is no data on hazard assessment for soil ecosystems. Methylparaben is an endocrine disrupting chemicals (EDCs) and is contained in many 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 biodegradable or not processed without any metabolites. It was therefore 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 collembo. Plants were exposed methylparaben from 0 to 400 mg/kg for 14 and 21 days. In plant toxicity tests, shoot and root growth, root development, stomatal opening size, chlorophyll contents and photosynthetic factors were monitored. In the collembo 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 collembo 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 Methylparaben is an endocrine disrupting chemicals (EDCs) and is contained in many 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 biodegradable or not processed without any metabolites. It was therefore 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 collembo. Plants were exposed methylparaben from 0 to 400 mg/kg for 14 and 21 days. In plant toxicity tests, shoot and root growth, root development, stomatal opening size, chlorophyll contents and photosynthetic factors were monitored. In the collembo 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 collembo 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
increases of reactive oxygen species, lipid peroxidation (TBARS), reduced glutathione, superoxide dismutase, and glutathione transferase. The level reached of TBARS in P. kessleri was 10 times lower than in M. contortum, while the levels of antioxidant defenses were 3.5 - 7 times higher. These results suggest that oxidative stress would be involved in the toxicity of the glyphosate formulation and that the differences in sensitivity between strains could be due to differences in their defense antioxidant levels. According to the EU Directive 93/67/EEC, the IC50 values estimated indicate that the glyphosate formulation assayed should be toxic for aquatic biota. Besides, results also warn about its possible effects on the composition of phytoplankton, which would put at risk the balance of the aquatic ecosystem.

**WE162**

Indicator, indigenous and invasive species: the need of risk-benefit considerations in PPP risk assessment

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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, accidently 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 specifically 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 invasive species to reach the new habitats degraded by the presence of alien species. 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

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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* species has been developed and is employed to control the spread of invasive species. The OECD 239 guideline can be adapted to include measurements of fresh and dry weights need to be recorded. The OECD 239 guideline requires that only a qualitative assessment of the roots is undertaken. Auxinic substances are known to exert their herbicidal activity by affecting growing tissues. As such, roots of *Myriophyllum* plants may be affected after exposure to auxins. The methodology described in the OECD 239 guideline can be adapted to include measurements of fresh and dry weights for whole plants, rather than just the shoots, thereby assessing, indirectly, also possible effects on the roots. However, it needs to be evaluated if an indirect quantitative assessment of the roots in the *Myriophyllum* studies with auxinic substances would result in significantly different endpoints that should be taken into account in the risk assessment of auxinic substances. Results will be presented to clarify if effects on the roots in a water sediment system are providing additional information relevant for the risk assessment.

**WE164**

Testing the emergent macrophyte, Glyceria maxima in a water-sediment system: Results of a ring-test with Isoproturon

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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 suitable control allowances, to determine the required test duration, 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 imazapyr, scheduled for Spring / Summer 2018.

Understanding the physiology of T. praecox 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.

Phytoextraction of heavy metals in Cieneaga de Tamasopo wetland, Mexico, by Typha latifolia
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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 [2]. Aquatic plant species are 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 released from the plant; (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 Cieneaga de Tamasopo Wetland, which were characterized by 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 for several days to determine the 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 (GPX) and ascorbate peroxidase (APX) was carried out by spectrophotometric method [5]. The results show that APX and GPX enzymes showed an increase indicating possible sublethal endpoints reaching an inhibition of almost 50% for 64 mg Zn/L and presenting an endpoint reaching an inhibition of almost 50% for 64 mg Zn/L and presenting a hazardous endpoint reaching an inhibition of almost 50% for 64 mg Zn/L and presenting a hazardous state bioaccumulation under constant metal concentrations. ANOVA revealed differences in metal concentrations among sites for all metals. Metal uptake reaches the steady state bioaccumulation under constant metal concentrations. A dose-dependent response of T. praecox was observed for Ni, Cu and Zn. At the highest applied Ni concentration, statistical significant reduction of total chlorophyl content and carotenoids was observed. 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.
WE169
Toxicity of the binary mixture Cd-Zn on Lemna gibba evaluated using
morphological and oxidative stress enzyme endpoints
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PRIET; M. Saenz, PRIET CONICET, National University of Luján
The presence of metals in the environment represents one of the mayor concerns as
they are persistent in nature, non-biodegradable and can bioaccumulate in living
animals and plants. Metals in aquatic ecosystems may have effects on primary
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
were 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 activities where higher than control,
however neither of both presented significative differences with it. For the mixture
analysis, multiple regression was used to fit the observed %frond number inhibition
(%FNI) to dissolved metal concentration ([Mdis]). 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 toxic units (∑TU) for each mixture test case based on single EC50s. The average
∑TU 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
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Martinez, s. curieses, J. Alberdi, CONICET PRIET UNLU; W.D. Di Marzio,
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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 mining,
chrome plating, leather tanning and wood preservation. The aim of this study was 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, Scenedesmus quadricuada and Nannochloris oculata. These two 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
to1,73 mg/l. These concentrations were chosen base on previous experiments
through range finding tests. Subletal solutions were renewed monthly and algal
cells were subcultured in new medium. After the preadapted period, each subletal
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 dilucidated 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 relative 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
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Sanitation; O.A. Bandeira, T.J. Pinto, L.P. Figueiredo, University of São Paulo
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In Brazil it is very common to have mining waste placed in dams, especially in the

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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 worst environmental
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, Crotalaria
juncea, Canavalia ensiformis, Cajanus cajan, Cajanus cajan, Dolichos lablab,
Mucuna pruriens gray, Mucuna pruriens black and Lupinus albus). The
ecotoxicological assays were run using five treatments, which consisted in 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 avaluated
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 EC20 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 (97.54%). The height of the first two species was affected by
20% in the proportions 78.61 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 of
tolerance to the mining waste; the waste that outpoured of the Fundão dam 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
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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 [3] which fate and impact on agro-ecosystems
is still largely unknown. Several studies showed the deleterious effects of metal
nanoparticles (NPs) on soil microbial communities (1) and reported the importance
of organic matter (OM) content in the NPs ecotoxicity due to its role as dispersing
and stabilizing agent (2). A high OM content is likely to increase NPs toxicity by
favoring their dispersion. Based on this assumption, our goal was to assess 1)
whether the plant modifies the microbial ecotoxicity of NPs because of organic
matter enrichment in the rhizosphere through the root exudation and 2) whether the
plant variety mitigates the ecotoxicity according to plant traits. Endpoints that relate
to soil fertility (ie abundance and activity of microbial communities involved in
carbon and nitrogen cycle) were used to assess NPs effects. The experimental
design consisted in planted and unplanted soil microcosms contaminated or not
with two doses of CuO-NPs (1mg and 100 mg/Kg) before the seedling of wheat.
We compared the effect of two conventional varieties (Arrezzo® and Skerzzo®)
exhibiting contrasted traits reflecting different root exudation. Ecotoxicological
effects were assessed after 30 and 50 days by measuring plant traits on each variety,
microbial activities (respiration, nitrification and denitrification) and microbial
abundance by qPCR targeting 16S RNA gene and function genes. The main
physico-chemical properties of NPs were characterized by Dynamic Light
Scattering in rhizosphere and unplanted soil solutions in which ionic strength, pH
and dissolved organic carbon were also measured. The results showed that the NPs
hydrodynamic diameter was higher in planted soil solutions compared to unplanted
one. Comparison between planted and unplanted soil showed that the plant
hampered ecotoxic effects on the microbial activity of functional microbial groups
without significant changes in their abundance. Arrezzo® limited the reduction of
nitrification and denitrification suggesting that NPs ecotoxicity depends on the
wheat variety likely because of the effect of roots on NPs and /or the microbial
populations recruited in the rhizosphere that can be more or less sensitive to NPs.
Water Res
WE173
Use of Posidonia oceanica as a potential bioindicator species of metal
pollutants: cellular and molecular responses to mercury exposure
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Laboratory Networking Ecotoxicology Area; F. Onorati, ISPRA Institute for
Environmental Protection and Research; A. Cicero, ISPRA Institute for
Environmental Protection and Research / National Center for Laboratory Network
The marine phanerogam Posidonia oceanica, thanks to its worldwide distribution

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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. 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. Biological markers of oxidative stress of gill tissue, such as the glutathione S-transferase activities, the ascorbate peroxidase activity, the total antioxidant capacity, the phenols content, the level of lipid peroxidation and the micronuclei 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

W. Kim, J. Park, Gwangju Institute of Science and Technology / School of Earth Sciences and Environmental Engineering; S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering 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 multivariate statistical analysis. 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 evaluated using Citrus reticulata, and Pteris L. adunca. 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

(\textit{Rosmarinus officinalis} L.) plants

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Rosemary (\textit{Rosmarinus officinalis} L., \textit{Lamiaceae}) is an aromatic shrub native from Mediterranean regions and 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 response to changes of the concentrations and availability of 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) were incorporated into the 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 (\textit{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 increase grows, but also leads to decreased concentrations of carbon-based secondary metabolites, such as phenolic compounds.

**WE176**

Leaf litter originating from trees treated with systemic fungicides - a new exposure pathway for defense trigger systems

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Leaf litter decomposition, an important process in freshwater ecosystems, is mediated by microbial decomposers and leaf-shredding macroinvertebrates. This process can, however, be modified by chemical stressors such as fungicides. Although fungicide stress has increasingly been assessed in recent years, the systemic nature of some fungicides, which enables the uptake and distribution within treated plants, is not yet considered. Therefore, as a first step, we treated \textit{Alnus glutinosa} with a mixture of systemic fungicides (SFs; azoxystrobin, cyprodinil, quinoxyfen and tebuconazole) via soil drenching at three levels (control, field application rate (FR), and 10 times the field application rate (FRx10)). During leaf fall, we collected the leaves and assessed the potential impact of the fungicides on microbial decomposers and leaf-shredding macroinvertebrates. We quantified microbial leaf litter decomposition, their community composition and the palatability of leaves after microbial conditioning for the model shredder \textit{Gammarus fossarum} Koch. By assessing growth and physiological fitness of this species over multiple weeks, we additionally estimated the nutritious quality of leaf litter. Gammarids preferred conditioned FRx10 over control leaves, which may reflect changes in microbial community structure. This increase in palatability as a consequence of SF may be related to the fungicides’ ability to reduce fungal pest pressure, allowing trees to divert energy and carbon from defense to growth or storage. The same treatment resulted in a 300% increase in gammrild growth, while FRx10 was significantly inhibited. We suggest that SF may indeed have implications for microbial decomposers and leaf-shredding macroinvertebrates, while the underlying mechanisms are still not fully understood.

**WE177**

SETAC Plants Interest Group

S. Lustsei, DuPont De Nemour Hellas S.A.

**WE178**

Environmental Risk Assessment in Sediments (P)

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Biotic interaction of sediment-dwelling organisms promotes the release of contaminants across the benthic-pelagic ecosystem boundary, thereby affecting the exposure to and uptake of sediment associated contaminants at the sediment-water interface by themselves and the entire community around them. This way, bioturbation activity may contribute to species specific sensitivities to sediment associated compounds. Therefore we assessed if invertebrate bioturbation activity determines species specific sensitivities to sediment contamination. For two metals, Ni and Cu, sufficient data were available to construct Species Sensitivity Distributions (SSD). The position of the species in the SSDs could indeed have implications for microbial decomposers and leaf-shredding macroinvertebrates, while the underlying mechanisms are still not fully understood.

**WE179**

Effect based sediment quality assessment incorporating chemical fingerprinting

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Sediment quality assessment is often based on chemical analysis only, occasionally including toxicological assays. Full sediment quality assessment, including chemical and macrofauna analysis and toxicological assays, is not the standard procedure due to high costs. Based on chemical analysis only, it is not always clear whether sediment management in form of dredging and landfill or remediation is necessary. To reduce costs of sediment management on the one hand and to increase environmental benefits on the other, the right priorities need to be set. To do so, the University of Lorraine / Laboratoire Interdisciplinaire des Ecosystèmes et des Bioassays and macrofauna analyses. Chemical, toxicological and ecological data is available from freshwater/sediment monitoring campaigns in the Netherlands. A multivariate analysis was performed to identify contaminants with high impacts on the bioassays. From the 49 chemicals included in the dataset, 28 were significantly relevant to the outcome of the bioassays of Daphnia magna and Chironomus riparius. The Species Sensitivity Distribution (SSD) 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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Sediment quality assessment needs to be included in the EU WFD. Sediment quality assessment is often based on chemical analysis only, occasionally including toxicological assays. Full sediment quality assessment, including chemical and macrofauna analysis and toxicological assays, is not the standard procedure due to high costs. Based on chemical analysis only, it is not always clear whether sediment management in form of dredging and landfill or remediation is necessary. To reduce costs of sediment management on the one hand and to increase environmental benefits on the other, the right priorities need to be set. To do so, the University of Lorraine / Laboratoire Interdisciplinaire des Ecosystèmes et des Bioassays and macrofauna analyses. Chemical, toxicological and ecological data is available from freshwater/sediment monitoring campaigns in the Netherlands. A multivariate analysis was performed to identify contaminants with high impacts on the bioassays. From the 49 chemicals included in the dataset, 28 were significantly relevant to the outcome of the bioassays of Daphnia magna and Chironomus riparius. The Species Sensitivity Distribution (SSD) 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.

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 (Beijing), in both 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 in active 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 Bioturbation in contaminated sediments: effects on exposure, toxicity and biochemistry. T.M. Remaillé; W. Bennett, Griffith University / Environmental Futures Research Institute; S.L. Simpson, School of Land and Water / Centre for Environmental Contaminants Research; E.D. Bowmer, University of Antwerp / Department of Biology; D.T. Welsh, Griffith University / Environmental Futures Research Institute; E. Lombi, University of South Australia / Future Industries Institute; D. Howard, Australian Synchrotron; D.F. Jolley, University of Wollongong / School of Chemistry.

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. $\text{O}_2$) 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 guidelines; (ii) assess potential use of bioturbation in the management and natural recovery of heavily degraded sediment ecosystems. Increased bioturbation in predominantly metal-contaminated sediments increased bivalve ($\text{Tellina deltoidalis}$) and amphipod ($\text{Victoriopisa australiensis}$) survival from 53 to 100% and 42 to 93%, respectively; and reproduction in a second amphipod ($\text{Melita plumulosa}$) 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 biorator present showed the indubitable role of bioturbation. Waters within the biorator 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 operated with increased accumulation of nickel 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 biochemistry 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 M. Gillmore; M. D. Bloom, University of Wollongong / School of Chemistry; G.A. Price, University of Wollongong / School of Chemistry; L.A. Golding, CSIRO Land and Water; J. Stauber, CSIRO / CSIRO Land and Water; M.S. Adams, CSIRO, S.L. Simpson, CSIRO Land and Water / Centre for Environmental Contaminants Research; D.F. Jolley, University of Wollongong / School of Chemistry

Mining of laterritic 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, $\text{Melita plumulosa}$ 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 expected or response-to-exposure relationships obtained using traditional metal extraction 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, 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 reproducive 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/m$^2$h 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/m$^2$h DGT-labile Ni) and Site 2 (1.0 mg/m$^2$h 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 Guangzhou, China using a integrated method of TIE and EDA J. Yao, H. Li, F. Cheng, Jinan University / School of Environment Development; Y. Gu, Guangzhou University / School of Chemistry; Z. Huang, Guangzhou University / School of Chemistry; G.A. Price, Griffith University / Environmental Futures Research Institute; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems; L. De Villeurbanne / Microbial ecology of anthropised river systems.

Bioaccumulation and passive sampling measurements at the sediment water interface and in the water column. Relationships will be determined between the diffusion of reactive chemical species (e.g. $\text{O}_2$) 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 guidelines; (ii) assess potential use of bioturbation in the management and natural recovery of heavily degraded sediment ecosystems. Increased bioturbation in predominantly metal-contaminated sediments increased bivalve ($\text{Tellina deltoidalis}$) and amphipod ($\text{Victoriopisa australiensis}$) survival from 53 to 100% and 42 to 93%, respectively; and reproduction in a second amphipod ($\text{Melita plumulosa}$) 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 biorator present showed the indubitable role of bioturbation. Waters within the biorator 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 nickel 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 biochemistry and contaminant exposure to surrounding ecosystems.

WE187 Water discharges from the city of Lausanne during rainfall in Lake Geneva: Use of a triad anpheluces as an bioassay for environmental health monitoring M. Cardoso-Martinez, T. Benejam, R. Vivien, Centre Ecotoc; S. Pesce, Iotea Lyon-Villeurbanne / Microbial ecological of anthorised river systems; L. De Alencastro, Ecole Polytéchnique Fédérale de Lausanne / Central Environmental Laboratory; N. Dubois, Eawag; L. Rossi, Hyridique Ingénieurs; S. Höss, Ecossia / Animal Ecology; B.J. Ferrari, Centre Ecotox EAWAGEPFL.

Sediments represent an important compartment in surface waters. It constitutes a habitat or spawning site for many organisms and is an essential trophic resource for higher level organisms. It can be impacted by anthropogenic activities, particularly through urban wet weather discharges like stormwater and combined sewer overflows. In Switzerland, the Vidy Bay located in the middle of the northern shore of Lake Leman, in front of the city of Lausanne, is of particular interest as it receives a large portion of stormwater from the city of Lausanne via the Flon River. In this context, this study aimed to evaluate the impact of the Flon river stormwater overflows on the sediment quality of the Vidy Bay using a triad approach combining chemistry, ecotoxicology and the study of in situ benthic communities.
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. The ecotoxicity of the chemistry of sediment samples from the 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 biotests 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, elutriates and extracts. The analysis of the benthic meiofauna 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.

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 Demo2 AC-project aimed to assess the ecotoxicological status of the River Wurm 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 Elendorf, 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 g SEQ/mL) were applied for the fish embryo toxicity test with Danio rerio as well as the oxygen demand test. Enzyme activities (2-DCCD test) were performed on the organic extracts. The results showed estrogenic and mutagenic potential in sediment extracts upstream the tested WWTPs. The embryotoxic 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 samples 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 Demo2 AC-project will contain comparative studies in order to evaluate the possible influence to sediment toxicity after implementation of full-scale ozonation.

WE191
Swimming in turbulent water: impacts of suspended fine sediments on fish physiology
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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, sediment resuspension and dam releases 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 (Oncorhynchus mykiss) and roach (Rutilus rutilus) were exposed in semi-static conditions (0.4, 20 and 1000 mg/L of non-contaminated fine sediments) for 28 days, mimicking dredging operations in terms of duration and environmental concentrations. They were sampled weekly, and mortality, condition index and histological gill lesions were evaluated. Several physiological 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 as 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 point out the resilience of fish whereas they were exposed for 28 days to high fine sediment concentrations. Roach exposure to suspended fine sediments did not induce 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-related metals can be present in various chemical and physicochemical 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 however, classical techniques and methods such as the 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) invertebrate species 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 mortality. A semi-standardized Risk Assessment Team (28d) is 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 Entropy rule informed by environmental conditions (benthic invertebrates). A battery of bioassays that considers the use of three species belonging to different trophic levels has to be applied both to solid phase and liquid phase (pore water or elutriate) of sediment. The results of ecotoxicological analyses are then assessed as a whole at the level of “battery” (not of single bioassay), weighing the biological relevance of the measured effects, the sensitivity of organism, the statistical significance of measured results and the degree of correlation in both the bioaccumulation and exposure. Chemical and ecotoxicological data are finally integrated for sediment quality assessment, following the weight of evidence (WOE) criteria, this representing an innovative approach respect to previous regulation, where chemical classification was determined by at least one parameter exceeding the threshold level and ecotoxicological classification was determined by the worst bioassay result of the whole battery. In this work, a comparison between “old” and “new” sediment quality assessment was performed, applying the two classification methods to ecotoxicological data obtained on dredging sediments from different study sites. Results obtained underline the importance of using an integrated and weighted approach (WOE) respect to a “pass to fail” criteria.

**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-spiked laboratory bioassays with benthic arthropods belonging to different taxonomic groups are presented. In the 10-d sediment-spiked toxicity tests the LC50 values showed the following order from low to high LC50: Caenis horaius > Chironomus riparius > Gammarus pulex > Chironomus dilutus > Sericostoma personatum > Sialis lutaria > Hyallella azteca > Asellus aquaticus and Sialis lutaria. The Hazardous Concentration to 5% of the tested species (HCS and 95% confidence limit) derived from these 10-d LC50 values was 2.2 (1.2-5.7) µg/g organic carbon (OC). This HCS value is approximately a factor of 2 lower than the 10-d LC50 estimate (4.37 ± 0.75) 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 28-d LC10 values showed the following order from low to high LC10: Asellus aquaticus > Chironomus riparius > Caenis horaius > Ephemerida danica > Hyallela azteca > Gammarus pulex > Sialis lutaria. The HCS and 95 confidence interval derived from these 28d-LC10 values was 0.13 (0.02-1.50) µg/g OC. This HCS value is approximately a factor of 3 lower that 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 HCS obtained from 28-10LC 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 10x-LC50’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 fish and mollusc production in Portuguese estuaries. D. Faber, Bayer AG, Crop Science Division / BCS D. Eix, ECOTAXOLOGY / K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; E. Hellpointner, Bayer AG, Research & Development, Crop Science / Environmental Safety; E. Bruns, Bayer AG, Division Bayer CropScience / ECOTOXICOLOGY; M. Jäger, Hochschule Niederrhein / Department of Chemistry 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-waters 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 (log P ow 1) and B (log P ow >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/Biology 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 the assessment of the alternative means of producing animal protein, releasing, at the same time, pressure over wild populations. Production from capture fisheries has relatively stabilized for the past decades, whereas aquaculture production of aquatic animals has followed a rising trend, amounting, by 2014, 73.8 million tonnes produced. Asia contributed for about 89 percent of that production, followed by America, with a production of around 5 percent. Europe, contributing for about 4 percent of the world’s aquatic animals’ production in 2014. Portugal is a traditional fishing country, yet with little aquaculture activity, with yet little 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 differences may come as a disadvantage, as it is expected to provide equal nutritional properties and benefits within the scientific and regulatory community, primarily producers or benthic invertebrates. The objective of this study was to evaluate the potential side effects of two antibiotics used in aquaculture (oxytetracycline and flumequine) on the community composition of marine biofilms exposed to these substances and on the surrounding marine ecosystem. Marine biofilms were exposed to 1, 10, 100 and 1000 µg/L of oxytetracycline and flumequine for one week under laboratory conditions. Subsequently, the exposed biofilms were used to feed G. aequicuicuda organisms for two weeks. The G. aequicuicuda 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 a higher biorescence up to 0.01 µg/L, while the highest tested concentration contributed to the loss of biorescence. 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. aequicuada 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 biofilm 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. Increasing antibiotic use in aquaculture has led to unbound developments with the general diseases, the treatment of which requires the intensive use of antibiotics. The objective of this study was to evaluate the potential side effect of two antibiotics used in aquaculture (oxytetracycline and flumequine) on the diatom assemblage structure and biological traits of marine biofilms exposed to these substances. Biofilms were grown during two weeks under natural conditions on glass slides. Two experiments were carried out in each laboratory for each antibiotic. In both experiments, diatoms were determined to the lowest possible taxonomic level under the microscope (Nikon Eclipse TE2000-U). The taxon abundance of the sampled quadrants 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-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 apicina and Coccomis placenta. High exposure concentrations of oxytetracycline and flumequine (100 and 1000 µg/L) resulted in an abundance decrease of the genera Skeletonema pseudocostatum and Licmophora WE203

An updated version of the SEPA BathAuto tool for assessing anti-parasitic chemical treatments in marine fish farms

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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: azamethipos, 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. Azamethipos, 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 Gilibrand and Turrell (1999; MLA Report No 20/99) 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 paper 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 calculations (opendisp.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 in other jurisdictions and regulatory frameworks. 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


As 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), marine biofilms and harvesting techniques and new antibiotics continue to be used. 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 ecotoxicological risks of veterinary medicines applied in aquaculture production. 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.

WE205

Effects of an aquaculture parasiticide (diflubenzuron) on non-target shrimp
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 moultling 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 shown 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 the risk-dependent processes). The degree of exposure to potential of DFB can vary in different distances from aquaculture farms is also uncertain. We have therefore developed a set model scenarios representing different medical 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, Al and As were analysed in the muscles and livers of farmed and wild milkfish and mullerits from Tanzanian coast. Fish samples were collected from January 2016 to April 2016 and analysed for heavy metals by using Atomic Absorption Spectrophotometer while head was discarded. The Hazard Quotient (HQ) was calculated at distance of 1.5 km from aquaculture farms. Hazard Quotient at distance of 20 km is disregard to fish species and in order of decreasing dominance, the overall range of concentrations in (mg/kg ww) of heavy metals were: Fe (< LOD-11.96), Pb (0.54-1.96), Zn (< LOD-2.81), Cu (< LOD-2.31), Ni (0.015-0.098), Co (< LOD-0.086), Cd (< LOD-0.024, Cr (< LOD-0.079) and Fe (< LOD-147.9), Pb (0.92-47.37), Zn (< LOD-15.08), Cu (< LOD-12.24), Ni (0.027-0.994), Co (< LOD-0.034) and Cr (0.013-0.103) in muscles and livers respectively. Al and As were not detected above limit of detection in any of the analysed samples. Metals showed different affinity to muscles and livers of milkfish and mullerits, whereas Co and Cd had higher levels in the muscles than in the livers, Cu, Pb, Fe, Zn and Ni were higher in livers than in the muscles of milkfish. In Mullet, Fe, Co, Cr, and Cd levels were higher in the muscles than in the livers and Cu, Pb, Fe, Zn, and Ni levels were higher in the livers than in the muscles. The concentration of Pb in the muscles of all analysed fish were above the Maximum Recommended Levels (MRLs) by FAO/WHO, EU and USFDA for human consumption. Other metals were below the MRLs. The THQ for all analysed metals were below 1 indicating that the metals are likely to present insignificant health risk to fish consumers. However, based on MRLs recommended by FAO/WHO for human consumption, the metals may present health risk to human as fish consumer and threatens fish biodiversity. Therefore, this research calls for regular monitoring of heavy metals in fish for consumption and strengthening pollution control measures.

WE207 Potential Toxic and Phototoxic Effects of Benzobicyclon on Crayfish

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Benzobicyclon is the active ingredient in the herbicide, BUTTE®. In 2001, various formulations using benzobicyclon as the active ingredient were approved for use in Japan. It was approved for use in the United States and registered in California as of 2016, and was first applied to rice fields in the 2017 growing season. Benzobicyclon is a pyridophosphate that acts as a HPPD inhibitor, leading to the bleaching of weed species to ultimately kill them. With an increase in herbicide-resistant weed species, new formulations of herbicides to combat this in rice fields is advantageous. California is the second largest producer of rice in the United States, with Louisiana as the third largest producer; Arkansas leads the country in rice production. Unlike California, Louisiana has a unique system where rice fields not only grow rice but also grow crayfish for harvest and consumption. The craved rice fields are prime habitat for crayfish to reside; therefore, crayfish are at risk for exposure to pesticides used in the rice production. Benzobicyclon readily hydrolyzes to benzobicyclon hydrolysate, and therefore it likely undergoes photolysis as well. The potential for toxic or phototoxic impacts of benzobicyclon or its intermediate degradation products to crayfish is important to know for the possible future registration of BUTTE® in Louisiana.

WE208 Effects of the isoflavones, genistein and daidzein, on Acetylcholinesterase from head of Solea Senegalensis.

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The aquaculture is an important economic activity in our area, and one of the main concerns of this sector is fish feed. One of the proposed alternatives to fish meal in feeds are vegetable proteins, and among plant protein sources, soybean is noteworthy. This plant has flavonoids including the isoflavones daidzein and genistein. Solea senegalensis is a flatfish of high commercial importance both in aquaculture and fisheries in Southern Europe with a wide geographic distribution and availability, and its biological cycle is well known. The aim of this work was to study the effects of the isoflavones, genistein and daidzein, on juveniles (weight 1.23 ±0.41 g) of Solea senegalensis. The 96-h toxicity tests were conducted with continuous aeration and water renewal every 24 hours, at a temperature of 19-20°C and a photoperiod of 12h light/12h dark. Juveniles were exposed to the isoflavones at concentrations of genistein (range 1.25-20 mg/l) and daidzein (range 0.625-10 mg/l), plus an untreated control and a solvent control (DMSO). Mortality was recorded and fish head acetylcholinesterase (ACHE) was measured with acetylthiocholine as a substrate after inhibiting butyrylcholinesterase with iso-OMPA. No mortality was observed within the period of the test when the fishes were exposed up to 20 mg/l genistein and 10 mg/l daidzein. Besides, head ACHE activity was not altered in fish exposed to genistein, but daidzein was found to enhance ACHE activity at a concentration equal or higher than 2.5 mg/l after 4 days of exposure. Acknowledgements: Authors are grateful to the Laboratory of Marine Culture at University of Cádiz for providing the experimental fish used in this study. This work was supported by National R&D&I Plan Ministry of Economy, Industry and Competitiveness (Project: AGL2014-52906-R) and in part by the Andalusian Plan for Research, Development and Innovation (PAIDI group: RNM-345).

Luminescent biomonitoring via bioassays of different complexity - from cells trough enzyme reactions to proteins (P)

WE209 Comparison between results of LumiMARA and Microtox tests

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In a regulatory context of ever increasing environmental responsibility (OSPAR convention, BREF CWW), there is a need to have biomonitoring tools to evaluate waste water quality. To date, and for several decades, the standard toxicity testing tool used for rapid analysis of waste water has been Microtox®. However, recently a new tool has become available: LumiMARA®, an acute ecotoxicity bioassay which measures the inhibition of luminescence on bacteria in a similar way to Microtox®. It is mainly advantageous in low-light conditions and in the presence of numerous bacterial species (all in all 11 species, 9 of which are marine and include Vibrio fischeri, plus 2 freshwater bacteria) against only one for Microtox® (Vibrio fischeri). Using a set of narcotic substances with different hydrophobicities and two mixtures (one home-made formulation comprising equimolar concentrations of four of the narcotics tested and one petroleum based complex substance) a comparison of both tools was realised with the aim to determine whether LumiMARA® is the better tool to evaluate effects on bacterial species. While it was recognised that the substances tested would not be directly comparable to whole effluents, this method was felt to be appropriate to compare the two assays. The in-built EC50 calculation software were also verified. Some errors were detected with the LumiMARA® effects calculation software, and these were recalculated by hand using Rotor squadron software, when possible. We observed that Microtox® is more sensitive than LumiMARA® and that in the latter test, freshwater bacteria toxicity is generally lower than that of marine bacteria. This suggests that there may be a small but real difference between freshwater and marine bacteria toxicity however this cannot be concluded on the basis of this research. Moreover, Vibrio fischeri is the bacterium that usually gives the lowest EC50 compared to other bacteria. Thus, the marine bacterium Vibrio fischeri classically used to determine the biotic effect
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 (C60, C80), 2,3,5,6-tetrafluorobenzene, C6H4(NO2)2, and humic substances (HS) are used here as bioactive compounds. Fullerene derivatives 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 effect of organic (1,4-benzoquinone) and inorganic (K2Fe(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±10-4 M and 10-4 M for bacterial and enzymatic assays, respectively, while the EC50 values of K2Fe(CN)6 were 10-4 M and 2-10-4 M. Also we studied the influence of bioactive compounds on the assays. They suppressed bioluminescence of the bacterial and enzymatic systems at concentrations >10-4/g/L and >5±10-3/g/L, respectively. Deterioration 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 test the bioluminescence intensity as the physiological parameter tested. To investigate the sensitivity of the bacteria to the low-dose gamma-radiation exposure (≤250 mGy), the irradiation conditions were varied as follows: bioluminescence intensity was measured at 5, 10, and 20° for 175, 100, and 47 h, respectively, at different dose rates (up to 4100 mGy/h). There was no noticeable effect of gamma-radiation on bioluminescence intensity, while the 20° results revealed a decline in 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 did not demonstrate monotonic dose-effect dependencies. 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 radiotoxicity evaluation under conditions of chronic low-dose gamma irradiation[1]. 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 hydrated fullerene 60 (C60H4F). This assay specifically detects the influence of substances on parameters of the soluble or immobilised coupled enzyme system of luminous bacteria: NADP(+) / FMN-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 bioluminescence intensity 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 > C60H4F. 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 immobilised enzyme system was less sensitive to C60H4F 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

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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 pollution effects. Fluorescence of chlorophyll is widely used for quick assessment of photosynthesis condition. A prompt fluorescence that can be registered by PAM fluorimeters 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 fluorimeter which automatically measures the relative indicator of delayed fluorescence (RIDF) in 24 plant samples. Simultaneously with RIDF, 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 times 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 RIDF up to 5 times. Our method also showed a high sensitivity in biosassay of toxicity of natural and waste waters where Chlorella vulgaris alga was used as a model organism. RIDF of DF of photosynthesis 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. Gaevsky, Siberian Federal University

The main regulating factor for the transition of plants from active vegetative to winter dormancy is the change in the duration of daylight. However, the temperature factor and air pollution also have a significant influence both during the autumn photoperiod reaction and at different phases of winter dormancy. This research aims towards 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 photosynthesis into the phase of winter dormancy, the photosynthetic apparatus of coniferous trees 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 for quick assessment of photosynthesis condition. A prompt fluorescence that can be registered by PAM fluorimeters 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 fluorimeter which automatically measures the relative indicator of delayed fluorescence (RIDF) in 24 plant samples. Simultaneously with RIDF, 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 times 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 RIDF up to 5 times. Our method also showed a high sensitivity in biosassay of toxicity of natural and waste waters where Chlorella vulgaris alga was used as a model organism. RIDF of DF of photosynthesis 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.
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 the age of needles, the depth of winter dormancy of both species clearly correlates with air pressure and the trees growing in industrial zones 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

Luminescent microscopy in the bioindication of the Baikal pollution with oil products and polycyclic 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 a luminescent microscope can be used. It has been experimentally revealed that many pollutants of water bodies, it is necessary to isolate oil products and polycyclic 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 biotesting areas is 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. Eucapitum baikalensis Sars (Copepoda, Copepodea) endemic of Lake Baikal - dwells practically in the entire water column of the pelagic lake. E. baikalensis accounts for up to 70% of the total biomass of zooplankton. Crustaceans Copepoda, having fat inclusions, accumulate them in oil products. This can, in particular, be observed by the blue-violet glow in a luminescent microscope. The accumulation of diesel fuel by fatty inclusions of Copepoda crustaceans and the high sensitivity of luminescence microscopy makes it possible to provide indication 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 luminescent microscope. It has been experimentally revealed that 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 7cogepoda crustaceans in fat drops was proposed.

WE216

The correlation between fluorescent properties of water extract from soil and its effect on bioluminescent enzymatic bioassay

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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 properties of the soils or/and the level of their contamination was under consideration. The research was aimed at the assessment of the luminescent 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 mg/kg were recorded and their stages were determined as follows – absence of effect, activation, and inhibition for genetic activity. The absorption spectra at different stages of the bioluminescent kinetics ensuring that the doses accumulated by the samples were close or a little higher than a tentative limit of a low-dose interval: 0.10 and 0.85 Gy for 241Am, or 0.11 and 0.18 Gy for 137Cs. The 16S ribosomal RNA gene was chosen as a target one for sequence analysis aimed to test whether low dose radiation triggers any alterations in this universal throughout bacterial world and probably an important gene. The sequencing was performed for each sample with the use of the primers 27F and 1522R. Nucleotide sequences of target DNA fragments were obtained and compared in bacteria exposed to 241Am or HTO and control bacterial suspension not exposed to radiation. All compared gene sequences were discovered to be identical which does not indicate any occurrences of mutation events in the analyzed gene under the applied conditions of low-dose alpha and beta radiation inducing changes in bacterial luminescence. Previous results on bacterial DNA exposed to low-dose gamma radiation (0.25 Gy) were analyzed and compared to those for alpha and beta irradiation. It is concluded that bioluminescence activation and/or inhibition under the applied conditions of low-dose alpha, beta and gamma radioactive exposure is...
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: halauuxifen-methyl (Arylex™ active)
C. J. Viggiano, S. Cavana, Dow AgroSciences Italia srl; A. Haley, Dow AgroSciences Ltd; W. Jones, DuPont

The need and 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 production 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 azaic herbicide, halauuxifen-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 halauuxifen-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 halauuxifen-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
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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 Kirah, North Lebanon (34°31'N, 35°0'E) from the raw material phase until the end-of-life phase through a Life Cycle Assessment (LCA); and (2) compare the environmental impacts of an EGR mock-up to a traditional gravel ballasted roof (TGR) 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 rainfall through their growing matrix, 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. Haas, Leitat Technological Center; 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 comparison of the costs and environmental impacts of 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-García, L. Lijó, Universidad de Santiago de Compostela / Chemical Engineering; A. Núñez, L. González Louro, FEGAMP - 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 based on the social and economic well-being of its people can only be sustained 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 system under 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 unsustainability 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). Acknowledgements This work was financially supported by the Spanish Ministry of Economy and Competitiveness (project ref. CTQ2016-75316-P) and by Xunta de Galicia (project ref. ED431F 2016/001). D. González-García would like to express her gratitude to the Spanish Ministry of Economy and Competitivity 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 Horizont 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, acquisition and end of life sequence, 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 and indicators that should be included in a road - or other - operational decision-making tools are essential to help authorities to accept them. In this regard, LCA has become popular in pavement engineering but there is still a lack of detailing, consensus (especially in terms of system boundaries) and reliable data. Beyond ISO 14040:2006, ISO 14044:2006, ISO/TS 14067:2013, EN 15980:2012 and GHG Protocol 2013, there is no specific methodology for selecting the parameters and activities that should be included in either asphalt or asphalt mixtures LCA. 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 at highlighting 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 tourist facilities: DemEAUmed solution A. Claret, C. Hidalgo, Leitat Technological Center / Sustainability Division; S. Vermeulen, A. JIMÉNEZ DEL BARCO CARRION, THE UNIVERSITY OF NOTTINGHAM; T. MEAUTE, 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 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; D. Van Hulle, KU Leuven / Environmental Engineering; C. Hidalgo, Leitat Technological Center; G. Sauve, KU Leuven / Materials Engineering; K. Van Acker, KU Leuven / Materials Engineering. Life Cycle Assessment (LCA) is a commonly used tool to assess 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 and other decision-making tools are essential to help authorities to accept them. In this regards, metal speciation is considered an important aspect to include in the estimation of the emission potential, as the emissions and eco-toxicological 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. However, many uncertainties remain related to 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. Ramos, INEGI / INEGI; A. 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 evaluated through a life cycle analysis (LCA) performed and compared so as 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, 1tonne of debris saving up to 1.3 million kg of resources and materials, while environmental indicators such as global warming potential (GWP), eutrophication potential and ecosystem quality impact (EEQ) depicted enhanced results. Regular gasification uses higher temperatures, thermally decomposing waste and originating 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, using 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.

Y. KURAHARA, N. Isuigo, 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 action in the most significant economic, environmental, 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). Several GGRT have been proposed: direct burial or the United Kingdom (GB). To carry out the life cycle impact assessment (LCA) 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 CCA-LC tool
Tomasi-Montenegro, KIT, Karlsruhe Institute for Technology; M. Weil, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis ITAS
Tomasi-Montenegro, C a, Weil, M a, b, c, HHI, Helmholz-Institute Ulin, 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, 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 (ERB) or the United Kingdom (GB). To carry out the life cycle impact assessment (LCIA) phase, the method that has been considered is LCID 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.

WE231 Development of Environment Hotspots of Analysis and the consideration of availability of eco-labeling program
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 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 (ERB) 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.

WE232 Development of Environment Hotspots of Analysis and the consideration of availability of eco-labeling program

P. Goglio, Carnfield University / School of Water, Energy and Environment; A.G. Williams, N. Balta-Ozkaz, 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 (CETRE)
Climate change targets could only be achieved with the contribution of greenhouse gas (GHG) mitigation in several GGR technologies. This study presents some methodological approaches for LCA of GGR technologies and discusses their strengths and weaknesses. The preliminary comparison and development of approaches was based on a bibliographic review combined with expert discussions. The approaches have been classified according to their completeness, uncertainty and complexity. Several approaches were discussed: comparing LCA with climate scenarios from integrated assessment model IAM (IAM) used to assess future trajectories for GHG and climate change; comparing socio-political factors with consequential LCA or using agent-based modelling for the socio-political factors affecting the choice of results from the equilibrium
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, both approaches require 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 comprehensive 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. Espí Gallart, Fundació CTM, Centre Tecnològic; J. Benzaúa, L. Vendrell, Fundació CTM Centre; F. Clarens, Fundació CTM Centre Tecnològic
More often, methodologies to assess socioeconomic impact are focused just on determining just a few indicators instead impacts, which don’t use to cover the whole spectrum of socioeconomic insights. In this sense, the hybrid fulfilment-importance matrix emerges with the aim to solve these limitations and to capture the socioecosystem. As a case study, this methodology has been applied to LIFE RELEACH project, which is aimed at managing leachates coming from landfill by producing the thermoplastic. The paper may contribute to the eco-design of a new bio-based product using a renewable residue from the horticulture industry 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 biomarkers (P)

WE239 Geostatistically estimating spatial structures of heavy metals and nitrogen accumulation in mosses sampled between 1990 and 2015 thorough Germany and Iceland
S. Dierking, W. Schröder, University of Vechta / 2
Mosses are used to spatially complement the collection of atmospheric deposition by technical samplers and to validate deposition modelling results. Since 1990, the Environmental Moss Survey has been providing data on element concentrations in moss every five years at up to 7300 sampling sites. In the moss specimens, 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 pre-condition to spatially join the moss survey data with data collected at other locations within different environmental networks. The case study maps reveal a clear and statistically 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.


WE240 Semi-volatile organic contaminants (SVOCs) in pine needles from Iceland in the recent past
M. Moe, J.A. Silva, S. Ramos, LEPABE - University of Porto / H. Ingerason, T. Eyfsteinsson, T. Jónsson, A. Sigurgeirsson, Icelandic Forest Research; N. Ratolo, Faculty of Engineering - University of Porto / Laboratory for Process Engineering Environment, Biotechnology and Energy
Iceland is famous for its great number of things, but vegetation (particularly forestry) is not one of them. However, trees do exist in this country and pine stands and recently widespread are a direct result of reforestation efforts. In addition, of these sites it was possible to collect needles from more than one species, allowing a comparison between their respective uptake abilities for SVOCs. In this work the levels of polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), polbrominated diphenyl ethers (PBDEs), organochlorine pesticides (OCPs) and musks were analysed and allowed an original description of the status of the art of these contaminants in this remote location. Acknowledgements: This work was the result of the project: (i) POCI-01-0145-FEDER-069393 (LEPABE – UID/EQUI/00511/2013) funded by the European Regional
The use of lichens as biomonitors of air quality is inexpensive and effective. The association between mercury and lichens is stable over a one year period with broad spatial resolution was the initial focus and the use of naturally occurring epiphytic lichens can be costly, whereas the use of naturally occurring epiphytic lichens can be effective tool for monitoring air quality as it is inexpensive. Therefore, the use of lichens as biomonitors of air quality is an effective and inexpensive approach. In the present study, we investigated the use of lichens to monitor air pollution in a region with high levels of air pollution. The results suggest that lichens can be used as effective biomonitors of air pollution, providing valuable information about the sources and distribution of pollutants in the environment.
the main 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
F. Jouni, Hokkaido University / Graduate School of Veterinary Medicine; J. Yabe, The University of Zambia / Veterinary Medicine, Paraclinical Studies; S.M. Nakayama, Hokkaido University / Graduate School of Veterinary Medicine; Y.B. Yohannes, Hokkaido University / Laboratory of Toxicology; K.M. Muzanda, University of Zambia; H. Nakaoka, Hokkaido University / Environmental Veterinary Sciences; Y. Ikenaka, Hokkaido University / Graduate School of Veterinary Medicine; H. Nakata, Hokkaido University; R. Downling, J. Caravans, Pure Earth; M. Iishizuka, Hokkaido University / Graduate School of Veterinary Medicine

Lead (Pb) toxicity on both 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 2.24 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 distances of dog movements per day was 17.6 km. There was no significant relationship between distances of dog movements per day and the gap of BLLs in dogs 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 BLLs and the distance between the mining area and dogs’ 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
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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 with steamed sweet potatoes at 25°C. The earthworms used in this study were sexually developed with an average body weight of 100 to 200 mg. The artificial soils were composed of industrial sand (70%, 50 to 100 micron particle), kaolin (20%, pH 4.5 to 7.0), and peat (10%). After mixing the components, pH was set in a range of 6.0 to 6.5. At least five diluted serial solutions were used to determine LC₅₀ values, whereas pure acetone was used in the control group. LC₅₀ values of sulfuric acid, methanol, methylhexylketone, nitric acid, formic acid, and toluene were 1.41, 5.71, 2.16, 1.76, 1.24, and 2.86 g/kg soil, respectively. These results are very different from the acute toxicities using filter papers, which toluene exhibited 26-fold lower acute toxicity than sulfuric acid, the strongest toxicant among the tested chemicals. Using the filter papers, methanol and methylhexylketone did not possess a negative effects on the earthworm. With these results, earthworms may act differently to the chemical incidents in relation to their residential condition when they expose to the chemicals.

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. Here, two endogeic and abundant species in the agroecosystem (Allotomophora chlorotica and Aporrectodea caliginosa) were inoculated 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 among and between A. chlorotica and A. caliginosa, 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 OP sensitivity. However, an in vitro inhibition trial with ethyl parathion evidenced a higher sensitivity of A. caliginosa AChE activity compared with that of A. chlorotica, 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 vermicompost tannery wastes
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Untreated waste water 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, mainly in 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 tannery waste transport from the vermicompost 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 begging and post-harvest), roots, stalk, leaves, and mature fruits. Cr (III) and Cr (VI) were determined through graphite furnace atomic absorption spectroscopy (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 accumulation decreased following the increase of the leaf. 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 organophosphate insecticide.
A. Le Navenant, Equipe Ecologie de la Production Intégrée, Site Agroparc; Y. Capowiez, INRA

Insecticide resistance in the natural enemy F. auricularia: detoxification pathways and sensitivity of acetylcholinesterase to organophosphate insecticide. A. Le Navenant, Equipe Ecologie de la Production Intégrée, Site Agroparc; Y. Capowiez, INRA

Forficula auricularia is a common insect in the European context of reduced pesticide use thanks to mating disruption and sexual pheromone. In the last 10 years, the insecticide resistance has been investigated for a better understanding of pesticide impact at population level. Forficula auricularia is known to be a generalist predator. The threshold activities of enzymes that belong to detoxification pathways involved in insecticide resistance have been investigated for a better understanding of pesticide impact at population level. Forficula auricularia is known to be a generalist predator. The threshold activities of enzymes that belong to detoxification pathways involved in insecticide resistance have been investigated for a better understanding of pesticide impact at population level.
disruption practice), and organic ones. Two frequently involved in pesticides resistance enzyme families: Glutathione-S-transferases (GST) and Carboxylesterases (CcEs) were studied, by measuring their activities on earwigs 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 their origin, with higher mortality in earwigs reared in contaminated orchards. 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 CcEs. Moreover, we observed that basal-activities of CcEs 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 CcEs 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, and 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 (DDTs), 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 metamorphosis type, collected from an e-waste pollution region in South China. Stable isotopic analysis showed grasshopper have the highest δ13C value, indicating a C4-plant based food source. In contrast, the butterfly, moth, and litchi stinkbug all represent a C3-based diet preference characterized by lower δ13C values. Moreover, enrichment of the heavy N isotopes 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 perfluorooctanesulfonate (PFOS) in the dragonfly and litchi stinkbug. The results of PCA analysis showed differential enrichment and redistribution of contaminants during metamorphosis, which can be used as a useful tool to assess the bioaccumulation process in different species of insects.

**WE252**  
**Glyphosate: toxic or not toxic, this is the question.**  
M. Verderame, R. Scudiero, University Federico II / Department of Biology Insect pest management: biological and toxicological impact of the insecticidal bioactive compound glyphosate (GH). 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 “no carcinogenic to humans”, converging evidence suggests that GBHs, such as Roundup (Monsanto), pose serious health risk on non-target wildlife. Many studies demonstrate that GBHs threaten the reproduction environmental pollution. Adult *P. sicula* specimens were divided in 3 groups (n=6): group 1 and 2 were exposed to pure Gly 0.1 and 1 μg/L respectively, via gavage every other day for 3 weeks; group 3 received by gavage the same dose of tap water (100μl). The results demonstrate that both Gly doses are toxic for the liver that shows an increase of melanocytes degranulation and the appearance of nodular/cystic formations mainly consisting of collagen fibers, typical of hepatic fibrosis. The liver of Gly-treated males also displays the biosynthetic alterations typical of an estrogenic contamination: hepatocytes, in fact, contain transcripts for both vitellogenin and estrogen receptors. At reproductive level, male gonad is affected by the treatment. Spermatogenesis is slightly slower, at low dose of Gly scattered spermatocytes II luse a cone-shaped arrangement, at high dose the number of rosettes increases and spermatozoids are dispersed in the lumen of the tubules. Alterations in the expression of estrogen and androgen receptors and aromatase are also detected. Interestingly, in females, the ovary is not affected by Gly exposure, no matter the dose. Our results suggest that Gly exposure in a terrestrial vertebrate commonly inhabiting the fields potentially exposed to GBHs causes tissue toxicity, with possible serious health implications for wild and breeding animals as well as human populations.

**WE253**  
**Concentration of perfluoralkyl substances decreases according to the laying order in the yolk of yellow-legged gull eggs (Larus michahellis)**  
C. De Filippi, University of Milano; M. Mazzon, University of Insubria, DISTA / Water Research Institute; B. De Felice, Università degli Studi di Milano; F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNIR; S. Polese, Water Research Institute- CNR / Water Research Institute; N. Sinno, University of Milano; M. Parolini, University of Milan / Department of Environmental Science and Policy, S. Valsecchi, Water Research Institute, Italian National Research Council IRSA-CNIR; M. Vedolin, University of Milano; T. H. Trevizani, Universidade de Sao Paulo / Instituto Oceanográfico; M. Petti, University of São Paulo / Departamento de Biologia Invertebrada; R. C. Filgueira, University of Sao Paulo / Institute of Oceanography The crab *Messomioecetes minuans* 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 conventional methods to determine the bioavailability of contaminants 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 e Zn) in different tissues (muscle, hepatopancreas and gills) of *L. michahellis* and compare populations from contaminated and noncontaminated areas. Samples were collected in two mangrove areas in Sáo Paulo State: Marambaia, a highly contaminated area, and Campinas, 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. cruenta* to these contaminants. In general, the hierarchical pattern of metals concentration in organs was represented by gills > hepatopancreas > 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 concluded 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 the 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 chenisinis)

X. Liao, Guangzhou Institute of Geochemistry / State Key Lab. Organic Geochem.; L. Bai, Guangzhou Institute of 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 has been observed in oviparous species, i.e. fish, bird and frog. Few studies are focus on viviparous species, but ovoviviparous species have not yet been studied. It is known that watersnake *(Enhydris chenisinis)* was ovoviviparous species. Their fertilized eggs develop into new individuals in the maternal body before producing offspring. The source of contaminants in watersnake eggs mainly derived from maternal tissues. In order to fully elucidate the deposition of contaminants in eggs, Firstly, the lipid-normalized concentration ratios of egg to muscle (EMR) were usually used to assess maternal transfer efficiency of contaminants in oviparous organisms. Secondly, due to relatively high lipid and weight of egg in watersnake, 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, PBB, 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 DBDE, 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 *K*<sub>OW</sub> of the chemicals for the watersnake. For compounds with high hydrophobicity (log *K*<sub>OW</sub> > 8), a negative relationship between EMER and log *K*<sub>OW</sub> is observed (*p* < 0.01). While for compounds with log *K*<sub>OW</sub> < 8, the values of EMER and EMER showed significant correlation (*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-EI-MS

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The ultra-fast 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. Dechlorane 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 chemical substance groups from Dechloran Plus and other dechloranes to brominated benzenes and alkyl benzenes, ethers and esters (TBA, ATE, BATE, PBT, PBVE, HBBz, DPTE, BEHTBP, EHTBB, BTBPE, Dec602, Dec603, Dec604, DPMa, Cl10-antiDP, Cl11-antiDP, syn-DF, anti-DF, DBDE), the analytes cover different chemical substance groups from Dechloran Plus and other dechloranes to brominated benzenes and alkyl benzenes, ethers and esters (TBA, ATE, BATE, PBT, PBVE, HBBz, DPTE, BEHTBP, EHTBB, BTBPE, Dec602, Dec603, Dec604, DPMa, Cl10-antiDP, Cl11-antiDP, syn-DF, anti-DF, DBDE). In this way, it gives an analytical basis for further extension towards other compounds. We will show details of different analytical aspects of the method, 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 handprint

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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 not go through life costing methods and is built on the principle that reducing one’s own footprint is not a handprint. Instead, the handprint comes through improvements caused in the performance of another actor. The most fundamental parts of defining the carbon handprint 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 unnecessary 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 handprint 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 to life cycle stage. The footprints are based on the baseline solution, the new solution and the 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

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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 surging 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 indicators and reveal 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 that these transformations reduce the ecosystem service value of the transformed natural land in the life cycle upstream 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 the 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.

**WE259**

Recent advances in natural capital accounting

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At the recent World Forum on Natural Capital (27-28 November 2017) a wide range of corporations, investors and policy makers came together 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 natural capital?” (Cohen et al, 2017) have highlighted the 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 to quantify and communicate these impacts that are also called handprints. This paper demonstrates situations where different approach for 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 interdependencies between ‘capitals’ as most cost-benefit analyses used in everyday decision 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 regimes based on scientifically informed political decisions.

WE260 A Life Cycle Costing and Analysis of a Hybrid-Electric Engine

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The sustainability of hybrid-electric vehicles (HEVs) has been called into question (Bailey et al., 2017, Hickman, 2012, Publishers, 2008, Biello, 2016). The current 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 sustainability and profitability of a HEV motor. This study computes total life cycle costs of a HEV motor. The analyzes considered capital and operating costs in order to define the total vehicle cost of ownership over 10, 15, and 20 year life expectancy. 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., Mangeri, N., & Van Acker, K. 2017. Sustainability of Permanent Rare Earth Magnet Motors in (H)EV Industry. Journal of Sustainable Metallurgy, 3, 611-626. Bailey, G. 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. 8, 2008. Electric Hybrid Vehicles Not As Green As They Are Painted (On Content Online). Inderscience PublishersAvailable: www.sciencedaily.com/releases/2008/02/080207043431.htm [Accessed November 27, 2017].

WE261 Developing a National Food Inventory to estimate the Carbon Footprint of the diet of an average Spanish citizen. Future requirements and policy recommendations

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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 consider 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 foreseen rate and the per capita GDP of considered countries. Moreover, the damage factor of PM 2.5 have been divided in three different damage factors taking into account the different population density of urban, suburban and rural areas. The complexity of production processes and products combined with an increasing dynamic competitiveness environment 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 or upstream of fossil fuel). 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 environmental impacts. The project have been modified taking into account the recent environmental impacts, to be able to aggregate them with the economic costs. Responding to SCORE LCA’s enquiring about LCC and its complementarity with LCA, the purpose of this study is to provide the keys to understanding LCC, its concepts, its scope of use and its theoretical and conceptual limits. It also presents operational implementation elements such as: current methods of applying the LCC, means available for its implementation and possibilities of coupling it with LCA. In a first part, we present the theoretical bases and the state of LCC practice, i.e. definition and history, usefulness and reasons why the method is still not widely used. The second part presents the different types of LCC and the cost perimeters considered for each of them. The third part presents how to implement LCC, with detailed recommendations to decision makers. The fourth part presents the possibilities of coupling LCC and LCA in practice by first showing the usefulness of such a combination, then by presenting the software that allows coupling methods. We conclude by identifying the necessary work to improve the LCC and LCC coupled with LCA.

WE264 Pizza: it is dangerously delicious!

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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 component of release; the population density of the area where the emission take place; the average level of income of the country in which the emission take place. A complete LCA of an electric, gasoline and petrol VW Golf has been carried out considering city cycle real consumptions from EPA (fueleconomy.gov) and real emissions from national inventory on transport air emission factors (http://www.sinanet.isprambiente.it/it/sia-ispra/fetranspo). The use phase of the vehicles occurs in Italy, the energy used for battery charging is the Italian marginal mix, the vehicles assembly occurs in Germany while batteries are assembled in Austria. The upstream of fossil fuel is consistent with the nowadays actual national import mix. Emissions of PM2.5, PM10, NOx, SOx, NH3, NMVOC, CO2 have been taken into account for externalities evaluations considering that the internal processes where involved, for each LCA phase accounting for more than 2% of the weighed emission of PM2.5, PM10, NOx, SOx, NH3, NMVOC, a specific height of realis and geographical area have been assinged distinguished by: Italy (were car are used and most of electricity produced), Germany (where car are produced), Austria (where battery are assembled) Lybia, Algeria, Holland, Russia (use of upstream of fossil fuel). An Bees Rccle 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 environmental impacts. The project have been modified taking into account the recent environmental impacts, to be able to aggregate them with the economic costs. Responding to SCORE LCA’s enquiring about LCC and its complementarity with LCA, the purpose of this study is to provide the keys to understanding LCC, its concepts, its scope of use and its theoretical and conceptual limits. It also presents operational implementation elements such as: current methods of applying the LCC, means available for its implementation and possibilities of coupling it with LCA. In a first part, we present the theoretical bases and the state of LCC practice, i.e. definition and history, usefulness and reasons why the method is still not widely used. The second part presents the different types of LCC and the cost perimeters considered for each of them. The third part presents how to implement LCC, with detailed recommendations to decision makers. The fourth part presents the possibilities of coupling LCC and LCA in practice by first showing the usefulness of such a combination, then by presenting the software that allows coupling methods. We conclude by identifying the necessary work to improve the LCC and LCC coupled with LCA.
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 benefits and demonstrate 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 outcome-specific incidence rates with risk ratios and severity factors from the Global Burden of Disease, measuring benefits (+) and impacts (-) in avoided \( \text{mDALY}/g \). 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 \( \text{mDALY}/g \). Human health scores for pizzas range from -35 avoided \( \text{mDALY/serving} \) pizza with extra meat to 2 avoided \( \text{mDALY/serving} \) pizza with no cheese. For the environmental impact assessment, global warming estimates varies 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 \( \text{mDALY/serving} \), 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 coherent metric: the more precise FU is to compare protein sources based on their outcome of all food items and diets in LCA. Expanding this approach to various food items could help decision-makers and consumers not only make better comparisons but also identify sustainable food items and adopt sustainable 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
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Proteins, which are made up of amino acids (AA), are essential for human health. Most of AA can be synthesized by the body but 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 of ~14.5% of all human caused greenhouse gas emissions but also requires large amount of land and water due to the high quantity of crops required for feeding animal. Consequently, comparing protein sources using Life cycle assessment (LCA) is important for decision-making. However, choosing the functional unit (FU) is often a critical issue for food systems. The quantity of food (i.e. 1 kg) is the most used FU currently. Nevertheless, this FU does not represent the function of food that is to provide nutrients. A more precise FU is to compare protein sources based on their protein content (i.e. 1 kg protein). To have a more holistic approach, nutritional and qualitative aspects should also be included in the FU. Actually, most plant protein sources do not bring all the EAA required. In this study, the Protein Quality Index (PQI) developed by Sonesson (Sonesson et al., 2016) was applied as a FU. It takes into account aspects such as EAA digestibility, AA requirements but also food habits. In our study, PQI was conducted on several protein sources: conventional (pork, chicken meat, salmon and tofu) and non-conventional sources (insects and algae). The role of the AA supplementation in animal feed was also investigated. On the one side, the analysis has shown that non-conventional protein sources perform better in all environmental categories, independently of the choice of the functional unit. Tofu performs better than animal protein but the difference between animal and vegetable based proteins becomes much lower when a more elaborated FU is used. On the other side, the supplementation in AA allows a reduction of the environmental impact of chicken and pork. Using the PQI as a FU, the impact of chicken and pork with AA supplementation is even lower than the one of tofu in some categories. Using the PQI as a FU is a step toward a more holistic assessment. A next step might be to include other nutrients such as iron and vitamins in the FU.

WE266
The ISO/DIS 14008 standard: Monetary valuation of environmental impacts and related environmental aspects – Principles, requirements and guidelines - an overview
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Key words: monetary valuation, framework, standard, ISO 14008 The use of monetised environmental impacts and associated substances has substantially increased in the last couple of years. This trend is mirrored since 2016 by efforts at ISO level to develop standards on environmental costs and benefits. This contribution will present the result of the work achieved in ISO/TC 207/SC 1/WG 7, developing ISO 14008 whose current title is “Monetary valuation of environmental impacts and related environmental aspects — Principles, requirements and guidelines”. The work started in February 2016. After five WG meetings, ISO 14008 has reached the Draft International Standard (DIS) stage in fall 2017. Many organizations have experience in assessing environmental aspects and related environmental impacts resulting from their activities in physical units (e.g. tons of CO2 emitted or numbers of disability adjusted life years, DALYs). To further integrate this information into the decision making process, it is useful to name the monetary impacts and also of related environmental aspects. Monetary valuations enable comparisons and trade-offs between different environmental issues. The aim of this standard is to increase the awareness, understanding, comparability and transparency of monetary valuation of environmental impacts and related environmental aspects. To achieve this purpose, standardised and transparent documentation of the methods used to derive monetary values is essential. The multiplicity of monetary values, methods to determine monetary values, and ethical perspectives on money, requires careful consideration and prudent communication. ISO/DIS 14008 provides a framework that includes principles, requirements and guidance for monetary valuation of environmental impacts and related environmental aspects. Many methodological requirements or recommendations are intended for persons assessing monetary values. Following these requirements and recommendations enables good practice. The requirements in the reporting clause assist the user of monetary values in assessing the quality of the monetary valuation study. The presentation will give an overview of the ISO/DIS 14008 document.
 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 lost in the trade-offs. A 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. Henrique, 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 Et 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-sequencing molecular method (BOX-PCR). Results of growth showed that long-term exposure to NaCl slightly increased the tolerance of *E. toletana* to this salt, EtC₅₀ for growth were: 22.5 g/L (8.64-36.4) for Et-LB; 30.3 g/L (23.2-37.4) 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 in saline ecosystems: when the low salinity cannot be an advantage but an impact J. Alvarado-Rojas, BBVA Bancomer Superficie Ingeniería Agroústica. Universidad Politécnica de Cartagena / Ciencia y Tecnología Agraria: F. Jiménez-Cárceles, BIOCYMA, Consulting in Environment and Quality, S.L. Murcia, Spain.; M. González-Alcaraz, Department of Biology & CESAM - University of Aveiro / Biología / CESAM

Salinity is widely considered one of the most detrimental factors of soil and water degradation worldwide. Anthropogenic salinization due to the use of low quality water and/or 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 flow from areas with intense agriculture. This water led to an increase of flooding periods, a decrease of soil salinity in the most saline sites and an increase 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? B. J. Jefford, J. Reich, J. Bray, University of Canberra / Institute for Applied Ecology

The effects of 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 microbe) 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 S. Bogart, University of Lethbridge / Department of Biological Sciences; E. Stock, University of Lethbridge; A. Manek, University of Saskatchewan; A. Tillmanns, C. S. Bogart, British Columbia Ministry of Environment & Climate Change Strategy; G.G. Pyle, University of Lethbridge / Biological Sciences

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 their mixtures. Water hardness can be increased to 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 data were collected for a range of fish species, and 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, phosphorus and micropollutants) 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 taxa response was tested 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, who were associated with both the highest and lowest environmental quality (USA) that should not be exceeded to achieve good ecological status according to the water framework directive. Thus, lowering of oxygenation in the salinity gradient 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 amongst 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 mussels can live for decades, we used the 300th percentile of the proposed receiving stream data: salinity and associated ions 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.

WE275
LIFE LAGOON REFINISH - Coastal lagoon habitat (1150*) and species recovery by restoring the salt gradient increasing fresh water input. Management measures in the northern Venice Lagoon (NE, Italy) F. Cacciarelli, ISPRA-Institute for Environmental Protection and Research / Mitigation and Prevention of Impacts; A. Bonometto, A. Feola, E. Ponis, ISPRA Institute for Environmental Protection and Research; A. Sfriso, University Ca Foscari of Venice; B. Matticchio, IPROS; M. Lizier, Regione del Veneto; V. Volpe, Provveditorato OO. PP. Veneto, Trentino Alto Adige e Friuli Venezia Giulia; M. Ferla, R. Boscolo Brusà, ISPRA - Institute for Environmental Protection and Research

The northern Venice Lagoon (SCI IT3250031) 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 IT3250031, but it is still unfavourable in the inner landward area due to lack of ecological gradients, favouring self-regulation processes, between lagoon and mainland. In the past, the project area was occupied by reedbeds in large amounts, now significantly reeded 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, is minimised. The LIFE LAGOON REFINISH 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 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 serially-cladified experiments and synthetic solutions (e.g. NaCl/CaCl2) of systatic salinities. There were different growth responses to the wastewater and saline concentrations among both plant species. F. rubra was exhibited a significant higher survival percentage than T. pratense. After four weeks of exposure, T. pratense exhibited greater sensitivity and lethality. Interestingly, biomasses of both plants were greater from industrial wastewater than the comparable brine solution. Furthermore, the study presents the challenge to verify the ecological effects of the Cocktail of industrial wastewater containing a variety of inorganic and organic substances that may have triggered plant survival and salt-tolerance. F. rubra grew under salts stress, and presented a mechanism to crystalize salt on their leaves. Hence, plant uptake, under certain conditions, may be promoted as an alternative treatment for high salt concentrations.

WE276
Comparing the growth of fescue and clover plants in petroleum industrial effluents and solutions of similar salinity P. Srikhunsuk, 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 increasing in volume and toxicity to the environment, particularly in regards 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 8 weeks under hydropic conditions to compare their growth in brine effluents from tertiary recovery operations. Experiments further compared similarly-diluted effluents and synthetic solutions (e.g., NaCl/CaCl2) of systatic 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 greater sensitivity and lethality. Interestingly, biomasses of both plants were greater from industrial wastewater than the comparable brine solution. Furthermore, the study presents the challenge to verify the ecological effects of the Cocktail of industrial wastewater containing a variety of inorganic and organic substances that may have triggered plant survival and salt-tolerance. F. rubra grew under salts stress, and presented a mechanism to crystalize salt on their leaves. Hence, plant uptake, under certain conditions, may be promoted as an alternative treatment for high salt concentrations.

WE277
Contribution to the salinization risk assessment, under drought conditions, in the Alqueva irrigation area (South Portugal) A. Tomaz, S. Fialho, A. Lima, Instituto Politécnico de Beja; A. Penha, H. Novais, M. Potes, M. Iakunin, G. Rodrigues, Instituto de Ciências da Terra; P. Alvarenga, L. Ferreira, Instituto de Investigação Agrária, UEFIFUT; M. Moreira, Instituto Superior de Agronomia, Universidade de Lisboa; M. Costa, M. Morais, R. Salgado, Instituto de Ciências da Terra; P. Palma, Instituto Politécnico de Beja / Department of Tecnologies and Applied Sciences

In Mediterranean regions, climate changes have enlarged water limitation for crops, leading to an increased demand for irrigation water. During the hydrological years of 2017 and 2018, Portugal suffered a drought that affected throughout the entire mainland territory reaching a severe drought level. Under water scarcity conditions and high atmosphere evaporative demand, the risk of land salinization is one of the main threats to the sustainability of irrigated agriculture. Therefore, it is very important to assess the quality of irrigation water and the risks of salinity for crop production, in order to adopt appropriate management practices in irrigated areas. This study is focused on the salinity risks for the production of the most representative crops grown in the Alqueva irrigation area. This is a large irrigation scheme with a total area of 120 000 ha centered in the Alqueva reservoir. For the purpose of the study, a chemical assessment of some
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 crusting problems, as well as, potential yield reductions in the most sensitive crops of the Alqueva perimeter. Higher ion concentration related with the proximity to water salinization. We selected water salinity and 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 different diet composition macros in the first tier risk assessment. The so-called PD factor (concentration of a diet component), is produced by the partial ingestion rate of various species. The main indicator for calculating the partial ingestion rate is the standard refinement parameters which intend to add realism to higher tier risk assessments. Publicly 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 studies are based on monitored factors, study or samples, or stomach flushing. 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 diet assessment defined diet fractions which have different default residue levels and different residue limits or monovalent 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 / Fincaica y de Fluidos Molecular endpoints are used nowadays under different conditions for inclusion in 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 pathways related to toxic effects in selected 42 of these genes plus six genes used as reference to design an array for Real-Time PCR analysis. Stress response, detoxification mechanisms, endocrine system, or epigenetics were some of the pathways analyzed in the array. In order to validate the toxicological and ecological interest of this approach, individuals were treated with an antibiotic, tetracycline, for seven days or were left starving for 7 and 10 days. Results obtained for these experiments are presented, showing the interest of designing specific arrays to perform more detailed analysis of molecular endpoints that can be related to toxicant mode of action and stress situation. We hope that the methodology presented here can serve as an example for the study of other species in order to improve our knowledge of their biology. This work has been funded by the Ministerio de Economía y Competitividad, CICYT (SPAIN), CTM2015-64915-R.

**WE281 Temperature of transcription of the marine copepod Temora longicornis**

J. Semmoun, Ghent University (UGent) / Animal Sciences and Aquatic Ecology; J. Asselman, Ghent University / Laboratory for Environmental Toxicology and Aquatic Ecol; R. Jansen, Ghent University / Applied Ecology and Environmental Biology; K. De Schampaert, 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 increasing. Understanding the impacts of this changing environmental condition in zooplankton communities is crucial, as alterations in the zooplankton communities can affect 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-sequencing 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 us with new insights on how exposure to increased sea water temperatures may affect the fitness of the most dominant zooplankton species of the southern part of the North Sea.

**WE282 A functional approach to modern endpoints - quantitative assessment of stress gene expression response to a range of copper concentrations in the freshwater mussel Anodonta anatina**

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In the field of ecotoxicology, modern transcriptomics technologies have the potential to improve and complement current toxicity assessment methods and biomonitoring protocols. Early warnings of general stress and specific toxic modes of action could in theory be used as biomarkers of pollutant exposure or adverse effects. However, necessary base level understanding is currently lacking concerning how gene expression may vary under realistic exposure scenarios. Therefore, we adopted an approach of quantitative assessment as an alternative to more descriptive methods. We chose the freshwater mussel Anodonta anatina as our model organism. Being a stationary filter feeder, it shows promise for use in exposure studies under both laboratory and field conditions. Furthermore, it is the most abundant freshwater mussel species in Sweden, and occurs in freshwater ecosystems over most of Europe. For the present study, mussels were collected locally in Vinne å (southern Sweden), on a location free from point source pollution. After two weeks of acclimatization to laboratory conditions, mussels were exposed for 96 h to one of three copper treatments (nominal concentrations of 1, 10 and 100 μg Cu²⁺), or a control treatment (n= 5 per treatment). Using RT-qPCR, relative expression of a selection of general stress genes will be quantified in extracted digestive gland and gill tissue. Preliminary data will be presented, testing the hypotheses that the amplitude (fold-change) of relative expression differs (i) between the copper source and (ii) between genes in the same treatment. Results from this initial experiment will be used to design a follow-up experiment, in order to test the dose-dependence of gene expression responses. Gradually, successively increased exposure scenario complexity (e.g. duration, chemical composition) will help us to better understand how expression patterns potentially vary under environmental exposure. By subsequent incorporation of biochemical and physiological biomarkers, we also aim to link stress gene expression patterns to effects at higher biological levels. Ultimately, a more thorough understanding of natural and pollution-induced variation in gene expression may allow transcriptomics to be usefully and successfully incorporated into various ecotoxicological assessment protocols.

**WE283 Validating a contamination assessment tool from lab to the field: Folosoma candida exposed to a fungicide-based formulation**

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Folosoma 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 toxicology 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 trials, 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 more realistic field scenario, by targeting specific molecular biomarkers retrieved from a previous laboratory study and an extensive molecular and biochemical 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 laboratory conditions, organisms were now exposed under field conditions for 4 days to the same concentration as for laboratory exposure (concentrations up to a 75% reduction 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 defaunated. 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 accimation period to the field by the organisms. RNA was extracted from each pool of organisms using the TRIzol® methodology. According to previous laboratory “omics” results with the same set of specific genes were selected for a targetted 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 such 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, spinosad 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. As expected, the pesticides exposure triggered different responses at the proteome level. Changes caused by spinosad were more noticeable than for indoxacarb exposure. Our results revealed a general decrease in the expression of globin proteins with the increase of spinosad concentration. Additionally, for spinosad, a significant decrease in the expression of an actin and a cuticle protein were also observed. Moreover, correlations between proteomics data and specific modes of action was found for both compounds. Our results suggest that protein profile changes have the potential to be used as early warning biomarkers of pesticide exposure, providing an interpretation of molecular pathways of toxicity behind the organismal response, therefore supporting the risk assessment of pollutants. This work contributes to the growing knowledge of sub-lethal effects of pesticides in invertebrates and their molecular targets. *Chironomus riparius*, a model organism in aquatic toxicology, is also presented as a promising model organism for environmental proteomics.

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**WE286**

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 have been extensively studied as model organisms for laboratory experiments to elucidate the cellular responses of diatoms to Cu toxicity. The freshwater diatom *Tabellaria flocculosa* (TFL), 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, frustulin), antioxidan (SOD, CAT) and detoxifying (GSTs) enzymes activity and low molecular weight antioxidants (GSH). These mechanisms were induced by a higher energy production (ETS activity, use of sugars and lipids). At the highest Cu concentration (10 µg/L), the oxidative 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-palmitoylgllycerol, glycerol and diterpenoid 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/8bh1jzedldq3vmw/graph%20a%20copper.tif?dl=0

**WE287**

Non-targeted approach to identify metabolic perturbations in gilt-head bream liver and brain exposed to benzophenone-3

H. Ziarrusta, L. Mijangos, University of the Basque Country UPV/EHU / Department of Analytical Chemistry; S. Picat, A. Perera, Polytechnic university of Catalonia UPC / ESSAI and control tanks were the control of the experiment. M. Olivaros, O. Zuloaga, University of the Basque Country UPV/EHU / Plentzia Marine Station (PIE-UPV/EHU) & Dep Analytical Chemistry

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 biota. Although some studies reported adverse effects in both invertebrates and fish, 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 then stored at -80ºC 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-Orbitrap MS in positive and negative modes with both C18 and HILIC separation. Metabolites were identified using Compound Discoverer (Thermo) interfaced to MZmine 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 altered during the treatment, a supervised statistical approach was used to discriminate metabolic pathways. Both for liver and brain, multiple metabolites were affected at different time points. A time-series statistical analysis was carried out to identify the major trends (adjusted p-value < 0.05) associated with the interaction between exposure day and animal group (exposed or control). Metabolites driving group separation were further investigated using the Kyoto Encyclopedia for Genes and Genomes (KEGG) in order to determine affected pathways. Overall, these data demonstrate the toxic effect of BP-3 and the involvement of the main biochemical and metabolic processes affected, such as energy metabolism, amino acids and other metabolites. Keywords: 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.

**WE288**

EFFLUENTS FROM PULP AND PAPER MILLS PROMOTE 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 the pulp and paper industry. The experimental design comprised the following: the establishment of embryos 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 28ºC and a light / dark cycle of 12/12 h 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 once every two days 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

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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 alga Chlamydomonas reinhardtii

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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 focusing 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 Chlamydomonas reinhardtii. 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 intersexuality 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 data will be carried out, thereby understanding of the biological functions impaired after amphipods wastewater exposure. A biological pathway analysis will also be 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

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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, applying a toxicological high-throughput genome-wide scaling multi-omics technologies. The approach towards achieving this end was a suite of targeted (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) toxics. To enable the 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 Elucidating 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 μgL) and Pb (5, 25 μgL) 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’, acetate/CFOX and CPT activity, inductions of micronuclei (MN), DNA strand breaks (i.e. Comet assay) and measurement of protein profile (i.e. proteomics using LC-MS). The clearance rate shows that the mussels exposed to all treatments containing 32 μgL of Cu have a significantly decreased feeding capability compared to controls, regardless of singular or binary exposure. This was in contrast to the comet assay results which suggests a general pro-oxidant relationship for copper treatments and the highest binary mixture (32, 25 μgL Cu and Pb, respectively). Mussels exposed to 25 μgL of 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. Zhou, S. He, University of Birmingham / School of Pharmacy & Biomedical Science; D. Butler, Unilever; G. Hodges, Unilever / Safety and Environmental Assurance Centre SEAC; 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 mechanisms prevents accurate predictive risk assessment. Adverse outcome pathway (AOP) discovery for pollution assessment 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 literary 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 toxicokinetic 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 phytoene accumulation was observed, whilst concentration dependent b-carotene depletion was shown at later times following exposure. For a concentration of 0.125 mgL of norflurazon, was identified 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 2000 μgL/L after 24 hours. This study highlighted the necessity for use of synchronous algal cultures for accurately understanding mechanism, as this would enable more accurate determination of time- and concentration- responses due to diurnal algal life cycles. Ultimately, this work has shown proof-of-concept and laid the foundation for development of a quantitative AOP for phytoene desaturase inhibition leading to growth inhibition and population decline.
WE293  
Effects of water-borne benzo[a]pyrene on early life stages of the fathead minnow (Pimephales promelas)  
M.T. Schmitt, RWTH Aachen University; M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; N. Bastu, McGill University / Faculty of Agricultural and Environmental Sciences; H. Hollett, RWTH Aachen University / Institute for Environmental Research; N. Hogan, University of Saskatchewan / Toxicology Centre and Department of Animal and Poultry Science, College of Agriculture and Bioresources; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre; A. Alcaraz, University of Saskatchewan - Toxicology Centre / Toxicology Centre; D. Green, University of Saskatchewan - Toxicology Centre / Toxicology; K. Bluhm, University of Saskatchewan / School of Environment and Sustainability; T. Lane, University of Saskatchewan; N. Baldwin, J. Taghavimehr, A. Masse, University of Saskatchewan - Toxicology Centre / Toxicology Centre; D. Crump, Environment and Climate Change Canada / National Wildlife Research Centre; N. Basu, McGill University / Faculty of Agricultural and Environmental Sciences; H. Hollett, RWTH Aachen University / Institute for Environmental Research; N. Hogan, University of Saskatchewan / Toxicology Centre and Department of Animal and Poultry Science, College of Agriculture and Bioresources; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre.  

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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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 freshwaters. 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 DNA methylation and/or histone modification leading to altered key processes resulting in increased 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 toxicology (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 is known about the effect of warming whether it increased the bioaccumulation of copper nanoparticles in freshwater fish. The purpose of this study was 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°C) for periods of 7 days for uptake and 7 days for depuration, to analyze the accumulation and toxic effects of copper nanoparticle on muscle. Results showed that the copper accumulation in muscle of 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°C group was significantly higher than of 26 and 28°C groups (p<0.01). However, they were 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**

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A review of the existing literature on mixture effects of nanomaterials (NM) and chemicals in environmental organisms was conducted in order to evaluate the current state of knowledge. More than 120 studies were assessed to explore the relationship between changes in contaminant and NM uptake, bioconcentration, and toxicity. The specific aim of the review was to describe more specifically the interactions that have been observed and to classify the most common mechanisms. As the literature evaluation demonstrated, the existing evidence for interference of NM and chemical mixture exposure with uptake and toxicity is rather diverse. Based on the observations made, we could discriminate at least 7 different categories to capture the evidence ranging from no changes in uptake and toxicity to an increase in uptake and toxicity upon mixture exposure to chemicals and NM: (1) increase in accumulation and toxicity (2) increase in accumulation and no change in toxicity (3) increase in accumulation and decrease in toxicity (4) no change in accumulation and toxicity (5) no change in accumulation and decrease in toxicity (6) decrease in accumulation and increase in toxicity. However, we assume that these observations were caused by different underlying mechanisms and processes, hence we developed a process oriented, tiered approach considering (1) Adsorption / Interaction between NM and chemicals, (2) Uptake of NM by organisms, (3) Desorption of chemical inside / outside the organism, and (4) Toxicity. J. Hammers, T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; N. Bianchi, 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

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**WE302 Nano silver based products and environmental challenges: toxicity and accumulation in a marine sentinel species**

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The use of nanoparticles (NP) based commercial 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 marine bivalve sentinel species as 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 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 assayed 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 vivo. Chemical analysis confirm Ag exposure and showed a dose-dependent increase of Ag in exposed mussels. In conclusion we can state that this nano silver-based commercial product can induce toxicity even at low concentrations and in short-term exposure scenarios. The observed toxicity of NanArgen underlines the need to further test commercial formulations of nanotechnology-based consumer products instead of bare nanoparticles in order to properly address any risk associated to their use and release into aquatic environment and in non-target aquatic species.

**WE303 Effect of gold nanoparticles on feeding, growth and enzymes activity of amphibians**

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The gold nanoparticles are widely used in medical therapy and cosmetics. However, the impact of these particles on the environment is barely understood. This study presents a relative abundance of papers focusing on engineered nanoparticles, including both freshwater and saltwater species. The knowledge about the potential ecotoxicity of these nanoparticles is essential before their use by society at a large scale, since they will ultimately be released in the environment. Thus, the aim of this study was to determine the effect of gold nanorods (Au-NR, 45nm) in the feeding rate, growth and enzymatic activity of the European common newt (Triturus vulgaris). A significant decrease in feeding rate and snout to vent length (SVL) of tadpoles was observed at concentration equal or higher than 0.004 μg/ml. For biomass a significant effect was observed at concentration 0.007 μg/ml or higher, though, a decrease in weight gain rate was observed at a lower concentration (0.004 μg/ml). At the biochemical level, the activity of enzyme lactate dehydrogenase (LDH) increased at 0.002 μg/ml of Au-NR, that of catalase (CAT) was significantly reduced at 0.005 μg/ml or higher, and glutathione S-transferase (GST) and acetylcholinesterase activity (ACHE) was significantly higher, relatively to the control, in the two highest tested concentrations 0.007 and 0.01 μg/ml. The
observed reduction in SVL, added to decreased feeding rates, in tadpoles exposed to Au-NP, are important effects that may compromise the fitness of the organisms, since they may cause a delay in the metamorphism, 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-NP) a detoxification pathway induced by Au. Furthermore, an enhancement of reactive oxygen species (ROS) may have led to the inactivation of catalase and the induction of cell death related with the 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**

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The disposal of various plastic nanocomposites containing multiwalled carbon nanotubes (wMWCNT) is anticipated to increase in the next decades. Thus, MWCNT 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. MWCNT undergo thereby structural changes compared to their pristine state, like formation of hydroxyl and carboxyl groups on the surface of the MWCNT. As a consequence, weathered MWCNT (wMWCNT) have an altered agglomeration- and sorption behavior to other contaminants like 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 ‘Trojan 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 concentration (60 µg/L TCC) 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 + 10 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) yields effective 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 caused by 60 µg TCC/L. Regarding to mixture effects of TCC and wMWCNT to algae, bioaccumulation of wMWCNT 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 Project "Q-Transfer" that receives funding from the Bundesministerium für Bildung und Forschung (BMBF) under agreement with the FP7 ERA-NET SIINN.

**WE305 Comparative assessment of the interactive effects of Carbon-based nanoparticles and Benzo(a)pyrene on zebrafish embryos**

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This study aimed to assess the ecotoxicological 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), two carbonaceous materials that are classified as contaminants linked to 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 due to 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 proteomics showed that different stress responses were detected by the two CNMs alone and their combination. 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 NANOPARTICLES ON HEMOCYTES OF MUSSEL Mytilus galloprovincialis**

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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 galloprovincialis. 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⁻¹), dispersed with a probe sonicator, and ZnCl₂ (10 and 25 µg mL⁻¹; 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⁻¹, with maximum values (>50%) of cell death after exposure to ZnO NPs 50 µg mL⁻¹. Furthermore, hemocytes treated with sub-lethal concentrations of ZnO NPs (5-25 µg mL⁻¹), showed a significant increase of O₂⁻, 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 nanoparticles, 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 relevant 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 < 0.05).

**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 AS49 and the Daphnia magna experiments, respectively. In AS49 cells, the incubation of Zn with ha-IONPs did not change the Zn effects on cell viability after 24h in terms of IC₅₀ (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 IC₅₀ for Zn was 0.070 g/L in the presence of 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 microscope images showed that ha-IONPs aggregates were uptake by the cells during the experiments. Therefore, even if adsorbed on ha-IONPs, NMs may 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 EC₅₀ 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 uptake and co-predication 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

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 excellent electrical, mechanical, thermal, and optical 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, Poeciliopsis lucida) 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 essential in order to assess their environmental fate. 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
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The number of engineered nanomaterials (ENMs) is rising continuously in consumer products and industrial fields. Therefore, knowledge about their ecotoxicity in aquatic and soil systems is very important but rare. Nanoscale titanium dioxide (TiO₂) is probably among the most relevant ENMs with a projected accumulation rate in European river sediments of 1.4 mg kg⁻¹ yr⁻¹ (Gottschalk et al., 2009). In experiments of Angelstorf et al. (2014) it was shown that TiO₂ is far more toxic to the nematode Caenorhabditis elegans than bulk TiO₂, especially under simulated solar radiation (SSR), probably a consequence of its photocatalytic property. Further experiments by Samet (2017) focused on the interaction of TiO₂ with cadmium (Cd), another environmental contaminant. C. elegans was exposed to Cd in the presence of CdNPs, another environmental contaminant. C. elegans was exposed to CdNPs (P25, primary particle size of 21 nm) and Cd in single and co-exposure for 72h under dark conditions and SSR. Choosing growth and reproduction as toxic endpoints, co-exposure with 40 μg L⁻¹ CdNPs and 50 μg L⁻¹ Cd under SSR led to a synergistic inhibitory effect of 80% of reproduction, twice as high compared to CdNPs alone. As Cd is known to induce intracellular calcium signaling as part of protective cell processes (Thévenod, 2009), in the study presented here, the effect of the mixture on intracellular calcium release will be investigated applying the following methods: 1) The molecular mechanism of nTiO₂ and Cd will be investigated with NS8593, a known human TRPM7 ion channel blocker. Because of high reproduction inhibition, the TRPM-like channel gene gon-2 could interact with the mixture. Gon-2 is responsible for gonadal cell division in C. elegans. If Cd or a CaNPs capping agent inhibits the blocker, this would show the same effects under SSR. 2) The mode of action of nTiO₂-Cd-agglomerates is still not identified. They could interact if Cd is bound to nTiO₂, or if Cd and nTiO₂ are in close proximity. The impact of nTiO₂-Cd-agglomerates will be examined using calcium as a potential competitive ligand. 3) The photocatalytic activity of nTiO₂ could damage cell membranes under SSR and Cd could enter the cell. Measurements of membrane integrity with propidium iodide and hexokinase will be tested. First results will be presented. Angelstorf et al., 2014. Environ. Toxicol. Chem., 33, 2288-2296. Gottschalk et al., et al., 2009. Environ. Sci. Technol., 43, 9216-9222. Samet, Abstract SETAC Brussels 2017. Thévenod 2009. Toxicol. Appl. Pharmacol., 238, 221-39.

WE311 Influence of temperature and salinity on toxicity of zinc oxide nanoparticle on the marine copepod Tigripus japonicus
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Zinc oxide nanoparticles (ZnO NPs) are ranked as the 5th most prevalent nanomaterials (NMs) in commercial products. About 93% of ZnO-NP-containing products are paints, cleaning and personal-care products, from which ZnO-NP can be easily released. 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 (Tigripus 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 ZnSO₄·7H₂O (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-NPs may exhibit a higher sensitivity (due to phenotypic plasticity mechanisms) to the nanoparticle. The aim of this study was to evaluate the multigenerational effects of gold nanorods to Raphidocelis subcapitata and Chlorella vulgaris
C. Monteiro, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; C. Venâncio, 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. Soares, University of Aveiro / Department of Chemistry / CICECO; L. Leao, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro. In real scenarios, exposure to nanoparticles may occur over several generations, which may exhibit a higher sensitivity (due to the accumulation of adverse effects) or tolerance (due to phenotypic plasticity mechanisms) to the nanoparticle. The aim of this study was to evaluate the multigenerational effects of gold nanorods (Au-NR) ranging from 8 to 90 μg mL⁻¹, for 72h. At the end of the assays, growth rate was computed for all generations of each alga. The following physico-chemical parameters of Au-NR concentrations were monitored: size, morphology and total concentration of Au. In addition, the toxicity of the capping agent
cetyltrimethylammonium bromide (CTAB) was quantified by exposing both algae to the concentration of CTAB present in the highest tested concentration of Au NR: 90 μg/L for *C. vulgaris* corresponding to 0.257 mC of CTAB band 53 μg/L for *R. subcapitata* corresponding to 0.152 mM of CTAB. *Chlorella vulgaris* exhibited a higher tolerance to Au NR than *R. subcapitata*: EC50/72h for *F0* was 79 μg/L and 39 μg/L, respectively. For *C. vulgaris*, a gradual increase of its tolerance to Au NR was observed over generations; after being exposed for four generations to this concentration, there was a significant effect on growth rate were monitored between all concentrations and the control. A different pattern of response was observed for *R. subcapitata*. This species significantly increased its sensitivity to Au NR 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 NR to freshwater microalgae. 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 aquatic 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 these environmental stressors on aquatic species have not been well studied. The purpose of this study is to assess the effects of aquatic acidification and warming combined with copper nanoparticle on survival and hatchability of early development of Japanese medaka (*Oryzias latipes*).Fig 1 shows the embryonic exposure to 25 μg/L of 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, 28°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 hatching 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**

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Nanoparticles (NPs) are widespread used in consumer products and industry; they are showing increasing interaction with the immune system and 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-engineered materials and developing predictive markers of risk vs. safety, with a collaborative international 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 science. The main objective of the analysis is to assess the cellular/molecular immune parameters (e.g. lysesosomal 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 the presence 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 hemocyes* 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 Skłodowska-Curie grant agreement PANDORA No 671881.*

**WE315**

**Influence of warming and acidification on copper nanoparticle bioaccumulation in medaka (Oryzias latipes) embryos**

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The level of atmospheric CO2 has elevated significantly since the Industrial Revolution, leading to global warming and ocean acidification. With the development of industry and technology, many emerging contaminants such as copper nanoparticle (CuNPs) may be exposed to environment. However, it is unclear whether the accumulation of copper nanoparticles in organism will increase under the warming and acidification scenarios. Therefore, the purpose of this study is to investigate whether CuNPs (25 nm, 0.03 mg/L) will accumulate in Japanese medaka (*Oryzias latipes*) embryos under the condition of elevated temperature and/or pH. The medaka embryo was followed for four consecutive days at 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, 28°C/5.5, 30°C/7.5, 30°C/6.5 and 30°C/5.5). Results showed that the temperature and pH did not affect the accumulation of CuNPs, respectively. When combined with temperature and acidification factors, Cu accumulation of group 30°C/6.5 was increased significantly than that of groups of 26°C/6.5, 26°C/5.5, and 30°C/5.5. However, it revealed that there was no significant evidence of warming and acidification on increased Cu accumulation of medaka embryo. Therefore, we concluded that there was no influence of Cu accumulation in medaka embryo when warming and acidification occurs in the future.

**WE316**

**Assessing the combined toxicity of metals and metal-oxide nanoparticles in a benthic estuarine microalgae**

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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 technology in the last decade have 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 mixtures due to heavy metal-ENMs interactions. Studies suggests 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 the characteristics of the exposure medium. Meta-analysis of published studies that investigate the dissolution of ENMs has revealed that the dissolution is not constant but varies depending on the exposure conditions. The data presented here is the result of a meta-analysis of published studies that investigate the dissolution of ENMs in the exposure medium. The data presented here is the result of a meta-analysis of published studies that investigate the dissolution of ENMs in the exposure medium. Therefore, the purpose of this study is to assess the effects of temperature and acidification on copper nanoparticle bioaccumulation in medaka embryos.

**WE317**

**Comparative toxicity of silver nanocolloids and titanium dioxide nanoparticles using medaka**

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Silver nanoparticles and titanium dioxide nanoparticles are representative nanomaterials and may be useful for multiple purposes in human life. Hence, environmental fate and unintentional ecological effects and/or toxicities have been considered and many studies are reported using model organisms. We have been investigating fish toxicity and ecological risk of silver nanocolloids (SNCs, 400 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, shortened length, small eye development, ischemia, reduced heart beating, and caused some oxidative stresses such as GSH reduction and lipid peroxidation. To adults, SNC 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 infectious 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 hands, titanium dioxide nanoparticles (TiO2-NP, Φ 90 nm) are well-known cause oxidative stress by UV radiation; however, there are some reports that TiO2-NP does not have significant toxic effect to fish other than
hyper trophy of gill nuss. 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), immuno-toxicity, and tolerance to acute and chronic bacterial 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 Genotoxicity assessment of aluminium oxide nanoparticles in relation to Escherichia coli and Aeromonas hydrophila
n. doskocz, M. Zalęska-Radziwill, A. Affleck, 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 physical and chemical 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 ecotoxicology. The randomized 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 used successfully 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 b-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 (nano Al₂O₃). L. Manusadzian, L. Tzortzi, E. Telari, E. Vakalid, 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. Determination of genetic sensitivity of obtained profiles bands for primer OPA2 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 Escherichia 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. Z. M. Sims, P. G. 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. Toxictans 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 combination with As throughout the whole life cycle of rice plants. No significant effect was observed on 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 (NP). 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, 50, 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 pharmaceutical compounds on aquatic specimens G. AMARIEL, Universidad de Alcala; K. Boltes, University of Alcala / Chemical Engineering: P. Letón, University of Alcala

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 aquatic environment, exerting toxic effects at the level of cells/tissues or the whole organisms. Therefore, 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 sludge, by exploring concentration-dependent effect and changes induced due to the presence of Ibuprofen (Ibu) or Levofloxacin (Levo). 72h algae growth-inhibition, marine bacteria luminescence reduction and 24h sludge enzymatic activity and oxidative stress were used as endpoints. Nanoparticles concentration ranged from 0.6 to 160 mg/L. The particle size and the ζ-potential of NPs in the culture media were measured to analyze 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 pharmaceutical compounds did not produce significant changes on nano-CeO₂ toxicity, but the following n-CeO₂ foraminates: 1) not exposed with a concentration of 0.625 mg/L n-CeO₂ (corresponding to the EC₅₀ for D. longispongia), ii) food (microalgae) spiked with nano-TiO₂ (after being exposed for 3 days to a concentration of 0.615 mg/L n-CeO₂, 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). D. longispongia showed a potent marine nanoparticle behavior for activated sludge. 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 BI. 2015. Behaviour of titanium dioxide and zinc oxide nanoparticles in the presence of wastewater-derived organic matter and implications for algal toxicity. [2] Sahle M, Venâncio L, M. Załęska, I. Ł. Manusadzianas, E. Padilla, A. D. longispina, M. Załęska, I. Ł. Manusadzianas, E. Padilla, A. Nano Toxicranis. 12, 10.1016/j.inoan.2015.08.022. [3] Załęska M, 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/MAE-2716 REMFVARES.

WE321 Toxicity of nanoparticles of titanium dioxide to Daphnia longispongia: waterborne versus dietary exposure E. Padilla, Institute for Environmental Sciences / University Koblenz-Landau; C. Venâncio, Department of Biology / Biology; I. Lopes, University of Aveiro / Department of Biology / Department of Biology / Biology; 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/MAE-2716 REMFVARES.

WE322 Dynamics of Cu accumulation in charophyte cell compartments after its exposure to nCuO Manusardzianas, NatureResearchCentre / Institute of Botany, Laboratory of Aquatic Ecotoxicology; B. Gylte, S. JURKONIEN, R. Vikutis, Nature Research Centre / Institute of Botany

In plant and bacterial cells, prior to be internalized, NPs have to pass separation of the main cell compartments, which enable mechanical separation of intermolecular cell of characean green alga posses features such as big size and clear internodal cell of characean green alga posses features such as big size and clear

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malate dehydrogenase and α Mannosidase, respectively. A high-purity vacuum (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 rCrO₃ 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? G. Nicolussi, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology, Science and Technology Faculty and Plentzia Marine Station, University of the Basque Country (UPV/EHU). Basque Country, Spain; A. Katsumiti, M.P. Cajaraville, 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 In the aquatic environment, complex mixtures of pollutants are usually found. Polycyclic aromatic hydrocarbons (PAHs) are prioritory 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 possible “Trojan horse” effect of graphene nanoplatelets (GnP). This work was funded by the EU H2020 (GRACE project grant 679266), Spanish MINECO (project NACE, CTM2016-8130-R), Basque Government (consolidated research group IT810-13) 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 L.A. Ellis, The University of Birmingham / GEES; E. Valsami-Jones, University of Birmingham / School of Geography Earth and Environmental Sciences; I. Lynch, University of Birmingham / Geography Earth Environmental Science. 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 change to their environments. Herein, we investigated key biological endpoints, such as survival, growth, reproduction, and morphological pathways in response to exposure to silver (AgNPs) and titanium dioxide (TiO₂) nanoparticles (NPs). Pathways were either pristine or aged, uncoated or stabilized with 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 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 µg/mL) compared to nanoplatelets alone (25 µg/mL), 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, CTM2016-8130-R), Basque Government (consolidated research group IT810-13) and University of the Basque Country (UIF 11/37).

WE325 Drivers of PAH and TEQ concentrations in marine sediments. L.M. Basirico, Louisiana State University; H. Rockett, R.J. Portier, Louisiana State University / Environmental Sciences 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 nearshore of the northern Gulf of Mexico including water column salinity, dissolved oxygen and 3) temperature of the overlying water column, and the percent sediment moisture and 5) percent organic matter of the aquatic sediment. A main effects-model was implemented in order to assess the impact of environmental variables on the concentrations of ten PAHs and three toxicity indicators in freshwater and marine sediments. A backwards, step-wise linear regression analysis of variance (ANOVA), generalized linear 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 of 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. The current research suggests that sediment organic content may be a significant driver and environmental 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 Microbial resistance to chemical pollution by urban effluents might be triggered by desiccation events. F. Romero, S. Sabater, ICRA Catalan Institute for Water Research; O. Pereda, University of the Basque Country; l. sabater, CSIC-IDAEA / Department of Environmental Chemistry; C. Font, V. Acuña, ICRA Catalan Institute for Water Research; O. Pereda, University of the Basque Country. 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 methaneogenesis. 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 stressors 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 disproportionally 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 fitmess 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 increased variability in temperature. Although fluoxetine and variability in temperature are the two major stressors, it remains unclear to what extent these two stressors can interact and jointly affect the response of organisms or their capacity to recover after exposure to 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 28 days to multiple human induced stressors. Our results indicated that mussels face greater fitness risk when exposed to multiple stressors at the same time than when each stressor acts on alone. We start the study highlights 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. Moreover, increased temperature regimes can not only affect the response of organisms but their capacity to recover from pollution events. In this study, we present the work aiming 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 28 days to multiple human induced stressors. Our results indicated that mussels face greater fitness risk when exposed to multiple stressors at the same time than when each stressor acts on alone. We start the study highlights 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.

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 pol brominated diphenyl ethyl 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), P 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 (50.6-67.8%), 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 viscerosomatic 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 response and energy metabolism. Our results indicate that the energy impacts, and those associated with other stressors, need to be further addressed, as understanding and managing these phenomena will be critical to maintain the sustainability of marine ecosystems. Further, the combined effect of warming and contamination, the energy budget of marine fish species is expected to be greatly affected, leading to impacts on fish fitness response and energy metabolism. Our results indicate that the energy impacts, and those associated with other stressors, need to be further addressed, as understanding and managing these phenomena will be critical to maintain the sustainability of marine ecosystems.
WE332 Functional and structural soil-vegetation indicators of ecosystem functioning in metal-contaminated environments: a case study in SE Spain

J. Alvarez-Rogel, A. Peralver Alcalá, M. Tercero Gómez, H. Conde Alcaraz, O. Martínez de Ozcar, E. Arquín-Vázquez, E. Pérez del Val, A. Astillero, I. Martínez Oró, Escuela Técnica Superior de Ingeniería Agronómica. Universidad Politécnica de Cartagena / Ciencia y Tecnología Agraria; F. Jiménez-Cárceles, BIOCYMA, Consulting in Environment and Quality, S.L. Murcia, Spain.; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science; M. González-Alcaraz, Departamento de Biología & CESAM - University of Aveiro / Biology & CESAM Functional and structural soil parameters of six environments defined by stages of vegetation succession were measured in 2016 in SE Spain. The objective of the present 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 (Lemna minor) using a combination of genomics, 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 UV light reduced L. minor reproductive rate at a high 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). Multiple stressors may also be enhanced by radiation from human activity such as nuclear power plants accident, nuclear medicine and weapon tests. (Van Hoec 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 and the interaction of radiation with metals in aquatic ecosystems of SE Spain. The objective of the present work was to study the relationship of metal contamination and radiation in SE Spain. The hypothesis must be confirmed by performing more specific tests.

WE333 Effects of imidacloprid and a neonicotinoid mixture on aquatic invertebrate communities under Mediterranean conditions

A. Riina, A. A. Sánchez, IMDEA Water Institute / Aquatic Ecotoxicology; J. Pasqualini, A. García-Astillero, L. Cherta, L. Nozal, IMDEA Water Institute; M. Vigh, IMDEA Water Institute / Earth and Environmental Sciences Neonicotinoids are a group of insecticides that are used worldwide in agriculture to treat piercing–sucking and chewing insect pests. These insecticides are considered contaminants of emerging concern due to their high toxicity to non-target organisms. In this study we measured the effect of imidacloprid (the most used neonicotinoid) and a mixture of five neonicotinoids (imidacloprid, acetamiprid, thiacloprid, thiamethoxam, and clothianidin) on freshwater macroinvertebrate and zooplankton communities. The experiment was performed using lentic mesocosms in Central Spain under Mediterranean conditions. This study demonstrated that the Concentration Addition (CA) model for the prediction of the toxic effect of mixtures of chemicals with the same mode of action may be applied to describe the short term effects of complex communities and not only individual organisms. However, some of the recorded indirect effects and the recovery of some populations showed slight differences between the imidacloprid and the neonicotinoid mixture treatments. This was attributed to the different dissipation rates of some of the test compounds 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 recovery. Some aquatic insect taxa (Chironomidae) showed a general trend 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 protective 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 (UVR) radiation on the IR duckweed (Lemna minor)

L. Xie, NIVA - Norwegian Institute for Water Research; Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management; O.C. Linn, K.A. Solhaug, B. Salbu, Norwegian University of Life Sciences; K. Toltenek, NIVA / Ecotoxicology and Risk Assessment In nature environment, aquatic biota is facing to the ionizing radiation emitted from natural radioactivity and non ionizing electromagnetic fields. These effects may also be enhanced by radiation from human activity such as nuclear power plants accident, nuclear medicine and weapon tests. (Van Hoec 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 and the interaction of radiation with metals in aquatic ecosystems of SE Spain. The objective of the present 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 (Lemna minor) using a combination of genomics, 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 UV light reduced L. minor reproductive rate at a high 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). Multiple stressors may also be enhanced by radiation from human activity such as nuclear power plants accident, nuclear medicine and weapon tests. (Van Hoec 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 and the interaction of radiation with metals in aquatic ecosystems of SE Spain. The objective of the present work was to study the relationship of metal contamination and radiation in SE Spain. 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- desorption behavior, and thermoresistance seem to be crucial for 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 clothing, and directly released into the surface waters, 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. exposed to sunscreen showed 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 biodegradation, constituting a major risk to marine organisms. Corals rely on the photosynthetic 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 decline, and direct exposure released to the surface waters, 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, 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 metallothionein (MT). 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 Gerridae, 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 metal 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 inductor 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 biomarkers for metal exposure in freshwater systems.

WE338 
Mollusks as indicators of environmental pollution (case studies in marine mussel Mytilus galloprovincialis Lam. and terrestrial snail Bradybaena truticum 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 biodiagnostics a 3-4 years old genetically modified morphs of bush snails Bradybaena truticum (Mull.) were used collected from Moscow city (Kartmazo, Kuz/mi/knee, Imsalovsk Park) with respectively annual and 10 years of mussel bioaccumulation test. Pairwise comparison was done based on cardiac activity monitoring in mentioned groups of snails under thermal treatment (20-50min, 50±5°C). It was revealed that snails of the same genotype (striped and without strips) from chemically polluted sites (Kartmazo, Kuz/mi/knee) differed in low thermoresistance from those of the reference side and Imsalovsk Park demonstrated in dynamics of HRs. The duration of cardiac activity monitoring (4 hours) of mollusks bioindication test, was performed in Sevastopol Bays using salinity change test. Possible links between chemical pollution by dioxin (in terrestrial snails) and heavy metals contents in mossier'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 0.08 ng/L and 0.6 ng/L respectively and annual average and maximum allowable concentration in surface waters are 0.008 ng/L and 0.06 ng/L respectively, one of the lowest 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 including the pyrethrroids which cypermethrin is part of. The 48 hour half maximal concentration (EC50) and median effective time (ET50) values were tested with cutoxemus 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 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 however 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. We refer to a number of multiple pattern recognition methods to identify and describe. 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 Rudipatus philippinorum from the Vallona lagoon (northern Adriatic Sea, NE Italy): Application of Contaminant/shell weight indices
E. Caiati, ISPRA-Italian Institute for Environmental Protection and Research / Mitigation and Prevention of Impacts; V. Bernarello, R. Boscolo Brusà, G. Frascinetti, G. Sesta, C. Maggi, D. Berto, M. Gabellini, C. Virno Lambert, ISPRA Institute for Environmental Protection and Research Rudipatus philippinorum (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 mussels 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 mussels. 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, Polycyclic 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. philippinorum 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 abovementioned factors. The main abiotic 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 mussels, Contaminant/shell weight indices, which consist of normalization achieved multiplying each contaminant concentration 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 contaminant 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. Vong, National University of Singapore Science Institute; 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 mangroves 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 Acetylhydroxysterosterase (AChE) were measured in the bivalves’ digestive tissue. Mussel’s haemolymph was also used to evaluate immunological parameters (Total Haemocyte counts, phagocytosis and lysozyme levels) and the level of haemocytes’ 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 disruptives 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 mangroves patches were potentially more at risk than others towards chemical contamination.

WE343 Impacts of climate change on mercury bioaccumulation in large ocean predators
Many marine ecosystems are being affected by climate driven changes in freshwater discharge, circulation, productivity and seawater temperature. Large ocean predators such as tuna account for a large proportion of methylmercury exposure in many countries (almost 40% of U.S. population wide intake between 2010-2012). Methylmercury is a potent neurotoxicant, 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, and trophic status). 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.

WE344 Chemical stress on aquatic communities under semi-arid conditions: towards an improved multimetric approach
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 chemical stress than those adapted to more constant water flows. The aim of this study was to evaluate the combined effects of multiple stressors on aquatic ecosystems in scenarios that are characteristic of (semi-)arid regions. In particular, this study focused on identifying the main stress factors that are influencing aquatic communities in the semi-arid region of Madrid (central Spain). Sixteen sites were selected in the watershed of the Tagus River (Madrid, Spain) and sampled in three consecutive years (spring, summer and fall). Hydrological and physico-chemical parameters of aquatic ecosystems were monitored, together with concentrations of metals and organic contaminants (pharmaceuticals, home-care products, pesticides). With respect to organic contaminants, the results from a screening analysis revealed the presence, at detectable levels, of 100 compounds in water samples. A group of 42 contaminants were selected for quantification due to their high toxicity and potential to aquatic organisms and frequent detection. Complex mixtures of pharmaceuticals, as well as highly toxic pesticides were identified. Through a multivariate analysis including pollution data, flow variability and related physico-chemical parameters, the main stressors and possible differences at
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 multimeteric 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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The influence of selected seasonal and anthropogenic phenomena on a perennial river in South Africa.

The quality of surface waters worldwide is declining fast. This is due to anthropogenic impacts from pollution and natural occurring floods and droughts 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 seasons. 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. The highest values of soluble metals were obtained during the low flow seasons, when compared to the high flow seasons. A combination of biological indices (Taxa Richness, Shannon-Weiner 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 taxa 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 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 that from single heat wave treatment as revealed by higher level of malondialdehide content and considerably stronger stimulation of antioxidative enzymes. Full recovery of photosynthetic processes and water content were observed during the 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 stress treatment compared to control plants 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. Threw exposure to extreme events many plants are stressed and show symptoms of heat stress and becomes thermally stressed. Characteristic symptoms are leaf bleaching, leaf scorches and loss of green color. Photosynthesis decreases as a result of heat wave stress. The extent of the damage to plants depends on the duration of the heat wave and the intensity of the heat. The study aimed to investigate the effect of heat waves on photosynthesis and growth of barley (Hordeum vulgare L.) and wild mustard (Sinapis arvensis L.) under controlled conditions. We evaluated the long term effects of a single heat wave in combination with drought (35°C vs. 21°C) on photosynthesis and growth. We found that increased temperatures decreased net photosynthesis, growth and biomass accumulation of Hordeum vulgare and Sinapis arvensis. The photosynthetic parameters of both species were not recovered after heat wave treatment as revealed by lower levels of chlorophyll a and lower RuBisCO activity in stressed plants. The importance of heat wave stress was evident in the physiological responses of both species. The combination of heat and drought treatments had a negative effect on photosynthesis and growth, whereas the single treatments did not affect the photosynthetic activity and growth. The results of the study showed that elevated CO2 does not protect plants against heat wave stresses.
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 competitive interactions. The aim of this study was 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 2:1, were sprayed with herbicide sprays solutions equivalent to 0.5 to 2.0 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 antioxidative 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 is known to pose at risk non-target insects due to effects on physiology and behaviour. Under natural conditions, there might be a development of resistance towards a specific substance over time. It can be expected, however, that the community resulting from the historic exposure to the stressors is not uniformly well optimized to utilize available resources in the best possible way. The hypothesis behind this study is drawn from the following concept: The history of community exposure to chemical pollution in the environment influences the sensitivity of responses to contemporary stressors. Such knowledge may persuade us to develop strategies of management in order to minimize the impact on sensitive species. Tadpoles and other aquatic animals from tropical areas also face multiple stresses, such as the plication of warming and increasing herbicide and organochlorine pollution. The aim of this study was to examine 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 shredder species and the associated production of fine particulate organic matter (FPOM). For that, trials were performed using a simplified trophic chain: Alnus glutinosa leaves as food resource, the shredder Sericostoma vitattum, the collector C. riparius and their natural predator the dragonfly Cordulegaster boltonti. A full factorial design tested the effects of the CAP (0 or 2 µg/L), presence/absence of the predator C. boltonti and of the shredder S. vitattum loss of FPOM and the feeding rate of C. riparius on the response of the model compound to environmental relevant concentrations of CAP decreased leaf decomposition in all treatments. Predation risk marginally reduced shredder effects on leaf decomposition. Considering detritivores interaction, an interspecific competition is suggested since the presence of shredders impaired chironomids performance despite the increase in leaf fragmentation. C. riparius growth rate was thus decreased independently by all factors (CAP exposure, predation risk and shredder presence) and a marginal interaction between CAP and predation risk was also observed. To conclude, the 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 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 sulfluron) 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 herbicides genes expression (dio2, dio3, thi6, tra, trfB 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 Ephippium nattereri had their antioxidant defense system affected by exposure to the herbicides clomazone and sulfluron, respectively. Sulf fed tadpoles were more resistant towards (SOD, CAT, G6PDH) and biotransformation enzymes activities and induced lipid peroxidation with temperature associated responses in tadpoles of both species. Clomazone also increase carbonylsterase activities in tadpoles exposed at higher temperatures. Integrated Biomarker responses (IBR) index showed a synergic effect of temperature and sulflurazon or clomazone in R. schneideri and E. nattereri. Our results demonstrated that temperature changes will influence survivorship and survival of tadpoles, and its effects can be pronounced at higher temperatures. These findings imply that the effects of abiotic factors should be taken into account to evaluate the real risks of exposure of amphibians to commonly used pesticides, mainly in tropical areas.

WE353 Adaptation vs. acclimation of natural phytoplankton communities towards herbicide exposure
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Freshwater ecosystems are subject to natural and anthropogenic disturbances such as climate change, landscape management, natural resources overexploitation and also pollution. Chronic background contamination by pesticides applied in agriculture poses a selective pressure on natural phytoplankton communities, favouring species and strains that can handle herbicide exposure better than others. Consequently, when faced with herbicide exposure, there might be a development of herbicide resistance (RUE). First results indicate less pronounced differences between the community responses at the two thermal regimes, and of the two species. Clomazone also influenced the formation of bacterial communities from tropical areas. Tadpoles and other aquatic animals from tropical areas of the world also face multiple stresses, such as the plication of warming and increasing herbicide and organochlorine pollution. The aim of this study was to examine 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 shredder species and the associated production of fine particulate organic matter (FPOM). For that, trials were performed using a simplified trophic chain: Alnus glutinosa leaves as food resource, the shredder Sericostoma vitattum, the collector C. riparius and their natural predator the dragonfly Cordulegaster boltonti. A full factorial design tested the effects of the CAP (0 or 2 µg/L), presence/absence of the predator C. boltonti and of the shredder S. vitattum loss of FPOM and the feeding rate of C. riparius on the response of the model compound to environmental relevant concentrations of CAP decreased leaf decomposition in all treatments. Predation risk marginally reduced shredder effects on leaf decomposition. Considering detritivores interaction, an interspecific competition is suggested since the presence of shredders impaired chironomids performance despite the increase in leaf fragmentation. C. riparius growth rate was thus decreased independently by all factors (CAP exposure, predation risk and shredder 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.

WE354 Impacts of climate change on freshwater pesticide exposure
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Climate change will modify environmental conditions which will likely have knock-on effects on the usage and environmental fate and behaviour of active
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 pollutants, covering a range of physico-chemical properties and uses, were modelled in scenarios 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. Naere, 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 effluent monitoring programs have been conducted for limited, but prioritized pollutants. In general, the prioritization has been mainly based on effect/toxicity information rather than exposure for relevancies. Thus, easily relevant 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 chromatographic 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 ranks, about 20 micropollutants were orthogonally confirmed and roughly quantified. The quantified micropollutants were mostly pharmaceuticals and personal care products including 9 groups such as analgesics/anti-inflammatory (acetaminophen, mefenamic acid), antibiotics/antifungal (climbazole, fluconazole, sulfamethoxazole, sulfamethazine), anticonvulsant (carbamazepine, carbamazepine-epoxide, oxcarbazepine), antistatins (diphenhydramine, fexofenadine), antihypertensive agent (ibresartan, valsartan), antipsychotic (amitriptyline), antibiotics (amoxicillin, clindamycin), and angiotensin receptor blocker (enalapril). 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> ibresartan> fluconazole> dehydrocyclohexylamine> 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. Hug, 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), endoperoxidases (S-transferase (GST) enzymatic 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 35.3ºC and 32ºC, respectively, in 7 days of exposure to temperature of 2ºC per day. In T. kingsejongensis, LT50 and NOEL were determined to be 28.4ºC and 12ºC, respectively. Levels of OS and GST activity were slightly elevated (<em>P</em>)

WE357
Effects of water browning on zooplankton physiology and fitness driven by foraging characteristics in a long-term enclosure experiment
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ECotoxocological 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 stressor is 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 seston characteristics and the elemental and biochemical composition of the daphnids. Surprisingly, daphnids kept in brown water showed higher reproductive performance than daphnids kept in clear water (A). This unexpected outcome is explained by higher seston quantity and quality in enclosures receiving tDOC, related to a higher abundance and biomass of nutritious 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 specimens in the recipient conditions. However, the exposure of aquatic systems in the UK. 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 infrastructure) 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 identify pattern 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-trrophic test system
V. Riedl, Environment Department, University of York / Environment Department; A. Agatz, IBACON GmbH / Environment Department; R. Bensted, Fera Science Ltd / Centre for Chemical Safety and Stewardship; R. Ashauer, University of York / Environment
Concerns, 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 ensure 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 proportionately translate to impacts at the population and community level. The use of testing procedures that are ecologically more realistic but ideally comply with regulatory needs thus should be a priority to risk assessors and regulators. The tri-trrophic 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 interacting 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 of 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 synthetic stressors in life. First generation acute toxicity bioassays and 48h chronic toxicity bioassays were conducted with two strains of Brachionus sp. Cayman (MRS10 and IB3), 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 psu 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 non-lethal temperature shocks (cold and heat), and after a recovery period, survival tests were performed. 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 greater resistance 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, freshwater producers provide important services which leads to a strong societal demand concerning the preservation of their quality. They are the receptors of many contaminants emitted by human activities and more specifically, Polywater pollution 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 DIADEMP project suggests developing and spreading out a cross-border multidisciplinary approach to improve the diagnosis and the chemical and biological (biomarkers) monitoring of freshwater producers 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 models. In order to calibrate and validate these models, a lotic mesocosm experiment was set up. Five substances were chosen: diclofenac, carbacholamine, naproxen, paracetamol and ibesartan. An environmental realistic mixture M of the five substances was tested along with MX10 and MX100. The study was carried out in twelve 20 m long lotic channels. The mesocosms were set up with artificial sediments, macrophytes, periphyton, benthic and pelagic invertebrates decomposers and one fish species (Gasterosteus aculeatus), zebra mussels, Fontinalis antipyretica and Gammarus fossarum were also caged in the mesocosms. After 3 months of stabilization, treatment lasted 5 months. Periphyton biomass, macrophyte biovolume, zebra mussel biomarkers and growth, G. fossarum survival, reproduction and growth, F. antipyretica biomass and growth, zooplankton and invertebrate abundance and diversity, and fish individual physiological responses to these substances were then measured biologically endpoints. The concentrations of each substance in water was monitored monthly along with some physico-chemical parameters. The overall experimental design will be presented along with the results related to the monitoring of substance concentrations in water, physico-chemical parameters, macrophyte biomarkers, invertebrate community response, fish larval densities. A brief discussion of the direct and/or indirect effects will then be performed.

Improving the Quality of Ecotoxicological Testing and Assessment (P)

WE362 Relationships between aquatic toxicity, chemical hydrophobicity and mode of action: log kow QSRs revisited

Quantitative structure toxicity relationships (QSRs) 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 MOA classifications. Log kow-QMOA relationships were developed as linear regressions 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), acetylcholinesterase 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 models showed that MOA-based QSRs 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, Eurofins 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 endpoint (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. The magnitude and exposure duration information about relevant sensitive life stages. This additional information may be relevant in context with higher-tier testing strategies as revised exposure testing or to justify the deployment of time-weighted average surface water concentrations for risk evaluation. Gaining of this additional information should be taken into consideration for planning of lower-tier studies with the most relevant organisms. For example spacing of the test concentrations or additional assessment dates during the test period can maximize the knowledge that may be retrieved from these tests with regard to potential risk refinement. This poster presentation gives examples on how results of standard ecotoxicity studies can more efficiently be used as basis for higher-tier approaches in the environmental risk assessment of agrochemicals.

WE365 Comparison of models and tools for derivation of species sensitivity distributions (SSDs) for use in pesticide risk assessment
L. Azevedo, BASF SE, Agrарzentrum Limburgerhof / Global Toxicology; G. Schmidt, BASF SE

EFSA’s guidance document for the risk assessment of edge-of-field aquatic organisms (EFSA, 2013;11(7):3290) recommends the use of species sensitivity distributions (SSD) as a second-tier approach for the aquatic risk assessment of plant protection products. For macrophytes, the hazardous concentration to 5% (HC₅) of tested species can be attained by deriving a species sensitivity distribution (SSD) composed of effective concentrations to a 50% effect on the growth rate of primary producers (ER₅₀). Various probability distributions are available for the derivation of a SSD (e.g., lognormal, logisitc) as well as publicly-available tools (RIVM’s ETX, MOASAIC-SSD from the University of Lyon, US EPA’s SSD
WE366
Effects on NTA communities: HCx vs NOEC designs
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 HCl, 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 tests were performed to a limited extent. The NOEC was found to be 56% (11%) of control 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 HCl, study consistent dose-response curves were obtained within 4 major arthropod taxa (63 out of 776; 8%) and SSD’s 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 66 from 596 (11%) taxa were valid for univariate analysis, representing all major 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 yielded biologically and statistically valid results. Whereas the HCl-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 MSD-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, value are key endpoints to assess safety of pest control measures, they are particularly challenged when 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-findings 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. The former can be known as Type-I and Type-II errors that lead to formulas for the calculation of each of them. Due to frequencies α and β, respectively. This contribution challenge 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 > α) does not necessarily equate to biological insignificance (P > β), whereas it only means that the producer’s risk is low. Safety stems from rejecting the null hypothesis when false and therefore the complement of β is known as the power of an experiment. We show how power criteria can be derived and used to construct a biology based confidence profile for studies addressing NTA communities. We also show how relaxing α helps to identify those taxa for which an experiment does not provide sufficient conclusive data to draw meaningful conclusions. In a multi-rate study design, the proposed increase of α to 10% is shown to be off-set by applying expert criteria such as inconsistencies in dose or time.

WE368
Defining simple toxicity values (EC, BMD) is not so simple
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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 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 (BMD) concept has been proposed in the field 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 hazardously 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 control. We will illustrate why this feature makes it more robust and usable 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. Fremdt, 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 and LC50 is the concentration 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 ECx/LCx due to the ignorance of control variability, the improper use of binominal 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 the baseline 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. Fremdt, Bayer AG Crop Science Division; T. Preuss, Bayer AG / Environmental Safety
Low effect EC values (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 is a power to detect effects, whereas the ECx/LCx derived from 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. Comparisons in which 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 parameterizations and curve shapes between the so-called linear and non-linear models are clarified in the manuscript. The characteristics of these approaches without caution are emphasized. 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 over 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

N.D. Forsberg, Arcadis U.S., Inc. / Environmental and Molecular Toxicology; B. Magee, S. Sager, ARCADIS US Inc

Derived no effect levels (DNELs) are indispensable tools needed to quantitatively evaluate the safety 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 default conservative estimations that when combined lead to a phenomenon termed “compound 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

G. Eck, F. Eschenbach, Eurofins Regulatory AG

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 a 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 testing 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 complicacy 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 conditions 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 lead to many scenarios. A critical point is to cover that the 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 practical scenarios. The poster will describe how to develop and implement realistic exposure profiles using parameters used in standard exposure models and designing field fate studies to derive more realistic parameters; analysing the exposure profiles associated with the maximum predicted exposure concentration in surface water (PECsw) compared to the exposure conditions used in standard aquatic ecotoxicology studies and designing modified exposure studies to more accurately mimic these exposure profiles; etc. The aim of this poster is to illustrate how different disciplines can work together to challenge the default assumptions of standard aquatic risk assessment, thus enabling appropriate refinement options to be derived and together design optimal solutions that are closer to addressing the real risks, rather than the theoretical ones.

WE374
Critical aspects of higher-tier laboratory exposure testing with different aquatic organisms

G. Consier, Eurofins Agroscience Ecotox GmbH; U. Memmert, G. Eck, F. Eschenbach, Eurofins Regulatory AG; C. Hafner, Eurofins Agroscience Ecotox GmbH / Aquatic Ecotoxicology

Repeated pulsed exposure in a partial life cycle test with zebrasfish: Keep it realistic!

M. Teigeler, Fraunhofer IME / Ecotoxicology; B. Weber, D. Warnecke, RIFCON GmbH Goldbeckstr Hirschberg Germany

Reefline exposure testing is proposed in the current EFSA guidance document for aquatic risk assessments providing the possibility to define ecotoxicologically relevant test conditions and a 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 testing 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 complicacy 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 conditions 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.

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WE376
Pulsed exposure of fish at sensitive life stages: The ‘worst case’ challenge.

SETAC Europe 28th Annual Meeting Abstract Book
WE377
TIER2+: Developing the Tools for Future Risk Assessment - New Chronic Invertebrate Test Systems and the Application of Realistic Exposure Scenarios
A. Dabrunz, F. Kümmich, 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. An Experimental data of newly established (sub)chronic test systems, which are suitable meeting Tier 2 criteria 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 a typical laboratory exposure is unrealistic.

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. Peiffer, Innovative Environmental Services IES Ltd; S. Högner, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Kopp, 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 throughout the test duration, exposure 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 system was carried out. This comparative test allowed studying stress parameters by comparing the effect level concentrations and the ECx 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 copper for offspring per survivor over 21 days. 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. Lancher, C. Dupuy, A. Jouand, Groupe SGS France; J. Berot, SGS Multilab / 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 HOCN guideline, for example. The embryo and larvae sensitivity of turbot (Scophthalmus maximus) to different concentrations of the test substance (labrax) 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, BASF SE; M. Mathis, 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 51 Xenopus laevis larvae to different concentrations of the test substance for 21-days and the following endpoints are measured: mortality, hind limb length (HLL), body length (snout to -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. Currently, the relevance of normalized 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 hindlimb length and body weight was found in 6 of the 7 studies with the asynchronous stage development (r²=0.347-0.156, 0.429, 0.564). For the censored studies, correlation 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 determined 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 test using Mediterranean fish species (Dicentarchus labrax L., 1758): Inter-calibration exercises towards standardized procedure

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M. Teigeler, E. Eilebrecht, Fraunhofer IME / Ecotoxicology; A.J. Jones, DuPont Crop Protection / Institute of Environmental Toxicology
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 values in the field are significantly shorter than in the standard laboratory test. However, this challenge is often to cover exposure values 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 a typical laboratory exposure is unrealistic.
but also standard test organisms for the RA for pharmaceuticals and chemicals. Diatoms are not only part of the risk assessment (RA) for plant protection products, S. Höger during the testing of difficult substances, this presentation also intends to underline can be successfully handled using all available technical options, which are cooperation with an external company specialized on providing flow through a sophisticated flow through test device to guarantee the success of the test. In and difficult tests, substance to be test chronic fish tests very low concentrations have to be tested and in many cases the requested at a later stage from the competent authorities. Typically, for these additives towards the endocrine system (and finally the reproduction) cannot be excluded. Full life cycle tests. All these tests include the evaluation of sublethal effects on the aquatic 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 have to carry out assays on sea bass larvae (50-70 days old) to the toxicant reference (Sodium Dodecyl Sulfate) concentrations: 6.31-3.98-2.51-1.58-1.00 mg/L and control. The LC50 (Trimmed Spearman-Karber method: TSK) mean valued ranged from 2.93±0.52 mg/L to 3.98±0.99 mg/L to 24h; and from 2.90±0.50 mg/L to 3.78±1.03 mg/L to 48h, respectively. The intra and inter laboratory variability of the tests were verified and Z scores were calculated. 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.

**WEB382**

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 test toxicity 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 toward the endocrine system is expected. For example in some cases the endocrine endpoints are additional tests, which may be requested at a later stage from the competent authorities. Typically, for these chronic fish tests very low concentrations have to be tested and in many cases the substance to be tested can be classified as difficult according to the OECD criteria, for instance low water solubility, high toxicity to fish, volatility or degradation during the test period. Due to the need for a flow test fish tests preferable a flow through test design is used and difficult test items with specific properties as described above request a highly sophisticated flow through test device to guarantee the success of the test. In cooperation with an external company specialized on providing flow through technique to science and industry, IES developed a new, highly flexible dosing system. This very flexible and computer controlled dosing device is a modular system which provides several new technical features for important steps during the test e.g. dosing of the test substance, preparation of the test media and distribution of the test medium to replicates. In this presentation several examples for the testing of difficult substances are shown and the advantages of this dosing system are explained. The biological and analytical results demonstrate that difficult test items can be successfully dosed and all available test protocols can be provided by the presented flexible flow through dosing system. Considering the increasing complexity of ecotoxicological tests and the methodical challenges during the testing of difficult substances, this presentation also intends to underline the importance of a continuous improvement of the technical setup for a successful performance of ecotoxicological test.

**WEB383**

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 for the “standard test items” with difficult test design to 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). To provide the necessary biomass for the presented working procedures demonstrate that every test item - independent from its characteristics - can be tested according to established OECD Test Guidelines, but in some cases extensive biological and chemical background and innovative capacity is required to find the best test design. To make it even more complicated, there are different ways to interpret the data and the most appropriate is clearly the result of the required endpoints. The different possibilities are introduced and discussed as well.

**WEB384**

Activity based Collombola sampling may improve the data of field studies for regulatory purposes

P. Mack, A. Appeltauer, J. Illig, Eurofins Agrosciences Ecotox GmbH; S. Knaebe, EAS Ecotox GmbH / Ecotoxic 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 Römhke 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 migrate 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 slide traps which were presented at SETAC 2016 by Dehelen et al. 2016. Our poster will discuss possible advantages for the combination of soil core and slide trap sampling and will present first the results from the comparison of soil core and slide trap catches. Römhke, J., Schmelz, R., Knaibe, 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

**WEB385**

New Technology evaluating Acartia tonsa as a biological model

S. Abreu, 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 / DEIT 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 the bottleneck for its massive utilization relies on the time consuming 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 differentiation in real time of copepod 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 indicate a high significant correlation between manual and automatic counting, constituting the first step for the use of this biological model on experimental studies.

**WEB386**

Solubility limits of lanthanides in standardized ecotoxicological media
Limonene is a stereoisomeric 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 considered safe when used as fragrance and flavour. However, the presence of possible LN -containing precursors can further complicate the interpretation of the corresponding biological responses. In this contribution, we use thermodynamic speciation modelling to examine the equilibrium theoretical speciation of LN in standardized ecotoxicological media for algae (chlorophyll a), in particular species; sweep nets are used to capture fast swimming organisms in test media. In medium aliquots amended with Cr(III) (range 0.005 to 1.25 mg/L), renewal every 24 h was not sufficient to obtain stable exposure concentrations (i.e. ±20% of the initial value) throughout the duration of the test. The actual exposure range (estimated as time weighted mean concentrations) was between 5 and 275 μg/L and was used to estimate the Cr(III) EC50. On the other hand, concentrations remained stable over time and agreed with expected values in comparative studies where the presence of dissolved organic matter can cause important differences in 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, LN can rapidly form complexes with phosphates or carbonates or undergo hydrolysis. Due to the low solubility of LN-phosphates and LN-carbonates in water, the formation of these is highly unlikely and LN solubility and reduce to a fraction of the expected value the concentrations to which organisms are exposed. The presence of possible LN-containing precipitates can 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.

WE387
Improving ecotoxicity tests for trace elements forming poorly soluble chemical species in test media
D.A. Vignati, CNRS / LIEC UMR7360; F.G. 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 EAWGEPFL; 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 hydroxides for Cr(III) and Sn, Ga, In species 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. We further studied the possibility of evaluating variable vs. colloidal/particulate elemental species to biological effects by testing the ecotoxicity of solutions aged for different periods. Chromium(III) was chosen as a model contaminant, but the general approach is applicable to all elements forming poorly soluble species and potential colloidal precipitates in ecotoxicological test media. In medium aliquots amended with Cr(III) (range 0.005 to 1.25 mg/L), renewal every 24 h was not sufficient to obtain stable exposure concentrations (i.e. ±20% of the initial value) throughout the duration of the test. The actual exposure range (estimated as time weighted mean concentrations) was between 5 and 275 μg/L and was used to estimate the Cr(III) EC50. On the other hand, concentrations remained stable over time and agreed with expected values in comparative studies where the presence of dissolved organic matter can cause important differences in 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, LN can rapidly form complexes with phosphates or carbonates or undergo hydrolysis. Due to the low solubility of LN-phosphates and LN-carbonates in water, the formation of these is highly unlikely and LN solubility and reduce to a fraction of the expected value the concentrations to which organisms are exposed. The presence of possible LN-containing precipitates can 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 stereoisomeric 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 considered safe when used as fragrance and flavour. However, the presence of possible LN -containing precursors can further complicate the interpretation of the corresponding biological responses. In this contribution, we use thermodynamic speciation modelling to examine the equilibrium theoretical speciation of LN in standardized ecotoxicological media for algae (chlorophyll a), in particular species; sweep nets are used to capture fast swimming organisms in test media. In medium aliquots amended with Cr(III) (range 0.005 to 1.25 mg/L), renewal every 24 h was not sufficient to obtain stable exposure concentrations (i.e. ±20% of the initial value) throughout the duration of the test. The actual exposure range (estimated as time weighted mean concentrations) was between 5 and 275 μg/L and was used to estimate the Cr(III) EC50. On the other hand, concentrations remained stable over time and agreed with expected values in comparative studies where the presence of dissolved organic matter can cause important differences in 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, LN can rapidly form complexes with phosphates or carbonates or undergo hydrolysis. Due to the low solubility of LN-phosphates and LN-carbonates in water, the formation of these is highly unlikely and LN solubility and reduce to a fraction of the expected value the concentrations to which organisms are exposed. The presence of possible LN-containing precipitates can 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.

WE389
Is that an effect? The importance of using all relevant data in mesocosm studies
J. Ashford, Cambridge Environmental Assessments; A.C. Brooks, Cambridge Environmental Assessments / Department of Environmental Science; A. Lawrence, Cambridge Environmental Assessments / Regulatory Ecotoxicology; M. Allen, Cambridge Environmental Assessments / Regulatory Ecotoxicology. In long-term multispecies studies, such as mesocosms, a complex statistical analysis is required to decipher the data and determine whether a test-item effect has occurred. It is sometimes the case that regulators and applicants have differing opinions as to what the no observed effect concentration (NOEC) or no observed adverse effect concentration (NOAEC) should be, based on the accompanying data and an expectation that chemical species in the environment (III) is sometimes viewed in isolation from the rest of the available data from the study. The aim of this poster is to urge regulators to not just focus on statistically significant differences, but to take into account all available and relevant data to assess the biological relevance of any differences observed. At CEA we use a range of sampling methods to collect data on aquatic macroinvertebrates within our mesocosm studies, including emergence traps, colonisers and sweep nets; each method captures different species; sweep nets are used to capture fast-moving pelagic organisms whereas colonisers are left in-situ to allow benthic organisms to enter the trap. In some cases, the same species can be caught using different sampling methods, such as mayflies, damselflies and caddisflies, and this can be from various stages in their life cycle. Comparing the data for the same taxa from different sampling methods can be helpful in creating and overall picture of how that taxa is responding within the mesocosm study. This can be helpful in deciding whether statistically significant differences are biologically relevant; for example, if a statistically significant difference to the control is only observed in one out of three sampling methods used for a particular taxa, this may indicate this is due to natural variation rather than the influence of the test item. Here, we will review literature, damselfly and mayfly data from past CEA studies where statistically significant differences have been observed and highlight cases where different sampling methods (emergent, colonisers and sweep nets) support or contradict a test-item effect.

WE390
Evaluation of the environmental risk assessment procedure according to Directive 2001/18/EC for Gene Modified Organisms used as medicinal products
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The deliberate release of genetically modified organisms (GMOs) including GMOs designed to be biodegradable, hydrophobic, non-polar narcotic substance like limonene under chronic conditions, how these were countered by the monitoring team and after a careful assessment of the data, both contributed to the development of a long adventure lasting several years with results supporting a chronic 3 classification. The subsequent regulatory procedure to implement the classification in the EU regulations is currently ongoing.

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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 EU regulation. 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. The information includes, 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 closely modified Adenoviruses, the most frequently used vector in gene therapy trials worldwide. Relevant information were entered into a database and categorized, applied 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 heavily confused with common vectors used 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 heavily confused with common vectors used in gene therapy trials worldwide.

WE391 PBT evaluation 20 years on: is it time to reconsider the technical progress made in risk assessment methodology? P. Thomas, C. Durou, CEHTRA 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 compounds has been changed in recent years. To prevent misinterpretation 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/vP criteria were originally designed. The numerical criteria were established in the late 1990s. OASAP. All the primary data has been transferred into an environmental database 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 heterogeneous group. Interestingly, the criteria were originally defined by using data from a set of chemicals known as highly hazardous for the environment. Such chemicals can have influence on development of hypopharyngeal glands and therefore 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 protein 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 even minor sub-lethal effects, hypopharyngeal glands do not develop correctly in these bees. Therefore, it is important to consider these changes in the evaluation of the current guidelines (EFSA and OECD) for the evaluation of hypopharyngeal glands as an endpoint in toxicity testing of chemicals on bees.

WE394 Assessing toxicity to Daphnia magna using movement parameters T. Derd, Department of Biology, University of Osijek / Department of Biology; D. Hackenberger, GeoNatura; B. Hackenberger, Department of Biology, University of Osijek; M. Hrnjak, Institute for Ecotoxicological Research of Industrial Organic Chemistry, Branch Pszczyna; A. Daniel-Wójcik, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies 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 usually in a more general sense. Bees are few steps forward and they also find application in the food, pharmaceutical and other 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 protein 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 even minor sub-lethal effects, hypopharyngeal glands do not develop correctly in these bees. Therefore, it is important to consider these changes in the evaluation of the current guidelines (EFSA and OECD) for the evaluation of hypopharyngeal glands as an endpoint in toxicity testing of chemicals on bees.
registration process for plant protection products. When Regulation (EC) No. 1107/2009 came into force, adherence to the SANCO/3029/09 rev 4 Guidelines became obligatory. This has resulted in serious implications for the registration process because ecotoxicological studies may well now be rejected on the basis of inadequate analytical methodology or incomplete analytical data although the studies have previously been accepted. Here we give an overview of current requirements and provide a checklist that can be used to evaluate analytical methods in ecotoxicological studies.

WE396 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

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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 72 hour stage exposure phases on Days 0 and 14. 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 exposure. The study was designed to be fast, 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 exposure. The study was designed to be fast, 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 exposure. The study was designed to be fast, 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 exposure. The study was designed to be fast, 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 exposure. The study was designed to be fast, 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 exposure. The study was designed to be fast,
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 is currently an area of focus. Much of the research using simplified, synthetic mediums has demonstrated that ENM stability is strongly influenced by the engineered surface coating enveloping the ENM. However, it is unknown 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 Samples using 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 time was monitored. As interactions with the surrounding medium, attachment efficiency factors (α_attach) were then calculated. As an indicator of ENM stability, α_attach provides a quantitative metric to determine whether the engineered surface coatings influence ENM stability in the representative medium chosen for this work. Despite the uniformity in the medium, significant differences in the stability of the model ENMs were observed. As was expected, the ENMs that maintained a positive surface charge after interacting with the surrounding medium were stabilized. More importantly, however, was that both the neutral and negatively-charged ENMs remained stable throughout the duration of the experiment (8 hrs.). This suggests that the surrounding environment did not affect the stability of these ENMs and demonstrates that ENM stability is influenced by the engineered surface coating, even after interacting with a complex, natural medium.

WE401 Engineered Nanoparticles interactions in secondary wastewater treatment: removal kinetic and efficiency during activated sludge stage. Y. Yang, University of Strathclyde / Civil and Environmental Engineering; R. Skuce, Scottish Water Horizons Ltd; C. Knapp, V. Phoenix, University of Strathclyde / Civil and Environmental Engineering

The rapid evolution of nanotechnology poses a unique and significant challenge for wastewater treatment plants (WWTPs). Engineered Nanoparticles (ENPs) are already utilized in a diverse array of applications, including cosmetics, optics, medical devices, and semiconductors. As the interflow time, we will see an increasing 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 a complex, natural medium. 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 ENP-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 denitriﬁcation of sewage involved in the removal of ENPs from sewage 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 “spoon” 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 (LCA) 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 sediment near a production site in Vieux-Thann (68) to determine parameters which control the fate of TiO2 NPs and several ENP removal methods are used in particular inductively coupled plasma with atomic emission spectrometry (ICP-AES), transmission electron microscopy (TEM) and conductivity or potential of hydrogen (pH) measurements. Then, parameters determined in the previous steps are used to calculate a fate factor of TiO2 ENPs in a natural environment according to the life cycle impact assessment method calculation. During the study, it was found that ionic strength, pH, percentage of organic matter, soil composition (percentage of clay, silt and sand) or size and concentration of TiO2 NPs are parameters which matter in TiO2 ENPs fate in soils, water and sediments. Furthermore, the first results obtained show that the sampling point located upstream of the production site has the lowest concentrations of titanium dioxide in soil and sediments. This point is used as a reference and allows evaluating the effect of wastewater and wastewater treatment on the transport of TiO2 NPs. As well as experimental attachment efficiency monitoring, methods for ENPs. However, previous developed models have not considered fate and transport of ENPs in intermediate path before reaching the each environmental compartment in spite of its importance. For example, more than 60% of TiO2 ENPs does not directly enter into natural surface water after usage, but they first transported into domestic wastewater. This study analyzed spatiotemporal concentration changes of TiO2 ENPs, a network through newly developed dynamic model, SWNano. Generally, heteroaggregation of ENPs with suspended particulate matters (SPM) is major process determining the fate and transport of ENPs in water. Water quality such as pH, DOM contents, ionic strength and Suspended solids, etc. and characteristics of ENPs such as shape, size and surface treatment strongly affect aggregation rate of ENPs. Therefore, we estimated attachment efficiency through sedimentation experiment of TiO2 NP in real wastewater and we compared sedimentation rate with the values experimented in other kinds of wastewater from previous studies. As well as experimental attachment efficiency, various input data of SWNano model such as SPM particle size, number concentration of SPM, zeta potential, etc. were also obtained through measurement by dynamic light scattering (DLS) and nanotag tracking (NTA), etc. It was confirmed that the TiO2 NPs aggregated relatively quickly with the SPM in sewage considering that the attachment efficiency was estimated to be about 10⁻¹ to 10⁻². When applying experimental values to input parameters of SWNano model, the range of predicted concentration of SWNano model was almost matched the concentration range from other model results. The model results showed that decreased TiO2 NPs decrease with increasing SPM through heteroaggregation and dilution by SPM when the concentration of SPM increased with time. Besides, it was also verified that the degree of decrease of dispersed TiO2 NPs concentration in sewer with time is significantly different according to the difference of attachment efficiency.

WE403 Assessing the fate and transport of engineered TiO2 nanoparticles in sewer pipes through a dynamic multimedia model (SWNano)

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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. As a result there is a need for an improvement of models to include ENPs for monitoring techniques for ENPs. However, previous developed models have not considered fate and transport of ENPs in intermediate path before reaching the each environmental compartment in spite of its importance. For example, more than 60% of TiO2 ENPs does not directly enter into natural surface water after usage, but they first transported into domestic wastewater. This study analyzed spatiotemporal concentration changes of TiO2 ENPs, a network through newly developed dynamic model, SWNano. Generally, heteroaggregation of ENPs with suspended particulate matters (SPM) is major process determining the fate and transport of ENPs in water. Water quality such as pH, DOM contents, ionic strength and Suspended solids, etc. and characteristics of ENPs such as shape, size and surface treatment strongly affect aggregation rate of ENPs. Therefore, we estimated attachment efficiency through sedimentation experiment of TiO2 NP in real wastewater and we compared sedimentation rate with the values experimented in other kinds of water from previous studies. As well as experimental attachment efficiency, various input data of SWNano model such as SPM particle size, number concentration of SPM, zeta potential, etc. were also obtained through measurement by dynamic light scattering (DLS) and nanotag tracking (NTA), etc. It was confirmed that the TiO2 NPs aggregated relatively quickly with the SPM in sewage considering that the attachment efficiency was estimated to be about 10⁻¹ to 10⁻². When applying experimental values to input parameters of SWNano model, the range of predicted concentration of SWNano model was almost matched the concentration range from other model results. The model results showed that decreased TiO2 NPs decrease with increasing SPM through heteroaggregation and dilution by SPM when the concentration of SPM increased with time. Besides, it was also verified that the degree of decrease of dispersed TiO2 NPs concentration in sewer with time is significantly different according to the difference of attachment efficiency.
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 organisms. Therefore, the hypothesis in this work is that microalgae lacking of cell wall will be more vulnerable to toxic effects of NPs than species with a typical cell wall. To test this hypothesis two microalgae species, Dunaliella salina, lacking cell wall, and Chlorella autotrophica, with a typical cellulosic cell wall were chosen. Species were exposed to (AgNO₃ and Ce(NO₃)₃) 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 microalgae 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-discretions, the measured z-averages ranged from 600 nm (CuO-27-Ni) up 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 NPMs have a primary particle size between 200 nm and several micrometres. In freshly sonicated stock-discretions, the measured z-averages ranging from 600 nm (CuO-27-Ni) up to 8 µm (HKUST), Zn-CPO and CPO-27-Ni had the most negative zeta-potential of -25 and -20 mV respectively, with Al (OH) fumarate and FeBTC-JM-AR forming a positive surface charge. Uio-66-COOH and HKUST had very weak surface potentials, which was also reflected in their instability in the stock and exposure media. In a first dissolution study, 5 out of 6 materials (100 mg/L) caused an increase in specific dissolved metals or elements in the exposure media. In a first dissolution study, 5 out of 6 materials (100 mg/L) caused an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting an increase in specific dissolved metals or elements in the exposure media.
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 Grlx-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 monitoring the transcript abundance of AgNPs-exposed E. coli 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 C. elegans in response to toxicants. Acknowledgements: Karl Andreas Jensen and Solfrid Lohne. This work was supported by the Norwegian Research Council funded NanoCharm (221390/440) 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 emission 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 (AgNist) were selected as test nanomaterials and 4 different exposure scenarios were considered: (1) control, (2) layer of AgNPs with low concentration (Low-Layer), (3) layer of AgNPs with high concentration (High-Layer), and (4) mixture of AgNPs and soil with low concentration (Low-Mix). Plant microcosm experiment was conducted in the greenhouse for 9 weeks. Soybean plant was most inhibited in Low-Mix exposure group which mimicked whole mixture of nanomaterials and soil because the soil surface was much more likely to be covered by AgNPs than Low-Layer and High-Layer. In case of soil enzymes, activities were dependent on exposure concentration. This study concluded that exposure concentration of nanomaterials as well as depth of nanomaterials layer should be considered in the soil ecotoxicity research area. 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 (2016R1A2B301045).

WE410 Fragmentation of nano- and microplastics from expanded polystyrene exposed to sunlight Y. Song, Korea Institute of Ocean Science and Technology; W. Shin, S. Hong, Korea Institute of Ocean Science and Technology / Oil and POPs research group; S. Eo, Korea Institute of Ocean Science and Technology

Production of nano- and micro-sized plastics 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 sunlight exposure dose difference were observed by scanning electron microscopy. Nano- and micro-sized particles were 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 was qualitatively and quantitatively determined from the expanded polystyrene (EPS) exposed to sunlight for 9 months. The exposed EPS cubes (3x3 cm surface area) were sampled in duplicate at 2 (2M), 5 (5M) and 9 month (9M) after sunlight exposure. The exposed EPS cubes were collected in 2 ml solution consisting of HPLC grade pure water with 0.1% Triton X-100. The exposure conditions were performed with plastic particles (micro < 1mm, MPs, to nano < 1µm, NPs) are among the most prominent environmental issues faced by government and research agencies. The accumulated exposure of plastic particles to ambient conditions can be substantial; 1) control, 2) layer of AgNPs with low concentration (Low-Layer), and 3) mixture of AgNPs and soil with low concentration (Low-Mix). Plant microcosm experiment was conducted in the greenhouse for 9 weeks. Soybean plant was most inhibited in Low-Mix exposure group which mimicked whole mixture of nanomaterials and soil because the soil surface was much more likely to be covered by AgNPs than Low-Layer and High-Layer. In case of soil enzymes, activities were dependent on exposure concentration. This study concluded that exposure concentration of nanomaterials as well as depth of nanomaterials layer should be considered in the soil ecotoxicity research area. 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 (2016R1A2B301045).

WE411 Effects of nano-plastics on natural marine aggregates and their associated microbial communities S. Summers, SCELS E Nanyang Technological University / SCELS; 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 micron 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 concentration of nano-plastics with MS would include plastics to the total pool of suspended particulates in the environment. Studies also showed that the impact 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-plastic particles. To assess this, we generated MS-associated nano-plastics from plastic debris collected from a subarctic northeast Atlantic region and nanometre-sized polystyrene spheres. Analysis of the nano-plastic-MS particles by 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. This study results suggest that pristine plastics are the major influencer in altering the bacterial communities associated with MS particles.

WE412 Tracking nanoplastics in marine bivalves at environmentally realistic concentrations M. AL SID 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, Pesches 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 < 1µm, NPs) are among the most important environmental issues faced by government and research agencies. The environmental concentration is expected to be above a part per billion. Detection 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 nanoplastic 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 nanoplastics at environmentally realistic concentrations in marine bivalves.


Plastic pollution is a widespread concern worldwide. Substantial amounts of plastics are produced and discharged into the environment every year, which will potentially impact aquatic ecosystems and consequently aquatic organisms. Plastic in the aquatic environment can undergo mechanical, chemical and biological degradation that can give rise to the formation of nano-plastics, which can be denominated as micro- (< 1 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, nanoplastics (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
environmental samples will be evaluated at the nanoscale in three key marine species, the cryptophyte algae Rhodomonas sp, the harpacticoid copepod Tisbe battagliai and the blue mussel Mytilus edulis and compared to its microscale counterpart. The uptake, accumulation and elimination kinetics of NPs in the three species will be evaluated under ecologically relevant conditions, as well as their potential transfer 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. Vijver, CML Leiden University / Conservation Biology probability.

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 studies is caused by interactions 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 ENMs (e.g. shape, 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 quantitative 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 microorganisms 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

H. PARK, Hanyang Univ. / Regulatory Chemical Analysis & Risk assessment Center; S. ok, Kitech / Regulatory Chemical Analysis & Risk assessment Center; M. Song, J. Ra, Korea Institute of Industrial Technology

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 rough road and raised road 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 diesel power and 24hr operation capacity and 10kWh supplementary battery system of 100kW assisting and initiating the system on arrival and in between generator exchange. Vibration testing shakers 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 Enneering; 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 biochemical processes and trophic interactions 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 smaller accumulation in the gills. The highest concentration of Ag was 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 (magnehite) nanoparticles in the guppy Poecilia reticulata

G. Qualhato, Federal University of Goias / Department of Morphology; T.L. Rocha, University of Algarve / CIMA; R.M. Saboia-Moraes, 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 organism 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 have 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; -95.9 ± 6.5 mV). The histopathological results showed an increase in the frequency 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) 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; nanoecotoxicity; guppy. Session: Ecotoxicology and human toxicology; from molecules to organisms, from omics to in vivo (Fish model species in human and environmental toxicology) Presentation preference: Poster presentation

(Edo)cotoxicity tests for hazard evaluation of recycling materials and waste (P)

WE418 Biotoxest for Hazardous Waste Classification (HP14): benchmarking Limits for Tolerable Ecotoxicity.

R. Welters, VITO / ABS; E. Rossi, OVAM; G. 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 used to label the waste. A new classification method is introduced where the chemical composition is taken into account to propose the chemical evaluation as step 1 biotests in steps 2 (acute aquatic tests on eluate fractions) and step 3 (acute terrestrial 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 biotest results against waste materials that were proven to be toxic in step 1, and it was conclusion that LDL As TE was a suitable option for our data set. The main conclusions were: The proposed set of biotests is essential for proper HP14 evaluation. Poor mass balance in the chemical data should be the trigger for the additional use of biotests. LID 4 is proposed as TE
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 of the different chemicals present with the 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, additives and antagonists) 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 (HPI4). Test strategies will allow wood wastes to be recovered or recycled.

WE420
QUALITY STANDARDS FOR URBAN WASTE FERTILIZERS: PUTTING ECOTOXICOLOGY IN THE PICTURE
S. Chelinho, CFE - Centre for Functional Ecology / Department of Life Sciences of University of Coimbra; C. Venâncio, Department of Biology / Biology; L. 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. viridisissmu > R. subcapitata > C. vulgaris > H. incongruens > B. calyculus. The observed toxicity was probably related with 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. Janke, H. 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 are carried 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
N. Bandow, Federal Institute for Materials Research and Testing / Contaminant Transfer and Environmental Technologies; F. Jürgens, BAM Federal Institute
Materials Research and Testing; U. Schoknecht, BAM Federal Institute for Materials Research and Testing
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 16637-2 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 concentration do not represent necessarily environmental concentrations. Thus, the concentration in the leaching test cannot simply be converted to limit values of 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 in situ amendments in soils and sediments (P)

WE423
Assessment and management of stormwater on sediment recontamination: you don't need to measure everything, just the right things
I. Drygrainnakki, Texas Tech University / Department of Civil Environmental and Construction Engineering; C. G. Pitt, The University of Alabama; E. Strecker, B. Steets, M. Otto, GeoEye Consultants
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. Water samples 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 stormwater. 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 Se and Ni. The study 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
L. Sem, Norwegian Geotechnical Institute; P. di palma, IRSACNR; C. Riccardi, INAIL; E. Eck, s.e. hale, Norwegian Geotechnical Institute; p. viotti, Università La Sapienza; M. P. Papini, Università La Sapienza / Chemistry
Petroleum is extensively used for making oil-based chemical and energy; its daily 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. viridisissmu > R. subcapitata > C. vulgaris > H. incongruens > B. calyculus. The observed toxicity was probably related with 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.
The impact of two biochar-based amendments (one from pruning trees and other from sewage sludge) on metal toxicity and bioavailability was assessed in an acidic environment. Benthic invertebrate community composition, and chemical analysis was measured during a chamber study design, where biochars were exposed to field conditions. 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 th...
WE429 Identification, Quantification, and Risk Assessment of Polycyclic Aromatic Hydrocarbons and their Polar Derivatives in Soil After Steam Enhanced Extraction L.S. Ting, Oregon State University / Chemistry / Environmental and Molecular Toxicology; E. Z. Harger, Oregon State University Environmental Protection Agency / Ground Water & Ecosystems Restoration Division; S.L. Massey Simonich, Oregon State University / Department of Environmental and Molecular Toxicology

Polycyclic aromatic hydrocarbons (PAHs) are environmental contaminants produced from the incomplete combustion and pyrolysis of organic matter, and are among the major contaminants in soils. Steam enhanced extraction (SEE) is an in situ thermal remediation technique that uses the addition of steam to soil subsurface to increase the removal efficiency and recovery of volatile and semi-volatile contaminants, like PAHs. However, there is limited research on formation of PAH derivatives during SEE of PAHs. Polar PAH derivatives are more mobile in the environment than PAHs, and some are more toxic than corresponding PAHs. There is an urgent need for analytical methods that can accurately quantify PAH derivatives in complex matrices, to better understand the chemistry occurring during SEE, and how it interferes with the remediation of soils. In this study creosote-contaminated soil from the Wyckoff/Eagle Harbor Superfund Site in Washington, USA was thermally treated with laboratory-scale SEE. Soil pre- and post-SEE, effluent collected during SEE, and pre- and post-SEE leachate samples (mimicking rain runoff and groundwater) were collected and analyzed with gas chromatography/mass spectrometry (GC/MS). Lor PAHs, polar PAHs, and MW302-PAHs (n=97). Most of analyzed PAHs were quantified in all pre-, post- and effluent samples. PAHs decreased significantly, while polar PAHs increased in mean concentration post-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 volatiles and polar derivatives to risk assessments to account the full effectiveness of SEE and prevent underestimation of potential risks. A quantitative risk assessment will be performed by calculating B[a]P₄, 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-leachate 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.

WE430 Enhanced total petroleum hydrocarbon removal without soil disturbance by serial surfactant foam spraying R. Bajagain, Y. Park, Kunsan National University; S. Jeong, Kunsan National University / Department of Environmental Engineering

Fuel spills are a complex mixture of hydrocarbons. Molecular 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 surfactant foam spraying technology, which avoids disturbing the soil, to deliver chemical oxidant and oil-degrading microbes to unsaturated soil. Hydrogen peroxide was used for the pretreatment of diesel oil. 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 three 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)).

WE431 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. Seo, 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 ionicize 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 phenols played important roles. There 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 existing 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 upgraded biochar as a sorbent for various types of contaminants.

WE432 Biochar for soil management: interactions with legacy contaminants and current-use pesticides L. Bielski, 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 many benefits for the uptake and adsorption of contaminants. Its efficient sorption properties make 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 soil structure and cation availability, protection of microorganisms, and increased 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 reducing bioavailability 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 epoxidazol and tbeconazol as representative of legacy contaminants and broadly used fungicides, respectively. Bioavailability was assessed by means of solid-phase microextraction as well as the fractionation of chromate to a set of fractions. The simulated exposure of BC as a model ecotoxicologically relevant organism. The influence of biochar properties and biochar dose was considered. At the same time, the well-being of earthworms in biochar-amended control soils was detected. This study aimed at balancing the dosing of biochar in soil management to ensure both an efficient and sustainable control of diffusive contamination and of pests.

WE433 PREPARATION AND CHARACTERIZATION OF COMPOSITIONS OF TYPE CLAY / POLYMERS AND THEIR USE IN THE REMOVAL OF CONTAMINANTS ORGANICS OF AQUATIC ENVIRONMENTS. M.S. Rodrigues, Instituto Federal do Maranhão; L. Aquil Vieira, Universidade Federal do Maranhão; A. Costa Filho, S.G. RIBEIRO, Universidade Federal do Maranhão

Clays have been used 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 material that can be gained through classical and scientific techniques. Adsorption 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-bromoanilino polymer, the process of cation exchange, which generated organophilic characteristics for samples, allowing combodispersants of organic compounds were used. Serious damage to fauna and flora are caused by the use of phosphate and hydrophobic hydrocarbons. This leads to search for new materials aimed at removing it mainly aquatic environment. Given that the application of this work aimed at the preparation and characterization of organophilic clays, as well as to evaluate its adsorption rate of these compounds organic. 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 Dibromophenol, this leads to search for new materials aimed at removing it mainly aquatic environment. Given that the application of this work aims at enhancing the preparation and characterization of organophilic clays, as well as to evaluate its adsorption rate of these compounds organic. Para it was used a natural clay from the Carolina-MA region.
ad Sorption capacity the material was 40% in nature, reaching a value of 78.4% after modification, demonstrating 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. Boulemtou, RioTinto; L. Poizat, 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 and phytostabilisation, reusing them seems 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 mostly 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 RHIZOTests (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 chemical extractions 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 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. Strand, University of Helsinki / Department of Ecological Sciences; C. Giestel, 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 invertebrates, using sublethal concentrations of 0.02 %, 0.06 %, 0.17 %, 0.5 % and 1.5 % of PES 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 enchytraeid 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 0.17 % and 0.5 %, 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 orbibatid 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 enchytraeid worms, whilst arthropods were not markedly affected by the fibers. However, not only enchytraeid worms (E. crypticus), but also isopods (P. scaber) and the earthworm E. fetida ingested fibers. Exposure of earthworms to the polymers was not related with the fiber concentration in the soil. As the accumulation of microplastics in soils is evident, the effects of different types of microplastics and their possible role as carriers of xenobiotics to soil organisms need to be further investigated. This study is part of the project IMPASSE – Impacts of Microplastics in Agro Systems and Stream Environment.

TH002 Effects of microplastic particles of polyhydroxybutyrate towards photosynthetic aquatic organisms
M.G. Pleiter, Universidad Autónoma de Madrid; G. AMARIEL, Universidad de Alcalá; M. Tamayo-Belda, Universidad Autónoma de Madrid; F. Leganes, Universidad Autónoma de Madrid / Biology; R. Rosal, Universidad de Alcalá; F. Fernandez-Pilhas, Universidad Autónoma de Madrid / Biology
Nowadays, the ecological impact of microplastics in freshwaters are not well understood [1]. Here, we have investigated the effects of microplastic particles (nominal size 5 nm) 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 by nanomaterials characterization (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 photoautotrophic algae and their effects in 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
M. Revel, Catholic University of the West / UBL, Mer Molecules Santé; I, lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Biotechnology; M. Fossi, C. Panti, University of Siena / Department of Physical Sciences, Ettajani, Catholic University of the West / UBL, Mer Molecules Santé; M. Bruneau, Catholic University of the West / UBL MMS Angers; F. Akcha, R. Sussarellu, J. Rouxel, IFREMER / Laboratoire décotoxicologie; P. Decottignies, B. Cognie, Universite of Nantes / UBL MMS Nantes; A. Chatel, Catholic University of the West / UBL, Mer Molecules Santé; C. Mouneyrac, Université Catholique de Louvain / UBL, Mer Molecules Santé
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. Several studies have reported the 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, France, in exposure tests 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 evaluated as well as LPO measurement (immunoassay) and DNA damage. 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 polyethylene microplastics at molecular and histological levels.
G. Limon, Universidad de Sevilla / Department of Physical Sciences, Earth and Environment; A. Mancia, L. Abelli, Universidad de Ferrara / Department of Life Sciences and Biotechnology; M. Fossi, C. Panti, Universidad de Sevilla / Department of Physical Sciences, Earth and Environment

421 SETAC Europe 28th Annual Meeting Abstract Book
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 complex mixture 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 polyurethane 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 sequencing to identify the transcriptome changes induced by the MPs. In addition, the gastrointestinal tract and the gills were dissected and fixed for histology and immunohistochemistry. The exposure to polyethylene and polyurethane 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.

TI005 Uptake and Effects of Synthetic and Natural Microparticles in the Shrimp Palaeomon varians
M. Weidung, University Duisburg-Essen; R. Saborowski, 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 artificial and natural particles on the induction of cellular stress in the Atlantic ditch shrimp (Palaeomon 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 suprarectal mesh and the digestive gland. Decapods were fed 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 Schyff, North-West University / Unit for Environmental Sciences and Management; J. Karstensen, GEOMAR

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 complex mixture 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 polyurethane 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 sequencing to identify the transcriptome changes induced by the MPs. In addition, the gastrointestinal tract and the gills were dissected and fixed for histology and immunohistochemistry. The exposure to polyethylene and polyurethane 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.

TI007 Effects of dietary microplastic exposure on fish intestinal physiology
G. Asmonaite, 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 animals, including fish. 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 polyurethane (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-virgin) or particles exposed to environmental degradation conditions (TEPA, TEPA/PAH, SCCIA/A) together with uptake rate of H-llysine. Overall morphology was observed using histology. Gene expression analysis of immune related genes (TGFβ, TNFα, IL-8, IL-10, IL-17, IL4/13A) and tight junction proteins (Occludin, ZO-1, Tricellulin) was performed to examine if PS particles and chemical contaminants induced inflammation in intestinal tissue. The innate immune response (lysozyme stability and complement system) in blood plasma was evaluated to assess the presence of systemic inflammation. 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
M. Martins, Faculty of Sciences and Technology, Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering; I. Neves, University of Lisbon / Department of Sciences and Engineering; P. Sobral, MARE - Faculty of Sciences and Technology, Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering; J. Neves, Faculty of Sciences and Technology, Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering; P. Sobral, MARE - Faculty of Sciences and Technology, Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering; M. Costa, MARE - Faculty of Sciences and Technology, Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering

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 and 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 juvenile Mediterranean sea bream (Solea senegalensis), using diet as the microplastic vehicle. For this purpose, four distinct diets were prepared using commercial fish pellets incorporated with two MP 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 sampled from each treatment and excised. The liver and stomach, from which fish were excised and 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 assayed, namely the lipid peroxides, glutathione and the activity of catalase, superoxide dismutase and acetylcholinesterase. 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
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 Ocean 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 nanoparticles (NP, 50 nm) on the fitness of the marine mussel *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 NP-treated mussels this effect was accompanied by a decreased phagocytic activity. Lysozyme activity in hemolymph was affected by either MP or NP treatments. In general, the mussel showed a decreased 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 150 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 scaled 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 treatments (1.5 and 15 ng/L) was significantly increased at 1.5 ng/L NP (HSI = C) and at 15 ng/L NP (HSI = D). Overall, results show 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.

**TH012**

**The sub-lethal impact of polystyrene microplastics and nanoparticles on the Mediterranean mussel *M. galloprovincialis***

M. Capolupo, erasmus muddus Ph.D in Marine and coastal management (MACOMA) - University of Cadiz / Inter-Departmental Research Centre for Environmental Science (CIRSIA); P. Valbonesi, University of Bologna / Department of Biological, Geological and Environmental Science (BiGea); S. Fracastoro, University of Cagliari / Department of Biological, Geological and Environmental Science (BiGea); E. Fabbi, University of Bologna / Bigea Department via Selmi Bologna

Although 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 Ocean 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 nanoparticles (NP, 50 nm) on the fitness of the marine mussel *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 NP-treated mussels this effect was accompanied by a decreased phagocytic activity. Lysozyme activity in hemolymph was affected by either MP or NP treatments. In general, the mussel showed a decreased 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 150 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 scaled 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 treatments (1.5 and 15 ng/L) was significantly increased at 1.5 ng/L NP (HSI = C) and at 15 ng/L NP (HSI = D). Overall, results show 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.
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 E. Bergami, G. Liberato, University of Siena / Department of Physical, Earth and Environmental Sciences; C. Manno, C.M. Waluda, British Antarctic Survey; S. Cappello, CNR IAMC; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

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 stressor on krill population. In this study we investigated the effects of model microplastics (< 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‰, 2°C) was also investigated by Dynamic Light Scattering. PS-COOH formed nanoscale aggregates (average size of 862 nm) in Antarctic NSW, 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 αβ γ gene in new findings reported for other Antarctic 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 our control. Our findings demonstrate that PS NPs are able to affect swimming behaviour, cuticle formation and FP 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 I. Brandt, M. Teles, Universidade Autonoma de Barcelona; A.P. Gonçalves, B. Becerro, University of Aveiro / Biology Department & CESAM; L. Guirado, D. Franco-Martinez, A. Tvarijonavicute, Universidad de Murcia; M.A. Martins, University of Aveiro / Chemistry Department & CICECO; A.M. Soares, University of Aveiro / Department of Biology & CESAM; L. Tort, Universitat Autonoma Barcelona / Department of Cell Biology Physiology and Immunology; M. Oliveira, University of Aveiro

Physico-chemical and 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 carbamazepine (cbz) toxicity on bivalve Mytilus galloprovincialis. Mussles were exposed for 96 h to the EC50 and EC10 concentrations, in new findings for the species. Molecular and biochemical biomarkers were evaluated in digestive gland, gills and hemolymph. Abundance of mRNA in digestive gland and gills revealed significant alterations in expression of genes associated with biotransformation, DNA repair, cell stress-response and innate immunity. Combined exposure induced significant down regulation in gene expression when compared to individual exposure. Total antioxidant status values suggest oxidative stress after exposure to 0.5 mg/L PSNP, whereas increased total antioxidant capacity and esterase activity suggest activation of antioxidant defenses after exposure to 50 mg/L. PSNP Exposure to 0.05 and 0.5 mg/L PSNP induced effects on neurotransmission in hemolymph. In gills, almost all experimental exposures induced inhibition of AST and ALT values. Genotoxicity was found in hemocytes after exposure to PSNP, cbz and their mixture.

TH016 The role of microplastic size and type on PAH sorption and bioavailability to copepods L. Sorensen, SINTEF Ocean / Environmental Technology; E. Rogers, Norwegian University of Science and Technology; M.U. Ronsberg, SINTEF Ocean / Environmental Technology; D. Aitn, BioTrix; A. Booth, SINTEF Ocean / Environmental Technology

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, MP's 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 accurate determination of PAH release from adsorbed compounds or fragments 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-dimethylpyrroline) to a range of different MP's in natural seawater. The selected PAHs exhibit different sizes and hydrophobicities, thus having varying surface solubility (two to three orders of magnitude). In the case of the low molecular weight 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 microbads, 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 finnarchicus) 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. ’

TH017 Limited influence of microplastics on the effects of an endocrine disruptor on the African clawed frog (Xenopus laevis) S. Rehse, Leibniz-Institute of Freshwater Ecology and Inland Fisheries / Ecophysiology and Aquaculture; A. Zikova, W. Kleiner, W. Kloas, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; C. Zarfl, University of Leibniz-Ingeningen / Centre for Environmental Research, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; S. Rehse, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; C. Zarfl, University of Leibniz-Ingeningen / Centre for Environmental Research, Leibniz-Institute of Freshwater Ecology and Inland Fisheries

Microplastics are ubiquitously distributed in freshwater ecosystems. Besides effects on the algae, 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) are of high relevance for aquatic ecosystems. In a study focused on potential and specific endpoints of microplastics on African clawed frog (Xenopus laevis) we tested microplastics with different sizes, shape (particles, fibres) and polymer compositions (polystyrene and polystyrene microbads, polyster microfibres) were used. Using a novel approach, the influence of MP sorption on PAH bioavailability to two marine copepod species (Acrtia tonsa and Calanus finnarchicus) 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. ’

TH018 Genetics of POPs sorption and plastic additives release to a variety of polymers under aquatic conditions D. Herze, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; K. Sakaguchi-Soeder, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; R. Sempere, V. Faurelle, Mediterranean Institute of Oceanography, Marseille; A. Booth, SINTEF Ocean / Environmental Technology

The PLASTOX project investigates the ingestion, food-web transfer, and ecotoxicological impact of microplastics (MPs), together with the persistent organic pollutants (POPs), metals and plastic additive chemicals associated with them, on key European marine species and ecosystems. PLASTOX combines field-based observations, laboratory tests and manipulative field experiments to study the ecological effects of MPs. The use of common microplastic reference materials, including a marine litter-derived MP produced from an environmentally weathered fish box, allows a meaningful comparison of data generated by different partners and across the different activities of PLASTOX. As part of a long-term field experiment conducted at marine locations across Europe (Mediterranean to

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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 SPMD 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, 6 months and 12 months after deployment. Hydrophobic persistent organic pollutants such as PCBs, PCDDs, PBDEs and PAHs were used to establish the adsorption kinetics in seawater under Arctic conditions. Samples were extracted using ultrasound and nonpolar solvents, followed by GC and SPE clean up. Analytical chemistries using GC/MS/MS and GCxGCMS was done in the laboratories of the TU Darmstadt and NILU, Tromsø. In addition, release kinetics of common persistent organic pollutants (polychlorinated biphenyls, organophosphate esters, bisphenol A and fluorinated chemicals) 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.

TII019
Characterization of microplastics present in personal care products and the study of its toxicity mixed with chlorpyrifos on juveniles of Solea senegalensis. G. Albení, Facultad de Ciencias de la Salud (Spain) / Toxicology Area; I. Cabrera, Universidad de Cadiz / Toxicology Area; M. Ríos, Universidad de Cadiz / Environmental Technology; B. Carrero, Universidad de Cádiz / Toxicology Area.

In the last years, different components from personal care products have arrived at aquatic ecosystems because these products are not biodegraded or removed in wastewater treatment plants. Some of the personal care products contain plastic microbeads such as exfoliating shower gel, toothpaste and make-up creams. Commonly used and available in supermarkets of our area were used by these assays. The microphases available in these samples were separated and chloroprepurified. The particles were identified by Fourier transform-infrared spectroscopy (FT-IR) analysis. The spectra are recorded in reflection mode in the spectral range 4000-650 cm\(^{-1}\) by adding 128 scans at a resolution of 4 cm\(^{-1}\). The particles were identified by comparing the FT-IR absorbance spectra of the microplastics to those in a polymer reference library. The microplastics were used in toxicity test. 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 12 h light/12 h 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: 5-10-20 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 biomarkers to diagnose exposure of natural populations to harmful chemicals by monitoring changes in their activity and different hydrophobic organic chemicals (HOCs) in the aquatic environment. There is an ongoing debate about MPs as vectors for chemical contaminants and their relative importance compared to other naturally occurring particulates. The goal of present study was to quantify particle-mediated chemical transfer and using biomarker approach examine associated biological effects in three spined stickleback (Gasterosteus aculeatus) and daphnia (Daphnia magna). AChE and BChE were then quantified at mRNA level in the liver and gut. Acetylcholinesterase (AChE) activity was measured by a number of bioluminescent reactions. Results showed that all treatments containing chemical mixtures caused changes in gene expression and altered enzymatic AChE activity. Differences could also be seen between particle types, where PS contaminated particles showed similar pattern with the non-particle plastics, while PE contaminated particles showed some peculiarities with respect to the chemical transfer of chemicals. The chemical control was for some treatments lower than the particle groups indicating a particle-mediated chemical transfer in fish.

TII020
Are microplastics inhibitory to Daphnia magna and are they significant vectors for hydrophobic organic pollutants? C. K. Frydryker, 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 studies aiming at elucidating the adverse effects resulting from dietary exposures to PS particles ranging in size 50-250 µm. In this study, rainbow trout (Oncorhynchus mykiss) were exposed to diets, enriched with PS particles (10 ng of PS particles/Fish/day) for 28 days. We used environmental contaminated PS particles from a site exposure from two environmental matrices (undiluted sewage effluent and industrial harbor runoff). As PS particles largely exceeded 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 via 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 exposure. The findings of this study revealed an indication of NRF2-mediated oxidative stress regulation.

TII021
Microplastics as vectors for hydrophobic organic chemicals in fish: a comparison of two polymers and silica particles, using three different model compounds M. Trefil, University of Gothenburg Sweden; A. G. Asmonaite, University of Gothenburg / Department of Biological and Environmental Sciences; E. Westberg, IVL Swedish Environmental Research Institute; B. Carney Almroth, University of Gothenburg Sweden / Department of Biology and Environmental Sciences

Plastic pollution is a recognized global issue and the ubiquity of microplastics (MPs) in aquatic environment is a cause for concern. Potential effects on organisms are still not fully understood, and mechanistic understanding, required to fully reveal consequences of exposure, especially in connection to chemical mixtures, is still lacking. Studies have shown that MPs have capacity to adsorb and concentrate hydrophobic organic chemicals (HOCs) in the aquatic environment. There is an ongoing debate about MPs as vectors for chemical contaminants and their relative importance compared to other naturally occurring particulates. The goal of present study was to quantify particle-mediated chemical transfer and using biomarker approach examine associated biological effects in three spined stickleback (Gasterosteus aculeatus) and daphnia (Daphnia magna). AChE and BChE were then quantified at mRNA level in the liver and gut. Acetylcholinesterase (AChE) activity was measured in brain. Results showed that all treatments containing chemical mixtures caused changes in gene expression and altered enzymatic AChE activity. Differences could also be seen between particle types, where PS contaminated particles showed similar pattern with the non-particle plastics, while PE contaminated particles showed some peculiarities with respect to the chemical transfer of chemicals. The chemical control was for some treatments lower than the particle groups indicating a particle-mediated chemical transfer in fish.
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 MPs 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 A. Catarrino, A. Homer, Heriot Watt University / ILES; L. Duran Saja, Heriot Watt University / ILES; G. D. Drymon, University of Plymouth / EPOC, University of Plymouth / EPOC Centre for Environmental Sciences and Engineering; A. dos Santos, Faculdade de Ciências Farmacêuticas - USP / Departamento de Análises Clínicas e Toxicológicas; M. Al-Sid-Chiekh, ISMERUQAR; A. J. Sweetman, Lancaster University / Lancaster Environment Centre; T. B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society

Nanopolystyrene (nPS, ≤ 1 µm) may result from larger plastic debris released in the environment and can pose a risk to marine organisms and ecosystems. The risk of MPs can be exacerbated because toxics sorbed to MPs may be transported to and become more bioavailable in organisms. It is likely that MPs 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 MPs on benthic meiofauna assemblages. It is critical to understand impacts of MPs 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 nanopolystyrene (nPS) and nPS with the sorbed co-contaminant Tributyltin (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). Nematodes were collected from the sediments using a microfuge equipped with oxygen microsensors. Changes in the nematode community structure were measured by assessment of changes nematode diversity (nematodes identified to genus) and dose responses analysed 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 MPs and their co-contaminants within a relevant scenario.

TH024 Nanopolystyrene Induces a Decrease in the Oxygen Uptake of Zebrafish Larvae by Enabling Sorbed Benzo[a]Pyrene Bioavailability A. Catarrino, Heriot-Watt University / ILES; M. Clement, Polytex Nice Sophia; M. Tait, Heriot-Watt University; D. Boyle, Plymouth University; M. AL SID CHEIEKH, University of Plymouth / Marine sciences and engineering; T. B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society

Microplastics (MPs, 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 nanoplastics (nPS, 500 nm) by measuring oxygen uptake in the sediment (cytocrictomachia reni) and in Zebrafish larvae (Danio rerio) larvae (9th postfertilization, hpf). The effects of BaP and nPS with sorbed BaP on larval (96 hpf) metabolic rate were assessed over a 24 h exposure. The concentrations tested for nPS and nPS with sorbed BaP were 0–50 µg/ml and 0–40 µg/l for BaP. Proof of particle ingestion by larvae was observed using fluorescent SPS (500 nm, µg/ml). Whole-organism metabolism rate (O2) and organ specific oxygen uptake rates (g oxygen uptake chomophore mass spec spectrometer (GC-MS)). Preliminary dose-response analysis showed that nPS, BaP and nPS with sorbed BaP induced a decrease in O2 by zebrafish larvae, indicating a higher energetic cost of physiological functions maintenance. The expression of cyto1A was up to 9 fold change in the highest concentration of nPS (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 more environmental 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; 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. Navarro, University of the Basque country UPV/EHU; M. P. Cajaraville, 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

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 pollutants (0.5 and 4) on marine bivalves. We detected the adsorption of benzo[a]pyrene (BaP) on mussels Mytilus galloprovincialis in order to elucidate the effects of MP size and the presence of adsorbed contaminants on the organism. MPs were provided daily, mixed with algae, at 1000 particles/ml/day, during 7 and 26 days. Effects were determined on early cellular biomarkers (catalase activity [CAT], neutral red uptake [NR] and DNA damage in hemocytes; histopathology in digestive gland) and on cellular biomarkers (scope for growth [SGF] and condition index). Chemical analysis showed that BaP concentrations in mussels increased with time (up to 150 times greater than background levels) and that smaller MPs posed an increased hazard in terms of the transfer of adsorbed BaP. In histology, large MPs were abundant in the lumen of stomach, mixed with stomach contents, and in the lumen of digestive tubules (DTs), associated to cell debris. Occasionally they appeared within epithelial cells of the stomach, ducts and DTs and in the connective tissue. Small MPs were also abundant in the lumen of stomach. In all samples, DT appearance indicated a high digestive activity, confirmed by hexosaminidase histochemistry. Overall, effects in all treatments increased with exposure time. Increased effects of MPs+BaP compared to MPs alone were found in NR and CAT activity, DNA damage and biomarkers (scope for growth [SGF] and condition index). We anticipated that our results (ongoing data analysis) will contribute to a better understanding of the effects of a variety of plastic type, size, shape combinations together with a wide variety of pollutants. Funded by Spanish MINECO (NACE project CTM2016-81130-R), Basque Government (consolidated group IT8810-13) and UPV/EHU (UFI 11/37, VRI grant PLASTOX). Work carried out within EU project PLASTOX (IPI Oceans 005/2015).

TH026 Characterization of the adsorption/desorption of benzo[a]pyrene from polystyrene micro- and nanoplastics for further toxicity assessment I. Martinez, University of Basque Country; K. Le Menach, UMR CNRS EPOC Université Bordeaux / EPOC UMR 5805; M. Dèvier, University of Bordeaux / EPOC / LPTC; UMR 5805 CNRS, M.P. Cajaraville, 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; A. Orbea, 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; J. Lacave, 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

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 sorbed on other pollutants present 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 nanoplastics (nPS, 500 nm) by measuring oxygen uptake in the sediment (cytocrictomachia reni) and in Zebrafish larvae (Danio rerio) larvae (9th postfertilization, hpf). The effects of BaP and nPS with sorbed BaP on larval (96 hpf) metabolic rate were assessed over a 24 h exposure. The concentrations tested for nPS and nPS with sorbed BaP were 0–50 µg/ml and 0–40 µg/l for BaP. Proof of particle ingestion by larvae was observed using fluorescent SPS (500 nm, µg/ml). Whole-organism metabolism rate (O2) and organ specific oxygen uptake rates (g oxygen uptake chomophore mass spec spectrometer (GC-MS)). Preliminary dose-response analysis showed that nPS, BaP and nPS with sorbed BaP induced a decrease in O2 by zebrafish larvae, indicating a higher energetic cost of physiological functions maintenance. The expression of
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 adsorption 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 adsorption of polystyrene microbeads and whether this process was directly dependent on plastic size. Results indicated that the presence of a higher adsorption capacity of BaP than 4.5 µm MPs. The percentages of adsorbed BaP/P from the total Ba/P 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 adsorption process of Ba/P on MPs and is currently being applied to NPs. Funded by French ANR (N°: 10-EXP-03-10, Cluster Excellence COTE-ANR:10-LABX 45), Spanish MINECO (NACE project — CTM2016-8130-R), Basque Government (consolidated research group IT8010-13) and UPV/EHU (UFI 11/37 and grant to IMA).

TH027 Occurrence of microplastics in epibenthic and sediment-dwelling species in a Norwegian fjord a borough. ECOLAB UMR2545 CNRS UPS INPT; C.G. Avio, L. Pittura, S. Gorbi, Department of Life and Environmental Sciences, Politecnico 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 aimed to report the presence of microplastics in ten sediment-dwelling and epibenthic species representative of different feeding modes and trophic levels. The species analysed 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 (fibres, 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 35%, 25% and 15% respectively. We hypothesize that maritime and fish feeding activities are 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, polycrylic, 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. Neve, Toulouse University Darmstadt / Department of Civil and Environmental Engineering; A. Goharnia, 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; M. Gottschling, Y. Zhou, I. Schwabe, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; A. van Oyen, Plastic Partner GmbH; L. Schebek, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; K. Sakaguchi-Soeder, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany. 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 = 3.3), phenanthrene (log Kow = 4.46) and fluoranthene (log Kow = 5.16). The plastic samples tested here are LDPE pellets with a low amount of additives. LDPE pellets were previously characterised by CARAT GmbH (Bochum, Germany). For the spiking method, batch reactors 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, on 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. Riet, DTU (Technical University of Denmark) / Department of Environmental Engineering; B.C. Alnroth, University of Gothenburg / Department of Biological and Environmental Engineering; T. Neve, Technische Universitaet Darmstadt / Department of Civil and Environmental Engineering; 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 estimation of the impact of POPs-loaded MPs on a range of marine organisms. In general, hydrophobic pollutants like POPs are first extracted from the matrix, in this case the plastic debris, using non-organic solvents. Then the purified POPs will be cleaned-up and analysed in e.g. gas-chromatography/mass-spectrometry (GC/MS). Some non-polar solvents applied for POP extraction, however, may dissolve plastic debris partially or completely, which disturb subsequent analyses. A number of methods have been reported for the extraction of POPs from MPs. Yet, the validity of these methods have not been fully discussed and the influence of polymers in extraction solutions in subsequent POP analysis has not been thoroughly investigated. The goal of the current study is the development of an optimal analytical protocol to extract POPs from different MPs. Known amounts of POPs were artificially charged on the surface of selected polymer particles, including preproduction resin pellets from different polymer type (polystyrene, PS; polyethylene terephthalate PET; polypropylene, PP; poly vinyl chloride, PVC) in the laboratory. The POPs on plastic particles were extracted in selected solvents using soaking and sonication methods under different conditions. Solvents used in this study include n-hexane (nHex), isopropanol (iPrOH) and dichloromethane (DCM). Extraction methods and conditions were evaluated for a high extraction recovery, a high reproducibility, as well as for a minimal damage of polymer particles, i.e. carriers of POPs. The recovery rate and analytical reproducibility of POP was determined using gas chromatography-mass spectrometry (GC/MS). The loss of plastic weight was measured for the evaluation of the stability of plastic particles under given extraction conditions. Further, we investigated the influence of polymers dissolved in solvent on quantitative analysis for POPs. TU Darmstadt and CARAT are participants of an EU project “PLASTOX”, a consortium of a JPI Oceans’ Joint Action. TU Darmstadt is funded by BMBF.
Is the Arctic threatened by plastics? Identifying sources and determining the distribution of microplastics around Svalbard

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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 potential source, 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 depths throughout the water column, microplastics 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 species already burdened by multiple stressors, knowledge about sources, fate and concentration of microplastics in different environmental compartments is crucial.

Microplastics - an ecotoxicological issue? How to balance facts and perception without marginalizing an environmental problem

C. Völkel, ISOE - Institute for Social-Ecological Research / Water infrastructure and risk analyses; J. Kramm, ISOE - Institute for Social-Ecological Research

While plastic has 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 have led to an upswing of the debate. In the 2000s, small plastic particles that had already been detected in seawater in the 1970s were labeled “microplastics” for the first time. Since then, the number of studies on the occurrence and effects of microplastics has risen exponentially. 13 years after the publication of the article by Thompson et al. the question of whether microplastics actually pose a risk to the environment could not yet be answered conclusively. In laboratory studies, biological effects have so far only been detected at high particle concentrations with limited environmental relevance. Independently of this, the impacts of microplastics are perceived by the general public as a serious factor for environmental pollution. 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 depths throughout the water column, microplastics 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 species already burdened by multiple stressors, knowledge about sources, fate and concentration of microplastics in different environmental compartments is crucial.

Addressing species diversity in biotransformation: variability in expressed transcripts 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 expression transcriptome data from 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 were selected for broad phylogenetic coverage, as well as economic, research, and conservation importance, and included: sea lamprey (Petromyzon marinus), lake sturgeon (Acipenser floridanus), American eel (Anguilla rostrata), alligator gar (Atractosteus spatula), paddlefish (Polyodon spathula), rainbow trout (Oncorhynchus mykiss), rainbow smelt (Osmerus mordax), fathead minnow (Pimephales promelas), Antarctic icefish (Trematomus loyneri), common carp (Cyprinus carpio), and channel catfish (Ictalurus punctatus). In addition to comparing information across fish species, the resolved isoforms were compared to human xenobiotic metabolizing enzymes. This comparison aids in evaluating the utility of human-based biotransformation tools such as ToxCast chemical screening assays or metabolism prediction software for potential relevance in fish. The content of this presentation neither constitute nor necessarily reflect US EPA policy.

Metabolism of Organophosphate Flame Retardants (OPFRs) in Freshwater Fish: Field and Laboratory Studies

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Organophosphate flame retardants (PFRs), as widely used alternatives of bromine flame retardants, result in a wide array of organophosphate matrices. Despite the adverse effects of PFRs, many researchers have paid their attention on the absorption, bioaccumulation, metabolism and internal exposure processes of PFRs in wildlife and human. PFRs can be rapidly metabolized in the body. The general metabolic pathway of PFRs revealed by certain in vitro studies includes dealkylation, hydroxylation, carboxylation and oxidative dehalogenation and phase II conjugation, resulting in a wide array of metabolites. D-alkyl phosphates (DAPs) from the dealkylation metabolism were recently deemed important biomarkers in human biomonitoring studies. As very limited information is available on DAP metabolites in environmental biotic samples, we first investigated
the accumulation and tissue distribution of eight common OPs 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 exact contribution of PFRs and DAPs in wild animal studies. In the consequent laboratory control study, we screened the metabolites of alkyl-PFRs by in vivo exposure of *Gobiocypris rarus*. Metabolites of alkyl-OFPs 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 glucuronidation for all the tested three alkyl-PFRs. We identified and accurately quantified the metabolites 3-OH-TBOEP, BBOHEP, 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 alkyl-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 the improvement of new analytical techniques, many of these chemicals, the so-called “emerging pollutants” (EPs), are being currently identified and their bioaccumulation in the environment is being assessed. In this context, the present work evaluates the bioaccumulation and fate in PBs versus their RS prey is substantially influenced by the chemical nature of OPs. The bioaccumulation factor was calculated to be 110 L/kg. Finally, the data will be modelled using a kinetic model and the predicted bioaccumulation and transformation rate constants will enable determining the role of biotransformation in the detoxification of prochloraz in *Hyalella azteca*.

**TH037**

Toxicokinetics and metabolite identification of two emerging pollutants, Acesulfame-K and 4-MBC, in the manila clam *Ruditapes philippinarum*

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Marine ecosystems have been historically sinks for many pollutants and chemicals whose effects are known 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 toxicokinetics (TK) of two OPs (the UV filter 4-Methylbenzylidene-camphor (4-MBC) and the artificial sweetener acesulfame K (ACE-K)) in the Manila clam *Ruditapes philippinarum*, focusing on determining the bioconcentration factors (BCF) and identifying metabolites and biotransformation products. During 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 (MetabolynxTM) 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 53913 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 metabolised 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 transformation of 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 MetabolynxTM 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: Adipose Tissue Concentrations and In Vitro Depletion and Metabolite Formation

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Organophosphate esters are known to bioaccumulate and occur in the marine food web. In East Greenland is a contamination “hot spot” for long-range transported anthropogenic chemicals, including organophosphate esters (OPEs). High concentrations of OPEs have been reported in Arctic air (particles) while very little is known for wildlife although recent reports for Hudson Bay polar bears (*Ursus maritimus*) indicate that OPE residue levels in fat tissues are very low or non-detectable and appear to be strongly influenced by biotransformation. In the present study, the hepatic in vitro metabolism of six environmentally relevant organophosphate (OP) triesters and corresponding OP diester formation were investigated in East Greenland (Scoresby Sound region) polar bears (PBs) and ringed seals (RSs; *Pusa hispida*). The in vitro OP triester metabolism assay results were compared to fat (adipose) levels of selected OP triesters in field samples from the same individual animals. 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 tri (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 assay. Tris (1,3-dichloro-2-propyl) phosphate was completely converted to its corresponding diester. However, the mass balances showed that OP diester formation corresponding to tris (2-ethylhexyl) phosphate, tri (n-butyl) phosphate, and tris (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.

**TH039**

Proteomics of a metabolic simulation system - a look inside rat S9

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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 used in vivo bioassays that are not by themselves capable of a metabolic transformation. The most prominent example is the Ames bacterial reverse mutation assay according to the ISO 11350 or the OECD 471. However, the application for S9 is much more diverse and spans from various not guideline-based bioassays towards the ad-hoc production of metabolites for chemical analysis. It is also applied for stability testing of pharmaceuticals and the observation of the potential in bioaccumulation of chemicals and their metabolites. In this study, we look at the proteomics of multiple rat S9 products and compare them with an animal component free biotechnological alternative.

**TH040**

A critically evaluated database of in vitro and in vivo toxicokinetic data for

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mammals and fish
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Toxicokinetics (TK) plays an important role in ecological and human health assessments. In vitro testing is compared to TK data to evaluate the impact of chemicals subject to 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 ($k_t$) are key determinants and sources of uncertainty in bioaccumulation assessment. $k_s$ can be determined in vivo with whole animal models or from in vitro 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.C931336.X0) containing TK data (i.e., biotransformation rates) for fish and mammal species (i.e., rat, mice) derived from in vivo and in vitro (59 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 4,000 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 modellling - in vitro testing approach: derivation of kinetic rate constants for mammalian models
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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 extrapols of TK parameters, and predicted TK and in vitro–in vivo extrapolation methods. In a first step, we derived at a database of candidate chemicals for in vitro testing based on model discrepancies, availability of reliable in vivo BCF and BMD data, and availability of kinetic data for in vitro biotransformation rates. The resulting chemicals were divided into three $K_a$ categories based on predominant exposure route(s) to guide in vitro testing: 1) log $K_{ow} >$ 4 (aqueous exposure dominates – to be tested in gill and liver models); 2) log $K_{ow} = 3.5$ (mixed exposure routes – to be tested in gill, liver and intestine models); and 3) log $K_{ow} <$ 3.5 (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 models 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
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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 used in OECD guideline GL 195 for hepatic monooxygenase activation. A 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 / Physical and Environmental Science; K.L. Foster, 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), intrinsic clearance using cryopreserved rainbow trout hepatocytes and intrinsic hepatic clearance using cryopreserved rainbow trout S9 sub-cellular fractions. These metrics are 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
Effects of endocrine disruptors on reproductive health: A new approach to integrating ecotoxicological and human health data

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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 the 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 existence 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 and 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 biocriteria for each of the EDs.

Pros and cons of fish toxicity tests in detecting chemicals with endocrine disrupting activities

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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 adressing the pathway endpoints that indicate chemicals with EAS (endocrine 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 and 377 ecotoxicological studies, 377 fish tests and 295 include tests and 422 guidance 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 the EU legal frameworks. In the EU legal frameworks, these fish tests are required 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 selection should always consider the most factors as much as possible. 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 lifespan, the investigated EDs-related endpoints, their robustness (and to which extent these have been validated) and the species sensitivity in response to chemicals with EAS modes of action will be compared. To this aim, publicly available data on different fish species tested according to TGs or TG-like protocols will be collected and analyzed. Analyzing 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.

Towards developing a list of reference chemicals for endocrine assay validation

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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 public and environmental hazard, 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 in vitro and 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 inter-laboratory 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 considered throughout the testing. This presents challenges to regulators and researchers in selecting assays with needed sensitivity and/or appropriateness of use. Here, we attempt to identify the parameters that should be evaluated when selecting validation chemicals. These range from simple physical/chemical properties, to more complex information related to a known mode of action. Additionally, reference chemicals used during assay validation should span a range of potencies incorporating both positive and negative controls. Some well accepted and commonly used chemicals are provided as a realistic starting point to compile a set 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
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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.) were 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 the strategies to fill the matrix, is an important source of scientifically relevant information available for a certain chemical thus facilitating the data analysis to identify EDs. For example the data-matrix visually reports if certain toxicological endpoint has been observed across different studies to support evaluation of biological plausibility of a causative endocrine disruption. When evaluating 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 function 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 MoA 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. Affinity and activity can be measured in silico or in vitro screens may predict endocrine-related bioactivity and the potential for a chemical to interact with the endocrine system, affinity and activity, also known as mechanistic potency, can be used in comparison to that of the endogenous ligands to support or reject the biological plausibility of a causal link between an endocrine MoA and an adverse effect. A hypothesis testing, weight of evidence framework and case study examples will be used to illustrate the use of bioactivity and mechanistic potency data, along with other lines of evidence, in the assessment of endocrine activity.

TH053 Addressing endocrine concerns for the environment in dossier evaluations with an FSdT - possibility to avoid further vertebrate tests
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In the context of the REACH regulation, long-term toxicity testing on fish is a standard information requirement for substances manufactured or imported in quantities of 100 or more tonnes per year. Additionally, some substance properties, for example a low water solubility, lead to the necessity to conduct a long-term toxicity test on fish. If a data gap in a registration for long-term toxicity to fish is identified in the process of a dossier evaluation (Dev), the preferred option is to request a Fish early-life-stage Test (FELS - OECD 210). However for a substance with hints for endocrine disrupting properties, further tests would be needed to clarify the concern in a substance dossier. If possible, efforts should be made to avoid additional vertebrate tests in a SEV by requesting a Fish Sexual Development Test (FSdT – OECD 234) as a standard long term fish toxicity test under dossier evaluation. This would make it possible to clarify the endocrine disrupting properties of the substance, if they are revealable in an FSdT, what is the case for most oestrogenic or androgenic acting chemicals. Similar to the FELS the FSdT assesse early life-stage events. Therefore a test which will fill a data gap. The investigation of the endocrine disruptive potential of substances to environmental organisms is not explicitly part of the standard information requirements under the REACH Regulation. However, the legal text of REACH Regulation does not refer to a specific OECD test guideline, but to a type of the test. ECHA’s Guidance on Information Requirements R.7b indicates that the need to conduct further tests is triggered e.g. by information on a specific mode of action and unexpected sensitivity of a group of organisms to the substance under investigation. As the FSdT is a level 4 test, according to the OECD conceptual framework no other fish (vertebrate) test may be necessary to identify the substance as an endocrine
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 endocrinological characteristics (VTG, glucose). Both urban and agricultural exposures resulted in growth alterations between treatments most likely due to density-dependent growth. Urban exposure indicated higher fecundity (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 fewer adverse effects on fish, including disruption of TH signaling, including thyroid hormone receptors and deiodinases. Zebrafish embryos were then tested using TH055.

TH056

Assessing species sensitivity using environmental mixtures

S. Knoho, St. Cloud State University / Aquatic Toxicology Laboratory; N. Cipoletti, St. Cloud State University / Aquatic Toxicology Laboratory; L. Wang, St. Cloud State University; U. Hasbay, 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 both mixtures. In addition to estrogenicities of 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 agricultural or urban CECs. Utilizing the same method in vitro, alligator PPAR-gamma and RXR-alpha were examined. A PPAR-gamma activation is an effect of both AG and UB mixtures were estrogenic, however, their estrogenicities varied depending on isofoms of ESRs and 4 species. Human ESR1 was the most sensitive to AG based on their estrogenicities, while minnow ESR1 was the least sensitive to AG. Bluegill ESR1 was the most sensitive to UB based on their estrogenicities, whereas BG ESR2 was the least sensitive UB in receptors we tested. Both AG and UB CEC mixtures did not activate neither alligator PPAR-gamma nor RXR-alpha. Although further investigations of PPAR/RXR signal are required in fathead minnow, PRAR/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 endurance 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 disrupting effects and changes in fathead minnow larval performance exposed to diverse CECs. 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 change on apical endpoints as the complexity of CEC mixtures increases. We tested the hypothesis that as the complexity of CECs in exposures increase, the apical endpoints observed will differentiate 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. We also observed indication 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.

TH059

Contaminants of emerging concern in the North American Great Lakes: Load reduction and biological recovery after wastewater treatment upgrades

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Many urban aquatic ecosystems are becoming effluent dominated, resulting in the presence of contaminants of emerging concern and subsequent adverse effects on aquatic wildlife. Despite these dramatic alterations, effluent dominated urban systems support many ecosystem services and are used by the nearby human population for recreation. The Metropolitan Water Reclamation District of Greater Chicago upgraded two wastewater treatment plants (one million cubic meters/day) to disinfection (UV; chlorination/de-chlorination). The receiving aquatic ecosystem adjacent to these two wastewater treatment plants has been the focus of intense biological and chemical study for the past seven years and provides a unique opportunity to assess two divergent treatment technologies of wastewater. We also observed indication 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 multiparallel detection of biological effects caused by anthropogenic micro-pollutants

C.E. Reigard, German Federal Institute of Hydrology; L. Moscovici, 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 aquatic environment is challenging. The project "TREESS" [1] (TRacking Effects of Environmental organic micro-pollutants in the Subsurface) aims to develop an innovative technological platform for monitoring MPs based on the assessment of their biological effects. The proposed setup will be composed of the following steps: (a) Extraction and pre-concentration of MPs and their possible transformation products in soil or water samples by solid phase extraction. (b) Separation of the extracts using high performance thin layer chromatography (HPTLC). (c) Biological effect measurement of the individual separated constituents by using genetically engineered yeast (Saccharomyces cerevisiae) or bacterial (Escherichia coli) bioreporters. A main goal of our study is to develop tools and methods for a multiparallel effect detection covering a range of potentially adverse biological effects. This can be achieved by the construction of yeast strains using specific fluorescence reporters for the various endpoints to be detected. By coupling these strains with HPTLC and mass spectrometry, a wide variety of compounds with biological activity could be screened simultaneously. The first step of coupling HPTLC with different bioreporters was performed. (d) Determination of the endpoints (androgenic, thyroidogenic, genotoxic, dioxin-like effects, effects on the vitamin D and the retinoic acid receptor) was successfully performed by using various reference compounds. Furthermore, mixing yeast strains with different endpoints yielded to the detection of different adverse effects at the same time. Next steps will include (i) the analysis of real samples, (ii) the further development of additional sensor-strains and complementary testing methodologies and (iii) method development for the detection of compounds by chemical analysis after separation by HPTLC. <br clear="all"/> [1] A German-Israeli research and development project in the field of water technology within the framework of the BMBF-MOST cooperation, FKZ: 02W11387.

TH062

Endocrine disruptors used in polymers in the offshore oil and gas industry

C. Phillips, Cefas Lowestoft Laboratory / Science Directorate - advice and assessment; R. Suerhing, University of Toronto; A. Smith, Cefas / Ecotoxicology and Molecular Ecology

Contrary to the advice and assessment of regulatory assessors that a number of polymer substances found in products registered for use and discharged offshore as a result of the activities of the oil and gas industry are based on monomers that are known and suspected endocrine disruptors. These polymers were described by chemical analysis after separation by HPTLC. Endocrine disrupting monomer groups were identified by chemical analysis after separation by HPTLC. Endocrine disrupting monomer groups were identified by chemical analysis after separation by HPTLC. The detection of a series of biological endpoints (androgene, thyroidogenic, genotoxic, dioxin-like effects, effects on the vitamin D and the retinoic acid receptor) was successfully performed by using various reference compounds. Furthermore, mixing yeast strains with different endpoints yielded to the detection of different adverse effects at the same time. Next steps will include (i) the analysis of real samples, (ii) the further development of additional sensor-strains and complementary testing methodologies and (iii) method development for the detection of compounds by chemical analysis after separation by HPTLC. <br clear="all"/> [1] A German-Israeli research and development project in the field of water technology within the framework of the BMBF-MOST cooperation, FKZ: 02W11387.
Thyroid disruptor screening using zebrafish as vertebrate model

I. Iurria, O. Jaka, C. Martí, A. Alzuále, 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 a TR-β dependent fluorescent reporter construct, 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.

TH064 Development of stably transfected cell lines with zebrafish thyroid hormone receptors α/a and β/a for assessing endocrine disruption in environmental samples

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Endocrine-disrupting chemicals (EDCs) are ubiquitous in our environment and can be found in many products including food/feed, containers as plastic bottles or metal food cans, cosmetics, pesticides, flame retardants, detergents... Accordingly, they suppose a threat to animal and human health through different exposure routes. In vitro bioassays are valuable tools for detecting and studying EDCs action and provide a sensitive and rapid system to evaluate their potential effects. In addition, the transgenic vertebrates are 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. The aim of this work was to develop two cell lines stably expressing zebrafish (Danio rerio) thyroid receptors (TR) α/a and β/a that could be used for the assessment of thyroid disrupting activity of the hypothalamus-thyroidal axis. To do this, the commercial HEK-293 cells was transfected with the zebrafish TRs-α/a and TRβ1-pcDNA3 constructs, together with luciferase gene (DR4-TK-Luc reporter construct) under the control of thyroid hormone response elements (TRE). Forty-eight hours post-transfection, cells were harvested and seeded in 96-well plates in Dulbecco’s Modified Eagle’s Medium (DMEM) supplemented with 0.4 mg/ml G-418, 10% fetal bovine serum and 1% penicillin/streptomycin (selection medium). To test the stable integration as well as the expression level of the receptor positive clones were plated in 96-well plates and exposed to a range (from 1 µM to 10 µM) of triiodothyronine (T3) concentrations. Finally, the clones showing better EC50 values were selected to determine the presence of thyroidal activity in livestock residues including agricultural amendments. These transactivation results allowed distinguishing the contribution of each TR to the residue-induced thyroidal activity. Acknowledgments - Supported by RTA2012-00053-00-00, RTA2015-00041-00-00 and AGL2016-74857-C3-3-R.

TH065 Screening endocrine disrupting potentials of alternative plasticizers using the zebrafish assay

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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 (DOTP), trictoyl trimellitate (TOTM), bis(2-ethylhexyl) adipate, and diheptyl 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 cytotoxicity assays for each cell line. While not all alternative plasticizers showed significant activity, the 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, ish β 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 disrupt balance of important hormones. Further investigations using in vivo models are warranted.

TH066 Development of reporter gene system for assessing cherry shrimp ecdysone receptor agonists using mammalian cells

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Ecdysoid is a key steroid hormone that regulates growth, development and molting in animals under the phylum of Arthropod, which includes the insects and crustaceans. The hormone targets the receptor component which inhibits the ecdysone receptor (EcR) and retinoid X receptor (RXr). The activated complex anchoring on the ecdysone responsive element (EcRE) 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 ecdysone derivatives (DAH or BAH) have been developed to disrupt ecdysone/receptor signalling. They work as the ecdysone receptor agonists, which cause premature launching of the molting process and subsequently death. Crustaceans, as a subphylum closely related to insects phylogenetically, also adopt this ecdysone signalling system, as they share the hormone, hormone synthetises enzymes and the receptors. Thus, these ecdysone disrupting insecticides, together with other untested potential endocrine disruptors, may post a threat on the crustaceans. Here we report the development of an in vitro reporter assay for the screening of ecdysone receptor agonist in cherry shrimp. The assay is done by transiently transfecting mammalian cells with plasmid vectors expressing cherry shrimp EcR and RXr, together with a vector carrying a luciferase reporter gene fused to a minimal promoter linked to five copies of EcRE. The results show that the system responds well to the native ecdysones hormones in a dosage-dependent manner. The adaptation of mammalian cells in in vitro assay for heterogenous receptor is satisfactory. Three DAH/BAH insecticides were also tested and gave minimal to moderate signals. The results suggest that these DAH insecticides aimed for insect pest control can be potential hazards to crustaceans. More studies on different mammalian cells and competition study with mixtures of chemicals are being carried out to validate this reporter gene system.

TH067 Micro-injection as an alternative for aquatic exposure? A case study in zebrafish embryos with 17α-ethinylestradiol

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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 (ED) properties are mainly carried out in in vitro or aquatic toxicity tests. However, it is often difficult to expose fish to poorly water soluble EDs via micro-injection in the yolk is therefore proposed as an alternative and ecologically relevant exposure route because the yolk of zebrafish embryos contains many lipids, and this route mimics maternal transfer. To be used as an exposure method, micro-injection needs to be characterized and compared to the traditional exposure route via water. In this study, 17α-ethinylestradiol (EE2, an estrogen receptor (ER) agonist) was chosen as a model compound to compare both exposure routes. Zebrafish embryos were exposed either via water or via injection within the first two hours post fertilization (hpf) until 120 hpf. Different endpoints at different levels of biological organization were assessed. Morphological (i.e., different types of abnormalities) and physiological (e.g., heart rate and swimming performance) endpoints were scored, as well as ER binding and qPCR analysis of 14 genes. An LC-MS/MS method was optimized for measuring EE2 levels in medium of the aquatic exposure experiment and the internal dose in embryos after aquatic exposure or injection. The pattern of brain aromatase mRNA expression
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-binding 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 afterwards. 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 in the hepatopancreas. Adult female crayfish (Procambarus clarkii) were exposed during one month to atrazine at concentrations of 1 or 5 mg/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 mg/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. Moreover, in the hepatopancreas, we observed data showing 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. Gismondi, 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 model systems, the limited knowledge of the endocrine system of invertebrates makes the evaluation of EDCs effects difficult. However, invertebrates represent the most part of the biodiversity, such as insects, which are crucial for their functioning (e.g. litter degradation, food resource). Moreover, gammarids are hosts of hidden parasites such as vertically-transmitted microsporida (microsporidia VT), 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 variation of these two genes have been measured by RT-qPCR after an exposure of four EDCs: ethinylestradiol (4HT), 17a-methyltestosterone (17MT) and the cyproterone acetate (CPA), all commonly studied in vertebrates. Sequence research allowed to obtain a 204 bp length and 255 bp length amplifiers for EcR and MIH, respectively. The EcR sequence encodes for 68 amino acid fragment while the MIH sequence encodes for an 85 amino acid fragment. Exposure of G. pulex males at each EDC highlighted an increased of the MIH mRNA expression and a decrease of EcR mRNA expression, a trend to increase was observed for the EcR expression only in uninfected gammarids. This work allowed to identify two main proteins involved in the endocrine system of amphipods. Exposure to each ECD highlighted EDCs affecting vertebrates could also impact invertebrates species. In addition, the presence of microsporidia VT appeared to be a confounding factor which could lead to misinterpretation the endocrine risk assessment. Finally, results the premises is promising in the development of PE biomarkers in invertebrates, since this is a tool that is currently missing. However, further studies will be needed to study the variations of these genes and understand their regularization, before to use them as biomarkers.

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

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Exposure to each ECD highlighted EDCs affecting vertebrates could also impact invertebrates, since further studies will be needed to study the variations of these genes and understand their biomarkers in invertebrates, since it is of great importance to know how sediment-bound chemicals affects aquatic organisms. The synthetic hormone 17α-Ethinylestradiol (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. reproductive development and the potential of sediments-bound E2 to bind to hormone receptors). MIH expression, whatever the parasitic status. However, a tendency to increase was observed when MIH expression was measured in 4HT and CPA treatments and decreased regardless of the exposure concentration. The internal doses were the same 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.

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 eco-toxicity data (48h-EC50) on Daphnia magna showed a significant discrepancy of 3.9-16.0 mg/L. Therefore, our aim is to determine the reliable toxicity of daphnia to BPA. Passive dosing as well as the existing toxicity testing protocol (i.e., spiking with co-solvent) were used to administer BPA to daphnia. Conventional spiking method often fails to control the exposure concentration of (semi-)hydrophobic organic compounds due to the loss of target compounds from sorption to the test vessel and volatilization. Here, passive dosing technique compensates for the bioavailability of EE2, inferred through the YES assay and LC/MS-MS analysis, provides insights into the bioaccumulation of EE2 in C. riparius larvae. Knowledge about the bioavailability, bioaccumulation, and estrogenicity of 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

Assessing acute toxicity of Bisphenol A on Daphnia magna by passive dosing approach

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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 main constituents to the aquatic ecosystem that provide a variety of ecological and economic services. Nowadays, the increasing quantities of insecticides leached in agricultural fields lead to the severe impact on the crustaceans. Among these insecticides, juvenile hormone analogue (JHA) insecticides are a kind of endocrine disruptors known to interfere with the normal hormone action in insects by mimicking the juvenile hormones. However, the structure and functions of the methyl farnesoate (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 mg/L (4.64, 3.20 mmol/L) and 1.96, 1.26 mg/L (6.32, 4.06 mmol/L) methoprene, the body length decreased by 20.5% and 11.8% as compared to control, respectively. And the total number of molts of 20 shrimp over this period reduced by 29.2% and 17.7%. Differential expressions of JH signaling pathway genes were investigated in this study. The genes h3 (hormone receptor 3) and e75 in N. davidi were up-regulated, while Chd64 (colipainlike protein), CHH (crustacean hyperglycemic hormone), e74, JHE (JH esterase), JHEH (JH epoxide hydrolyase) and JHAM (JH acid methyltransferase) were down-regulated in shrimp treated with fenoxycarb and methoprene. The results indicate the interference of these two JHA insecticides on the juvenile hormone system.

TH075 Development of Multimedia Fate Model for Human Risk Assessment of EDCs in the Asan Lake Watershed, Korea M. Choi, J. Kim, Greencos Inc.; Y. Kim, Greencos Inc.; CEO

Multimedia fate model (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 constructing of surrounding environment. The meteorological data and mensuration data of these two chemicals have adverse effects on the development of juvenile N. davidi. After six weeks chronic exposure to 100 μg/L (0.33 mmol/L) fenoxycarb and 200 μg/L (0.64 μmol/L) methoprene, the body length decreased by 20.5% and 11.8% as compared to control, respectively. And the total number of molts of 20 shrimp over this period reduced by 29.2% and 17.7%. Differential expressions of JH signaling pathway genes were investigated in this study. The genes h3 (hormone receptor 3) and e75 in N. davidi were up-regulated, while Chd64 (colipainlike protein), CHH (crustacean hyperglycemic hormone), e74, JHE (JH esterase), JHEH (JH epoxide hydrolyase) and JHAM (JH acid methyltransferase) were down-regulated in shrimp treated with fenoxycarb and methoprene. The results indicate the interference of these two JHA insecticides on the juvenile hormone system.

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, Envigo / 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 immunoassay format. A method was developed with fetus compartment for these chemicals. Results indicate that for both, BPA and DEHP, diet is the main contributor to the total exposure. However, dermal exposure also contribute significantly to the total DEHP exposure. Levels of both EDs were modelled in maternal blood and in fetuses 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).

TH073 Dietary and non-dietary prenatal exposure to endocrine disruptors (BPA and DEHP). Spanish case study. M. MARÍN, INSTITUTO DE MEDICINA Y ENSEÑANZA, J. Rovira, Universitat Rovira i Virgili; R.P. Sharma, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Nadal, Universitat Rovira i Virgili / School of Medicine, IISPV; V. Kumar, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química

Endocrine disruptors (EDs) are chemicals compounds that send confusing messages causing various dysfunctions 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, dermal exposure also contribute significantly to the total DEHP exposure. Levels of both EDs were modelled in maternal blood and in fetuses 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).

TH074 Comparative toxicity and endocrine disruption potential of urban and rural aerosol extracts from PM-2.5 in human placental 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 to 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 and extracts from the rural samples collected in winter 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.

Hormones (T3 and T4) in Fetus, Pup and Adult Rat Serum - Sampling Considerations. J. Rovira, Universitat Rovira i Virgili; R. Sharma, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Nadal, Universitat Rovira i Virgili / School of Medicine, IISPV; V. Kumar, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química

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 immunoassay format. A method was developed with fetus compartment for these chemicals. Results indicate that for both, BPA and DEHP, diet is the main contributor to the total exposure. However, dermal exposure also contribute significantly to the total DEHP exposure. Levels of both EDs were modelled in maternal blood and in fetuses 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).
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 estrogenicity activity associated in dairy farm watersheds regardless of effluent management practices
L. A. Tremblay, Catchwater Institute; J. B. Gadd, NiWA / Department of Chemistry; G. Northcott, Northcott Research Consultants Limited
Steroid estrogens contamination has been linked to adverse effects on exposed 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 minimally treated 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.

TH078 Toxic receipt: Why You Should Avoid it?
J. M. Miller, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; V. Mart, J. Randjelovic, L. Šojić, ALHem - Safer Chemicals
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 system 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 equivalent EEq) was present at low levels in 83% of the stream samples (highest: 1.44 ng L⁻¹ EEq). Most of the samples were found (≤0.15 ng L⁻¹ EEq). While estrogenic activity was generally present 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 estrogen concentrations and estrogenicity 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.

TH079 SETAC Endocrine Disruptor Testing and Risk Assessment Interest Group
H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory
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. Preuss, VITO / ABS; 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). Environmental risk assessment (ERA) regarding 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 (i.e. redesigning the product) or prevents the predicted exposure (i.e. redesign the production process) to minimize the potential risk. Case study: ERA Ag-NP metal based ink. Potential effluent 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
K. Ariis, ARCHE; J. Mertens, Precious Metals and Rhenium Consortium c/o EPFM
As part of the REACH Substance Evaluation for silver, new data was required to be generated, in order to further justify the read-across from ionic silver to silver nanomaterials. Information on aquatic and soil ecotoxicity of the smallest silver nanoform with the highest specific surface area registered under REACH as compared to ionic silver. The tested nanof orm should 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. 201): 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 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
S.F. Hansen, Technical University of Denmark / DTU Environment; S. N. Sørensen, DTU Environment / DTU Environment; L. Skjolding, DTU / DTU Environment; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; A. Bądziak, Technical University of Denmark / DTU Environment
The Environmental Chemical Agency (ECHA) is in the process of revising its guidance documents on how to address the challenges of ecotoxicological testing of nanomaterials. In these revisions, outset is taken in the hypothesis that ecotoxicological test methods, developed for soluble chemicals, can be made applicable to nanomaterials. European Research Council project EnvNano - Environmental Effects and Risk Evaluation of Engineered, which ran from 2011-2016, took another outset by assuming that: ‘The behaviour of nanoparticles

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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 varieties of ENM, the effort for an individual investigation and assessment would be enormous. Therefore grouping of ENM 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 ENM regarding their environmental potential in our project. A key aspect is the behavior of the pristine ENM 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 code (factors for low 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 considered compartment 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 effects 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 on the production volume of the ENM, that portion which is relevant for the considered use, in used 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 for an identification and characterization of the environmental potential of 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 monophasic 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 exposure bond (5 groups of exposure bond). Finally, the so called ecotox bond. The latter one is based on information about ecotoxicity of the bulk material, morphological information 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-ZnO and nano-TiO2 used in sunscreen products. Key words: release, fate, ecotox bond

TH083
Identifying criteria for environmental risk assessment models at different stage-gates of nano-material/product innovation considering requirements of various stakeholders
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The EU H2020 project calibrate aims to establish a state-of-the-art versatile risk governance framework for assessment and management of human and environmental risks of manufactured nanomaterials (MN) and MN-enabled products. The approach is based on the identification and development of innovative best practices for the assessment of these materials and products. Initial efforts have focused on identifying criteria for environmental risk assessment (ERA) models and tools for such governance framework. It was recognized that some criteria are applicable to both environmental and human risk assessment (HRA), and these so-called “overall” criteria were identified through joint efforts of the ERA and HRA working group experts in calIBRate. The identified “overall” criteria relate to RA model features and resources needed to use the tools, whereas the criteria specific to ERA models relate to model outcome on hazard, exposure and risks. The identified criteria were listed against the Cooper stage-gates®, thus forming a table in which the importance or relevance of each criterion could be assessed for each of the stage-gates. This was formed into questionnaires with defined response options for each environmental hazard assessment (HRA) and 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 considered compartment 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 effects 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 on the production volume of the ENM, that portion which is relevant for the considered use, in used 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 for an identification and characterization of the environmental potential of 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 monophasic 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 exposure bond (5 groups of exposure bond). Finally, the so called ecotox bond. The latter one is based on information about ecotoxicity of the bulk material, morphological information 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-ZnO and nano-TiO2 used in sunscreen products. Key words: release, fate, ecotox bond

TH085
Matrix to predict possible environmental risk of nanomaterials during use stage-gate evaluation
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Engineered nanomaterials (ENM) are used in different products. Key words: release, fate, ecotox bond

TH084
Considerations of nanomaterial’s environmental fate to support grouping and environmental risk prediction
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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 differ 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, ion exchange capacity, morphology and toxicity of each ENM as well as ecotoxicology and their chemical 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 questions which, 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 discards and is important test 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.

Forms of released engineered nanomaterials: A systematic assessment in material flow analysis

V. Adam, EMPA Technology & Society Lab / Technology and Society Lab; A. Caballer-Guzman, EMPA / Technology and Society Lab; B. Nowack, Empa Swiss Federal Laboratories for Materials Science and Technology / Technology and Society Lab

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 the environmental fate. The flowchart identifies the processes that contribute to the 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.

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A new test method to determine the bioaccumulation of manufactured nanomaterials in filtering organisms (Bivalvia) using the freshwater mussel Corbicula fluminea


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 standardized 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 through conditions. First studies were carried out with the freshwater mussel Corbicula fluminea. 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 released silver in water was carried out by ICP-MS or ICP-OES. The subsequent time and water 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 global distribution of certain perfluoroalkyl and polyfluoroalkyl substances (PFASs) in the environment is of concern given their environmental persistence and possible adverse effects. The aim of the present study was to investigate the sorption to organic soil constituents of a variety of PFASs added as a fine film of foam (AFFF) impacted sites may be contaminated by the relatively limited number of certified standards to ensure a rigorous quantification. A possible solution is the implementation of a surrogate approach such as the total oxidizable precursor (TOP) assay, relying on the oxidative conversion of potential perfluoroalkyl acid precursors (Pre-PFASs) into readily measurable perfluorocarboxylic acids. However, in order to ensure a fully legitimate comparison between conventional (i.e. before oxidation) and after TOP 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 ultrahigh performance liquid chromatography tandem mass spectrometry (TOP-UHPLC-MS/MS) analysis was therefore evaluated using various environmental waters. The validation endpoints ascertained included, notably, the evaluation of oxidation yields in the various matrices as well as the TOP assay as a crucial assay used in the study of aromatic oxyfunctional groups 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 (FTSs) were the target pre-PFASs predominantly reported before oxidation in most instances, they could only partially account for the observed AFFF (molar concentration increases upon oxidation). The unexplained AFFF portion likely results from the oxidation of untargeted, pre-PFASs for which oxidation yields are yet to be determined.

**TH096**

Use of biochars for the sorption of poly- and perfluorinated alkyl substances (PFASs) 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 are industrial contaminants, which are highly accumulated in soils and wastewater treatment plants. These 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 atom. Perfluorinated acids and alkyl and perfluorooctane sulfonamide (FOSA), and for these substances, the SOM bulk net negative charge was the better sorption predictor as compared to the pH value alone. However, for sorption of the most long-chained substances (i.e. the C8−C13, PFCA, PFOS and FOSA), cation effects were small and instead sorption was more strongly related to the pH value. This suggests that the most long-chained PFASs have a binding preference towards the highly condensed parts of the humin 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**

Environmental degradation rates for new PFAS via decarboxylation potential in water, in a MS-collision cell and in silico

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Straight-chain perfluorooctylalkanoic 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 by CO2 so fast at room temperature that its spontaneous decomposition is a synthetic method for nonfluorosubstituted. There are indications that novel oxygen-containing analogs of PFOA are less stable towards decarboxylation. A typical detection method for PFCSs 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. Despite the high sensitivity and selectivity of the decarboxylation 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**

Perfluoroalkylated acids (PFAAs) in soil and invertebrates (Isopoda) near a fluoroclean plant in Antwerp, Belgium

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Perfluoroalkylated acids (PFAAs) have been produced for over five decades. Due to their hydrophobic and lipophilic character they are suitable for a wide range of applications. However, PFAAs may enter the environment, accumulate in wildlife and may cause detrimental effects. The widespread use of PFAAs has resulted in a global presence. Therefore the major global manufacturer, 3M, phased out the production of perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) in 2002. Nevertheless, these compounds are still detected in high concentrations in the environment and biota. Especially the fluorochemical plant has been characterized as a potential source for environmental contamination. In the present study we measured the concentration of 12 PFAAs (8 perfluoralkyl carboxylic acid PFCA's and 4 perfluoralkyl sulfonic acids (PFAS's) in soil and isopods collected at a fluoroclean plant in Antwerp, Belgium. In addition, samples from four other areas were collected, representing a gradient in distance from the pollution source. We tested for both correlations between soil properties (e.g. total organic carbon (TOC) and PFAA concentrations in soil, as well as correlations between PFAA concentrations in soil and invertebrates. In the soil, PFOA, PFOS and PFOA were the only compounds that were detected at all sites. Soil concentrations of all other compounds, with exception of PFDoA and PFBS, were <LOQ in all sites except for the plant site. Median concentrations of 606 ng/g ww for PFOS and 8 ng/g ww for PFOS were detected in soil at the plant site, which were high compared to what has been reported in previous studies conducted in the area. Furthermore, these concentrations decreased significantly with distance from the plant. However, concentrations did not differ between the three locations that were situated farthest away from the plant. No significant differences in TOC were
observed among the studied sites, but TOC was positively correlated with multiple
PFAAs, including PFOS and PFOA. At this moment (November 2017), isopods
have not been tested for PFAAs concentrations yet, but based on the soil
concentrations and concentrations detected in previous studies near the
fluorochemical plant in Antwerp, we expect high concentrations of multiple
PFAAs. The outcome of the present study will be used in further monitoring studies
on the effects of soil type on PFAAs bioavailability to invertebrates, as well as
effects of PFAAs on multiple biomarkers.
TH100
Occurrence and distribution of legacy per- and polyfluoroalkyl substances
(PFASs) and fluorinated alternatives in coastal waters of the German North
and Baltic Seas
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Chemistry
Long-chain per- and polyfluoroalkyl substances (PFASs) are recognized as global
contaminants of high concern as they have been shown to be persistent,
bioaccumulative, toxic, and ubiquitously present in the environment. This has led to
a number of actions by industry and regulatory authorities aiming at restricting the
production, use, and release of long-chain PFASs. Consequently, an industrial shift
has been taking place, moving away from long-chain PFASs toward alternative
substances, such as per- and polyfluoroether carboxylic and sulfonic acids
(PFECAs and PFESAs). Due to structural similarities, the question arises whether
the alternatives represent a substantial improvement on their predecessors. Public
data on their properties and environmental exposure is still limited. This study aims
at investigating occurrence and distribution of legacy PFASs and fluorinated
alternatives in surface water samples from coastal areas of the German North and
Baltic Seas. In summer 2017, two sampling campaigns were realized using the
research vessel Ludwig Prandtl, during which 94 water samples were taken along
the German coastlines. The analytical method included 26 legacy PFASs and 5
fluorinated alternatives, among them the PFECAs GenX and ADONA. Filtered 1 L
water samples were spiked with mass-labelled internal standards (50 µL, 60 pg/µL)
and loaded onto preconditioned solid phase extraction cartridges (Waters Oasis
WAX; 6cc, 500 mg, 60 µm). After a washing step, the target compounds were
eluted using methanol and 0.1 % ammonium hydroxide in methanol. The eluates
were reduced to 150 μL under nitrogen and 13C8-PFOA was added as injection
standard (10 μL, 100 pg/μL). Instrumental analysis was performed by
HPLC-MS/MS, using an Agilent HP 1100 LC system coupled to an AB Sciex API
4000 triple quadrupole mass spectrometer. First results show that the fluorinated
alternative GenX can not only be detected in all water samples along the German
North Sea coast, but is one of the dominating PFASs with average concentrations of
1.4 ± 0.2 ng/L. Based on these and further results, it will be discussed if regulations
on long-chain PFASs and the subsequent ongoing shift to fluorinated alternatives
lead to changes in the coastal environment.
TH101
Suspect screening for short chain PFAS in environmental water samples,
waste water treatment plants, and building materials
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Perfluoroalkyl and polyfluoroalkyl substances (PFASs) are very persistent
anthropogenic fluorinated chemicals that have been detected in remote areas and all
compartments of the environment. Historically perfluorooctanoic acid (PFOA) and
perfluorooctane sulfonate (PFOS) are the two most frequently used and most well
studied PFASs. As a consequence of the bioaccumulative and toxic properties of
long chain PFASs, their voluntary phase-out started in the year 2000 and led to an
increased prevalence of short chain homologues (C4 to C6) in the aquatic
environment. Short and ultra-short (>C4) chain PFASs are quickly eliminated from
organisms and thus do not bioaccumulate. However, they are more mobile in the
water cycle then their long chain homologues, thus exhibiting higher tendencies to
reach raw and drinking water, and are expected to accumulate in the edible parts of
plants, which may lead to an increased exposure through drinking water and
vegetable consumption. While perfuorobutanoic acid (PFBA) and perfluorobutane
sulfonate (PFBS) have been extensively studied information about ultra-short chain
PFAS is still scarce and, if available, limited to perfluorocarboxylic acids (PFCAs)
and perfluorosulfonic acids (PFSAs). Trifluoroacetic acid has been detected in
concentrations in excess of 20 µg/L in tap water, while perfluoropropane sulfonate
(PFPrS) and perfluoroethane sulfonate (PFEtS) have been detected in a study of tap
water samples from China, Japan, India, the United States of America, and Canada.
In 2016, the first C1-homologue of a legacy PFAS class was detected in the form of
trifluoromethane sulfonic acid (TFMSA), which was present in various
compartments of the water cycle ranging from waste water treatment plant effluents
to finished drinking water. Information about ultra-short chain homologues of other
PFASs like perfluoroalkyl phosphonic acids (PFPAs), perfluoroalkane
sulfonamides (FASAs), perfluoroalkane sulfonamidoethanols (FASEs), and
perfluoroalkane sulfonamidoacetic acids (FASAAs) are to the best of our
knowledge not available so far. In an attempt to close this gap in knowledge, we

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performed a suspect screening for (ultra-)short chain PFASs of several substance
classes in environmental water samples, waste water treatment plants and building
materials.
TH102
Utilization of passive samplers to detect poly- and perfluoroalkyl substances
(PFASs) in wastewater treatment plants and estuarine environments
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Oceanography
Poly- and perfluoroalkyl substances (PFASs) are of growing concern worldwide,
due to the linkage of these compounds to adverse effects in humans and the
environment. Surface waters in the northeastern United States in particular have
displayed elevated concentrations of PFASs. Here we utilize passive samplers to
gain a better understanding of the sources and spread of these contaminants.
Thirty-two microporous polyethylene (PE) passive samplers (containing
Hydrophilic-Lipophilic-Balanced sorbent) were deployed across nine sites in
Narragansett Bay (RI, USA) in the fall of 2017 for a one month duration each.
Deployment sites ranged from wastewater treatment plant and industrial outfall,
military and fire training bases, and more pristine areas. 25 PFASs (including
sulfonates, carboxylic acids, and GenX) were measured across all sites in the
passive samplers, as well as water and sediment samples. For a more direct point
source evaluation, 10 additional samplers were deployed in two waste water
treatment plants of a large urban area. By analyzing the spatial and temporal trends
of these fluorinated compounds we plan to assess their longevity in water and
sediment of the Bay. Lastly, we aim to understand and predict potential effects on
the environment and better advise on regulatory practices.
TH103
Distribution of per and polyfluoroalkyl substances in sediments of the Spanish
coast
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Department; D. Prada-Rodríguez, Universidade da Coruña / Grupo Química
Analítica Aplicada (QANAP); L. Viñas, Instituto Español de Oceanografia / Centro
Oceanográfico de Vigo; J.A. Campillo, Instituto Español de Oceanografia / Centro
Oceanográfico de Murcia; S. Muniategui, Universidade da Coruña / Analytical
Chemistry
Per- and polyfluoroalkyl substances (PFASs) configuration, consisting in an
alkylated hydrophobic chain fully or partially fluorinated, hydrophilic group
terminated, provides to PFASs simultaneous hydrophobicity and lipophobicity.
Their persistence, bioaccumulation and toxicity make them a source of increasing
environmental and public health concern. Presence of PFASs in sea environment is
caused by discharges of wastewater effluents or river flows, urban runoff,
atmospheric deposition of volatile precursors and subsequent transformation, or
direct application of fire-fighting foams containing PFASs, among others. Samples
were collected in two semiconfined coastal areas, one of them an area with high
industrial and port activities (Ría de Vigo) and the other one with high touristic and
agricultural activity (Mar Menor). PFOA, PFOS, PFOSA, n-MeFOSA and
n-EtFOSA were extracted from sediments by sonication, cleaned up by dispersive
solid phase extraction and the analyzed by LC-LTQ-Orbitrap-HRMS in full mode
(Concha-Graña E. et al, 2017). This is the first time that these compounds were
measured in these areas. N-MeFOSA and N-EtFOSA were not detected in any
sample, whereas PFOSA was only detected in two samples, but below the
quantitation limit. PFOS was measured in 39 % of samples, most of them from Mar
Menor. In Ría de Vigo PFOS was detected in a point close to a ceramic factory.
Regarding PFOA, this compound was measured at level higher than quantitation
limit in 2 Mar Menor samples. Sum of PFASs in each sampling point was below 0.4
ng/g in Mar Menor and below 0.1 ng/g in Ría de Vigo, being the total concentration
of PFOS similar than the detected in similar areas. Some characteristic of the
sediments were taking into account in order to find the correlation between these
parameters and the obtained data. Moreover, the environmental risk was evaluated.
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reference: CTM2013-48194-C3-1-R/2-R, and ARPA-ACUA, project reference:
CTM2016-77945-C3-3-R). References: Concha-Graña E. et al, VIII Reunión de la
Sociedad Española de Espectrometría de masas, V Reunión Nacional de Dioxinas,
Furanos y Compuestos Orgánicos Persistentes Relacionados (2017)
TH104
Utilization of Polyethylene Passive Samplers to Detect volatile PFAS
precursors in water and air
E. Dixon-Anderson, R. Lohmann, University of Rhode Island / Graduate School of

SETAC Europe 28th Annual Meeting Abstract Book


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 performed 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 KPC, were determined for several PFASs at a Waste Water Treatment Plant (WWTP) and were also performed in 2016, and sampling rates derived from the loss of performance reference compounds. Best-fit curves were used to determine polyethylene-water partitioning constants, log KPW, during the 3-week uptake experiments. Derived log KPW values for 6, 8, 2, 8 and 10,2 FTOHs were 3.8, 4.4 and 4.8, respectively. For MeFOSE and EiFOSE, derived log KPW 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, MeFOSE and EiFOSE.

TH105 Occurrence and Removal of perfluoralkyl and polyfluoroalkyl substances (PFASs) in full-scale water and wastewater treatment plants 

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TH106 Perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) occurrence in biota in Czech rivers

V. Kodes, D. Leniortyova, Czech Hydrometeorological Institute / Section of water quality, R. Grubic, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Objective of the Study The comparsion PFOS and PFOA concentrations detected in biota within CHMI bioaccumulation monitoring program from years 2010 – 2016. Material and Methods A bioaccumulation monitoring of selected perfluoroalkyl substances in biota (fishes and aquatic organisms) was performed in the Czech Republic. Monitoring comprises two profile sets containing 21 and 22 monitoring sites. Sampling at those two site sets alternates in the three-year cycles. Sites are located at important parts of main Czech rivers (country borders, before confluences, downstream industrial sites or large cities, etc.). An assessment was made for following matrices: juvenile fish, benthos (Hydropispsche sp., Epipomatus sp., Gammaridae sp., Oligochaeta sp., Gammaridae sp., Oligochaeta sp., etc.), microalgae, macroalgae, periphyton and Diatoms. The analyses of fish were conducted for following tissues: muscle, blood and liver. In total, following number of samples of various matrices were analysed using LC-MS/MS and LC-HRMS: fish blood 105, fish liver 15, fish muscle 78, juvenile fish 149, benthic organisms 126, mussels 73. Results PFOS highest values were detected in fish blood (10.3030 g/L), weight wet concentrations in fish liver (50.5 - 317 g/kg), juvenile fish (1.2 - 312 g/kg) and benthic organisms (0.05 - 0.01 - 0.01 g/kg) significantly exceeded levels of PFOS found in fish muscle (0.04 - 0.038 g/kg). The lowest PFOS concentrations were found in mussels (0.01 - 0.02 g/kg). PFOA concentrations compared to PFOS reached significantly lower levels in all monitored matrices. Range of values was between 0.001 - 3.1 μg/kg, whereas minimum represents the smallest concentration found in mussels and the maximum represents concentrations in juvenile fish. PFOA highest values were detected in juvenile fish (0.01 - 3.1 μg/kg), followed by benthic organisms (0.02 - 2.5 μg/kg) and fish blood (0.06 - 1.8 μg/L). Small concentrations were found in mussels (0.01 - 1 μg/g), fish muscle (0.02 - 0.5 μg/g) and fish liver (0.02 - 0.07 μg/g). Concentration in general the PFOA concentration was higher than PFOS except fish muscle was not detected. PFOA concentrations in fish blood, kidney and liver were higher than concentrations found in muscle tissues due to its binding to proteins in a blood and a liver. All collected fish blood samples and more than 50% collected samples of fish liver and juvenile fish exceeded EQS for PFOA (9.1 μg/L).

TH107 Analytical strategy to study the distribution of perfluoralkyl substances in fish tissue of Italian deep subalpine lakes


Perfluoralkyl substances, such as perfluorinated sulfonic acids (PFASs) and perfluorinated carboxylic acids (PFCAs), are ubiquitous contaminants in the aquatic environment, including wildlife and humans. Perfluoralkyl acids bind to proteins and the binding in bioaccumulation behaviour differently 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 contaminants. Several fish species from Italian deep lakes were seasonally collected for the analysis of 10 perfluorocarboxylates, 7 perfluorosulfonates and 5 perfluorosulphonamides. Individual fish were measured, weighed and dissected in three fractions: whole viscera, the muscle and the rest of the carcases (head, fishbone, skin and fins). The fractions of fish six were analysed separately or pooled in one or two samples for the subsequent analysis. The dry weight, the lipid and the protein content were measured in each fish fraction (muscle, viscera and the rest of carcase). PFAS analysis were carried out with fresh samples but some samples of fillet were also freeze-dried in order to compare the concentrations. Extraction of the animal tissues (2-5 g) was performed by sonication with ACN/H2O mixture enhanced by salting out and acidification; extracts were purifed on HydroSSEP and then analysed by gas chromatography–mass spectrometry using GC-MS/MS. PFSA concentrations in samples expressed on fresh weight basis are lower than ones determined in fresh samples probably due to evaporation of analytes. The mixed-effect model and were fitted for 24 C1-C6 PFAs and C3-C7 PFCAs, as well as some pre-PFAs such as 8:2 and 10:2 FTAs, as well as FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP were also frequently detected (60% of variability was explained for each expression effects by phospholipids. Perfluoralkyl compounds were determined by liquid chromatography tandem mass spectrometry (HPLC-MS/MS) coupled to an on-line turbulent flow chromatography (TFC) for on-line purification of the extracts. PFAS concentrations in lyophilised samples (expressed on fresh weight basis) are lower than ones determined in fresh samples probably due to evaporation of analytes. The mixed-effect model and were fitted for 24 C1-C6 PFAs and C3-C7 PFCAs, as well as some pre-PFAs such as 8:2 and 10:2 FTAs, as well as FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP. However, a significant decrease in pre-PFCAs/PFCAs concentration ratios with trophic level suggested a possible contribution of...
precursors to the apparent biomagnification of PFCAAs, via their biotransformation. In addition, the Total Oxidisable Precursor (TOP) assay was applied to sediments and, for the first time, to biota samples. Results revealed the presence of large proportions of unknown pre-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


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 PFCA), medium and long chain PFAS (e.g. C6 to C14 PFAS and PFPeA), and also precursors (e.g. PAPs, diPAPs, 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 (Oncorhyncus 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 RIVERLY Laboratoire Ecotoxicologie; J. Koschorreck, Umweltbundesamt

Per- and poly-fluorinated substances (PFASs) are ubiquitous in the environment, especially 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 estimate the accumulation of individual species and the impact on these PFASs. We performed experiments on two fish species: Zebrafish (Danio rerio) and Rainbow Trout (Oncorhyncus mykiss) exposed through the diet to two selected PFASs, namely perfluoroctanoic sulfonic acid - PFOS and perflurohexane 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. The concentration in fish was measured through several technical and biological methods. The test concentrations were 53B constituents in bream liver samples afar from production sites, and provide indications on distribution patterns.

**TH111**

Does water temperature influence the toxicokinetics of perfluorinated substances? Comparison of two dietary experiments in rainbow trout (Oncorhyncus mykiss)

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Per- and poly-fluorinated substances (PFASs) are widely found in fresh and marine water environments and accumulate in aquatic organisms. Fish are poliketheromes and subject to large seasonal changes of temperature, which control physiological processes such as feeding, respiration, fecal egestion, and ultimately growth. 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 after 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 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 a commercial diet enriched 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

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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), perfluorooctane sulfonate (PFHxS) and perfluorobutanoic acid (PFA) in the zebrafish embryo (Danio rerio) (Danio rerio ZFET), to develop an alternative test model for toxicity testing. We exposed ZFETs at three concentrations of each PFAS for 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 ZFETs were sampled separately at nine time points. Water samples from chemical controls without ZFETs 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< PFBA). 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 ZFET indicate biphasic uptake kinetics with slow uptake before reaching a faster uptake after hatching. Apparent steady-state concentrations were reached at 96 hpf for PFOS, PFHxS and PFA, while PFBA did not reach steady-state within 120 hpf. Moreover, PFOS and PFHxS (sulfonic acid end group) showed a higher biocaccumulation than PFOA and PFBA (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

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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, FPO-902 or PFOSomega). 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.
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 matrix concentration. Higher safety factors need be determined because of the difference in toxicokinetic half-life between humans 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 Perfluorooether carboxylic acids - are these substances appropriate PFOA-alternatives regarding their environmental concerns? C. Stuhr, G. Gobert, A. Biegel-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 in the production, use and disposal (PUP) under 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 perfluorooether carboxylic acids (PFECA). PFECAE are structurally similar to perfluorooakyl carboxylic acids such as PFOA but do not form the perfluoralkyl ether chain. Due to this structural similarity it could be expected that PFECAE 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 PFECAE such as ADONA (ammonium 2,2,3,3-tetrafluoroalkyl ether) and GenX (ammonium 2,3,3,3-tetrafluoro-2-(heptafluoropropoxy) propionate). The poster will present a summary of the substance evaluations. PFECAE are expected to be very persistent under environmental conditions. The substances have a low bioaccumulation potential in aquatic organisms. However, just as PFOA, PFECAE may not fit into the common accumulation pattern. Furthermore, the substances are probably mobile in the aquatic environment and can reach groundwater and consequently drinking water resources. PFECAE 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 PFECAE already demonstrates that these substances are hazardous for the environment and further risk management measures are needed. [1] Gebbink WA, van Assen, The L., van Leeuwen SRF. 2017. Environ. Sci. Technol. Lett. 4: 507-510 (PUP) [2] Sun M, Arevalo E, Strynar M, Lindstrom A, Richardson M, Kearns B, Pickett 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. Diploma thesis [4] Heydebreck F, Tang J, Xie Z, Ebinghaus R. 2015. Environ. Sci. Technol. 49: 8386-8395: 49: 14742-14743

TH115 Fluoropolymers: Polymeric PFAS That Satisfy Global Polymer of Low Concern Criteria B. Henry, T. Kennedy, W.L. Gore & Associates, Inc. Fluoropolymers, such as polytetrafluoroethylene (PTFE), constitute a distinct class of chemicals. Fluoropolymers are resistant to chemical, hydrolytic, oxidative, photochemical and biological degradation. They are uniquely stable within their intended processing temperatures (e.g., 260°C for PTFE). Fluoropolymers have negligible residual monomer, low molecular weight (MW) compounds, and can reach groundwater and consequently drinking water resources. 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 Monomer 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- and 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 VPs (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 J. Vollinger, Eurofins AgroScience Services EcoChem GmbH / Field Ecotoxicology; O. Klein, Eurofins AgroScience Services EcoTox GmbH / Ecotox Field; S. Knaebe, EAS Ecotox GmbH / Ecotox 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 adequacy 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 location 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-site 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 Products 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, but also by 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 on soil as statistically evaluate and is 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).

THI19
Adaptation of the earthworm field test method: conceptual overview and first results
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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 anonymous regulations. This data on soil as a statistically evaluate and is 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).

THI20
Soil ecotoxicology and ecological risk assessment in southern African mining landscapes
M. Mabuza, North-West University / Unit for Environmental Sciences and Management; H. Elsajackers, 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 toward soil life 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 difficult for biological and terrestrial organisms, and subsequently 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

THI12
Establishment of tiered risk assessment approach of pesticides for soil 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, soil 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 on soil as a statistically evaluate and is 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).

THI121
Establishment of tiered risk assessment approach of pesticides for soil 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, soil 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 on soil as a statistically evaluate and is 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).

THI122
Ecological recovery and terrestrial Non Target Arthropods: abundance, functional roles and networks
M. Haen-Kissling, Eurofins-Mitot; S. Aldershoff, Bioresource and Evaluation; F. Thunberg, 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 disturbance of the application of pesticides, which is 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 functionality 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. Hğerbäumer, R. Raschke, Bielefeld University / Animal Ecology; S. Hoss, Ecosa / 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. Informations 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 metazoan organisms and hence, they are a key component in non-target population dynamics. Nematodes 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 Chromomus 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 microarthropods facilitate OM breakdown in an arable field soil? - Implications on specific protection goal setting for soil risk assessment of plant protection products
G. Ernst, TH123; A. Sharples, FMC Agricultural Solutions; F. Staab, BASF SE / Ecotoxicology
This work was initiated and funded by the ECPA non-target arthropod group Protection Goals for soil micro-arthropods in field-areas. This foresees that even short-term effects on single species in a magnitude of >65% are considered unacceptable to ensure the provision of Ecosystem Services in agricultural soils. The present study shows that a reduction of the total soil micro-arthropod community by 80% over a period of 6 months has no unacceptable effect on the mesofauna driven OM degradation in a minicontainer test on an arable field. Thus, the relevance of the structural endpoints on soil micro-arthropods (i.e. single species populations) within an in-field soil risk assessment for PPP, which focus on maintenance of soil fertility (protection of soil functions), is questionable.

TH125 The role of source sink dynamics in the assessment of risk to non-target arthropods from the use of plant protection products
G. Lewis, JSC International Ltd; S. Braecker, 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 dependant. However, this issue does raise important questions about endpoints which will need to be considered when establishing the role of non-target 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 age of representative surrogatespecies 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 concept of source-sink dynamics in regulatory risk assessment. Practically it means that 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 ECPA non-target arthropod group

TH126 Classification of uncertainty in ecological risk assessment of pesticides
A. Hunka, Hulma University / School of Business, Engineering and Science; M. Meli, ADAMA Agricultural Solutions; S. Pashani, S. Waara, Hulma 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 a better understanding of the species in question, making it, making this an issue of significant concern. The lack of straightforward presentation of all sources of uncertainty puts an extra burden on risk managers. This issue has been recognized by EFSA recently, but still there is very little research into classifying, visualizing and addressing uncertainty in ERA of pesticides. Currently EFSA recognizes standard and non-standard uncertainties in ERA of pesticides. This classification offers little insight into how these two categories impact ERA conclusions and further risk management decisions. In order to address this gap, we have undertaken an appraisal of a large subset of ERAs (102 up to date) conducted for approval of active substances at the EU level. We have been working on ERA data, conclusions on pesticide peer-reviews and Draft Assessment Reports in ERA. We have considered most of the methods and sources of uncertainty, classify different uncertainties and link them to recognizable points of concern, data gaps and risk management decisions. At the moment it is still unclear which sources of uncertainty influence the decision outcome more than others and our preliminary results indicate that it is possible to uncover non-obvious relationships between uncertainty and risk assessment outcomes. It is, for instance, possible to describe how different sources of uncertainty affect the process of ERA (e.g., duration, effectiveness of a Rapporteur Member State in producing a draft assessment report) and compare how uncertainty is addressed in risk assessment for different environmental compartments, especially soil versus ERA for aquatic organism which is much more developed. We aim to provide a typology of recognized uncertainties in ERA and discuss how it could help inform the establishment of the surrogate reference tier and the subsequent calibration of lower tiers in the new risk assessment scheme for soil organisms, which is currently being developed by EFSA. Ultimately, we aim to link the typology of uncertainties in ERA to risk management techniques, in order to help ERA practitioners to better
Derivation of soil threshold concentrations for arsenic: consideration of bioavailability through combination of ecotoxicological and analytical data

J. Roembke, B. Forster, ECT Oekotoxikologie GmbH; E. Giese, Federal Environment Agency; S. Janisch, RWTH Aachen University / Institute for Environmental Research; S. Nickoll, B. Scholz-Starke, RWTH Aachen University; Institute for Environmental Research; J. Brack, Federal Environment Agency / Institute for Environmental Research BioV; M. Ross-Nickoll, B. Scholz-Starke, RWTH Aachen University; Institute for Environmental Research; S. Ottermanns, RWTH Aachen University / Institute for Environmental Research; S. Pieper, German Federal Environment Agency (UBA) / 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 is the first time the performance of CPCAT was assessed 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 more precise approach than is currently used for earthworm field studies because data distribution and variance are adequately considered and smaller differences between control and treatments can be detected.

Derivation of soil threshold concentrations for arsenic: consideration of bioavailability through combination of ecotoxicological and analytical data

J. Roembke, B. Forster, ECT Oekotoxikologie GmbH; E. Giese, Federal Environment Agency; S. Janisch, RWTH Aachen University / Institute for Environmental Research; S. Nickoll, B. Scholz-Starke, RWTH Aachen University; Institute for Environmental Research BioV; 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 is the first time the performance of CPCAT was assessed 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 more precise approach than is currently used for earthworm field studies because data distribution and variance are adequately considered and smaller differences between control and treatments can be detected.

TH130

Relationship between soil microbial biomass methods used in environmental fate laboratory studies

P. Massey, Smithers Viscient; P. Pearson-Davies, B. Earnshaw, Smithers Viscient; S. Swales, Smithers Viscient 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 microbial biomass is the fumigation extraction method. This method determines the carbon content of the soil biomass, via extraction, 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 used to determine microbial biomass. The fumigation extraction method is used for laboratory studies. In work currently being undertaken by Smithers Viscient 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.

TH131

Where are the Springtails? New data on the vertical distribution of Folsomia candida (Collembola) and its population dynamic in artificial soil

L.S. Tschoppke, RWTH Aachen University / Institute for Environmental Research BioV; V. Roeben, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research BioV; M. Nickoll, RWTH Aachen University / Institute for Environmental Research / Institute for Environmental Research BioV; M. Nickoll, RWTH Aachen University / Institute for Environmental Research / Institute for Environmental Research BioV;

Folsomia candida is a non-target arthropod species which is often referred as the „Standard Soil Arthropod“ (Fountain and Hopkin, 2005). It is part of the regulatory framework of pesticide risk assessment and in the last years an increasingly important model organism in ecological and effect modelling. However, the knowledge on the population dynamics on a long-term scale and the vertical dispersal within the soil column is still scarce. We will present the results of two experimental studies exploring those unknown topics – one on the population dynamics over time and one on the vertical dispersal in relation to food location.
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 oscillating phase during its maximum at 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 gauze 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. Septa 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 untreated, the top, the bottom and in the ground 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. Fajana, 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 the reproduction of *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, stress biomarkers, and bioavailable zinc were determined 28 days after exposure. Habitat quality did not change zinc bioavailability which remained at 2% across all three habitat indices. Instead, mite fitness improved with increasing habitat quality and mites were able to tolerate higher zinc body burdens in better habitat qualities. Furthermore, the zinc response (measured as the slope of the EC50) was more pronounced in lower habitat qualities. Our data suggest that habitat quality is more important than metal concentration for soil protection. Ecorestorisation, rather than remediation, will likely be a more effective means of ameliorating zinc toxicity.

**TH133** Effects of atmospheric hydrogen chloride and ammonia on Paronychiurus infusionum (Collembola: Onychiuridae)

J. Wei, 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 accidents 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 reactivity with soil biota such as *Collembola* and earthworms. The experiment carried out in PS container filled with 30g of 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 concentration of toxic substances in the enclosed chamber for 20 minutes. After exposure, the test vessels with *P. kimi* were transferred to an incubator (20°, constant darkness) in a closed state, and the mortality and reproduction rate of *P. kimi* were observed after 1 hours, 2 weeks, and 4 weeks. There were no deaths after 1 hour, but the mortality rate was increased over time from 2 weeks. Also, 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.

**TH134** 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 andrei and soil algae Chlamydomonas reinhardtii and Chlorococcum infusionum. *Eisenia andrei* were exposed with 25 g of control or MEK soils with closed system. After 7 days exposed, mortality and abnormalities including spawnings, fragments, swelling, bleeding, and mucous secretion were measured. For soil algae, *Chlamydomonas reinhardtii* and Chlorococcum 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, 7th-LOEC and 7d-EC50 of MEK to *Eisenia andrei* 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 (KEITI) through "The Chemical Accident Prevention Technology Development Project", funded by Korea Ministry of Environment (MOE) (No. 20160019070001). *<strong>Key word: methyl ethyl ketone, earthworm, soil algae</strong>*

**TH135** Effects of endocrine disruptor 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 disruptor 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 use in soil algae.* Soil algae were exposed with low to high concentration of BPA, DEHP, and nonylphenol. As results, 7-day-LOEC and 7d-EC50 of BPA, DEHP, and nonylphenol in soil 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 Chlororococcum 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*

**TH136** 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 biondicato and *Folsomia candida*. The effects of bisphenol A (BPA), bis(2-ethylhexyl)phthalate, nonylphenol, soil vinasse in natura and after pH adjustment was valid for the evaluations of vinasse may have on reproductive behavior of these animals. Earthworms of the species *Eisenia andrei* (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 both tests, the animals were exposed to 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 biosauss 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

R.G. Kuperman, Edgewood Chemical Biological Center / Molecular Toxicology Branch, R. Checkai, U.S. Army Edgewood Chemical Biological Ctr / Molecular Toxicology / Environmental; M. Simini, U.S. Army Edgewood CB Center / National Institute for Occupational Safety and Health

The study investigated the toxicity of nitrogen-based energetic soil contaminants in acute tests with the earthworm *Chromopogon arenosus*. The toxicity of each contaminant was determined using the guideline OECD 207. *Chromopogon arenosus* were exposed to different concentration of the contaminant and mortality and reproduction rate were measured 10 days after exposure. The results show that the toxicity of the contaminant varies significantly, from non-toxic to highly toxic. The study highlights the importance of assessing the toxicity of nitrogen-based energetic soil contaminants in acute tests with earthworms in order to better understand their ecological impact.
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-hexaazaisowurtzitane (CL-20), 2,4,6-trinitrotoluene (TNT), 1,3,5-trinitrobenzene (TNB), 2,4-dinitrotoluene (2,4-DNT), 2,6-dinitrotoluene (2,6-DNT). 2-amino-4-hydroxydinitrotoluene (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 (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration. These results further confirm the Cu tolerance potential of these content (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration. These results further confirm the Cu tolerance potential of these content (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration. These results further confirm the Cu tolerance potential of these content (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration. These results further confirm the Cu tolerance potential of these content (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration. These results further confirm the Cu tolerance potential of these content (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration. These results further confirm the Cu tolerance potential of these content (comparable to the control) than non-preferential growth rate, survival, cocoon and juvenile counts and soil Cu concentration.

Alternatives to soil invertebrates and terrestrial plants were used to derive test 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 Cleanup Values (SCVs). Based on established SSD and Eco-SSL values developed in these studies will be submitted to the USEPA Eco-SSL Work Group for use in establishing soil invertebrate- or plant-based Eco-SSLs for the individual EMs, and will be made available for use in Ecological Risk Assessment of terrestrial habitats at U.S. Army testing and training sites and other military locations. The EC20s can provide site managers and regulators with a risk assessment tool which allows them to make decisions specific to their local site conditions (e.g. HC5 or HC90 protection level) that they wish to use to derive a site-specific SCV protective of plants, soil invertebrates, and critical soil processes.

TH139

Occurrence responses of oligochaeta 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 control, contains 60% copper. This high copper content may significantly contribute to the copper volcano 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 (COPRO). Bacterial data were obtained from the inoculated soils and discovered the presence of copper oxychloride in soils and earthworm tissues were determined. Findings revealed that **A. andrei** in inoculated substrates (200 mg kg\(^{-1}\)) exhibited significantly higher (p < 0.05) preference and reproductive success was recorded. However, at 1000 mg kg\(^{-1}\) copper oxychloride soils, no distinct effect was observed on both **E. andrei** and *E. albidus* in bacterial inoculated and non-inoculated substrates. In conclusion, **Achromobacter sp.** - *Bacillus cereus* bacterial consortium decreased the ecotoxicity of metal-based fungicide towards **E. andrei** and E. albidus; *E. andrei* 200 mg kg\(^{-1}\) copper oxychloride. These results further confirm the Cu tolerance potential of these bacterial strains at 200 mg kg\(^{-1}\). **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. Oligochaeta

TH140

Development of a terrestrial biotic ligand model (TBLM) for predicting acute toxicity of cadmium and zinc to soil collembolan Paronychiurus kimi

J. Son, K. Cho, Korea University / Division of Environmental Science and Ecological Engineering

Cadmium and zinc are toxic to many species of humans and the environment. However, their bioavailability of organic chemicals. Data for reproduction (soil invertebrates), growth (plants), and critical soil processes (basal and substrate-induced radiation, 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.

**Toxicological benchmarks** developed in studies with soil invertebrate and terrestrial plants were used to derive test 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 Cleanup Values (SCVs). Based on established SSD and Eco-SSL values developed in these studies will be submitted to the USEPA Eco-SSL Work Group for use in establishing soil invertebrate- or plant-based Eco-SSLs for the individual EMs, and will be made available for use in Ecological Risk Assessment of terrestrial habitats at U.S. Army testing and training sites and other military locations. The EC20s can provide site managers and regulators with a risk assessment tool which allows them to make decisions specific to their local site conditions (e.g. HC5 or HC90 protection level) that they wish to use to derive a site-specific SCV protective of plants, soil invertebrates, and critical soil processes.

TH141

Characteristics of metal-tolerant bacterial plasmids from a platinum mine tailings dam

T. Mahlatsi, C. Bezuidenhout, M. Maboeta, North-West University / Unit for Environmental Sciences and Management

The presence of mine tailings may 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 IncQ, IncP-9 and IncW specific primers, where only IncW 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 Ni/Al(+3)Pb2+ and Ba2+ with metal resistance order of Ni/Al(+3)Pb2+>Ba2+. 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 AlNi alloy containing media. Two-dimensional electrophoresis PAGE and western blots 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 new metal-tolerant bacterial strain, which was shown to be resistant to metal-plasmid characteristics 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, Yuukianura szeptycki (Collembola: Neanuridae), to cadmium and copper

Y. Lee, Korea University; J. Wee, J. Son, Korea University / Division of Environmental Science and Ecological Engineering; Y. Kim, Korea University; K. Cho, Korea University / Division of Environmental Science and Ecological Engineering

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 **Yuukianura szeptycki**, known as the species in which they live water side, and their bioaccumulation amount 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. szeptycki** were also compared to those of other collembolan species (**F. candida** and **Paranymphius 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 production of **Y. szeptycki** was tested in a concentration dependent manner after 28 days of exposure duration. Although the response of **Y. szeptycki** to the tested metals was not highly sensitive to the other collembolan species reported in literature, the study of the response of **Y. szeptycki** 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

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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 DeJong et al. 2010.

THI48 Bioaccumulation kinetics of pesticides chlorpyrifos and tebuconazole in the earthworm Eisenia andrei in two different soils
M. Svobodová, Masaryk University RECETOX; K. Smidova, Masaryk University RECETOX / Research Centre for Toxic Compounds in the Environment (RECETOX); M. Hvezdova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); L. Bielski, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX; J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX
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 tebuconazole. 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 tebuconazole. Bioaccumulation factors were also calculated as the ratio of uptake and bioaccumulation to sorption with results comparable with steady-state bioaccumulation factors. 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. In the clay content also probably contributed, namely to degradation of chlorpyrifos by clay-catalysed hydrolysis and to decreased bioavailability of tebuconazole by binding to clay minerals. The lower Koc, and hydrophobicity of tebuconazole relative to chlorpyrifos probably lead to higher availability of tebuconazole through pore water exposure. On the other hand, higher hydrophobicity of chlorpyrifos probably caused an increase in availability by its additional uptake via ingestion.

THI49 Effects of diuron and imidacloprid on eight nematode species
J.N. Neury-Ormaiň, Iresta / EABX-CARMA; C.N. Doose, INRS - Centre Eau Terre Environnement; N. Majdi, Ecolia / UMR 5245 CNRS; J. Vedrenne, S. Morin, Iresta Bordeaux / UR EABX; S. Hłosz, Ecosa / Animal Ecology; W. Traunspurger, Bielefeld University / Animal Ecology
To assess how terrestrial nematodes extract 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 360 days for diuron. With a toxicological constant with results comparable with steady-state bioaccumulation factors. 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. In the clay content also probably contributed, namely to degradation of chlorpyrifos by clay-catalysed hydrolysis and to decreased bioavailability of tebuconazole by binding to clay minerals. The lower Koc, and hydrophobicity of tebuconazole relative to chlorpyrifos probably lead to higher availability of tebuconazole through pore water exposure. On the other hand, higher hydrophobicity of chlorpyrifos probably caused an increase in availability by its additional uptake via ingestion.

THI50 Multigeneration effects of pentachlorophenol and 2,2',4,4'-tetrabromodiphenyl ether on Folsomia candida
M. Qián, O. Zhang / Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, China
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 unpoolluted 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 on 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 than the reproductive capacity of adults. The affected endpoints of springtails can be inferred by the two methods. PCP and BDE47 do not influence completely the same endpoints.

THI51 Bioaccumulation of lead in earthworms: a comprehensive study to derive a bioaccumulation factor (BSAF) for risk assessment
K. Oorts, ARCHE; J. Chowdhury, International Lead Association / Senior Scientist -Environment
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, endogeic 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 representative for soils in Europe. Overall 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 an eCEC of 3 and 30 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.

THI52 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 relatively 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, quinolines, carbazole derivatives, benzyltoluenes and dibenzyltoluene 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 samples. The coefficients of correlation for LOHCs (Quin-2Me) as examples. Soil ecotoxicity was estimated for the quinaldines in the soil bacteria Arthrobacter globiformis and Collembola Folsomia candida in pore-water and soil exposure scenarios. The log Koc values generally increased following indoles < quinolines < carbazole derivatives < benzyltoluenes < dibenzyltoluene. 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-2Me-Ph) in soils. The H2-rich form (Quin-2Me-H10) appeared the highest leaching capacity through the soil followed by the H2-leant form (Quin-2Me) implying the risk of groundwater contamination. Ionic-interaction was considered dominant in the adsorption of Quin-2Me-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 Arthrobacter at the highest test concentrations (500 mg L-1 and 750 mg kg-1 dry weight (dw) soil). Higher toxicity was found in the Collembola and malformations.
of cuticle in the pore-water scenario were observed. Dose-response modeling showed 10 < LC₅₀ < 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: adsorption, bioavailability, hazard assessment

TH153
Combining field measurements and biotest to assess lead and zinc phytoavailability in contaminated urban soils
M. Berzin, C. Chevassus-Rosset, CIRAD; L. Lemal, MetRHZlab; M. Montes, G. Moussard, E. Simon, M. Tella, CIRAD; M. Valimier, MetRHZlab; E. Doelsch, CIRAD / UPR Recyclage et risque; F. Feder, CIRAD; S. Legros, CIRAD / LITEN According with the French legislation on the recycling of wastes from wastewater treatment plants, a guideline was provided to stakeholders to test 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 RHIZObiotest) 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 RHIZObiotest and measuring the uptake flux of Pb and Zn in the whole plants exposed to the soil. As expected, field-collected plants exhibited a large range of Pb and 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, RHIZObiotest measurements showed that only the contaminated soil 15 exhibited a significantly higher phytoavailability than other soils and have consequently to be regarded 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?
Although the soil contamination regulation, side effects of chemical substances on the soil microflora focus on the determination of the nitrogen transformation (OECD 216). However, according to EFSA a 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 realistic conditions may be one option. Standardised field protocols are mainly addressing a small, mainly autotrophic bacterial community with comparable low diversity; (ii) respiration activity of the heterotrophic microflora as indicator for the effects on abundance and biodiversity of earthworms, taking into consideration that the available standardised field methods evaluate the 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. Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (P)

TH157
SETAC Soils Interest Group
M.H. Wagelmans, Bioeclear earth

Re-calibration of the earthworm Tier 1 risk assessment of plant protection products - an update
G. Frist, Bayer AG / Ecotoxicology; J. Bendall, Dow Agrosciences; T. Carro, FMC; H. Cunningham, Syngenta / Environmental Safety; A. Koutsafis, ADAMA; S. Loutseti, DuPont De Nemour Hellas S.A.; M. Marx, Bayer AG Crop Science Division / Environmental Safety - Ecotoxicology; B. O’Neill, DuPont Crop Protection; A. Sharple, FMG Agricultural Solutions; F. Staab, BASF SE
The assessment in tier 1 risk assessment of plant protection products (PPP) is expected to increase due to revision of the PEC conservatives modeling guidance. The new EFSA guidance foresees to use worst case PECcon 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 PECcon 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 PECcon 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 set of 5 field studies complied by EFCA companies. In this exercise, the relevant soil layer for PECSoil 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). However, uncertainty linked to soil intra- and interspecies variability and the 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.
A novel analytical method for simultaneous quantification of Bracken fern produced carcinogetic ptaquiloside-like compounds and their derivatives V. Kisielius, Metropolitan University College; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences; M. Rodamer, Agilent Technologies; D. Lindqvist, L. H. Rasmussen, Metropolitan University College

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 aqueous environments adjacent to drinking water supplies is needed. Nevertheless, their quantification and rapid determination of toxic compounds are largely unknown. This study aims to identify the environmental risk factors that predetermine release of glycosidic natural toxins from non-agricultural lands to aqueous ecosystems. Bracken ferns (Pteridium aquilinum) are known to produce up to 6 kg/ha of carcinogetic ptaquiloside. Previous studies demonstrate leaching of ptaquiloside from Bracken to soils and upper ground waters. The ptaquiloside-like compounds – ptaquiloside, caudatoside and pteroselentenolside – 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 pteroselentenolside and their respective pterosin-derivatives (6 compounds in total) to be used for the abovementioned study. The novel LC/MS method (Agilent 1260 Infinity HPLC/MS System) 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 pterosin B (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 sorption to soil and sediment, and hence reaches 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 was treated with a preservation method and a recovery percentage of the compounds was 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 demonstrating this method, we facilitate 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 – NaToxAq, which is funded by the European Union’s Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 722493.

A novel method for ptaquiloside and pterosin B preservation in groundwater samples N. Škrbić, University of Copenhagen / Plant and Environmental Sciences; S.C. Christensen, A. Pedersen, HOFOR A/S, Copenhagen; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences; L.H. Rasmussen, Metropolitan University College

Analyzing natural toxins in groundwater is challenging due to their labile and transitory nature. Ensuring sample integrity for analyses 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 pterosin B (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 sorption to soil and sediment, and hence reaches 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 was treated with a preservation method and a recovery percentage of the compounds was 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 demonstrating this method, we facilitate 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 – NaToxAq, which is funded by the European Union’s Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 722493.


Northern Kentucky University and the U.S. EPA Office of Research Development in Cincinnati Agency are collaborating to develop a harmful algal bloom detection algorithm that estimates the presence of cyanobacteria in freshwater systems by image analysis. Green and blue-green algae exhibit different Hue-Saturation-Value color histograms in digital photographs. These differences are exploited by machine learning techniques to train a smart device (cellular phone, tablet, or similar) to detect the presence of cyanobacteria in a small surface portion of a freshwater system. The Harmful Algal Bloom Classification Application (HAB APP) has been field tested and verified to classify both green and blue-green algae. Specifically, the APP has been tested on several small streams and ponds, correctly classifying green algae blooms and has been tested on the Ohio River, correctly classifying blue-green algae in the 636-mile cyanobacteria bloom in summer 2015. The application is being tested via fixed camera monitoring stations and optimized at several locations along the Ohio River and in Lake Harsha, a 22,000-acre reservoir which supplies six million gallons per day of drinking water to the Ohio county in which it lies and is a source of many recreational activities, including swimming, boating, and fishing. The presence will be verified by other detection instruments and in vitro by agency scientists and hysteresis techniques will be used to drinking water (presumably). A pre-treatment on a per-reservoir (daily) basis is planned at the monitoring stations. Further, the APP is being extended to classify harmful algae macroscopically at the genus level using a convolutional neural network approach.

Matrix-assisted laser desorption/ionization–time of flight mass spectrometry application for rapid screening of microcystins occurrence in northern Taiwan tap-water reservoirs W. Ding, National Central University / Department of Chemistry

Microcystins (MCs) are the most common hepatotoxins and tumour promoters produced by freshwater cyanobacteria. Due to the dangerousness the liver through inhibition of protein phosphatases 1 and 2A, they pose a serious health threat to humans and animals, and even inducing death. MC-LR and MC-YR are probably the most concern and toxic microcystics. They are also widely distributed and detected in the freshwater system worldwide. In this study, matrix-assisted laser desorption/ionization–time of flight mass spectrometry (MALDI-TOF-MS) technique was developed for the rapid screening of these toxins in tap-water reservoirs in northern Taiwan. In addition, solid phase extraction (SPE) was used for sample cleaning-up and target analytes enrichment. The extracted target toxins were separated on a C18 column with washing of 10% methanolic solution, and then eluting with methanol. The limit of quantitation of MC-LR and MC-YR was 0.06 µg/L, which was below the limits recommended by WHO guidelines for drinking water (1 µg/L). A preliminary result revealed that C. raciborskii from MC-LR and MC-YR were detected in two reservoirs water samples by using the developed method.

Smelly HABs: response-surface optimized HS-SPME/GC/MS method for monitoring multi-class HAB odor compounds in water C. Avagianos, M. Pisansia, T. Kaloudis, EYDAP SA / WATER QUALITY CONTROL

HABs are known to produce a wide range of malodorous compounds belonging to various chemical classes such as terpenoids, ionones, amines, aldehydes, ketones and sulfurous compounds. Such compounds have detrimental effects to the aesthetic quality of water, making drinking water unbearable by consumers and damaging recreational and tourism enterprises in lakes. Although HAB odorous compounds are generally non-toxic to humans at environmentally relevant concentrations and they do not inevitably indicate the co-occurrence of cyanotoxins, there is a need for water utilities and water authorities to apply fast and reliable monitoring for early warning and control of off-odor events. The objective of this study was to develop and optimize an efficient method for monitoring of multi-class HAB odors in freshwaters using automated HS-SPME/GC–MS. The study focused on optimization of the SPME factors, including salting-out effect, pre-incubation and extraction times, extraction temperature and stirring rate. 20 model compounds of various chemical classes were selected as indicative of the wide range of odorous compounds, ranging from volatile, early-eluting (e.g. ionyl sulfides) to late-eluting compounds (e.g. ionones). Factor effects on the responses (extracted ion peak areas) were assessed. Design of experiments (DoE) techniques included preliminary Plackett-Burman factorial designs for screening of significant factors, followed by a 4-factor Box-Behnken design to assess linear and quadratic main effects and factor interactions and to optimize the response. Optimization was based on an evaluation of the response that the objectives for maximum sensitivity screening of the whole range or of certain classes/groups of compounds. Optimization experiments resulted in full- quadratic response models for individual compounds, while desirability functions can be
Cyanobacteria are one of the components of the microalgae in suspension in periphyton formation. The community distribution is affected by water quality, flow regime, climate, and geology. During the past decades, there has been a noticeable increase in 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 determination of natural toxins from various kingdoms, those produced by aquatic organisms that are very useful to discriminate these species with respect to both the presence and identity of co-occurring cyanotoxins. The method was applied to develop the study and characterization of cyanotoxin concentrations in Catalonia freshwater reservoirs. **Keywords:** cyanotoxins, microcystins, high-resolution mass spectrometry.

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**TH165 Suspected screening of cyanatoxins in freshwater by high performance liquid chromatography coupled to high-resolution mass spectrometry**

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Cyanobacterial toxins are responsible for both acute and chronic poisoning in animals and humans. 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 (Lucertini and Ortatti, 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, PlAgD03, 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 fixative 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; algal bloom **References** Groben R. and Medlin L., 2005. In *situ hybridization of phytoplankton using fluorescently labeled RNA* probes. Methods in enzymology. 359: 299-310, I. M. Current L. 2011. Cyanotoxins: Microcystis aeruginosa for water human consumption: Guidelines for risk management. **National Group for cyanobacteria risk management in water for human consumption**, vol. 2, VIII, p. 67 Rapporti ISTISAN 11/35 Pt 2

**TH166 Adequacy of EPI Suite prediction models to estimate physicochemical properties of natural toxins potentially present in surface water**

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Natural toxins constitute a potential risk to water supplies in Europe. Only a few studies have assessed natural toxins in water according to the persistence conducted in Europe. There is thus a need to conduct new risk assessments, especially to reflect possible effects of climate change on the distribution of agricultural plants throughout the continent and to reflect increasing prevalence of monoculture farming. Furthermore, screening-level assessment of many natural toxins that have been identified but not fully assessed is needed (Bucheli 2014). Persistence and mobility of natural toxins might be useful for developing 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., Kow) 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 (Q SAR) 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 dataset to prioritize natural toxins in water according to their persistence and mobility. **References** Bucheli, Thomas D. 2014. “Phytoxins: Environmental Micropolutants of Concern?” *Environmental Science & Technology* 48 (22):13027–33. Gramatica, Paola. 2007. “Principles of Q SAR Models Validation: Internal and External.” *Q SAR & 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.

**TH167 Cyanobacterial oligopeptides of environmental concern and (co)production dynamics**

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Our ecosystems and drinking water resources are not only vulnerable towards anthropogenic pollutants. Natural toxins present an additional threat for which we still lack comprehensive knowledge. Cyanobacterial oligopeptides are a group of natural toxins from various kingdoms, those produced by aquatic organisms have a direct entry into our water resources. More frequent and intense surface water blooms of cyanobacteria have triggered particular scientific interest in their secondary metabolites as potential aquatic toxins. The variety of cyanopeptides is well documented since the 1990s and the growing publication record reflects an increasing scientific awareness. Cyanopeptides can be divided into subclasses characterized by indicative monomeric building blocks. Cyanopeptides are by far the most intensively studied class of cyanopeptides. While it is known that many cyanopeptides are produced simultaneously from one species, the co-production of these potential toxins has not been explored comprehensively for cyanopeptides beyond microcystins. This project focused specifically on the production and co-production dynamics of cyanopeptides under different culturing condition of common cyanobacterial strains. Our targeted LC-HRMS analysis of biomass samples of single strain cultures show that besides microcystins, cyclamides and various cyanopeptolins are co-produced. Our data shows the evolution of the
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 ptasquiloside under alkaline conditions
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The carcinogenic ptasquiloside (PTA) is found in several species of ferns worldwide. The distribution and occurrence is well described for genus Pteris (Bracken fern) which 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 streams. PTA (4,700ppb) was deglycosylated using 0.10/10/1.0 M NaOH and 3 different 0.025M buffer systems (approx. pH 7-12; NaHPO4/NaH2BO3/H3PO4; 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 area distribution of the main mass trace. 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 dienone (BDE), a ultimate carcinogenic. 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. Schoenees, Agroscope / Environmental Analytics; K. McNeill, ETH Zurich / Institute of Cytosine Biocatalysis 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.(1) 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 be transported in surface and ground water resources 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.(2) 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 toxic analysis. A multi-step molecular size and functional group specific calibration procedure is applied in indirect evaluation of natural toxic 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 isoflavonoids as reference compounds in addition to representative compounds of the aquatic environment. Thus, experimental data will help in prioritization 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] Schenkel, et al.; [3] Environ Sci Technol2012.

TH169
Phytoxins as aquatic micropollutants: a procedure for prioritization
B.F. Guenthardt, Agroscope / Environmental Analytics; J. Hollender, Eawag / Environmental Chemistry; M. Scheringer, ETH Zurich / Institute for Chemical and Bioengineering; K. Hungerbuehler, ETH Zurich / Bioengineering; T. Bucheli, Agroscope ART / Environmental Analytics
Phytoxins are natural toxins produced by plants with widely varying molecular structures and toxic effects. Despite possibly high concentrations of natural toxins in vegetation, crops and freshwater, they are not yet commonly perceived as environmental contaminants of possible concern. This far, environmental exposure and effect studies have only been conducted for a very limited number of phytoxins, and systematic and larger monitoring campaigns are completely lacking. A crucial challenge is to systematically identify among the plethora of phytoxins those that actually present a serious risk for the aquatic environment. For this purpose, we ranked 1586 phytoxins from over 800 plant species compiled in a previously developed database based on three critical properties: toxicity, plant frequency and environmental behavior of the phytoxins. Toxicity was included as descriptor of the effect and parametrized by both plant, and estimated compound toxicity. Plant frequency, obtained from InfoFlora, was used to estimate the occurrence of all plant species producing a certain secondary metabolite class and serves as an approximation of exposure. To identify the phytoxins relevant for the aquatic environment we used a procedure, which systematically ranks substances suspected of causing Human gastric cancer. Pterosin B is formed from ptasquiloside upon hydrolysis. Similar pterosins are formed from caudatoside and ptesculentoside ('the ptasquiloside group'). Brackens are classified by WHO/JARC in Group 2B due their carcinogenic properties at cellular level and due to their association with several veterinary diseases. Brackens cause bovine urinary bladder cancers and are suspected of causing Human gastric cancer. Pterosin B is formed from ptaquiloside for being persistent and mobile organic compounds (PMOCs) based on volatilization, biodegradation and hydrolysis as measure of persistence and Day, the pH dependent Kow, as measure of mobility The PMOC analysis, using predicted phytoxin properties, showed that approximately 70% of phytoxins are mobile in the environment. However, over 50% of the secondary metabolite classes are not of priority due to their fast degradation in the environment. Other second order priority classes are mainly characterized by 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 streams. PTA (4,700ppb) was deglycosylated using 0.10/10/1.0 M NaOH and 3 different 0.025M buffer systems (approx. pH 7-12; NaHPO4/NaH2BO3/H3PO4; 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 area distribution of the main mass trace. 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 dienone (BDE), a ultimate carcinogenic. 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.

TH170
Sorption of pterosin B to soil materials
J. Andersen, L. Rasmussen, Metropolitan University College
Bracken ferns (Pteridium sp.) are considered environmentally problematic due to their content of the carcinogens ptasquiloside, caudatoside and ptesculentoside ('the ptasquiloside group'). Brackens are classified by WHO/JARC in Group 2B due their carcinogenic properties at cellular level and due to their association with several veterinary diseases. Brackens cause bovine urinary bladder cancers and are suspected of causing Human gastric cancer. Pterosin B is formed from ptaquiloside for being persistent and mobile organic compounds (PMOCs) based on volatilization, biodegradation and hydrolysis as measure of persistence and Day, the pH dependent Kow, as measure of mobility The PMOC analysis, using predicted phytoxin properties, showed that approximately 70% of phytoxins are mobile in the environment. However, over 50% of the secondary metabolite classes are not of priority due to their fast degradation in the environment. Other second order priority classes are mainly characterized by 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 streams. PTA (4,700ppb) was deglycosylated using 0.10/10/1.0 M NaOH and 3 different 0.025M buffer systems (approx. pH 7-12; NaHPO4/NaH2BO3/H3PO4; 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 area distribution of the main mass trace. 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 dienone (BDE), a ultimate carcinogenic. 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.

TH171
Modelling the fate of natural toxins in the soil using DAISY- a case study of pterosin B.
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Cyanobacteria form blooms in freshwaters due to environmental pollution and can exhibit toxic effects on human and ecosystem health. Modelling of natural toxin fate presents several challenges compared with xenobiotics: many and partly continuous sources, variable and poorly studied physicochemical properties of the toxins, highly variable temporal and spatial rates of transfer of the toxins from the source plant to soils, often linked to specific environmental conditions, and other mechanisms. Hydroxyl radicals and other mechanisms are non-selective and randomly attacking oxidants, usually reacting with rate constants orders of magnitude higher than for other oxidants. So far, most research focuses on treatment of microcystins, but other toxin classes such as nodularins, saxitoxins, cylindropermopsin and anatoxins have also been shown to be susceptible to be removed by AOP treatment.}

The most often reported AOPs for the removal of cyanotoxins include ozonation, photo- or sonolysis, direct and catalyst-enhanced photo- or sonolysis, and combinations of these or with hydrogen peroxide. Lesser studied, but still very promising AOPs for the removal of cyanotoxins from drinking water are sonochemical and hydrodynamic cavitation, electrochemical oxidation, radiolysis and other novel approaches such as those based on non-thermal plasmas. The present paper summarizes pros and cons of AOP technologies for the removal of cyanotoxins from drinking water and presents the first experimental evidence for the use of alginate nanoparticles to deliver ultrasound to the aquatic environment, providing an alternative AOP approach to those currently used. The results of these experiments highlight the potential of sonochemical approaches for the treatment of cyanotoxins and provide a basis for future development of sonochemical technologies for the removal of cyanotoxins from drinking water. Further research is needed to optimize the treatment conditions and to evaluate the effectiveness of sonochemical technologies for the removal of cyanotoxins from drinking water.
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, and pistachios with intact cuticles are more resistant to the A. flavus colonization. The paper should be of interest both for readers in the areas of hazard analysis for monitoring purpose, and for other researchers in mycotoxin field, due to the great utility of low-cost, rapid and reliable methods of analysis for aflatoxin determination.

TH176
Impact of climate change drivers on toxin contamination and genotoxicity in Mytilus galloprovincialis: combined effects of warming, acidification and harmful algal blooms.

A.R. Braga, Biology Department CESAM, Aveiro University; C. Camacho, IPMA, LP.; V. Pereira, R. Marçal, A.M. Marques, Biology Department CESAM, Aveiro University; S. Guillerme, Biology Department CESAM, Aveiro University / Bussels were exposed to G. catenatum, DNA damage in both gills and M. Pacheco, Biology Department CESAM, Aveiro University / Dept of Biology; P. Costa, IPMA, LP.

Mytilus galloprovincialis under current environmental conditions and at conditions simulating scenarios of climate change, namely warming, acidification and the combination of these two factors were exposed to paralytic shellfish poisoning (PSP) toxin-producing Alexandrium catenatum. Shellfish toxicity derived from accumulation of alga toxin was assessed in mussels at the four treatments as well as the damage at DNA level via the comet assay. Mussels were acclimated for 21 days and then exposed to G. catenatum, during 5 days (upake), followed by 10 days with non-toxic diet (elimination). The analyses of PSP toxins in the mussels were carried out by Liquid Chromatography with Fluorescence detection. The highest PSP content was observed at day 5 in mussels in the actual conditions (1493.8 ± 202.4 µg STX eq/kg⁻¹), which exceeded the international seafood safety limits (800 µg STX eq/kg⁻¹). Significantly lower PSP content was observed in mussels under climate change scenarios. The lowest levels (661.9 ± 22.8 µg STX eq/kg⁻¹) were found in warm-acclimated mussels, followed by acidification (761.2 ± 62.3 µg STX eq/kg⁻¹). However, interaction of both parameters did not reveal an additive effect. Lower toxin elimination was observed in warm-acclimated mussels. Genotoxicity was assessed in gills and hepatopancreas of mussels sampled at the end of each period. In mussels not exposed to toxic algae, the comet assay revealed highest damage levels in mussels under combined effects of warming and acidification at the end of the experiment (i.e after 36 days). When mussels were exposed to G. catenatum, DNA damage in both gills and hepatopancreas significantly increased at an earlier stage, i.e just after the uptake period. The treatments representing the acidification scenario and the interaction of warming with acidification revealed higher DNA damage than the actual conditions, highlighting a synergistic impact. DNA damage decreased in all treatments at the end of the elimination period, although reduction was subtle in mussels under interaction of warming and acidification. This is the first study assessing the impact of the combined effect of warming, acidification and biotoxins in shellfish. In conclusion, it was provided evidences that changes of global conditions may lead to lower PSP contents, but also to slower elimination rates and to a synergistic effect on DNA damage implying possible consequences for the mussels populations.

TH177
Interest of bivalves for the biosurvey of cyanotoxins in aquatic ecosystems

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The eutrophication of aquatic ecosystems, associated to climate change, enhance the frequency and the severity of cyanobacterial proliferations. Cyanobacteria are photosynthetic organisms producing endotoxins such as neosaxitoxins, hepatoxins, dermatotoxins, and cytotoxins, threatening target organisms and humans. The bivalves are target organisms in aquatic ecosystems, these impacts on organisms is overall quite well documented. However, the neuretoxin β-methylamino-L-alanine (BMAA), suspected to be a causative agent in the human degenerative disease amyotrophic lateral sclerosis (SLA), is less studied. The bioaccumulation of BMAA has recently been demonstrated with highly selective analytical methods in various marine organisms (zooplankton, mussel, oyster, fish), but rarely in freshwater organisms. Bivalves are known to consume phytoplanktonic species such as cyanobacteria or diatoms, both known to produce BMAA, and can be used as sentinel organisms to reveal the environmental contamination. A dual approach, in the laboratory and in situ, is used to evaluate the pertinence of the bivalves Anodonta anodonta, Dreissena polymorpha and Mytilus edulis as bioindicators of the contamination of fresh and estuarine waters by MCs and BMAA. The laboratory approach consist in the evaluation of the kinetics of BMAA and MCs accumulation and detoxification in bivalves at various times and concentrations of exposure. The in situ approach consist in the evaluation of the MCs and BMAA accumulation in caged bivalves along a river continuum from with nutrient freshwater to interconnecting estuarine and coastal areas used for mussel aquacultures. First results show MC and BMAA accumulation on laboratory-exposed D. polymorpha and A. anodonta, with varying kinetics. Freshwater and marine bivalves also accumulated MCs in situ and a MC transfer from fresh to estuarine waters occurred, highlighted by an accumulation in the marine bivalve M. edulis. The results of this project will facilitate the long-term tracking of the contaminant ecosystems by cyanotoxins, which will provide an advance in the knowledge about the ecodynamics of cyanotoxins and the changes conditions of human exposure.

TH178
Tetrodotoxin an Emerging Threat to Humans in the Mediterranean Area: First detection in Italian Mussels

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Tetrodotoxin (TTX) is one of the most potent neurotoxins, originally found in ovary and liver of pufferfish (Tetraodontidae) [1]. Succesively, 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 shells poisoning toxins (PST) producing organisms – was proposed as potential biogenetic 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 cultivated puffer fish Fugu rubripes. Toxicon 45:851-859. [2] Yasumoto T, Kojima N, Hayasuma D, Yotsu M, Michishita T, Endo A, Kotak Y. 1986. Bacterial production of tetrodotoxin and anhydrotetrodotoxin. 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.

TH179
The first report on neurotoxic anatoxin-a occurrence in cyanobacterial blooms in the Czech Republic

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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 cyanotoxins 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,
-RR, -YR, -LF, -LW, -LA, -LY, -WR; nodularin; cylindrospermopsin and anatoxin-a) as well as other bioactive metabolites of cyanobacteria (isomers of lipopeptide puwainaphycin F). The quality of the anatoxin-a analysis was assured by the use of D5-phenylalanine internal standard. Cylindrospermopsin (CYN) has been confirmed (4.25 microgram/dL) in a single bloom from the pond Pisecky (South Moravian region close to Slovakian and Austrian borders) detected by invasive species Cylindrospermopsis raciborskii. The other species found in CYN-positive bloom are Cylindrospermum and Chroomonas. Cylindrospermopsis aphanoizemone (formerly known as Aphanabaena aphanoizemone). Pseudoanabaena limnetica and Planktothrya limnetica. For the first time we have identified anatoxin-a in total 3 samples from the Czech Republic (concentration ranging 0.34 - 2.82 microgram/L), all originated from South Bohemian region around lake Bajer (sampling years 2014/2015). Three species of Dolichosphurmus sp. were found in all three anatoxin-a positive samples (different species in different samples - D. planctonica, D. smithii and D. flos-aquae). The phytoplankton communities of the anatoxin-a positive blooms were fairly rich in composition containing also Aphanopsaca sp., Aphanizomenon sp., Microcystis sp., Woronichinia sp., Sphaerospermopsis sp. In addition to CYN and anatoxin-a, the paper discusses concentrations of other above mentioned cyanotoxic bioactive metabolites and their risks.

TH180
Toxic cyanobacteria succession during a drier summer in a water reservoir in Sicily, Southern Italy. Implications for monitoring programs and risk assessment.


Sicily, in the middle of the Mediterranean Sea, is the largest Italian island and is characterized by a dry hot climate. During the fourties-sixties of the last century several water reservoirs have been built for drinking and irrigation water, and some of them have been interested by harmful cyanobacteria blooms. However, monitoring programs have been discontinuous, and no recent data are available for most of them. Therefore, a quarterly two years survey of main lakes used for drinking and irrigation water started in 2016, with a complete (chemical, physical, microbiological and microscopic) analysis of samples, according to the Italian D.Lgs 152/2006, combined with a water toxicity assessment through a Vibrio fischeri ecotoxicological test (ISO 11348:3-2007), Lake Disueri (37°11'26"N 14°17'16"E) was the only one in which a persistent bloom occurred during 2017 summer. After the July sampling when a Microcystis sp. bloom was first detected, the frequency of sampling was increased, to assess the risk of the exposed population and wild and domestic animals. Between mid-Jul and mid-Sept the composition of the cyanobacterial community changed dramatically. In July the bloom was dominated by Microcystis sp. and Cylindrospermopsis raciborskii (in the order of 10⁸ and 10⁷ cell/L, respectively), the only two species detected. By mid-Aug these two species disappeared and have been substituted by Anabaenopsis sp. and Plankthotrix rubescens, which in mid-Sept were still growing (10⁷ and 10⁶ cell/L, respectively). Disueri Lake is among the largest lakes, with a surface of 1,85 km² and a maximum depth of 31 and 15.2 m. However, due to landfill and increasing and persistent dryness, depth can be reduced as much as to 1 m. Cyanobacteria are not always present (from Nov 2016 until Jul 2017 no species has been detected) and sediments represent probably the source for the inocula triggering the blooms, as well as a sink-source for nutrients, in the agricultural area of the water catchment. Results on chemical (nutrients and cyanotoxins) and molecular analysis will be discussed with a risk-based approach, to determine the risk for health for the population and to plan future management strategies.

TH181
Cyanobacteria taste and odor compounds; a study in freshwaters of Greece: T. Pla, D. Christodoulopoulos, CYANOCOST COST Action ES 1105; J. B. Diaz-Alvarez, M. Panou, Aristotle University of Thessaloniki / School of Biology; S. Zervou, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; M. Psiania, EYDAP SA / WATER QUALITY CONTROL; C. Christophoridis, T.M. Triantis, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; A. Hiskia, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; S. Gkelis, Aristotle University of Thessaloniki / School of Biology; C. Avagianos, EYDAP SA / WATER QUALITY CONTROL; J. M. F. Lemos, Polytechnic Institute of Leiria; T. F. Simoes, S. C. Novais, Polytechnic Institute of Leiria

Invasive alien species represent a worldwide threat to the integrity of native communities, which increasingly compete with autochthonous species and global changes is now presenting concerns as chemical pollutants do, and are thus often referred to as biological pollution. The red seaweed Asparagopsis armata exhibits a strong invasive behavior and it is included in the list of the “Worst invasive alien species threatening biodiversity in Europe”. This alga has been shown to produce a large diversity of halogenated volatile organic compounds with potent biological effects. It can be found on tide pools during the low tide, where its exudate released into the water represent a threat to the organisms present in just a few hours, leading to a reduction in abundance of native species. Marine organisms, in particular invertebrates, have proven to be a major source of unique fatty acids (FAs). Membrane lipids, results showed that a wide range of T&O compounds were present in natural water samples and cyanobacterial extracts. Examples of odorants (odors) include trimethylamine (fishy), dimethyl- and trimethyl-sulfide (septic), methanethiol (septic), b-cyclotricr (tobacco), a- and b-ironones (floral). Interestingly, results showed that in surface water bodies of Greece geosmin and MIB have a minor role, while other T&O compounds having characteristic odor (e.g. fishy, swampy) may be more important. Based on these results, T&O profiles of cyanobacteria strains were developed. It is concluded that non-TH182
Determination of multi-class cyanotoxins in fish tissues

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The analysis of cyanotoxins in aquatic organisms, particularly in fish, has lately received increasing interest, due to environmental concerns and public health issues. This study presents the development and optimization of novel, sensitive and accurate analytical methods for the simultaneous determination of multi-class cyanotoxins i.e. Cylindrospermopsin (CYN), Anatoxin-a (ANA-a) and 12 Microcystins (MCs), in freshwater fish tissues. For the efficient extraction of selected cyanotoxins from fish tissue (muscle and liver), prior to LC-MS/MS analysis, several combinations of extraction solvents at different pH were tested. Additionally, various treatment techniques, i.e. protein precipitation using addition of salts and hexane extraction of lipids, were also tested in order to eliminate matrix effect. The LC-MS/MS method was then applied to the target toxins. Matrix interferences are a major issue in the analysis of toxins in fish tissues, beyond geosmin and MIB. Acknowledgement - The authors thank CYANOCOST COST Action ES 1105 www.cyanocost.net. C. Christophoridis acknowledges 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. First, after calculating the lethal concentrations of the alga exudate, Gibbula umbilicalis and Palaemon 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 shrimp hepatopancreas. Result 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 on toxicity 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
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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 alga 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 Palaemon serratus and the marine snail Gibbula umbilicalis to the exudate of this macroalgae. The seaweed collected at the coast of Peniche, (Portugal) was left in laboratory tanks, for 12 hours, in the dark at 20ºC±1. Effects of macroalgae exudates toxicity in invertebrates were tested. After assessing the lethal concentrations of the algae exudate, 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 analyzed on invertebrates showed an impairment of their physiological status after exposure to this algae exudate, with both species suffering from disruptions on their own normal 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 on this macroalgae on the invaded ecosystems under a global change scenario.

TH185 Assessing consumption risks through cadmium-contaminated shellfish amplified by ocean acidification
When, Kashef Dehghan University / Dept Biological Science and Environmental Biology; H. Lin, National Taiwan University; S. Chen, Chung Shan Medical University / Public Health
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 model and projected emission scenarios representative concentration pathways 8.5, and Cd 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 rates 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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Amphiphilic eutrophic purpose, climate change results in the increase occurrence and intensity of toxic cyanobacterial blooms. Cylindrospermopsin (CYN), a (sub-)tropical cyanobacterial toxin of emerging concern, is detected in temperate climates with increasing frequency, thus driving the scientific effort to investigate health risks linked to CYN-producing blooms. Exposure to CYN occurs primarily orally, causing hepatoxic effects. However, extrahepatic manifestations of CYN toxicity have also been reported and adverse respiratory conditions have been frequently linked to cyanobacterial blooms. Detection of cyanobacterial toxins in aerosols and dust particles raises the question of potential associated hazard of human exposure via inhalation. The susceptibility and vulnerability of human bronchial epithelia to CYN were investigated in vitro. To assess inhalation toxicity on airway epithelia, monolayers of immortalized human bronchial epithelial cells A549 and 16HBE14o- were exposed to a concentration range of 0.1-5 µM CYN. Cytotoxic endpoints were assessed as morphologic alterations, resazurin reduction capacity, esterase activity, membrane integrity and by real-time cell analysis. Both cell lines were sensitive to CYN. Depending on the endpoint assessed, EC50 values ranged between 0.8-2.1 µM (HBE1) and 1.6-4.8 µM (16HBE14o-). To evaluate alterations of other cellular events by sub-cytotoxic concentrations of CYN, phosphorylation of regulatory switches, mitogen-activated protein kinases (MAPKs) ERK and p38, was evaluated. After prolonged exposure (8-48 h), stress-activated MAPK p38 was hyperphosphorylated in both cell lines, while elevated phosphorylation levels of ERK following CYN treatment were detected only in 16HBE14o- cells. This study suggests possible hazards of cyanotoxin inhalation, which might impact the integrity of airway epithelia and epithelial cell signalling, including chronic inflammation due to extended p38 hyperphosphorylation. Further research of CYN-induced toxicity and underlying mechanisms is needed, as well as more data on environmental concentrations of cyanotoxins in aerosols and exposure assessment. The research was supported by the Czech Science Foundation Grant No. G17-23579Y and from H2020-MSCA-ITN-2016 Project No.722493 NaToxAq.

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 cyanotoxin detected in the environment, known to induce primarily hepatotoxic effects. However, we investigated effects of MCLR on in vivo and intraperitoneal administration of the toxin. Therefore, we investigated effects of MCLR in human bronchial epithelial cell lines (HBE1, 16HBE14o-, BEAS-2B). Cyanobacterial lipopolysaccharides (LPS) represent another bioactive component of cyanobacterial biomass, which is likely to expose human beings simultaneously with MCLR, thus we studied also effects of LPS isolated from a culture of Microcystis aeruginosa PCC7806. Dose- and time-dependent formation of MCLR-protein adducts was observed in the exposed human bronchial cells. Several genes from OATP family previously implicated in the cell uptake of MCLR were found to be expressed in HBE1 and/or 16HBE14o- cells. Nevertheless, MCLR (up to 20 µM and 48 h) did not induce significant cytotoxic effects. MCLR targets protein phosphatases (PP1/PP2A), which are the major regulators of MAPKs ERK and p38. Although protein adducts with the molecular weight corresponding to MCLR-PP2A complex were detected, MCLR did not alter phosphorylation of MAPKs ERK1/2 and p38 in bronchial cell lines. Short
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 MCLR cell 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. Inhalation toxicity of other hazardous cyanobacterial blooms components and their complex mixtures, such as extracts and LPS isolated from different cyanobacterial strains and natural water blooms, should be 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 NPeToxQ.

TH188
Estrogene and retinoid-like activity in stagnant waters
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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 human. Recent investigations indicate that cyanotoxins like cyanotoxins 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, alkylphenols or phytosterogens. Other compounds might play a role in these effects as well. Maximal detected retinoid-like activity in water samples reached 256 ng REQ/L. We analysed the presence of nine retinoic substances, where 4-keto all trans retinoic acid and retinal were the most common forms detected in the samples. Retinoid-like activity was almost fully explained based on concentration and relative potency of individual for liver retinols. However, we also suggest that still other compounds with retinoic acid receptor-mediated modes of action are present. Our study highlights the ability of common species of cyanobacteria to produce retinoids naturally and excrete them directly into the environment in concentrations that can reach hazardous level for vertebrates in aquatic environment such as fish or amphibians. The SOLUTIONS Project is supported by the Seventh Framework Programme (FP7-ENV-2013) of the European Union under grant agreement no. 603437.

TH189
Excitative effects of 2,4 -diaminobutyric acid on leech Retzius nerve cell membrane potential
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Neurotoxicity of 2,4-diaminobutyric acid (DABA), a non-protein amino acid, was first shown after TH189

Generating ectotoxic information on microcystins and prymnesins: A different approach
There is a lack of information for estimating safe levels for aquatic life concerning the toxicity of natural toxins produced by cyanobacteria and algae. Current literature indicates that LC50 for cyanobacterial toxins range from 21-2 mg/L. There is even less ectotoxicity information available for prymnesin which is produced from the estuarine algae Prymnesium parvum. This flagellated 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 (2015) conducted toxicity tests on several different cladoceran species using lyophilized phytoplankton samples collected from hydroelectric/drinking 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 toxoproducing 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 4°C. The F. aequorae cells were not freeze/thawed. Forty-eight hour acute tests were conducted with Ceriodaphnia dubia, Hyalella azteca larval Pimephales promelas and Neolecomotriangularis 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. triangularis and H. azteca only when tested in Moderately Hard Reconstructed Water but not in Reformulated Moderately Hard Reconstructed 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 microcystin chronic results will also be presented.

TH191
Protemic analysis of rice plant exposed to long-term microcystin LR exposure
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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 MCs induced inhibition in cell growth and migration 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 of the molecular mechanisms underlying adverse effects in terrestrial plants exposed to water containing microcystins (MCs). Keywords: rice, microcystin-LR, photosynthesis, proteomics Acknowledgements This research was financially supported by the National Natural Science Foundation of China (Grant number 21407056).

Developments in the use of bioassays for chemical and environmental risk assessment (P)

TH194 Responses to PFOA and PFBS exposure in the sediment dwelling invertebrate Dendrobena veneta (Annelida)
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Toxicity of Perfluorooctane sulfonate (PFOA) and perfluorooctanoate (PFOS) for short (72 h) and longer (14 and 28 days) times. The exposures were carried out in soil microcosms prepared with glass containers filled with 300 ml of soil humidified at 30% with PFOA or PFBS spiked water. An As for the 72 h tests the exposures were performed, at two different nominal concentrations, i.e. 30% of 1 or 10× 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 5× MAC-EQS fw values. Our results show, for the two 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.

TH195 Toxicity of Per- and Polyfluoroalkyl substances on Chironomus dilutus for use in a relative toxicity model
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Per- and polyfluoroalkyl substances (PFASs), including perfluorooctanoic acid (PFOA) and perfluorooctanoate (PFOA) are commonly elevated in soil and groundwater. High detection frequency and concentration has resulted in identification of PFAS as compounds of interest and as emerging contaminants due to their regulatory uncertainty. Published toxicological research to date relates to PFOA 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 PFAS 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 dilutus 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 deformation 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 models. 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 toxicity 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.
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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 which the activity stopped about 100 years ago (the “TRIPLE” project - OH 2017). Among the conclusions, it was noticed that the selection of the control soil may have a significant influence on the expression of the results and therefore on the risk assessment. This impact is particularly obvious for the assessment of a heterogeneous site and for the first TIER (screening level) of the TRIAD method. This statement is illustrated by observing the consequences on the assessment conclusion when the results of bioassays are expressed according different control/reference soils.

TH197
Estimating the hazardous concentrations of nonylphenol for soil ecosystem protection with protection levels approach
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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, secernentea, chitellata and collombella) were tested. Also, for the chronic assay, four soil species from four different taxonomic groups were investigated. Finally, acute and chronic hazardous concentrations for H2C, H2O, HC2, HC3 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
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The toxicological properties of organophosphate (OP) triesters that are used as flame retardant and plasticizer additives are currently not well understood. However, there is increasing evidence that suggests OP triesters can affect the thyroid system. Perturbation of thyroid hormone (TH) transport is one mechanism of action that may affect thyroid function. The present study applied an in vitro competitive protein binding assay with thyroxine (T4) and human transthyretin (hTTR) to determine the potential for the OP triesters, TDCIPP (tris(1,3-dichloro-2-propyl) phosphate), TBOEP (tris(butoxyethyl) phosphate), TEP (triethyl phosphate), TPFP (triphenyl phosphate), p-OH-TPFP (para-hydroxy triphenyl phosphate), and the OP diester DPH (diphenyl phosphate), to competitively displace T4 from hTTR. Enhancement of T4 binding to hTTR, rather than the hypothesized competition, was observed for the six OP triesters and DPH and in a concentration-dependent manner. For example, T4-hTTR binding was significantly increased at concentrations of TBOEP as low as 64 mM, and to 184 % at 5,000 mM. A plausible explanation of these results, which to our knowledge has not been previously reported, may be allosteric interactions of the OP esters with hTTR allowing T4 to access the second site of the TH binding pocket. It is plausible that OP triester and diesters can covalently bond to residues of serine, lysine or tyrosine on the surface of hTTR, resulting in a conformational change in the dimer–dimer interface and allowing for both TH binding pockets to be accessible for T4. These in vitro results suggest a novel mechanism of OP ester toxicity via T4 binding enhancement, and possible dysregulation of T4-hTTR interactions.

TH199
In Vitro and In Silico Competitive Binding of Brominated Polyphenyl Ether Contaminants With Human and Gull Thyroid Hormone Transport Proteins
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Tetradeacabromo-1,4-diphenoxoxenzenyloxy (TeDB-DiPhOBOz) is a highly brominated additive flame retardant (FR). Debrominated photodegradates of TeDB-DiPhOBOz have been shown to be enzymatically hydroxylated in vitro in herring gulls (Larus argentatus) liver assays, including one metabolite identified as 4′-OH-2,2′,4′-tetrabromo-DiPhOBOz. Chemically related methoxylated tetraacerbo- to hexabromo-DiPhOBOz are known contaminants in herring gulls from the Laurentian Great Lakes of North America. To our knowledge, nothing is currently known about the biological effects of these polybrominated (PB) DiPhOBOz-based compounds. The present study investigated the potential thyroid hormone affinity of 2,2′,4′-tetrabromo and DiphOBOz. Three strains of S. oryzae – and hydroxy-analogues, using an in vitro competitive protein binding assay with human thyroid hormone (TH) transport proteins transthyretin (TTR) and albumin (ALB). T4a-OH-tetrabromo-DiPhOBOz was found to be capable of competing with thyroxine (T4) for the binding site on human TTR and ALB. The para-MeO-tetrabromo-DiPhOBOz and the tetrabromo-DiPhOBOz were much less competitive. The results suggest that these tetraacerbo-DiPhOBOz-based compounds may also act as ligands for an avian TH protein despite evolutionary differences compared with human TTR. This analysis found all three tetrabromo-DiPhOBOz analogues to be potential ligands for gull TTR, and with similar binding efficiencies to THs. Overall, the results indicated both species- and structure-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
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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 to phosphine resistance. Three strains of 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 6.65 mM for C, MR, and R strains. Lineeweaver-Burk plot for COX using ethyl formate exhibited different modes from C strains to C strains. And six genes wnt7, wnt11, casp, jhip, wnt7 gene was also upregulated in the R 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 jhip 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
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Glutathione is an important non-protein compound and existed in both internal and external of 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 very essential with development of high-sensitivity 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 GSH 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 5mM 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 detection was also validated by adding larger amount of water improves both the contaminants detection within aquatic ecosystems and the ecological risk evaluation. The ecological risk assessment aspect is 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 original 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.

TH202
Rapid analysis of bivalves' xenometabolome using High Resolution Mass Spectrometry
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A new method for the analysis of contaminants 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 xenometabolome, 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 xenometabolome, 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 in 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 environmental concentrations (PECs) or EQS values) and can identify potential stressors among large assemblies of pollutants that can give rise to a given AO. The NIVA RAd™ 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 development of an 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-EEDRISK (www.niva.no/eedrisk), 268294 MixRisk (www.niva.no/mixrisk), 223268 CERAD (www.nmbu.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, poses impacts on marine organisms. However, little is explored for potential risks of environmentally relevant concentrations of MPs to benthic organisms. ORBi: Assessing the risk of the marine bivalve Mytilus posed by environmentally relevant concentrations of polystyrene microplastics (PS-MPs) and MPs based on bioassays results from related published literature. METHODS: We used Hil-based dose-response model to simulate the effects of PS-MPs on the lysosomal destabilization and phagocytosis in bivalves. The predicted no-effect concentrations (PNECs) causing 1% inhibition of immune functions were also estimated. A risk-based probabilistic model was used to characterize the potential hazards of marine bivalves in response to predicted environmental concentrations (PECs) of PS-MPs or MPs by quantifying exceedance risks (ERs) and hazard quotients (HQs) in five plastic-filled gyres.
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 (un)known (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 innovate 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 known that the daphnid bioassay 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 compounds that have serious impacts on water quality, most of which are not listed as priority substances. Consequently, a large portion of the observed toxic effects of surface waters cannot be attributed to compounds measured by the water authorities. Hence, there is an urgent need for an effect-based monitoring strategy that employs bioassays to identify environmental risks. Therefore, the aim of the present study was to implement innovative tools in a smart, integrated monitoring strategy, applied in a nationwide water quality assessment campaign in The Netherlands. The monitoring strategy entails a passive sampling (PS) with a battery of bioassays to investigate ecotoxicological risk to aquatic biota. At 47 locations silicone rubbers and Polar Organic Chemical Integrative Samplers (POCIS) were exposed to surface water for 6 weeks. Alongside the PS a 7-day in-situ daphnid test was performed at all locations. Subsequent to field exposure, accumulated compounds were extracted from the PS after which a battery of 3 in vitro bioassays was performed: a yeast-based LUCiferase gene eXpression (CALUX) bioassays was exposed to the re-dissolved extracts. The bioassay battery was selected such that it can identify the risk posed by a wide range of chemical pollutants and their transformation products, while simultaneously allowing for more targeted identification of groups of compounds that cause specific effects. Bioassay responses were compared to effect-based trigger values to identify potential ecotoxicological risks at the investigated locations. Subsequently, the SIMONI model was applied to rank sites based on ecotoxicological risk, rather than on the presence of priority compounds. It is concluded that the Smart Monitoring strategy allowed prioritization of sites based on ecotoxicological risks, identified the presence of hazardous compounds, regardless of being 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 - exposimetry of in vitro bioassays for priority compounds
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EU commission Water Framework Directive considers employment of passive sampling and use of effect-based tools in the monitoring of aquatic pollution. A combination of both 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 sampler device with silicone rubber (SR) and SDB-RPS Empore™ (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 literature (POCIS) or calculated from the obtained data. In both cases the samples were detected both in adsorption (ED) and partitioning samplers (SR). The samples were analyzed for aryl hydrogen-, estrogen- and androgen receptor-mediated effects using in vitro bioassays. The effects were expressed as bioanalytical equivalents (BEQobs) of respective model compounds in water. The BEQobs levels were significantly 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 (BEQcal). The comparison of bioanalytical vs. bioanalytical equivalents showed that the detected chemicals explained mostly a low fraction of the BEQobs. Only in the case of estrogenicity in extracts of the samplers collecting polar chemicals, the BEQobs was comparable with the BEQcal 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 Nisokora 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 bioassay 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 integrative samplers (e.g. SPMD, SR) an extraction is needed before sphering of biotest 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 the reconstitution of environmentally realistic contaminant mixtures in terms of qualitative chemical composition. In the current 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 Nisokora 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: acetoneitrile, ethyl acetate and dichloromethane. We exposed 80 larve divided into 8 replicates in a fully randomized setup including controls and solvent controls to each of the Speedisk™ extracts and counted larvae and copepodites after 466 SETAC Europe 28th Annual Meeting Abstract Book
Identification of Gestagen(s) and Corticosteroid(s) from Danube River wastewater sample by using LC-HRMS and non-target screening approach

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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 gestagenic and corticosteroid 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 (LV SPE) and was pre-screened on genetically modified bioassays for agonistic and antagonistic hormonal activity for progesterone and glucocorticoid receptors (PR and GR). The extracts were further cytotoxicity and estrogenic response by relevant in vitro assays. The cytotoxicity, sample was fractionated by using reversed phase-high performance liquid chromatography (RP-HPLC) by using C-18 silica based column. Two minute fractions were collected (total 30 fractions) and applied on respective bioassays and identified one agonistic active fraction for both PR and GR. Second step fractionation was performed on the only active fraction by using aminopropyl column with gradient elution with methanol/water (30:70) with 0.1% formic acid. One to two minute fractions (total 28 fractions) were collected and biological analysis of these sub-fractions revealed again one active fraction with reduced potency as compared to F18 (parent fraction). For unravelling the compounds responsible for gestagenic 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 eco-toxicological effects using effect-directed analysis (EDA)

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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 aid sustainable water management. To assess the ecotoxicological state of the Musi River, water samples (40-150 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 toxic potential to aquatic organisms by 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 lyticea 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 river and ecotoxicological 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

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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 (IITM), 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 comprehensive ecotoxicological characterisation of an urban Indian river and potentially raise awareness of possibly related risks.

TH217 NAWA SPEZ 2015: Ecotoxicological risks in five small Swiss streams within agricultural catchments

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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 surface waters in catchments adjacent to 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 benchmarks and regulatory criteria (QC), we calculated risk quotients (RQ). In a second step RQs 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 to this, the impact was 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 assessment. In Lake Mondsee this 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 WWTP inflow and outflow, plus pre-thickening (PS) and thickening stage (PS) samples were also collected. The toxicity assessment for summer 2015 was made by performing 48-h population growth (r) assays with the rotifer Daphnia pulex (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 WWTP inflow samples presented high toxicity to B. calyciflorus (EC50 > 60%). Samples of S, PS and TS were extremely/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 with the invertebrate Heterocyclops 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, S, PS and TS samples. Samples collected in the summer 2016, showed similar results for the growth inhibition for R. subcapitata and the feeding inhibition test 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 endpoints applied onto 96-well plates in the LYES

M. Ragan, Swiss Centre for Applied Ecotoxicology Eawag-EPFL / E.

SETAC Europe 28th Annual Meeting Abstract Book
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 in small concentrations. Several studies have found that SDS concentration caused temporary damage in the number of fry on the females exposed to both concentrations of SDS in the bioassay. However, there is scant information on the kinetics of the redissolving effect on test substances on 96-well plates. Furthermore, a redissolving step can be circumvented by adding samples directly to the assay, either dissolved in water or medium (or DMSO). In this study we compared the availability of four estrogenic compounds (E2, E1, EE2 and BPA) on 96-well plates in the lyticase-based yeast estrogen screen assay (LYES; this test was recently adopted as an ISO standard). Two-fold dilution series of compounds were added directly to the wells via medium (aqueous; i.e. as suggested in the LYES ISO protocol) or using ethanol (ethanolic) which was evaporated before medium was added. We tested different redissolving times by shaking the plates, using shaking times between 0 to 120 min. For redissolution, medium was transferred to new wells for further testing (redissolved) and emptied wells were given fresh assay medium and yeast cells and were also tested (rest). We evaluated the recovery of test substances in “redissolved” and “rest” wells. Results revealed that, for all test substances: 1) less activity was observed after ethanolic application compared to aqueous application, while their relative potency towards the reference substance remained comparable; 2) only about 50% (for the compounds E2, E1, EE2 and BPA) of the nominal activity appeared in the “redissolved” wells and ca. 50% of the activity remained in the “rest” wells; and 3) shaking times beyond 10 min did not further enhance redissolving. The fact that less activity was observed following ethanolic application compared to aqueous application may be because: 1) a fraction of the compounds remained sorbed to the wells and never become available to the assay medium and were only gradually evaporated along with the ethanolic solvent. To gather more information on these aspects – and to determine the actual concentrations in the two application methods – LC-MS/MS measurements of E2, E1, EE2 and BPA were performed in parallel to the bioassay. These data are currently being evaluated.

TH222
EFFECTS OF POTASSIUM BROMATE ON THE EMBRYOLOGICAL DEVELOPMENT OF THE SEA URCHIN Arbacia lixula (Linnaeus, 1758)
A. Aksu, University Ege / Hydrobiology; D. Ayvar, Les Établissements Scolaires Tétev Fikret; G. Kenanoglu, Turkish Education Foundation İnönü, Türkiye private high school; M.A. Karaaslan, University of Ege; S. tez, Ege University Potassium bromate is a powerful oxidizing agent that chemically ages flour much faster than the air. Partially brominated leavened dough, which plasticizes faster than the formation of tin, thin-walled bubbles as the bread rises. The product is fluffy, soft and naturally white. In this investigation, the embryotoxic, sporophytic effects of Potassium bromate analyzed during the development of the sea urchin Arbacia lixula from the post-fertilization to pluteus stage (72-h). Moreover, effects of Potassium bromate on fertilization success were observed. Sea urchin sperms and eggs were exposed to 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
Effect of thermal stress on endocrine disruption in Daphnia magna

J. Na, Korea University; H. IM, J. Jung, Korea University / Environmental 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 fish and mammals. Recently, several studies reported that daphnia species which reproduce by parthenogenesis may undergo male to female sex change 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 performed to test 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 a insight into the endocrine disrupting effects of EDCs on aquatic organisms under influence of thermal effluents discharged into streams and rivers.

TH225

Microplate Alga Growth-Inhibition Bioassay

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The development of new chemical compounds is a long and costly process 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 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 readout 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.

Challenges, methodological developments and practical solutions for Social Life Cycle Assessment in industry and policy (P)

TH226

Applying Social-LCA and Social Hot Spot Analysis including a SDG Evaluation to Product Assessments with SEEBALANCE®

P. Saling, BASF SE / Sustainability Strategy; A. Alba Perez, T. Gruenenwald, P. Koelch, BASF SE / CDS/S

Social criteria and objectives – such as education, health or working conditions – are becoming increasingly important which is why these factors are also addressed by the SDGs (Sustainable Development Goals). For this reason, social aspects also have an increasing impact on marketing and management decision-making processes. The SEEBALANCE® methodology, measures the ecological and economic consequences of alternate products or processes. The Eco-Efficiency Analysis is integrated to an overall result together with the Social Analysis (Figure 1).

TH227

Pilot Responsible Research and Innovation in Industry

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There is now only limited experience with Responsible Research a Innovation (RRI) in industry and there is also limited evidence of the added value of opening up the innovation process in industry for social engagement and gender considerations. In the PRISMA project (http://www.rri-prisma.eu), we overcome these current limitations by carrying out eight RRI pilot projects in a real-world industrial context. To establish the added value of the RRI approach and life gender dimension in and for industry, we assess the pilot projects on a number of product and process RRI dimensions and compare the pilots on the relevant RRI dimensions with similar projects in the same companies in which the RRI approach has not been followed. We focus on implementing RRI for some of the major technological challenges in the EU including nanotechnology, synthetic biology, Internet of Things (IoT) and self-driving or automated cars.

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) for SGI 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 Dingang, further to “Integrating Smallholders within the 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 cycle 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.

TH229

How can the social pillar be properly integrated into sustainability evaluation methodology? Evidence from bio-based products case study

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Abstract

Along with environmental and economic assessment, social sustainability of the bioeconomy have become a growing challenge, with important effects on the market uptake of bio-based products. In recent years, social and socioeconomic aspects have progressively been included in both the discourses and sustainability analyses concerning the bioeconomy. Yet, when it comes to bio-based products the situation still lags behind (Siebert et al. 2017[1]), given that bio-based products involve longer and more complex value chains (Bell et al. 2014[2]) that make the assessment of social and socio-economic impacts extremely challenging. Furthermore, the success of a sustainable bioeconomy relies on stakeholders’ acceptance – especially consumers and manufacturers – leading to a growth in demand for such products. The choice of ‘what is to be measured’ is the critical point in S-LCA, and, by employing recognised participative techniques, the stakeholders’ involvement can be used to shape the final sustainability criteria and regulatory recommendations. Against this background, our study aims at investigating social impact of bio-based products, by identifying and validating the main social impact categories pertaining to the bio-based products realm. In doing so, we employ a robust three-step methodological framework encompassing: impact categories identification, stakeholders mapping, and social impact categories validation. In order to operationalize the methodological framework, empirical data is gathered by means of in-depth literature review, stakeholders’ interviews, and focus groups. By providing empirical evidence on the social dimension, which incorporates different visions of the stakeholders involved in the bio-based value chains, our study paves the way for further developments concerning the integration of social assessments within bioeconomy context. Keywords: bio-based products, social assessment, stakeholders analysis - bio-based products - sustainable development

TH230

Methodological considerations for applying social LCA to modelled future European energy systems in the REFLEx project

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A methodology has been developed for the social assessment from a life cycle perspective of supply chains for future energy systems for the European Union
(modelled techno-economically 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 handbook 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 system model is the single point of network entry together with all upstream necessary 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 represented 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 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 modelled shall be based on projected data for the current system. However, if doing so it is necessary to be clear about how 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 Investigación, UNAM / Ingeniería Ambiental; L. Güereca, Engineering Institute Universidad Nacional Autónoma de México / 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 operation of the implications 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 USEPA/SETAC. LCI and other instruments of social impact assessment. Where, if considered five tables of the water system: water abstraction and treatment, distribution, storage, waste water collection and wastewater treatment. The method 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 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, with 0 is the worst and 1 the best; it determines a score of 0.6 for all the analysed system, which places the system analysed in Good social performance, but is identified as priority needs like decrease of overtime in the drinking water and wastewater treatment plants; improve security conditions in the facilities in order to reduce the risks to health. The welfare of workers requires attention on these points to get closer to the definition of decent work.

Improvements in environmental exposure assessment: Development and application of tools across industry sectors, regulatory agencies, and international boundaries (P)

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 allowed us to determine specific concentration levels eligible for seawater 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 concentrations considered (including releases of 1.45 mg/l for constant/frequent release and 5900 mg/l for intermittent release) are lower than those allowed 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: S. 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.

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 it has created several difficulties in its evaluation. For regulators, there is a need for comprehension of the unique agronomic practices, application techniques, environment and management concerns, both from an ecotoxicology and environmental fate perspective considering the majority of rice cultivated within the Europe is grown in paddies fields. This gap in knowledge causes 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 manufactures 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, A. Canha, in conjunction with the lack of technical 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 need of a bottom up multidisciplinary 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
The applicability of the assessment entity concept in the REACH registration of complex mixtures. A case study for fragrance substances.

K. Jenner, Givaudan / Global Regulatory Affairs & Product Safety; G. Kreutzer, Givaudan SA; S. Kern, Givaudan Schweiz AG; M. Pacella, M. Torres Sanchez, Givaudan Suisse SA

The assessment entity (AE) concept was developed by ECHA together with industry and regulatory authorities in an attempt to address the inherent difficulty of differentiating between mixture constituents and mixtures. The tool was introduced in IUCLID 6 and aims to assist users in documenting complex assessment cases in a transparent and systematic way. The assessment entities (AEs) may be imported in Chesar 3 for the purpose of exposure assessment. The relevance and applicability of the AE concept to multi-substituent substances is illustrated by a fragrance ingredient case study. For the purpose of the worker and consumer exposure assessment, data on the individual fragrance components were used. However, to assess environmental exposure and risk, a constituent block approach was used because the substance consists of components with different environmental fate properties (e.g. water solubility, log Kow, adsorption coefficient) and ecotoxicity profiles (e.g. acute EC50/LC50 values). The use of whole substance testing versus constituent data is explored. The adaptation of standard tests, such as the traditional whole substance whole-life approach, was based on using the different HPLC partitioning characteristics of the components and the use of analytical detection methods was exploited to obtain water solubility information for the individual blocks from a test performed on the whole substance.

The case study also demonstrates the combined applicability of experimental data, QSAR and read-across in the assessment of the aquatic toxicity of the individual constituents and impurities in order to derive appropriate PNECs for each assessment entity.

TH237
Canada's Approach to Determining Causes of Impact at Federal Contaminated Sites

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Canada's Federal Contaminated Sites Action Plan (FCSAP) was developed to reduce risks to human health and the environment from--and to reduce the financial liability associated with--contaminated sites. One result of this initiative has been the common use of ecological risk assessment (ERA) as a site management tool, a Focus Group developed guidance for conducting ERAs under FCSAP. One element of that guidance is a technical guidance module on conducting causality assessment. Causality assessment has the overarching goal of differentiating ecological impairment due to 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 CADESS guidance, though it is simplified for use at FCSAP sites.

The Guidance and the presentation close with an overview of the case studies of each example of causality assessment at contaminated aquatic sites from the peer-reviewed literature.

TH238
Improving "man via the environment" exposure assessment for lead: a case study with lead and recycling uses

S. Navis, Arche consulting / Laboratory of Aquatic Ecology, Evolution and Conservation; F. Verdonck, ARCHE; K. De Brouwere, VITO NV / Health; L. Geerts, VITO NV; J. Chowdhury, International Lead Association / Senior Scientist

Environment; L. Allen, S. Binks, International Lead Association

Current chemical safety assessments for metals under REACH typically include a guess, worst-case scenario pathway assessment to limit risks to human populations resulting from exposure via the environment. As a result of comprehensive biological monitoring campaigns undertaken by European Member States in the past and supplemented modelling data, a large body of data is available on lead body burden in the general European population (children and adults). However, little information is reported for blood lead in populations surrounding lead manufacturing facilities.

The guidance framework is based on multiple lines of evidence on dose, exposure, and effect. Each potential cause of impairment is transparently evaluated with respect to coherence, plausibility, consistency of association, biological gradient, complete exposure pathway, specificity, and predictive performance. Multiple lines of evidence on each candidate cause are evaluated for both consistency and coherence of evidence. The Guidance and the presentation close with an overview of the case examples of the use of causality assessment at contaminated aquatic sites from the peer-reviewed literature.
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 applied temperature. The measured chemical fate in industrial STPs (iTreat; Straiss 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. The bioaccumulation rate constants of the non-parametrized iTreat model 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 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 result from two-fold higher elimination rates which reflect 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.

TH240
Combination of remote sensing and coarse statistical data for determination of precise spatial distribution of a pesticide load onto soils at a national scale
V. Kodeš, Czech Hydrometeorological Institute / Section of water quality; L. Brodsky, Mapradix Ltd.; T. Herza, Hydrosoft Veleslavín Ltd.
Objective of the Study In order to calculate an annual pesticide load over a certain area, one needs detailed data on pesticides' application that are hard to find in a real world. One way is to collect desired data from the farmers, but this is feasible just in relatively small areas. Due to missing detailed data, we computed more precise pesticide application and related pesticide residues (in mg/kg) at each site using a coarser data (7x7 km) grid. The combination of coarse statistical data and remote sensing imagery was used for a more detailed analysis. The remote sensing product with the highest resolution (10 m per pixel) and the highest coverage of Czech soil was used. The raw false colour composite was converted to a gray scale image with 256 levels. The image was then vectorized using specific software to obtain the contour lines with data of pesticide application. The area of application was divided to 2 classes: the area covered by the pesticide and the area not covered by the pesticide. The area was divided into 5438 pixels. The spatial pattern of the pesticide application was analyzed in the R software.

TH241
A Bayesian approach to estimate biodynamic model parameters: bioaccumulation of PCB 153 by the freshwater crustacean Gammarus fossarum.
A. Ratier, Irstea 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 EPOC; L. Peluhet, CNRS / EPOC Universite Bordeaux / UMR 5805 EPOC; L. Delorme, L. Garnier, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; M.P. Babut, Irstea / Water

TH242
Bioaccumulation and biotransformation of Hexabromocyclododecan (HBCD) by the freshwater crustacean Gammarus fossarum: a Bayesian approach to estimate biodynamic model parameters.
A. Ratier, Irstea 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 EPOC; L. Peluhet, CNRS / UMR EPOC LPTC; N. Delorme, L. Garnier, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; M.P. Babut, Irstea / Water

TH243
Chemical Exposure Disparities by Demographic Traits in the US Population 1999-2014
V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; J. Colacino, University of Michigan / Department of Environmental Health Sciences; B. Loomis, University of Michigan / Department of Environmental Health Sciences
Identifying individuals or populations at high risk for adverse health outcomes due to chemical exposure requires understanding how chemical exposure patterns vary by inherent traits. Currently, we lack of comprehensive screening to study the thousands of chemicals populations are exposed to on a daily basis. The purpose of this study is to develop a systematic approach that quantifies chemical exposure disparities by demographic traits with the help of a broad set of sources. First, we identify populations at high risk for exposure. We used the National Health and Nutrition Examination Survey (NHANES) datasets to collect information on chemical biomarker measurements and demographic traits for the years 1999-2014 (n = 74,942), focusing on 229 chemical biomarkers from 16 different classes of chemicals. Poverty income ratio (PIR) was used as a surrogate variable for socioeconomic status, while cotinine levels was used as a proxy for smoking habits. We evaluated the association of each individual biomarker and various demographic factors (age, gender, race/ethnicity, PIR, and smoking status) by using generalized linear model while controlling for relevant confounders and covariates.
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 triclosan, a common antibacterial and antifungal agent, were observed to elevated in African Americans when compared to White Americans. In addition, higher levels of parabens were observed in women, 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 triclosan, which could possibly be explained by more accessibility 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

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The amount of e-waste produced globally is growing dramatically. National numbers suggest in PM 10, PM 2.5 and ultrafine particles in the air of dismantling facilities. Levels of FRs in air collected from this Canadian e-waste dismantling facility in Southern Ontario, Canada, and to estimate occupational exposure of dismantlers at the facility to these FRs. Sampling was conducted daily over a total of five days in February 2017. Thirty-three dust samples were collected using vacuum cleaners and air samples were collected using polydimethylsiloxane passive air samplers (PDMS-PASs) co-deployed with active low-volume air samplers (LV-As). A Micro-Orifice Uniform Deposition impactor (MOUDI) was used to obtain particle size distribution of air samples. Post-deployment, samples were extracted and analysed for 12 target FRs, including novel brominated flame retardants (NFRs), polybrominated diphenyl ethers (PBDEs) and organophosphate esters (OPEs), using gas chromatography mass spectrometry (GC-MS). The most abundant FRs in air and dust samples were the novel and quantified FRs in indoor air and e-waste dismantling facility in Southern Ontario, Canada, and to estimate occupational exposure of dismantlers at the facility to these FRs. Results for the MOUDI samples showed that triphenyl phosphate (TPH) and other replacement FRs were much lower than target FRs in air and dust samples. Several on-line sample preparation techniques such as liquid chromatography and spectroscopy, standard samples of arsenite, arsenate, monomethylarsonic acid, and dimethylarsinic acid were preconcentrated from 6,300- to 45,000-fold. The limits of detection obtained with UV absorbance detection were 0.08 – 0.3 ppb As. For a spring water sample spiked with the four arsenic species, LODs of 2–9 ppb As were obtained, which are lower than the WHO guideline of 10 ppb total As.

TH245

Global approaches to environmental exposure assessment of e-wastes

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Obsole 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, plastics, flame-retardants, microbes and valuable materials, including gold, silver, platinum, and palladium, among others: manufacturers, scientists, economists, 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” (#2014-031-3-000), supported by the International Union of Pure and Applied Chemistry (IUPAC), brings together multidisciplinary global experts to explore different aspects of the e-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 speciation analysis (e.g. to assess bioavailable or bioaccessible fractions) to evaluate e-waste contaminant risk.

TH246

Droplets deposition pattern from a prototype of a fixed spraying system in a sloping vineyard

S. Otsu, Italian National Research Council; G. Innererbe, A. Schmid, C. Roschatt, Laimburg Research Centre; D. Loddo, University of Padova / DAFANE, M. in water, but its unaudited 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 at Laimburg Research Centre and a trial was performed with the aim of measuring the deposition pattern of droplets on the vine 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.

TH247

Sensitive Arsenic Speciation by Capillary Electrophoresis Using UV Absorbance Detection with On-Line Sample Preconcentration Techniques

H. Lee, J. Kwon, Seoul National University; D. Chung, Seoul National University / Chemistry

The World Health Organization (WHO) guideline states that the total arsenic concentration in drinking water must not exceed 10 ppb. However, arsenic toxicity varies significantly, with inorganic arsenic species being more toxic than organic species. Arsenic speciation is therefore important for the evaluation of health risks from arsenic-contaminated drinking water. Capillary electrophoresis (CE) provides the necessary high performance separation for the determination of arsenic species including arsenite and arsenate. Using a coated capillary, several on-line sample preconcentration techniques such as liquid chromatography and spectroscopy, standard samples of arsenite, arsenate, monomethylarsonic acid, and dimethylarsinic acid were preconcentrated from 6,300- to 45,000-fold. The limits of detection obtained with UV absorbance detection were 0.08 – 0.3 ppb As. For a spring water sample spiked with the four arsenic species, LODs of 2–9 ppb As were obtained, which are lower than the WHO guideline of 10 ppb total As.

TH248

Determination of background levels of free cyanides in surface waters


Natural background concentrations of cyanide can originate from the degradation of cyanogenic glucosides, cyanide in anthropogenic sources. Recently, environmental quality standards (EQSs) for free cyanide were proposed under the European Water Framework Directive (WFD). The EU Joint Research Centre, for example, has proposed an annual average EQS of 0.5 μg/L free cyanide. Since there is a lack of reliable data on background concentrations of free cyanide in surface waters it is not clear whether the proposed EQS values can be practically implemented. To this end a project was initiated to implement and test a method that allows reliable measurements of free cyanide background concentrations in surface waters. Current methods for the measurement of free cyanide in waters only achieve limits of quantifications (LOQs) of about 1
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 up to 12 and storing 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 the sites, with levels mainly below the limit of detection (LOD, 1/3 of the LOQ), and downstream sampling points where free 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 test 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 WTW Y Waal. H. K. W. Jonker, K. L. Ghyselen, K. Van Herck, H. E. Meyer, E. Fijnvankan, H. Beck, Saarland University; K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaefeer, 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 in EU Water Framework Directive (WFD) includes priority substances, which also have to be monitored for 1 year monitoring program for priority substances, which is efficient approach in water management. However, the capturing variable contaminant level is also critical for eco-toxicological 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 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_{wia}) over the full sampling period. PDMS sheets with two different thicknesses (76 and 22 µm) were deployed without the application of performance reference compounds. From the concentration ratio from two PDMS sheets, true in situ concentration (C_{wia}) 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 made with Oasis HLB® drench group. Throughout four seasons, several monitoring sites were established in southern Germany were selected as sampling sites and duplicate passive samplers were deployed for two weeks. After sample recovery, targeted analysis via LC/MS/MS analysis was followed. Based on earlier results, both sampler types performed well and all contaminants were detected in total including 8 priority substances in EU WFD. C_{wia} values can be used as representative values for the comparison with environmental quality standards and C_{ena} values can be regarded as variable exposure level. Complementary use of kinetic and equilibrium passive sampler enabled us to comprehensively identify multiple aspect of water quality. Studies on coupling passive sampling with passive dosing mode for risk assessment are ongoing.

TH250
Improvement of relationship between water pesticide contamination and land used at a large scale using the Polar Organic Chemical Integrative Sampler (POCIS). M. Bonnaud, B. Bonnaud, M. Bernard, B. Sarr, Centre National de la Recherche Scientifique, Montpellier, France

The water quality monitoring of organic micropollutants, is generally achieved by collecting grab water samples. Some disadvantages have been highlighted with this technique, such as the lack of sufficient control of temporal and spatial representativeness. Currently, grab sampling only provides a snapshot of the contamination, e.g. leading to a partial picture during a flood event. To overcome such issues, an alternative sampling strategy can be the use of passive samplers which allow in situ pre-concentration of analytes and offer an integrative capacity of several weeks. This study focuses on the Polar Organic Chemical Integrative Sampler (POCIS), widely used for polar pesticide sampling due to its capacity and relevance of POCIS has already been demonstrated to estimate pesticide loads in small watershed monitoring. Our study aims to show that POCIS provides more comprehensive information during a large scale deployment, and then a better correlation between water quality and urban or various agricultural land uses. A selection of 46 polar pesticides was investigated in 51 monitoring network stations of the Adour Garonne basin (SW of France). These stations were selected in function of the agrochemical pressures and the land uses, assuming different contamination profiles in the different sub-watersheds. Six sampling periods of 14-days were performed over 2016. Firstly, a low loss of POCIS (i.e. < 10%) was recorded, proving a good implementation despite the complexity of field conditions, especially for large scale deployment. Secondly, this study demonstrated that the use of POCIS can provide valuable and unprecedented knowledge about pesticide contamination of the Adour Garonne basin. With the large amount of data collected during this 1-year monitoring, correlations between the targeted pesticides and the various land uses over this large watershed (116 000 km²) were established. For example, principal component analysis allowed to develop the o...
application group. Although both haemal setal 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 ingredient. A complete evaluation 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 Sediment Variation*

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 ecosystem. Studies using the OECD 308 Guideline ‘Aerobic and Anaerobic Transformation in Aquatic Sediment Systems’ and the EPA Guideline OCSPP 835.4300 ‘Aerobic Aquatic Metabolism’. Although there is no strict recommendation for the level of microbial biomass that should be contained in sediments used for testing, like there is for soils (i.e., 1% of the soil organic carbon content, OC), it is still a useful parameter to assess viability of the aquatic ecosystems. Studies using the OECD 308 Guideline in sediments is typically determined by fumigation/extraction procedure prior to test initiation (post-handling/pretreatment), near test initiation and near test termination. 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 of initial 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 0.32 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.)*

A. Drzewiecka, M. Napora-Rutkowski, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies; E. Kulec-Ploszczyna, P. Parma, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies

The hypopharyngeal glands (HGP) of Honeybees consist of many acini connected through production of “milk” containing proteinic substances to feed larvae and the 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 four concentrations (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 0.32 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.

**TH255**

*Comparison of International Quality Assurance and Quality Control Standards for High Resolution Mass Spectrometry Dioxin Analysis*

D. Thal, E. Ogban, Environmental Standards Inc; R. Vitale, Environmental Standards; D. Bly, Environmental Standards

The analysis of polychlorinated dibenzo-p-dioxins and polychlorinated dibenzo-p-dioxin (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 compared. Official method validation is based on technology, equipment and scientifically based standards 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. Greysy, University of Saskatchewan / Department of Veterinary and Biomedical Sciences and Toxicology Centre

Recent developments of new, faster and more 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 congeners even in the same sample, which can potentially interfere. Validation of a GC/OrbiTrap system brings levels of mass resolution not previously available for analysis of POPs by GC chromatography coupled with ultra-high resolution mass spectrometry (GC-UCHRMs). Here we report use of GC-UCHRMs 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 23,7,8-substituted congeners even in an extract of used motor oil. Reanalysis of fish tissues previously analyzed by a magnetic sector instrument demonstrate accurate identification and quantification of concentrations and provide valid 20°C methods. Analyses were also 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 extraction 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*

M. O’Brien, D. Doran, C. Garcia, J. Uzyczac, M. Kirby, D. Sheahan, Cefas

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 season and so the highest concentrations of HNS were at 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 relatively frequent transport in bulk quantities. Additionally, benzalkonium chloride and a surfactant are also used as disinfectants and so be of interest. The release of a GC/OrbiTrap system brings levels of mass resolution not previously available for analysis of POPs by GC chromatography coupled with ultra-high resolution mass spectrometry (GC-UCHRMs). Here we report use of GC-UCHRMs 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 23,7,8-substituted congeners even in an extract of used motor oil. Reanalysis of fish tissues previously analyzed by a magnetic sector instrument demonstrate accurate identification and quantification of concentrations and provide valid 20°C methods. Analyses were also 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 extraction 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.
Acetylcholinesterase inhibition: a comparison of available methods for determination of acetylcholinesterase in muscle tissue of Limanda limanda. J. Uyczak, 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 the inactive products choline and acetic acid. The role of AChE is to regulate the nervous transmission by reducing the concentration of ACh. When AChE is inactivated by an organophosphorus 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/IUCN 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 organophosphate 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 management. For regulatory and forensic legal approaches. 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 with 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 Ruiz, C. Rubio Montejano, Spanish Medicines Agency / Department of Veterinary Medicines

Our conclusion 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 regulatory and forensic legal approaches. 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 pursing 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, CFE - Centre for Functional Ecology; T. Natal da Luz, University of Coimbra / Department of Life Sciences; Coimbra; S. Siciliano, University of Saskatchewan / Department of Soil Science; J. Sousa, University of Coimbra / Department of Life Sciences

Due to 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 toxicity. However, the addition of other metals is not 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 metal element 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

TH258 Using Correlations of Biological Toxicity Equivalent Quotients and Toxicity Equivalent Quotients to Derive Threshold Values for Dioxin-Like Compounds in Sediment J.D. Duckworth, R.W. Walker / Aachen University; Department of Ecosystem Analysis; M. Brinkmann, University of Saskatchewan; School of Environment and Sustainability and Toxicology Centre; C. Gembé, Institute for Environmental Research RWTTH / Aachen University; Department of Ecosystem Analysis; S. Buchinger, Federal Institute of Hydrology; Department G Biochemistry Ecotoxicology; G. Reifferscheid, German Federal Institute of Hydrology; H. Hettler, R.W. Walker / 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 bioanalytical 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 waveries 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/F (r²=0.940, p<0.001) and DL-PCBs (r²=0.924, p<0.003). 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. Kierkegaard, C.L. Chen, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. McLachlan, 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. Astrup, Institute for Environmental Research - Niels Bohr Institute in Physics Hørsholm; J. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology

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 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. These data will be used to evaluate the BIONIC model, a mechanistically 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 amenable to measurement. Our first experiments are being performed with a series of primary, secondary, tertiary and quaternary amines ranging 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 uL of aquatic water is transferred with a pipette to a vial containing 600 uL 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 a much longer, 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 marine traffic and potentially greater risk of marine incidents.
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 communities demonstrated an 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, ccx, cat, gr, gst) and thyroid-related genes (trt, trf, dio1, dio2, nis) in zebrafish larvae was examined upon lead acetate treatments to have a complete analysis of lead toxicity on larvae. The comparisons of both trt, gr, gst, at the transcription level and nis were evidenced in the experiment. The expression profiles of trt, trf, dio1, dio2, nis, sult1-s1, sult1-s2, sult3-s3, utilg1, utilg2a1, 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 trt, trf and nis levels increased with the increase of oxidative stress balance. 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 cycles have already been observed. Most of the available data on LN toxicity focuses on the effect of 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 LN compounds using both individual and cumulative interactions as bioindicators of LN toxicity. From the seven organism studied (A. fischeri, R. subcapitata, C. vulgaris, B. calyciflorus), the algae B. calyciflorus, 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 major 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 different proteins and ligands. Although metals, these complexes lowered with the decrease in theionicradii, and, in detriment of these complexes, species with LNCO22− and LNIH22+ 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 algae R. subcapitata; whereas less than additive toxicity was instead observed for the rotifer B. calyciflorus. Overall, our multi-toxicity approach should not be assumed LN toxicity as 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 UNL; Y. Gopalapillai, Environment and Climate Change Canada; M. Saen, PRIET CONICET, National University of Luján; B. Hale, School of Environmental Sciences, University of Guelph, W.D. De Martino, CONICET-PIREI / 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 were exposed to Ni, Cd and Zn individually and as ternary mixtures. The influence of the uptake and toxicity of dissolved organic carbon (DOC) as an environmental light LNs, respectively, on seven aquatic species - A. fischeri, R. subcapitata, C. vulgaris, B. calyciflorus, D. magna and D. rerio - was investigated upon both individual exposures and co-exposure. Results show that both iron and cadmium levels, as evaluated from guideline values (Canadian soil PNEC values) and a ratio based on the average concentrations in a contaminated site (Sudbury) 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 communities demonstrated an 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.
guidelines for surface waters, in order to reach a good ecological water quality status for all water bodies. Nevertheless, many rivers and streams are still experiencing trace metal concentrations that exceed the current Environmental Quality Standards (EQS). In combination with other stressors, this situation may lead to an unfavorable shift in the composition of the ecological community due to a variety of direct and indirect effects. The range of concomitant contributing pressure factors is manifold, such as eutrophication. In this way, both the aquatic environment and community will respond to the presence of a stressor(s). To gain insights into the contributing factors, we are investigating eleven sites for which apparently contradictory effects are observed. That is, based on monitoring data (www.vmm.be/geoview) gathered by the Flanders Environment Agency (VMM), the sites that have an exceedance of the EQS, yet a good ecological quality is observed. This paradox was explained by biometric, methodological, and chemical differences (MMIF). We hypothesize that the macroinvertebrate communities at these locations have (i) adapted to high trace metal concentrations and/or (ii) experienced a lower metal bioavailability due to the water chemistry. To sort out the involved processes, we will systematically characterize the bioaccumulation and exposure patterns of trace metals in a suite of macroinvertebrate taxa collected at these sites and determine the trace metal concentrations in the different ecological compartments (water, sediment and biota). The results, together with general water quality parameters (pH, conductivity, temperature, DOC and macronutrients) will identify whether the ecological quality is primarily governed by chemical or biological factors, or a combination of the two. The outcomes of our research will provide mechanistic insights into the determinants of ecological quality and facilitate development of a more differentiated basis for the setting of EQS.

TH269 Effects of heavy metal mixtures on bioaccumulation and defence mechanisms in common carp, Cyprinus carpio G. Casamassimi, University of Antwerp / Biology; G. De Breec, University of Antwerp / Biology SPHERE; T. De Neve, University of Antwerp / Department of Biology (SPHERE Research Group)

The aquatic environment is continuously under threat because it is the final receptor of a large number of toxic compounds including heavy metals that can harm the health of aquatic organisms. The main goal of the present study is to better understand the effects of waterborne heavy metals and their mixtures on a freshwater fish. Common carp were exposed to sub-lethal concentrations of Cu and Zn and different combinations thereof for a period of one week at a temperature of 20°C. Our aim is to assess the effect of sub-lethal concentrations of Cu and Zn on fish survival rate, determine the bioaccumulation of heavy metal in the gills and assess changes in gene expression of two transcriptional factors, oxidative stress and defence mechanisms. Preliminary results indicate that metal accumulation induced expression of metal binding and stress proteins, and metal specific compensatory effects were seen in genes related to ionoregulation and oxidative stress. Further analysis will determine whether antagonistic, additive or synergistic effects occurred.

TH270 Silver nanoparticles exposure inhibits glycans synthesis and induces cytotoxicity in human cell line K. Shimizu, Toyo University; M. Horie, Advanced Industrial Science and Technology; S. Kashiwada, Toyo University / Graduate School of Life Sciences

Silver nanoparticles (SNPs) are used in industrial products worldwide. Hence, there are concerns about environmental and health-related issues. Although silver nanoparticles have been reported having induction of cytotoxicity and ROS accumulation, there is limited information of the toxic mechanisms. In our previous study using embryos of medaka, we have revealed that glycans are one of the targets of silver nanocolloids (SNCs). SNCs is a kind of SNPs and nano-sized particles composed of aggregated silver ions; SNCs keep balance with dissociated silver ions. Glycans have roles of cell-protective, stabilizing and barrier function, we assumed SNP would disrupt glycans function. Beyond medaka research as a vertebrate model in nanotoxicology, in order to evaluate toxic effects of SNPs on humans, we evaluated cytotoxicity of SNPs using human cell line. In this study, we employed four different SNPs including SNCs to compare their different toxicities using three human cell lines. Of SNPs, one was coated with sulfide and diameter was ca. 30 nm. Another one’s coating material and diameter are unknown. Of SNCs, one SNCs was non-coated and its diameter was ca. 30 nm. The other SNCs was coated with nitrogen and diameter is ca. 20 nm. We used three kinds of human cell lines: lung cancer-derived A549, epithelial-derived HaCaT, and monocYTE-derived THP-1 because we supposed SNPs have a chance to contact to alveolus of lungs, epidermis and blood. To evaluate cytotoxicity, each cells were exposed to SNPs or SNCs (10 μg/mL) and incubated for 24 hours, and then we measured survival rate, membrane damage, inflammatory response, ROS accumulation, caspase-3 induction, intracellular ion concentration, and gene expression. In results, SNPs suppressed survival rates. SNPs and SNCs exposures exhibited membrane disturbance and inflammatory response. However, ROS accumulation and caspase-3 induction were observed in only SNCs exposure. Measurement of concentration of intracellular silver found that higher silver concentration in SNCs exposure rather than SNPs exposure. Finally, to investigate effects of SNP/SNC exposure on glycans, we measured glycan-relative genes (ALG2, BAGALT2 and GNS) expressions. Tested gene expression levels were all suppressed by SNPs and SNCs exposures. Since this study demonstrated that SNPs inhibited glycans synthesis in medaka in vivo and human in vitro models, toxic effects of SNPs on glycans is probably universal among vertebrate organisms.

TH271 Mixture toxicity of ZnO and silver nitrate to Daphnia magna M. Baek, KIST Europe; Y. Seol, University of Science and Technology; H. Kwon, Y. Kim, KIST Europe / Environmental Safety Group

Zinc oxide nanoparticles (ZnO NPs) and silver nanoparticles (AgNPs) as Engineered nanomaterials (ENMs) can be found in numerous materials or consumer products. These applications of metal (oxide-) nanoparticles indicate that the exposure to the aquatic environment may lead to mixture forms of by biological system. In this study, the acute toxicity tests using Daphnia magna were conducted for examining the single- and mixture toxicity. The methodological approaches for mixture toxicity (Mixture I – 5:5; Mixuture II – 7:3 and Mixuture III – 3:7) were conducted as three binary mixtures of AgNO3 and ZnO based on the estimated toxicity data (i.e., EC50 values) of single substance. To compare with control response and mixture results, the mode of action in mixtures, the effects of mixture were analyzed using the MIXTOX model. The EC50 values of AgNO3 and ZnO were 0.0009 mg/L (with a 95% CI of 0.0007-0.0011 mg/L) and 2.2884 mg/L (with a 95% CI of 1.3702-3.2066 mg/L), respectively. Among the 3 mixtures, Mixure III was the highest toxicity at the low concentration. With reference at the concentration addition (CA) and independent action (IA) model of all considered, the minor cumulative effects observed can be managed by effective mitigation measures such as vegetated filter strips, conservation tillage practices or green cover crops in addition to reducing point source pollution.
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 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

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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-metolachlo 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 estuary of the Vistula River in Poland. We focused on non-target, 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), which is an important model organism. In conclusion, an indication of a novel mode of action of the chronic pesticide toxicity 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.

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The first results of the genetic and biological monitoring of the relative abundance of non-target organisms (invertebrates, algae, bacteria, fungi) in the course of 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 reached 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.

TH276 MODELLING ACUTE AND CHRONIC RISKS OF PESTICIDES RESIDUES IN SOUR CHERRIES

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The topic of tank mixes has been identified by ctgb (the Board for the authorization of plant protection products and biocides in the Netherlands) as one of the most important knowledge gap. However, 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/abstr}

TH277 Environmental and Human Cumulative Risk Assessment of Pesticides Using Local Monitoring Data: A Case Study from the Pucara River Basin, Bolivia

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Local Monitoring Data: A Case Study from the Pucara River Basin, Bolivia

L. Herrero Nogareda, University of Copenhagen / Department of Plant and Environmental Sciences; M. Álvarez Caero, H. Antzaneza Fernández, Universidad Mayor de San Simon / Facultad de Ciencias y Tecnología; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences

In Bolivia, pesticides are used intensively cultivated crop with sequential applications of products and mixtures of plant protection products and biopesticides. The current product by its chemical composition and possible multistress in risk assessment of pesticides. Current state of knowledge, based on a realistic application schedule and spray drift on surface water in the river basin. Pesticide concentrations exceeded the current risk safety thresholds for the environment and the human health at approximately 50% and 20% of the sampling points, respectively. The thresholds were especially surpassed at the discharge zone of the river basin, where SumTU reached values of approximately 0.5 for D. magna, fish and C. riparius, and HI for chronic exposure reached values of 4.70 and 1.57 for children and adults, respectively. The results suggest that pesticide pollution likely impairs the stream system biota in multiple points, while water was not acceptable for a human daily intake in two sampling points, especially for children. The detected pesticides that caused most concern 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.

TH278 Developing a strategy to improve the environmental risk assessment of pesticide mixtures using local 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 the risk of complex mixtures of pesticide mixtures. This workshop brought together stakeholders from academia, government, industry and non-governmental organizations to identify and prioritise research needs to improve risk assessment of complex mixtures. The workshop discussions highlighted the need for more research on: characterizing complex mixtures, using different exposure scenarios across the life cycle, and identifying gaps in risk assessment methodologies. The workshop also identified several research needs and priorities for future work in this area. The research needs were grouped into four categories: characterizing complex mixtures, exposure assessment and human health, risk characterization and assessment, and risk communication and education. The workshop recommendations are intended to guide future research on the risk assessment of complex mixtures and to identify gaps in current risk assessment methodologies.
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 Agriculture, proportion of 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 save 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 respective CnA, without considering the mixture as a whole, seems insufficient. Thus, a mixture-oriented approach has been developed.

TH280 Natural complex mixtures: Ecotoxic behaviour, what we know and what is next?
R. Samsara, Cehtra SAS; N. Delpeit, Laboratoires des Pyrénées et des Landes; P. Bichere, Kreatis; J. Rivera, A. Barret, C. Durou, Cehtra SAS; P. C. Thomas,
Cehtra SAS / Ecotoxicology and Environment. One of the most challenging groups was fragrances. In fragrance chemicals, natural, synthetic, member of a natural family: gums, resinoids and concretes, sub-categories of essential oils. They were challenging because they have a high number of compounds, and each compound has a characteristic odor and behavior. Additionally, the environmental fate of these substances is not well understood. The main challenges that were identified were: (1) the availability of hazard and exposure data for many compounds, (2) the need for more research on the environmental fate of these substances, and (3) the lack of standard methods for testing these 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, when 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 methods are available, the mixture 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. Cerriani, M. Fuart-Gatnik, M. Pavan, S-IN Solutioni Informatiche Srl; A. Giatti, European Chemicals Agency; E. Saouter, EU Commission JRC / Sustainable Assessment Unit
The Product and Organisational 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, USEtox requires the most up-to-date data base 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, 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 E5D0, 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, Kliminchik 1/2) were included in the first two levels. This presentation is intending to discuss these issues and to bring some elements of response based on case studies.

TH283 Deriving USEtox aquatic freshwater toxicity Effect factors from OpenFoodTox database 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. Cerriani, S-IN Solutioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAscientific Committee and Emerging Risks Unit Department of Ralisation Competence Management; J. Richerdeau, EFSA AMU; D. Meoeg, Eurofins SCAFE LLC Product Environmental Footprint (PEF) and Organisational Environmental Footprint (OEF) form 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). There are currently 25 PEF and 2 OEF pilots testing the method and developing Product Category Rules (PCRs) to be used as potential input data in the calculation of impact factor emitted during the life cycle of a product. This impact factor is assessed via the USEtox multimedia fate model [3]. This model requires ecotoxicity data to freshwater aquatic life. For PEF/LCA, those data are required for thousands of chemicals using the most up-to-date information [4,5]. The USEtox OpenFoodTox database was used to extract the information required to calculate effect factor for Plant Protection Products. EFSAscience has populated a chemical hazards database with the most up-to-date data available from the OpenTox database, which is a freely accessible database of ecotoxicological data. This presentation is intending to discuss these issues and to bring some elements of response based on case studies.

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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. Acute 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 multiple stressors from food contact materials
K. van Gils, Foundation; J. Müncke, 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 FCAs 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 FCAs, basic information on migrating chemicals must be available, such as their chemical identity. However, this is often not the case for all migrating FCAs, 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 migrating FCAs migrate simultaneously, forming the ‘overall migrate’ 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 noxiousness of stable and radioactive substances for both human health and ecosystems
K. van Gils, Foundation; J. Müncke, Food Packaging Forum Foundation; J. Muncke, Food Packaging Forum Foundation / General Management
Inspired by methods and tools developed in the field of life cycle analysis (LCA), we develop a new index of integrated toxic pressure to assess the impacts of contaminant substances and wastes for human and environment health. 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 radioxicity. For ecosystems, a comparative toxic unit has already been defined from which we derived our noxiousness index. It is based on the concept of Potentially Affected Fractions (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 PAFAF (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 calculation of a single index. According to acknowledged practices in LCIA and corresponding available data, we suggested declining this approach to human health, taking into account exposures resulting from both ingestion and inhalation pathways. This led to eight basic indexes, 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 noxiousness.

TH286 Solution-focused application of mixture modelling and chemical footprints
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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 100 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 compounds (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 integrated model. In SOLUTIONS, the modeling train is to result in complex footprint models. 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 extends the potential transfer of information from one watercourse to another, for example, in case monitoring early warning systems. 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
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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 exposure levels, profiles, and exposure pathways to phthalates for different population groups. Eighteen phthalate metabolites including mono-ethyl phthalate (MEP), mono-ethyl phthalate (MEP), mono(2-ethyl-5-oxohexyl) phthalate (MEHHP), mono(2-ethyl-5-hydroxyhexyl) phthalate (MEHP), mono(2-ethyl-5-oxo-hexyl) phthalate (MEHHP), mono(2-ethyl-5-carboxymethyl) phthalate (MCMPH), and monoo(2-ethyl-5-carboxypentyl) phthalate (MECPP) were analyzed. Among 18 phthalate metabolites, MEP, MEHP, MEHHP, MCMPH, MECPP, MBP, MEP, and MB2P were detected in almost all of the urine samples (detection rate >97%). However, MEP, MiPP, MNP, MEP, and MCPP were rarely detected in all of the urine samples (<10%). Total concentrations of phthalate metabolites ranged from 3.12 to 8300 ng/mL with a median concentration of 104 ng/mL. Five DEHP metabolites: Concentration (lowest: MEP, 63 ng/mL; MiBP, 8.4 ng/mL), MnBP (6.8 ng/mL) and MEHP (5.2 ng/mL) showed relatively higher concentrations than other phthalate metabolites. Our findings suggest 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 consumptions 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.
products can enter the aquatic environment after use if they are not completely biodegraded or degraded in situ. Detection studies on fish and aquatic invertebrates showed the presence of the major pesticide triclosan. The effects of these and other emerging substances on aquatic organisms are still poorly characterized in vitro models are problematic for knowledge-based predictions.

**TH289**

**CENTRAL ASIA POLLUTION: OBsolete TAILINGS, OBsolete PESTICIDES, OBsolete GASOLINE AND HUMAN HEALTH DISORDERS**

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We study the radioactive and toxic wasteage health impact: in areas of tailings and obsolete pesticides, and severe air-environment pollution by obsolete gasoline. They all have the same targets - immunity, genetic, endocrine system. The old radioactive tailings of former USSR military industry in Central Asia are poorly characterized. These three health danger sources: 1) poorly characterized radioactive tailings; 2) resettled seventeen families for most dander points; 3) freshwaters of obsolete gasoline areas. We made map of most danger areas and how environmental change influences on population health.

**TH292**

**Risk assessment of chemical mixtures in the Erft river basin**

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Mixture toxicity was assessed using extensive chemical monitoring data from the river Erft, its tributaries, and ten municipal waste water treatment plants (WWTPs) discharging into the Erft river basin. Toxic Unit (TU) approach was applied for mixture toxicity assessment. We observed 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-detects 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 comparing 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 macrophyte 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 as a potential candidate to become a toxic compound even at concentrations below LOD. Pesticides often generated high TU but due to the seasonal application substance patterns varied strongly between sampling dates and different locations. Highest pesticide concentrations in surface waters were measured during heavy rainfall which caused run-off from arable land. Single substance risk assessment identified mainly Triclosan, Ibuprofen and Dichlofenac as substances with a possible risk for the aquatic organisms. In water bodies strongly influenced by WWTP discharges Dichlofenac and Ibuprofen 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
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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 to be a 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 adaptions 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 WE-NEED (Water JPI- WATERWORKS2014 ERA-NET) focused 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 the real groundwaters from the two University of Aveiro’s case studies. 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 larvae (OECD 203). To raise models’ predictive power and to estimate binary mixtures was used to predict the effects of the ternary mixtures. Deviations from the Concentration Addition reference model indicated 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
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In coastal environments, chemical contaminants are not present on their own but in mixtures. A chemical monitoring survey detected more than 60 organic chemicals 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 Stensundsg, an area with multiple harbours and home to Sweden’s biggest chemical industry cluster. Dibutyl phthalate (DBP) and Sodium dodecyl sulphate 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 μM DBP and 0.32 μM DBP (DBP), respectively. The combination of structural endpoints as well as toxic microexperiments 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
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The Great Barrier Reef (GBR) is a protected ecosystem, listed as a UNESCO World Heritage site since 1981. It runs for approximately 2500km 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 sediment, 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 pesticides and other emerging contaminants and to the GBR we analysed the risk posed by the 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 greater 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 Stylphora pistillata to inorganic sunscreen exposure
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Between 16000 and 25000 tons of sunscreen are used annually in tropical countries with tourism linked to coral reefs considered to be one of fastest growing tourism sector worldwide. Sunscreens are complex mixture of UV filters, emollients, and emulsifiers, and at least 25% of the cream is washed off from the skin and enters the environment. Sunscreens can reside and cause persistence and the resistance against degradation of these compounds, sunscreen products can reside in coastal waters and potentially biocummulate in aquatic animals. Therefore sunscreens may pose a major threat for marine organisms in the shallow water near tourist beaches. Organic UV-filters have been documented to cause bleaching both in adult and larval stages of corals but only few studies have addressed the impact of inorganic sunscreens. A common component of inorganic sunscreen are titanium dioxide nanoparticles (TiO₂) which are widely used as UV-filters in the cosmetic industry due to their transparency and broad spectrum protection along with absence of skin irritation. Results from previous experiments on corals’ symbiotic algae Symbiodinium indicate that sunscreen toxicity is likely driven by the oil based emulsifiers in a sunscreen and not the titanium dioxide nanoparticles contained in it. A series of short-term (5 days) experiments was carried out to test the effects of these sunscreens on corals, by studying coral photophysiology, coral respiration, symbiont density and chlorophyll content. The expression of genes involved in thermal stress (HS70), carbon absorption (intra and extracellular carbonic anhydrase) and calcium and ATP exchange (CA-ATPase) were also analysed to characterize Stylphora pistillata transcriptomic response to sunscreen exposure. Results from this work will be compared to other studies carried out with organic sunscreens. Results from the present studies are essential to understand how the exposure to inorganic sunscreens affects reef-building corals, and they will contribute to the development of effective conservation programs and support eco-tourism.
The effect of antibiotic mixtures on the growth of Anabaena flos-aquae

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Antibiotics can be released to the environment, following use in both human and veterinary medicine. As a wide range of antibiotics active ingredients are in use, the natural environment will be likely exposed to mixtures of these compounds. The environmental risks of these mixtures, however, are not yet fully understood. In this study, the toxicity of single and mixture of six major human antibiotics from different classes; amoxicillin (AMO), oxytetracycline (OXY), clarithromycin (CLAR), 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 mg/L (CLAR, 0.001 mg/L); CEP, 0.003 mg/L; CIP, 0.008 mg/L; OXY, 0.006 mg/L; MER, 0.02 mg/L and AMO, 0.03 mg/L. Use of these toxicity data along with 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, CLAR, MER, CIP and CEP respectively, suggesting that five of the six compounds may be adversely affecting the aquatic environment. The toxicity mixtures studies are ongoing bit of the data available; these will be used to evaluate the concentration addition (CA) and independent action (IA) for estimating the toxicity mixture. The best performing model will then be used alongside exposure modelling approaches to explore the risks of mixture for different scenarios.

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 Liège / GIGA-R. Laboratory for Organogenesis and Regeneration; H. Bernts, NMBU-School of Veterinary Science / Department of Production Animal Clinical Sciences; K.E. 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 Liège / Department of Food Science, FARAH AhR transactivation activities are often persistent in the environment at concentrations of the POP mixture corresponding to 75 times the blood level, which corresponds to an interpolated antagonistic equivalent of 0.165 µM BDE-47, while only 0.0047 µM BDE-47 present in the mixture at the IC50. In addition, the isobole coefficient of the mixture is 0.3 (~1) according to additive mixture effect model. This indicates that AhR antagonistic activities are significantly enhanced in real mixtures.

The acute immobilization test resulted in a EC50 of 0.0047 mgl-1 for 1 D. magna at an EC50 of 14.7 mg/L for DnBe and 1-Octanol as single substances. The determined values with data of other biofuels (2-methylfuran, 2-methylyrathofuran) showed a higher toxicity of the mixture. DnBe showed a higher toxicity on D. magna than the tested mixture. For the comprehensive ecotoxicological assessment of the mixture and there compounds as single substances further tests are required. Especially regarding to the possible impairment of the slippage 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 work was performed as part of the Research Cluster “Tailoring biofuels from biomass” funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

Mixture toxicity of abamectin and difenoconazole to zebrafish embryos (Danio rerio)

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There are many agrochemicals used in agriculture, among them the insecticide abamectin and the fungicide difenoconazole. They are widely used compounds in strawberry crops in regions of tropical climate, although they are compounds classified as extremely toxic and very dangerous to the environment. The use of fish as test organisms stands out in ecotoxicology due to its representativeness and critical role in aquatic environments. Due to ethical issues and to reduce costs, space and waste generated, alternative methods such as assays using fish embryos are currently widely used. The FET - Fish Embryo Toxicity Test is an example of a standardized test that use Danio rerio embryos. Considering the ecological risks
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; 2.4; 5.3 and 11.7 mg L\(^{-1}\) of abamectin and 0.2; 0.5; 1.0; 2.3 and 5.0 mg L\(^{-1}\) of difenoconazole. The factorial design was used combining 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 MIXED procedure of SAS that considered the effect of treatments, the binomial nature of abamectin and difenoconazole promotes 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 have been obtained in other studies with other species of fish, documenting one 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 bioreactor-systems and zebrafish embryos

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There is an ever-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 organic chemicals 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 similar mixtures is 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 zebrafish embryos. They will be exposed to selected priority pollutants and their mixtures (e.g. polychlorinated biphenyls, heavy metals, polycyclic aromatic compounds). 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/gasification 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 through exergy and Ecoinvent 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 MWh kg\(^{-1}\) [1]. The pyrolysis/gasification 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/gasification 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 damage to resources 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 an high impact, except for land occupation. The LCA study made it possible to carry out a preliminary assessment supporting the pyrolysis/gasification pilot plant design, to identify critical aspects and strengths of each scenarios. [1] Legislative Decre 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

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REELCOOP, an EU-FFP funded project which stands for Renewable Electric COOPeration (www.reelcoop.com), aims to develop and test novel prototypes of solar photovoltaic (PV) ventilated façade (6-kW) and involves the development of c-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 prototypic 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 used in addressing the task that also requires a focus 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 (CEENE) 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


The aim of the present study is to investigate embryotoxic and teratogenic, but also mechanism-specific effects using zebrafish 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.
The scope of the present study is to investigate the environmental sustainability of different routes of terephthalic acid (TA) production, comparing the results against the equivalent batch synthesis. 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 HMF and ethylene: it involves the production of HMF (5-hydroxymethylfurfural) from starch, its conversion to DMF and the Diels–Alder reaction with bio-ethylen to obtain p-xylene; Alternative pathway from p-cymene: it consists in the oxidation, using O₃ 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 impacts of the scenarios, a simulation of the selected processes was carried out using ChemCad 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, residues and their potential use in different sectors. The most cited LCA has been published in 1999. 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.

TH310 LCA of nanomaterials production for the emerging technology: the case of printing batteries

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BASMATI is an ambitious project which main goal is to develop active nanomaterials and electrochemical inks for printing technologies to transfer and up-scale to pilots at SMEs and industry facilities. This project is co-funded by the European Union under The Framework Programme for Research and Innovation HORIZON 2020. The project is developed by 10 different partners, where LEITAT is in charge of the Life Cycling Assessment. For that task, the environmental impact has been performed during the whole life cycle considering synthesis, formulation and disposal of printed batteries. The LCA has been focused on estimating the potential impacts of Cu, LFP (LiFePO₄) and NMC (Ni-Mn-Co) nanoparticles synthesized, the inks which contain these nanoparticles and the batteries printed with these inks. The LCA applied in the study is based on the standard ISO-framework for LCA (ISO 14040:2006 and ISO 14044:2000). Calculations have been done using the software GABI, and taking as a basis the GABI Database, Ecoinvent Database and the LCA impact assessment method. The functional unit has been defined as “a printed flexible battery to be used for power source” and the scope has been based on the “cradle to grave” approach. Primary data have been priorised and secondary data from databases and literature have been used when needed. The inventory process has been completed with the information of inputs and outputs gathered from partners. Then, the impacts related to inventory flows have been calculated for ten impact categories. Results show that the impact of nanoparticles synthesis is mainly dominated by raw materials. Moreover, for the inks compared, and results it can be seen that inks with NMC nanoparticles have higher impacts in most of the impact categories. Specifically, NMC inks represent the highest weight due to raw materials used during NMC synthesis. In addition, the highest impacts in climate change and resource depletion are dominated by Cu inks. A holistic trend toward the CF system as a greener and more efficient way of ENPs production has been seen. After that, the manufacturing of the printed lithium-ion batteries has been analysed. The stack and interdigitated battery has been chosen as demonstrators to develop the LCA. Landfill and recycling have been assumed as end of life scenarios. Finally, the conclusions take into consideration the new generation of technologies and their environmental performance.
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 to develop a suitable process looking for high performance materials but environmentally friendly. Some improvements are being made such as use of aluminium 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 Eiseneser, Austria. The case of RICAS2020 PROJECT. A. Claret, Leitat Technological Center / Sustainability Division; G. Ferrer, Leitat Technological Center / Quantitat Ge en/ RD; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division; M.R. Riera, LEITAT / Sustainability Division; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division. 

Europa 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 for the electric grid, where the peak production of energy is usually not in phase with the peak demand; the developing of large scale electric storage systems. The innovative AA-CAES developed within the RICAS2020 project can solve this problem. In a CAES the air is compressed in a storage unit when electric energy overproduction is available, and by the inverse process, is reintroduced in the grid when required in the high demand periods. Additionally, AA-CAES collects the heat produced by compression in a specific Thermal Energy Storage (TES) and returns it to the air when the air is expanded to generate power, delivering higher efficiencies via a zero CO2 emissions process. RICAS2020 is being assessed under the Environmental and Social LCA, in order to define improvement measures to guarantee its sustainable performance. The scope of this LCA covers the construction and the operation stage of the AA-CAES. Regarding the construction stage are being assessed: the site excavation methods and the manufacture of materials needed for the construction of the Cavern and TES. Respect to the operation stage, the impact of machinery used (turbines, compressors, coolers) are being considered. The main goals of this assessment are to: (i) study the environmental efficiency of the new CAES generation compared to the conventional setups, 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 coating simulation using algebraic differential equations and the physics of the process as well as the material and energy balances of the unit operations involved. The Life Cycle Assessment is a methodology aiming at analysing the overall life cycle of products, processes or service. In this work, we present the analysis of the complete life cycle of polyurethane rigid foam from cradle to grave in tight connection with process simulation methods, thus obtaining the characteristic equations of the production process, such as pressure drop, energy consumption and other, with impact assessment. This combination will allow us to identify the best solution for the production of polyurethane rigid foam both in terms of end of life scenario and environmental impact.

TH314
Anticipatory life cycle assessment of sol-gel derived anti-reflective coating for greenhouse glass N. Tsao, 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 

Reflective greenhouse technologies are gaining significant interest as a means to increase crop production. The critical parameter for these systems is the reflective greenhouse coating, which is expected to have high light transmittance than the conventional coatings by 2.5%. This study, three reference coatings in addition to the novel coating were assessed. The functional unit of the coating was the mass of tomatoes obtained under a certain area of the 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 is based on the amount of energy required, the emissions of each coating, and the economic results of the greenhouse glass coated with novel coating. The results of this study show that the new coating is competitive to the conventional coatings in terms of energy saving and greenhouse glass coated with novel coating performs better than the conventional coatings by 10%.

TH313
Upgrading wastewater treatment technologies in the framework of current renewable energy policies - an environmental assessment A. Petit-Boix, University of Freiburg / Chair of Societal Transition and Circular Economy; M. Riera-Salas, Universitat Autònoma de Barcelona / Institute of Environmental Science and Technology (ICTA); V. Balbuena, Universitat Autònoma de Barcelona / Institute of Environmental Science and Technology & Department of Chemical, Biological and Environmental Engineering; E. Moliné, Depuración de Aguas del Mediterráneo; M. Suárez-Ojeda, Universitat Autònoma de Barcelona / Department of Chemical, Biological and Environmental Engineering; R. Boix, University of Freiburg / Chair of Societal Transition and Circular Economy.
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 diclofenac, to disrupted Jαtun and Wnt signaling
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Embryotoxicity 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 diclofenac, 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 populations. The intermedial step involved with bladder inflation is formation of lipid peroxidation (LPO) and impairment of antioxidant defences. Essential biological macromolecules such as DNA, protein and lipids can be damaged by ROS through oxidation and peroxidation, thus leading to diverse types of adverse effects, such as growth arrest, developmental abnormalities, reproductive failure and lethality. A wide range of oxidative stressors such as ionizing radiation, ultraviolet (UV) radiation, metals and organics are known to induce oxidative stress. The consequences of oxidative stress have been well studied in vertebrates, 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 key components in the AOPs 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 damage, DNA fragmentation, protein and lipid storage, and abnormal ovaries structure and oogenesis in D. magna, thus verifying some 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** Quantity of AOP by Bayesian network modelling: linking 3,5-DCP exposure to adverse outcomes in Lemma minor

J. Moe, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; W. G. Landis, Western Washington University / Institute of Coastal and Aquatic Research; J. Xie, NIVA - Norwegian Institute for Water Research; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment

AOPs 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 at different concentrations. 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 Event 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 the DCP concentration to assess the probability at linking 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

J. Jeong, University of Seoul; N. Chatterjee, University of Seoul / Environmental Engineering; S. Choi, University of Seoul / Environmental Engineering; J. Choi, University of Seoul / School of Environmental Engineering

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 systemic 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 bronchoscopic epithelial cell (Beas2B). Beas2B cell was exposed to CMIT/MIT (a biocide 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 (key initiating events; MIEs), intermediary 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-million 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 building 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 FPT 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 sewage 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. Fish were exposed to the wastewaters from three WWTPs located 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 tachykinins (tachykinin 3a and tachykinin 3b involved in neuroendocrine regulation of reproduction), myelin basic protein (responsible for myelination of axons and neuroprotection). The activity of acetylcholine esterase, enzyme that terminates action potential transmission in chemical synapses of cholinergic type, was also examined. A trend of slight upregulation for the expression of the genes encoding for tachykinin 3a and tachykinin 3b, GABA receptors and myelin basic protein 10 was observed for all three studied sites when compared to 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 (FPT-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 alcoholic 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 fertirrigation should not affect the soil 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 detoxification reactions that usually, occur in rhizosphere of plants, they 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 analyzes 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 (Gobiodes broussonneti - Gobiidae) from one of the most productive estuaries in Brazil
L. Salgado, Universidade Federal do Paraná / Farmacologia; A.M. Maques, UFPR / Genetics; F. Garrido de Oliveira, UFPR / Pharmacology; S.L. Moretto, M.M. Cestari, UFPR / Genetics; H. Silva de Assis, UFPR / Pharmacology

The estuarine-lagoon complex of Subaúma and Sítio do Cananéia (São Paulo state south 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 (Gobiodes broussonneti - 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. broussonneti the studied area. Fishes were sampled near Subaúma, 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 GPx and GST activities, GSH concentrations and LPO damage in liver were similar. However, it was observed genotoxic effects (micronuclei 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 region and the bioavailability of contaminants may interfere in the responses. Necropsy and autonomic 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 analyzes 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 biomarkers
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The Aachen-Soers (Germany) wastewater treatment plant (WWTP) is one of the most important wastewater treatment plants (WWTPs) in the Aachen region. Nevertheless, these chemicals can have adverse effects on the river biota. The diet of the Purple Goby (Gobius niger) (Aachen-Soers WWTP) is mainly made up of fish, crustaceans and molluscs. The violet goby is a demersal species with a social behavior, which is very common in estuaries. The Violet Goby (Gobioides broussonneti - 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. broussonneti the studied area. Fishes were sampled near Subaúma, 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 GPx and GST activities, GSH concentrations and LPO damage in liver were similar. However, it was observed genotoxic effects (micronuclei 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 region and the bioavailability of contaminants may interfere in the responses. Necropsy and autonomic 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 analyzes are being performed in the studied points in order to support a better understanding of these responses.
during late summer in 2017 to evaluate the status quo of the stream and the performance of the “Aachen Soers” WWTP. This project is funded by the Ministry for Climate Protection, Environment, Agriculture, Conservation and Consumer Protection of North Rhine-Westphalia.

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, large numbers of chemicals, in particular pesticides, have been identified with 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 spiggin-1 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 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 of the presence of a number of pesticides previously 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 RTL-W1
S. Weeks Santos, EPOC University of Bordeaux; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS S805; J. Groussin, EPOC University of Bordeaux / UMR EPOC; Q. Papin, University of Bordeaux / UMR EPOC; C. Céradeau, EPOC University of Bordeaux / EPOC UMR; B. Morin, University of Bordeaux / EPOC; B. Cormier, Université of Bordeaux / UMR EPOC; P. Gourves, University of Bordeaux / UMR EPOC CNRS S805; J. Cachot, University of Bordeaux / EPOC VIA. The vineyard uses the most pest-free farming methods. 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 (RTL-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, Parodier, Grand Village and Vignolles) and one site Reguignon from Les Souches (a small stream highly impacted by viticulture activity that rejects in La Livenne). Pollutants from 1 L water column had been extracted by SPE (Solid Phase Extraction) method and from sediments had been extracted by elutriates. In the first part of the study, RTL-W1 cells were exposed separately to extracts of water and sediment samples from the three campaigns and different toxicity tests were performed as cytotoxicity (MTT test) and ROS (Reactive Oxygen Species) induction. In the second part of the study, larvae had been exposed 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 Reguignon were cytotoxic 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 Reguignon when compared to non-exposed larvae. In particular, the head size was significantly smaller than control larval, and yokol 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 Particular Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment (PC)

MOPC07
Optimization and Automation of Raman Microspectroscopy for Microplastic Analysis
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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 considered as emerging pollutants because of their tiny size. They can be a source of adverse effects on organisms. Various spectroscopic techniques have been developed to address the issue of MP, however, they are not suitable in the analysis of complex samples, for example environmental samples. To address this need we developed a simple methodology that allows one to prepare small microparticles of PS, PVC, PE and PP in a simple and fast way. The automated Raman Microspectroscopy (RM) is a versatile tool for MP analytics.[2] Optimization and automation of RM measurements as well as automation of spectral data evaluation are of high importance for monitoring programs and enable quantitative results. We advanced RM-based analysis by optimizing measurement parameters, measurement automation and rapid and automated spectral data evaluation. First, a filter holder was constructed that flattens the filter surface. This filter holder is superior compared to filters deposited on, or glued to, glass slides. However, a flat filter surface is only a first step for successful measurements. For an automated particle recognition we also optimized the contrast between particles and filter. To this end, a variety of polymers (e.g. PE, PP, PS) of different sizes (10−3−10−6 µm) and forms (spheres and irregular shapes) were analyzed on different filter materials (e.g. PC, gold coated PC, nitrocellulose, etc.) under different modes of illumination (bright and dark field, fluorescence mode). We found that reflecting filter materials combined with dark field illumination yield superior contrast. Finally, we tested these optimized parameters with samples of different complexity (incl. environmental samples) and three RM modes: semi- and automated in regard of particle recognition and Raman measurements). Samples with a low loading are accessible via an automated approach, whereas the analysis of more loaded samples is better done with manual particle recognition.[3] The results brought forward in this work aim to catalyse advances towards automated methods for a better assessment of environmental risks arising from MP. In the future, the challenges lie in developing automated methods into these samples, especially for poorly dispersed samples. Experimental and automated approaches for the rapid and automated analysis of complex samples are needed. The authors thank the German Federal Ministry of Education and Research for funding the project MiWa and the Raman microspectroscopy alpha 300R (WITec GmbH).

MOPC08
Preparation of model small microplastics and nanoplastics
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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 nanoplastics (<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 toxicity. Here, we present a simple methodology that allows one to prepare small microparticles of PE with sizes between 0.7 µm to 3 µm. These particles were obtained by dissolving PE pellets in toluene at high temperatures, which was then solidified in water and dried by ultrasonication. 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
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 composition, degradation, and ecotoxicological 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 bases of the marine food chain, but may also provide an exposure route for toxic additive chemicals present in tires to marine organisms. Furthermore, the rubber particles may act as a vector 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 out 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, DDTs, biphenols, as well as metals were measured to establish the adsorption and leaching kinetics in seawater under in situ conditions. Samples were extracted using ultrasound 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, Trondheim. Exposure to demonstrate the detection of nanostructured MPs in environmental matrices and in particular, micronized tire rubber, making up >90% of the particles at some sites. Zero point microplastics from various sources exhibit different properties and high mortality rates were found for different marine zooplankton species.

MOPC10
Micronized tire rubber: abundance and distribution within microplastic litter of the Charleston Harbor Estuary, South Carolina, USA
R. Leads, College of Charleston / Biology; J.E. Weinstein, The Citadel / Department of Biology

Micropolastics (2acet present on shorelines worldwide. A previous survey of Charleston Harbor, South Carolina, USA reported an average of 591±103 microplastic particles/m² in intertidal sediments, with black fragments suspected to be micronized tire rubber making up >90% of the particles at some sites. The objective of the present study was to further characterize the abundance and distribution of micropolastics, and in particular, micronized tire rubber, in an effort to identify the sources and pathways into Charleston Harbor. As rivers are thought to be a contributor of non-point and point source micropolastics, 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 microlayers (n=3) samples were collected from three sites (upstream, midstream, downstream) along each of the rivers and were analyzed for micropolastics (630-500 µm). Intertidal sediment micronized rubber abundance ranged from 3-4375 particles/kg wet weight. Sea surface microlayer micronized abundance ranged from 3-36 particles/L. Micropolastic 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 micronized rubber fibers and micronized tire rubber presented the highest occurrence and accounted for 62% of all micropolastics observed, constituting 26.2% and 17.1%, respectively, of total micropolastics collected. Furthermore, micronized tire rubber was collected at every sampling location in and every environmental sample type (intertidal sediment, subtidal sediment, sea surface microlayer). These results suggest that micropolastics in Charleston Harbor originate primarily from non-point sources and that micronized tire rubber is a significant contributor of micropolastic litter in Charleston Harbor. These results are the first to report on the abundance and distribution of micronized tire rubber as micropolastic litter in a southeastern U.S. estuary and contribute to the understanding of the worldwide environmental presence of micronized tire rubber.
Minor concentrations. Venlafaxine and carbamazepine proved to be persistent pharmaceuticals, in consistency with literature.

**MOPC20**
Ciprofloxacin By-Products in Seawater Environment in the Presence and Absence of Gilt Head Bream

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The widespread use of pharmaceuticals has caused a growing concern on the presence of pharmaceuticals such as the antibiotic ciprofloxacin (CIPRO) in the aquate 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 biofluids (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 zwiterionic 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 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 glycine 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 deamination and both glycine and glutamine conjugation gained importance since 4 of the 10 new BPs had suffered both reaction transformations. Although CIPRO metabolites were searched in gill-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 deamination gained importance in bile, neither glycine nor glutamine conjugates were observed in bile BPs.

This work was financially supported by the Ministry of Economy and Competitiveness through the project CTM2014-56628-C3-1-R. H. Zurrasta 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. Montegudo, 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 anions 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 $\text{SO}_4^{2-}$ (with no generation of the very effective $\text{SO}_3^-$). Sulfate and hydroxyl radicals were involved in the main reaction pathway of diclofenac. Diclofenic amide and three hydroxy-diclofenac isomers (3'-hydroxy-diclofenac, 4'-hydroxy-diclofenac, 4'-hydroxy-diclofenic 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.

Mercury Biogeo sciences - Fate, Effects and Policy (PC)

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 production site in Italy to illustrate this approach to Hg source characterisation. 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 across the GTA in July and August 2016 for time periods ranging from 34 to 46 days. In Italy, PASs were deployed at two spatial scales (a 0.56 km² square comprising the former Abbadia San Salvatore mercury mine and a 41.6 km² square covering the eastern slope of Mt. Amiata). Both squares were divided into a grid of 7x7 cells and a sampling site was selected within each of the 49 cells. The finer spatial resolution grid was sampled twice with one-week long deployments in Oct. 2015 and Jul. 6 2016. The coarser spatial resolution grid was sampled for an entire year (Oct. 2015-Oct. 2016), in four seasonal deployments of approx. 3 months each. Mean gaseous Hg concentrations in downtown Toronto (1.77 ± 0.28 ng m⁻³) were slightly, but significantly elevated relative to other parts of the GTA (1.42 ± 0.20 ng m⁻³). Concentrations at sites close to waste/recycling (1.61 ± 0.22 ng m⁻³) and hospitals/dental facilities (1.63 ± 0.21 ng m⁻³) were significantly higher than at sites presumably distant from potential sources (1.37 ± 0.20 ng m⁻³). In the mine area in Italy concentrations reached as high as 123,500 ng m⁻³ and declined rapidly with distance from the most contaminated site. Concentrations were higher in July than in October. At both spatial scales, concentrations declined less steeply towards the East, consistent with prevailing westerly winds. Atmospheric emission from the mine was estimated to range from 50 to 100 kg annually. Clearly, the PAS’s ability to precisely and accurately discriminate small differences in gaseous Hg concentration (≤ 0.2 ng m⁻³) across a wide range of concentrations, including 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 emission sources.

MOPC24
Mercury trend as a possible result of changes in cod age distribution A. Rugg, NIVA / NIVA; D. Hjerrmann, NIVA; J. Schjønn, S. Øxnevad, NIVA; Norwegian Institute for Water Research; B. Beylich, NIVA; M. Schjønn, University of 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. The 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. The 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.
Polymer inclusion membranes followed by X-ray fluorescence analysis as a new tool for mercury monitoring in natural waters at low concentration level

Suwannee River humic acid (SRHA) and in natural water rich in DOM from Onego

The data was selected following strict criteria as regards to the endpoints, measured values were calculated on the basis of the standard test species used in the acute first tier (Daphnia magna, Oncorhynchus mykiss) and further inhibition to the photodemethylation pathway and a strong seasonal difference due to variation in concentration level

Glycine

The results showed that adsorbed and intracellular Hg concentrations decreased 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, if once a sample is 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.

Dissolved organic matter as a modifier of Mercury bioavailability to phytoplankton

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 responsible 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 thioclate (TOMATS) as extractant. PIMs were contacted with Hg in natural waters and, once the metal was collected, membranes were dried and immersed in one of the two following apparatus (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 were observed when 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, if once a sample is 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.

Dissolved organic matter as a modifier of Mercury bioavailability to phytoplankton

Mercury (Hg) is a priority toxic 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 the cell wall, of the cell wall. The mercury interaction (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 intracellular mercury concentrations by Direct mercury analyzer on freeze-dried pellets. Concentrations of Hg in the exposure media were measured with the MERX Automated Total Mercury Analytical System. Chemical speciation of Hg in the absence 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 0.1 µM 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.

Fungicides - an overlooked compound group? Fate, effects, risk assessment and mitigation (PC)

TUCP01

Overview of 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. Bandschuh, Swedish University of Agricultural Sciences / Department of Aquaculture 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) with a particular emphasis on the functional and ecosystem level. Related contributions reviewed fungicide exposure and mitigation measures. Within aquatic systems, aquatic fungi appear to be particularly at risk of adverse effects because fungicides are designed to control their terrestrial relatives during crop production. Indeed, structural and functional interactions in aquatic fungal communities have been reported in field and laboratory studies. As fungi positively (e.g., conditioning of detritus) and negatively (e.g., via parasitism) interact with other organisms, such effects have been shown to result in indirect fungicide effects on other taxonomic groups. In addition, other taxonomic groups can also be directly affected by fungicides because these substances act on biological processes that are highly conserved (e.g., energy production). Direct effects of fungicides on non-fungal 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 will 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.

TUCP02

Relative tolerance of aquatic organisms to fungicides

A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; M. Daam, New University of Lisbon

Fungicide compounds are routinely used in intensive agriculture production to treat a wide range of plant pests and microbial pathogens. These compounds may reach aquatic ecosystems by spray-drift, leaching and runoff, posing a potential threat to aquatic organisms. In this study we evaluated the sensitivity and relative tolerance of a range of aquatic organisms of importance to European aquatic systems and further act 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, if once a sample is 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.

determination of the metal can be performed. Applied toxicology and risk assessment in aquatic organisms; J. Schmid, University of Geneva / Département F.

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 responsible 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 thioclate (TOMATS) as extractant. PIMs were contacted with Hg in natural waters and, once the metal was collected, membranes were dried and immersed in one of the two following apparatus (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 were observed when 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, if once a sample is 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.

Dissolved organic matter as a modifier of Mercury bioavailability to phytoplankton

Mercury (Hg) is a priority toxic 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 the cell wall, of the cell wall. The mercury interaction (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 intracellular mercury concentrations by Direct mercury analyzer on freeze-dried pellets. Concentrations 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 0.1 µM 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.
Fungicides affect spray drift 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: Chaetopterygium 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 lowest in the highest fungicide treatment compared to the lowest fungicide treatment and the untreated control. Fungal biomass has been put forward. A densely vegetated and wide buffer strip achieves the highest reduction potential 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, row drift, or in drainage. To mitigate fungicide exposure a range of 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 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 time of fungicides and thereby the probability for adsorption and degradation processes to take place. Mitigating the fungicide exposure via spray drift and runoff may 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, we first 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

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 (genecos and SSD) and tier-3 (micro/mesocosm) 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 yeastslike fungus, Auroebasidium pullulans, and the response of evaluating authorities C. Donat, bio-ferm GmbH

In the course of inclusion of two strains of the species Auroebasidium pullulans as active substances to be used in plant protection products in Annex I of the Directive (EU) 91/414, a data package was developed to assess the ecotoxicity of these yeastslike 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 the Flying 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 Auroebasidium pullulans to Annex I without requiring any additional study. National decision for the registration of the product varied, some authorities authorized the use, others did not, whereas some nations demanded 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 microorganism 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

TUPC06

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

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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, Ctb
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 microbials, where test 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 only weakly persistent in the environment and exposure levels may be too high to achieve actual effects. 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**
Health 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 (SUD; Dir. 2009/128/EC) strengthens a targeted use of integrated control of pests and diseases where non-chemical measures are preferred. Plant protection products with a microorganisms as active substance could be a solution. Microorganisms have to be approved in Europe in order to be used as active ingredients in biopesticide products. Data requirements for pesticides based on microorganisms are available as separate part of regulation EC 283/2013 and EC 284/2013. The regulation widen the scope of the data required above current testing 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. Künzlich, A. Dabrunz, C. Lang, Eurofins Agroscience Ecotex GmbH / Aquatic Ecotoxicology
In the last decade the number of biopesticide registrations in the EU and US have steadily increased. In the EU biopesticides 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/3030/99 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 pesticides, microbial actives need to be tested under all conditions which allow growth and reproduction of the microorganism. Experimental data will be presented with focus on the applicability of microbial quantification methods considering different microorganisms and substrates.

**TUPC17**
Modelling bioaccumulation of persistent organic pollutants in Arctic food chains
R. Scholten, 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 maritimus), 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. King, Korea Polar Research Institute / Division of polar paleoenvironment; 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 correlations for the TMF analysis. After the trophic 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 Anarctic Environment by the New and Legacy POPs. The levels of most POPs were magnified through trophic levels, while Decloranes, emerging contaminants, appeared not to enough TMF values. The insufficient detection rate of Decloranes, compexity of the food web structure, and the overestimation due to migrant animals arose the uncertainties in TMFs, and therefore need to be taken into consieration to interpret the TMF results in this study.

TUPC19
Bioconcentration as the predominant mechanism for fish PCB contamination in alpine lakes.
T. Massee, Universite Savoie Mont Blanc; M. Perga, University of Lausanne / Faculty of Geosciences and Environment; N. Cottin, Universite Savoie Mont Blanc; S. Cacheria, CISA LB; C. Piot, E. Naffreuch, 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 concentrations 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 d13C) and the influence of trophic parameters using d15N 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 254±132 ng.g⁻¹ w/w and 45±28 ng·g⁻¹ w/w respectively. Individual’s PCB contaminations in both species were not tied to feeding habitats (p>0.05). Trophic position (characterized with d13C) was also not correlated with intra-species concentration 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 discrepancies in arctic char (p=0.002) and whitefish (p=10⁻¹). 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: organohalon accumulation in an avian top predator
M.E. Loseth, The Norwegian University of Science and Technology / Biology; N. Brieis, Norwegian University of Science and Technology / Biology; I. Eulaers, 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 Tromso; G. Poma, G. Malavarran, University of Antwerp / Toxicological Center; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical 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 WTE 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 low food chain potentially results in biomagnification of OHCs. The nestlings 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 congeners are reduced and the bioavailability changes, thus the biological accumulation of OHCs 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, Smela and Steigen. In total, 14 polychlorinated 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 investigated OHCs. Furthermore, 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 nestlings and between locations, suggesting that siblings may not always share prey. 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
M. Tran, F. Alliot, EPHE / UMR Metis; H. Budzinski, University of Bordeaux; M. Cheveu, EPHE / UMR METIS 7619; R. Santos, Hepia, University Applied Sciences Western Switzerland / Ecology and Engineering of Aquatic systems research group; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC.
Trophic magnification factors have been extensively assessed for persistant 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 prey. 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 organism 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.

Epigenetic and evolutionary toxicology: from mechanisms to risk assessment (PC)
WEPC01
Does pre-exposure to bisphenol A affect the susceptibility of breeding zebrafish upon re-exposure?
H. Lüttler, University of Exeter / Biosciences College of Life and Environmental Sciences; L.V. Laing, University of Exeter / Biological Sciences; R. Boreham, M. Griffiths, University of Exeter / Biosciences College of Life and Environmental Sciences; M. Trznadel. University of Exeter / Biosciences; J. Fitzgerald, University of Exeter / Biosciences College of Life and Environmental Sciences; G.C. Paull, University of Exeter; R. van Aarle, Centre for Environment Fisheries and Aquaculture Science / Biosciences College of Life and Environmental Sciences; J. Mill, University of Exeter / Exeter Medical School; E.M. Santos, University of Exeter / Biosciences College of Life and Environmental Sciences.
Bisphenol A (BPA) is a commercially 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 signalling 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/AL 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 0-72μg/ml 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 (amh) was significantly downregulated only in fish re-exposed to BPA to 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 analyse the promoter DNA methylation of amh 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 do not assess these effects over multiple generations. It is hypothesised 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 exposures to 5AC and MEHP, many changes of DNA methylation were found in Violace, 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 would be expected from 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) / Biosphere Impact Studies; E. Eeckhoudt, SCK-CEN; J. Selles, SCK-CEN / Biosphere Impact Studies; S. Gaschak, Chernobyl 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 radiation exposure setting. In particular, we focused on 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 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 toxicology: tools to understand impacts of past, present and future environmental contamination S. E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Toxicology; D. Wecker, 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 Bioindication; H. Hollett, RWTH Aachen University / Institute for Environmental Research

This research will utilize environmental reconstruction methods along with palaeoecological, palaeoarchaeological, and palaeoanthropological techniques to understand historical, current and potential 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 future use 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 contaminant exposures and the micro-evolutionary adaptations of genes that influence the response to contaminants and environmental conditions. Further, evolutionary change 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 in 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. Vera-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, competitive exclusion of species to altered genetic diversity. Furthermore, evolutionary change 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 in long-term and potentially future evolutionary responses of natural populations to environmental changes resulting from environmental stressors, including contamination and climate change.

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)

WEP09
Dangerous misconceptions - Consumers need help!
U. Klawchka, 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%) and 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%) as 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 and 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.

WEP08
The European Union Observatory for Nanomaterials (EUON): A new platform for communicating information on the safety of nanomaterials
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 refinement 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.

WEP09
Roadmap for the unknown
M. Luitwieler, M.H. Wagelmans, Bioclear earth
The main environmental themes have been addressed in the last decades. Think about acidification, 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. This 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 therefore the reasons 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. Bioclear 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 develop 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, company and RWS. The results of this workshop will also be highlighted.

WEP10
EVOEKED: enhancing the value of climate data - translating risk and uncertainty utilizing a Living Labs approach
A. M. Deo, Norwegian Geotechnical Institute / Natural Hazards; L. Van Well, M. Zetterlund, Norwegian 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 (trends, 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 for tools to engage knowledge providers, users, and translators to identify improvements to climate services through co-design, co-development and co-evaluation. The EVOEKED 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 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 national level and the level 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, EVOEKED supports the development of the field of climate services to improve our capacity to manage climate-related risks.

WEP11
Communicating monetary values of environmental impacts - case studies related to ISO DIS 14044
B. Steen, Chalmers University of Technology; K. Hallberg, AkzoNobel; P. Hanarp, Volvo Group; J. Lindberg, IVL Swedish Environmental Research Institute; E. Riese, Essity; M. Romare, IVL Swedish Environmental Research Institute; T.
Monetary valuation of environmental impacts from human activities is a way of understanding and communicating its societal significance. However, monetary values are easily accepted 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 platform working standard on monetary valuation of environmental impacts and related aspects (sensitivities 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 method and to find out which metadata that had the greatest influence on decisions made. The cases represented three choices of ways to produce energy, one between natural gas and waste and heat from waste incineration, one between different vehicle propulsion techniques, and one between different ways of sludge treatment and energy recovery. We have used the EPS 2015dx method to value emissions and resources and a national Swedish database used for cost-benefit studies. The results indicate that it is important to report the system boundaries of the impact valuation i.e. which impacts 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 impacts such as public information have considerable influence on the choices. We discuss here 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.

WEPC1D Full STEAM Ahead: Merging Science and Communications to Investigate Environmental Questions

G.K. Biedenrfraser, 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 benefit studies. The results form 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 publicized using modern communication tools such as social media, in tandem with more typical scientific means such as presentations at academic conferences from the university. The emphasis was on the mechanics of how the collaboration lead to grant 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 get visual, a picture is worth a thousand words: How to explain Emerging Contaminants using animations

N. Osipina-Alvarez, N. 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 kinds 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) and their environmental fate can have severe implications on the environment, 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 materials related to emerging contaminants in the environment. In this presentation, we will 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 teaching and learning: developing textbooks and open educational resources

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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 online open access book on Environmental Toxicology that is useful for training at BSc, MSc and higher levels. The book will be designed in a modular way, each module having a clear teaching goal/attendance level and flagged 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 about 10 environmental toxicologists and chemists from six 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 testing methods are expensive and time consuming, requiring large numbers of animals to deliver confidence in the results. Such recognized shortcomings have led to sub-optimal policy outcomes that are increasingly being acknowledged by all stakeholders. For example, the general public and regulatory agencies are being forced to overlook the potential effects of most chemicals in the environment due to a lack of data. On the other hand, the chemical industry is being frustrated by slower access to potentially lucrative markets. Beyond human welfare concerns, the welfare of test animals is also a major consideration, particularly for groups mobilized around the issue of animal rights. Alternative testing methods are being increasingly available, such as using in silico computational models and in
Thinking green and circularly about microparticles, nanomaterials and composite materials: approaches for recovery, recycling and reuse (PC)

WEPC16
SETAC Science and Risk Communication Interest Group
T. Seiler, RWTH Aachen University / Ecosystem Analysis

WEPC17
Biochar-mortar composites for construction materials
S. Ohi, T. Seo, University of Ulsan / Department of Civil and Environmental Engineering; Y. Sou, University of Ulsan / Civil and Environmental Engineering
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, Helmholtz 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, QSRAR methods can be utilized in the development of chelating ligands designed for affinity toward strategic elements. Through this, novel ligand concepts can be rapidly assessed and synthesis development of chelating ligands designed for high affinity toward strategic elements. QSAR methods can be utilized in the construction of the ligand design against given trends for strategic elements. By comparing the predicted no-effect concentration (e.g. Zn\(^{2+}\), Al\(^{3+}\)), stabilized by anions (e.g. NO\(_3^-\)) and water molecules between layers, LSDH have remarkd kinetic release in aqueous environments if compared with materials obtained without CA. The ongoing NanoBonD project is focused on the use of these sponge-like materials for soil and water remediation. We have demonstrated that these materials exhibit superb performances in removing contaminants both from fresh and from sea water. In particular we are able to remove heavy metals (Zn\(^{2+}\), Cd\(^{2+}\), Pb\(^{2+}\), Cr\(^{3+}\) and Cu\(^{2+}\)) and organic contaminants (e.g. pyrrole and benzene) and biopolymers from an ecotoxicological point of view by testing their performances towards exposure to marine and freshwater species such as algae and bivalves, hence the definition of eco-friendly materials.

WEPC20
Zn-Al layered double hydroxides: a promising eco-friendly engineered nanomaterial
R. Martins, Department of Biology, University of Aveiro / Department of Biology and CESAM; J. Figueiredo, University of Aveiro / Biology; A.M. Soares, University of Aveiro / department of Biology & CESAM; J. Tedini, University of Aveiro / Department of Material and Ceramic Engineering CICECO; S. Loureiro, Universidade de Aveiro / Biology
Layered double hydroxides (LDH), also known as anionic nanohybrids, 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\(^{2+}\), Al\(^{3+}\)), stabilized by anions (e.g. NO\(_3^-\)) and water molecules between layers. LDH have remarkable properties in aqueous environments, such as intercalation and exchange capacity, controlled release capacity, high specific surface area and stability. The safe-by-design structure and composition as well as the properties of Zn-Al LDH have led their use in several industrial and material engineering greener applications, as well as in medicine and pharmacuetics for a safe controlled release of drugs. Despite LDH have been regarded as having low toxicity to human or toxic to marine organisms, recent data on local toxicity to 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 (Helix diversicolor), crustaceans (Acartia tonsa, Artemia salina, Palaemon serratus) 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 mg/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 noted in bivalves and polychaetes. For 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.

WEPC21
Studying microfiber release from textiles towards improved clothing design
R. Johansson, Helly Hansen; S. Kubowicz, SINTEF Materials and Chemistry; I. Yousf, S. W. Haugen, Helly Hansen; A. Booth, SINTEF Ocean / Environmental Technology
Clothing manufactured from synthetic fabrics such as polyester and nylon can release hundreds of microfibers to waste water systems when washed in domestic washing machines. Fleece fabrics have been of particular focus, however, fleece clothing can be made from a wide variety of different fabric constructions, which may exhibit different microfiber release characteristics. Mechanical and chemical finishing of the yarn and fabric will influence the size and

vitro cell- or genomics-based testing strategies (Waters and Fostel 2004). For more than a decade, these alternatives have been discussed and debated in a range of high profile forums (National Research Council 2007) as offering potential answers to the various challenges facing chemical risk assessment. However, the accepted regulatory approaches to determining the risk of chemicals in environmental toxicology have remained, for the most part, unaffected. This poster explores the role of SETAC in policy learning using primary survey data collected from participants in previous SETAC forums. We will summarize the instrumental and core policy beliefs concerning alternative testing methods of respondents and assess their self-reported policy learning experiences at SETAC. We will then consider the significance of SETAC as a professional forum through which policy actors learn and adapt to emerging challenges in regulatory science.

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volume of the microfibers released. In the current study, we assess the release of microfibers 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 (140 cm x 90 cm) was prepared by overlocking the edges to prevent loss of fabric threads due to subsequent washing. The fabrics were washed in a standard synthetic clothing program (60 °C, 1 hr, 40 °C). Weights inside the washing machine assured the same mass for each material assayed 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 released fibres adhered to fabric shedding. 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 hosing 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 was 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.

WEPC22 Exploring a Potential Nanofertilizer: Effects of Silica Nanoparticles on Alfalfa (Medicago sativa)
Nano-agrochemicals 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 substance for conventional pesticides, the broadband fungicide tebuconazole was tested. Seed germination and infection tests, and a plant growth test were conducted. The Si was quantified by inductively coupled plasma –!-

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

WEPC23 Environmental Footprint for pasta production - the PEF pasta pilot
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WEPC25 Life Cycle Assessment of applying Algal Oil in salmon aquaculture: challenges for methodology and tool development
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Dutch DSM found 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 overfished. As there is 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
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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. In 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 associated with the 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 associated with the 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. 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.
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 human 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.
Degradation.

Chronic toxicity.

Ecotoxicology.

Development.

Case study.

Degradation.

Cytotoxicity.

Decision analysis.

Chronic toxicity.

Chronic toxicity.

Degradation.

Chemical sampling.

Degradation.

Development.

Chronic toxicity.

Case study.

Degradation.

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Chronic toxicity.

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Chronic toxicity.

Degradation.

Chronic toxicity.
Risk assessment.

Risk management.

Sediment.

Soil.

Sorption.

Spatial.

Sustainability.

Toxicity.

Transportation.

Sediment.

Surface water.

Water quality.

Water quality management.
Uncertainty.

Urban.

Waste water.

Water quality.

Weight of evidence.

Wetlands.
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