Final report V1.0 for the CORE Organic II funded project: Coordinating Organic Breeding Activities for Diversity - COBRA

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Final report V1.0

for the CORE Organic II funded project

“Coordinating Organic Breeding Activities for Diversity - COBRA”

Period covered: 03/2013 – 02/2016
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<tr>
<th><strong>Project acronym</strong></th>
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Projects website: www.cobra-div.eu
Background

Plant breeding is crucial in creating organic crop production systems that can better cope with interacting stresses such as pests and diseases (especially seed-borne diseases), weeds and the increasingly erratic and unpredictable variation in climate and weather. In this context, COBRA aimed to support and develop organic plant breeding and seed production with a focus on increasing the use and potential of plant material with high genetic diversity in cereals (wheat and barley) and grain legumes (pea and faba bean), through coordinating, linking and expanding existing breeding and research. Although systems based on high, within-crop diversity have shown promising results in organic systems and are now subject to intensive research, their benefits cannot be exploited currently, due to agronomic, regulatory and other hurdles. Also, it is currently unclear which plant breeding approaches, high diversity or otherwise, are most efficient to breed varieties for organic agriculture. To help these aims, COBRA’s work was arranged into a management workpackage to coordinate the work and the following five sub-programmes:

(1) To improve methods ensuring seed quality and health
Progress was made in handling individual seeds in terms of their actual and potential resistance to seed-borne disease. One of the most important problems, bunt of wheat, was advanced considerably in terms of the ‘gene for gene’ interaction between host and pathogen and in observing the performance of the, currently, most effective resistance genes. Wheat populations, grown over two generations, did not change in their response to bunt. In barley, progress was made in identifying and confirming known and novel resistances to a range of the most important seed-borne diseases. Benign sprays were confirmed as potentially useful for Ascochyta blight control in peas.

(2) To determine the potential to increase resilience, adaptability, and overall performance in organic systems by using crop diversity at various levels
Valuable progress was made in confirming, expanding and understanding the resilience of the performance of composite cross populations of wheat. A wide range of molecular markers were identified in barley which will help in selecting genotypes adapted to expected future changes in climate and weather. Progress was also made with organic trials of grain legumes. The early
development stages of composite cross populations in the field is now better understood in relation to nutrient use efficiency.

(3) To improve breeding efficiency and to develop novel breeding methods to enhance and maintain crop diversity
A range of different technologies has been improved for selecting within composite cross populations using NIR spectrometry, colour markers and molecular markers; many of these are non-destructive. Of more immediate value, new composite cross populations involving winter and spring wheat genotypes and including bunt resistant genotypes have now been made. Progress has also been made with bulk breeding of peas, although single genotypes may still be preferred for cropping applications.

(4) To identify and remove structural barriers to organic plant breeding and seed production
It was important to bring together interested individuals and groups from different European countries to cover available experience and discuss further developments. Most importantly, this was also done directly in consultation with DGSanco, and booklets on the discussions and findings were published and distributed.

(5) To improve networking and dissemination in organic plant breeding
COBRA successfully established and utilised its website, produced regular newsletters and undertook training and farm days throughout the life of the project to raise the awareness of the project and to also communicate and discuss the finds and outcomes of the work undertaken by its various partners.

Conclusion.
COBRA’s strength is its focus on coordinating, linking and expanding ongoing organic breeding activities in cereals and grain legumes across Europe, drawing together experts from previously separated areas.

Pre-project summary
Organic plant production is currently challenged by several pressure factors. Along with perennial problems such as weed control, climate change is threatening to affect crop production through increasing weather variability. Plant breeding is a crucial factor in creating organic crop production systems that can better cope with such interacting stresses and producers need crop varieties with (a) good resistance against pests and diseases, esp. seed borne diseases; (b) the ability to react to environmental, esp. climatic variability; and (c) high competitiveness against weeds. COBRA aims to support and develop organic plant breeding and seed production with a focus on increasing the use and potential of plant material with High genetic Diversity (Hi-D) in cereals (wheat and barley) and grain legumes (pea and faba bean) through coordinating, linking and expanding existing breeding and research. Although Hi-D-based systems have shown promising results in organic systems and are currently subject to intensive research, their benefits can at present not be exploited, due to agronomic, regulatory and other hurdles. Also, it is currently unclear which plant breeding approaches, Hi-D-based or else, are most efficient to breed varieties for organic agriculture. Therefore, COBRA aims (1): To improve methods ensuring seed quality and health; (2) to determine the potential to increase resilience, adaptability, and overall performance in organic systems by using crop diversity at various levels; (3) to improve breeding efficiency and to develop novel breeding methods to enhance and maintain crop diversity; (4) to identify and remove structural barriers to organic plant breeding and seed production; and (5) to improve networking and dissemination in organic plant breeding. COBRA’s strength is its focus on coordinating, linking and expanding ongoing organic breeding activities in cereals and grain legumes across Europe, drawing together experts from previously fragmented areas.
1. Main results, conclusions and fulfilment of objectives

1.1 Summary of main results and conclusions

Plant breeding is crucial in creating organic crop production systems that can better cope with interacting stresses such as pests and diseases (especially seed-borne diseases), weeds and the increasingly erratic and unpredictable variation in climate and weather. In this context, COBRA aimed to support and develop organic plant breeding and seed production with a focus on increasing the use and potential of plant material with high genetic diversity in cereals (wheat and barley) and grain legumes (pea and faba bean), through coordinating, linking and expanding existing breeding and research. Although systems based on high, within-crop diversity have shown promising results in organic systems and are now subject to intensive research, their benefits cannot be exploited currently, due to agronomic, regulatory and other hurdles. Also, it is currently unclear which plant breeding approaches, high diversity or otherwise, are most efficient to breed varieties for organic agriculture. Therefore, COBRA aimed (1): To improve methods ensuring seed quality and health; (2) to determine the potential to increase resilience, adaptability, and overall performance in organic systems by using crop diversity at various levels; (3) to improve breeding efficiency and to develop novel breeding methods to enhance and maintain crop diversity; (4) to identify and remove structural barriers to organic plant breeding and seed production; and (5) to improve networking and dissemination in organic plant breeding. COBRA’s strength is its focus on coordinating, linking and expanding ongoing organic breeding activities in cereals and grain legumes across Europe, drawing together experts from previously separated areas.

(1) To improve methods ensuring seed quality and health

Progress was made in determining the vitality of single barley seeds through application of a new method for measuring oxygen variation. The related software application produced values that help to indicate samples with potential high germination.

For wheat and triticale, multispectral imaging and Single Kernel Near-Infrared Spectroscopy (SKNIR) proved valuable for distinguishing uninfected and infected parts of the surface of a seed and to distinguish between different varieties.

In wheat, considerable progress was made in unravelling the range of bunt resistance genes in the host and of virulence genes in the pathogen together with their variation in frequency in time and place. In field resistance testing, two winter wheat varieties, SW Magnific and Inna, did not have any infection in either of two test years.

A large scale analysis using SNP markers on mapping populations involving parents with different resistance genes revealed the presence of major gene resistances to dwarf and common bunt, modified by QTLs. Partners in different countries were able to show, however, that only Bt11 and Bt12 provided resistance to dwarf and common bunt at all locations. Several resistance genes including these two were effective in tests of a range of winter wheat varieties at the Bavarian Research Centre for Agriculture.

More than 1,000 landraces and cultivars of barley were tested at a range of European centres for resistance to seed-borne diseases, including net blotch, leaf stripe and loose smut. Many resistant lines were noted, including some with previously unknown resistance to leaf stripe. Tests were also made for nutrient use efficiency, exposing the positive relation between N and P use efficiency, probably related to artificial breeding activities. In tests in Estonia and Latvia, a number of potentially useful breeding lines were identified, including some with resistance to loose smut. Tests with RILS that carry markers for loose smut resistance revealed that some were susceptible to smut indicating that current markers for resistance are not wholly reliable.

In a separate Task, observations were made on a series of composite cross populations developed from different single crosses of wheat varieties. Although some carried bunt resistance genes, there was no evidence of adaptation of the populations to the disease, which may indicate a more rapid response of the pathogen to changes in the host, rather than the reverse.
Clove oil, tea-tree oil and thyme oil were tested in the field for their ability to control the Ascochyta blight fungal complex in peas. Though seed treatment was found to be damaging, foliar sprays appeared to be as effective as copper sulphate sprays, indicating that they could be a useful alternative to help reduce copper applications in the field. Extracts of clove and thyme oil applied to pea, field bean and lupin in vitro were found to be effective against other important seed-borne pathogens, which was not the case for oils from tea-tree and common juniper. The activities that were found now need to be trialled in the field.

(2) To determine the potential to increase resilience, adaptability, and overall performance in organic systems by using crop diversity at various levels

Coping with climate change
More than 12 partners were involved in field trials with a wheat Composite Cross Population across Europe. The CCPs performed exceptionally well with respect to an emerging epidemic of yellow rust across Europe at all test sites: the disease data showed the superiority of genetic diversity for coping with unexpected stress. The same was true for the CCP performance during a severe drought in Germany compared to pure wheat genotypes. A total of 16 CCPs that were either cycled among partners since 2008 or that had stayed in Germany, Hungary or the UK were compared for two years in Germany (KU). In addition, the German population was grown on six participating farms for two or three years and then included in the comparison trial in the second year. Evolutionary changes occurred for morphological traits such as stem length, ear length and degree of ear awnedness, but not for grain yield, disease incidence or molecular markers. The results suggest that selection for morphological changes can occur within only two to three years, while agronomic traits such as grain yield and disease resistance are evidently not affected. Baking quality parameters (sedimentation, falling number, etc.) of the CCPs often suggested lower potential performance than pure stands, however, in practice, baking volume usually did not differ from the high baking quality of reference varieties. The results confirm the plasticity of the CCP populations and their ability to adapt and change within short time periods of only two to three years under selection.

Some 200 accessions of landraces and varieties of, mostly, barley were grown in a phytotron through their whole life-cycles under a range of expected future CO2, O3 and temperature conditions and their combinations. Relatively dense association mapping using SNPs identified characters associated with such conditions which could be used for future MAS in breeding. Many of these lines were also tested for resistance to five major pathogens in the field and for performance under high CO2 (FACE) under different forms of cultivation. Some old and some new lines performed well under these conditions; most of these carried the SNP markers identified in the phytotron study. Some cultivars were also subjected to LCA assessments under different possible future cultivation scenarios to determine their eco-efficiency. The impacts were variable, but generally negative. The form and outcomes from most of these studies have already been (or are about to be) published.

A range of grain legume species and varieties was tested in spring and winter trials in five different countries under organic conditions. Performance of all was highly variable although faba beans appeared to be the most reliable. Promising advanced lines of faba bean, soybean and cowpea from these trials are already going forward for registration in Turkey.

Coping with weeds and intercropping
It has been thought that one of the factors associated with the resilient performance of wheat populations was effective competition with weeds relative to the parents or other monoculture varieties. However, the project studies showed that although root and seedling development in the populations appeared to improve over generations, at least under organic conditions, such development was associated more with nutrient use efficiency and competition for light than with direct weed competition since no allelopathic interactions were detected. Early root and seedling
vigour was more strongly expressed in the quality population than in the yield populations, further indicating the importance of NUE in relation to the expression of high quality. Despite the lack of overall allelopathic interactions, it was possible to find individual genotypes in the populations with strong allelopathic activity, indicating a potential for selection for this characteristic.

In a more detailed study, it appears that the improved root development in the populations grown organically was probably due to greater density and length of root hairs.

Other trials on intercropping grain legumes with cereals indicated that, both for winter and spring types, intercropping provided more stable yields although it was difficult to predict the proportions of the components, which was environmentally sensitive. Mixtures of grain legumes also showed useful interactions in terms of weed suppression.

(3) To improve breeding efficiency and to develop novel breeding methods to enhance and maintain crop diversity

Objectives were to identify methods to improve the constitution and performance of high diversity crops particularly in terms of adaptation and productivity under the conditions of organic farming, to create tools to make genetic resources better available for breeding and to further develop breeding methodologies and techniques adapted to participatory plant breeding.

One approach to improving breeding methodology was to apply NIR spectrometry as a form of mass selection to select for single grains with greater protein content. Although successful in itself, a familiar side-effect was a correlated reduction in yield.

Another approach was to use a colour marker as a simple method for identifying new recombinants in the population either from spontaneous outcrossing or from the introduction of a new variety. Seeds containing the blue aleurone layer can now be sorted artificially in experimental designs that allow identification of F1 hybrids.

There will be an increasing need to select crops and varieties adapted to future combinations of higher temperature with higher CO2 and ozone. A genome-wide association study (GWAS) on 167 diverse spring barley genotypes identified markers for increased primary production under artificially elevated levels of temperature, CO2 and ozone. The study identified markers and chromosome regions to be targeted in breeding for development of climate resilient cultivars. In pea breeding studies, bulk breeding produced higher yielding lines and evolutionary mixtures were higher yielding than lines derived from bulks or simple mixtures. However, pure lines or mixtures were preferred in practice.

Though farmers and breeders selected for similar characters in breeding material, it was found that farmer-selected lines were higher yielding than those selected by breeders, suggesting that experienced farmers should be more involved in the breeding process.

High throughput genotyping-by-sequencing provided many polymorphic SNP markers in three crosses among pea varieties with high accuracy of genomic selection, indicating the potential value of this approach for organic breeding.

For wheat, non-destructive phenotyping methods were compared for assessment of early vigour, biomass, LAI and leaf chlorophyll content. These involved spectral measurements and digital analysis of thermal and colour images. The latter proved to be very accurate for determination of ground cover and thus indirectly of early vigour, LAI and biomass. Colour analysis appeared suitable for detection of long-term effects of biotic and abiotic stresses, while thermal imaging may be used for direct detection of drought stress.

Within wheat, a large participatory breeding programme has been carried out and new approaches for the management of on-farm experiments have been further developed, comprising improved experimental designs, the development of a bayesian model designed to analyze the unbalanced data obtained in PPB experiments and the establishment of a database system for the management of these data. Importantly, several new bulk populations of bred wheat, adapted to central European conditions, were created in 2012-2014 based on commercial varieties adapted to Germany and surrounding countries. The parents comprised nine winter and seven spring wheat genotypes carrying xenia markers as well as germplasm resistant to common and dwarf bunt.
effectively creates a sound base for further research and practical application of participatory breeding and dynamic management of populations.

(4) To identify and remove structural barriers to organic plant breeding and seed production

The overall objective was to lower the socio-economic and legal hurdles to organic seed production plant breeding. It was achieved through two tasks, linking Europe-wide inventories with national-level case studies.

In the first task, financing models and case studies were presented at a socio-economic workshop in June 2015 in Freisingen, Germany, and in a booklet, “Breeding for diversity – political implications and new pathways for the future”. Key areas included were:

- Progress in breeding for organics: impacts of the seed derogation system
- Implementation of populations (CCPs) in the Netherlands
- The situation of composite cross populations in Denmark
- The challenge of how to finance plant breeding for organic farming
- Breeding pure line varieties of spring wheat for organic agriculture in the Netherlands
- Breeding for improved weed competitiveness in spring barley
- Conventional vs. organic soybean production in northeast Italy
- Status quo analysis of seed production and breeding of organic wheat in Slovenia

Partners also contributed to an inventory of alternative breeding initiatives.

The work resulted in an increased focus on specific potentials and barriers for different organic breeding initiatives.

In the second task, a number of meetings involving several partners in different countries discussed present needs for the regulatory framework. These included workshops with DG Sanco. Related to this, several partners are involved in the Commission Decision on the “Call for participation for a temporary experiment providing for certain derogations for the marketing of populations of the plant species wheat, barley, oats and maize.”

A questionnaire for breeders and seed companies was distributed within the partners network in September 2013, followed by presentation of the results at a legislative workshop in October 2013 in Brussels. An Italian country report was published in 2015.

Contributions from this Task are presented in the booklet: “Breeding for diversity – political implications and new pathways for the future”, which includes:

- Plant genetic diversity, farmer’s rights and the European seed legislation
- Seed marketing in Europe: an opening for diversity?
- Organic seeds and plant breeding from the perspective of the seed companies.

(5) To improve networking and dissemination in organic plant breeding

The COBRA website (www.cobra-div.eu) was successfully launched and maintained throughout the project which was the home for information about and reports on outputs of the project. Two stakeholder workshops were undertaken to encourage interactions between researchers and end users and a successful international conference was organised jointly with the FP7 SOLIBAM project. The other key tool for networking used in COBRA was the newsletter. Six newsletters have been done during the project lifespan. The newsletter reached around 90 people that included all project partners, people from other projects on organic breeding, individual interested people that could subscribe the newsletter for free from the COBRA webpage.

Formal and informal training opportunities were publicised and undertaken. Four partners gave input on formal training opportunities at their sites. A total of 25 field days during the whole project have been communicated through the website and even more have been probably organized by partners.

Overall Conclusion
Facing an ambitious programme over a short time scale, the project summary indicates considerable progress (much of which is now published or in the process of being published), both in theoretical and practical developments, including breeding material that is of immediate practical use, and, of course, the expected European capacity-building which is essential for further and more rapid progress in all areas.

1.2 Fulfilment of objectives
To what extent did the project achieve its objectives?

COBRA is an extremely broad project and, with such a large number of partners, the objectives stated were by necessity equally broad and difficult to quantify. The objectives are;

- To improve methods to ensure seed quality and health;
- To determine the potential to increase resilience, adaptability, and overall performance in organic systems by using crop diversity at various levels;
- To improve breeding efficiency and to develop novel breeding methods to enhance and maintain crop diversity;
- To identify and remove structural barriers to organic plant breeding and seed production; and
- To improve networking and dissemination in organic plant breeding.

At the end of the project it can be argued that COBRA has been successful in meeting all of its objectives. The work on improve methods to ensure seed health, use of crop diversity in breeding and methods to enhance and maintain crop diversity have improved our knowledge considerably. The work on barriers has elucidated useful insight that will feed inform both the industry and policy makers. The improvement in networking and dissemination in organic plant breeding will be a lasting legacy of COBRA. In part just bringing the consortium together achieved this but the institutional and individual contacts made within the project will allow increased formal and informal cooperation in the years to come.

2. Milestones and deliverables status

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<sup>2</sup> Please use the numbering convention <WP number>.<number of milestone/deliverable within that WP>. For example, deliverable 4.2 would be the second deliverable from work package 4.

<sup>3</sup> Measured in months from the project start date (month 1).
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<td>PU XVIII Biennial International Workshop on the Smuts and Bunts Tune/Copenhagen 3rd-5th February 2014.</td>
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<td>18 (accepted with revisions required, see 3.1.3)</td>
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<td>D1.7</td>
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<td>Prototype</td>
<td>PU Well on its way, to be completed in September 2016</td>
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<td>42 post project</td>
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4 Please indicate the nature of the deliverable. For example Report, Paper, Book, Protocol, Prototype, Website, Database, Demonstrator, Meeting, Workshop...

5 Please indicate the dissemination level using one of the following codes: PU = Public; INT= Internal (Restricted to other project participants); RE = Restricted to a group specified by the consortium; CO = Confidential, only for members of the consortium.
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<td>Protocol</td>
<td>PU The protocol was completed in 2013 and will be published in this paper: Bertholdsson, N.-O.; Weedon, O.; Brumlop, S. and Finckh, M.R. 2016: Evolutionary changes of weed competitive traits in winter wheat composite cross populations in organic and conventional farming systems. European J. of Agronomy (accepted with revision)</td>
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* Depending on legislation at the time of delivery. **Workshops will be quasi-public, but participation will be limited by available places; key stakeholders will have priority; *** prelim. date; this will be coordinated with international meetings planned for SOLIBAM, ECO-PB, and the EUCARPIA organic section

**Additional comments (in case of major changes or deviation from the original list)**

2.7: Root hair measurements on plant material produced at Tingvoll in spring 2016 will be studied in the first part of 2016. A scientific paper will be written between Anne-Kristin Løes (Norsok) together with Nils-Ove Bertholdsson et al. RCH has accepted that we postponed this work due to sick leave in 2015, and it will be completed during 2016.
3. Work package description and results:

<table>
<thead>
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<th>WP 0</th>
<th>Management</th>
</tr>
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<tbody>
<tr>
<td>Responsible partner: 1. ORC, Bruce Pearce</td>
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**Original description of work:**
The project organisational structure consists of a Coordinator and a Deputy coordinator, Workpackage Leaders and Task Leaders, a General Assembly and an advisory external Stakeholder Group. The **Coordinator** (Dr Thomas Döring, ORC) will be responsible for project management and administration, and will be supported in these tasks by a coordination team of three researchers based at ORC and the deputy coordinator (Dr Sally Howlett, ORC). Responsibilities of the coordinator include: (a) establishing a Consortium Agreement with all COBRA partners where all Tasks, Milestones and Deliverables are agreed, (b) ensuring that efficient collaboration takes place within the project team and that all participants comply with their contractual obligations, (c) acting as the contact point between the COBRA consortium and Core-Organic II, (d) facilitating within-project communications, i.e. through internal project website, project meetings (M3, 16, 33), and exchange visits among the partners and the coordinator, and (e) organising the report writing (project implementation, interim report, final report).

The **WP Leaders (WPL)** report to the Coordinator and are responsible for all Tasks in their respective WPs. Within a WP each Task is led by a Task Leader who reports to the WPL. The WPL (a) guarantee that all Milestones and Deliverables are provided on time, and (b), building on input from the Task Leaders, prepare scientific WP reports for delivery to the coordinator. The **Executive Board (EB)**, consists of the coordinator, the deputy coordinator and the WP Leaders. It monitors the effective and efficient implementation of COBRA, arranges the meetings and sets the agenda for project meetings. The EB meets every 6 months. The **General Assembly** is the democratic decision making body of the project and consists of one Representative per partner organization. The Representatives are responsible to their national funders for implementation and delivery of the project. All project participants, under the supervision of the Task Leaders, will contribute to (a) the specific Tasks within each Workpackage (b) scientific reporting on their activities to the WP leaders, (c) scientific and other output and (d) project meetings. The **Stakeholder group (SG)**, consisting of breeders, scientists, advisors and farmers from Europe with experience and expertise in organic plant breeding, will advise the Executive board on the general direction of the project, alert them to new developments in the field that may impact on project outcomes and support the dissemination of project findings. The SG (established in M2), will be organised by the Coordinator with the help of national feedback from all COBRA partners.

To facilitate coordination and cooperation among partners, regular bilateral phone/electronic meetings will be held around each of three focus crops (wheat, barley, grain legumes), with **Focus Crop Coordinators** from the ORC coordination team contacting partners for individual updates and collating highlight information for all project participants. This additional, more informal structure will help to flag up problems early and to facilitate exchange of essential project relevant information. The focus crop coordinators report to the Coordinator.

**Report on results obtained and changes to the original plan/WP aims:**
(max 1 page, Arial, size 11)

A- results obtained:

**Management meetings**
Executive committee meetings were held on a regular basis (12/14, 06/15 and a final meeting at the final congress held between 24-26/11/15 in Denmark). The final General Assembly was held during the Danish meeting. At this meeting we discussed project close down, the final report and publications.

The role of crop co-ordinators continued but at the latter stages of the project was not particularly...
successful or needed. The role of WP leaders was such that although a challenge coordination and communication across the project was good.

**Internal website**
The internal website continued to be used throughout the project as a management and communication tool.

**CORE Organic webpage update**
The COBRA webpage continued to be updated where appropriate.

**Annual report on project progress**
An internal update-report monitoring the progress within COBRA was produced by WP coordinators in autumn 2015. This was more informal that previous updates as it was an iteration of the mid-term report and close to this final report.

**Conference participation**
COBRA co-organised the COBRA – EUCARPIA – ECO-BP Meeting in June 2015 in Munich, Germany where the work of COBRA was presented.

Robbie Girling attended the CORE Research Seminar on behalf of COBRA in October 2014.

**Change of management**
The project co-ordinator Dr Robbie Girling left his position at the ORC at the end of March 2015 to take up a position at Reading University, UK. Therefore, the coordination of the project has been passed back to the Deputy Director of the Organic Research Centre, Dr Bruce Pearce who had held the reigns of the project between Dr Thomas Döring leaving ORC and Dr Girling starting. This transition between coordinators has been conducted in discussions with CORE II and ORC’s national funders (Defra), who have been extremely supportive.

ORC moved its crops team from the Wakelyns Agroforestry (WAF) site to its main office location in October 2015. This resulted in the researchers working at WAF deciding to leave ORC rather than move. The impacts on COBRA have been minimal as most of ORCs work had been completed by this time and the late autumn/winter period that was the end of the project has been mainly management by Dr Pearce and report writing.

**B- comments on deviations from the original plan:**

As reported in the mid-term report the stakeholder group was to be a flexible and arranged by the local organiser of the COBRA meetings. The final meeting was in Denmark with open both closed and open sessions. Due to time constraints no formal stakeholder group meeting was held but the open forum of the final day of the meeting allowed stakeholders to engage directly with the project via questions to platform speakers as well as posters.

**WP 1  Seed health and quality**

**Responsible partner:** Agrologica, Anders Borgen,

**Original description of work:**
Objectives are to study and coordinate methods of controlling seed borne diseases (SBD) and seed quality control by (1) improving diagnostic tools (2) coordinating and improving SBD resistance screening methods; (3) improving SBD resistance breeding methods in Hi-D populations; and (4) developing a novel method for direct control of SBD.

**Task 1.1 Rapid detection of germination capacity, vitality and SBD** [Leader: J. Jørgensen, AU, DK], COBRA will develop methods to determine seed vitality and health in single seeds. Recent studies
show that a single seed near infra-red spectroscopy (NIRS) in combination with an Extended Canonical Variates Analysis (ECVA) are potential methods for accurate prediction of seed viability. Thereby it is possible to predict germination potential before germination actually occurs. During the last decade novel non-destructive technologies have also been developed for seed quality testing including detection of seed infection in maize and wheat. A new approach using a multispectral vision system for identifying surface properties of different fungal infections has been developed and successfully tested for *Fusarium* in barley kernels and for *Alternaria, Fusarium* and other diseases in spinach seeds. COBRA will develop and validate (1) NIRS spectra for seed vitality in cereals (wheat and barley) and grain legumes (pea and faba bean) and (2) novel rapid non-destructive ways of assessing SBD, such as *Fusarium, Microdochium, Stagonospora* and *Pyrenophora*. With advanced multispectral light these can be detected even when not visible to the naked eye. COBRA will exploit and validate these novel methods and compare their accuracy and speed properties to those of established approaches. This will include comparisons with the single seed sorter IQ1002S (TriQ and IQ technology, BoMill). Integration with other COBRA WPs will be achieved by testing material from Tasks 1.2, 1.4, 2.1, 2.3 and 3.1.

**Task 1.2 Coordination of resistance screening for seed borne diseases [H. Buerstmayr, BOKU, AT].** This Task will exploit, link and expand existing screening and breeding programmes of the partners in the consortium to increase the efficiency of breeding for resistance to SBD. It will focus on common and dwarf bunt in wheat, as well as *Fusarium, Ustilago*, and *Pyrenophora graminea* in barley. (1) COBRA will coordinate resistance breeding activities by holding 2 stakeholder workshops (M6 & M30) and by creating a European database on SBD in wheat, barley and grain legumes (see Table 3). (2) We will expand pathogen collections to increase the range of races of common and dwarf bunt available and assess the stability and range of the different resistances in wheat varieties; this builds on ongoing work at Agrologica (DK) with development of collection of common bunt races. The lines from various breeding institutions across Europe will be cross-inoculated with spore originating from other varieties. Based on the reaction from different spore origin, the type of resistance in the different varieties can be characterised. (3) We will further develop genetic markers for resistance to common bunt and dwarf bunt, evaluate genotypes and develop SMART breeding tools for efficient and durable resistance to dwarf and common bunt in winter wheat, complementing ongoing work in BioBreed (see Table 3). Using closely linked DNA markers identified in COBRA, marker assisted breeding will become feasible to introgress the highly relevant trait dwarf bunt resistance into high quality and agronomically well adapted cultivars in relatively short time and at comparatively low cost. (4) We will coordinate current activities in resistance screening to SBD in pea and faba bean. (5) Finally, COBRA will assess a range of landraces and cultivars of barley for susceptibility to SBD and evaluate the environmental plasticity of the accessions in relation to infections under climate extremes. Association mapping of resistance genes and production characters will be performed in natural and manipulated environments. Suitable material will be identified in collaboration with Nordic barley breeders. The work will be closely linked to WP2. COBRA will broaden the existing activities in the Danish climate phytotron and in a Free Air Carbon Enrichment facility by screening for 3 new diseases with high relevance in organic barley (*Fusarium, Ustilago, Drechslera graminea*) in the disease nurseries. The core collection of 140 accessions is supplemented with accessions from outside Scandinavia (e.g. from Southern Europe, Latvia).

**Task 1.3 Improving bunt resistance in CCPs [A. Borgen, Agrologica, DK].** This Task will focus on common bunt in *Hi-D* CCPs to develop methods to improve resistance in populations with a minimal loss of intra-varietal genetic diversity. Specifically, COBRA will (1) determine how effectively existing wheat CCPs can be improved in terms of their bunt resistance by subjecting them to high bunt pressure as a selection force; 3 CCPs with different parental backgrounds and histories will be grown in the field with and without bunt infection at Agrologica (DK) for two consecutive years. In order to determine effects of resistance in the field, these 6 populations will then be grown in plot trials under natural bunt pressure in yr 3 on 4 organic sites across Europe (UK 2 sites, DK 1 site, F 1 site). In addition, Agrologica will grow seed from 210 crosses (F3) between 30 parents separately with bunt pressure for 2 yrs to assess bunt infection levels within each cross-population. A bulked population between the crosses will be grown with and without bunt infection. In yr 3, the separate crosses will be bulked, and the resulting population will be
grown to compare the infection level in the different populations. Further, COBRA will (2) compare breeding methods in CCPs (selection pre-composing or post-composing) for ensuring high bunt resistance in a CCP while maintaining diversity and agronomic performance in field trials over 3 yrs. Field trials at Agrologica will be sown already in 2012 on own expense to allow for 3 growing seasons within the project period.

**Task 1.4 Control of seed-borne diseases with essential oils [L. Pecetti, CRA-FLC, IT]**. Access to seed treatment is very limited in organic production, and no control measures are currently available for anthracnose of pea. Preliminary work has indicated antifungal activity in several essential oils. This Task will develop and field-test formulations of these oils for seed treatments on peas and faba beans. A major seed borne disease in pea is anthracnose, caused by *Peyronellaea pinodes* (formerly *Mycosphaerella pinodes*), *P. pinodella* (formerly *Ascochyta pinodella*) and *A. pisi*. Preliminary work carried out within the Italian National Organic Seed Plan is testing potentially useful essential oils from several species (*Thymus vulgaris*; *Melaleuca alternifolia*; etc.) at different concentrations for antifungal activity and seed toxicity on different pea varieties. COBRA will complement and expand this innovative programme by testing the two most promising oils at each of two concentrations and an untreated control under field conditions on pea seed of two varieties in two climatically contrasting locations of Italy in one test year; using seed inoculated simultaneously with all 3 pathogen species to ensure the presence of the disease and the wide applicability of the results. In year 2 and year 3 oil-treated and non-treated pea seed of two varieties (non-inoculated) will be tested at 4 locations across Europe (DK: KPB, UK: ORC, BE: Inagro and Hogent). Infection levels of selected samples from the experiments in Italy will also be tested by the multispectral vision system in Task 1.1. Finally, essential oils will be tested at 2 concentrations for antifungal activity and seed toxicity on two faba bean varieties (for anthracnose control) in the field in LUX (CRP-GL) and BE (Inagro and Hogent).

**A- results obtained:**

**Task 1.1- Rapid detection of germination capacity, vitality and SBD [Leader: J. Jørgensen, AU, DK].**

**Sub-Task 1.1.1 development of methods to predict seed vitality in single seeds.** The respiration of barley seeds during germination was investigated with a new oxygen sensitive technology (Q2). The Q2 technology was able to detect the variation between the different samples to some degree. The Q2 software calculated a set of values which are related to certain steps in the germination process. About half of these values provide useful information of the germination capacity of barley samples which can be beneficial for the prediction of high vitality seed lots with rapid germinating prior to seeding or use as malting barley.

**Sub-Task 1.1.2 Assessment of seed borne diseases by multispectral light reflection.** The use of multispectral imaging and Single Kernel Near-Infrared Spectroscopy (SKNIR) for determination of seed health and variety separation of wheat and triticale as a diagnostic method for seed health testing and variety identification have been studied. *Fusarium* sp. and black point disease-infected parts of the seed surface could successfully be distinguished from uninfected parts with use of a multispectral imaging device. The study produced a successful distinguishing between the infected and uninfected parts of the seed surface. Furthermore, the study showed the ability to distinguish between varieties.

**Task 1.2 - Coordination of resistance screening for seed borne diseases [H. Buerstmayr, BOKU, AT]**

**Sub-Task 1.2.1 Resistance to bunt (Tilletia spp) in wheat.** Studies at Agrologica shows that it is possible to purify bunt spores by maintaining them on selected wheat varieties. Field studies demonstrate that virulence is present against several bunt resistance genes except Bt3, Bt4, Bt5, Bt6, Bt8, Bt9, Bt11, Bt12. It was possible to group varieties with unknown resistances according to the uniform reaction against different bunt virulence races. New genes different from the known Bt-genes have been postulated, improving the possibility to piramid resistance genes in wheat. In
Sweden, Lantmännen found no virulence to Bt11, Bt12 and BtP in 2014 and in 2015 no virulence to Bt5, Bt8, Bt11, Bt13 and BtP.

A partly developed differential set of near isogenic lines of wheat from NordGen with different resistance genes to common bunt has been multiplied and tested by BOKU, Agrologica and SLU, and a program has been started to fill in missing resistances in the set. Work will continue in subsequent projects.

Lantmännen, Agrologica and BOKU have incorporated with other stakeholders performed a ring trial where 69 wheat varieties and breeding lines has been test for bunt resistance in 10 locations in Europe, Turkey and USA to further study the virulence pattern of European bunt races.

Estimation of resistance against *Tilletia caries* were set up at the State Stende Cereal Breeding Institute for 113 winter wheat varieties in 2013 and 60 genotypes in 2014. In 2014 spring only 46 winter wheat genotypes from 113 passed the winter. Good winter hardiness had 16 genotypes (7-9 points). Infection level of *T. caries* for tested varieties was 0-60.9%. Winter wheat varieties ‘SW Magnific’ and ‘Inna’ did not have any *Tilletia tritici* infected plants during two testing years.

**Subtask 1.2.2: Genetic markers for resistance to common bunt and dwarf bunt.** Three mapping populations of 100 to 120 recombinant inbred lines (RIL) were developed from crosses between bunt resistant exotic material. These RIL populations along with parents and checks were tested for common and dwarf bunt resistance in field trials and genotyped with SNP markers. All parental lines and the whole set of 14 bunt differential lines (*Bt1-Bt13, BtP*) were tested with COBRA partners in Czech Republic, Germany and Sweden for dwarf bunt and common bunt resistance. The testing showed that solely Bt11 and Bt12 confer stable resistance to dwarf bunt and common bunt across diverse locations. Segregation pattern pointed towards the presence of major QTL in each of these populations and QTL analyses indicated the presence of major bunt resistance genes conferring bunt resistance in collaboration with minor effect QTL.

Also at The Bavarian Research Center for Agriculture (LfL), studies were made on dwarf bunt and common bunt. In 2013/14, 40 winter wheat varieties and breeding lines were tested for their susceptibility to dwarf bunt infection. The local dwarf bunt race showed virulence against Bt7 and Bt2, whereas no virulence was observed against resistance genes Bt12, Bt10, Bt13, Bt6, Bt11, and Bt1. This specific behaviour of virulence is best attributed to dwarf bunt race D-10.

**Subtask 1.2.3: Assessment of landraces and cultivars of barley for susceptibility to seed borne diseases and evaluate the environmental plasticity of the accessions in relation to infections under climate extremes.** DTU/KU Denmark have in close cooperation with WP2 assessed natural occurring diseases on 11 selected cultivars of barley grown in a Free Air Carbon Enrichment facility (FACE) in three years. The cultivars were grown in field plots at either ambient CO2 level (ca. 400 ppm) or in an enriched CO2 atmosphere (ca. 550 ppm), mimicking future climate conditions. Increased CO2 had a protective effect on biotrophic diseases like powdery mildew whereas no significant effect was observed for any leaf spotting diseases.

At LUKE, Finland, 985 barley cultivars and landraces with different geographical origin were screened for net blotch resistance in greenhouse. Frequency of resistant genotypes against net type net blotch was highest among European barley cultivars and Syrian landraces. Of the Nordic landraces only a Swedish landrace NGB15162 showed good net blotch resistance. Also resistance to leaf stripe and loose smut was tested in 125 barley genotypes (38 European landraces). The most resistant genotype against leaf stripe was a Finnish landrace Ylenjoki AP0301 (NGB4413A). Of the 125 barley genotypes tested, 13 resistant landraces and 11 resistant and four susceptible cultivars were characterized further using published leaf stripe resistance molecular markers for Rdg1a gene (Vada resistance). Four resistant cultivars and four resistant landraces (NGB13021, NGB16881, NGB9315 and NGB9410) showed to have leaf stripe resistance which genetic background differed from Vada resistance. Moderate loose smut resistance was found among the tested barley landraces. Landrace Ljubljana KVL 15 showed best resistance for loose smut. Also nutrient use efficacy was tested in the material. 195 barley genotypes were grown in two N regimes. Clear positive breeding effect on N and P use efficiency in barley cultivars was found. N
and P use efficiency seemed to be coupled. Efficient N use and utilization correlated positively with grain yield and negatively with stem length.

Estonian breeding lines with comparatively short stem and planophyle growth type ranged the highest according to average yield in trials in Estonia and Latvia. The highest yielding line 4628.6.6.3 showed adaptability to favourable environments and was susceptible to loose smut. The second best line 4491.3.3.9 had a trend to adaptation to unfavourable environments and relatively low infection with loose smut; This line can be recommended as appropriate for organic growing. The most stable over environments according to coefficient of regression was line ST-12945 (Latvia, Stende) with low number of loose smut infected plants and good early vigour. No plants of Latvian variety 'Rasa' were infected by loose smut and leaf stripe. Recently registered hulless barley variety 'Irbe' selected for conventional farming was adapted to favourable conditions, whereas the other hulless barley genotypes (landraces, old or organic varieties) tended to be better adapted to unfavourable environments.

At the breeding station Priekuli, Latvia, at special task was made focusing on seed borne diseases in barley. The aim of this study was to identify resistant spring barley genotypes to loose smut (*Ustilago nuda*) and leaf stripe (*Pyrenophora graminea*) and to evaluate effectiveness of molecular marker for loose smut resistance gene Un8. No infection with loose smut was found for 67 genotypes and no infection with leaf stripe for 17 genotypes. 7 genotypes did not have infected plants by both diseases. Marker data was fully corresponding to the loose smut occurrence phenotype data for 72 RIL lines (73.5%), and clear contradictions were found for 10 lines. 10 lines in which Un8 gene was identified got loose smut infected plants. It shows that the marker is not fully credible and the selected perspective breeding lines has to be double-checked by artificial inoculation. 23 RIL lines showed resistance and can be used in further breeding as resistance source.

Research with *Ustilago nuda* in barley was performed at the State Stende Cereal Breeding Institute. The number of tested spring barley genotypes was 235 in 2013 including genotypes from working collection and breeding lines from spring barley breeding programs in Latvia. During three test years 30 spring barley accessions did not have any loose smut infected plants. Screening for resistance to loose smut was carried out for spring barley (41 in 2013; 30 in 2014). Infection level among susceptible genotypes varied from 1.1 to 83.3% of damaged ears. Only two six-row spring barley accessions (Fox, Milton) showed complete resistance under artificial and provocative infection backgrounds.

At Stende Plant Breeding Station, Latvia, evaluation of hulless barley was carried out in organically managed field with 47 genotypes (8 two-row, 47 six-row) from 2013 to 2015. Grain yield of genotypes (average of 2014 and 2015) varied from 1.76-4.70 t ha⁻¹. Grain yield was higher for two-row hulless barley compare to six-row ones. Genotypes of both types had high variation according to 1000 kernel weight (29.4-55.2 g), crude protein (10.2-15.4%) and beta-glucan content (3.8-6.3%). Loose smut disease-free genotypes (27) under natural infection background was identified. The highest grain yield at Stende location showed two-row genotype Canadian/uncnown (4.70 t ha⁻¹), and six-row genotype Italian naked/14293 (4.24 t ha⁻¹). Both hulless barley genotypes did not have any loose smut infected plants during two testing years.

Task 1.3 - Improving bunt resistance in CCPs [A. Borgen, Agrologica, DK]. 220 single cross populations were infested with bunt spores by Agrologica and grown separately for 3 years with natural development of common bunt. In total 15 CCPs were developed. Each year, the frequency of common bunt was recorded. In the first year, all cross populations had a high infection level. Single cross populations with one parent having a resistance gene for which no virulence were found developed increased resistance during the period, whereas other cross populations maintaind a high infection level with no decrease. The complex CCPs showed no sign of decreased infection level during the period. The lack of decreased bunt infection level in the CCPs indicates that diversity of resistance mechanisms against bunt in a population cannot compensate for the development of virulence within a spore population.
Task 1.4 - Develop formulations of essential oils for seed treatments on peas. [L Pecetti, CRA-FLC, IT]. Clove (Syzygium aromaticum) oil, tea tree (Melaleuca alternifolia) oil and thyme (Thymus vulgaris) oil were tested for their efficacy against the ascocytta blight fungi complex. Pathogen control by oil application with seed submersion was promising in in vitro tests. However, plant establishment after the seed submersion treatment was consistently lowest in field trials. Some intrinsic toxicity of the essential oils on germinating seeds could not be excluded based on laboratory tests. In addition to any oil effect, an excessive imbibition during seed submersion may have somehow damaged the outer seed coats. In field trials, the spray application of essential oils with film coating was as effective as the application of copper sulphate in favouring the establishment of plants deriving from infected seeds (double application of either clove oil at 0.2% + pinolene as coating, or tea tree oil at 3% + pinolene, were most promising). If further confirmed, this technique would offer the possibility of using the oils as alternative natural and biodegradable antifungal agents.

In NW Europe with regular rainfall during the year Botrytis cinerea on pea and field bean, Colletotrichum acutatum and Pleiochaeta setosa on lupin species are a much bigger phytosanitary challenge. Therefore a laboratory screening experiment with different essential oils and formulations of essential oils at the experimental farm of the Ghent University and the University College Ghent of was set up. Tendencies of inhibition of spore germination and of mycelial growth by different essential oils and their doses were similar for all tested isolates within the different species and between species. Extracts based on Thymus vulgaris and Syzigium aromaticum had a good inhibition on the mycelial growth and spore germination of all tested isolates. Under in vitro circumstances doses of 0.5 l/ha and higher result in a total control of the pathogens. The activity of tea tree (Melaleuca alternifolia) oil and oil of common juniper (Juniperus communis) was insufficient to control the selected plant pathogens. If the effect of the application of the essential oils was evaluated, it turned out to be necessary to encapsulate the lipophilic components. Furthermore, the impact of the highly concentrated emulsions on the seed germination was smaller. This could mainly be explained by a more superficial penetration compared to the lower concentrated formulations.

Task 1.4.2: Developing resistance to the complex of foliar diseases Mycosphaerella pinodes, Aschochyta pisi and Phoma medicaginis in pea based on pure lines and CCP-mass selection. Task 1.4.2 was terminated, as the subtask responsible Jens Knudsen left COBRA during the project period.

B- comments on deviations from the original plan:

See Task 1.4.2

WP 2 Breeding for resilience

Responsible partner: 17. KU. Maria Frinckh

Original description of work:

Objectives are to (1) study the potential of Hi-D (CCPs) and specific genotypes to adapt to climatic fluctuations; (2) to find determinants of early vigour and competitiveness against weeds; (3) to identify breeding strategies for coping with multiple stressors through studying the effects of variable degrees of functional diversity in crops.

Task 2.1 Coping with climate change [M Finckh, KU, DE]. This Task will study adaptation to climatic fluctuations in wheat, barley and grain legumes. (1) In 2005, researchers from several European countries initiated coordinated field trials with the F5 of a wheat CCP from the UK. Since the F8, the CCPs are either staying in one place (home CCPs) or moving around among 8 partners and some farmers in Germany (i.e. ‘cycling’ CCPs). By 2013, there will be ca 18-20 F13 CCPs available, all originating from one seed batch with vastly different histories. Exploiting this unique opportunity, COBRA will test the hypothesis that exposing genetically diverse material to varying environments under natural selection increases general adaptability to climate change. For this,
the cycling CCPs will be compared with ‘home’ CCPs in the field and by molecular markers. (2) For barley, trials will expand work conducted in the SUSTAIN-NordForsk project where a genetically wide array of European spring barley accessions has been analyzed in extreme climatic conditions. A smaller array of ca. 50 interesting accessions will be explored for their environmental plasticity in single- and multifactor treatments to allow breeders to integrate best genotypes in their programmes. (3) Finally, to identify grain legume accessions with high resilience to climatic fluctuations and determine underlying plant traits COBRA will test pea and faba bean accessions for morphology, yield and feed value (protein contents and antinutritive substances) in organic field trials in six countries with a wide geographical range (BE: INAGRO, Hogent, AARI (TUR), DK, LUX). Partners will choose 1-2 additional grain legume species with high relevance or innovative potential for their region for additional evaluation trials (see Part A, partner role descriptions for details). The partners will also map the existing legume breeding activities, the geographic potential for each variety and define the most important breeding goals for organically grown grain legumes in the different regions of Europe.

Task 2.2 Coping with weeds and intercropping [N.-O. Bertholdsson, SLU, SE]. COBRA (1) will evaluate wheat improved for allelopathy and selections from the UK CCP for their weed suppression ability and general performance in the field in Northern and Central Europe. Further, evolutionary changes in root growth and allelopathic activity will be studied (a) in evolving CCPs after several years of natural selection in organic vs. conventional systems and (b) in CCPs grown for several generations with/without mechanical weed control or undersown companion crops to test if this has improved the CCP’s competitiveness against weeds. This natural selection process will be compared against the use of a hydroponic system as a novel mass selection tool for root growth and early vigour. Coordinated with these Subtasks COBRA will (2) conduct root morphological analyses especially of root hairs to study genetic variation in root hair length and density, to ensure that cereal varieties selected for efficient nutrient uptake and competitiveness due to other root parameters are also characterized by sufficient root hair growth. (3) The grain legume trials in Task 2.1 will also be assessed for weed biomass and early vigour. In this context of studying plant competitiveness, additional trials will explore co-breeding pea for intercropping with cereals.

Task 2.3 Diversity for coping with multiple stressors [R B Jørgensen, DTU, DK]. To understand the fundamental ecological principles underlying plant diversity effects in response to multiple stresses, controlled experiments will be conducted in the UK and DK. (1) We assess yield and stability effects of increased genetic diversity within drought tolerant wheat and drought/temperature tolerant barley varieties that vary in their response to fungal diseases and low nutrient levels. In greenhouse and field experiments we expose monocultures and mixtures of varieties to combinations of drought, temperature and the other stresses e.g. also nutrient deficiency. We quantify effects of stepwise increased genetic diversity by growing varieties in increasingly complex mixtures, measuring their yield responses under different stresses. (2) We study how evolving crop populations respond to multiple stresses. When seed of grain crops is saved and re-sown, genotypes well adapted to stress conditions contribute more seed to the next year’s population than less adapted plants. Following plant populations over several generations under varied stress regimes allows us to extract evolutionary stable strategies to guide plant breeding and variety selection. Experiments are performed with wheat and spring barley, where large arrays of genetic diverse genotypes are tested in singe and multifactor experiments. COBRA will link the experiments up with other organic breeding activities by including material tested in other COBRA Tasks, e.g. Task 1.2.

Report on results obtained and changes to the original plan/WP aims:

Task 2.1 - Coping with climate change
Sub-task 2.1.1 Coping with climate change
The variety tests carried out by the partners involved across Europe (Scotland, Sweden, Estonia, Belgium, Germany, Slovenia, etc.) resulted in the identification of widely adapted wheat and/or barley germplasm. In addition to the planned partners (ORC and KU) for the trials with CCPs, at
least 12 partners conducted experiments with the CCPs. The CCPs performed exceptionally well with respect to an emerging epidemic of yellow rust across Europe in all sites where they were tested and disease data reported showing the superiority of genetic diversity for coping with unexpected stress. The same was true for the CCP performance during a severe drought in Germany compared to wheat pure stand lines. A total of 16 CCPs that were either cycled among partners since 2008 or had stayed in Germany, Hungary or the UK were compared for two years in Germany (KU). In addition, the German population was grown by six participating farms for two or three years on their farm and these populations were also included in the comparison trial in the second year. Evolutionary changes occurred for morphological traits such as stem length, ear length and degree of ear awnedness, but not for grain yield, disease incidence or molecular markers. The results suggest that within a timeframe of only two to three years morphological traits can already be selected for while agronomic traits such as grain yield and disease resistance do not change that fast. Baking quality parameters (sedimentation, falling number, etc.) of the CCPs often suggested lower performance than pure stands, however, these were not reflected in baking volume that usually did not differ from the high baking quality reference varieties. The results confirm the plasticity of the CC populations and their ability to adapt and change within very short time periods of even two to three years under adequate selection pressures.

Sub-task 2.1.2 - Evaluation of Barley for adaptation to extreme climates

In a technically advanced climate phytotron about 200 accessions of landraces, old and newer barley cultivars, a few wheat lines and a CCP were grown and phenotyped for their entire life cycle under future environmental conditions of elevated CO$_2$, O$_3$ and temperature and their combinations. Grain yield, vegetative biomass, number of ears and seeds and their environmental stability (all accessions) and protein contents in a large selection of these cultivars were analyzed (Ingvorsen et al., 2015a 2016). Association mapping using approximately 7000 SNP markers resulted in 60 very solid associations between traits expressed under elevated CO$_2$, O$_3$ and temperature and their combinations (Ingvorsen et al., 2015b). This information can be used in MAS breeding for cultivars adapted to the future climate conditions. Approximately 140 of the barley accessions were also analyzed in the field for resistance to five of the most common leaf fungal diseases (results to be published in 2016). Three years of field trials were performed under ambient and elevated CO$_2$ (on average 560 ppm) in a FACE facility (Free Air Carbon) comparing old and new cultivars of barley and mixtures under traiditional and low input cultivation. Grain yield, vegetative biomass and foliar diseases were recorded as well as WUE in the first year of the trial. Some of the new and of the old genotypes were better at exploiting the extra CO$_2$. Most of the well performing cultivars harbored the SNP markers found in our association mapping performed under elevated CO2 in the phytotron (results to be published in 2016).

The eco-efficiency of spring barley cultivation for malting in Denmark in a future changed climate has been evaluated through Life Cycle Assessment (LCA) comparing alternative future cultivation scenarios, both excluding and including earlier sowing and cultivar selection as measures of adaptation to a changed climate, changed climate, as well as impacts of a long heat-wave. LCA results showed that all environmental impact categories experienced increased impact for all investigated scenarios, except under the very optimistic assumption that the pace of yield improvement by breeding in the future will be the same as it was in the last decades. If yield measurements are based on relative protein content, the negative effects of the future climate seem to be reduced, though remaining negative in almost all scenarios. Results are presented in Niero et al. 2015 a and b.

Subtask 2.1.3 - Evaluation of grain legumes for adaptation to extreme climates

In Belgium, Luxembourg, Germany, Latvia, and Turkey peas, faba beans, lupins and/or soybeans were tested for performance under organic conditions in spring and winter trials. Besides performance important aspects were feed value, potential for weed suppression (see also task 2.2.3) and pre-crop value. Performance of spring and winter crops in central Europe was very variable with faba beans being overall most reliable. All grain legumes showed a better previous crop value than the non-nitrogen-fixing control triticale with respect to grain yield and protein yield.
In Turkey the promising advanced lines in terms of yield and quality of faba bean, soybean and cowpea screened in COBRA project are transferred to period of registrations and three different private companies declared to MoFAL (AARI) that they are willing to have manufacturing permits some of the materials officially. Agro-morphologically characterized field pea landraces will be evaluated in pre-breeding and breeding projects planned in near future.

Task 2.2 Coping with weeds and intercropping

Subtask 2.2.1 Winter wheat CCPs: Early vigour, growth and allelopathy
Evolutionary trends of seedling root and shoot growth in hydroponics and allelopathic activity were studied using a bioassay. The study was conducted on seeds from generation 6 ($F_6$) and 11 ($F_{11}$) from winter wheat CCPs produced for quality, yield and a combined yield and quality population that are maintained at Neu Eichenberg, Germany under either organic or conventional conditions. Seedling root length, seedling root - and shoot weight in the $F_{11}$ of the organically-managed CCPs were significantly greater than in the organic $F_6$ CCPs. In the conventionally-managed CCPs no such differences were observed. Both organic and conventional CCPs produced for quality showed higher early root and shoot growth than those produced for yield, stressing the importance of early vigour for nutrient use efficiency NUE. The observed changes are related to differences in management rather than chance. There were no significant differences in the allelopathic activity of the populations and between generations. It is concluded that observed differences are more related to NUE and intra-specific competition for light rather than from a direct effect of increased weed pressure in the organic system (Bertholdsson et al. 2016).

Field trials were conducted in five environments in Sweden, Scotland and Estonia. Winter wheat CCPs, mixtures and modern varieties had high yield stability. Pure lines selected from the CCP as being strongly allelopathic showed higher WCA than the CCP itself indicating that a direct selection for a trait with importance to WCA could be successful. Mixture effects in spring wheat were lower and even negative compared with the winter wheat material. However, the mixture of the breeding lines was more stable over all three locations than the components.

Subtask 2.2.2 Root morphological analyses
This study found significant differences in root hair length. This was also affected by growing conditions, as shown by increased density, and tendency of increased length, with increased N level found here. The CCPorg had high values for both root hair density and length, but did not have more or longer primary roots than other accessions. Hence, these data do not indicate a generally higher growth vigour in the CCP, but the CCPorg has good characteristics for root hair morphology since root hairs are important to acquire nutrients especially by organic growing conditions with a relatively slow release of nutrients.

Subtask 2.2.3 Field trials of intercropping of grain legumes with cereals
As during the screenings in 2.1.3, it became clear that grain legumes cultured in their region of selection performed better; winter varieties can provide an alternative. Winter and summer varieties of grain legumes were combined with cereals usually with more stable yields than in the pure stands. However, the amount of legume and cereal harvested is strongly influenced by year and site. A challenge remains to find the correct combinations to insure synchronous ripening. Direct weed suppression in mixtures of grain legumes were evident in autumn and spring sown mixtures. In Turkey advanced lines of soybean and faba bean superior for weed suppression were identified.

Task 2.3 - Diversity for coping with multiple stressors
As described above, the task was coordinated together with subtask 2.1.1 and 2.1.2 and results are reported there.

B- comments on deviations from the original plan:
The only deviations from the original plans were the integration of additional trials.
**WP 3**  **Improving breeding efficiency**

**Responsible partner:** 25. TUM. Peter Baresel

**Original description of work:**
Objectives are to (1) identify best methods allowing the constitution and continuous improvement of Hi-D; (2) improve Hi-D adaptation to and productivity under the conditions of organic farming; (3) improve the possibilities of introgression of genetic resources into the breeders’ genebanks and germplasm exchange through the establishment of networks; (4) further develop breeding methodologies and techniques adapted to participatory plant breeding.

**Task 3.1** Improving breeding methodology and strategies in wheat [P Baresel, TUM, DE]. COBRA will assess the potential of improved mass selection or similar methods to produce a high number of genotypes for maintaining the character of a population. Several methods will be assessed: (a) automated and non-destructive high-throughput selection for quality parameters based on single seeds; (b) selection among single plants or nanoplots distributed in regular patterns, permitting, to a certain extent also high-throughput-selection for quantitative traits and to perform neighbour analysis; (c) Marker Assisted Selection; (d) dynamic management of populations, i.e. exploitation of natural selection with and without additional mass selection; and (e) Recurrent selection (RS) based on the above-mentioned selection methods. For the latter, several techniques to generate mass-crosses will be studied: Use of male-sterile lines, exploitation of spontaneous cross-pollination using Xenia as marker to identify cross-pollinated progenies (xenia-assisted recurrent selection; XARS), and others. All these methods will be compared directly in an experimental programme carried out by 4 partners in 4 countries. The trial design will allow us to calculate heritabilities and effectiveness of the selection methods and to simulate various scenarios, e.g. high and low selection intensities, or “fuzzy selection” i.e. incomplete removal of genotypes.

**Task 3.2** Evolutionary, participatory and marker-assisted selection approaches for grain legumes [P Annicchiarico, CRA-FLC, IT]. This Task has 4 Subtasks. Subtask 1: assessing low-cost evolutionary bulk breeding approaches for pea selection and comparing them with the conventional single-seed descent (SSD) approach. The two-year comparison under organic farming of bulk- vs. SSD-derived lines from each of three crosses will verify the ability of the evolutionary selection scheme (which is cheaper than SSD) to also provide greater selection gains; the frequency of elite lines observed in 9 crosses subjected to evolutionary selection will be related to earlier bulk data of these crosses, to verify whether early bulk data can be exploited to exclude less promising crosses. Subtask 2: comparing farmers’ vs. breeders’ selection criteria and their implications on pea selection. We will assess the consistency between farmers’ and breeders’ evaluation scores and priority traits on large samples of pea lines, and compare farmers’ vs. breeders’ selections for grain yield and other traits. Subtask 3: investigating marker-assisted selection for pea grain yield and quality. Some 270 recombinant inbred lines (RILs; 90 for each of 3 connected crosses) will be phenotyped in 3 organically-managed environments and genotyped through 384 SNP. Subtask 4: producing higher-yielding potential varieties for organic systems (based on the overall phenotyping of about 450 pea lines). COBRA will fund one phenotyping experiment for each of the 4 Subtasks.

**Task 3.3** Phenotyping methods [P Baresel, TUM, DE]. This is aimed at developing appropriate phenotyping protocols that are applicable non-destructively on high numbers of plants and in early generations for the above-mentioned breeding methods. Such phenotyping methods are necessary for the direct use as selection tools, but also to establish associations between molecular markers and phenotypic traits, where suitable high-throughput phenotyping methods with reasonable costs are often lacking. COBRA will test methods of non-destructive quality assessment on single grains, non-destructive determination of biomass, and yield by digital image analysis, as well as remote sensing by spectroscopy.

**Task 3.4** Strategies to increase diversity in populations [I. Goldringer, INRA, F]. (1) Based on the methodology developed in Tasks 3.1 and 3.2, hierarchical systems of population enrichment by recurrent introgression will be designed and implemented in different environments, consisting of a set of several populations, differing in the degree of adaptation, genetic variability and performance, e.g. landraces, broad or narrow CCPs. Each population is regularly improved by...
recurrrent selection {Kannenberg, 1995 #85; Kannenberg, 2001 #84}. Before each selection cycle, the basal population is enriched with new more or less adapted material. Part of each population is transferred to the population of the next level after each selection cycle. Exchange of material from several populations originating from participatory breeding and natural adaptation will lead to networks with continuous flow of genetic material between different environments in organic farming. (2) In addition, COBRA will study the development of wheat dynamic populations and the comparison with other strategies within a PPB context. Populations with increasing diversity (improved landraces, CCPs, dynamic populations) will be submitted to farmers’ evaluation and selection in the framework of a French network of farmers involved in PPB. In connection with WP2 aims (Task2.2, Coping with climate change), we will assess the adaptability and temporal stability of the dynamic populations compared to the other genetic structures; in relation with Task 2.2 (Coping with weeds and intercropping), dynamic populations' weed competitiveness will be assessed and co-breeding with legumes strategies will be initiated based on SOLIBAM activities; in relation to Task 2.3 (Diversity for coping with multiple stressors), organic farmers who are often facing multiple stress situations with very poor soil, will assess crop performance of the dynamic population and the impact of within-population diversity to cope with these stresses; AMF symbiosis will be evaluated in a subset of populations to complement the breeding criteria. Using the information from phenotypic evaluation in several farms over multiple years will provide an assessment of the yield response under multiples stressors. Environmental effects, GxE interactions will be characterised and used to explain the evolution over time of dynamic populations and other wheat CCPs in the network of French farms.

Report on results obtained and changes to the original plan/WP aims:

A- results obtained:

**Task 3.1 Improving breeding methodology and strategies in wheat** Aim of this task was to test innovative methods of mass selection and molecular methods for the improvement of bulk populations and for selection of more resilient varieties. Mass selection for grain protein content was papplied on a bulk population of wheat for two consecutive generations using an automatic single seed sorter based on NIR spectrometry. The experiment showed that, despite the high variability within genotypes, it is possible to increase the protein content in populations in this way, but, as in pure line varieties, this is on the expense of a yield loss of about 10% for each percent of protein content. Another important aspect of breeding wheat populations is recombination, e.g. in order to introduce additional genotypes or joint different populations, or after mass selection. One approach, is to exploit spontaneous outcrossing using xenia as marker. Adapted populations of winter and spring wheat, carrying genes conferring blue aleurone genes, have been established at the technical university of Munich; using appropriate experimental designs, F1 seeds can now easily be identified and sorted out using automatic seed sorters. Approximately 2 % of cross pollinated seeds could be identified in an experiment carried out at Freising in 2015. Future cereal cultivars will have to produce under the constraints of higher temperature in combination with increased concentrations of atmospheric carbon dioxide and ozone as a consequence of climate change. Phenotypic selection for these future growth conditions is currently not possible under field conditions. Therefore, a genome-wide association study (GWAS) on a diverse set of 167 spring barley genotypes was performed to identify markers for increased primary production under artificially elevated levels of temperature, CO2 and Ozone. The study identified markers and chromosome regions to be targeted in breeding for development of climate resilient cultivars.

**Tasks 3.2 Evolutionary, participatory and marker-assisted selection approaches for grain legumes**

Evolutionary bulk breeding approaches for pea were compared with traditional single-seed-descent methods in two environments in Italy. Moreover the effectiveness of visual selection of professional breeders and farmers on grain yield was compared, in order to assess the suitability of participatory breeding approaches. Finally, marker-assisted selection was applied on a germplasm sample phenotyped under organically-managed conditions, in order to develop a genomic selection model for organic agriculture. Bulk breeding, was better able to produce higher yielding...
lines. Moreover, in environments similar to those in which they evolved, evolutionary mixtures were higher yielding than bulk breeding-derived pure lines or narrow-based mixtures. However, pure lines (or mixtures) seemed to be preferable in a wider adaptation context. Though farmers and breeders included similar priority for visually assessed traits in selection indices, in a subsequent evaluation in two contrasting sites, farmers’ selected lines tended to be actually higher yielding than breeders’ selected lines. Consequently, there seem to be no reason not to involve experienced farmers in the breeding process. The genotyping-by-sequencing (GBS) high-throughput technique provided thousands of polymorphic SNP markers of three interconnected crosses among pea varieties. The accuracy of genomic selection was large to moderate, suggesting a possible implementation of this approach for organic agriculture.

**Task 3.3 Phenotyping methods** Several non-destructive phenotyping methods were compared for assessment of early vigor, biomass, LAI and leaf Chlorophyll content in wheat. These comprised mainly spectral measurements and digital analysis of thermal and color images. The latter proved to be very accurate for determination of ground cover and thus indirectly of early vigor, LAI and biomass. Color analysis turned out to be suitable for detection of long-term effects of biotic and abiotic stresses, while thermal imaging may be used for direct detection of drought stress.

**Task 3.4 Strategies to increase diversity in populations** A large participatory breeding programme has been carried out and new approaches for the management of on-farm experiments have been further developed, comprising improved experimental designs, the development of a bayesian model specifically designed to analyze the unbalanced the data obtained in the PPB experiments and the establishment of a database system for the management of these data. Moreover, several new bulk populations of bred wheat, adapted to central European conditions were created in 2012-2014 based on commercial varieties adapted to Germany and confining countries. The parents comprised 9 winter wheat and 7 spring wheat, genotypes carrying xenia markers as well as germplasm resistant to common and dwarf bunt. Thereby, the basis for further research and practical application of participatory breeding and dynamic management of populations has been created.

**B- comments on deviations from the original plan:** There were no major deviations from the plan.

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### WP 4 Socio-economics and legislation

**Responsible partner:** 7. NIBIO. Tove Mariegaard Pedersen.

**Original description of work:**
This WP is organised in two Tasks which link Europe-wide inventories with in-depth case studies at national level. The general objective is to lower socio-economic and legal hurdles to organic seed production and organic plant breeding.

**Task 4.1 Regional chains, organisation and funding of organic plant breeding [A Stubsgaard, VFL, DK].** The Task aims to lower breeders’ thresholds to engage with the organic sector by increasing communication and cooperation through the production chain, thus discovering possible shortcuts that have not been exploited before, because the organic sector has largely been ignored by professional breeders. COBRA will make an inventory of initiatives for alternative breeding approaches, including Hi-D initiatives. Several case studies will determine how quality requirements in regional production chains are related to breeding and selection criteria. These will focus on wheat (bread and pasta), barley and grain legumes, to study in depth how local breeding initiatives can be better embedded in local production chains. VFL, ISD, ORC, and UniUD with AIAB will do at least one case study each; UniUD will run supply-chain analyses (winter cereals and soya bean) focusing on the views of plant breeders, farmers, customers, processors in both low-input and organic systems. Descriptions of the country-specific barriers and drivers will be performed through a European Workshop on organizational and financial models for organic plant breeding. At this Workshop (M13), the 2007 Proceedings of the ECO-PB International Workshop on different models to finance plant breeding will be updated and extended with new promising
examples of how breeding efforts can be regionally integrated in the food chain. All stakeholders contribute with their models, either with posters or with presentations. Conference proceedings will be edited and published. Thus, COBRA will identify the most successful models and propose innovative financing models for organic plant breeding, e.g. by creating shortcuts in the production chain through improved communication and cooperation.

Task 4.2 Legislation & Registration: how to deal with genetically diverse material [R Bocci, AIAB, IT]. COBRA will (1) identify and discuss the present needs concerning the regulatory framework for organic seed systems with stakeholders in all the participating European countries (seed industries, breeders (large and small), public research institutes, farmers’ associations, policy makers, and NGOs in Europe); and (2) produce a country based report on legal constraints and opportunities of organic and diversity-focused breeding and seed production. The activities planned in this Task include a comprehensive literature review on current constraints of seed laws and registration procedures regarding organic seed; a joint meeting with ECO-PB and other expert groups discussing the outcomes of the survey (M26), with the involvement of officials involved in seed testing or registration at national and EU level (e.g. DG SANCO).

Report on results obtained and changes to the original plan/WP aims:
(max 1 page, Arial, size 11)

A- results obtained:

Task 4.1

Financing models and case studies performed by a number of partners were presented at a socio-economic workshop in June 2015 in Freisingen, Germany. Contributions are also presented in the booklet: “Breeding for diversity – political implications and new pathways for the future”:

- Progress in breeding for organics: impacts of the seed derogation system
- Implementation of populations (CCPs) in the Netherlands
- The situation of composite cross populations in Denmark
- The challenge of how to finance plant breeding for organic farming
- Breeding pure line varieties of spring wheat for organic agriculture in the Netherlands
- Breeding for improved weed competitiveness in spring barley
- Conventional vs. organic soybean production in northeast Italy
- Status quo analysis of seed production and breeding of organic wheat in Slovenia

Partners have also contributed to an inventory of alternative breeding initiatives.

The work has resulted in an increased focus on specific barriers and possibilities in different organic breeding initiatives.

Task 4.2

Meetings (involving several partners in different partner countries) discussing present needs for the regulatory framework including workshops with DG Sanco has taken place.

Several partners are taking part in the Commission Decision on the “Call for Participation for a temporary experiment providing for certain derogations for the marketing of populations of the plant species wheat, barley, oats and maize.”

Questionnaire for breeders and seed companies were distributed within the partners network in September 2013

Legislative workshop was held in October 2013 in Brussels with ECO-PB and results from the questionnaire were presented.
An Italian country report was published in 2015.

Contributions from task 4.2 are presented in the booklet: “Breeding for diversity – political implications and new pathways for the future”:
- Plant genetic diversity, farmers rights and the European seed legislation
- Seed marketing in Europe: an opening for diversity?
- Organic seeds and plant breeding from the seed companies perspective

B- comments on deviations from the original plan:
Delivered as planned.

<table>
<thead>
<tr>
<th>WP 5</th>
<th>Dissemination and networking</th>
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</thead>
<tbody>
<tr>
<td><strong>Responsible partner:</strong> 5. AIAB. Livia Ortolani.</td>
<td></td>
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<tr>
<td><strong>Original description of work:</strong></td>
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<tr>
<td>This WP will have the contribution of all the partners of the project with the aim of (1) make sure that COBRA is intensely linked with relevant research projects (see table 3 below) other current breeding initiatives and the existing networks of organic farmers or initiatives. This includes the creation and circulation of a common newsletter which highlights upcoming relevant events and reports on new developments both from COBRA and from other (organic) breeding projects. WP 5 will also (2) coordinate knowledge transfer activities, through (a) two stakeholder workshops on organic plant breeding (one on cereals, one on grain legumes); (b) a dedicated training programme (see below, B7); (c) a joint meeting with the EUCARPIA organic plant breeding section (International Conference, end of 2013 in Göttingen, Germany), in collaboration with SOLIBAM and ECOPB; (d) a brief online video translated in different languages presenting the major COBRA objectives to a larger public; (e) field days organized by partners in each country (1 per year), facilitating exchanging and networking on organic plant breeding issues; (f) the creation of a public web page for COBRA, which links to all partners’ websites and to the Core Organic web site, where relevant materials from all the WPs will be collected and published with the aim of facilitating the practical implementation of project output and further research in the field starting from project results; the CCP initiatives identified in WP4 will be mapped on a specific page that can be easily updated by the users.</td>
<td></td>
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</table>

| Report on results obtained and changes to the original plan/WP aims: |
| The large network of partners in COBRA could assure high quality information to be updated on the webpage and links with other projects, however a continuous activity from the AIAB team was needed to encourage partners to contribute in the newsletter and website. |

Two stakeholders workshops have been organized one at the beginning and one at the end of the project, with the aim of encouraging the researcher interaction with end users, following a multi actor approach. This approach is particularly relevant for organic plant breeding, as a need to further develop the sector, with the support of research, exists. The first stakeholder workshop was held in October 2014, in Istanbul within the preconference of IFOAM OWC assuring the connection with the organic sector as a whole. Other events at local level have been organized by different partners and advised in the webpage (e.g. “Filigrane” in Italy in October 2015 on cereals). A final workshop on Grain Legume took place in Denmark in November 2015 during the final conference of the COBRA project with the local organic sector. While the first workshop had a large participation of actors from the organic sector, with some COBRA partners as speakers that presented the research constrains in organic plant breeding, the second workshop, included in the final conference of the project, had the larger participation of researchers, with people from seeds companies as speakers that put to the attention their requirements to research and commented on project results.
An **International Conference** was organized in 2014 joint with SOLIBAM final conference in France, Nantes from 7 to 9 of July 2014. This was a really good opportunity for both projects. The idea of organize joint conferences to present project results allow to both project to have more than just partners funded to participate in the conference and make the event a good opportunity of mutual learning for all participants. A specific printed publication of book of abstract for paper presented by COBRA partners at the international conference has been produced after the conference.

The main tool for dissemination created in WP5 during the project lifespan was the **web page** [www.cobra-div.eu](http://www.cobra-div.eu). It contains general information about the project, the list of partners, their locations, project aims, objectives and structure. However it was not static, but the webpage represented for the large partnership involved in COBRA the space to post all information related to dissemination and networking activities: specific section have been dedicated to training opportunities, field days and news. The webpage links to connected project and initiatives website and to all partners websites. Photos of field trials and field days have been uploaded on the webpage.

In addition to the website a **Facebook page** of the project was created with the aim to spread to a wider public the news about the project results and activities. The page was promptly updated with the information provided by partners by e-mail to the AIAB team. The Facebook page create connections with other initiatives and give the possibility for people using facebook (quite well spread among farmers and advisers nowadays) to easy “share” news and events that are of their interest.

The other key tool for networking used in COBRA was the **newsletter**. All the partners gave their contribution to the newsletter, coordinate by Maria Barletta from AIAB. 6 Newsletters have been done during the project lifespan (August 2014, November 2014, February 2015, May 2015, August 2015, December 2015). The newsletter reached around 90 people that included all project partners, people from other projects on organic breeding, individual interested people that could subscribe the newsletter for free from the COBRA webpage.

The newsletter was organised with an editorial written by one of the COBRA WP leaders and 6-7 news on coming events, field days reports, training opportunities, report and experimental activities. Each report published in the newsletter is available on the webpage.

Formal and informal **training opportunities** within COBRA partnership has been published in the COBRA website and newsletter. 4 partners gave input on formal training opportunities at their place: BOKU –Austria, ITAB- France, Humbolt University – Germany, AARI Turkey. A total of 25 **field days** during the whole project have been communicated through the website (5 in 2013, 11 in 2014 and 9 in 2015), and even more have been probably organized by partners. They had several objectives at local level, from the presentation of project activities to local actors to the development of local networking on organic plant breeding. They always represented important training opportunities for farmers, breeders, students and external researchers and positive feedback are directly producing synergies and relationships at local level.

**B- comments on deviations from the original plan:**

The main deviation from the original plan was the online video, it was canceled at the beginning of the project in agreement with the Italian funding body. However during the project lifespan the Facebook page was added as communication tool and this required also an additional effort that was not foreseen in the project.

The international conference was not joint with the EUCARPIA meeting in 2013 and was joint with the SOLIBAM final conference instead. This was an opportunity that the project partners approved due to interest by more partners to be involved in the SOLIBAM final conference. It was definitly a good choice.
The training programme did not have a good success. This is due to the lack of resources to develop a dedicated training programme within a CO project, but also to the fact that not all the partners offer often training opportunities or identified the space of the project as a way to advise their training opportunities, probably both students and professors use different networks than research project to look at training opportunities. However, a higher success was observed concerning informal training opportunities such as field days that are less costly to be organized and encourage the development of a multi actor approach at local level.

4. Publications and dissemination activities

4.1 List extracted from Organic Eprints (as of 06/07/2016)

Conference paper, poster, etc.


Beeckman, Annelies; Dewaele, Karel and Delanote, Lieven (2014) Rassenkeuzen erwten en veldbonen in combinatie met triticale. [Variety choice of peas and fava beans together with triticale.] Working paper.


Delanote, Lieven; Temmerman, Femke and Dewaele, Karel (2014) Composite cross populations keep up with winter wheat varieties. Inagro vzw.


Dewaele, Karel and Delanote, Lieven (2015) A focus on cereals in conference on organic breeding and propagation material with Belgian stakeholders. COBRA.


Dewaele, Karel; Delanote, Lieven; Dewitte, Kevin and Haesaert, Geert (2015) Organic field trials of winter and summer crop mixtures of grain legumes and cereals in Belgium. Poster at: COBRA Final Conference, Denmark, 24th-25th November 2015.


Dewaele, Karel; Delanote, Lieven and Temmerman, Femke (2014) Samengestelde kruisingspopulaties doen niet onder voor winteratarwassen. [Composite cross populations keep up with winter wheat varieties.] Working paper.


Ingvorsen, Cathrine Heinz; Backes, Gunter; Lyngkjær, M.; Peltonen-Sainio, Pirjo; Jensen, Jens D.; Jalli, Marja; Jahoor, Ahmed; Rasmussen, Morten; Mikkelsen, Tesi N.; Stockmarr, A. and Jørgensen, Rikke Bagger (2015) Significant decrease in yield under future climate conditions.


Miceli, F.; Centa, M; De Infanti, R and Signor, M (2014) *Food Soybean Varieties in Low-Input Conditions Grain Yield and Quality from Three NE Italy Environments*. In: Chable, Véronique; Goldringer, I.; Howlett, Sally; Barberi, Paolo; Miko, P; Mendes-Moreira, P; Rakszegi, M; Ostergaard, H; Borgen, Anders; Finckh, Maria R.; Pedersen, T and Boci, Riccardo (Eds.) *Book of abstracts of SOLIBAM final congress*.

Miceli, F.; Centa, M.; De Infanti, R. and Signor, M. (2014) *Food Soybean Varieties in Low Input Conditions. Grain Yield and Quality from three NE Italy Environments*. In: Chable, Véronique; Goldringer, Isabelle; Howlett, Sally; Barberi, Paolo; Miko, Peter; Mendes-Moreira, Pedro; Rakszegi, Marianna; Ostergaard, Hanne; Borgen, Anders; Finckh, Maria R.; Pedersen, Tove and Boci, Riccardo (Eds.) *Book of Abstracts of SOLIBAM final congress*, pp. 65-66.

Mikkelsen, B. L.; Jørgensen, R. B. and Lyngkjær, M. F. (2014) *Complex interplay of future climate levels of CO2, ozone and temperature on susceptibility to fungal diseases in barley*. *Plant Pathology*, n/a, n/a/n/a.


Weedon, Odette; Brumlop, Sarah; Heinrich, Sven; Boening, Andreas; Elsner, Maren; Finckh, Maria R.; Lammerts van Bueren, E.T.; Nuijten, E.; Messmer, Monika; Baresel, Jörg Peter; Goldringer, I.; Péter, Mikó; Megyeri, Maria; Borgen, Anders; Rasmussen, Søren K.; Pearce, Bruce and Wolfe, Martin (2016) *Agronomic performance of two generations (F12 and F13) of thirteen winter wheat composite cross wheat populations with differing cultivation histories in 2014/15*. Poster at: EUCARPIA Conference 2016, Zürich, Switzerland, 29-1 September 2016. [Submitted]


Weedon, Odette; Haak, Jette; Brumlop, Sarah; Heinrich, Sven and Finckh, Maria R. (2015) *The effect of changing environmental conditions on composite cross winter wheat populations over six years from the F8 to the F14*. In: *Conference Proceedings COBRA*, 1 (1).


**Project description**

4.2 Publications:


Bertholdsson, N.-O.; Weedon, O.; Brumlop, S. and Finckh, M.R. 2016: Evolutionary changes of weed competitive traits in winter wheat composite cross populations in organic and conventional farming systems. European J. of Agronomy (accepted with revision)

Borgen, Anders (2014) Virulence pattern in Danish races of common bunt (Tilletia caries) Common Bunt. In: Chable, Véronique; Goldringer, I.; Howlett, Sally; Barberi, Paolo; Miko, P; Mendes-Moreira, P; Rakszegi, M; Ostergaard, H; Borgen, Anders; Finckh, Maria R.; Pedersen, T and Bocci, Riccardo (Eds.) Diversity strategies for organic and low systems. Book of abstracts of SOLIBAM final congress, Nantes, 7-9 July 2014,


Borgen, Anders (2014) Økologisk Planteforædling holder hus med vores ressourcer, Global Økologi, 2, p. 11.


Dewaele, Karel; Delanote, Lieven; Dewitte, Kevin and Haesaert, Geert (2015) Organic field trials of winter and summer crop mixtures of grain legumes and cereals in Belgium. Poster at: COBRA Final Conference, Denmark, 24th-25th November 2015.


Finckh, Maria R.; Heinrich, Sven and Brumlop, Sarah (2014) Performance of wheat composite crosses on station and on-farm. In: Chable, Véronique; Goldringer, I.; Howlett, Sally; Barberi, Paolo; Miko, P; Mendes-Moreira, P; Rakszegi, M; Ostergaard, H; Borgen, Anders; Finckh, Maria R.; Pedersen, T and Boci, Riccardo (Eds.) Diversity strategies for organic and low systems. Book of abstracts of SOLIBAM final congress, Nantes, 7-9 July 2014, pp. 107-108.


Miceli, F; Centa, M; De Infanti, R and Signor, M (2014) Food Soybean Varieties in Low-Input Conditions Grain Yield and Quality from Three NE Italy Environments. In: Chable, Véronique; Goldringer, I.; Howlett, Sally; Barberi, Paolo; Miko, P; Mendes-Moreira, P; Rakszegi,
M; Ostergaard, H; Borgen, Anders; Finckh, Maria R.; Pedersen, T and Bocci, Riccardo (Eds.) Book of abstracts of SOLIBAM final congress.


Publications submitted:


Jørgensen RB, Mikkelsen TN, Ingvordens CH, Lyngkjær M. mf. Three years of FACE experiment with cultivar and cultivar mixes of barley (Hordeum vulgare) – Traits of importance for CO2 exploitation (to be submitted primo 2016).

Weedon, Odette; Brumlop, Sarah; Heinrich, Sven; Boening, Andreas; Elsner, Maren; Finckh, Maria R.; Lammerts van Bueren, E.T.; Nuijten, E.; Messmer, Monika; Baresel, Jörg Peter; Goldringer, I.; Péter, Mikó; Megyenyi, Maria; Borgen, Anders; Rasmussen, Søren K.; Pearce, Bruce and Wolfe, Martin (2016) Agronomic performance of two generations (F12 and F13) of thirteen winter wheat composite cross wheat populations with differing cultivation histories in 2014/15. Poster at: EUCARPIA Conference 2016, Zürich, Switzerland, 29-1 September 2016. [Submitted]

Dissertation S. Zimmer: Title. This will be available from the University of Kassel server after final corrections were done under: https://kobra.bibliothek.uni-kassel.de/handle/urn:nbn:de:hesis:34-2006060612928 as Dissertation


Paper planned on the grain legume species trial conducted at the University of Kassel in Frankenhausen, Germany. (target journal: Field Crops Research)

Oral presentations:


Oral presentation (09.04.2004) in organic research seminar at Estonian Agricultural University (Tartu).

Oral presentation of Task 1.2 by Borgen, A. 2014: Virulence pattern in Danish races of common bunt (Tilletia caries) at the 7th ISTA Seed Health Symposium 12-14th June 2014.

The XVIII Biennial International Workshop on the Smuts and Bunts Tune/Copenhagen 3rd-5th February 2014 was organised as part of the COBRA project (M6) with oral presentation from Søren Rasmussen (Copenhagen University), Anders Borgen (Agrologica), Almuth Müller (BOKU).


Zimmer, S.: Auswirkungen verschiedener Körnerleguminosen-Arten auf die Nachfrucht Winterweizen. LSG Sorteninformationsversammlung, Beringen, Luxembourg. 04. September 2014 (Oral presentation)


Zimmer, S.: Anbauwürdigkeit von Körnerleguminosen als regionales Futtermittel im biologischen Landbau in Luxemburg. LSG Sorteninformationsversammlung, Beringen, Luxembourg. 11. September 2012 (Oral presentation)

COBRA Conference 24th-25th November. Click here for the book of all abstracts.

24th November
Presentations:
1 “COBRA” Bruce Pearce, WP0
2 “Mapping bunt resistance in winter wheat” Almuth_Elise_Müllner
3 “Detection of fusarium in wheat by multispectral imaging” Johannes_Ravn_Jørgensen
4 “COBRA WP2 Overview” Maria_Finckh
5 “WP1 Overview” Anders_Borgen
6 “Weed competitive ability and yield stability of winter wheat populations, cultivar mixtures and cultivars” Nils-Ove_Bertholdsson
7 “WP4 – Socio-economics and legislation” Tove_Pedersen
8 “COBRA WP5 activities” Riccardo_Bocci
9 Rikke_Bagger
10 Berta_Killermann

25th November
Grain Legume Workshop
Maria_Finckh
Steffi_Zimmer
Alev_Kir

Presentations
1 Edwin_Nuijten
2 Odette_Weedon
3 Isabelle_Goldringe
4 Odette_Weedon
5 Lene_Krusell
6 Riccardo_Bocci
7 Regine_Andersen
8 Bruce_Pearce

Other dissemination activities:

Döring, Thomas (2013) Unleashing the potential of genitic diversity for organic plant breeding, CORE Organic newsletter, April 2013, p. 2.

Newsletter ‘Biopraktijk’ which provides Flemish organic farmers with news on research in organic agriculture, we included 5 articles about the project COBRA and the course of our work in June 2013, July and September 2014 and 2015 and in April 2016. These articles were republished or were the start for articles in some other newsletters, publications and newspapers such as BioKennis, Management&Techniek, Landbouwleven, which have a larger and very diverse readers public in Belgian and Dutch agriculture. Inagro received questions from interested farmers and sector relatives, both organic and non-organic.
Field days and seminars etc.

Fuller details, summaries, reports and pictures of field days can be found on the COBRA website at [http://www.cobra-div.eu/field-days/](http://www.cobra-div.eu/field-days/)

2013

In Italy:
- Farmers and researchers coming from Lodi farm “Tre Cascine” and from the University of Perugia, respectively implemented two Italian field days on May.

In Estonia:
- ECRI members arranged a field visit in Jõgeva (Estonia), involving farmers, advisors, researchers on July.

In Belgium:
- At Rumbeke – Beitem (Belgium), farmers and extension workers met on the experimental organic farm of Inagro in order to briefly present the experiment on grain legumes.

In Luxemburg:
- In Luxemburg (Karelshaff, Colmar-Berg), the CRP-GL and IBLA partners held their official field day in July at the presence of Minister of Agronomy, farmers and other stakeholders.

2014

In Italy:
- A group of organic farmers (at least six in each of the two locations, Lodi, northern Italy, and Perugia, central Italy) visited the field pea evaluation trials and took part in a participatory selection approach by carrying out a synthetic visual evaluation of about 360 pure lines under selection, and ranking several possible selection criteria (on a questionnaire specifically designed) according to their priorities and expectations.

In Luxemburg:
- Official field day in presence of the Minister of Agriculture in Bou
- Field day in Colmar-Berg

In UK:
- ORC open day in Wakelyns Agroforestry, Fressingfield, Suffolk.

In Germany:
- Field day in Neu-Eichenberg: experimental farm Excursion to the dwarf bunt of wheat susceptibility field trials, Wolfersdorf near Freising
- Excursion to the dwarf bunt of wheat susceptibility field trials, Wolfersdorf near Freising
- Excursion to the dwarf bunt of wheat susceptibility field trials, Wolfersdorf near Freising

In Belgium:
- Open field day in a research organic farm.

In Denmark:
- Open field day. Presentation of organic plant breeding in Agrologica, Houvej 55, DK-9550 Mariager.

In Estonia:
- Organic research seminar at Tartu, Estonian University of Life Sciences (09.04.2014);
- Seminar to Baltic seed producers at Jõgeva, ECRI, Estonia (15.04.2014);
- Training day for organic farmers at Jõgeva, ECRI; Estonia (20.04.2014);
- Field day for organic farmers at Jõgeva, ECRI, Estonia (08.07.2014);
- Estonian cereal forum at Paide, Estonia (08.04.2014);

In Latvia:
- Field day in Priekuli
In Turkey

2015
In Turkey
- Event was held with the broad participation of stakeholders of our region at the Aegean Agricultural Research Institute (AARI) (Menemen / Izmir-TURKEY).

In Estonia:
- Seminar for organic farmers at Rakvere, Estonia (6.03.2015);
- Seminar for organic farmers at Saku, Estonia (9.03.2015);
- Seminar for organic farmers at Jõgeva, ECRI, Estonia (06.06.2015);
- Conference „Organic farming 2015” at Avinurme, Estonia (09.06.2015);
- Seminar for Baltic plant breeders at Olustvere, ECRI, Estonia (7-8.07.2015);
- Field day for cereal producers at Jõgeva, ECRI, Estonia (09.07.2015);
- Field day for organic farmers at Jõgeva, ECRI, Estonia (14.07.2015).

In the UK:
- Field day at National Organic Combinable Crops 2015.

In Belgium:
- Open field day in a research organic farm.

4.3 Further possible actions for dissemination
- List publications/deliverables arising from your project that Funding Bodies should consider disseminating (e.g. to reach a broader audience)


In prep:
- K. Dewitte¹, J. Latré¹, K. Dewaele², L. Delanoote² and G. Haesaert¹. Effectiveness of essential oils to control Botrytis cinerea in pea and Colletotrichum acutatum in lupin. Crop Protection

- Indicate publications/deliverables that could usefully be translated (if this has not been done, and indicate target language)

COBRA (2016). Breeding for diversity – political implications and new pathways for the future. Eds. Tove Pedersen, SEGES, Denmark and Frédéric Rey, ITAB, France. (COBRA publication in press). This is a user-friendly summary of the outcomes of COBRA which is currently in its final stages of publication in English. Translation into other languages would allow greater access to the information.

The book of abstracts from the COBRA conference in November 2015.
4.4 Specific questions regarding dissemination and publications

- Is the project website up-to-date?
  Yes up to the end of the project, by the WP5 leader with the support of all partners.

- List the categories of end-users/main users of the research results and how they have been addressed/will be addressed by dissemination activities

The main users of the research activities are: farmers and breeders, researchers and students and official authorities. In particular:

The COBRA website and latterly social media has been used to publicise the work and outputs of the project. Social media has been useful to access an audience that may not usually find such a project ie students, public and has signposted them to the website as well as other information that is relevant to COBRA but not generated by the project. As with many research projects the publication of results starts within the timeframe of the project but will continue for years to come with specific publications coming form the COBRA funded work but also with wider publications that COBRA will contribute.

- **Farmers and breeders**: they are generally informed and subsequently trained on the developments of the research activities (with a particular focus on the portability of those results in field) by means of workshops and field days organized by Focus Crop Coordinators in collaboration with the involved researchers. COBRA has organised specific field days as well as engaged with field days organised by other organisations/projects to ensure that farmers and breeders are exposed to the project and the outcomes of the project.

- **Researchers**: they could keep up to date on progress status by periodic appointments (on Skype or in person) and international assemblies/meetings with the other partners. Moreover, they could obtain desired information on the website, as it will be more and more updated promptly.

- **Official authorities**: they are always invited to both local and international events (field days, meetings, etc) so that an updated and complete description of COBRA progress status is persistently provided to them. Members of the COBRA consortia also have close relationships with their national officials allowing a continuous flow of information.

- Impact of the project in relation to main beneficiaries of the project results
  (Note: for the different categories of end-users/main users of the research results, explain how well the project has been able to reach these target groups, and any known impact)

The impact of the COBRA project on the main beneficiaries of the project (Breeders, Farmers, Researchers and students and Official Authorities).

- **Breeders**: COBRA has provided a range of outputs and deliverables that will have direct impact on breeders. Bunt resistant markers once published by BOKU in the later part of 2016 will provide vital information to inform breeding programmes for both organic as well as conventional breeders. Similarly the protocols germplasm resistant to bunt and dwarf bunt as well as the protocols for CCP breeding will be of direct benefit to breeders. Breeders can also use the protocols for diagnostics for seed vitality and health.

- **Farmers**: There are indirect impacts for farmers from COBRA via breeders above. Direct impact will be information of performance of crops including grain legumes.

- **Researchers and students**: An overarching impact of COBRA was the ability of the project to bring together an incredibly wide range of European organic plant breeding stakeholders for the first time and to facilitate the joint working towards common and
agreed goals. Many researchers were working in relative isolation within their own countries and COBRA has allowed them to work with a cohort of like-minded and experienced researchers in this area to further the science and to offer support. Over the lifetime of the project new scientists have been trained via PhDs or post-docs associated with the work. These new scientists have first hand knowledge of the work of the project but also have made personal contacts with a wide range of other researchers spread across Europe that they will be able to draw on for the rest of their career increasing the efficiency and performance of the European Research Area.

- **Official Authorities:** The work within COBRA has allowed the researchers to continue to engage and work with the official authorities within their own countries to further discuss seed legislation (and in particular the current Marketing Experiment of populations). Although COBRA has worked hard to engage a range of stakeholders during its relatively short timeframe the major impacts for its work will be seen in the years that follow. The new germplasm, protocols, breeding methods/approaches will take time to be ‘road tested’ within the industry before they become accepted or rejected.

5. **Added value of the transnational cooperation in relation to the subject**

COBRA’s strength is its focus on coordinating, linking and expanding ongoing organic breeding activities in cereals and grain legumes across Europe, drawing together experts from previously fragmented areas. In particular, links are established through transnationally sharing germplasm, data, methodological knowledge and discussing results and implications in a diverse consortium. Access to genetic diversity is crucial to enable adaptability of the European crop industry; COBRA will provide long-lasting added value to member states after the project’s lifespan by providing researchers and industry partners with greater access to plant diversity, plant breeding innovations and know-how.

COBRA’s approach focuses on the potential of plant material with high genetic diversity. This approach provides transnationally relevant added value by developing and providing access to plant material with the ability to respond to future stresses through genetic adaptation. Innovation and progress made in previously and current funded EU projects (e.g. SOLIBAM, BioBreed, DIVERSIFOOD, OSCAR) is being added to by the transnational sharing within the COBRA consortium. The project enables formal data exchange between EU member states and provides the added value of building relationships between previously disjunct researchers, industry partners, and farmers. This is creating synergies which have facilitated direct and immediate dissemination that will continue beyond the project’s lifespan, and unite experts across the EU for future collaborations.

**ANNEX 1: CHANGES IN WORK PLAN AND PROBLEMS ENCOUNTERED**

**Changes in consortium and work plan**

The project co-ordinator Dr Robbie Girling left his position at the ORC at the end of March 2015 to take up a position at Reading University, UK. Therefore, the coordination of the project has been passed back to the Deputy Director of the Organic Research Centre, Dr Bruce Pearce who had held the reigns of the project between Dr Thomas Döring leaving ORC and Dr Girling starting. This transition between coordinators has been conducted in discussions with CORE II and ORC’s national funders (Defra), who have been extremely supportive.

**Problems encountered delays and corrective actions planned or taken, if any:**

The completion of this final report has been delayed largely due to over commitment of ORC staff leading the project and getting financial and output information from the consortium members. The final report is now in draft and a final push will be undertaken with all consortium members to get full financial and output information.
ANNEX 2: COST OVERVIEW AND DEVIATIONS FROM BUDGET

Project budget and costs in € (if in National currencies, please indicate)

<table>
<thead>
<tr>
<th>Partner no.</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
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</thead>
<tbody>
<tr>
<td>TOTAL BUDGET</td>
<td>£148734</td>
<td>€150000</td>
<td>37300 DKK</td>
<td>£160000</td>
<td>€69993</td>
<td>960000 DKK</td>
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* By April 1, 2016 330 722 NOK of funding, comprising about 36 750 Euro, remains from RCN for COBRA work. The reason is sick leave during part of 2015. Norwegian partners have agreed with RCN to use this funding during the rest of 2016 for additional root studies on seed samples received from COBRA partners, and for publishing the results in cooperation with Nils-Ove Bertholdsson and others. Additionally, output from the project will be disseminated for Norwegian stakeholders, such as newspaper and website articles.

Total budget is 1 600 000 NOKJ form Research Council + 256 000 own financing.

Total PM spent at 31/03/2016: 1 454 278

Of this – spent at Mid-term: 563 265 NKK and 2nd period 891 013 NKK

1 By Partner 29
2 Partner 16 JNK Plant Breeding withdrew after 1st year.

3 Due to increases in personnel costs the budget was adjusted in November 2015. Original budget was €155,758

4 Includes 8015 DKK own contribution.

5 Sejet Plant Breeding, partner 22, only had a budget for travel costs in relation to meetings. But finally we did not succeed to join the meetings due to other tasks.
Person months (PM) spent on the project:

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\footnote{12} Total budget was 572,307 DKK. From this 372,000 DKK was to be financed by GUDP (Core Organic II). The rest 200,307 DKK was own financing.

\footnote{13} The Slovenian partners: 15 – ITR and partner 29 - University of Maribor used one budget according to one agreement from Slovenian Ministry coordinator by partner 29. From the total sum ITR (partner 15) got 34.74 % of the budget. On request of partners their data has been merged.

\footnote{14} The remaining ca. 3 months will be used to complete root hair measurements on plant material produced at Tingvoll this spring, and write a scientific paper together with Nils-Ove Bertholdsson et al. As you are informed of, RCH has accepted that we postponed this work due to sick leave in 2015, and it will be completed during 2016.

\footnote{15} See 29

\footnote{16} Partner 16 JNK Plant Breeding withdrew after 1st year.
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**Reasons for major deviations in spending compared to original budget:**

Comments are contained within the footnotes of the previous and this page.

Sejet Plant Breeding, partner 22, only had a budget for travel costs in relation to meetings. But did not succeed to join the meetings due to other tasks.

**ANNEX 3: RECOMMENDATIONS TO THE CORE ORGANIC CONSORTIUM IN RELATION TO LAUNCHING AND MONITORING OF FUTURE TRANSNATIONALLY FUNDED RESEARCH PROJECTS**

(Max ½ page)

Most of the recommendations to the CORE Organic Consortium have already been communicated and discussed throughout the life of COBRA. The size of COBRA was one of its real benefits but also a risk. The coordination focus of COBRA was a great success. Bringing together what were small and sometimes disparate national activities and allowing researchers and breeders to work and communicate across the continent was a success and I would recommend that coordination activities be continued with future programme, as the sum of the whole is clearly greater than the sum of the parts.

ORC took on the role of leading and coordinating the project and we understood the concerns from CORE Organic at the beginning that COBRA was such a large project. The challenge was also complicated by senior staff changes within ORC. CORE Organic have already implemented a maximum size of a project which I understand and in many way agree with as the management of such a large project is a very big task and the financial commitment from any one national funder can easily be completely used in management rather than under taking science. However, if this can be overcome and there is a clear rational for such a large project in the future I hope CORE would look at ways to allow such a project to be given life as two smaller projects would not have allowed so many partners to share and coordinate their work and build new relationships that will outlive COBRA.

---

17 The Slovenian partners: 15 – ITR and partner 29 - University of Maribor used one budget according to one agreement from Slovenian Ministry coordinator by partner 29. From the total sum ITR (partner 15) got 34.74 % of the budget. On request of partners their data has been merged.