COBRA

Coordinating Organic plant BReeding Activities – for diversity

Program and abstracts for the COBRA Final Conference 24th and 25th November 2015 at Vingsted hotel & conferencecentre, Denmark

Financial support for this project is provided by funding bodies within the FP7 ERA-Net CORE Organic II

CORE organic II

Coordinating Organic Plant Breeding Activities for Diversity

C O B R A

Organic plant breeding

Breeding of plant material adapted for organic agriculture is important in order to cope with stresses such as climate change, weeds and seed borne diseases. Conventional varieties may not meet the specific needs of organic agriculture. The use of plant material adapted to conditions of organic agriculture will have a positive effect on the productivity and sustainability of organic crop production.

COBRA (Coordinating Organic plant Breeding Activities for Diversity) is a European research project which aims to unleash the potential of plant genetic diversity for organic agriculture by linking up efforts on both pure line breeding and High genetic Diversity (Hi-D) systems in cereals and grain legumes. The COBRA project is part of the CORE Organic II program and is led by the Organic Research Centre (UK). It started in March 2013 and brings together 42 partner organizations from 18 countries. COBRA focuses on four major arable crops: wheat, barley, pea and faba bean.

The project deals with

- seed health
- response of crops to multiple stresses
- improvements in breeding efficiency for organic systems
- structural issues such as funding for breeding and the regulatory framework
- networking and coordination

The COBRA final conference takes place on 24th and 25th November in connection with the Danish Organic Congress. International experts from 16 different countries will attend the conference. On 25th November the international experts will take part in sessions about organic plant breeding at the Danish Organic Congress and this will be open to a broad audience.

The sessions at the Danish Organic Congress will introduce organic plant breeding to the listeners (session A6), and examples of breeding activities will be presented (session B6). Policy issues and farmers rights will be the subject of session C9.

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Program

24th November: COBRA final conference

12.00 – 13.00 Registration and lunch

13.00 – 13.15 Welcome by Bruce Pearce

Seed Health and quality (WP1)

13.15 – 13.30 Anders Borgen: Summary of achievements in the project period and future perspectives
"Manipulating protein content in diverse populations using NIRS single seed sorting"

13.30 - 13.45 Berta Killermann: "Screening of wheat cross material and breeding lines regarding susceptibility for dwarf bunt on naturally infested and ecologically farmed fields"

13.45 – 14.00 Almuth Elise Müllner: "Mapping bunt resistance in winter wheat"

14.00 – 14.15 Johannes Ravn Jørgensen: "Detection of fusarium in wheat by multispectral Imaging"

14.15 – 14.45 **Poster presentations:**

Mara Bleidere: "Results of evaluation of spring barley and winter wheat genotypes for resistance against loose smut and common bunt"

Linda Legzdina: "Testing for resistance to seed borne diseases in various spring barley genotypes"

Linda Legzdina: "Yield stability and tolerance to seed borne diseases of various spring barley genotypes under Baltic climatic conditions"

Franci Bavec: "The use of image-spectroscopy technology as a diagnostic method for seed health tests and variety identification"

Jalli Marja: "Nutrient use efficacy and resistance to seed borne diseases in European spring barley cultivars and landraces"

Anders Borgen: "Virulence pattern in Danish races of common bunt"

14.45 – 15.15 Coffee break

24th November: COBRA final conference

continued…

Breeding for resilience (WP2)

15.15 – 15.45 Maria Finckh: Summary of achievements in the project period and future perspectives

15.45 – 16.05 Rikke Bagger Jørgensen "Adaptation of spring barley for extreme climates"

16.05 – 16.20 Nils-Ove Bertholdsson: "Weed competitive ability and yield stability of winter- and spring wheat populations, cultivar mixtures and cultivars"

16.20 – 16.50 **Poster presentations:**

Bruce Pearce: "Winter wheat variety performance in organic and conventional farming systems and the potential for organic plant breeding in the UK"

Silva Grobelnik Mlakar: "Adaptation of specific winter wheat genotypes (CCPs) to the Slovenian climate"

Martina Robačer: "Adaptation of 10 barley genotypes to the Slovenian climate"

Manfred Jakop: "Ranking the most resistant winter wheat varieties from the common Introductional progamme of organic farming in Slovenian climate"

Karel Dewaele: "Performance of winter wheat CCP's in comparison to reference varieties in organic field trials in Belgium"

Riccardo Bocci: "Using populations in organic agriculture: the role of local adaptation, farmers' selection and the awareness of the actors involved"

Improving breeding efficiency (WP3)

16.50 – 17.10 Peter Baresel: Summary of achievements in the project period and future perspectives

17.10 – 17.20 **Poster presentations:**

Isabelle Goldringer: "Agronomic evaluation of the first population–varieties developed within the wheat participatory breeding program in France"

Isabelle Goldringer on behalf of Veronique Chable: "On farm breeding strategies for diversity in winter soft wheat"

24th November: COBRA final conference

continued…

Socio-economics and legislation (WP4)

17.20 – 17.40 Tove Mariegaard Pedersen and Riccardo Bocci: Summary of achievements in the project period and future perspectives

17.40 – 17.50 Regine Andersen: Seed Legislation in the context of Plant Breeding for Diversity in Organic Agriculture

Poster presentations:

Isabelle Goldringer on behalf of Frederic Rey: “Organic seeds and plant breeding from the seed companies’ perspective”

Dissemination and networking (WP5)

17.50 – 18.00 Riccardo Bocci: Summary of achievements in the project period and future perspectives

18.00 – 19.00 Dinner

19.30 – 21.00 COBRA work package meetings, - this part of the program is for more practical purposes and discussions and will be organized by work package leaders.

25th November: COBRA, grain legume workshop

9.00 – 9.15 Introduction (Maria Finckh)

9.15 – 9.30 Paolo Annicchiarico: “Pea breeding for organic systems. II. Genomic selection for higher grain yield“

9.30 – 9.45 Steffi Zimmer: “Suitability as protein-rich animal fodder and previous crop value of different grain legume cropping systems in organic agriculture in Luxembourg”

9.45 – 10.00 Alev Kir: “Screening Advanced Lines of Soybean (Glycine max L.) under Organic Management in Turkey”

10.00 – 10.30 **Poster presentations:**

Martina Bavec: “Genetic diversity - Assessment of Vicia faba accessions from the Slovenian plant gene bank”

Silva Grobelnik Mlakar: “Allelopathy of barley plant on in vitro pea emergence”

Evelyne Stoll: “Suitability of different grain legume cropping systems as protein rich animal fodder in an ON-FARM field trial under organic growing conditions in Luxembourg”

Karel Dewaele: “Organic field trials of winter and summer protein crop associations in Belgium”

Luciano Pecetti: “Effect of seed treatments with essential oils on plant emergence of ascochyta-infected pea”

Aina Kokare: “Comparison of pea genotypes under organic growing conditions”

10.30 – 11.00 Discussions and future perspectives.

25th November: COBRA sessions at Danish Organic Congress

11.30 – 13.00 Organic and High Diversity Plant Breeding

IFOAM has outlined principles for organic plant breeding; the main principles will be presented, as will different possible pathways to new varieties for use in organic farming.
Organic producers need crops that can cope with multiple stresses such as weeds, pathogens and environmental stresses. The use of populations of specific species has gained significant interest as a tool to breed for increased resilience and tolerance for climate variability. This breeding method favours the forces of natural selection, and farmers are enabled to take part in the breeding process.

Key questions

- What is the difference between organic breeding and breeding for organic farming?
- What is the definition of a population, and what is the difference between a population and a variety mixture?
- How can populations be beneficial in organic and low input systems?
- Is evolutionary breeding an applicable tool in organic plant production?

Principles and pathways in Breeding for organic farming
PhD Edwin Nuijten, Senior Researcher, Louis Bolk, The Netherlands

The agronomic potential of populations
MSc Organic Agriculture Odette Weedon, University of Kassel, Germany

On farm management of diversity and participatory plant breeding
Dr. Isabelle Goldringer, Head of research team Diversity, Evolution and Adaptation of Populations (DEAP), French National Institute for Agricultural Research - INRA, France

Chairman
Professor Dr. Maria R. Finckh, University of Kassel, Germany

25th November: COBRA sessions at Danish Organic Congress

continued..

14.00 – 15.30 Breeding of Cereals and Grain Legumes for Organic Farming

Breeding for organic farming can range from evolutionary and participatory plant breeding of diverse plant material to breeding of varieties in conventional breeding programs. Here you will be presented with different examples of breeding activities all targeted for organic farming: Cycling populations of wheat in different countries, participatory breeding of pea in Italy and breeding activities for organic farming in the Danish breeding companies.

Key questions

- What are the specific needs for varieties in organic production systems?
- Is it possible to have high yielding varieties which at the same time are adapted to the growth conditions of organic farming?
- Is participatory plant breeding an option for e.g. Danish farmers?

Observations in Cycling populations of wheat
MSc Organic Agriculture Odette Weedon, University of Kassel, Germany

Evolutionary and participatory breeding approaches in Pea Breeding for Organic Systems
*Paolo Annicchiarico, Centro di Ricerca per le Produzioni Foraggere e Lattiero-Casearie (CRA-FLC)**,** Italy*

Breeding of cereals for organic farming in Danish Breeding Companies.
Barley breeder Lene Krusell, Sejet Plant Breeding, Denmark

Chairman
Organic plant breeder Anders Borgen, Agrologica, Denmark

25th November: COBRA sessions at Danish Organic Congress

continued..

16.15 – 17.45 Farmers Rights and Policy issues in the context of Plant Breeding for Diversity

Plant breeding and marketing is tightly regulated in order to protect both breeders and farmer’s interests regarding pure line breeding. Breeding for higher diversity on the other hand has met legal hurdles within the EU, since diverse plant material cannot meet requirements of standard variety testing. Within the EU a trial period for marketing populations of some cereals has been implemented and examples are given from different countries.
The session will also offer insight into Farmers Rights according to international treaties and other commitments.

Key questions

- Can a farmer legally sell or purchase a population or other diverse plant material?
- Is it possible to protect the farmer’s interests when purchasing a population?
- What are the Farmer’s Rights in the context of breeding for higher diversity?

Farmers Rights - Seed Legislation in the context of Plant breeding for Diversity in Organic Agriculture
Dr. Regine Andersen, Executive Director Oikos – Organic Norway, Norway

Marketing of Populations
Dr. Bruce Pearce, Deputy Director, Organic Research Centre, United Kingdom

Chairman
Riccardo Bocci, Italian Association for Organic Agriculture - AIAB, Italy

COBRA, list of participants (COBRA partners)

Aina Kokare	State Priekuli Plant breeding Institute	Latvia
Alev Kir	Aegean Agriculture Research Institute (AARI)	Turkey
Almuth Elise Müllner	BOKU Vienna	Austria
Anders Borgen	Agrologica	Denmark
Anne Ingver	Estonian Crop Research Institute	Estonia
Anne-Kristin Løes	Norwegian Institute for Bioeconomy Research, NIBIO	Norway
Berta Killermann	Bayerische Landesanstalt für Landwirtschaft	Germany
Bruce Pearce	The Organic Research Centre	United Kingdom
Edwin Nuijten	Louis Bolk Instituut	Netherlands
Evelyne Stoll	Institute for Organic Agriculture Luxembourg	Luxembourg
Franci Bavec	University of Maribor, Faculty of Agriculture and Life Sci.	Slovenia
Frederic Rey	ITAB	France
Hans Haldrup	Nordic Seed	Denmark
Ilmar Tamm	Estonian Crop Research Institute	Estonia
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Johannes Ravn Jørgensen	Aarhus University	Denmark
Kaija Hakala	Natural Resources Institute Finland (Luke)	Finland
Karel Dewaele	Inagro vzw	Belgium
Kevin Dewitte	Ghent University	Belgium
Lene Krusell	Sejet Plant Breeding	Denmark
Lieven Delanote	Inagro vzw	Belgium
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Maria Finckh	University of Kassel	Germany
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Martina Robačer	University of Maribor, Faculty of Agriculture and Life Sci.	Slovenia
Matthew Heaton	DEFRA	United Kingdom

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Odette Weedon	University of Kassel	Germany
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Regine Andersen	Oikos - Organic Norway	Norway
Reine Koppel	Estonian Crop Research Insitute	Estonia
Riccardo Bocci	AIAB	Italy
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Robin Walker	SRUC	United Kingdom
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Veronique Chable	INRA	France
Ülle Tamm	Estonian Crop Research Institute	Estonia

Improving quality and health in diverse populations of wheat

By:
Anders Borgen, Agrologica, Houvej 55, 9550 Mariager, Denmark E-mail: borgen@agrologica.dk

Introduction

Biodiversity is one of the keystones in the concept of organic agriculture, both as a goal in it self, and as a mean to control pests and diseases, and to optimal exploitation of resources. On the other hand, monocultures have advantages in producing a homogeneous crop easy to harvest and process. In cereal production, composite cross populations have been proposed as a compromise between the benefits of biodiversity and monoculture fit for organic farming (Döring *et al.* 2011).

A change from pure line pedigree breeding to population breeding calls for at new set of breeding tools to develop optimal quality and yield compatible with pure line varieties. The aim of this work is to test different population breeding methods, where unfit germplasm are discarded while still maintaining some degree of diversity of adapted plants.

Common bunt (*Tilletia caries*) is a devastating pseudo seed borne disease of wheat intensively controlled by fungicide seed treatment in conventional agriculture. In organic agriculture, control measure are limited to mechanical treatments such as brush cleaning (Borgen 2015) or heat treatments (Kristen and Borgen 2001, Borgen *et al* 2005), application of natural compounds like acetic acid (Borgen and Nielsen 2001), mustard, milkpowder, Tillekur or bio-agents like Cedomon (Borgen and Kristensen 2001,Borgen and Davanlou 2001). Therefore, control of common bunt in organic agriculture must follow an integrated strategy including several different measures including also genetic resistance. Measures for breeding pure line varieties for resistance to plant diseases are well known, but these cannot be used in population, at least not without adjustments.

Common bunt drastically reduce seed production in infected plants, and natural selection will therefore theoretically reduce susceptible lines within a population. However, a quantification of this effect has not previous been made.

Protein content is an important quality parameter in cereals whether used for baking or feed. It has previous been shown that it is indeed possible to increase protein content in populations by single seed sorting based on Near Infrared Reflection (Borgen *et al.* 2014). However, seed in this trial were sorted and sown directly. Seed sorting will affect not only the genetic composition of the seed, but possibly also the vigour of the seed, potentially affecting the yield and quality of the harvested grain. The longer term genetic effect of NIR seed sorting is therefore still a bit unclear.

Materials and methods

In 2008, 40 crosses were made between 20 parents of winter wheat, and in 2008 another 200 crosses were made between these and additional 11 parents. Each cross were propagated separately. In 2010 samples from each of the crosses from 2008 were merged, and inoculated with spores of common bunt (*Tilletia caries*). Each year onwards, the population were threshed and sown without seed treatment, allowing the pathogen to multiply naturally within the population (Steffan 2014). In 2011, samples of all crosses were merged, following the same procedure. In 2012, each individual cross population were inoculated with bunt spores, and grown separately with bunt in 1m^2 plots maintaining their own pathogen population. In crosses without infected plants to maintain selection pressure, new spores were added annually.

Each of the parents were in 2013 and 2014 grown in separate micro plots, inoculated both with a blend of new spore, and with propagation of their own spores within each variety (Borgen 2015). The frequency of infected heads were recoded based on macro symptoms in both pure line parents and the populations.

All winter wheat varieties selected as parents had in one or more publications been recorded as resistant to bunt.

A British CCP (ORC Wakelyns CCP) well described in other papers of this publication was included in 2013 with infection of bunt, and multiplied for one year with infection. The same population had also been grown in Hungary, where it had been naturally infested with bunt.

In 2008, 15 crosses were made between 13 spring wheat varieties, of which 5 had purple bran. Each cross were grown separately each year onwards in 1m^2 plots. Each year, crosses with a purple wheat parent were colour sorted, discarding light kernels. In 2013, each cross were sorted into 3 fractions based on protein on each kernel using a BoMill IQ Grain Quality Sorter 1002. The highest and lowest protein fraction each contained 20% of the seed sample. Only the highest and the lowest protein fraction was grown. In 2014, each sample was again sorted in the same way, but only the highest fraction were sown from the high protein fraction seed, and the low fraction of the low fraction seed. All seed from each fraction were sown increasing plot size to 3-15m^2.

Results and discussion

<u>Bunt in winter wheat</u>

Infection level of bunt in the populations seems to be stable from year to year in both the narrow population with 40 crosses, and the broader CCP with 220 crosses. It did increase the last three years, but whether this is a trend or a case of climatic fluctuation is unclear. In the wheat populations, the pathogen population was propagated within the wheat population allowed it to adapt to the genetic composition of the wheat. Compared with the pure lines grown in the same way, the bunt infection level was much lower, comparable with pure lines grown with new spores each year. I think this means that despite a stable infection level, resistance may increases in the populations from year to year, but in the same time, the virulence of the pathogen increases and eliminate this effect.

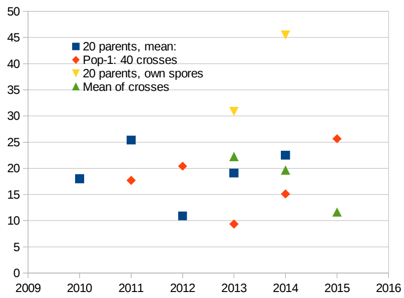

Figure 1: Common bunt in CCPs, single cross populations and in pure lines

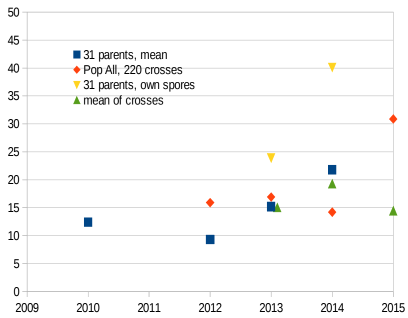

Figure 2: Common bunt in CCPs, single cross populations and in pure lines

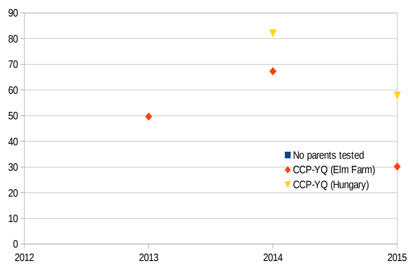

Figure 3: Common bunt in ORC Wakelyns CCP

The ORC Wakelyns CCP had a much higher infection level as my own CCP, and the CCP grown in Hungary was even higher. This can be explained by the fact that the parents of the ORC Wakelyns CCP were not selected for their resistance to bunt, leading to a more susceptible CCP.

Most of the parents were resistant to some virulence races of bunt, but susceptible to others (Borgen 2015). Crosses with parents with resistance to all tested virulence races had a stable low infection level over the three years (<10%), while crosses where parents were susceptible to some virulence races had higher and in many cases increasing infection level over the years. This indicates that virulence had build up within the individual populations. The average infection level between the individual crossings was comparable with the the infection level in the CCP of the same parents indicating that no synergistic effect was achieved by the higher diversity of the CCPs.

Protein content in spring wheat

After two generations of seed sorting, the protein content of the high protein fraction was higher than the content in the low protein fraction (Borgen *et al* 2014). Growing the same fraction without further seed sorting maintained the differences in protein content, meaning that the achieved differences was indeed heritable traits as shown in Figure 4. The protein content was in average 11.5% in the samples grown from the low protein fraction seed, and 12.3% in the high protein fraction. The yield on the other hand decreased from 5674 to 5252 kg/ha respectably.

The experiment shows that it is possible with automatic single seed sorting to increase the protein content in populations, but that this (like it is known from pure line varieties) is on the expense of a yield loss of about 10% for each protein content increase.

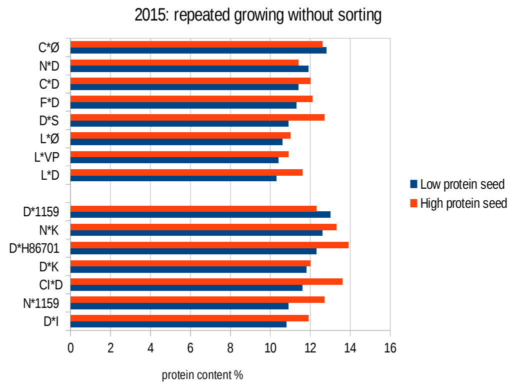

Figure 4: Protein content in cross populations of spring wheat

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Screening of wheat cross material and breeding lines regarding susceptibility for dwarf bunt on naturally infested and ecologically farmed fields

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Introduction

In ecological farming, dwarf bunt of wheat (Tilletia controversa) is a serious disease that can cause high damage in wheat production. In the case of a heavy infestation, the harvested crop can be used for neither food nor feed and additionally the soil is strongly infested with bunt spores for several years. As part of the EU project COBRA, in a two years field trial breeding lines and varieties of winter wheat were tested under practical conditions for their susceptibility against dwarf bunt of wheat. Results of the first year are presented hereafter.

Material and methods

In 2013/14, 40 winter wheat varieties and breeding lines were tested for their susceptibility to dwarf bunt infection, among them the standard set of differential wheat cultivars according to Goates (1996, 2012) with the resistance genes Bt1-Bt7 and Bt10-Bt13. The test site was a field certified according to ecological accreditation at Wolfersdorf in Upper Bavaria, Germany. Bunt spore potential in soil at the test sites was of natural origin and determined according to the method described by Bauer et al. (2014). The average concentration of bunt spores in soil was approximately 3,500 bunt spores/10 g.
The wheat lines and varieties were grown in two replications in single rows of ten meters length each with 25 cm row spacing and 600 kernels per row.
Susceptibility to infection with dwarf bunt was evaluated by counting the number of infected ears per row and the number of spores per kernel according to ISTA (Working sheet No. 53).

Results and discussions

Field trials for resistance against dwarf bunt of wheat often present difficulties because of the special weather conditions the pathogen requires, e.g. low temperatures without freezing, high humidity, and light, as they can be found under snow cover for a sufficient length of time. In the case of an adequately high natural bunt spore potential in soil, representative results can be achieved under practical conditions.

Results realized by other research workers regarding resistance as well as susceptibility to dwarf bunt infection could be confirmed (Fig. 1). Lines and varieties that showed efficient resistance to infection with dwarf bunt earlier exhibited no or almost no symptoms in the first year of the field trial, while the known susceptibility of certain varieties could be likewise confirmed (Bürstmayr and Huss 2012, Huber and Buerstmayr 2006). On the basis of the infection results obtained through the standard set of differential cultivars a specific virulence pattern of the pathogen resulted, making it possible to produce a preliminary characterization of the pathogenic race occuring at the test site.

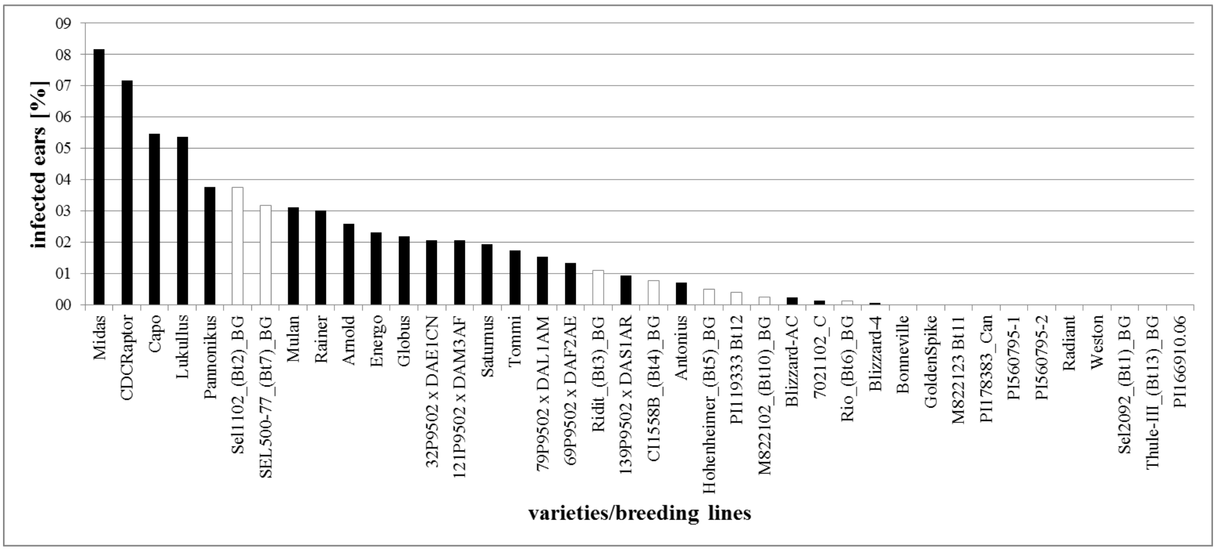

Fig 1: **Percentage of infected ears, one year results**

Acknowledgements

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Mapping bunt resistance in winter wheat

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Introduction

During the last two decades, bunt diseases have re-emerged in organic winter wheat throughout Europe. Currently, no bunt resistant and adapted varieties are available for organic farming in Austria. Whereas the predominantly seed-borne disease common bunt (*Tilletia tritici, Tilletia laevis*) can be managed by careful seed hygiene, control of the soil-borne dwarf bunt (*Tilletia controversa*) is more challenging: Dwarf bunt teliospores are long-lived and once contaminated, soils cannot be used for organic winter wheat production for a minimum of 10 years. Host resistance therefore constitutes the most important tool for bunt control in organic farming. Breeding for bunt resistance is time and cost intensive and molecular marker technology could greatly facilitate the development of resistant winter wheat varieties. Knowledge regarding the genetic basis of bunt resistance, however, is limited and little is known about the interaction between dwarf and common bunt resistance. Monogenic, race specific bunt resistance in winter wheat is conferred by 13 to date identified bunt resistance genes Bt1 to Bt13, of which seven have been allocated to individual wheat chromosomes. Molecular markers are available for Bt10 only. Quantitative, non-race specific bunt resistance has also been described in winter wheat.

Materials and methods

6 mapping populations of 100-120 recombinant inbred lines (RIL) were developed based on crosses between bunt resistant exotic material (Turkish landraces and North American cultivars) and susceptible but adapted winter wheat cultivars. These RIL populations were tested for bunt resistance in field trials (artificial inoculation) at multiple sites in 2014 and 2015 in Austria and Utah (USA); the third – and last – year of testing will take place in 2016. Parental lines were tested at three additional sites in Czech Republic, Germany and Sweden. Based on the segregation pattern of the resistance trait, 3 populations were selected for genotyping using the SNP chip technology.

Results and discussion

Resistant parental lines, representing monogenic as well as complex sources of resistance, conferred durable protection against a broad spectrum of bunt races across locations and years. Mapping populations were scored for common bunt resistance in 2014 and 2015. In contrast to common bunt, dwarf bunt disease development depends on favourable weather conditions during winter: multiple years of testing are therefore required to assess the resistance phenotype. In 2014, dwarf bunt infection levels were too low; in 2015 informative results were collected for the mapping populations. The most promising populations with regard to differentiation of the resistance trait will be genotyped with genome-wide SNP markers in order to map bunt resistance loci. In 2016, mapping populations will be re-evaluated for bunt resistance in field trials; the combined statistical analysis of phenotypic and genotypic data will identify genes (QTL) for durable bunt resistance in winter wheat and enable the development of molecular markers for application in organic wheat breeding.

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Detection of fusarium in wheat by multispectral Imaging

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Introduction

Wheat (*Triticum aestivum*) seeds are infected by a high number of fungi, including pathogens coursing head blight, such as *Fusarium graminearum*, *F. culmorum*, *F. poae*, *F. avenaceum* and *Microdochium nivale* (Stepien and Chelkowski 2010). *Fusarium spp*. is a widely distributed fungus causing yield reduction in wheat and other cereals. Many *Fusarium* species produce mycotoxins responsible for serious quality deterioration. Wheat produced by organic, low-input and conventional farming are all affected. Breeding for resistance to head blight in wheat is a non-trivial task for plant breeders to develop, and dependent on appropriate selection tools that allow breeders to identify the desired genotypes (reliable testing methods) (Buerstmayr, Buerstmayr et al. 2014). In the breeding program disease symptoms are scored visual in the field and also postharvest by visual scoring the percentage of diseased seeds in harvested samples.

Multispectral imaging provides fast non-destructive information from seed images, which has already been proposed as a useful tool to determine the presence of fungal spores on seeds and for phenotyping (Olesen, Carstensen et al. 2011, Hansen, Hay et al. 2015, Shrestha, Deleuran et al. 2015).

Materials and methods

To evaluate the application of multispectral seed imaging as a tool for scoring of the percentage of diseased seeds, we took seeds from 21 wheat cultivars inoculated with *F. culmorum* as well as healthy reference samples of the cultivars provided by Herman Buerstmayr, BOKU, from year 2014 harvest. We used the multispectral imaging system, VideometerLab 3, an instrument equipped with 19 light emitting diodes at wavelengths ranging from 375 to 970nm (ultraviolet, visual and near-infrared light) where the object is sequentially LED illuminated and image captured at each wavelength band.

Seed samples were divided into two set; calibration set and test set. Calibration set consisted of visually scored 352 infected (by *F. culmorum*) seeds and 120 healthy seeds from the 21 wheat cultivars. Similarly test set consisted of two seed samples with each sample containing at least 250 seeds from the 21 cultivars; one visually scored as 0% infection and other with visual score which ranged from 15 to 100%. Wheat seeds were placed in a petri dish on top of a blue background and subsequently multispectral images from dorsal and ventral side of seeds were captured using VideometerLab vision system. The captured images contained the information from seeds (region of interest) and blue background (noise). So each image was masked using default feature ‘Blue background with object’ of Videometerlab ver 3.0 software to acquire region of interest i.e. image of seed for further analysis. A normalized canonical discriminant analysis (nCDA) based multispectral imaging (MSI) transformations were calculated using the images of infected and healthy seeds from calibration set. The nCDA is a supervised classification method, which minimizes the distance to observations within classes and maximizes the distance to observations between classes. All seed images were collected into a blob database of the VideometerLab ver 3.0 software, where each blob was a representation of one seed. Different features from blob toolbox were extracted and calculated for discrimination of infected and healthy seeds. The features included were RegionMSImean, intensity (mean pixel intensity of the image), hue, saturation, CIELab L *, CIELab a * and CIELab b *. RegionMSImean calculates a trimmed mean of nCDA-transformed pixel values within the blob and were calculated using MSI transformation developed using the infected and healthy seeds from calibration set. Among the several features, RegionMSImean gave the best discrimination between the infected and healthy seeds. Threshold value of -0.5 on an arbitrary scale was used to classify the infected seeds. Any seed RegionMSImean values greater than -0.5 were regarded as infected seeds. The accuracy for healthy seeds was 95% whereas 87% of the infected seeds were correctly identified by the model.

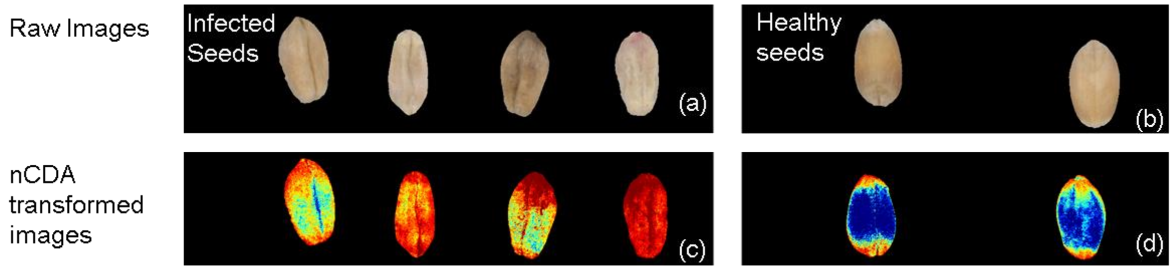

Figure 1. RGB images of wheat seeds (a) infected and (b) healthy seeds and nCDA transformed images of (c) infected and (d) healthy seeds

Results

The RegionMSImean values were calculated for the test set and were predicted using prior determined threshold value of -0.5. The model showed relatively little infection in the seed samples ranging from 8-25% in most of the seeds samples with 0% visually scored samples. However, few samples were predicted up to 63% of infection in 0% visually scored samples. The model predicted more than 95% of infection in the seed samples visually scored more than 35% of infection. The seed sample with 15% visual score was predicted with 84% of the infection. In average the model predicted 96% of the seeds in the infected test set to be infected whereas the average visual score was 61%. Overall the model estimated 60% of the presented seeds to be infected while only 31% of the seeds were visually scored to be infected.

Discussion and conclusion

The high positive scoring of infected seeds compared to the visual scoring could be due to that the imaging system is a quantitative measurement of infected seeds, whereas the visual scoring is a subjective scoring of significance of the infection. Likewise could the positive estimation of infected seeds found in the expected healthy lot be due to unregistered infections. A further evaluation could be carried out by microscopically evaluation for fungal infection of suspected seeds. However use of spectral imaging as a reliable tool for evaluation of fusarium resistance in breeding program is a promising tool but still to be proven. Based on our experience from this study we have in an attempt to make a more sensitive model selected 96 wheat seeds suspected to be infected with *fusarium* ssp. These are analysed by qPCR in order to detect the actual pathogens on the seeds. Using them in building a new model will lead to a more exact measurement of the number seeds with specific *fusarium* pathogens on the tested seeds.

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Results of evaluation of spring barley and winter wheat genotypes for resistance against loose smut and common bunt

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Introduction

The increasing drive towards sustainable agriculture, including organic production has increasingly leaded to the sowing of untreated seed. If no seed treatments are used among seed born diseases, loose smut (*Ustilago nuda*) in barley and common bunt in winter wheat (*Tilletia tritici*) are expected to cause one of the biggest problems in Latvia. Especially common bunt in wheat is subject to great concern.

A main problem for organic seed producers is that organically produced seeds have to fulfill the same regular phytosanitary requirements like those of conventional origin. For the production of certified seeds not more than five ears infected with smut and bunt for barley and wheat respectively are allowed on an area of 100 square meters in Latvia. In organic farming diseases should be dealt with through prevention rather than treatment. Therefore the use of resistant varieties is an important factor in an integrated strategy to control seed-borne diseases. The first problem is that of securing donors of resistance which might be used in crosses. There are resistance genes available but often we do not know their occurrence in the working collections. This problem could be partly solved by the extensive resistance tests.

Main goal of this study was to screen spring barley and winter wheat genotypes for working collections for their degree of loose smut and common bunt resistance respectively.

Materials and Methods

Research with *Ustilago nuda* in spring barley and *Tilletia tritici* in winter wheat was performed at the State Stende Cereal Breeding Institute during 2013-2015.

Spring barley. To imitate an inoculation under natural conditions by wind provocative background for smut testing was established. Plots highly infected genotypes were interspersed within the plot arrangement in every second plot column. Furthermore, spore-suspension was sprayed all over the field with a backpack sprayer repeatedly. Plot size was 2.0 square meters in two replications. The number of tested genotypes was 235 in 2013 including genotypes from working collection and breeding lines from spring barley breeding programs in Latvia, those genotypes which had no smut infection under natural conditions during previous years. Disease-free genotypes were selected (182 genotypes) for re-examination in 2014, and 81lines in 2015. The number of infected ears per one m^2 was estimated.

Screening for resistance to loose smut also under artificial background carried out for samples from working collection (41 genotypes in 2013; 30 in 2014). Inoculation was performed by vacuum method transferring aqueous spore-suspension in the ears at the beginning of flowering of each variety individually when pollen was green or yellowish green. One gram of dry spores and one liter water were mixed adding 10 g of sugar. Artificially infected ears (5 per plot) were protected by a paper bag and handpicked at harvest. In the next spring the artificially inoculated seeds were sown (rows 2-5). The degree of resistance was recorded as percentage of infected plants.

Winter wheat. For infection with *Ustilago tritici*, one gram of spores per kg of seed was used for inoculation by adding the dry spores to the seeds in a plastic bag and shaking them to disperse the spores.

Estimation of 113 winter wheat varieties against *Tilletia caries* were set up in Stende in 2013. Artificial inoculation by local common bunt spores and sowing: In 2013 113 genotypes (73 samples from Stende's material; 40 samples from the collection of A. Borgen (Denmark) was estimated but in 2014 60 genotypes (27 samples from Stende's material; 33 samples from Denmark/ A. Borgen) were screened. The sowing time was choosing later than optimum in Latvian agro-climatic conditions (28.09 in 2013; 26.09 in 2014) with purpose to provide the highest infection level. Inoculated material was sown in 0.2 to 1 m^2 size plots depending on quantities of seed. The degree of resistance was recorded as percentage of infected ears.

Results

Spring barley. Infection level among susceptible genotypes under provocative background of loose smut varied from 0.5 to 35.5 infected plants per m^2. Estimation results of maximum damage (out of all replications and years) of spring barley genotypes for resistance against loose smut by means of provocative infection showed that out of 235 varieties initially included in this trial 30 accessions did not have any loose smut infected plants during three test years. These selected genotypes repeatedly will be assessed under artificial inoculation loose smut background and could be valuable source of resistance in the future breeding work.

The results show great variation in susceptibility under artificial inoculation background for potential loose smut resistance. Infection level among susceptible genotypes varied from 1.1 to 83.3% of damaged ears. It was detected that germination of artificially infected seeds was negatively affected - on average 70% of plants were emerged with variation between genotypes from 30 to 100%. From 41 accessions included in this trial only two six-row spring barley accessions (Fox, Milton) showed complete resistance under artificial and provocative infection backgrounds. Besides of these varieties with full resistance ten genotypes (Svetlocok, PR 6000, PR 6096, Run 8/ 454, 3- 192, 3- 191) showed low level symptoms when artificially inoculated (damage level below <3%) and at the same time stayed healthy under the condition of natural infection. In this case besides of genetic basis of resistance a number of other factors could affect the development of the loose smut, for instance specific defense reactions which are existed in spring barley could be take into account. The most observed is cleistogamous or closed flowering character that could be as an alternative method of dealing with loose smut by barley breeders.

Winter wheat. In 2014 spring after resumption of vegetation only 46 samples 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%. The following genotypes without infection of *T. caries* were identified: SW Magnific, Stava, Inna, Nemchinovskaya 25, P/ 591981, PG 3540, Pi 554-099(Bt 9), Ridit, Kalininskaya. The results in 2015 showed great variation in susceptibility to bunt. Over all 60 genotypes tested for *T. carries* resistance under artifical background, the classified damage (0; >0–1; >1–5; >5–10; >10-30; >30-60; and >60%) was 18.3, 0, 15.0, 6.7, 20.0, 25.0 and 15% respectively. Within the group '0%' the following genotypes were selected: SW Magnific, Hansel, Inna, Sam, Andrew, Hostop, PI119 333, PI554 098, PI554 119, PI554 118, PI554 106. Varieties SW Magnific and Inna did not have any infected plants during two testing years.

The research results confirmed that currently total resistance from *Ustilago nuda* and *Tilletia tritici* was not expected from any of the present Latvian spring barley and winter wheat variety, and hereafter this must be sought by using as parents those introduced genotypes which had so far proved to be resistant.

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Testing for resistance to seed borne diseases in various spring barley genotypes

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Introduction

Loose smut (*Ustilago nuda*) and leaf stripe (*Pyrenophora graminea*) are the most hazardous seed borne diseases for spring barley (*Hordeum vulgare* L.) which can cause serious problems under organic production system where no chemical seed treatment is used. One of the tasks in breeding program for organic farming is to obtain varieties resistant to those diseases. For that purpose appropriate initial material with information on resistance is needed.

The aim of the research was to test resistance to loose smut and leaf stripe in large amount of spring barley genotypes under provocative background, using artificial inoculation and with molecular markers.

Materials and Methods

Testing was performed during 2013-2015 under conventional farming conditions. 188 genotypes with various origin were grown under provocative background in 2.3 m^2 plots in 2013. A mixture of several genotypes highly infected with loose smut (*Ustilago nuda*) and leaf stripe (*Pyrenophora graminea*) according to previous year data was grown in every second plot. 200 and 165 genotypes were included in the nursery with provocative background in 2014 and in 2015, respectively. The number of infected plants per plot was counted.

Accessions with known resistance genes and the ones with no infected plants found in previous trials were infected in 2014 artificially by entering one drop of local loose smut spore suspension in each flower of three spikes in flowering stage.

RIL population from cross CDC Freedom/ Samson consisting of 96 lines was sown under provocative background in 2013 and 2014 and infected artificially with loose smut in 2013. Canadian two-row hulless barley variety ‘CDC Freedom’ has Un8 gene providing resistance to loose smut according to the information obtained from the breeder, and ‘Samson’ is susceptible six-row covered variety originating from USA. Amplification products of SCAR marker for Un8 gene from Eckstein et al. (2002) were sequenced and co-dominant CAPS markers were designed based on single nucleotide polimorphisms that cause changes in DraI restriction site in resistant genotypes. The lines were genotyped with the markers and the results compared to the phenotyping data.

Results and discussion

In 2014, 168 out of 200 genotypes in the nursery with provocative background were included for 2nd year (seed of those was infected under provocative background in 2013) and the rest was added to the nursery for 1st year. The infection rate with loose smut was not as high as expected; the reason for that can be meteorological conditions during flowering time in 2013 when the infection occurred or during plant development in 2014. 163 accessions did not have infected plants and the rest had 1-7 infected plants (1.8 on average) per plot; the provocative background had on average 2.9 loose smut infected plants per plot. If all available data from 2013 and also from other trials was summarized, 60 accessions did not have any loose smut infected plants.

From the 32 accessions included in the trial because of potential resistance to leaf stripe 13 did not have any infected plants during two testing years. From the whole set of accessions 59 did not have leaf stripe infected plants during two years. In 2014 the susceptible samples were infected in range 1-21 plants per plot (3.5 on average); in provocative background plots there were 12.7 infected plants on average.

There were 25 genotypes without any seed born disease (including covered smut which was also observed in the trial) symptoms during two years testing, however, in 2013 the seed sown did not come from provocative background but from various locations. The data of 2015 has to be added to make final conclusions about resistance sources.

Marker data was fully corresponding to the loose smut occurrence phenotype data (from both provocative background and artificial inoculation) for 74 RIL lines (75.5%), and clear contradictions were found for 17 lines. Six lines in which Un8 gene was identified got loose smut infected plants. It shows that the marker is not fully trustable and the selected perspective breeding lines has to be double-checked by artificial inoculation. 22 RIL lines showed resistance and can be used in further breeding as resistance source.

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Yield stability and tolerance to seed borne diseases of various spring barley genotypes under Baltic climatic conditions

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Introduction

Selection for yield stability is challenge for barley breeding programms as high number of test environments usually exceeds the common capacity of this work (Mühleisen et al., 2014). Varieties suitable for organic and low-input systems should be more adapted to respond to increasingly frequent weather extremes. Seed-borne pathogens such as loose smut (*Ustilago nuda*), leaf stripe (*Pyrenophora graminea*) and net bloch (*Drechslera teres*) represent a major threat to barley establishment, thus should negatively affect grain yield, its stability and quality. In organic farming diseases should be dealt with through prevention rather than treatment and can be mainly controlled by choosing resistant varieties (Borgen, 2004).
In organic production system environmental conditions are known to have significant influence on yield of spring barley. A major challenge is to breed new varieties that are able to cope with increasing environmental variability. The varieties bred for organic farming have to be appropriate for this cultivation system showing high degree of adaptation to different stress factors. The aim of this study was to determine yield stability and infection level with loose smut, leaf stripe and net-bloch of 26 spring barley genotypes grown in organically managed fields in Latvia and Estonia during three seasons.

Materials and Methods

The trial included in total 26 spring barley (*Hordeum vulgare* L.) genotypes comprising Latvian and Estonian varieties and breeding lines with potential adaptability to organic farming, hulless barley accessions arising from a collection of A. Borgen (Denmark) and organically bred German hulless variety ‘Pirona’. Testing was performed during 2013-2014 in locations Priekuļi, Stende (Latvia) and Jõgeva (Estonia) under organic growing conditions and is being continued in 2015. The trials were arranged in three replications in randomized complete block design in 5 – 6.5 m^2 plots. 400 germinating seeds were sown per m^2. The soil was sod-podzolic sandy loam or loamy sand with organic matter varying from 1.7 (Priekuļi 2013) to 3.5% (Stende 2014) and pH from 5.5 (Stende 2013) to 6.7 (Jõgeva 2013). Weed management was performed by harrowing with exception of Priekuļi in 2013 where it was not possible due to rain; in Jõgeva harrowing was done twice per season.
Yield stability analysis was performed according to the joint regression analysis method proposed by Finlay and Wilkinson (1963) and Eberhart and Russell (1966) by calculating, the regression coefficient for the slope (b) of the Finlay-Wilkinson regression line, and variance due to deviation from regression (s^2d) as parameters of adaptability and stability, respectively. Barley plants infected with loose smut, covered smut (*Ustilago hordei*) and leaf stripe were counted in all trial plots and net-bloch was scored according to scale 0-9. In addition some traits related to competitive ability with weeds were estimated.

Results and discussion

The highest average yield level was reached in Jõgeva in both years (4.45 and 3.95 t/ha). Four lines selected in Jõgeva and one line selected in Stende were the highest yielding over all trials (Table). Estonian line 4533.4.3.6 was the most stable with the coefficient of regression close to 1 and a little deviation from regression (s^2d) showing wide adaptability. Line 4628.6.6.3 was the only one with b significantly different from 1 and consequently adaptability to favourable conditions. Slope for the rest of genotypes did not significantly differ from 1 or the significance of it was >0.05. Line PR-5105 selected in Priekuli had yield significantly above the average and a tendency to adaptability to unfavourable conditions. No loose smut and also leaf stripe infected plants were found for the variety 'Rasa' in either location; the yield of it was not significantly different from the average. Three genotypes ('Leeni', PR-5117 and PR-6262) had only one loose smut infected plant; no leaf stripe was found for eight genotypes. The German hulless variety 'Pirona' was highly infected with covered smut. 'Naked barley No.43' had the lowest average infection level with net-blotch.

Table. Yield stability parameters and infection with seed borne diseases of selected barley genotypes over six environments, 2013-2014

Genotype	Breeder/ source	Grain type	Average grain yield, t/ha	b[3]	Difference of b from 1	s^2d[4]	Loose smut, plants[6]	Leaf stripe, plants[6]	Net blotch, score 0-9
4491.3.3.9	Jogeva/EST	CO[1]	4.35*	1.18	b=1	0.21	9	5	4.1
4628.6.6.3	Jogeva/EST	CO	4.29*	1.67	b>1	0.10	67	1	4.8
ST-12955	Stende/LV	CO	4.27*	1.20	b=1	0.12	221	0	4.2
4533.4.3.6	Jogeva/EST	CO	4.26*	1.04	b=1	0.03	6	2	4.0
4439.5.5.2	Jogeva/EST	CO	4.21*	1.30	b=1	0.06	66	0	5.0
PR-5105	Priekuli/LV	CO	4.13*	0.80	b=1	0.09	30	1	4.4
Rubiola	Priekuli/LV	CO	4.02	1.27	b=1	0.13	21	0	4.3
Rasa	Stende/LV	CO	3.91	1.13	b=1	0.12	0	0	4.2
Hora	A.Borgen/DK	HB[2]	3.20	0.82	n.s.[5]	0.18	30	1	4.6
Irbe	Priekuli/LV	HB	3.00	1.21	b=1	0.06	43	2	4.0
Naked b. no. 43	A.Borgen/DK	HB	2.44	0.84	b=1	0.03	53	0	3.4
Average			3.64				32	2	4.5
Range			4.35-2.16	0.45-1.67		0.02-0.52	0-221	0-8	3.4-5.8

[1]covered barley; [2]hulless barley; [3] slope (coefficient of regression); [4] deviation from regression; [5] p>0.05; [6]total number of infected plants in all trials; * significantly above average yield (p=0.05)

According to the data obtained from the six environments the line 4533.4.3.6 can be recognized as the most stable yielding one with relatively low infection with seed borne diseases. No loose smut and leaf stripe infected plants were found for the variety 'Rasa'. However, data of more environments are needed to draw final conclusions.

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The Use of Image-spectroscopy Technology as a Diagnostic Method for Seed Health Tests and Variety Identification

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Introduction

The development of repaid and time efficient health diagnostic and identification technology in breeding programs could increase the availability of new varieties. The aim of the study was to evaluate the potential of multispectral imaging and single kernel near infra-red spectroscopy (SKNIR) for determining seed health and variety separation of winter wheat (*Triticum aestivum* L.) and winter triticale (*Triticosecale* Wittm. & Camus) which could be applied in to conventional as well as organic breeding programs.

Materials and methods

The analysis included twenty-seven winter wheat (*Triticum aestivum*) varieties and nine triticale varieties (*Triticosecale* Wittm. & Camus) for selecting the most resistant varieties under conventional cropping practice where each of the 36 treated varieties also had an untreated control set.
All digital images were captured with a Videometer Lab instrument (Videometer A/S, Hørsholm, Denmark). The multispectral images were captured at 19 different spectral bands from VIS to NIR wavelengths (375nm-970nm). After capturing, the seeds were incubated according to ISTA blotter test [1] as a control set for multispectral analysis. After a week, microscopic observations for determination of seedborne pathogens were conducted [13]. Multispectral data were handled with the Videometer software version 1.6 (Videometer A/S, Hørsholm, Denmark), using a normalized canonic discrimination analysis (nCDA) with all 19 bands. Extracted data were further handled in Excel, where the RegionMSImean values (feature based on an nCDA transformation between varieties image) were correlated to the different varieties. SKNIR was acquired from 900 to 1600 nm using a FT-NIR analyzer (Q-Interline A/S QFAflex 600; Tølløse, Denmark). A principal component analysis was applied after the spectra

preprocessing, in order to find similarities or dissimilarities between species.

Results and discussions

More than half of the evaluated seeds were infected by *Alternaria* spp. and *Fusarium* sp. could be detected on 672 seeds. *F. graminearum*, *F. avanaceum*, *F. poe* were the three most frequently determined *Fusarium* species.
It was possible to demonstrate that multispectral imagining enables the distinguishing between uninfected and with *Fusarium* spp. and *Alternaria* spp. infected parts of seeds.
The use of SKNIR and multispectral imaging were successfully employed just for a few variety separations with currently software available and nCD develop features. These two methods have perspective and an under researched capacity for seed health and variety identification diagnostics.

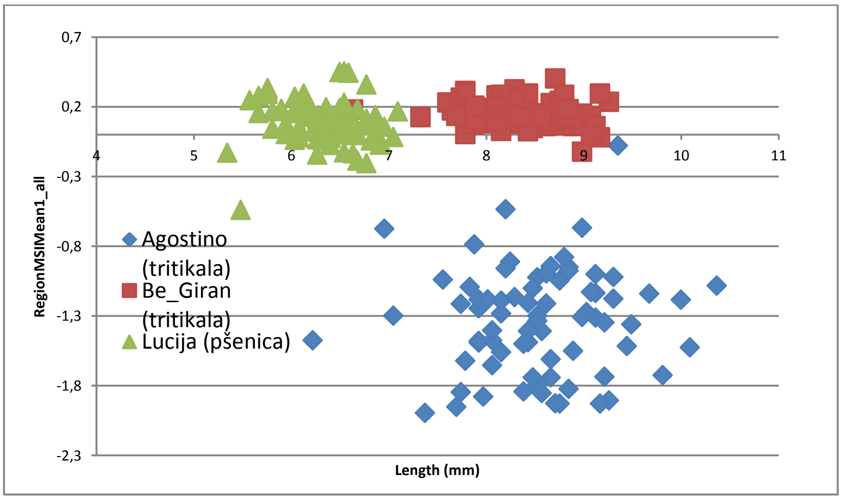

Figure 2: Example of successful varieties separated with multispectral imaging into three groups according to color differences (green/yellowish) and size (long/small) and according to the species (winter wheat/triticale).

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Nutrient use efficacy and resistance to seed borne diseases in European spring barley cultivars and landraces

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A two-year field experiment was conducted during 2011–2012 to study the breeding effect on N use efficiency traits of barley. All together 195 genotypes (72 the Nordic Genetic Resource Center landraces and 123 cultivars released during 1916-2010) were grown in two N regimes (35 and 70 kg N/ha) at the experimental farm of MTT Agrifood Research Finland (presently Natural Resources Institute Finland) in Jokioinen. The results revealed clear positive breeding effect on N use efficiency in barley cultivars. Breeding had also improved grain yield and traits related to yield, like grain number, grain weight and harvest index. Efficient N use and utilization correlated positively with grain yield and negatively with stem length. Accordingly, it appears that breeding induced changes in stem length and biomass allocation pattern, resulting in increased yield, has simultaneously improved N use efficiency in barley cultivars. Phosphorus was analysed from a smaller set of genotypes to reveal/study the breeding effect and potential association between N and P uptake and utilization dynamics. According to the results breeding had improved P use efficiency and N and P use efficiency seemed to be coupled. However, additional P analyses should be carried out to confirm these preliminary results.

Resistance of 152 barley genotypes (both Nordic cultivars and landraces) was tested in field against Fusarium head blight (*Fusarium culmorum*) in 2013 and 2014. Some variation was found in infection and germination level between the genotypes. However, no significant resistance was found. Also, there was a poor correlation in results between the two years and it was shown that reliable testing for quantitative Fusarium resistance needs a several years multilocational testing system. The resistance of 985 barley cultivars and landraces with different geographical origin for their net type (*Pyrenophora teres* f. *teres*) and spot type (*Pyrenophora teres* f. *maculata*) resistance was tested in a greenhouse. A significant improvement in the net blotch resistance level was found in the European barley cultivars released during the last 40 years. The frequency of resistant genotypes against net type net blotch was highest among the European barley cultivars and Syrian landraces. For the spot type net blotch, the average resistance level in the studied material was clearly weaker than for the net type net blotch. Most promising material was found in the landraces originating from Jordania. Of the Nordic landraces only a Swedish landrace NGB15162 showed net blotch resistance that could be useful in breeding programmes. Leaf stripe resistance (*Pyrenophora graminea*) was tested using a sandwich method in 125 barley genotypes (with 38 Nordic landraces). Significant differences in resistance was found. The most resistant genotype was a Finnish landrace Ylenjoki AP0301 (NGB4413A). The most resistant cultivars were Jyvä and Harbinger. Cultivars Pilvi, Elmeri, Posada, Rambler, Marthe, Sunshine, Prestige, Beatrix and Iron were highly susceptible against leaf stripe infection. Of the same material also loose smut (*Ustilago nuda*) resistance was tested in greenhouse. Moderate loose smut resistance was found among the tested barley landraces.

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). Two resistant landraces (NGB13482 and NGB6941) were shown to carry Vada resistance. Importantly, four resistant cultivars (Idumeja, Latvijas_Vietejie, Minttu and Mitja) and 4 resistant landraces (NGB13021, NGB16881, NGB9315 and NGB9410) were shown to have leaf stripe resistance which genetic background differed from Vada resistance. This is useful finding in search for new resistance sources. In general, the published PCR-based markers are very valid for marker-assisted selection in breeding programs when the resistance source is known and there is a specific marker to differentiate between resistant and susceptible genotypes. So far, however, there are no general molecular markers that could be used for screening leaf stripe resistance in all barley cultivars and landraces.

Purifying virulence races of common bunt (*Tilletia caries*) to identify resistance genes in wheat

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Introduction

Breeding wheat with resistance to common bunt (*Tilletia caries*) is an important part of a strategy to control the disease in organic farming. Many wheat varieties are described as resistant to the disease, and several major genes are described able to control the disease. However, there is a lack of consistency in field trials assessing wheat varieties for their susceptibility to the disease (Blažkova and Bartoš 1997).

No doubt, genetic markers will be a powerful tool to identify resistance genes in breeding lines, also when it comes to resistance against bunt in wheat, but in order to develop markers, reliable phenotypic assessments are needed. To get there, we need tools to distinguish between different resistances in wheat lines.

Most trials published on susceptibility of wheat varieties to bunt are done by adding spores to a seed sample before sowing, at assessing the frequency of of infected heads. This method is a more or less adapted method of trials testing for the effect of seed treatments (EPPO 1997). When testing for resistance, the virulence of the spores are of crucial importance, since a trial will not be able to identify resistance genes, if the spores used are virulent to the gene. Often, the origin and virulence of the spores in trials are more or less unknown, and most spore samples found in practical farming are genetic diverse. The may therefore be a mixture of virulent and avirulent spores against the resistances in question. Using such spores in a trial may result in data difficult to interpret, as a low infection level can be a result of a low frequency of virulent spores in the spore sample. Next year, the composition of spores may have changed, which will lead to a different result in the same varieties.

In this situation, I felt a need to work with less diverse spore samples in my bunt trials, and to achieve this, I developed a simple technique to develop more uniform and well described spores. Already Weston (1932) and Roemer og Bartholly (1933) showed that susceptibility of a variety increased when it was infected by spores from the same variety compared with spores from other varieties. Woolman (1930) showed that only up to 4 spores were able to infect the wheat meristem. Therefore, working with spores from a single plant will decrease diversity in a spore collection, and if this plant is resistant, it will after a few regenerations of such a variety be selective to a specific race, virulent to the resistance gene in question.

Materials and methods

Spores were collected in the ORGSEED project in 2001-5 by Bent Nielsen. Spores were collected from a broad range of fields grown with different varieties in different regions of Denmark. It is likely that they pretty much represent the genetic diversity of the pathogen present in Danish agriculture by the time of collection. The collection was maintained on susceptible varieties until 2011 and hereafter on a composite cross population.

From 2012 and onwards, resistant varieties were infected with spores collected on their own variety if infected spikes were found. In this way, a collection of 98 virulence races were build up specific to the 98 wheat varieties.

In 2013-14 and in 2014-5, spores from selected varieties were used to infect other varieties.

The varieties were sown in 0.5 rows without replication with 5g seed per row.

Results and discussion

Some of the varieties that had low infection in previous trials (Steffan 2014 , Steffan *et al.* 2014), and therefore were recorded as resistant, could indeed be infected if they were re-inoculated with the spores from the few infected plants of the variety. For example, varieties with the resistance gene Bt10 had no or low infection when infected with the diverse spore collection, and it was concluded that Danish spores were avirulent to Bt10. However, when varieties with Bt10 were inoculated with spores from their own variety, they turned out to be highly susceptible. The spore collection is therefore a mixture of virulent and avirulent spores against Bt10. The same is the case for Bt2, Bt7, Bt13 and BtZ.

When varieties were inoculated with spores that had been purified on other varieties, they were normally either more resistant or more susceptible compared with inoculation with the spores mixture. It is likely that varieties that react in a similar way to different origins of spores may have the same resistance genes. However, this is not always the case. Some (or maybe most) varieties carry more than on gene affecting the susceptibility, and certainly most virulence races of the pathogen are virulent to more than one resistance gene. For example, spores virulent to Bt10 are in this study also virulent to BtZ, and I have been unable to distinguish between these two genes.

Some varieties have so far been resistant I all studies, and even if a few plants were infected, spores from these plants have been unable to create a high infection level. Some of these varieties may have a resistance gene to which no virulence have been found, and some varieties may have a combination of genes. I have been unable to develop virulence races specific to the resistance genes Bt3, Bt4, Bt5, Bt6, Bt8, Bt9, Bt11, Bt12.

It is possible that the varieties NGB9014, NGB-9015, Tambor, Kuban, Begra, Maribos, Fold, Monopol, Tarso, Torrild, Cardos, Kranich, Türkis, Gluten, Folke have Bt7, since they react in a similar way to the 7 different sources of spores used in 2014-5.

It is possible that the varieties Format, Curier, Complet, Solstice, Bussard, Paroli, Dream, Butaro, Ochre, PG3540 and Hereward have Bt2.

The varieties Korrund, Aron, Karat, Tulsa, Xenos, Tataros, Erbachshofer Braun and Indigo have in some trials shown resistance, but in 2014-15, they have been susceptible to most or all virulence races.

Spores harvested on the variety Tommi by Svalöf/SLU in 2014 were able to give a high infection in the Tommi, Globus and Segor, and a medium infection was achieved with spores from BOKU, even though these varieties have so far been resistant or shown low susceptibility to spores from Denmark. The variety Quebon could be infected by spores purified from Czech Republic, but was resistant to all other spores tested on other varieties. This indicates that Tommi, Globus and Segor may carry the same resistance gene, which is different to the gene in Quebon, and different to the known Bt-genes.

Infecting 65 varieties of winter wheat with 7 sources of common bunt inoculum
"*Own spores*" means spores harvested previous year on the tested variety

	Vr: Own spores	Vr2: Complet	Vr: 10 (Weston)	Vr:1 (Pi554108)	Vr: 4 (Cardos)	Vr: 5 (Hohenheim)	Vr: 13 (Thule III)	Vr: 8 (Pi554120)
Pi554-121 (Bt3)	0,0	0,0	0,0	0,0	0,0		0,0	0,0
Ridit (Bt3)	11,1	0,0	5,0	3,4	0,0	0,0	0,0	0,0
PI 554-116 (Bt3	33,3	0,0	0,0	11,1	7,1	0,0	0,0	0,0
Nebred (Bt4)	16,7	0,0	0,0	14,3	0,0	3,7	0,0	2,3
PI-554-115 (Bt4)	66,7	0,0	4,8	0,0	14,8	3,8	0,0	0,0
Carlton	0,0	6,7		0,0	60,0	0,0	7,8	0,0
PI-554-117 (Bt6)	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
Erythrosperum 5221 (Bt14)	0,0	2,6	0,0	3,0	9,8	0,0	0,0	4,8
Promesse (Bt5)	0,0	0,0	0,0	2,7	0,0	0,0	0,0	0,0
Hohenheimer (Bt5)Ci-11458	50,0	27,6	16,7	0,0	0,0	6,9	0,0	12,5
Lars	0,0	8,3	35,3	0,0	0,0		2,4	6,3
Triple Dirk	0,0	3,2	13,0	0,0	0,0	4,8	0,0	0,0
Hypnos	0,0	2,9	18,2	3,7	3,4	9,7		0,0
Skagen	4,5	37,8	20,0	9,1	0,0	3,3	1,6	0,0
Skotte	0,0	26,7	0,0	0,0	0,0	0,0	0,0	0,0
PI-554-110 (Bt8)	0,0	0,0	20,0	33,3	40,0	0,0	0,0	0,0
PI-554-111 (Bt8)	50,0	0,0		100,0	20,0	47,1	0,0	0,0
Thule III (Bt13)	71,4	0,0	0,0	0,0	0,0	2,6	30,4	13,3
Inna	22	0,0	84,0	0,0	0,0	0,0	4,2	0,0
Nemchinovskaya 25 (BtZ) PI	0,0	3,4	55,6	3,8	7,1	0,0	0,0	2,9
Pi173437 (Btp)		2,0	18,8	0,0	0,0	0,0	0,0	13,3
Weston	33,3	0,0	52,6	0,0	0,0	0,0	0,0	13,6
PI-554-118 (Bt10)	45,5	0,0	53,6	0,0	0,0	0,0	0,0	0,0
PI-554-109 (Bt10)	33,3	0,0	50,0	10,5	7,1	15,8	0,0	0,0
Intaler	47,4	15,7	9,1	13,8	2,4	10,5	1,8	2,0
Lutescens 6028 (PI 591884)	0,0	12,5	25,0	30,0	10,0	17,1	4,0	4,8
Penta	20,0	9,5	0,0	0,0	0,0	4,8	0,0	0,0
PI-554-103 (Bt2)	25,0	8,8	0,0	45,0	0,0		0,0	0,0
PI-554-097 (Bt2)	57,1	61,3	5,9	31,6	0,0	0,0	0,0	0,0
Format	76,9	44,4	0,0	22,2	0,0	4,7	0,0	0,0
Curier	54,5	41,9	11,8	0,0	0,0	2,4	0,0	0,0
Complet	43,8	**69,7**	0,0	6,3	0,0	0,0	0,0	0,0
Solstice	75,0	40,3	0,0	12,5	2,4	4,2	0,0	0,0
Bussard	77,3	63,4	0,0	12,5	2,3	0,0	0,0	0,0
Paroli	80,8	53,3	0,0	14,8	2,0	4,8	0,0	0,0
Dream	61,5	28,1	0,0	16,7	0,0	2,3	0,0	0,0
Butaro (154)	37,5	64,7	7,1	25,0	0,0	3,0	2,6	0,0
Ochre	50,0	30,0	0,0	24,2	0,0	1,7	0,0	0,0
PG3540	50,0	10,0	0,0	15,0	0,0	0,0	0,0	0,0
Hereward	60,0	14,8	12,0	4,2	0,0	0,0	0,0	7,7
PI-554-108 (Bt1)			0,0		0,0		0,0	
PI-554-101 (Bt1)	100	11,1	0,0	37,5	7,7	12,5	0,0	0,0
Bill	16,7	0,0	3,8	0,0	2,0	25,0	0,0	2,8
NGB9014	57,1	3,2	15,6	0,0	0,0	30,0	10,0	0,0
NGB-9015	33,3	22,5	23,1	4,2	0,0	18,0	4,7	0,0
Tambor	0,0	31,6	12,5	0,0	2,0	19,7	4,0	3,0
Kuban	11,1	18,5	12,1	3,0	32,8	11,9	0,0	0,0
Begra	0,0	27,8	44,4	4,5	17,6	27,5	5,4	7,3
Maribos		12,1	21,4	3,2	37,5	8,6	2,3	10,0
Fold	16,7	12,2	14,6	5,6	8,6	9,8	0,0	4,3
Monopol	50,0	90,9	0,0	40,0	11,1	3,4	0,0	0,0
Tarso	7,1	23,4	34,4	16,0	28,6	6,3	8,7	2,1
Torrild	14,3	38,5	35,0	13,9	14,3	17,5	2,1	3,2
Cardos	17,6	26,2	57,9	31,3	**16,0**		8,9	0,0
Kranich	19,0	35,0	10,5	33,3	23,7	22,7	11,7	0,0
Türkis	45,0	40,0	25,0	11,1	42,2	28,6	6,8	0,0
Gluten	28,6	16,7	46,2	34,6	61,0	22,5	14,3	0,0
Folke	37,0	32,6	25,0	33,3	70,6	24,3	5,6	0,0
PI-554-114 (Bt7)	50,0	19,2	22,2	30,0	0,0	25,0	15,2	3,7
PI-554-100 (Bt7)	33,3	28,6	45,7	4,5	0,0	35,5	10,9	9,1
Korrund	20,0	25,0	55,6	0,0	4,2	17,0	14,8	35,7
Aron	33,3	50,0	55,6	15,8	2,9			
Karat	84,6	55,6		12,5	0,0			20,0
Tulsa	21,1		54,8	42,9	51,1	34,4	36,2	45,7
Xenos	70,7	62,0	66,7	39,4	18,4		13,2	26,3
Tataros	12,5	34,6	52,0	26,7	29,7	26,2	16,7	26,3
Erbachshofer Braun	55,6	75,0	45,5	55,0	46,4	57,1	12,5	0,0
Indigo	76,6	68,0	77,5	88,2	75,7	59,0	30,0	4,4

Colour legends: low plant number | 0,0% | <10% | 10-40 | >40%

Acknowledgement

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Infecting 34 highly resistant varieties of winter wheat with 4 sources of common bunt inoculum
"*Own spores*" means spores harvested previous year on the tested variety

	Vr: Own spores	Vr: Tommi Svalöf	Vr: Tommi BOKU	Vr: CCP mixture	Vr: RU11	Vr: Weihenstephan
Quebon	24,0	0,0	6,7	0,0	39,3	6,3
Tommi	26,7	17,1	14,3	10,3		
Globus	0,0	55,6	14,3	0,0		
Segor		43,8	0,0	13,3		
Huslop		0,0	0,0	0,0		
Winridge	0,0	0,0	0,0	0,0		
Wasatch	0,0	0,0	0,0	0,0		
Sprague		0,0	0,0	0,0		
PI-554-104 (Bt5)		0,0	0,0	0,0		
PI-554-112 (Bt9)		0,0	0,0	0,0		
Pi 554099 (Bt9)		0,0	0,0			
Pi 554099 (Bt9)		0,0				
Andrews (Bt-9?)		0,0	0,0	0,0		
BB118 (Philipps)	2,9		0,0	0,0		
BB152 (Philipps)	0,0	0,0	2,9	0,0		
PI-554-098 (Bt11)		0,0	0,0	0,0		
PI-554-119 (Bt11)		0,0	0,0	0,0		
PI-554-105 (Bt4)+5		0,0	0,0	0,0		
Hansel (Bt-8+9+10?)		2,4	0,0	0,0		
PI-119-333 (Bt12)	0,0					
PI-119-333 (Bt12)	7,4	0,0	0,0	0,0		
PI-554-106 (Bt12)		9,1	6,5	44,4		
Rio (Bt6)	0,0	0,0	8,3	0,0		
Sam	0,0	9,1	0,0	0,0		
Golda (Zarya-res.)		31,0	0,0	0,0		
Bill			10,4			
Marin (Ridit-res.)	43,8	27,0	0,0	4,0		
PI-554-120 (Bt8)	50,0		0,0			
Yayla	50,0		0,0	33,3		
Turkey (Pi11610) Bt4)		41,2	19,0	34,8		
Heines VII (no Bt genes)		33,3	50,0	30,0		
Rola (Trebelir-res.)	45,5	2,9	68,0	44,4		
PI 172201 (Doubbi resistens)			0,0			
PI 172201 (Doubbi resistens)			50,0			
Doubbi (Bt-14)	25,0		50,0			
Doubbi (Bt-14)			0,0			

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Adaptation of spring barley for extreme climates

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Abstract

Climate change is likely to decrease crop yields worldwide. Developing climate resilient cultivars is one way to combat this production scarcity, however, little is known on crop response to future climate conditions and in particular the variability within crops.

In Scandinavia, spring barley is widely cultivated, but yields have stagnated since the start of this century. In this study we cultivated 173 spring barley and 18 spring wheat accessions in a climate phytotron under four treatments mimicking forecasted levels of temperature, carbon dioxide concentration ([CO_2]) and ozone ([O_3]) at the end of the 21[st] century. The ambient control had 19/12°C (day/night) and [CO_2] at 385 ppm. Three single-factor treatments had elevated temperature +5°C day/night, elevated [CO_2] at 700 ppm or elevated [O_3] at 120 ppb, and in a two-factor treatment the combination of elevated temperature and elevated [CO_2] was applied.
Treatment effects were assessed on grain yield, grain protein concentration, grain protein harvested, number of grains, number of ears, aboveground vegetative biomass and harvest index. In addition, stability of the production was calculated over the applied treatments for the assessed parameters.

In the climate scenario of elevated temperature and [CO_2] the grain yield of barley decreased 29% and harvested grain protein declined 22%. Vast variation was identified among the individual barley accessions, which should be exploited by plant breeders in the development of climate resilient cultivars.

A genome-wide association study (GWAS) of recorded phenotypes and 3967 SNP-markers identified 60 marker-trait associations ($-\log_p>2.95$). Markers were found associated with grain yield under all three single factor treatments temperature, [CO_2] and [O_3], as well as with stability over treatments.

Effect of a 10-days heatwave at flowering superimposed on future climate scenarios was also analysed. We found a 52 % decrease in grain yield, when such an extreme heatwave was induced in the treatment where elevated [CO_2] and elevated temperature were combined. Change in allocation of biomass was recorded.

A subset of the spring barley accessions were analysed under elevated [CO_2] in a FACE (Free Air Carbon Enrichment) facility; also analysed were variety-mixes. Those accessions and variety-mixes with the best potential for exploiting the extra [CO2] were pinpointed. Fungal diseases were recorded, and the results showed changed abundance of diseases in response to the elevated [CO2].

Finally we performed a life cycle assessment (LCA) of the future barley production under climate change. This LCA stressed the importance of targeted breeding for climate change conditions to minimize the environmental impacts from spring barley production.

Weed competitive ability and yield stability of winter- and spring wheat populations, cultivar mixtures and cultivars.

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Introduction

COBRA aims to support and develop organic plant breeding and seed production in Europe with a focus on increasing the use and potential of plant material with High genetic Diversity (Hi-D), such as Composite Cross Populations (CCPs) and variety mixtures. Multi-site field trials with such material are hence essential. CCPs and variety mixtures have mainly been studied from a pest resistance perspective and less so for yield stability and weed competitive ability (WCA). Both in spring wheat and winter wheat differences in WCA can be explained by early growth vigour (Bertholdsson, 2010; Bertholdsson, 2011). Within the Cost action SUSVAR breeding lines of winter wheat have been selected for high allelopathic activity from a CCP in generation F_4 and within COBRA there has now been a possibility to test these lines in northern Europe from west to east. In SUSVAR and a number of national project breeding lines of spring wheat with improved allelopathic activity have also been produced and are now being further evaluated in broader environments. It is known that allelopathy is dependent on the environment and hence it is important to test such material across a range of these. Along with the breeding lines, their mixtures, national cultivars and CCPs have also been evaluated for yield, yield stability, WCA and disease resistance.

Material and Methods

Organic field trials have been conducted in Estonia, Scotland and south Sweden during 2 seasons of winter and 3 seasons of spring wheat, respectively.. Common to all sites in winter wheat are four breeding lines selected for high allelopathic activity from a composite cross population (CCP) in F_5 produced by ORC, various combinations of the breeding lines, the CCP grown both organically and conventionally for another 6 generation and the Swedish standard cultivars Harnesk and Kranich. In spring wheat, common to all sites are 3 breeding lines with improved allelopathic activity from a cross between Zebra and Mohan (Bertholdsson 2010), a mixture of the three, Zebra and a spring CCP from ORC selected out of the winter CCP. In both winter and spring wheat, at least 3 local cultivars and were also included. Besides the usual observation of early vigour (by a subsample of a 1 meter row at early stem elongation) and weed biomass(by sampling at heading, all weeds of an area of 2 x 0.25 m^2) were observed. Baking quality and weeds analysis were also done in some of the trials. Under controlled conditions, early vigour of shoot and roots were analysed in hydroponics as well as potential allelopathic activity by a bioassay with rye-grass as receiver plants (Bertholdsson 2010; Bertholdsson and Østergård, 2006).

Result and Discussions

The trial at SRUC in 2014 was hampered by wet and cool weather in the autumn and the results are hence based on 5 trials (environments). In winter wheat the CCPorg and CCPconv showed 10 and 11% higher yield than the mean of the pure lines selected from the CCP and thus support the hypothesis that CCPs have higher yield stability. Similar to this the mixtures also showed 2-8% higher yield than the components of the mixtures. The single trait that showed the highest correlation with the weed biomass was the no. of fertile ears (r=-0.77, p<0.01) followed by allelopathy (r=0.59, p<0.05). CCPorg and CCPconv showed 12% and 25% more weed biomass than the pure lines selected from the CCP for higher allelopathic activity. This may indicate that a direct selection for a trait with importance to WCA could be successful.

In spring wheat only the three lines selected and twice back-crossed to Zebra, Zebra and the spring form of the CCP from ORC were planted at all three sites. The results are preliminary and based on the first two years. Yield was 18% lower of the breeding lines than Zebra as well as the mixture of the three breeding lines. Zebra also out-yielded the CCP with 5 %. In spite of 26% lower allelopathic activity, Zebra showed the lowest weed biomass of all. The reason for this is probably an 18% higher early biomass growth. In spring wheat there was a strong correlation between early wheat biomass and weed biomass (p=-0.97, <0.0001). There was also a strong positive correlations between no. of tillers and weed biomass (p=0.77, p<0.01), indicating that high allelopathic activity may have negative effects on tiller formations in this material.

There were no clear results showing the importance of seedling root growth for yield and yield stability or N-uptake of mixtures as observed in barley (Bertholdsson and Østergård, 2006). In winter wheat the CCPs showed high yield stability as well as three of the four mixtures. However two of the breeding lines and one of the cultivars also showed high yield stability and thus no clear results supporting that mixtures and population show higher yield stability. The yield stability of the spring wheat material is not yet analysed.

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for the CCP seeds.

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Winter wheat variety performance in organic and conventional farming systems and the potential for organic plant breeding in the UK

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Introduction

Modern plant breeding has increased cereal yields through development of varieties with high response to external inputs such as mineral fertilisers and pesticides which regulate the cropping environment (Evenson and Gollin, 2003). Such varieties, however, often lack resilience in more marginal environments, including organic farming systems. As a consequence, the productivity of organic farming in the UK is limited by the lack of suitable cereal varieties. This could be resolved by utilising modern plant breeding technologies and selecting material for local adaptation to the target environment so as to develop varieties that perform well under low inputs (Wolfe *et al*., 2008). Evolutionary participatory plant breeding offers this opportunity (Dawson *et al*., 2008).

This study reports on variety trials on organic and non-organic farms in the UK to examine Genotype x Environment x Management (G x E x M) interactions together with trials currently in progress to assess the performance of winter wheat lines selected under organic conditions compared to commercial elite controls.

Materials and methods

Nineteen modern and heritage wheat varieties were trialled in a replicated randomised complete block design on two organic and two conventional sites in England over three trial seasons from harvest years 2003 to 2005. Trials which are currently in progress include six winter wheat selected lines and high yielding (c.v. Alchemy) and high quality (c.v. Solstice) control varieties. The lines were hand selected as single ears from an F9 Composite Cross Population (CCP) grown organically in 2011. Replicated plot trials were set up on two organic farms in the east of England and harvested mechanically for yield assessment.

Results and discussion

Principal Component Analysis of variety performance on organic and non-organic farms suggests differences between sites and the two management systems (Fig. 1). Trends in variety performance were similar for the two conventional sites whilst trends at the two organic sites differed from the conventional sites as well as from each other. The genotype by site interaction to genotype effect ratio was 0.29 for the two organic sites and 0.03 for the two conventional sites. Whilst this only includes two sites for each management system, the observations support the premise not only that conventionally managed farms provide a regulated growing environment for the crop, leading to predictable performance of elite varieties, but that organic farms require different varieties, as demonstrated by Murphy *et al.* (2007), and that these requirements are site specific because of the more variable nature of organic management practices and uncontrolled biotic and abiotic stresses. This highlights the potential value of evolutionary plant breeding and selection of breeding material in the target environment as an appropriate approach for breeding wheat for organic systems.

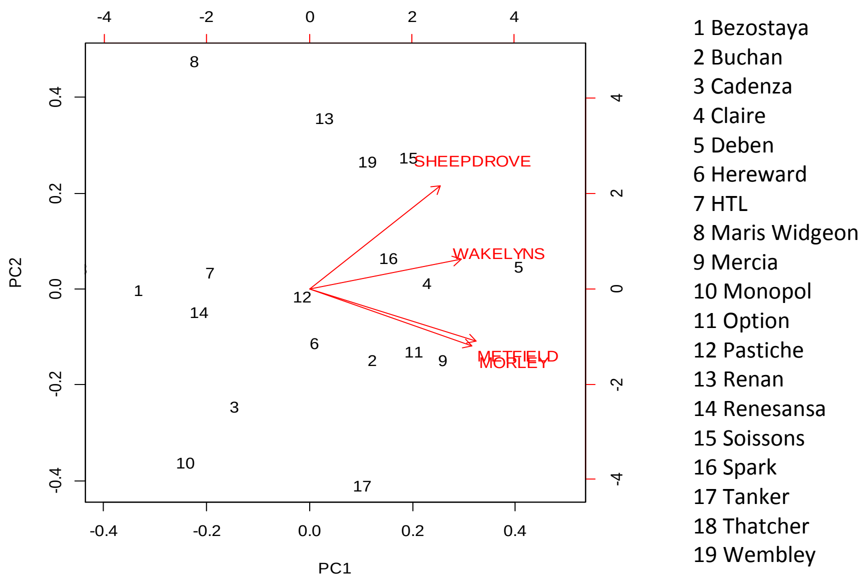

Figure 1. Biplot of the first and second principalcomponents of mean yield over three growing seasons of 19 winter wheat varieties represented by numbers at two organic sites (Sheepdrove and Wakelyns) and two non-organic sites (Metfield and Morley)

Current trials are expected to show that the performance of six winter lines which were selected under organic conditions at Wakelyns, outperform commercial elite varieties in terms of yield, disease resistance and weed competitiveness in the selection environment. However, performance of the organic selected lines appears to be worse away from the selection environment at a different, very low yielding organic site in the east of England.

These trials help to demonstrate the potential advantages of this approach for developing varieties with reduced input requirements and greater resilience as well as addressing the lack of suitable cereal varieties available to organic farmers.

Acknowledgements

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Adaptation of Specific Winter Wheat Genotypes (CCPs) to the Slovenian Climate

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Introduction

As a partner in the COBRA project, the Faculty of Agriculture and Life Sciences (FALS) of the University of Maribor plays an active role in the WP2 addressing of resilience and adaptability of different winter wheat Composite Cross Populations (CCPs) and their overall performance in different countries (cycling populations). CCPs were created in 2002 by crossing high yielding (Y) and high quality (Q) genotypes of a diverse genetic base (Döring et al., 2010).

Material and methods

Samples of winter wheat, CCP populations F_{12} (A I, A II, Q I, Q II, YI, Y II), were received from the project partner University of Kassel. At the experimental field of FALS samples were sown in October 2013 (half of the received F_{12} samples) and in 2014 (second half of the F_{12} samples and populations F_{13} – materials harvested in 2014). Samples were sown in organic (ORG) and conventional (CON) production systems treatments. The field experiment was set according to the split-plot randomized complete block design in four replications. The experimental plots were 7.5 m^2 (6×1.25 m) in size. Production system was the main and CCPs was the subplot.

Growth, development, morphological and physiological characteristics, infection with the most common diseases (foliar: *Septoria tritici, Puccinia graminis,* and ear diseases: *Fusarium* spp., *Septoria nodorum*), and the productivity of individual genotypes were monitored in accordance to the received protocol. Only some of the obtained data are presented in the present abstract. Results obtained in both production years were subjected to ANOVA. The comparisons of means were performed with the Duncan test. The results are presented as the mean of four replications ± standard error of mean (±SEM).

Results and discussion

Some of the results obtained over the two years are presented in Table 1. The height (80.5 to 133.0 cm) and grain yield (2,600 to 8.000 kg ha^{-1}) of the wheat were not affected by the genotype. The grain yield in CON is significantly higher than in the ORG production system: for 1,548 kg ha^{-1} in average, the difference is more pronounced in the less favourable year 2014/2015 (in average for 1,998 kg ha^{-1}). The specific weight of the harvested grain in the first year is relatively low and not affected by the production system, while in 2014/2015 both populations expressed higher values in the CON system. Both genotypes Q (the result of crossing varieties of high quality) show higher specific weights than genotypes Y (the result of crossing high-productive varieties), but in general show the same results as genotypes A (combination of Q and Y). The production year influenced them all, and interaction PS × CCP is not significant in any of the presented parameters.

Table 1: Influence of production system and genotypes on plant height, grain yield (14% moisture) and specific weight.

	Plant height (cm)	**Grain yield (kg ha^{-1})**	**Specific weight (kg 100L^{-1})**
		F_{12}: 2013/2014	
Prod. Syst. (PS)	**n.s.**	***	**n.s.**
CON	107.5 ± 4.32	6,719 ± 88.2a	74.0 ± 0.26
ORG	104.0 ± 4.47	6,075 ± 167.8b	73.7 ± 0.37
CCP	**n.s.**	**n.s.**	*****
A I	111.6 ± 3.11	6,418 ± 323.8	74.3 ± 0.34bc
A II	108.0 ± 3.01	6,321 ± 274.1	73.8 ± 0.23c
Q I	111.1 ± 3.18	6,289 ± 212.1	74.9 ± 0.33ab
Q II	101.0 ± 12.98	6,473 ± 287.4	75.4 ± 0.24a
Y I	95.1 ± 12.20	6,353 ± 198.8	72.3 ± 0.34d
Y II	107.5 ± 2.75	6,529 ± 299.3	72.1 ± 0.54d
		F_{12}: 2014/2015	
Prod. Syst. (PS)	**n.s.**	***	*****
CON	101.2 ± 1.38	6,006 ± 179.4a	76.1 ± 0.30a
ORG	98.2 ± 1.20	4,424 ± 276.7b	75.3 ± 0.33b
CCP	**n.s.**	**n.s.**	*****
A I	97.3 ± 2.90	5,449 ± 473.4	76.2 ± 0.29a
A II	95.5 ± 3.00	5,069 ± 472.0	76.0 ± 0.29a
Q I	104.9 ± 2.75	5,066 ± 501.1	76.9 ± 0.37a
Q II	101.9 ± 3.74	5,007 ± 546.8	76.8 ± 0.37a
Y I	98.5 ± 3.28	5,291 ± 574.2	73.9 ± 0.49b
Y II	100.1 ± 1.27	5,407 ± 505.4	74.4 ± 0.34b
		F_{13}: 2014/2015	
Prod. Syst. (PS)	*****	***	*****
CON	106.6 ± 1.70a	6,507 ± 114.5a	77.6 ± 0.26a
ORG	101.3 ± 1.35b	4,093 ± 183.9b	75.2 ± 0.31b
CCP	**n.s.**	**n.s.**	*****
A I	104.3 ± 2.82	5,116 ± 556.3	75. 6 ± 0.53d
A II	102.8 ± 3.06	5,757 ± 497.6	76. 6 ± 0.69bc
Q I	106.6 ± 4.09	5,257 ± 448.7	77.3 ± 0.40ab
Q II	108.5 ± 2.28	5,032 ± 440.8	78.0 ± 0.60a
Y I	100.4 ± 2.21	5,407 ± 604.8	75.0 ± 0.60d
Y II	101.3 ± 1.19	5,232 ± 587.7	75.9 ± 0.61cd

*** Significant at *P*<0.001,** significant at *P*<0.01, ,* significant at *P*<0.05, n.s. not significant
[a-d] Mean values (± SEM) followed by different letters within a column, treatment, year and population are significantly different (Duncan, α= 0.05)

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Adaptation of 10 Barley Genotypes to the Slovenian Climate

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Introduction

Within the COBRA project, the role of crop genetic diversity in spring barley has been investigated in Slovenia by the Faculty of Agriculture and Life Sciences (FALS) of the University of Maribor. FALS tested the growth and yield performance, as well as the disease susceptibility of 10 spring barley varieties received from the Technical University of Denmark.

Materials and methods

Samples of 10 spring barley accessions (‘Alliot', 'Brage', 'Brio', 'Evergreen', 'Edvin', 'Jacinta', 'Mari', 'Pallas', 'Sebastian' and 'Tron Sejet') were received in 2015 from the Technical University of Denmark. Cultivars have been chosen regarding their response in production parameters to projected future climate and analysed in RERAF phytotron (Ingvordsen et al., 2015), as well as regarding the description of Slovenian climate conditions.

A field experiment was arranged in a completely randomized block design, because a small quantity of seeds has been received, it was conducted in three replications, in experimental plots sized 3 m^2 (1×3 m). Varieties were evaluated based on heading date, plant height, tillering, grain yield, specific weight and susceptibility to, in our condition most prevalent, the diseases *Helminthosporium teres*, *Rhynchosporium secalis* and *Ramularia collo-cygni*. Infestation with diseases and damage caused by *Oulema melanopus* were assessed according to the protocol of the Agricultural Institute of Slovenia (logarithmic key based on scale from 1 to 9: 1 is 0, 9 more than 62% infected plants) as is used in national variety tests carried out at FALS every year.

Obtained results were subjected to analysis of variance (ANOVA) and a comparison of means was performed via the Duncan test. The results are presented as the means ± standard errors (SEM) of three replications.

Results and discussion

Results describing the growth and yield performance of 10 tested spring barley varieties are presented in Table 1.

Varieties which are ranked into the group of the highest grain yield (from 3,993 kg ha^{-1} in ‘Evergreen’, 4,018 kg ha^{-1} in ‘Tron Sejet’, 4,245 kg ha^{-1} in ‘Brage’, 4,412 kg ha^{-1} in ‘Alliot’ to 5,146 kg ha^{-1} in 'Sebastian') are rather shorter (58.7 cm 'Brage' to 67.1 cm 'Alliot') and mostly have the highest specific grain weight (54.3 kg 100 kg^{-1} in 'Tron Sejet' to 58.6 in 'Alliot'). Except in the 'Sebastian' (952 ears m^{-2}) variety, all the other most productive varieties revealed a lower number of ears (515 ears m^{-2} in 'Brage' to 840 ears m^{-2} in 'Tron Sejet'). The heading days of the tested varieties were ranked between 13th to 25th of May.

Results revealing disease infestation and damage caused by pests are not significantly affected by the variety (data not presented). Generally, varieties are rather insusceptible to *Rhynchosporium secalis* (values 1.5 to 2.5), and medium susceptible to *Ramularia collo-cygni* (2.0 to 4.0) and to *Helminthosporium teres* (values from 3.0 to 5.0). *Oulema melanopus* was abundant in all varieties, and the damage was estimated from 7.0 to 8.0 on a 9–point scale.

Table 1: Difference in plant height (PH, cm), number of ears (No. m^{-2}), specific weight (SW, kg 100 kg^{-1}), grain yield (GY, kg ha^{-1}, adjusted to 14% moisture) and heading date (day in May) among tested barley genotypes.

Genotype	PH (cm)	No. ears (per m^2)	SW (kg 100 kg^{-1})	GY (kg ha^{-1})	Heading (May)
ANOVA	***	***	**	**	***
'Alliot'	67.1 ± 0.12d	656 ±22.6d	58.6 ± 0.2ab	4,412 ± 554.6ab	13.d
'Pallas'	70. 7 ± 0.07c	1,008 ± 2.2a	61.6 ± 3.5a	3,130 ± 56.2cde	21.b
'Mari'	58.1 ± 0.06j	455 ± 16.0e	59.76 ± 0.5ab	3,892 ± 427.3bcde	15.c
'Brio'	78.6 ± 0.10a	596 ± 20.7d	43.06 ± 3.5c	2,920 ± 385.5de	25.a
'Tron Sejet'	75.4 ± 0.12b	840 ± 4.2c	54.3 ± 2.0ab	4,018 ± 324.9abcd	22.b
'Sebastian'	60.6 ± 0.09g	952 ± 44.7ab	57.1 ± 0.8ab	5,146 ± 425.8a	15.c
'Jacinta'	59.5 ± 0.17h	902 ± 17.2bc	52.4 ± 2.7b	2,7266 ± 237.5e	15.c
'Evergreen'	64.4 ± 0.18f	837 ± 5.8c	55.3 ± 3.3ab	3,993 ± 37.9abcd	24.a
'Edvin'	65.1 ± 0.03e	620 ± 13.7d	53.3 ± 0.8b	3,744 ± 264.0bcde	20.b
'Brage'	58. 7 ± 0.15i	515 ± 31.4e	52.7 ± 0.6b	4,245 ± 589.7abc	14.cd

***Significant at P<0.001,** significant at P<0.01

[a-i]Mean values (± SEM) followed by different letters within a column are significantly different (Duncan, α= 0.05)

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Ranking the Most Resistant Winter Wheat Varieties from the Common Introductional Progamme of Organic Farming in Slovenian Climate

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Introduction

One of the main problems in organic production of winter wheat are plants which are non-resistant to diseases, and the fact that synthetic pesticides are not allowed (EU Regulation No. 834/2007). ,This means that better specific plant protection measures need to be applied, such as crop rotation, high genetically expressed plant resistancy, healthy seeds, etc. (Finckh et al., 2015). The aim of this contribution is to evaluate existing and potential winter wheat varieties for the production and introduction into Slovenian production circumstances.

Material and methods

The used data were taken from Slovenian national crop variety trials (assessing their value for cultivation and use (National List of Varieries), and a special testing of varieties was carried out (for Descriptive Variety List)), from one location (Maribor) over three years of testing. Every year aprox. 70 varieties are tested. Among some other diseases prevalent in Slovenia, the infestation with the most important foliar (*Septoria tritici*) and ear disease (*Septoria nodorum*) was assessed from the part of non-sprayed trials at the EC 65/75 growing stage. The intensity of the infection is evaluated according to a logarithmic key based on a scale from 1 to 9, where 1 is 0, 2 – to 2%, 3 – to 5%, 4 – to 8%, 5 – to 14%, 6 – to 22%, 7 – to 37%, 8 – to 61% and 9 is more than 62% of the infected plants.

The results reveal the most resistant varieties regarding the assessed intensity of the infection with *S. tritici* and *S. nodorum* (values 4 and below) and the variety 'Alixan' which serves as a standard for assessing the disease (non-resistant: values 4 and above).

Results and discussion

The results of the variety tests, carried out over the last three years at the location in Maribor, exhibit a wide range of susceptibility to *S. tritici* and *S. nodorum*. Table 1 shows the most resistant varieties to *S. tritici* (scores 2.0 to 3.0 in average) and to *S. nodorum* (scores 3.0 to 4.5 in average) in comparison to the standard variety 'Alixan' which exhibits a higher infection rate in all years (scores 4.7 and 6.3 for *S. tritici* and *S. nodorum*, respectively).

The selection of adequately resistant varieties, the management practices that reduce wheat residues on the soil surface (broad crop rotation, appropriate agricultural techniques), healthy seeds, and sound fertilization, all help to keep the field crops healthy. According to the obtained results also among varieties officially tested in Slovenia there are some more resistant to Septorias and thereby more appropriate for organic production.

Table 1: Selected varieties of winter wheat compared with the standard ‘Alixan’ according to the assessed lowest intensity of the *S. tritici* (St) and *S. nodorum* (Sn) infection (1 – without)

Production Variety	**2012/2013**		**2012/2013**		**2012/2013**		**Average**	
	St	Sn	St	Sn	St	Sn	St	Sn
'Alixan' (standard)	4	4	6	9	4	6	4,7	6,3
'Angelus'	2	3	2	3	2	4	2,0	3,3
'Tacidus'	3	2	3	3	3	4	3,0	3,0
'Orcas'	/	/	2	4	3	3	2,5	3,5
'Moisson'	/	/	3	4	3	3	3,0	3,5
'Arezzo'	**3**	**3**	**2**	**2**	**3**	**4**	2,7	3,0
'Energo'	2	4	2	2	3	5	2,3	3,7
'Zp'	**/**	**/**	**3**	**5**	**3**	**4**	3,0	4,5
'Lukulus'	/	/	2	2	3	4	2,5	3,0
'Vulkan'	2	3	2	3	4	6	2,7	4,0

/ Variety was not tested in the particular year

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Performance of winter wheat CCP's in comparison to reference varieties in organic field trials in Belgium

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Composite cross populations of winter wheat were tested in field trials at the organic farm of Inagro in Beitem (BE) during the growing seasons 2013-2014 and 2014-2015. Up to 9 CCP's and 4 commercial varieties were assessed. The results confirm that these winter wheat CCP's produce resilient crops with overall similar performance as varieties.

Material and methods

The organic farm of Inagro is located in Beitem, Belgium which has a moderate maritime climate. The sandy loam soil can be considered as fertile without strong limitations. The trial was set up as a randomized block design with 4 replicate blocks with a net plot size of 20 m².

The CCP seed samples originated at the University of Kassel (D; 9 CCP's) and the Organic Research Centre (UK; 1 CCP). They were created in the UK by the ORC in the year 2001. Some CCP's have a smaller parental basis (OQ, OY) and all have different histories since 2001. As a reference, 4 winter wheat varieties were included of which organic seed was available. These are considered as local standard varieties for organic baking quality. The objects were scored or measured for traits such as early growth vigour, weed suppression, disease susceptibility, yield and yield quality at various times during the growing season.

Results and discussion

Although the large heterogeneity among plants in the CCP plots was apparent, among the different CCP objects the visual appearance was very similar and none of the assessed traits showed significant differences inside of this group. This in contrast to the variety group where strengths and weaknesses among objects were significant and recurring. In both years emergence rate, amount of tillering, early crop development and weed suppression did not significantly differ between the CCP group and the variety group. The CCP's were remarkably taller than the varieties but lodging did not occur thanks to favorable weather conditions. In terms of yield and protein content the CCP's and varieties performed equally (see Table 1). Yields in the season 2013-2014 averaged at 6,5 t/ha for the variety group (4 obj.) and 6,2 t/ha for the CCP group (4 obj.) and in the season 2014-2015 at 7,0 t/ha for the variety group (4 obj.) and 7,2 t/ha for the CCP group (9 obj.). By testing for protein content, sedimentation value (Zeleny) and falling number (Hagberg) it was found that the OY populations do not comply for baking. The OQ populations performed as good as the varieties for these parameters, the OA populations were intermediate.

Respectively yellow rust and drought were the limiting factors for growth in these two seasons. Initially in the season 2013-2014, the CCP's suffered more from yellow rust than the varieties but their infestation rate was stable in the period from April to June. Infestation of the varieties rised quickly in May to June and the highest infestation was observed for varieties Lukullus and Midas. In 2014-2015, yellow rust pressure was only moderate. Late

May, varieties Renan and Skerzzo were significantly less infested than the CCP's. The mild to severe drought in spring persisted through summer but had no apparent impact on the time of ripening and the yield of all objects.

Table 1: Origin and trial results of CCP's and varieties of winter wheat (harvest 2014 and 2015; Tukey HSD).

Name/var.	Origin	2013-2014			2014-2015		
		yield (kg/ha) *15% moisture*	protein (%) *15% moisture*	yellow rust *2014 June 10*	yield (kg/ha) *15% moisture*	protein (%) *15% moisture*	yellow rust *2015 May 29*
C CCD13	UK08-DK09-TUM10-HU11-NL12-D13	6056 a	9,3 ab	6,5 a	6557 b	8,6 abcd	4,3 de
C CCUK13	HU08-NL09-D10-CH11-F12-UK13	6527 a	9,0 ab	6,5 a			
CCD13 **CA** I	D13, conventional culture	6386 a	9,0 ab	6,5 a	7413 ab	8,3 abcd	4,8 cde
CCD13 **OA** I	D13, organic culture	5831 a	9,3 ab	6,3 a	7475 ab	8,2 abcd	4,0 e
OA I F13	D14, organic culture				7140 ab	8,9 a	4,5 de
OA II F13	D14, organic culture				7027 ab	7,9 bcd	4,5 de
OQ I F13	D14, organic culture				7138 ab	8,7 abc	5,3 cd
OQ II F13	D14, organic culture				6790 ab	8,5 abcd	5,0 cde
OY I F13	D14, organic culture				7568 a	7,8 cd	4,0 e
OY II F13	D14, organic culture				7589 a	7,8 d	5,0 cde
Lukullus	Biocer (France)	6363 a	9,5 a	4,8 b	6870 ab	8,5 abcd	5,8 bc
Midas	Biocer (France)	6908 a	8,9 ab	5,3 b	7228 ab	8,1 abcd	5,8 bc
Renan	Biocer (France)	6338 a	9,4 ab	6,3 a	6992 ab	8,8 ab	7,5 a
Skerzzo	Biocer (France)	6655 a	8,7 b	6,5 a	6949 ab	8,3 abcd	6,5 ab
Average		6383	9,1	6,1	7134	8,3	5,1
V.C.		5,4	2,9	5,4	5,5	4,5	9,7
p-value		0,19	0,01	0,00	0,01	0,00	0,00
Score 1= *9=*				*100% infest.* *no infest.*			*100% infest.* *no infest.*

Although, as expected, some differences between CCP's showed up in the results, no in-field characteristics could be proven to be significantly different. The high level of heterogeneity inside CCP plots may be one reason for these low statistical significances.

The results show that these CCP's of winter wheat can keep up with, if not equal the performance of modern reference varieties. The results also suggest that the CCP's have higher resilience, e.g. against yellow rust. Although based on only two years of trial, the results confirm observations by others who experimented with these CCP's.

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Using populations in organic agriculture: the role of local adaptation, farmers' selection and the awareness of the actors involved

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Introduction

Within the framework of the projects COBRA and DIVERSIFOOD, RSR and AIAB organised a trial in three organic farms in Italy. The trials aimed at better understanding the performances of soft wheat Composite Cross Populations in different environments, checking if they adapt to the growing environments, looking at the role of farmers in doing selection within populations and figuring out how the awareness of the actors involved in process of evolutionary plant breeding affect the process itself.

Materials and Methods

The three locations in Italy are organic farms in Toscana (farmer Rosario Floriddia – ENV1), Sicilia (farmer Giuseppe Li Rosi – ENV2) and Molise (farmer Vincenzo Battezzato – ENV3). Each trial has 10 entries of different materials: CCPs from different sources (SOLIBAM and COBRA projects), local varieties, one old variety and one modern variety as test (see table 1).

Table 1: Entries selected

N°	Entry	Type of Material
1	COBRA A	CCPs
2	COBRA Y	CCPs
3	COBRA Q	CCPs
4	SOLIBAM floriddia	CCPs
5	SOLIBAM li rosi	CCPs
6	selection floriddia	CCPs
7	maiorca	Local variety
8	andriolo	Local variety
9	frassineto	Old variety
10	emese	Modern variety

The entry SOLIBAM Floriddia was grown for 3 seasons in a plot and 1 season in open field in the farm in Toscana, the SOLIBAM Li Rosi on the contrary was grown the first year in a plot and then for three seasons in open field in Sicilia. The Selection Floriddia is a selection done by the farmer within the SOLIBAM Floriddia in 2013 and then grown for two seasons in the same farm in a plot for multiplication the seed. The COBRA entries were grown for two seasons in Floriddia farm in Toscana. The sowing density was 20gr/square metres and each plot was 10 square metres replicated twice in each farm. The sowing date was late for each trial compared to normal seasons: ENV1 05/01/2015; ENV2 19/12/2014; ENV3 13/12/2014. All the trials were harvested in the first half of July.

In all the farms we organised participatory evaluation trials during the farm days in June (see table 2 and poster for the outcomes). During the season we collected the following data from the plots: plant and spike height, presence of flag leaf, time of heading, ground coverage, lodging, presence of weeds and prevalent type of weeds.

Table 2: Evaluation field trials with different stakeholders

Farm	date	N° people	Participants
ENV1	12-13 June	39	23 Males, 16 females 11 farmers, 10 techinicians, 18 other stakeholders
ENV2	22 June	29	22 Males, 7 females 12 farmers, 6 techinicians, 11 other stakeholders
ENV3	26 June	13	23 Males, 2 females 7 farmers, 2 techinicians, 4 other stakeholders

Results and discussions

Based on the qualitative evaluation done by the participants to the farm days and on the yield data we have elaborated some preliminary results (see poster for the graphs). The best yielding varieties were Selection Floriddia in ENV1, SOLIBAM Li Rosi in ENV2 and Maiorca in ENV3. This result was expected because in ENV1 and 2 the two populations had time to adapt to these environments, whereas in ENV3 they arrived just for the experiment, so they were at the first season. The qualitative data confirm this outcome, but the preferences of the stakeholders were not directly linked to yield. In general COBRA CCPs had poor performances, maybe because they were selected for North Europe (see following graphs). The next step will be to analyse all the data collected and therefore better describe the performances of CCPs and understand the preferences expressed during the farm days.

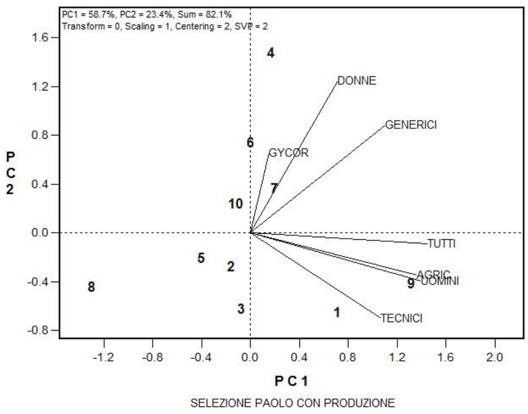

Figure 1: ENV3 outcomes

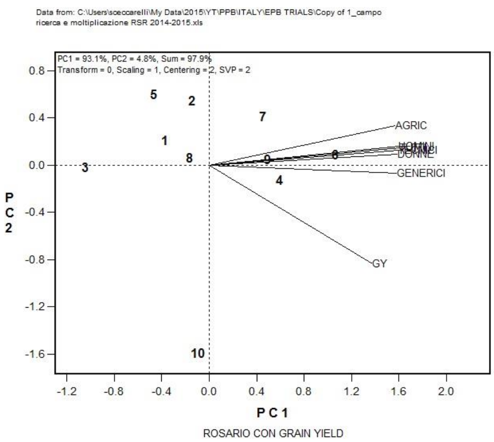

Figure 2: ENV1 outcomes

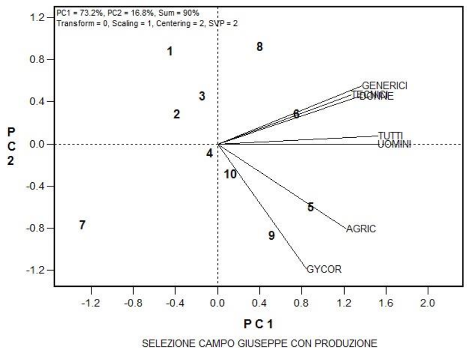

Figure 3: ENV2 outcomes

Agronomic evaluation of the first population-varieties developed within the wheat participatory breeding program in France

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Introduction

Participatory plant breeding (PPB) is based on the decentralization of selection in the farmers' fields and on their involvement in decision making at all steps of the breeding scheme. It allows for the development of varieties that may be finely adapted to the local pedo-climatic conditions, to the farmers agronomic practices and to the type of products and marketing that is applied to this production. Most often in such PPB programs, the varieties developed are population-varieties, i.e. genetically heterogeneous varieties derived from one or several crosses, or from the mixture of crosses and/or of landaces, where diversity has been maintained at a certain level determined by farmers selection practices.

Such a PPB approach has been applied on bread wheat in France since 2006 in a partnership among INRA Le Moulon (France) and groups of the farmers' NGO Réseau Semences Paysannes (RSP) (Dawson et al. 2011; Rivière et al. 2014). In this program, specific protocols, experimental designs and statistical methods have been developed for on-farm trials (Rivière et al. 2015). Moreover, a collective organization among farmers, facilitators and the research team has been set up. Here, we present preliminary results of a 2 years agronomic evaluation of the first population-varieties that have developed within this PPB program.

Materials and methods

Ten population-varieties developed within the bread wheat french PPB program (hereafter called *PPB varieties*) were proposed by five farmers invoved in the project. They cover a wide range of the possible types of population-varieties that are usually derived from PPB: a landrace grown and selected on farm for several years, a mass selection of one particular plant within a landrace, a single cross not fixed population, a mix of several landraces, a mix of several (up to 20) single-cross populations, a mix of both landraces and single-cross populations. The 10 PPB varieties have been evaluated during two years (2013-2014 and 2014-2015) in six organic farms of farmers involved in the PPB project among whom four had proposed their PPB varieties. Two commercial varieties Renan and Hendrix among the most frequently used by organic farmers in France have been included in the experiment. Each trial consisted in a two complete randomized blocks design. Agronomic management was organic but varied depending on the local usual practices of the farmers (input of organic manure or not, preceding crop, sowing date, harrowing or manual weeding or no intervention,…). Soil fertility and quality drastically veried among farms with some very superficial soil and some deeper and more fertile ones. Climates also were contrasted with one farm located in a very dry area in the south of France while others in the Alps had quite cold temperature in winter.

Grain yield, thousand kernel weight (TKW), protein content have been measured at the plot level while plant height, spike weight and some morphological traits have been measured on individual plants (25 plants / plot). Data were analyzed using an AMMI (Additive Main effects and Multiplicative

Interaction) model with *population*, *farm* and *year* as main effets, *block* within farm as a nested effect, and all three second order interactions effects among population, farm and year. Principal component analysis was run on the *Pop* x *Farm* and *Pop* x *Year* interactions terms and Wricke's ecovalence were estimated for each variable.

Results and discussions

Results from the analysis of variance showed that *Pop*, *Farm* and *Pop* x *Farm* interaction effects strongly varied in significance and magnitude according to the trait considered. While *Pop* and *Farm* effects were of similar importance and interaction was limited for plant height, the interaction was of the same order than *Pop* and *Farm* for spike length, spike weight and TKW; for grain yield, the *Farm* effect was the most important with *Pop* x *Farm* the second being three times larger than the main *Pop* effect. Moreover, when comparing the overall grain yield per population, only four PPB varieties were significantly less productive than the two commercial varieties, the six others did differ significantly. Depending on the farm and on the year, Renan performed best (on one or two farms), Hendrix performed best (on one farm) or some PPB varieties did better (on three or four farms), but in all cases, some PPB varieties had very interesting responses when considering grain yield as well as biomass and protein content. Therefore these preliminary results seemed promising to the farmers involved in the PPB process and the number of participants is increasing each year.
Statistical analyses are still ongoing and the agronomical results will be complemented with nutritional and sensory characterization of the varieties, which is of primary importance for the farmers (Vindras-Fouillet et al. 2014).

Acknowledgements

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On farm breeding strategies for diversity in winter soft wheat

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Introduction

In Europe and other developed countries, Participatory Plant Breeding (PBB) was an answer to the diversified needs of organic agricultures and their environments (Dawson et al, 2011). The basic hypotheses are: (1) inter and intra-varietal diversities are the best way to increase the sustainability of the agroecosystems (Wolfe et al, 2008) and (2) crops are best adapted if the plants are selected in the conditions for which they will be exploited (Dawson et al, 2012). In France, PBB started in the 2000 between the Peasant Seeds Network (RSP) and INRA teams (Chable et al, 2014). The RSP brings together 80 seed associations whose aim is to increase agricultural biodiversity and to sustain the development of an informal seed system. One issue is to evaluate strategies of plant breeding to increase the availability of new populations in diversified conditions of farming and baking. With farmers, a common experiment had been conceived to compare two strategies already met on farms: creation of **dynamic population** mixtures and **composite cross population** (CCP). In the framework of COBRA project, the aim was to create and assess two experimental populations from these both techniques on the basis on the same initial parents.

Materials and methods

A dynamic population and a CCP (Cross Composite population) were created during the first year of the project with the following populations which were previously evaluated by farmers from genetic resources provided by gene banks:
- Saint Priest et le Vernois rouge (VPRS)
- Bladette de Provence (BP)
- Redon Guer B7 (RGB7)
- Redon Guer 335 (RG335)
- Redon Sixt /Aff 346 (RSA)
- Redon Roux pale 1.13 (RRP)

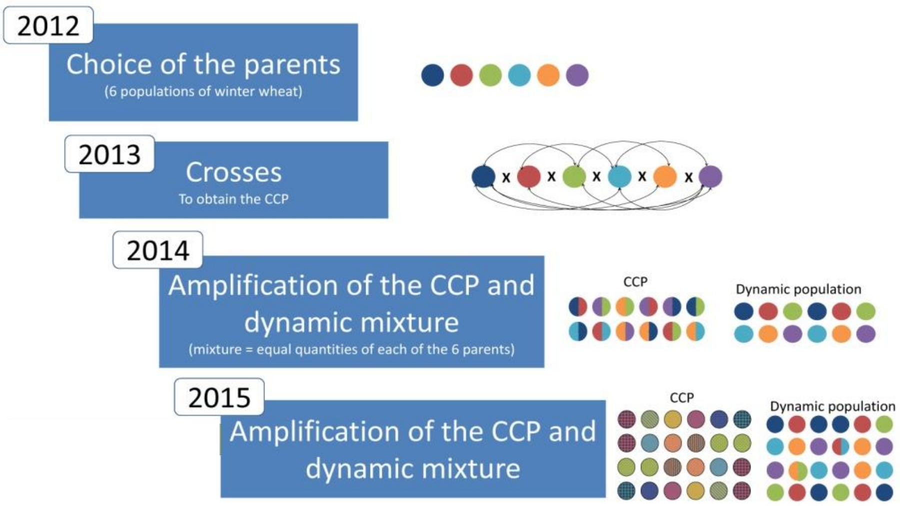

Figure 1: scheme of creation of two new populations from 6 landraces of soft wheat

The chosen wheat populations as parents are supposed to have a broad genetic base. The mixture of these parental populations is called **dynamic population** because it evolves year after year thanks to environmental constraints and farmers' practices; the evolution is designed through natural crosses between plants and the evolution of the relative rate of each initial parents' composition. The "CCP" (for "**Composite Cross Population**") is created by the mixture of the progenies of crosses two by two of each single population (see Figure 1). Several plants were crossed from each parental populations to represent most of the internal variability of the parents.

Results and discussions

The seeds from the experiment in 2014 have ensured the implementation of the 2015 trial, sown in autumn 2014. This trials were duplicated on two farms for further amplification. First preliminary observations have been performed on 2015, to observe the diversity inside the dynamic population and the CCP, on some phonological and morphological criteria. The CPP group has shown a larger variability.

From 2016, the experiment will be enlarged building a network of farmers interested in on farm selection. Then, in the framework of DIVERSIFOOD (Embedding crop diversity and networking for local high quality food systems, an H2020 project 2015-2019), we will study natural evolution and breeding impact on the diversity of both populations in contrasted conditions. The issue is to evaluate strategies according to expected agronomical performance and end-use/sensory qualities, in diversified conditions. This aims to help farmers to determine their own strategy taking into account previous observations on landraces and their own objectives.

Compared evaluation of the CCP and dynamic populations with different farmers will enhance methodological approach for (i) the creation population-varieties adapted to local conditions and practices of farmers, (ii) the development of methods and operational tools for management / biodiversity selection grown on the farm and (iii) strengthen skills and autonomy of farmers in management / seed selection .

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Organic seeds and plant breeding
from the seed companies' perspective

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Introduction

All organic agricultural systems suffer from a lack of plant cultivars adapted to organic production. Within the framework of the European projects SOLIBAM* (Strategies for Organic and Low Input Breeding and Management) and COBRA** (Coordinating Organic Plant Breeding Activities for diversity) a study was undertaken to provide an overview of the seed companies' breeding strategies for the organic sector and their viewpoints about the organic seed production.

Results highlight that the organic seed market has grown significantly in some countries (mostly in Northern Europe) in the last three years. This growth is however hampered by an easy granting derogation policy in some countries and technical difficulties in the field multiplication. Several companies surveyed are currently carrying out organic breeding programmes but they still remain relatively few because of the lack of return on investment and the absence of rules adapted to the registration of these varieties bred for the organic sector.

Materials and methods

Within the framework of the European projects SOLIBAM and COBRA a study was undertaken to provide an overview of the seed companies' breeding strategies for the organic sector and their viewpoints about the organic seed production. This study based on an online questionnaire was launched in September 2013 and supported by the projects' partners who allowed a wide and efficient dissemination across Europe. The preparation of the 7th European Workshop on Organic Seed Regulation (Oct. 13) by the European Consortium for Organic plant breeding (ECO-PB) provided a great opportunity to involve organic stakeholders in this study.

Results and discussions

Contributors' description

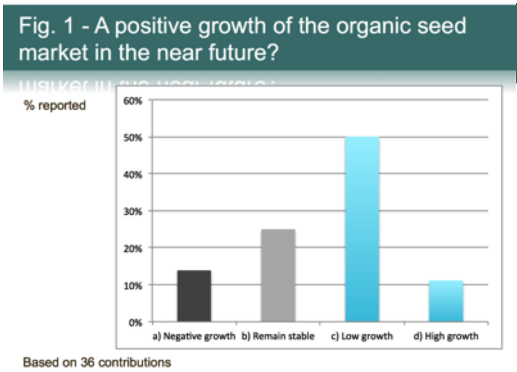
Almost half of the 36 contributors* came either from France or the United Kingdom, where the study had probably been more efficiently forwarded. Other responses came from The Netherlands, Austria, Denmark, Switzerland, Germany, Latvia and Bulgaria.

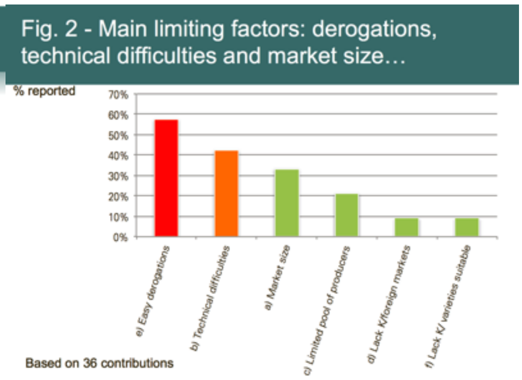
Most of the responders were companies producing vegetable and cereal seeds engaged both in conventional and organic seed production.

**List of contributors in § "Acknowledgements"*

Organic seed sales

According to the 36 contributors, it is in France, in the UK, in Germany and in Austria that their sales of organic seeds have increased the most over the last three years. The majority of the responders also estimated that the organic seed market will continue to grow in the near future, but quite slightly (Fig. 1), mainly because of the easy granting of derogations for conventional seeds treated after harvest. Technical difficulties and the lack of market opportunities were also cited as limiting factors (Fig. 2).

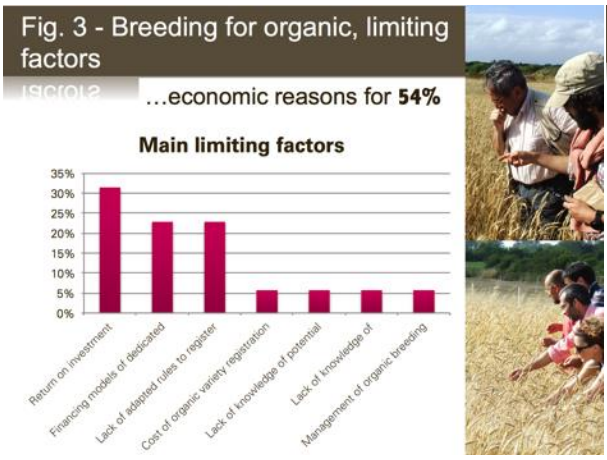


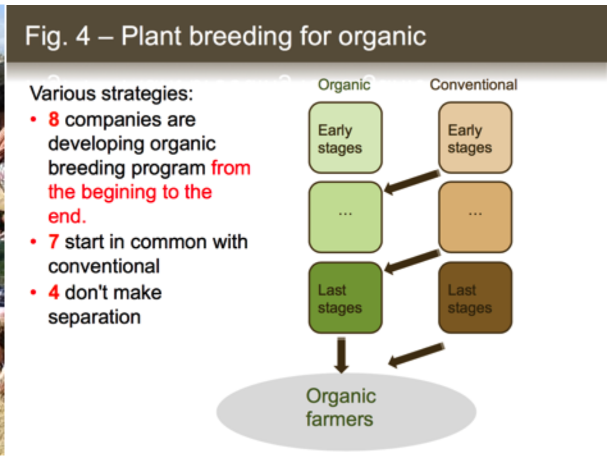


Plant breeding for organic

We can bring out 3 different breeding strategies for organic systems (Fig. 4): 1) Programmes that are organic from the very beginning of the breeding process; 2) Programmes starting with a conventional approach and switching to organic in later stages; 3) No dedicated organic breeding programmes (entirely conventional).

For 54% of the companies, the main limiting factor to further development of dedicated organic plant breeding programmes is economic (Fig. 3). 22 % of the responders have also cited the lack of adapted rules for organic seed registration as a major impediment.





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Pea breeding for organic systems: genomic selection for higher grain yield

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Genotyping-by-sequencing (GBS) can generate high numbers of SNP markers for a definitely lower cost per data point compared to SNP array platforms. Its use may allow to define genomic selection models for production traits (as established in animal breeding), in order to accelerate yield gains through greater selection intensity or to decrease phenotyping costs. This study provides an unprecedented assessment of the potential value of genomic selection for pea grain yield based on GBS data. Phenotyping data related to 105 pea inbred lines for each of three connected crosses [Attika × Isard (A×I); Attika × Kaspa (A×K); Kaspa × Isard (K×I)]. The 315 lines were phenotyped in two climatically-contrasting, organically-managed rainfed environments of Italy, i.e., Lodi (northern Italy) and Perugia (central Italy), using alfa-lattice experiments with three replications. Besides grain yield, we also assessed the ability of genomic selection to predict pea genotype value as estimated by a visual agronomic score ranging from 1=very poor to 9=excellent that was assigned to lines by groups of farmers during grain filling in a farmers' participatory breeding experience.

Grain yield across all lines ranged from 4.44 to 9.02 t/ha in Lodi, and from 1.75 to 4.42 t/ha in Perugia. Farmers' score ranged from 3.5 to 6.1 in Lodi, and from 3.1 to 5.7 in Perugia. GBS generated at least 4,000 and 2,500 polymorphic SNP markers per cross when requiring at least 4 and 6 reads for SNP genotype calling, respectively. The accuracy (i.e., the correlation between genome-enabled and actual data, assessed via cross-validation) of an rrBLUP-based genomic selection model differed between crosses, being low only for one cross featuring lower marker number and lower phenotypic variation (A×I). For the other two crosses, genome-enabled predictions were quite accurate ($r > 0.50$) for both mean yield and mean farmers' score across locations of the advanced lines.

Suitability as protein-rich animal fodder and previous crop value of different grain legume cropping systems in organic agriculture in Luxembourg

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In Central Europe, grain legumes are highly important crops needed as protein-rich animal fodder. There exist no studies comparing the most common grain legume cropping systems on one site, a commercial farm with less favorable soil conditions as present in Luxembourg, to show the suitability and the previous crop values of these cropping systems.

The aim of this study was to (i) determine the suitability of grain legumes in different cropping systems for cultivation as protein-rich animal fodder, (ii) compare the performance of winter and spring types of faba beans and peas, as well as to compare the performance of peas sown in pure stand and in mixture with cereals, and (iii) examine the previous crop value of different grain legume cropping systems on succeeding wheat in organic agriculture in Luxembourg. Field trials were undertaken on a commercial farm in Luxembourg. Eight different grain legume cropping systems (winter and spring peas in pure stand and in mixture with cereals, winter and spring faba beans, soybean, blue lupin) and a non-nitrogen fixing control crop (winter triticale) were cultivated followed by winter wheat in two consecutive seasons (2011/12 – 2012/13 and 2012/13- 2013/14).

Even under the given poor soil conditions, the tested grain legume cropping systems with the exception of winter pea in pure stand, showed good suitability for protein-rich fodder production. Faba beans were the best choice for cultivation as protein-rich animal fodder, in the year with sufficient soil moisture. Semi-leafless peas, which have a lower risk of lodging, perform better when sown in pure stand, while full-leaf types need a supporting crop to reach good yield. Spring pea performed better than winter pea, which again can be explained by the leaf type and not by the sowing time. For faba beans there was no difference between winter and spring faba beans.

Monocropped cereals had a lower previous crop value compared to cereals in mixture with grain legumes. In the first experimental sequence, non-nitrogen related beneficial effects of the grain legumes prevailed and it was spring pea in pure stand which showed the highest previous crop value. In the second experimental sequence grain legumes grown in pure showed a higher previous crop value compared to winter triticale or grain legumes grown in mixture with a cereals partner, due to a difference in mineralization of residues and consequently, a higher nitrogen supply.

Screening of Soybean (*Glycine max* L.) Advanced Lines under Organic Management in Turkey

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Key words: Soybean, *Glycine max* L., Advanced Lines, Breeding, Organic Management, Yield, Seed Protein Content.

Summary: The breeding research of soybean (*Glycine max* L.) began with advanced lines of "Soybean Breeding Project" supported by MFAL-GDAR in 2013 under organic management for comparing grain legume crops of organic breeding programme of COBRA (Coordinating Organic Plant Breeding Activities for Diversity) project because of priority for this species to be produced organically in Turkey for organic sector at Organic Open Field Experimental Area of AARI located in the Mediterranean Region. Main objective of the research is screening the advanced lines of soybean genotypes under organic management to determine appropriate, adapted and stable line/s of breeding programme. For this purpose, trials of 6 advanced lines of second crop soybean genotypes were set up successive two years in 2013 and 2014 during summer season. The trials of 6 soybean lines and 2 registered varieties in year 1 and 2 showed that many lines in terms of grain yield and some of yield components such as 100-seed weight, number of pod per plant, plant height, and number of days to flowering, number of lateral branch were significantly different and superior to standard registered varieties of AARI which have been produced in different parts of Turkey mainly in Aegean and South East Anatolia Regions. Additionally, min. and max. seed protein contents of soybean lines ranged as 31.8-41.2% in 2013 and 32.9-43.1% in 2014, respectively. Protein contents were found remarkable in terms of advanced lines especially for three genotypes which were showed higher contents than registered varieties although data was not determined significantly different, statistically. It is found that there is strong possibility and encouraging results to obtain high yielding and quality soybean varieties suitable for organic farming.

Introduction: Organic farming (O) requires varieties which are especially adapted to low input cropping system. As a consequence, researchers breeding approach to develop region-specific genotypes of soybean for organic conditions. O has been increasingly attaining interest in world wide. Turkey is one of the ten countries with the most organic producers in 2013 in the world. O area of Turkey occupied 1.9 % of the total agricultural area in 2013. For comparison, the share of O area in total agricultural area was 3.9% in France, 6.5% in Spain and 10.3% in Italy in 2013 (Anonymous, 2015). Turkey has O law (25659/3.12.2004) and legislations (27676/18.8.2010) modified in 2013 and they refrains from using agrochemical inputs. Major crops of organic products are dried fig, apricot and grape but number of crops reached 208 in 2013 whereas it was 150 in 2002. In Turkey 7 Soybean registered varieties have been using for production and the research is the first evaluation some of varieties lines for O. Turkey import considerable amount of soybean and soybean products for both conventional (C) and O sectors. Turkey's production of C and O soybean were 180 000 and 691 ton in 2013, respectively. It is well known that the use of modern cultivars in the organic context does not imply that these are the best cultivars for the O system. Main objective of the research is to develop soybean varieties adapted to Aegean Region which are competitive, high-yielding, herbicide and nematode tolerant, and low input usage, from advanced lines of AARI for O practices which were performed high yield and quality under trials of conventional management last year's to increase soybean production in Turkey.

Materials and Methods: The research was carried out with 6 advanced lines and 2 registered varieties (Cinsoy and Umut2002) of AARI in Randomized Block Design with 3 replicates, 4 rows, 5 m row length, 20 plant/m^2 densities (45x5 cm) for two successive years (2013 and 2014) as second crop at Organic Experimental Area of AARI located in the Aegean Region of Turkey. The characters examined were grain yield (kg/ha), 100 grain weight (g), Plant height (cm), Pod number per plant, branch number, number of days to flowering and seed protein content (%).UPOV and Bioversity International descriptors used for observations. UPOV and Bioversity International descriptors used for observations. Seed protein content was determined by Dumas combustion method using RapidN Cube and conversion factor was 6.25. Statistical data analysis was performed by JMP 7 (SAS, 1997).

Results and Discussion: The soybean breeding program of AARI selects high-yielding varieties through extensive testing in various locations in Turkey in order to release premium soybean varieties to benefit of soybean growers. This is achieved by establishing hundreds of populations with diverse genetic background and screening hundreds of distinct breeding lines. 100-seed weight (g) (17,5-21), number of pod per plant (56-87), plant height (96-140 cm), and number of days to flowering (27-33), number of lateral branch (1-7) were significantly different and superior to standard registered varieties of AARI (Cinsoy and Umut2002) which have been producing in different parts of Turkey mainly in Aegean and South East Anatolia Regions, however yield capacity of Cinsoy and Umut2002 are high under C (2500-4500 and 2500-3300 kg/ha, respectively) and protein contents approximately 35-40% depending on environment and genotype. Both are important for both feed and food utilization of soybean, so; seeds from all entries of trials are routinely tested for seed protein content. Totally high level results obtained for samples and mean values determined between 35.2-40.9%. The highest seed protein content was found for ETA3 while it showed the lowest grain yield for both years. Vollmann *et al.* (2000) reported negative correlation between soybean yield and seed protein content like many others. Grain yields were obtained between 2573-3849 and 1677-4116 kg/ha in 2013 and 2014, respectively. In 2013 the highest yield belonged to 68 (3849 kg/ha), in 2014 69 gave the highest yield (4116 kg/ha)(Figure 1, 2). Concerning with grain yield, yield components, and seed protein contents 68 and 69 gave promising results as evaluated totally. As a result, it is clear that at the end of the research new soybean varieties will be gained organic sector and usage of farmers producing traditionally.

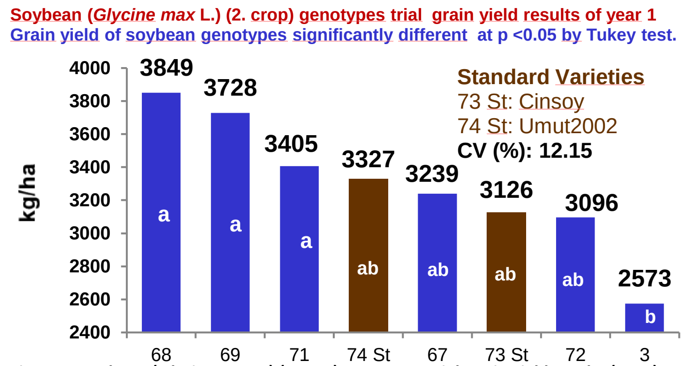

Figure 1. Soybean (*Glycine max* L.) (2.crop) genotypes trial grain yield results (2013)

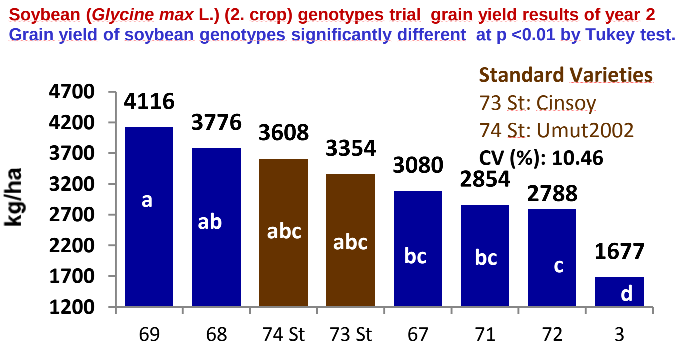

Figure 2. Soybean (*Glycine max* L.) (2.crop) genotypes trial grain yield results (2014)

Acknowledgements: The work reported here is supported by the EU Core Organic II project COBRA (Coordinating Organic Plant Breeding Activities for Diversity).

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Genetic Diversity Assessment of *Vicia faba* Accessions Taken from the Slovenian Plant Gene Bank

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Introduction

The Slovenian plant gene bank is rich in *Vicia faba* accessions. The first collection and evaluation of 34 accessions of *Vicia faba* showed that the most accessions appeared to be of the equina type, one minor and the rest of the major type (Meglič, 1988). At present the gene bank houses 41 *Vicia faba* acessions, of which 39 are abundant enough to be included in research. In the study reported here, all faba beans from the gene bank were assessed for genetic diversity using microsatellite markers with the aim to decipher the genetic relationships among the accessions and identify putative duplicates in the collection.

Material and methods

A total of 39 accessions of *Vicia faba* from the Slovenian plant gene bank, and three additional accessions from private faba producers were included in a preliminary assessment of genetic diversity using eight microsatellite markers. The DNA was extracted from two pools of four individual plants of each accession. A set of eight fluorescentlly labelled microsatellite primer pairs developed by Zeid et al. (2009) were used in the PCR. Based on the polymorphisms obtained from the 3130xL Genetic Analyzer (Life Technologies), a cluster analysis was deployed to reveal the genetic relationships among the accessions. Each SSR allele was scored for the presence (1) and absence (0) of each accession. The binary data matrix was used to calculate Jaccard's similarity coefficients (Jaccard, 1908); GSij = a/(a+b+c), where GSij is the similarity between two individuals i and j, a is the number of bands present in both i and j, b is the number of bands present in i and absent in j and c is the number of bands present in j and absent in i. Associations among the accessions were computed using the Neighbor-Joining algorithm in the NTSYS-pc package (Rohlf, 1998).

Results and discussion

In the analysis of eight SSR loci 155 alleles were detected (on average 19.4 alleles per primer pair), of which 154 (99.4%) were polymorphic. Only one allele was common to all of the studied accessions. Most alleles (30) were scored at locus VfG 1,

followed by VfG 41 (24) and VfG 9 (23). The smallest number of alleles (10) were scored at VfG 44.
The microsatellite data was used in a cluster analysis to evaluate the relationships between the accessions (Figure 1). All accessions were successfully distinguished, indicating the high efficiency of microsatellite markers to resolve the genetic identity of faba beans. Four clusters were identified in the NJ dendrogram, two large and two small ones. Since morphological characterization of the accessions is still in progress, phenotypical features that associate accessions into the clusters and traits that differentiate them, remain to be unravelled.

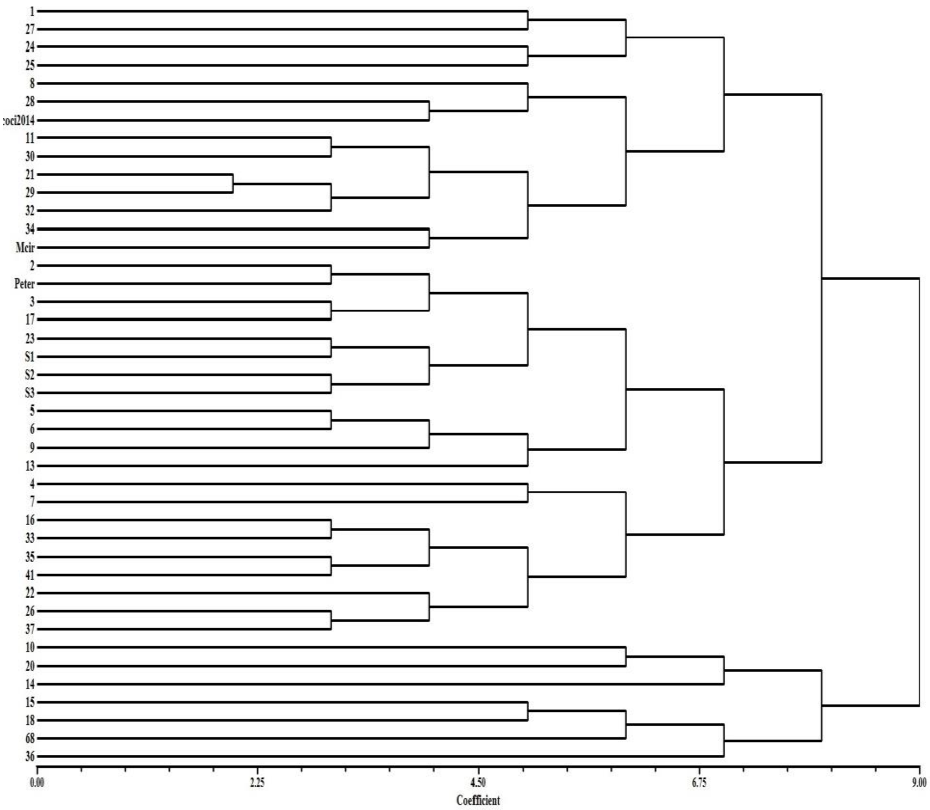

Figure 1. NJ dendrogram showing the genetic relationships among 42 faba bean genotypes

Acknowledgements
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Allelophathy of the Barley Plant on in Vitro Pea Emergence

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Introduction

Many species of weeds, as well as crop plants, are known to be allelopathic. Allelochemicals are secondary metabolites present in different plant organs (Rice, 1984), which may substantially differ in allelopathic activity (Ciarka et al., 2009; Grobelnik Mlakar et al., 2012). Besides other factors, allelopathy also plays an important role in successful intercropping systems. This particular study was derived from field observations carried out in barley–pea intercropping systems examined at the University Agricultural Centre of Faculty of Agriculture and Life Sciences, Maribor. A series of aqueous extracts from different plant parts of barley were bioassayed to determine the phytotoxic effects on pea germination.

Material and methods

The barley plants ('Sandra') and pea plants ('Kenzzo') were collected on the field in 2015, when barley was close to maturity, and the peas were in the milk-waxy maturity stage. The sampled plants were prepared as a whole, as aboveground parts, or separated into roots, stems, leaves, and the inflorescences (whole spikes in barley, pods and seeds in pea). The material was chopped (0.5 cm) and 25 g lots were soaked in 500 mL of distilled water (24 h) at an ambient temperature. Extracts were filtered and stored in dark bottles. In the bioassay, the extracts were used as the full-concentrated stock (100%) or diluted to 50%. Before storage, filtrates were filled with argon to displace the oxygen. In the bioassay, where allelopathic effects of peas were examined, barley went into the stage of seed-decaying (fungal spoilage), therefore only the allelopathic effects of barley are presented. Twenty-five pea seeds were evenly placed into a sterile Petri dish lined with filter paper. The filter paper was moist (10 mL) with extracts or with distilled water (control). The sealed Petri dishes were incubated at 25 °C, 12/12 h of light/darkness. Germination (radicle visible) was recorded every 24 h. Besides the total germination (Gt), the speed of germination (S) was also evaluated (Anjum and Bajwa, 2005). The average fresh weight of a seedling was calculated after weighing all germinated seedlings per Petri dish. The obtained results were subjected to ANOVA, and comparisons of means were performed by the Duncan and by the *t*-test. The results are presented as the means ± standard errors (SEM) of six replications.

Results and discussion

The inflorescence extract of barley (both concentrations) significantly reduced the total seed germination (Gt) of pea when compared to the control. Total seed germination was affected in stem and leaf extracts when applied in lower – 50% concentration. The speed of germination (S) is a more sensitive index than the Gt index; extracts from all plant parts negatively affected the speed of germination (averaged across concentrations 55.3 to 85.0) in comparison to the control (96.7). Extracts from the whole barley plant and from the inflorescence resulted in a higher seedling weight in comparison to the control (Table 1).

Table 1: Effects of the barley extracts and concentration (C) on germination indices (total germination, Gt; speed of germination, S) and on fresh weight (FW) of pea.

Plant part	C (%)	Gt (%)	S	FW (g)
Root	100	90.7±3.53a	62.9±4.18a	0.67±0.01a
	50	93.3±2.67a	55.3±2.72b*	0.70±0.02a
Mean		92.0	59.2*	0.69
Above-ground	100	94.7±1.33a	80.4±1011a	0.71±0.00b
	50	89.3±1.33a*	77.6±0.24b	0.78±0.02a*
Mean		92.0	79.0*	0.74
Whole plant	100	93.3±1.33a	67.5±7.31a*	0.72±0.01a
	50	90.7±2.67a	68.2±9.54a*	0.77±0.02a*
Mean		92.0	67.9*	0.75*
Stem	100	98.7±1.33a	79.8±3.21a	0.69±0.01b
	50	90.7±3.53b	70.6±2.34b*	0.76±0.01a*
Mean		94.7	75.2*	0.73
Leaf	100	89.3±4.81a*	58.1±6.43a*	0.61±0.03b*
	50	94.7±2.65b	72.4±13.23b*	0.74±0.03a*
Mean		92.0	65.2*	0.67
Inflore-scence	100	89.3±1.33a*	58.8±3.34b*	0.69±0.02b
	50	90.7±2.67a*	85.0±3.73a	0.81±0.01a*
Mean		90.0*	71.9*	0.75*
Control	0	97.3	96.7	0.68

$^{a–b}$ Means followed by different letters within plant parts are significantly different.
*Values followed by an asterisk are significantly different from those in the control.

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Suitability of different grain legume cropping systems as protein rich animal fodder in an ON-FARM field trial under organic growing conditions in Luxembourg

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Due to the protein deficit in Europe and the many negative impacts related to soy imports from overseas, it is important to increase the cultivation of grain legumes for regional fodder production. Therefore, an on-farm trial of different grain legume cropping systems was set up at two Luxembourgish locations, with the aim to study their fodder value and previous crop value, as well as their suitability for cultivation under organic growing conditions in Luxembourg.

In the growing season 2013/14, an ON-FARM strip trial with 7 different grain legume cropping systems was established at two commercial organic farms in Luxembourg (Colmar-Berg: 360 m above mean sea level (AMSL), mean annual precipitation 750 mm, mean annual temperature 8.8°C, soil type loam; and Bous: 210 m AMSL, mean annual precipitation 684 mm, mean annual temperature 9.4°C, soil type loam). The different grain legume cropping systems were winter pea in mixture with triticale (EFB 33/Massimo), winter faba bean (Hiverna), spring pea (Alvesta), spring faba bean (Fuego), blue lupin (Boregine), white lupin (Feodora) and winter triticale (Massimo), as a non-nitrogen-fixing control. These grain legume cropping systems were tested for their practical suitability with respect to cultivation techniques and fodder value. The previous crop value is also being assessed and the different grain legume cropping systems were followed by spelt in the growing season 2014/15.

The overall grain yield of the previous crops were markedly higher in Bous (26.6 dt/ha at 100 % DM) than in Colmar-Berg (17.0 dt/ha at 100% DM). The highest grain yield in Bous was reached with white lupin (33.3 dt/ha at 100% DM), and with spring pea (21.8 dt/ha at 100% DM) in Colmar-Berg. However, on both sites, the highest protein yield was achieved with white lupin (13.5 dt/ha at 100% DM in Bous and 6.1 dt/ha at 100% DM in Colmar-Berg), with the white lupin protein yield in Colmar-Berg being less than half that of Bous. As protein contents of the white lupin were about the same at both sites, the observed difference in protein yield can be explained by the difference in grain yield. The differences in grain yield levels in turn can be predominantly explained by the differences in soil composition: both soils are classified as loams, however, the soil in Colmar-Berg is very shallow with a large stone fraction, such that the root penetration and water holding capacity are much lower than in Bous. Overall, all the tested grain legumes, with the exception of winter faba bean in Colmar-Berg, showed good suitability for regional high-protein fodder production. This ON-FARM trial is being used to validate results under "real-life" conditions from an exact trial conducted in Colmar-Berg. In this exact trial in Colmar-Berg, it was observed that even faba bean is suitable for cultivation under the poor soil conditions at the site, when sufficient soil moisture can be assured.

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Organic field trials of winter and summer crop mixtures of grain legumes and cereals in Belgium

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Summer- and wintersown crop mixtures of faba beans or fodder peas together with wheat/barley/triticale were tested in field trials in the growing seasons 2013-2014 and 2014-2015. Different regionally available varieties and European plant material of faba bean and fodder pea were harvested together with cereals as dry grain. Special attention was paid to weed suppression capability and crop reliability.

Materials and methods
The trials were conducted at operating organic farms in Beitem and Lo-Reninge and at the conventional research farm in Bottelare (Belgium). All locations have a fertile sandy loam soil. The maritime climate is moderate. The trial was set up as a randomized block design with 4 replicate blocks with a net plot size of 20 m².

The small demand for organic cereal and legume seeds in Belgium is for the most part filled by foreign seed providers. The tested varieties are a selection of their suitable offer for the region. Five breeding lines of winter pea from the Getreidezüchtungsforschung Darzau (D) were tested. Also a separate trial with breeding lines of summer pea and faba bean from Latvia was conducted but is not in the scope of this article.

Summer crop mixtures
In these summer trials, varieties of faba bean together with summer wheat var. Epos (2014) and varieties of fodder pea together with summer barley var. Calcule (2014, 2015) were sown at the start of April. Early crop development and weed suppression were good but reliability towards the end of the season was low. Late ripening of faba bean together with bad weather conditions caused lodging and breaking and subsequent foraging by birds by the time of harvest (Figure 1). This was even more the case in the pure stand where the faba bean grew taller. The fodder peas were susceptible to lodging and foraging by birds, even though plant density was moderate and all varieties were semileafless.

Suggested focus:
Faba bean: early ripening, resistance against lodging (shortness, sturdiness)
Fodder pea: resistance against lodging (semileafless, short, twining types)

Winter crop mixtures
In these winter trials, varieties of faba bean and fodder pea together with triticale var. Borodine (2013-2014) and var. Vuka (2014-2015) were sown at the start of November. The vigorous triticale component suppressed weeds sufficiently in both seasons. Faba bean with triticale resulted in a reliable crop in both years. In the first year, high plant density of fodder pea caused lodging by the time of harvest. The second year, plant density of fodder pea was

too low because of foraging by slugs and wildlife during late winter and early spring (Figure 2). The 5 pea lines from Darzau could not be assessed due to very low presence in the crop.

Suggested focus:
Faba bean: winter hardiness, resistance against lodging (shortness, sturdiness)
Fodder pea: winter hardiness, post-winter resilience (slugs, birds, wildlife) and growth vigour, resistance against lodging (semileafless, short, twining types)

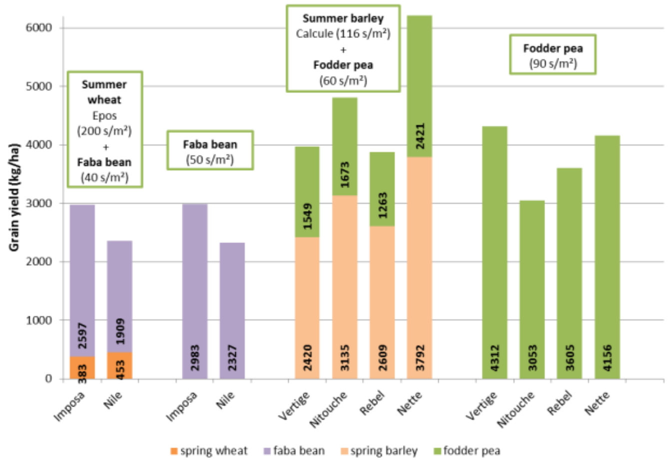

Figure 1: Summer crop mixtures of wheat/barley with faba bean/fodder pea and pure stands of faba bean and fodder pea in Beitem in 2014.

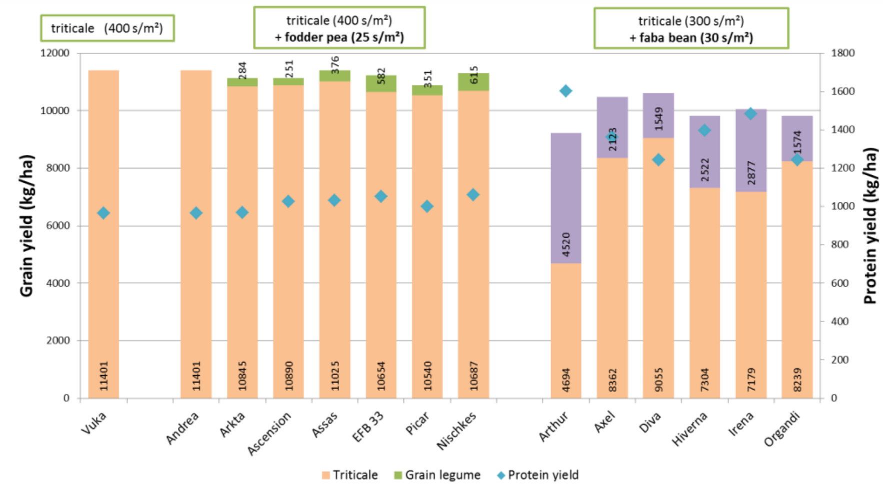

Figure 2: Winter crop mixtures triticale with faba bean/fodder pea in Lo-Reninge in 2014-2015.

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Effect of seed treatments with essential oils on plant emergence of ascochyta-infected pea

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Seed-borne diseases represent a critical problem for successful production, especially in organic agriculture where less plant protection agents are available. The ascochyta blight fungi complex (*Peyronellaea pinodella*, syn. *Phoma medicaginis* var. *pinodella*; *Peyronellaea pinodes*, syn. *Mycosphaerella pinodes*; *Ascochyta pisi*) is a major threat for field pea (*Pisum sativum*) cultivation.
Interest has grown in developing alternative measures to chemical treatments for crop and seed protection, including physical methods, biocontrol agents and plant extracts. Essential oils are metabolic plant products that have long been studied for their bactericidal and fungicidal properties due to the presence of various active compounds (e.g., alkaloids, phenols, flavonols, monoterpenes, sesquiterpenes and isoprenoids). Clove (*Syzygium aromaticum*) oil, tea tree (*Melaleuca alternifolia*) oil and thyme (*Thymus vulgaris*) oil are among those considered to be potentially useful in agriculture due to their effectiveness, non-toxicity to humans, biodegradability and easy availability. However, most results were obtained *in vitro*, and further investigations are needed with field experiments, to verify the effectiveness of essential oils as seed treatment in field conditions, and to assess any possible phytotoxicity on seeds.
Aim of this study was to test the efficacy in laboratory and field conditions of three essential oils and two seed application methods against artificially-inoculated ascochyta blight fungi. Pathogen control by oil application with seed submersion was promising in *in vitro* tests, thus confirming literature evidence. 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, although this effect seemed to be rather cultivar-specific. In addition to any oil effect, an excessive imbibition during seed submersion may have somehow damaged the outer seed coats, jeopardising proper germination of the embryo under field conditions. 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. If further confirmed, this technique would offer the possibility of using the oils as alternative natural and biodegradable antifungal agents.

Yield stability in winter wheat composite cross populations under organic and conventional conditions from the F_5 to the F_{13}.

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Keywords: Composite Cross Populations, evolutionary breeding, yield potential.

Introduction

The use of Composite Cross Populations (CCPs) as an alternative breeding approach, has gained significant interest, particularly due to the potential adaptability of evolving populations to specific locations. Three winter wheat (*Triticum aestivum* L.) CC populations were created in 2001 through collaboration between Elm Farm Research Centre (ORC) and the John Innes Institute in the UK. The populations were created through the inter-crossing of 20 parental varieties, in order to create a high yielding (Y) population; a high baking quality (Q) population and a population consisting of the 9 x 12 (YQ) intercrosses. Since 2005/6 (F_5), the populations have been maintained at Kassel University without conscious selection.

The objectives of the study are to increase the resilience, adaptability and performance of winter wheat through increased genetic diversity, as well as the improvement of breeding efficiency and the introduction of a novel breeding method that has the potential to provide an alternative source of germplasm in the form of CCPs. The adaptation of CCPs into mainstream practice is hampered by a number of factors including legal and quality aspects, but yield performance and stability of the CCPs are important criteria in view of increasing climatic unpredictability and for farmers (Finckh *et al*. 2000). Actual yields, as well as the yield stability of the populations relative to a common reference variety will be presented.

Materials and Methods

The CCPs have been grown under both organic and conventional conditions since the F_5 at the research fields of the University of Kassel in Neu Eichenberg, Germany (51°22"N, 9°54"E, 247m ASL, mean annual ppt:619mm, mean temp.: 7.9°C). The variety Capo was also grown alongside the CCPs under both organic and conventional conditions for comparison. The populations under both organic and conventional management have been maintained since 2005 in plot sizes >100m^2 and information regarding yield and its components have been recorded. The mean location yields per year, under organic and conventional management, were compared to the actual yields of the populations. Simple regression analysis (as described by Finckh *et al.*, 2000) was used to calculate the slope and fit of the model for the three populations (YQ, Q, Y) and of the reference variety Capo.

Results and Discussion

The mean combined yields of the organic and conventional populations over all years ranged from 4.54 t/ha for the Q population to 4.92 t/ha for the YQ population. The reference variety Capo had the highest mean combined yield of 5.60 t/ha. The regression of yields against the site mean show that the yields of YQ and Q CCPs increased proportional to the site potential (slopes close to 1.0 in Fig. 1). Yields of the Y CCPs increased more with site mean (1.06) indicating greater responsiveness to better growing conditions, while the slope of 0.86 of the reference variety Capo indicates that this variety performs relatively better at a lower yield potential. The high R^2 and low MSE (mean square error) values of the CCPs combined with slopes around 1 or greater indicate a better yield stability than for Capo where the slope was considerably lower.

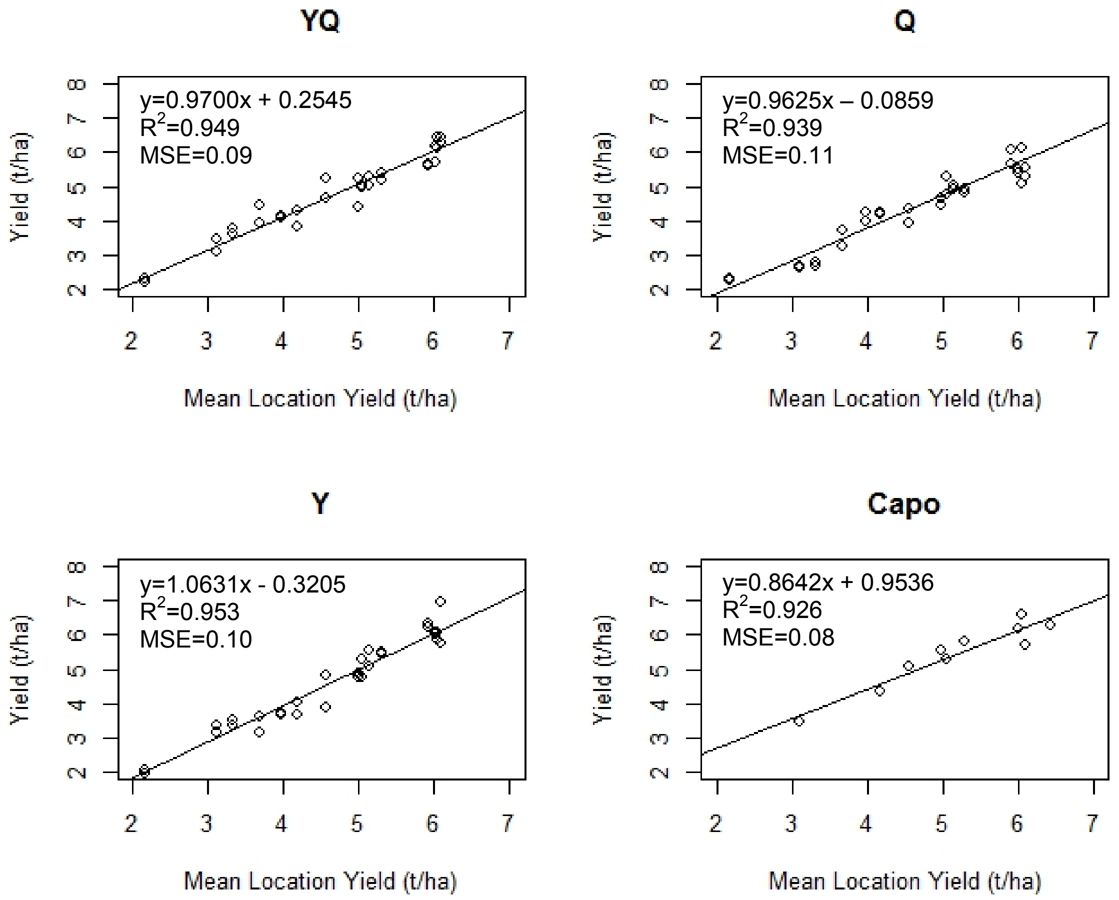

Figure 1: Effect of the overall yield potential of a site on the relative performance of wheat CCPs YQ, Y, Q, and the reference variety Capo over nine years (2006-2014). The actual yield of the CCPs and reference variety as compared to the annual mean location yield is plotted. The linear equation, adjusted R^2 and mean square error of the residuals (MSE) are shown.

Further analyses will be necessary to determine management system effects on yield stability of the CCPs, but also additional varieties over time. Additional data from 2015 will be included in the presentation.

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The effect of changing environmental conditions on composite cross winter wheat populations over six years from the F_8 to the F_{14}.

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Keywords: Composite cross populations, evolutionary breeding, genetic diversity

Introduction

A lack of appropriate crop varieties specifically bred for the higher biotic and abiotic stresses experienced in organic and low-input systems, has driven novel breeding approaches such as Composite Cross Populations (CCPs) and other genotype mixtures, thereby increasing both intra- and inter-varietal diversity. The creation of CCPs using carefully chosen parental varieties, coupled with the ability of the populations to adapt to local environments means that these populations are well suited for low-input and organic agricultural systems (Phillips and Wolfe, 2005). COBRA (Coordinating Organic plant BReeding Activities for Diversity) aims to “support and develop plant breeding and seed production in Europe by increasing the use of plant material with High genetic Diversity (Hi-D) through coordinating, linking and expanding existing breeding and research in cereals (wheat and barley) and grain legumes (pea and faba bean)”.

Three winter wheat (*Triticum aestivum* L.) CCPs were created in 2001, through collaboration with the Elm Farm Research Centre and the John Innes Institute. In 2005, seed batches of the F_4 of these populations were equally divided and distributed to three additional partners (France, Hungary and the University of Kassel). In 2007, it was decided to submit one of the CCPs to changes in environments every year. A pattern was developed between eight partners whereby these “cycling” populations would be grown in a plot of >100m^2 and sent to the next cycling partner the following year. The original partners (UK, Hungary and Germany) have also maintained their original “non-cycling” populations for comparison.

The aim of the project is to compare populations that all originated from the same seed batch in 2005, but that have been exposed to vastly different climatic conditions, in one site (Germany) for their performance and diversity (phenotypic diversity, yield and disease occurrence). 2014/15 is the second year of the project whereby both saved seed from 2013 and harvested seed from 2014 were sown, in order to compare two generations in one growing season and to ensure that the differences between populations were not in fact due to differing seed size and quality.

Materials and Methods

In 2013, seed from each of the eight “cycling” and three “non-cycling” populations were planted at the University of Kassel. Three commonly-used reference varieties (Achat, Akteur and Capo) were also used in the experiment for comparison. In 2014, saved seed from the original experiment was resown with newly harvested seed from the same populations (see Table 1). In addition, six CCPs that had been grown by six farmers in Germany for either two or three years were included in the experiment, as well as seven reference varieties. In both experimental years information regarding yield, disease incidence (foot and leaf) and phenotypic variation were recorded.

Table 1: CC populations, farmer populations and reference varieties sown in the second experimental year at the experimental farm Neu-Eichenberg, University of Kassel.

Populations (Code)*		**References**	**Farmer Populations**		**Cycling**
2013 Harvest	2014 Harvest (Uni Kassel)		Harvest 2013	Harvest 2014	
D13NCI**	D14NCI	Achat	Frankenhausen	Gut Fahrenbach	NL14
D13NCII**	D14NCII	Akteur	Maßhalderbuch	Sudershausen	
D13NC**	D14NC	Butaro	Niederbeisheim	Wartmannsroth	
HU13	HU13-D14	Capo			
DK13	DK13-D14	Naturastar			
TUM13	TUM13-D14	Scaro			
F13	F13-D14	Wiwa			
HU13NC**	HU13NC-D14				
UK13	UK13-D14				
CH13	CH13-D14				
D13	D13-D14				
NL13	NL13-D14				
UK13NC**	UK13NC-D14				

*Partner codes and institutes: CH = FIBL, Switzerland; D = Uni. Kassel, Germany; DK = Agrologica, Denmark; F = INRA, France; HU = Centre for Agricultural Research, Hungary; NL = Louis Bolk Institute, The Netherlands; TUM = Technical University München, Germany; UK = The Organic Research Centre, United Kingdom.
**NC = Non-cycling population

Results and Discussion

Results from the first experimental year indicate significant differences between the populations mainly in terms of morphological characteristics. Observations in the field during the growing period showed noticeable phenotypic differences between the populations in terms of plant height, ear length and colour and whether the ears were awned or awnless. These initial results indicate that these populations, although from the same origin, have evolved and maintained a high degree of diversity through the different climates and locations and over time. In addition, the yields of the populations were not significantly different from the reference varieties, indicating that the CC populations yield performance is comparable to commonly used reference varieties. The harvesting and phenotypic assessment of the populations and reference varieties in the 2014/2015 experimental year will be available by October 2015 and preliminary results will be included in the poster for the conference in November.

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Pea breeding for organic systems: evolutionary and farmer-participatory breeding approaches

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Single-seed descent (SSD), which usually implies nil or very limited selection until obtaining advanced lines, is the most popular breeding scheme for inbred crops such as pea. Bulk breeding schemes, in contrast, imply the occurrence of natural selection in segregating lines that are usually grown under intra-family competition. Late bulk breeding stages may be exploited either for isolating pure lines for further selection, or for obtaining evolutionary mixtures to introduce as such into cultivation. Both SSD and bulk breeding schemes could also be exploited to breed mixtures of elite pure lines (rather than pure lines). This study reports on the comparison of SSD and different bulk breeding schemes for ability to provide high-yielding pea material in organically-managed environments of two climatically-contrasting regions of Italy, i.e., northern Italy (Lodi) and central Italy (Perugia). We also report on the comparison of ordinary vs. farmers' participatory breeding approaches – as represented by indexes of selection established according to pea priority traits assigned by groups of breeders and farmers, respectively – applied to 315 inbred lines issued from three connected crosses (Attika × Isard; Attika × Kaspa; Kaspa × Isard) that were evaluated in northern and central Italy.

On the whole, our results suggested that bulk breeding, compared with SSD, not only is less expensive but also more capable of producing higher yielding pure lines - particularly for environments similar to those under which it was carried out. Evolutionary mixtures proved higher-yielding than bulk breeding-derived pure lines or narrow-based mixtures, in environments similar to those in which they evolved; whereas mixtures of best pure lines, or just pure lines, proved preferable in a wider adaptation context.

On average, farmers and breeders displayed a few differences in priority traits included in selection indexes, while showing fairly high consistency for line visual assessment. Farmers' selected lines tended to be higher yielding and more acceptable to farmers than breeders' selected lines. However, selections merely based on grain yield were nearly as good as selections based on farmers' selection index.

Insights about the temporary marketing experiment & the marketing of cereal populations.

BRUCE D PEARCE

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Increasing climatic variation has had, and will continue have an impact on crop production and the economic viability of farmers. A way to insure against these impacts is to increase the diversity on farm. Increased genetic diversity within the crop can be a component of this. Genetic diversity can be delivered by growing a greater number of crops or varieties separately or as a mixture or by growing composite cross populations (CCP). Since 2001 the Organic Research Centre has developed CCPs of winter wheat in organic and low input systems. Aligned with this work are activities with UK and EU policy makers to address the EU legal framework that would allow for the marketing of populations. After nearly 15 years of work we are now at a point where some cereal CCPs can be marketed under a temporary marketing experiment. The paper will cover the production and development of the populations as well as an analysis of their performance along with insights into the development of new seed regulation policies as well as our initial experiences of working within the new marketing experiment