Using crop-pathogen modeling to identify plant traits to control Zymoseptoria tritici epidemics on wheat

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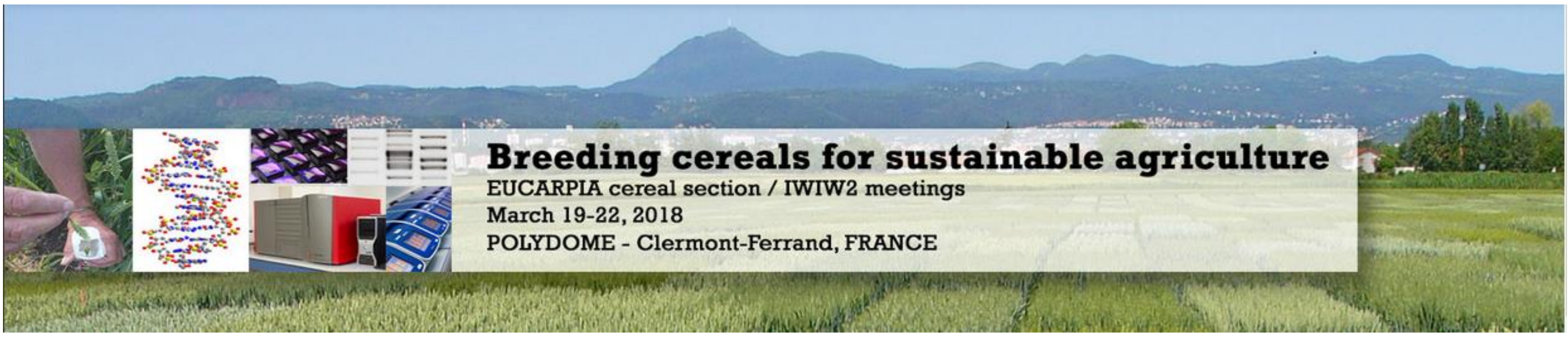

EUCARPIA

EUCARPIA Cereal section meeting

&

2nd International Wheat Innovation workshop

March 19th - 22th, 2018
Polydôme, Clermont-Ferrand, France

Polydôme, Clermont-Ferrand, France

EUCARPIA Cereal section meeting - March 19-21 2018 -

2nd International Wheat Innovation Workshop - March 22 2018

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Eucarpia Edito

Small grain cereals are the most widely grown annual crops worldwide. Wheat and barley are the two major cereals with cultivated surfaces of 230 and 60 Mha, respectively. Cereal grains have a huge range of utilization and wheat represents ~19% of human calorie intake, up to 50% in some countries. It is processed in many different products (breads, biscuits, breakfast, beer…). It is also widely used for ruminant and monogastric feed, and by biofuel (ethanol) or biomaterial industry.

Cereal production dramatically increased after the green revolution (1960-1990), but the production rate is now decreasing (see http://www.wheatinitiative.org/about-wheat/factsheets-infographics). The reason is a slight decrease of the surface cultivated with cereals because of competition with other crops or non-agricultural uses. Another reason is a slower yield progression, partially attributed to climate change. To face the challenge of food security for an ever-growing world population, cereal yield must be pushed upward, but not at any cost. Indeed, intensive production systems using fertilizers and pesticides are increasingly questioned for their negative impacts on the environment or human health. For example, nitrates in fresh or marine water cause eutrophication and pesticide residues may have carcinogenic effect or act as endocrine disruptors. Consumers and citizen are also more and more demanding for sustainable farming systems and safe products.

Plant breeding is often presented as a sustainable way to produce more and better. Recent developments in genomics, genotyping, phenotyping and breeding methodologies open new perspectives to design future varieties, with higher performance or resilience, targeting more specific conditions and utilizations, and more environmentally friendly.

The cereal section congress of EUCARPIA (www.eucarpia.org) is the opportunity to learn about recent developments in genetics and cereal breeding, from basic research on quantitative genetics, breeding methodology, crop modelling, genome organization and evolution to applied research on genome editing, QTL detection for marker assisted selection, plant-micro-organism interaction, biotic and abiotic stresses.

The 2nd edition of the International Wheat Innovation Workshop (IWIW2) is the opportunity to present complementary results obtained in major wheat programmes and pave the way for future collaborations and funding opportunities.

Gilles CHARMET
February 25th, 2018, Clermont-Ferrand, France
Head of the Eucarpia Organizing Committee

IWIW Edito

In recent years, many scientific and technological advances have opened up huge opportunities to select more efficiently for complex traits and accelerate the pace of genetic progress. These include the long-awaited availability of the wheat genome reference sequence and its annotation, the development of genomic selection and high throughput phenotyping methods, and the fine-tuning of increasingly precise and specific genome editing techniques...

How can we use these fantastic opportunities to meet the challenge of global change? The issue is now to define optimal strategies to combine all these results and tools, all the multidisciplinary skills and all the academic, private and institutional partners to breed for better adapted cereals cultivars. We designed this day of exchange and reflexion to help generate these strategies.

The second edition of the International Wheat Innovation Workshop (IWIW2) is indeed the opportunity to exchange on topics of interest with the wheat community and pave the way for future collaborations and funding opportunities. It is built around topics which have known major evolutions recently and will probably continue change rapidly: Genetic resources, Phenotyping, Genome editing, Genomic selection and Bio-informatics.

This workshop is organized at the initiative of the BreedWheat project, and associates some of the major wheat programmes: Design Future Wheat program (UK BBSRC), proWeizen (German research and breeding alliance) and CRP Wheat (CGIAR challenge program) as well as the Wheat Initiative.

We thank in advance the speakers that accepted to describe the state of the art and speculate about the future in introductory and impulse talks that will feed the group sessions. We hope that this day and the World Café will be fruitful to all and lead to new networks of collaborations and relevant projects.

Jacques LE GOUIS
March 2nd, 2018, Clermont-Ferrand, France
Head of the IWIW Organizing Committee

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EUCARPIA
SCIENTIFIC PROGRAMME

Monday March 19th, 2018

7h30 am - **9h30** am	**Registration & Welcome coffee** (poster hanging)
9h30 am - **10h10** am	Welcome talks by **Andreas BORNER** EUCARPIA cereal section chairman and organizers
10h10 am - **10h50** am	Introductory talk: **Raphaël DUMAIN** (Bayer - Germany): *Global context for wheat, Innovation & Bayer Activities*
Session I. Characterization and use of genetic resources **Chair: François Balfourier, INRA, France**	
10h50 am - **11h30** am	Keynote speaker: **Andreas GRANER** (IPK - Germany): *Ex situ collections: getting dressed up for another green revolution*
11h30 am - **11h55** am	**Davide GUERRA** (CREA - Italy): *Studying the genetic basis of barley adaptation to contrasting environments in the WHEALBI collection*
11h55 am - **12h30** am	**Ernesto IGARTUA** (EEAD-CSIC - Spain): *Association with high-resolution climate data reveals selection footprints in the genomes of barley landraces across the Iberian Peninsula*
12h30 pm - **2h00** pm	**Lunch break**
2h00 pm - **2h25** pm	**Marco MACCAFERRI** (U Bologna - Italy): *SNP-based characterization of a wide collection of tetraploid wheat reveals extensive domestication sweeps and haplotypes associated to traits of breeding interest*
2h25 pm - **2h40** pm	**Helmy M YOUSSEF** (IPK - Germany): *Natural Diversity of Inflorescence Architecture Traces Cryptic Domestication Genes in Barley (Hordeum vulgare L.)*
2h40 pm - **2h55** pm	**Constance LAVERGNE** (U Nottingham - UK): *Introduction of Aegilops sharonensis cytoplasm into common wheat and production of addition / wheat – Ae. sharonensis lines translocation lines*
Session II. Genomics and epigenomics in the post sequence era **Chair: Laura Rossini, University of Milano, Italy**	
2h55 pm - **3h35** pm	Keynote speaker: **Scott Allen JACKSON** (U Georgia - USA): *Sources of variation in crop legumes—genetic, epigenetic and structural*
3h35 pm - **4h00** pm	**Luigi CATTIVELLI** (CREA - Italy): *Durum wheat genome reveals the signature of 10,000 years of selection*
4h00 pm - **4h30** pm	**Coffee break & poster session**
4h30 pm - **4h55** pm	**Caroline JUERY** (INRA GDEC - France): *Linking genomic and epigenomic features in hexaploïd wheat genome*
4h55 pm - **5h20** pm	**Cécile MONAT** (IPK - Germany): *Hi-C and chromosome-scale assembly to detect large chromosomal rearrangements in wheat genomes*
5h20 pm - **5h35** pm	**Maria BUERSTMAYR** (BOKU - Austria): *High-resolution mapping of the pericentromeric region on wheat chromosome arm 5AS harboring the Fusarium head blight resistance QTL Qfhs.ifa-5A*
5h35 pm - **6h30** pm	**Romain DE OLIVEIRA** (INRA GDEC - France): *Deciphering Structural Variations in the Wheat Genome Using Resequencing Data*
6h30 pm - **8h00** pm	**Cocktail at the Clermont-Ferrand city hall**

Tuesday March 20th, 2018

Session III. Yield potential and resource efficiency **Chair: Frank Ordon, JKI, Germany**	
8h30 am - **9h10** am	Keynote speaker: **John FOULKES** (U Nottingham - UK): *Exploiting Genetic Diversity for Enhancing Yield Potential and Resource Efficiency in Wheat*
9h10 am - **9h35** am	**Bruno CONTRERAS-MOREIRA** (ADES-CSIC - Spain): *Agronomic advantage of presence of PpdH2 in winter barleys proven through a natural selection approach using GBS in pools of field-grown populations*
9h35 am - **10h00** am	**Gaëtan TOUZY (**Arvalis - France): *Improving Water Use Efficiency in Bread Wheat by Multi-trait multi-Environment Genome-Wide Association Studies*
10h00 am - **10h30** am	**Eric OBER** (NIAB - UK): *Implementing large-scale field phenotyping in genomic selection to accelerate wheat breeding*
10h30 am - **11h00** am	**Coffee break & poster session**
11h00 am - **11h15** am	**Kerstin NEUMANN** (IPK - Germany): *Non-invasive phenotyping reveals stress adaptive and constitutive biomass QTL in cereals*
11h15 am - **11h30** am	**Ulrike LOHWASSER** (IPK - Germany): *Searching for Frost Tolerance in Wheat (Triticum aestivum L.) – A genome wide association study*
Session IV. Improving tolerance to biotic and abiotic stress **Chair: Fiona Doohan, University of Dublin, Ireland**	
11h30 am - **12h10** pm	Keynote speaker: **Heribert HIRT** (KAUST - Saudia Arabia): *Using beneficial endophytes to enhance abiotic stress tolerance and yield of plants*
12h10 pm - **12h30** pm	**Ewen MULLINS** (Teagasc - Ireland): *Intensive field phenotyping to support breeding initiatives against septoria tritici blotch (STB) disease of wheat*
12h30 pm - **2h00** pm	**Lunch break**
2h00 pm - **2h25** pm	**Yvan MOËNNE-LOCCOZ** (U Lyon - France): *Interactions of plant-beneficial rhizosphere bacteria according to cereal genotype*
2h25 pm - **2h50** pm	**Laetitia WILLOCQUET** (INRA - France): *Developing phenotyping methods for quantitative host plant resistance using simulation modelling and ROC curves*
2h50 pm - **2h15** pm	**Hermann BUERSTMAYR** (BOKU - Austria): *Genomics assisted improvement of Fusarium head blight resistance in bread wheat, durum wheat and triticale*
3h15 pm - **3h30** pm	**Javier SANCHEZ-MARTIN** (U Zürich - Switzerland): *GWAS Reveals New Race-Specific Resistance Genes to Powdery Mildew in Wheat*
3h30 pm - **3h45** pm	**Ali NIAZ** (U Leicester - UK): *Novel rye-wheat translocations in Thinopyrum intermedium wheat introgression lines enhancing Wheat streak mosaic virus resistance*
3h45 pm - **4h00** pm	**Pierre-Antoine PRECIGOUT** (U Paris-Saclay- France): *Questioning the sustainability of quantitative physiological resistance: epidemiological and evolutionary responses of foliar fungal pathogens to changes in wheat plant traits*
4h00 pm - **4h30** pm	**Coffee break & poster session**
4h30 pm - **11h00** pm	**Excursion & Gala Dinner**

Wednesday March 21st, 2018

Session V. High quality and safe cereals for food, feed and industry **Chair: Dario Fossati, Agroscope Switzerland**	
8h30 am - **9h10** am	Keynote speaker: **Tatsuya IKEDA** (NARO - Japan): *Importance of international collaboration on wheat quality and safety*
9h10 am - **9h35** am	**Sebastian MICHEL** (U Vienna - Austria): *Multiple to conquer: Simultaneous selection for grain yield and baking quality in genomics-assisted wheat breeding*
9h35 am - **10h00** am	**Suong CU** (Flinders U - Australia): *Genome-wide association studies of zinc, iron, copper, manganese and phosphorus in wheat (Triticum aestivum L.) grain and rachis at two developmental stages*
10h00 am - **10h30** am	**Simon GRIFFITHS** (JIC - UK): *Biochemical and genetic analysis of arabinoxylan fibre in wheat grain*
10h30 am - **11h00** am	**Coffee break & poster session**
Session VI. Future challenges and innovations **Chair: Roberto Tuberosa, University of Bologna, Italy**	
11h00 am - **11h40** am	Keynote speaker: **Peter ROGOWSKY** (INRA Lyon - France): *CRISPR-Cas9 technology in plant science*
11h40 am - **12h05** pm	**Fred VAN EEUWIJK** (Wageningen U - The Netherlands): *Statistical methodology for modelling genotype by environment interactions in QTL mapping and genomic prediction, with applications to barley and wheat*
12h05 pm - **12h30** pm	**Andreas HUND** (ETH Zürich - Switzerland): *Zooming into GxE by means of modern field phenotyping techniques*
12h30 pm - **2h00** pm	**Lunch break**
2h00 pm - **2h25** pm	**Friedrich LONGIN** (U Hohenheim - Germany): *Optimum breeding schemes for line breeding in wheat*
2h25 pm - **2h40** pm	**Daniela BUSTOS-KORTS** (Wageningen U - The Netherlands): *A synthesis of crop growth and statistical genetic modelling to evaluate phenotyping strategies in wheat*
2h40 pm - **2h55** pm	**Renaud RINCENT** (INRA GDEC - France): *Genomic prediction of genotype by environment interactions by coupling genetic and physiological modelling*
2h55 pm - **3h10** pm	**Philipp BOEVEN** (Limagrain - France): *Rare and wanted! How to breed for favorable male floral traits required for hybrid wheat?*
3h10 pm - **3h30** pm	**Funmi LADEJOBI** (NIAB - UK): *Differentially penalized regression improves genomic prediction of wheat flowering time*
3h30 pm - **4h00** pm	**Concluding remarks and prizes for the three best posters**
4h00 pm - **4h30** pm	**Coffee break**

IWIW2

SCIENTIFIC PROGRAMME

Thursday March 22th, 2018

7h45 am - **8h30** am	Registration & Welcome coffee
Introduction	
8h30 am - **9h00** am	**Frank Ordon**, Julius Kühn-Institut (JKI) - *Wheat Initiative Research Committee* Achievements of the Wheat Initiative
9h00 am - **9h30** am	**Victor Kommerell**, *International Maize and Wheat Improvement Center* (CIMMYT) Progress update on major multi-country/institutions wheat research programs
9h30 am - **10h00** am	**Laurène Coral**, *Institut National de la Recherche Agronomique* (INRA); *Directorate for regional policy, higher education and Europe - management* (DARESE) European Commission funding opportunities
10h00 am - **10h30** am	**Viktor Korzun**, *KWS Cereals Breeding Technologies* Future important breeding targets
10h30 am - **11h00** am	**Coffee break**
Impulse talks	
11h00 am - **11h20** am	**SESSION I -** Genetic resources: **Simon Griffiths**, John Innes Centre (JIC)
11h20 am - **11h40** am	**SESSION II -** Phenotyping: **Jérémy Derory**, Limagrain Europe
11h40 am - **12h00** am	**SESSION III -** Genome editing: **Pierre Barret**, Institut National de la Recherche Agronomique (INRA)
12h00 am - **12h20** pm	**SESSION IV -** Genomic selection: **Friedrich Longin**, University of Hohenheim
12h20 pm - **12h40** pm	**SESSION V -** Bio Informatics: **Hillary Sullivan**, DuPont Pioneer
World café	
12h40 pm - **12h50** pm	**Introduction**
12h50 pm - **2h00** pm	**Lunch break**
2h00 pm - **2h45** pm	**Phase 1:** exchange on the 5 topics **(10 groups)**
3h00 pm - **3h45** pm	**Phase 2:** exchange on the 5 topics **(10 groups)**
4h00 am - **4h30** am	**Coffee break**
4h30 pm - **5h30** pm	Conclusion for each of the 5 topics discussed during the world café

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INTRODUCTION TALK

Introduction Talk

RAPHAEL DUMAIN
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Bayer CropScience

Global context for wheat, Innovation & Bayer Activities

Context for wheat and the challenges to deliver innovation

Wheat is the staple food for 1/3 of the world population. Future demand for wheat will be driven by population growth and to meet this predicted demand, global annual wheat yield growth will need to increase from around 1% to 1.6% p.a. This significant production increase will need to be achieved in the context of expected negative impacts of climate change (-6% global yield for each 1 degree C temperature rise) and the need to improve input efficiency and environmental stewardship in agricultural production. The wheat planted area is not expected to grow (ref. FAO, Wheat Initiative) and is even declining in countries like the US. Improvements in agronomy and management practices have already addressed much of the gap between average yield and potential yield, in major producing regions. As the stability of wheat production worldwide is under environmental and economic pressures, there is a critical need to increase wheat yield through new varieties, hybrids, traits and other agronomic innovations.

Wheat improvement has been lagging behind other major crops such as corn and rice. The enhanced enabling resources and methodologies developed in the past decade in genomics (e.g. the IWGSC wheat genome reference sequence), molecular genetics (e.g MAGIC populations accelerating QTL identification) and biotechnology (e.g. genome editing of wheat powdery mildew genes), provide new platforms for accelerating wheat breeding. These platforms encourage investment, but the timelines remain long (16-20 years from discovery to market) and the risk is high, thereby necessitating the need for investment and risk sharing by multiple players to ensure success. Public/private partnerships are essential to meet the scientific challenges of trait improvement in wheat and ensure that innovation reaches all farmers worldwide as fast as possible.

Wheat today is almost exclusively an open-pollinated crop in all markets, with the exception of a small hybrid wheat business in parts of Europe. As an open-pollinated crop, farmers purchase new genetic material and then retain seed-lots on-farm to plant for the next year. Consequently, we estimate that ~70% of the global wheat planted area is “farm-saved” seed. The nature of wheat plant breeding to date, given the lower technology deployment versus other crops and the relatively low-income stream, has necessitated a key role for public breeding and led to a highly fragmented private breeding structure with many markets having country specific local breeding companies. These companies are able to operate at relatively low cost yet effectively address the local market with adapted varieties. A small number of companies have developed multi-regional breeding programs, including partnerships with local companies and in some countries with public institutions, providing some economies of scale. However, the need to develop locally adapted varieties limits the synergies that can be realized from this approach and the opportunities to generate revenues for private breeders remains limited. As a consequence of the lack of market attractiveness, public breeding has played a critical role in the global breeding effort with CGIAR/CIMMYT estimating ~60% of released varieties globally being from public breeding sources.

(Source: Impacts of International Wheat Improvement Research 1994-2014, CGIAR, CIMMYT, ICARDA: 2014 Global Wheat Impacts survey) whereas investment by private companies in wheat research has been sporadic and not long term oriented. Quoting from the assessment made by the Wheat Initiative organization in May 2013 which reflects the low level of private-corporate investment – “In the last 20 years, wheat has become an orphan crop in terms of research investments considering its importance for global food security. As of 2010, global investments in maize research, mostly in the US and Europe, were more than four times greater than in wheat research.” (Source: Wheat Initiative May 2013 - www.wheatinitiative.org)

Hybrid wheat offers the opportunity to provide farmers with higher yield stability and improved revenues. It has been identified as a game changing breeding method to improve the effectiveness of breeding for yield potential as part of the Wheat Initiative strategic research agenda. Investments in hybrid wheat have been undertaken by a number of companies over the past decades but in the absence of efficient enabling platforms hybrid wheat varieties have been very challenging to deliver. Bayer believes that the new genomic and phenotyping technologies developed in large part by public-private initiatives in the last decade will now enable a long-term sustainable business in hybrid wheat. However, the commercial development of such technology will only slowly impact the market given the logistics of wheat seed production and the likely premium in the market to support higher production costs.

Bayer wheat research – Breeding and Traits

In the last years, Bayer committed to invest significantly in new capacities and programs to support wheat breeding and trait research as a long-term strategic plan to address the above challenges. As part of this investment, seven main breeding stations in Europe, North America and Australia were established and a major research program aiming at developing a hybrid wheat platform supplemented by project to improve traits underlying yield performance (in a so called crop efficiency program) has been deployed.

To produce Hybrid Wheat, Bayer has focused on developing a CMS based male sterility system for its future hybrid varieties. To date, Bayer reached several significant milestones and is continuing efforts to launch hybrids as soon as possible in the next decade. As for trait-based yield improvement, Bayer’s research program is focused on deciphering and improving multiple genetics component of grain yield through genes involved in resource utilization, radiation use efficiency, transport of sugars and minerals to seeds, etc… These research efforts are intentionally designed to complement incremental improvements provided by breeding to increase yield and protect it from disease and insect loss. Bayer strategy is first to deploy traits that will enable the production of hybrids while continuing to provide native traits with durable resistance to major diseases such as rusts, followed by the delivery of the first non-GM CE traits to improve yield performance.

To support this strategy, significant investments are also made into enabling technologies. This includes supporting the production of a high quality reference genome sequence of bread wheat through international collaborations with the IWGSC and internal sequencing of elite parent materials, developing integrated databases and deploying precision phenotyping platforms. Bayer has also developed expertise for wheat in the areas of cell biology, gene editing, transcriptomics, and doubled haploid methodology. Finally, future areas of research may include a better understanding the interactions between the genetics components of the crop and biological compounds used in the fields to stimulate yield increase.

Keywords: Introduction, Context, Innovation, Hybrid, Bayer

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Session I

CHARACTERIZATION AND USE OF GENETIC RESOURCES

Chair: François Balfourier, INRA, France

Keynote Lecture

ANDREAS GRANER
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IPK, Germany

Ex situ collections: getting dressed up for another green revolution

Conventional crop breeding essentially rests on repeated cycles of crossing and selection. This approach has warranted the development of superior cultivars over the past decades. However, it is only sustainable, if the genetic diversity that is lost in the process of selection is adequately replenished by introducing novel diversity into the genepool. At present about 7 million seed samples are stored in far more than 1000 *ex-situ* collections worldwide. Arguably, the vast diversity resting on the shelves of genebanks has been tapped into only marginally. Benefitting from genomics technologies, the conservation management of individual collections can be optimized in order to maintain the authenticity and the integrity of a given accession, and to provide access to the genetically defined material.

At the highest level of resolution, informed access to genetic resources means that each genebank accession will be tagged with information on individual alleles along with their phenotypic effects. The majority of crop genomes are characterized by their large size and inherent complexity hampering molecular analysis, meiotic recombination and, sometimes, their amenability to genetic modification. However, these limitations have been increasingly overcome by technical advances in several key areas including (i) structural and functional genomics, (ii) phenotypic cataloging of accessions using automated imaging and (iii) novel biotechnological approaches that will provide entry points for crop improvement. While the application of novel technology opens up a wealth of entry points for genetic analyses, it also generates and amasses humongous streams of data. Therefore, integrated concepts of data management and analysis will need serious consideration when aiming at the exploitation of novel technologies for the systematic phenotypic and genotypic characterization of genebank collections.

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Studying the genetic basis of barley adaptation to contrasting environments in the WHEALBI collection

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The EU-funded WHEALBI (Wheat and barley Legacy for Breeding Improvement) project is taking a multidisciplinary approach to identify, understand and utilise the genetic diversity available in wheat and barley formally-bred cultivars, landraces and wild relatives. This is strategic to meet the challenge to increase crop yields while reduce environmental impact. Here we focus on a carefully selected set of 403 barley accessions from extensive *ex situ* collections which cover the geographical and agro-ecological adaptive range of barley cultivation. Comprehensive molecular variant analysis by exome sequencing defined sets of 0.45 M and 1.9 M SNPs for GWAS and allele mining approaches, respectively. Agronomic and life history traits were also collected from multi-environment common garden experiments and provide unique datasets to decipher the genetic basis of adaptation to environment. Genomic regions and candidate genes driving phenotypic differences for heading date, plant height, grain weight, and awn length across a number of environments will be discussed. Relating the genomic diversity to tested accessions' sites of origin and breeding history will define promising sources of alleles for European breeding programmes.

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Association with high-resolution climate data reveals selection footprints in the genomes of barley landraces across the Iberian Peninsula

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Landraces are populations of crop plants adapted to a particular environment. Extant landraces are surviving genetic archives, keeping information of the selection processes experienced by them until settling in their current niches. Barley landraces were abundant in Spain and properly collected in germplasm banks over the last century. This study intends to establish relationships between genetic diversity of the landraces and the climate of their collection sites. A thorough study of a period of 30 years of past climate (1981-2010), extracted agro-climatic variables meaningful for cereal production at the collection sites of 135 barley landraces. Variables summarize temperature, precipitation, evapotranspiration, potential vernalization and frost probabilities at different moments of the year and time scales (season, month). A set of high-resolution maps of climatic variables, computed from over 2,000 temperature and 7,000 precipitation stations across peninsular Spain, was built to assess the relationship between barley accessions and the climate of the territory. SNP genotyping of the landraces was carried out combining Illumina Infinium assays and genotyping-by-sequencing, yielding 7,479 biallelic markers placed on the barley reference genome. These SNPS were then analysed to ascertain their association with agro-climatic variables at two levels of genetic diversity, with and without taking into account population structure. We report a number of associations that suggest the differential adaptation of the germplasm groups identified are dominated by reactions to cold temperature and frost. Several significant associations pointing at specific adaptations to agro-climatic features were found, and candidate genes underlying some of the main regions are proposed.

Keywords: agro-climatic indices, genome-wide association analysis, selection footprint, barley, adaptation

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SNP-based characterization of a wide collection of tetraploid wheat reveals extensive domestication sweeps and haplotypes associated to traits of breeding interest

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The iSelect wheat 90K SNP array and the recently assembled Svevo durum wheat genome were used to characterize a world-wide tetraploid wheat collection and to project the genotyping data onto the golden-standard durum wheat genome. We report on the diversity pattern of 1,854 non-redundant accessions from all known *Triticum turgidum* subspecies, from wild emmer to modern durum wheat. The whole-genome genetic diversity survey relied on a common genotype-calling pipeline developed by AgriBio (c/o Matt Hayden) supported by 17 tetraploid linkage maps. The pipeline yielded 17,416 informative single-locus SNPs anchored to the Svevo genome. Among the wild emmer (WEW), domesticated emmer (DEW), durum wheat landraces (DWL), durum wheat cultivars (DWC), WEW showed the highest and most uniform average p10Mb diversity level across the whole genome, thus providing a

valuable reference for cross-comparisons with DEW, DWL and DWC. Extended genetic diversity depletions were found particularly in pericentromeric regions. Our results suggest that 38.2% of modern DW genome has been affected by genetic bottleneck/selection events leading to strong diversity depletions. However, we also report the presence of six extended regions of increased genetic diversity associated to DEW-DWL and DWL-DWC transitions. We also confirm the presence of an extended region on pericentromeric 4A chromosome characterized by genetic diversity depletion in WEW. Population structure of the collection have been studied in details using ADMIXTURE and fineSTRUCTURE, revealing multiple subsequent events of population differentiation associated to human-driven dispersal routes. This analysis provides the basis for a more informative re-sequencing towards a tetraploid pangenome. Sub-panels of this collection have already been used for GWAS analysis. I.e. the dicoccum, landrace and modern durum panels have been characterized for response to yellow rust, allowing us to identify highly significant GWAS-QTL (Liu et al. 2017 Theor Appl Genet. 130:649-667) that can be readily used in breeding. GWAS-QTLs for grain yield components (grain size and grain number per spike) have been identified using a subpanel of Mediterranean DW landraces. These resources allowed us to map QTLs at an improved resolution (1 cM confidence interval) and readily scan the genome for underlying candidate genes.

We acknowledge the funding support of: Italian Ministry of Education and Research 251 through the Flagship InterOmics (PB05) project, CREA project Interomics, Fondazione in 252 rete per la ricerca agroalimentare (AGER) project From Seed to Pasta, FP7-KBBE Project 253 DROPS (ID244347), Genome Canada, Western Grain Research Foundation, Saskatchewan 254 Wheat Development Commission, Alberta wheat Development Commission, Saskatchewan 255 Ministry of Agriculture, the administrative support of Genome Prairie, Canadian Triticum 256 Applied Genomics (CTAG2), Binational Science Foundation (BSF) grant #2015409, USDA-257 Agricultural Research Service Current Research Information System project 3060-21000-258 037-00-D, German Federal Ministry of Food and Agriculture (2819103915), German 259 Ministry of Education and Research (031A536), and Natural Sciences and Engineering 260 Council of Canada (NSERC) (SPG 336119-06, RGPIN 92787).

Keywords: single nucleotide polymorphism, genetic diversity, GWAS, *Triticum turgidum* L. ssp. *durum* Desf., haplotype

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Natural Diversity of Inflorescence Architecture Traces Cryptic Domestication Genes in Barley (*Hordeum vulgare* L.)

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Many-grained mutants occurring spontaneously among their less well-endowed field mates may have appeared to early farmers as fortunate twists of fate foreboding wealth and abundance. In domesticated barley, the number of kernel rows in spike can be tripled by recessive mutant alleles at the Six-rowed spike 1 (vrs1) locus that abolish the suppression of lateral spikelet fertility. Genetic analyses of so-called intermedium barleys, where lateral spikelet development is intermediate between six-rowed and two-rowed types, and induced mutants have revealed that other genes can increase the size of florets and even stimulate occasional grain setting in lateral spikelets, in interaction with vrs1. Here, we show that a complete six-rowed phenotype occurs in a diverse panel of intermedium barley carrying wildtype Vrs1 in presence of the Int-c.a allele of the intermedium spike-c (int-c) gene, previously considered only as a modifier of lateral spikelet fertility. Int-c.a-type alleles had arisen before domestication and are associated with the enlargement of lateral florets in wild barley. Since Int-c.a cannot overcome the suppression of fertile lateral florets in the genomic background of wild barleys, we infer the existence of other gene loci at which novel alleles or allelic combinations were selected for after domestication to increase grain number of barley independently of Vrs1.

Related publication:
Youssef, H.M., Mascher, M., Ayoub, M.A. et al. (2017) Natural diversity of inflorescence architecture traces cryptic domestication genes in barley (*Hordeum vulgare* L.) Genet Resour Crop Evol 64: 843. https://doi.org/10.1007/s10722-017-0504-6

Keywords: Barley domestication, Int-c, intermedium barleys, spikelet fertility, Vrs1

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Introduction of *Aegilops sharonensis* cytoplasm into common wheat and production of addition / wheat – *Ae. sharonensis* lines translocation lines

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Exploitation of heterosis, or hybrid vigour, is a promising solution for yield improvement in wheat (*Triticum aestivum*) to meet the challenge of feeding the growing population. Cytoplasmic male sterility (CMS) is one option to produce hybrid wheat on a large scale. Today the main CMS system is based on *T. timopheevii* cytoplasm. Nevertheless, a combination of several restorer genes seems to be required to reach a complete restoration of fertility when other CMS could be restored using one single restorer gene. *Aegilops sharonensis* Eig (Sharon goatgrass) is a diploid species (2n = 14, SshSsh), which together with Aegilops speltoides, belongs to the Sitopsis. It has been reported that *Ae. sharonensis* caused sterility when its cytoplasm was transferred into common wheat (Tsunewaki et al., 1976). *Ae. sharonensis* could therefore be a potential source of sterile cytoplasm for common wheat. Cytoplasm of *Ae. sharonensis* was introduced into common wheat by repeated backcrosses. The initial cross was carried out by pollinating several accessions of *Ae. sharonensis* with common wheat. *Ae. sharonensis* carries a gametocidal gene, Gc, on chromosome 4Ssh, which will be preferentially transmitted to the next generation. To eliminate chromosome 4Ssh, a wheat translocation line carrying a small fragment of chromosome 4Ssh on the distal end of chromosome 4B with the Gc gene knocked out (Friebe et al., 2003) was used as the recurrent parent. Alloplasmic, addition and wheat – *Ae. sharonensis* translocation lines were identified using genomic in situ hybridization (GISH). If some of the back crossed lines appear to be sterile, the next steps will be to find a restorer line against *Ae. sharonensis* cytoplasm together with a maintainer line, to have a complete CMS system.

Related publication:

Tsunewaki, K., Y. Mukai, T.R. Endo, S. Tsuji and M. Murata (1976) Genetic diversity of the cytoplasm in *Triticum* and *Aegilops*. VI. Distribution of the haploid-inducing cytoplasms. Jpn. J. Genet. 51:193-200

Friebe, B., P. Zhang, S. Nasuda, and B.S. Gill (2003) Characterization of a knock-out mutation at the Gc2 locus in wheat. Chromosoma 111:509–517

Keywords: *Ae. sharonensis*, hybrid wheat, Gametocidal gene, GISH, alloplasmic lines, CMS, wild relatives

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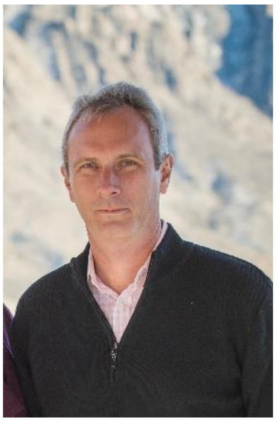
Session II

GENOMICS AND EPIGENOMICS IN THE POST SEQUENCE ERA

Chair: Laura Rossini, University of Milano, Italy

Keynote Lecture



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Sources of variation in crop legumes - genetic, epigenetic and structural

Using two legume crops, common bean (*Phaseolus vulgaris*) and soybean (*Glycine max*), I will discuss efforts to understand and describe genetic/epigenetic variation and how that might affect plant improvement programs. Both crops have undergone extensive genetic bottlenecks due to domestication and, in soybean, the US germplasm can be traced to just a few founder lines, an extreme bottleneck. Polyploidy has resulted in diversity within domesticated soybean that is often overlooked, e.g. differentially methylated genes, presence-absence variation facilitated by genetic redundancy, and potentially, fixed hybridity between duplicated genes. Through comparisons between common bean and soybean, I will describe and discuss some of the genome-level mechanisms of generating novel variation and the results of polyploidy on genes and genome structure.

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Durum wheat genome reveals the signature of 10,000 years of selection

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The domestication of wild emmer wheat ~10,000 years ago by early agrarian societies led to domesticated emmer and subsequently durum wheat through a process of selection for non-brittle rachis and free-threshing forms. Durum wheat became established as a prominent crop only ~1,500-2,000 years ago. We completed genome sequencing of the modern durum wheat cultivar Svevo, assembling the Illumina reads with DeNovoMAGIC software (NRGene, Israel) and scaffolding contigs using a high density genetic map and chromosome conformation capture sequencing (Hi-C). The final assembly covers 10.45 Gb and is characterized by a N50 of 6Mb. The comparison of this durum wheat assembly to a recently published wild emmer assembly allowed the identification of genome-wide modifications imposed by 10,000 years of selection and breeding on the genomic architecture of tetraploid wheat. The comparison revealed strong overall synteny, as well as several thousand regions subjected to copy number variation. The wheat iSelect 90K SNP Infinium assay was then employed to genotype a diversity panel of 1,854 tetraploid wheat accessions representing the main domestication- and breeding-related germplasm groups. Analysis of these data revaluated the number of regions exhibiting strong signatures of genetic divergence associated with speciation, genetic drift, and selection events. Further, we projected onto the durum wheat genome about 1,500 QTL for morphological, phenological and quality traits, grain yield components, and disease resistance reported from published biparental mapping studies and GWAS. The durum wheat genome contains more than 66,000 genes. Among them, we annotated ~1,500 complete NBS-LRR genes, a family of genes known to be prominently involved in signaling and plant disease resistance. The availability of the complete genome of durum wheat will accelerate the identification and the isolation of new sources of disease resistance, as well as other genes that will contribute to breeding high-yielding and more resilient cultivars.

Keywords: Durum wheat, Genome sequencing

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Linking genomic and epigenomic features in hexaploïd wheat genome

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Because of its size, allohexaploid nature and high repeat content, the bread wheat genome is a good model to study the impact of the genome structure on gene organization, function and regulation. Such analyses have long been hampered by the lack of a high quality reference sequence. However, thanks to the efforts of the International Wheat Genome Sequencing Consortium (IWGSC), such sequences are now available and can be used to decipher the wheat genome. By conducting a wide range of analyses on the reference sequence of chromosome 3B, including gene and transposable element annotation, recombination, transcriptomics and structural variations studies, we revealed unexpected relationships between gene and genome structure and function, that cannot be explained at the sole genomic level. These include a strong partitioning of chromosome 3B, the existence of chromosomal domains encompassing clusters of co-expressed genes as well as co-regulation islands involving distant co-expression clusters. In order to investigate the role of epigenetics, we conducted ChIP-seq and RNA-seq experiments on various tissues and analyzed the presence of different histone modifications in view of the gene and genome organization and regulation. Results of this study will be presented.

Keywords: Genomic, Epigenomic, Wheat, Histone modification

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Hi-C and chromosome-scale assembly to detect large chromosomal rearrangements in wheat genomes

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High-quality sequence assemblies of multiple individuals have emerged as important tools towards a deeper understanding of the full diversity within a species. The comprehensive and robust assessment of the different type of variation based on sequence assemblies can only be achieved through the collection of genome-wide sequence data with complementary technologies that retain information about physical linkage between sequence fragments. In the context of the Wheat Ten plus Genomes Project, in which we are assembling at pseudomolecules level wheat genomes with the final goal to get on overview of the wheat pan-genome, we are studying structural variation in bread wheat (*Triticum aestivum* L.). We have developed a pipeline to use chromosome capture (Hi-C) sequencing data to construct chromosome-scale sequence assemblies. Hi-C uses three-dimensional contact probabilities of chromatin in the nucleus to reconstruct the linear order of sequence scaffolds. The first step of our pipeline is the alignment of Hi-C data to a genetically anchored sequence assembly. Subsequently, residual misassemblies are detected and corrected and chromosome-scale physical genome maps are constructed from Hi-C contact matrices. Here, we present the outcome of this computational pipeline for three wheat cultivars. Highly contiguous sequence assemblies of three wheat genomes with scaffold N50 values > 10 Mb were constructed using the DeNovoMAGIC™ technology (NRGene, Ness Ziona, Israel) from paired-end and mate-pair Illumina data as well 10X Chromium linked-reads. These assemblies were ordered into 21 pseudomolecules representing > 95 % of the genome using Hi-C. Sequence alignments and Hi-C contact matrices revealed megabase-scale chromosomal rearrangements between cultivars such as inversions and inter-chromosomal translocations.

Keywords: genome assembly, pseudomolecules, wheat

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High-resolution mapping of the peri-centromeric region on wheat chromosome arm 5AS harboring the *Fusarium* head blight resistance QTL Qfhs.ifa-5A

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Delfina BARABASCHI[2], Andrea VOLANTE[2], Giampiero VALE[2], Luigi CATTIVELLI[2]
and Hermann BÜERSTMAYR[1]

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The relevant Qfhs.ifa-5A allele, contributing to enhanced *Fusarium* head blight resistance in wheat, resides in the pericentromeric interval of chromosome 5A (Buerstmayr et al. 2003). It is well documented that crossover events are distributed unevenly along chromosomes and almost absent around centromere. As an example, even among 3650 near isogenic recombinant inbred lines used for genetically fine-mapping Qfhs.ifa-5A, solely four individuals recombined within the C-5AS1-0.40 bin covering 40% of the physical length of 5AS. As a consequence, precisely locating of Qfhs.ifa-5A employing recombination dependent linkage mapping is practically impossible. To facilitate fine-mapping of Qfhs.ifa-5A we therefore used radiation hybrid (RH) mapping approach. RH mapping utilizes radiation to induce double strand breaks (DSBs) leading to deletions and these DSBs occur randomly and evenly distributed along a chromosome (Tiwari et al. 2016). We generated a radiation selfing (RS) panel by selfing plants from irradiated seeds of the experimental line NIL3 containing the Qfhs.ifa-5A resistance allele in the background of the cultivar Remus. In addition we generated a radiation hybrid (RH) panel by pollinating Chinese Spring (CS) Nullisomic-5A/Tetrasomic-5B plants with gamma-irradiated CS pollen. In total 5,157 radiation selfing and 276 radiation hybrid plants were screened for deletions on 5AS and plants containing deletions were analysed using 102 5AS specific DNA markers. Combining genotypic information of both panels yielded an 817-fold map improvement (cR/cM) for the centromeric bin and was 389-fold increased across the Qfhs.ifa-5A interval compared to the genetic map, with an average map resolution of 0.77 Mb/cR. Radiation induced deletion mapping demonstrated to be particularly efficient for high-resolution mapping of recombination poor stretches, such as pericentromeric regions. Selected RS lines will be used to associate losses of DNA stretches with the FHB phenotype and vice versa during evaluation in field tests, thus providing an unprecedented tool for fine localization of the Qfhs.ifa-5A gene locus. For more details please refer to Buerstmayr et al. (2017).

Supported by the Austrian Science Fund, project: SFB F3711

Related publication:

Buerstmayr et al. (2003) Molecular mapping of QTLs for Fusarium head blight resistance in spring wheat. II. Resistance to fungal penetration and spread. Theor Appl Genet 107:503–508. DOI:10.1007/s00122-003-1272-6

Buerstmayr et al. (2017) High-resolution mapping of the pericentromeric region on wheat chromosome arm 5AS harbouring the Fusarium head blight resistance QTL Qfhs.ifa-5A. Plant Biotechnol. J., in press. DOI: 10.1111/pbi.12850

Tiwari et al. (2016) A whole-genome, radiation hybrid mapping resource of hexaploid wheat. Plant J. 86, 195–207. DOI:10.1111/tpj.13153

Keywords: Radiation hybrid mapping, *Fusarium* head blight, *Triticum aestivum*, supressed recombination, gamma irradiation

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Deciphering structural variations in the wheat genome using resequencing data

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Structural variations (SVs) such as copy number and presence-absence variations (CNVs, PAVs) are polymorphisms that are known to be involved in the expression of phenotypes. In the absence of a reference genome sequence, their study has long been hampered in wheat. The recent advent of new wheat genomic resources has led to a paradigm shift, making possible to investigate the extent of SVs among cultivated and wild populations. Our project aims at characterizing SVs in a *Triticeae* diversity panel of 44 accessions from seven tetraploid and hexaploid *Triticeae* species. To cope with the wheat genome complexity, we developed strategies combining shotgun sequencing of sorted chromosomes 3B with bioinformatics tools and we studied SVs affecting not only genes but also transposable elements (TEs). Our results show that 14% of the genes are variable within this panel. In addition, they reveal a very high level of intra- and interspecific variability affecting TEs, contrasting with the weak polymorphism rate usually reported with SNPs. Chromosomal extremities are the regions where we see most of the variability, confirming previous hypotheses made when comparing wheat with the other grasses.

Keywords: Structural variation, CNV, PAV, Wheat, Comparative genomics, bioinformatics

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Session III

YIELD POTENTIAL AND RESOURCE EFFICIENCY

CHAIR: FRANK ORDON, JKI, GERMANY

Keynote Lecture

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Exploiting genetic diversity for enhancing yield potential and resource efficiency in wheat

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Future genetic progress in wheat grain yield will depend on increasing biological yield (aboveground dry-matter per unit area) and maintaining, or ideally increasing, harvest index (grain yield/biomass; HI). In recent years the rate of genetic progress in HI has not increased; indeed, there is evidence for a trade-off with increasing biomass (Aisawi et al., 2015). Therefore, it is important to identify new avenues for increased grain partitioning in high biomass backgrounds. A Genome Wide Association Study (GWAS) was carried out focused on grain traits to enhance HI in a High Biomass Association spring wheat panel (HiBAP) at CIMMYT, Ciudad Obregon, Mexico in two years. Our analysis has identified marker-trait associations for key grain traits associated with HI and elucidated underlying mechanisms in relation to stem internode partitioning and fruiting efficiency (grains g-1 spike DM at anthesis). Future genetic progress in grain yield will depend on raising biomass without commensurate increases in N fertilizer inputs to minimize environmental impacts. In the BBSRC Wheat Improvement Strategic Programme (WISP) we quantified variation in grain yield, biomass and N-use efficiency (NUE) and associated traits in panels of diverse hexaploid wheat germplasm comprising landraces (Watkins collection) and synthetic-derived hexaploid under low N and high N conditions. Intensive phenotyping has revealed novel diversity in leaf photosynthetic rate and senescence-related traits associated with improved biomass under both high N and low N conditions. Mapping of Single Seed Descent (SSD) lines derived from Watkins landraces × UK elite cultivar Paragon crosses has revealed novel QTL for biomass and NUE and related traits. In the WISP mapping populations yield was mainly explained by genetic variation in N uptake under low N implying root traits were determining improved yield. Our recent work in the ERC Future Roots project has developed high-throughput platforms for field phenotyping of root system architecture for application in wheat breeding. The perspectives for future goals to raise yield potential and resource efficiency and the challenge to transfer traits into commercial varieties will be discussed.

Related publication:
Aisawi K, Reynolds M, Singh RP, Foulkes, MJ 2015. Crop Science 55, 1749-64

Keywords: Genetic diversity, yield, resource use, wheat, phenomics, GWAS

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Agronomic advantage of presence of *PpdH2* in winter barleys proven through a natural selection approach using GBS in pools of field-grown populations

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Previous research suggests that winter barley in the Mediterranean region can benefit from the presence of a functional gene *PpdH2* which, in more northern latitudes, is typical only in spring cultivars. Previous evidence also indicates an agronomic advantage of reduced vernalization allele *VrnH1-4* in this same area.

A natural selection experiment was devised, using biparental RIL population SBCC097 x Plaisant. SBCC097 is an inbred line derived from a Spanish landrace, presenting an active PpdH2 allele and VrnH1-4. Plaisant is a typical winter cultivar, with a loss-of-function *ppdH2* allele and the winter *vrnH1* allele. Both parents have an active *VrnH2* allele, necessary to produce a winter growth habit.

Four bulks of 25 RILs each were assembled, stratified according to the four possible haplotypes resulting from the *PpdH2-VrnH1* combination, and ascertained using allele specific molecular markers. Bulks were carefully prepared for the whole series of trials, using the same amount of seed per each of the 25 constituent lines. Field trials were run for three years, at two sowing dates, standard (November) and late (February), in Zaragoza, Spain. Grain yield was superior for bulks having the active *PpdH2* allele, at both sowings, whereas yield advantage of VrnH1-4 allele was less general, restricted to late sowings and in interaction with *PpdH2*.

Besides the four bulks, a "metabulk" was constructed, using equal amounts of seed from the four bulks (hence, from the 100 lines). This metabulk was sown in the same field trials, at both sowing dates, with the difference that it was hand harvested every year, and the seed from each harvest was used for next season sowings. Thus, two series of metabulks, standard and late sowing, were actually subjected to natural selection for three years. The expectation was that the initial balanced contribution of the 100 lines would shift according to the reproductive fitness of the lines, i.e., lines producing more seeds would increase in frequency in the metabulks over the years.

After the field trials, the initial bulks and the final metabulks were genotyped, using double digest restriction-site associated DNA (ddRAD-Seq). For the initial bulks, DNA was extracted from each of the 25 constituent lines, and pooled in equal amounts. For the two final metabulks, DNA was extracted and pooled from three independent samples of 100 plants, thus providing true biological replicates. Parents were also genotyped.

After curation of the sequence reads and alignment to the current barley reference, 3087 markers were kept, which were used to analyse allele frequency shifts in the metabulks. Overall, there were more frequency shifts in the late sowing, with a majority of changes occurring in favour of the SBCC097 allele. In the standard sowings, the only significant shift occurred precisely in the *PpdH2* region, which showed a strong selection footprint towards the SBCC097 allele.

These experiments provide a double confirmation of the advantage of the presence of *PpdH2* in winter barleys, under a typical Mediterranean climate, validate the adaptation syndrome described for Spanish (and other Mediterranean) barleys, and demonstrate the validity of a DNA-pooling approach to answer relevant questions on crop adaptation.

Keywords: bulk, genotyping by sequencing, PpdH2, winter barley

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Improving water use efficiency in bread wheat by multi-trait multi-environment genome-wide association Sstudies

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Drought is one of the main abiotic stresses limiting bread wheat (*Triticum aestivum* L.) growth and productivity around the world. Many climate-based simulations have predicted an increase in the frequency and intensity of this abiotic stress. The delivery of new high yielding and stress-tolerant cultivars is now necessary and requires an improved understanding of the basis of the physiological and genetic response to drought.

A panel of 220 European elite cultivars, genotyped with the TaBW420K SNP chip, was evaluated in 39 field experiments; among them, specific drought field trials were conducted using monitored irrigation and rain shelters. Grain yield and yield components were scored for each trial. A crop model was run with detailed climatic data and soil water status, to identify the timing, intensity and history of stress for each genotype × trial combination.

A detailed study of genetic by environmental interactions (GxE) was conducted. By modelling water-deficit patterns, as Chenu et al. (2011), the trials were grouped in three scenarios: no stress, high stress at pre-anthesis stage, and moderate stress at post-anthesis stage. The grain yield loss in the two stressed scenarios ranged from 7 to 12% when compared to the no-stressed scenarios. A large genetic variability of grain yield was identified, with a genotypic coefficient of variation affecting the mean by ± 15%. In the same way, GxE interactions affected the grain yield mean by ± 12.5%. Reaction norm models were run using stress indicators to discriminate tolerant/sensitive varieties. Then, GWAS was performed with the stress-tolerant index and phenotypic values, using simple and multi-environment mixed models. Several quantitative traits loci (QTLs) were identified in the different stress scenarios and the allelic effects of these QTL were related to the environmental co-variables. Methods and results will be discussed especially those regarding the impacts of QTLxE interactions on stress indicators, grain yield and components and grain yield.

Related publication:

Chenu K, Cooper M, Hammer GL, Mathews KL, Dreccer MF, Chapman SC (2011) Environment characterization as an aid to wheat improvement: interpreting genotype-environment interactions by modelling water-deficit patterns in North-Eastern Australia. J Exp Bot 62: 1743–1755

Keywords: Drought stress, Genotype-by-environment interaction, GWAS

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Implementing large-scale field phenotyping in genomic selection to accelerate wheat breeding

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The process of breeding new plant varieties can be accelerated and made less costly by applying new methods of genomic selection (GS). GS uses high density molecular markers and relatively inexpensive genotyping, in combination with phenotypic and pedigree data to increase the selection intensity and decrease the cycle time between initial crosses and finished varieties. Increased precision comes from both exploiting genetic relationships between traits and reducing the effect of environmental influences. However, a bottleneck in applying GS is the cost of phenotyping the requisite training population, which for commercial breeding programmes must be large (several thousand individuals). Robust prediction algorithms developed from the training population can be applied to reference populations without needing to repeat the phenotyping process. While yield is the fundamental phenotypic variable of interest, we test the idea that additional physiological and morphological traits can increase the predictive power of algorithms, akin to increasing the number of genetic markers. The time and cost of phenotyping can be reduced by using remote sensing methods, largely based on spectral reflectance from crop surfaces using RGB, multi- and hyperspectral cameras. In a collaborative project with four commercial breeding companies, we are testing these principles using a combination of ground-based measurements, UAV and aircraft platforms to derive phenotypic markers in a large population of 2992 wheat (*Triticum aestivum*) lines (44 elite crosses, 68 lines per cross) planted in two field locations in 2016 and in 2017. Further prediction precision can be obtained by taking into account environmental covariates that vary spatially across the trial area, such as apparent differences in soil texture and water holding capacity. Initial results show heritabilities of 0.37 to 0.54 for yield predicted from genomic data, with optimal correlations obtained with at least 60 BLUEs per cross. Predictive linear models based solely on phenomic data (dominated by remotely sensed variables) showed a correlation of 0.58 between training and focal sets. The utility of models combining genomic and phenomic markers will be discussed, and the implications for wheat breeding programmes implementing GS.

Keywords: wheat, genomic selection, phenotyping, remote sensing

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Non-invasive phenotyping reveals stress-adaptive and constitutive biomass QTL in cereals

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Future yield improvement is hampered by biotic and abiotic stresses due to climate change and accordingly, wheat and barley yields in Germany showed a strong fluctuation during the last 15 years. One factor is the increased frequency and severity of drought stress events, especially in spring during the vegetative growth phase - affecting the formation of biomass. Our experiments in different wheat and barley collections show a general reduction of ~30% in grain yield caused by a drought period in the vegetative stage. Understanding the genetics of biomass accumulation under well-watered and seasonal drought stress conditions can provide a basis for stabilizing or even increasing grain yield. By daily non-invasive imaging dynamics of biomass accumulation could be resolved, both phenotypically and genetically. To evaluate the genetics of biomass, an automated phenotyping system with controlled watering and non-invasive imaging was used to evaluate diverse wheat and barley collections under greenhouse conditions. Genome-wide association scans (GWAS) were applied to identify genetic markers associated with variation in biomass formation. Drought stress treatment started in the tillering phase and inlcuded a recovery period until the imaging period ended around tipping time. We obtained high heritabilities for biomass under both conditions in wheat and barley and over the whole observation period. GWAS revealed constitutive biomass QTL that are shared between drought and well-watered treatments and stress-adaptive QTL that occur only under the influence of drought stress.

Related publication:
NEUMANN K., ZHAO Y., CHU J., KEILWAGEN J., REIF J.C., KILIAN B., GRANER A. (2017): Genetic architecture and temporal patterns of biomass accumulation in spring barley revealed by image analysis. BMC Plant Biol. 17 (2017) 137. dx.doi.org/10.1186/s12870-017-1085-4.

Keywords: biomass, growth, drought, genome-wide association mapping

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Searching for frost tolerance in wheat (*Triticum aestivum* L.) – A genome wide association study

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Wheat is one of the most important crops for human nutrition and is the most widely grown cereal crop in the world. Wheat yield stability has to be ensured in the context of climate change. Frost tolerance is essential for autumn-planted wheat to survive freezing temperatures during winter in temperate zones. It is a key trait with economic and agronomic importance but its molecular and genetic background (a complex trait with polygenic inheritance) is still poorly understood. The aims of the present study are (1) to investigate phenotypic variation for frost tolerance in a panel of 276 winter wheat accessions (2) to generate information about genetic variation and (3) to find molecular markers closely linked to the trait frost tolerance. Frost tolerance phenotype scores were collected from several locations in Germany and Russia during two seasons and were combined with the genotypic data in genome wide association studies (GWAS). The genotyping was done employing ILLUMINA infinium iSelect 90k wheat chip. The chip carries a total of 81,587 valid and functional SNPs. Finally, 17,900 polymorphic markers could be used for the genome wide analyses. SNP associations were performed using linear mixed models that evaluated the effects of SNPs with minor allele frequencies (MAF) > 10% individually, adjusting for kinship. GWAS analyses of the most significant SNP loci (highly significant associations, LOD>3) identified three and seven positive SNP associations on the chromosomes 1B and 5A, respectively. Haplotype analysis revealed that most of the significant SNP loci for these positions represent an advantage for the evaluated genotypes. Validation of SNPs and their respective haplotypes associated with frost tolerance together with candidate gene based-association studies, will be performed in future studies to determine the diagnostic value of markers for marker assisted selection in winter wheat breeding programs.

The project (code 0315953C) was funded by the German Federal Ministry of Education and Research (BMBF)

Keywords: wheat, frost tolerance, GWAS

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Session IV

IMPROVING TOLERANCE TO BIOTIC AND ABIOTIC STRESS

CHAIR: FIONA DOOHAN, UNIVERSITY OF DUBLIN, IRELAND

Keynote Lecture

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Next green revolution: Plant endophytes to enhance stress tolerance and yield of crops

We currently have more than 1 billion of undernourished people worldwide. This situation is not going to get better, unless we can increase food production by 50 % in the next decades. However, drought, salt and heat stress make up to more than 60 % of crop losses, so that improving abiotic stress resistance is one of the most important factors in securing food production. In the DARWIN21 project (http://www.darwin21.net), we isolate and employ endophytic desert microbes to enhance stress tolerance of plants. I will show that endophytic microbes isolated from desert plants can enhance yield of crops in adverse agriculture conditions, making beneficial microbes a powerful low-cost approach for subsistence farming in many under-producing areas of the world. To better understand the molecular processes in beneficial plant-microbe interactions, I will present the results from our molecular genetic approaches using as a model system *Arabidopsis thaliana*.

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Intensive field phenotyping to support breeding initiatives against *septoria tritici* blotch (STB) disease of wheat

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Zymoseptoria tritici, the causal agent of septoria tritici blotch (STB), remains a significant threat to European wheat production with the continuous emergence of fungicide resistance in *Z. tritici* strains eroding the economic sustainability of wheat production systems. For Irish wheat growers, STB is eroding the commercial viability of the national crop with wheat acreage dropping 40% in the last two years. In response, a national programme was established through the Virtual Irish Centre for Crop Improvement (VICCI) to investigate the aspects of the wheat-STB biotic interaction with a view to supporting commercial wheat breeders in delivering elite lines with higher STB resistance scores. One aspect of VICCI has focused on the pre-symptomatic phase (latent period, LP) after which the pathogen switches to an aggressive necrotrophic stage, when lesions bearing *pycnidia* quickly manifest on the leaf. As minimal knowledge on the possible role of the LP in supporting the resistance/susceptibility of wheat exists, the goal of this study was to investigate the spatial and temporal association between the LP and disease progression across three locations (Ireland – Waterford, Carlow; UK – Norwich) that represent commercially high, medium and low STB pressure environments. Completed over two seasons (2013-2015) with commercial cultivars, the potential of the LP in stalling STB epidemics was significant as identified with cv. Stigg, whose high level of partial resistance was characterised by a lengthened LP (~36 days) under the high disease pressure environment of Waterford. However, once the LP concluded it was followed by a rate of disease progression in cv. Stigg that was comparable to that observed in the more susceptible commercial varieties. Complementary analysis via logistic modelling of intensive disease assessments and qPCR-based quantifications of STB biomass made at Carlow and Waterford in 2015, further highlighted the value of identifying wheat germplasm in breeding programmes that support an extension of the LP. In parallel to this study we have been field phenotyping the NIAB MAGIC population for STB response under Irish conditions and have included the DiverseMAGIC population for 2,018 assessments. Data recorded on these multi-parent mapping populations will enable QTL associated with STB resistance to be identified, and integrated into genome-wide selection strategies for forward selection during varietal development.

Related publication:
Hehir et al. (2017) Plant Pathology. http://onlinelibrary.wiley.com/doi/10.1111/ppa.12780/full

Keywords: wheat, STB, resistance

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Interactions of plant-beneficial rhizosphere bacteria according to cereal genotype

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Roots of wheat and other cereals interact with a huge range of soil microorganisms, which colonize the rhizosphere and/or root tissues. This root-associated microbial community (sometimes termed the root microbiome) is important to consider because it includes both deleterious and symbiotic microorganisms. Therefore, it has a significant impact on plant growth, health and tolerance to stress. In this context, understanding root microbiome assembly and dynamics is of fundamental interest and could help design microbiome-based strategies for crop management. To address this issue, the significance of cereal genotype for root-microbiome interactions was studied. A relationship was found when comparing rhizobacterial communities with the phylogenetic distance between the corresponding plant genotypes, but this relationship held true only for a fraction of the community. It prompted us to assess how plant hosts selected for specific bacteria with plant-beneficial potential. This type of screening showed that the ability to interact with plant-beneficial bacteria had been largely counter-selected in modern wheat cultivars, yet it had been retained in some of them. The implications for plant-microbe interactions and breeding will be discussed.

Related publication:

Vacheron, J., G. Desbrosses, M.-L. Bouffaud, B. Touraine, Y. Moënne-Loccoz, D. Muller, L. Legendre, F. Wisniewski-Dyé, C. Prigent-Combaret. 2013. Plant growth-promoting rhizobacteria and root system functioning. Frontiers in Plant Science 4:356.

Bouffaud, M.-L., M.-A. Poirier, D. Muller, Y. Moënne-Loccoz. 2014. Root microbiome relates to plant host evolution in maize and other Poaceae. Environmental Microbiology 16:2804-2814.

Bruto, M., C. Prigent-Combaret, D. Muller, Y. Moënne-Loccoz. 2014. Analysis of genes contributing to plant-beneficial functions in plant growth-promoting rhizobacteria and related Proteobacteria. Scientific Reports 4:6261.

Bouffaud, M.-L., S. Renoud, Y. Moënne-Loccoz, D. Muller. 2016. Is plant evolutionary history impacting recruitment of diazotrophs and nifH expression in the rhizosphere? Scientific Reports 6:21690.

Vacheron, J., E. Combes-Meynet, V. Walker, B. Gouesnard, D. Muller, Y. Moënne-Loccoz, C. Prigent-Combaret. 2016. Expression on roots and contribution to maize phytostimulation of 1-aminocyclopropane-1-decarboxylate deaminase gene acdS in Pseudomonas fluorescens F113. Plant and Soil 407:187-202.

Cormier, F., J. Foulkes, B. Hirel, D. Gouache, Y. Moënne-Loccoz, J. Le Gouis. 2016. Breeding for increased nitrogen use efficiency: a review for wheat (*Triticum aestivum* L.). Plant Breeding 135:255–278.

Lemanceau, P., M. Blouin, D. Muller, Y. Moënne-Loccoz, Y. 2017. Let the core microbiota be functional. Trends in Plant Science 22:583–595.

Keywords: Rhizosphere, microbial community, microbiome, PGPR, phytoprotection, phytostimulation, QTL

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Developing phenotyping methods for quantitative host plant resistance using simulation modelling and ROC curves

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Components of resistance (CRs) constitute a conceptual stepping stone for resistance expression, from QTL (the geneticist's scale) to the cultivated field (the breeder's target). We propose an approach to identify predictors for field resistance, and to determine thresholds for genotype selection associated to sensitivity and specificity of the identified predictors. In a first stage, we use a process-based epidemiological simulation model that includes the effects of CRs on monocyclic parameters such as infection efficiency, latent period, infectious period, lesion expansion, and sporulation intensity. These effects correspond to relative resistances, RRs, ranging from 0 (susceptible) to 1 (resistant genotype). This model enables sensitivity analyses on the effects of RRs on simulates epidemics, which allow identifying CRs which affect most the speed of epidemics. This translates into a first result useful for breeding: the identification of traits (CRs) to target in phenotyping for field resistance. Each of the RRs of these traits represents a predictor for field resistance. In a second stage, epidemics are simulated considering genotypes displaying increasing levels of RR. The familiar decision framework for chemical control "spray / do not spray", with cases as epidemics and controls as non-epidemics, is translated into a decision framework for the breeder "retain / reject the genotype" where cases are resistant genotypes and controls are susceptible genotypes. Analyses using this framework allow identifying a value of RR to target in phenotyping according to levels of sensitivity or specificity using ROC curves. This approach can be applied to wheat diseases such as rusts and septoria to develop phenotyping methods for field resistance.

Related publication:
Willocquet L, Savary S, Yuen J. 2017. Multiscale phenotyping and decision strategies in breeding for resistance. Trends in Plant Science 22: 420-432. DOI : 10.1016/j.tplants.2017.01.009

Keywords: phenotyping, host plant resistance, plant disease epidemiology, breeding

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Genomics assisted improvement of *Fusarium* head blight resistance in bread wheat, durum wheat and triticale

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Fusarium Fungi are among the most feared pathogens of small grain cereals, particularly wheat. Resistance to *Fusarium* is therefore a top priority for resistance breeding, and receives high attention in research. The search string: TS=(*Fusarium* AND resistance AND wheat) in the ISI Web of Knowledge finds in the time frame 2000 -2017 more hits than any other wheat disease.

Apart from yield losses, the contamination of the crop with mycotoxins is the major issue associated with *Fusarium* head blight (FHB). *Fusarium* is an opportunistic pathogen, which preferentially penetrates and colonizes cereal florets during the flowering period. Environmental conditions which either favor or hamper the fungus and the status of the plant during flowering have therefore a huge impact on disease establishment and development.

Resistance to FHB can be classified into passive (morphological and developmental) factors and active (physiological, biochemical) factors, both of which play a role.

Typical passive resistance factors are plant height and the extent of anther extrusion during flowering. Based on previous comparative mapping studies we discovered that 1) anther extrusion is a clearly quantitative trait, 2) higher degree of anther extrusion is associated with reduced FHB susceptibility and 3) plant height genes, Rht-B1b and even more pronounced Rht-D1b are associated with increased FHB susceptibility and reduced anther extrusion, which could partly explain their effect on lowering FHB resistance.

The famous FHB resistance allele Fhb1, which was discovered in Chinese germplasm, residing on chromosome 3B, most likely has a different function. Fhb1 has not been linked to height or flowering traits. In our results, we always find Fhb1 associated with increased resistance to Fusarium spreading, and simultaneously to the toxin deoxynivalenol (DON). We could recently show that Fhb1 improves field resistance to FHB also in durum wheat and in triticale. Further tests on the functional characterization of Fhb1 in relation to DON detoxification are currently underway.

Apart from few large effect QTL for FHB resistance a great proportion of resistance is most likely due to numerous small effect genes. Breeders can make use of these small effect genes as well. Resistance selection utilizes often a skillful combination of phenotypic testing in provocation nurseries and of genome wide selection using genomic prediction approaches.

Related publication:

Prat N, Guilbert C, Prah U, Wachter E, Steiner B, Langin T, Robert O, Buerstmayr H (2017) QTL mapping of *Fusarium* head blight resistance in three related durum wheat populations. Theor Appl Genet 130:13-27 doi:10.1007/s00122-016-2785-0

Steiner B, Buerstmayr M, Michel S, Schweiger W, Lemmens M, Buerstmayr H (2017) Breeding strategies and advances in line selection for *Fusarium* head blight resistance in wheat. Tropical Plant Pathology 42:165-174 doi:10.1007/s40858-017-0127-7

Lemmens M, Steiner B, Sulyok M, Nicholson P, Mesterhazy A, Buerstmayr H (2016) Masked mycotoxins: does breeding for enhanced *Fusarium* head blight resistance result in more deoxynivalenol-3-glucoside in new wheat varieties? World Mycotoxin Journal 9:741-754 doi:10.3920/wmj2015.2029

Buerstmayr M, Buerstmayr H (2016) The Semidwarfing Alleles Rht-D1b and Rht-B1b Show Marked Differences in Their Associations with Anther-Retention in Wheat Heads and with *Fusarium* Head Blight Susceptibility. Phytopathology 106:1544-1552 doi:10.1094/phyto-05-16-0200-r

Buerstmayr M, Buerstmayr H (2015) Comparative mapping of quantitative trait loci for *Fusarium* head blight resistance and anther retention in the winter wheat population Capo x Arina. Theor Appl Genet 128:1519-1530 doi:10.1007/s00122-015-2527-8

Keywords: *Fusarium* head blight, resistance, wheat; triticale, durum wheat, food safety, feed safety

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GWAS reveals new race-specific resistance genes to powdery mildew in wheat

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Wheat powdery mildew, caused by *Blumeria graminis* f.sp. *tritici* (Bgt), is among the most important and persistent diseases of wheat and compromises its global production. To counter the disease, breeding for wheat powdery mildew resistance has been the main mean to control wheat powdery mildew. Quantitative resistance (QR) based on the joint action of several genes is of great importance. However, its use in breeding is very limited due to its multi-genetic nature, and, therefore, difficulty to select. Conversely, major resistance (R) genes have been extensively used in breeding as most of these genes show a simple dominant Mendelian inheritance, enabling an easy and rapid way to introgress resistance. R genes are also called race-specific resistance genes because they confer resistance to some but not all powdery mildew races through the recognition of pathogen effectors by proteins that generally have nucleotide-binding and leucine-rich repeats (NLRs). However, R-mediated resistance tends to be short-lived under agricultural conditions, as newly evolving pathogen races frequently defeat this type of resistance by modifying effectors recognized by the NLRs. Consequently, to ensure genetic resistance, a constant search and transfer of new R genes is necessary to counter the continuous evolution of virulence of the wheat powdery mildew.

To discover new sources of race-specific resistance against powdery mildew we performed a genome-wide association study (GWAS) using the WHEALBI collection of 506 geo-referenced inbred European and Middle Eastern wheat accessions genotyped by whole exome sequencing technology. The GWAS identified four genomic regions in chromosomes 1B, 1D, 2B and 5D associated with powdery mildew resistance. To identify candidate R genes, a genomic region of ~1Mb around the most significant SNP call for each association signal was defined. Afterwards, all open reading frames that potentially encoded proteins falling into those genomic regions were screened for NLR domains using InterProScan in search of full-length NLR type genes.

Strong associations were found for SNPs linked with the recently cloned Pm2a gene in chromosome 5D. Further haplotyping analysis and long-range PCR amplification and sequencing of the complete Pm2a coding region for the accessions showing polymorphism unveiled a new allele, temporarily named Pm2j. The Pm2j allele recognizes the AvrPm2 gene of the pathogen in the heterologous system *Nicotiana benthamiana* proving its biological functionality. Finally, the significant association of SNPs in chromosome 1D did not overlap with documented R genes conferring resistance to powdery mildew. Two predicted R genes in very close proximity to the peak signal show a high association between the haplotypes and the phenotypes and their functional validation is ongoing.

The identification and validation of a new allele in the Pm2 locus and two novel R genes in chromosome 1D demonstrates the power of GWAS for the rapid identification of major genes conferring resistance to powdery mildew. These results will enhance understanding of powdery mildew resistance and assist breeding for resistant wheat varieties.

Keywords: wheat, powdery mildew, GWAS, NLR genes, gene isolation

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Novel rye-wheat translocations in *Thinopyrum intermedium* wheat introgression lines enhancing *Wheat streak* mosaic virus resistance

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Pyramiding of alien-derived *Wheat streak mosaic virus* (WSMV) resistance genes in wheat is a cost-effective and environmentally safe strategy for disease control. PCR-based markers and genomic *in situ* hybridization (GISH) were applied to identify alien chromatin in four genetically diverse populations of wheat lines incorporating chromosome segments from *Thinopyrum intermedium* and *Secale cereale* (rye). Out of twenty experimental lines, ten carried *Th. intermedium* chromatin as 4Ai#2S.4DL translocation, one carried *Th. intermedium* chromatin on another D-genome chromosome, while seven lines were positive for alien chromatin (*Th. intermedium* or rye) on chromosome 1B. The newly described rye 1RS chromatin was associated with WSMV-resistance and has recombined with the 1BS arm. Under field conditions, 1RS chromatin alone showed some resistance, while 1RS chromatin together with the *Th. intermedium* 4Ai#2S offered superior resistance. Three translocation breakpoints between 1BS and alien chromatin of 1RS were recovered. Repetitive DNA probes proved excellent cytological markers for the detection of *Th. intermedium* 4Ai#2S arm in both alien-wheat lines and *Th. intermedium* genome. Labelled genomic *Th. intermedium* DNA hybridized to the rye 1RS chromatin under high stringency conditions, indicating micro-collinearity of the cereals group-1 and presence of potentially novel orthologous resistance and quality genes on *Th. intermedium* group-1. The presence of cryptic 1RS alien fragments may explain the difficulty of developing well-adapted lines carrying *Wsm1*. The results will facilitate development of agronomically desirable WSMV-resistant germplasm.

Related publication:
http://www.nature.com/hdy/journal/v117/n2/full/hdy201636a.html

Keywords: Wheat alien introgression, fluorescent in situ hybridization, chromosomes, molecular markers, *Thinopyrum intermedium*, Wheat streak mosaic virus

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Questioning the sustainability of quantitative physiological resistance: epidemiological and evolutionary responses of foliar fungal pathogens to changes in wheat plant traits

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Crop pathogens are known to rapidly adapt to agricultural practices. Although cultivar resistance breakdown and resistance to pesticides have been broadly studied, little is known about the adaptation of crop pathogens to more quantitative traits such as quantitative resistance. Quantitative resistance could be more sustainable than gene for gene resistance because it exerts a lower selective pressure on pathogens and relies on a variety of plant traits (and probably genes) rather than on a single one. Using a modelling approach, in this study we address the epidemiological and evolutionary responses of the pathogen to changes in several plant traits that impact epidemic development. With the model, we study life history evolution of biotrophic fungal pathogens of wheat. We focus on a single pathogen life history trait, the latent period, which directly determines the amount of resource allocated to growth and reproduction alongside the speed of canopy colonization. We investigate the evolutionary response of pathogens to changing several plant traits such as leaf metabolite concentration, leaf dimension and leaf lifespan. These plant traits impact epidemic development: disease severity is predicted to increase with metabolite content and leaf lifespan. We compare predictions of latent period evolution based on different "empirical" fitness measures such as annual spore production or within-season exponential growth rate, with predictions based on the more rigorous concept of invasion fitness from adaptive dynamics theory. For each of the studied plant traits, we use pairwise invisibility plots to identify evolutionarily stable strategies of the latent period (ESS). The ESS latent period responds differently to the different plant traits: it is longer on plants with long-lasting leaves and shorter on plants with bigger leaves leading to denser canopies. Our results further reveal that early canopy colonization during crop development might be a critical factor determining the issue of between-strain competition and shaping pathogen adaptation in the context of plant quantitative resistance. Finally, we argue that landscape-level heterogeneity may induce maladaptation of the pathogen that may be useful in stalling the evolutionary breakdown of quantitative resistance.

Related publication:
Précigout, P-A., Claessen, D., and Robert, C. (2017). Crop Fertilization Impacts Epidemics and Optimal Latent Period of Biotrophic Fungal Pathogens. Phytopathology PHYTO-01-17-001

Keywords: modelling, epidemiology, plant traits, ecophysiology, evolutionary ecology, plant pathogenic fungi, latent period, brown rust

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Session V

HIGH QUALITY AND SAFE CEREALS FOR FOOD, FEED AND INDUSTRY

Chair: Dario Fossati, Agroscope Switzerland

Keynote Lecture

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Importance of international collaboration on wheat quality and safety

Wheat quality and safety are very important for various end-use products. The Expert Working Groups (EWG) on improving wheat quality for processing and health was established under the Wheat Initiative (WI) in 2015 for maintaining and improving wheat quality and safety under varying environmental conditions. Our expert group focuses on wheat quality and safety in the broad sense, including seed proteins, allergens, carbohydrates, nutrition including micronutrients, processing and food safety. To share our data and advance our knowledge about the wheat quality and safety, it is important to standardize our methods and share standard cultivars representing allelic and phenotypic variations. We discuss to found a joint working group with ICC to standardize methods. For sharing standard cultivars, we selected standard cultivar sets representing the low-molecular-weight glutenin subunit alleles of bread and durum wheat. These sets have already become available for researchers at the CIMMYT gene bank. We are also working on developing other master sets for particular quality traits including carbohydrate diversity and low allergen content. We also have been collaborating with the EWG on Wheat Information System (WheatIS) to build connections between the Wheat Gene Catalogue and genome resources to facilitate the utilization of the genomic data for wheat breeding. I would like to discuss with participants the further promotion of international collaboration.

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Multiple to conquer: Simultaneous selection for grain yield and baking quality in genomics-assisted wheat breeding

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Genomic selection has been implemented in many national and international breeding programs in recent years. Numerous studies have shown the potential of this new breeding tool, few have however taking the simultaneous selection for multiple traits into account that is though common practice in applied breeding programs.

The simultaneous improvement of grain yield and baking quality is thereby a major challenge in wheat breeding due to the well-known negative trade-off. Accordingly, we investigated the potential and limits of multi-trait selection for this particular trait complex utilizing vast phenotypic and genomic data collected in a commercial wheat breeding program where genomic selection is implemented since 2012.

The accuracy of across-year predictions of both preliminary and multi-environment trials could be significantly improved when combining phenotypic and genomic information in a genomics-assisted selection approach, which surpassed both genomics-based and classical phenotypic selection methods. Genomic predictions for quality traits could furthermore be improved when additionally considering traditional measurements for indirect selection such as protein content and sedimentation value from the head-row stage.

Using these enhanced genomics-assisted breeding methods two strategies were compared, which 1) aimed to select high-quality genotypes with acceptable yield potential, and 2) develop high-yielding varieties, while maintaining baking quality characteristics. Different genomic selection indices were developed, whereby protein quality related to loaf volume and crumb structure was represented by dough rheological parameters. Forward prediction across years revealed that twice the response to selection could be achieved by genomics-assisted compared to phenotypic selection with both breeding strategies when considering protein yield as target criterion. Simultaneous selection for grain yield, protein content and protein quality showed a lower trade-off between grain yield and protein quality than protein content suggesting that it is feasible to develop varieties that combine superior yield potential with comparably high end-use quality, thus utilizing available nitrogen resources more efficiently.

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Genome-wide association studies of zinc, iron, copper, manganese and phosphorus in wheat (*Triticum aestivum* L.) grain and rachis at two developmental stages

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Globally, zinc and iron are two micronutrients that are most lacking in diets. The resulting micronutrient malnutrition can have detrimental impacts on overall health and wellbeing and affects approximately one third of the world's population, particularly young children [1-3]. Enhancing the densities of these minerals in bread wheat (*Triticum aestivum* L.) is a breeding objective to help alleviate these problems. A targeted biofortification breeding program initiated at the International Maize and Wheat Improvement Center (CIMMYT) established an association-mapping (AM) panel. This panel consists of 330 lines harbouring rich genetic diversity from resources possessing high Zn and Fe such as Triticum spelta, landraces and synthetic wheat [4]. Significant variation for grain Zn and Fe concentrations were found within the panel grown at various locations in India and Mexico [4, 5]. In this study, the panel was grown in Obregon (Mexico) over two seasons (2014-15 and 2015-16). Rachis and grain samples were taken at ten days after anthesis and at physiological maturity for the analysis of mineral concentrations. Phenology traits including days to heading, days to maturity and thousand kernel weight were also measured. Genome-wide association studies (GWAS) were carried out for five mineral concentrations, namely zinc, iron, copper, manganese and phosphorus with and without using phenology traits as covariates. For grain samples at physiological maturity, 40 to 60 significant marker-trait associations (MTA) ($p < 0.001$) were identified for each nutrient. Markers that showed pleiotropic effects on two or more nutrients were found on chromosomes 1B, 2D, 3B, 4A, 5B, 6B and 7B, of which those on chromosome 5BL were significantly associated with all five nutrients. Several significant MTAs were also identified for the nutrients in the rachis and grain at 10 days after anthesis. The genetic basis underlying the mechanisms that control the mineral concentrations at different developmental stages will be discussed.

Related publication:

Borrill, P., et al., Biofortification of wheat grain with iron and zinc: integrating novel genomic resources and knowledge from model crops. Frontiers in Plant Sciences, 2014. 5: p. 53.

Velu, G., et al., Biofortification strategies to increase grain zinc and iron concentrations in wheat. Journal of Cereal Science, 2014. 59(3): p. 365-372.

WHO The world health report 2002 - Reducing Risks, Promoting Healthy Life. 2002: Geneva.

Velu, G., et al., Genomic prediction for grain zinc and iron concentrations in spring wheat. Theoretical and Applied Genetics, 2016. 129(8): p. 1595-1605.

Velu, G., et al., Performance of biofortified spring wheat genotypes in target environments for grain zinc and iron concentrations. Field Crops Research, 2012. 137(Supplement C): p. 261-267.

Keywords: GWAS, iron, zinc, nutrition

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Biochemical and genetic analysis of arabinoxylan fibre in wheat grain

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Wheat is the most important source of dietary fibre in western diets, with the major component being arabinoxylan (accounting for about half of the total fibre in whole grain and about 70% in white flour). AX fibre occurs in insoluble and soluble forms, which may differ in their health benefits with soluble fibre being particularly important in modulating glycaemic response. Work carried out under the EU Healthgrain programme identified the Chinese breadmaking wheat cultivar Yumai 34 as having the highest content of AX in flour out of 150 diverse genotypes, with about half of this being water-soluble (compared with about 25% in most wheats). Analysis of AX structure by enzyme fingerprinting (digestion with endoxylanase followed by HP-AEC of the released oligosaccharides (AXOS)) also shows that the high content and solubility of AX in Yumai 34 are associated with structural differences. To determine the molecular basis for the high contents of total and soluble AX in Yumai 34, and to facilitate their exploitation by wheat breeders, we have analysed four populations (2RIL and 2DH) from crosses between Yumai 34 and either lines with normal fibre content (Ukrainka, Claire, Altai) or high fibre content (Valoris). We have mapped QTL for either viscosity of aqueous extracts (as a proxy for soluble AX) or total AX (by enzyme fingerprinting) and identified a major QTL, allowing the identification of candidate genes and the establishment of markers for selection.

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Session VI

FUTURE CHALLENGES AND INNOVATIONS

Chair: Roberto Tuberosa, University of Bologna, Italy

Keynote Lecture

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CRISPR-Cas9 technology in plant science

CRISPR-Cas9 technology has raised considerable interest among plant scientists, both in basic science and plant breeding. Presently the generation of random mutations at a predetermined site of the genome is well mastered just like the targeted insertion of transgenes, although both remain restricted to species or genotypes amenable for plant transformation. On the other hand, true gene editing, i. e. the deliberate replacement of one or several nucleotides of the genome in a predetermined fashion, is limited to some rather particular examples that generally concern genes allowing positive selection, for example tolerance to herbicides. Therefore, further technological developments are necessary to fully exploit the potential of gene editing in enlarging the gene pool beyond the natural variability available in a given species. In principle the technology can be applied to any quality related, agronomical or ecological trait, under the condition of upstream knowledge on the genes to be targeted and the precise modifications necessary to improve alleles. Published proof of concepts concern a wide range of agronomical traits, the most frequent being disease resistance, herbicide tolerance and the biochemical composition of harvested products. The regulatory status of the plants obtained by CRISPR-Cas9 technology raises numerous questions, in particular with regard to the plants that carry in their genomes the punctual modifications caused by the presence of the Cas9 nuclease but not the nuclease itself. Without clarification by the competent authorities, CRISPR-Cas9 technology would continue to be a powerful tool in functional genomics, but its potential in plant breeding would remain untapped.

Keywords: CRISPR-Cas9, plant breeding, genome editing, plant transformation

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Statistical methodology for modelling genotype by environment interactions in QTL mapping and genomic prediction, with applications to barley and wheat

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The most popular statistical methodology for QTL mapping and genomic prediction in plant breeding consists in some variations on linear mixed models with fixed QTL and random polygenic terms. The random polygenic effect is structured by a genotype x genotype matrix of relations to which both pedigree and marker information can contribute. In QTL mapping this random genetic effect is used as a correction term for background genetic variation in the search for QTLs, while in genomic prediction this term is the principal term producing the predictions. QTL and genomic prediction models for analysis of single traits and/or single environments are widely applied. Generalizations to multiple environments and traits while accounting for genotype by environment interactions are less common. Fixed QTL effects need to be defined in an environment specific way, while random polygenic effects need to be structured in a parsimonious way. For the latter, typically Kronecker products of structuring matrices on genotypes and environment are used. The computational requirements for fitting multi-environment mixed models strongly complicate model building and inference. For barley and wheat data from the EU-Whealbi project we present a mixed model framework for multi-environment QTL mapping and genomic prediction giving special attention to model building strategies, inference and interpretation.

Related publication:

Malosetti, M., Ribaut, J. M., & van Eeuwijk, F. A. (2013). The statistical analysis of multi-environment data: modeling genotype-by-environment interaction and its genetic basis. Frontiers in physiology, 4.

Millet, E. J., Welcker, C., Kruijer, W., Negro, S., Coupel-Ledru, A., Nicolas, S. D., ... & Presterl, T. (2016). Genome-wide analysis of yield in Europe: allelic effects vary with drought and heat scenarios. Plant physiology, 172(2), 749-764.

Thoen, M. P., Davila Olivas, N. H., Kloth, K. J., Coolen, S., Huang, P. P., Aarts, M. G., ... & Bucher, J. (2017). Genetic architecture of plant stress resistance: multi-trait genome-wide association mapping. New Phytologist, 213(3), 1346-1362.

Keywords: Adaptation, Genotype by environment interaction, genomic prediction, GWAS, multi-environment analysis, multi-trait analysis

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Zooming into GxE by means of modern field phenotyping techniques

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The combination of genomic prediction, modern crop phenotyping and modelling, will allow selecting crops in a way that was never possible before. Automated, sensor-based phenotyping techniques are well established under controlled conditions. Such techniques are now increasingly implemented in the field. This is an important step forward considering the large difficulties to mimic the field environment under controlled conditions. High throughput field phenotyping will completely alter the way we see plants. In the early days of quantitative genetics, mainly simple, static traits could be measured. Today, non-destructive, high throughput phenotyping offers the possibility to quantify allelic effects and their response to altering environmental conditions throughout the growing season.

We will demonstrate this concept using the example of wheat evaluated in the ETH field phenotyping platform (FIP; http://www.kp.ethz.ch/infrastructure/FIP.html). This unique prototype for automated, image-based crop phenotyping enables to follow the development of hundreds of plots. We evaluated a diverse panel of more than 300 genotypes consisting of the GABI wheat panel and important Swiss varieties. The overall aim within this project was to achieve a year-round phenotyping of wheat. We aimed to achieve a deeper understanding of wheat development during early growth (Grieder, Hund, and Walter 2015; Yu et al. 2017), stem elongation (Kronenberg et al. 2017) and senescence (See poster of Jonas Anderegg at this conference) including the response of these wheat genotypes to temperature and other environmental covariates. We will supply an overview of the heritability of the different traits assessed throughout the growing season and their dependencies. We will also highlight the major loci determining developmental processes across stages.

Related publication:

Grieder, Christoph, Andreas Hund, and Achim Walter. 2015. "Image Based Phenotyping during Winter: A Powerful Tool to Assess Wheat Genetic Variation in Growth Response to Temperature." Functional Plant Biology 42 (4):387. https://doi.org/10.1071/FP14226.

Kronenberg, Lukas, Kang Yu, Achim Walter, and Andreas Hund. 2017. "Monitoring the Dynamics of Wheat Stem Elongation: Genotypes Differ at Critical Stages." Euphytica 213 (7): UNSP 157. https://doi.org/10.1007/s10681-017-1940-2.

Yu, Kang, Norbert Kirchgessner, Christoph Grieder, Achim Walter, and Andreas Hund. 2017. "An Image Analysis Pipeline for Automated Classification of Imaging Light Conditions and for Quantification of Wheat Canopy Cover Time Series in Field Phenotyping." Plant Methods 13 (March):15. https://doi.org/10.1186/s13007-017-0168-4.

Keywords: Phenotyping, wheat, canopy development, association mapping, abiotic stress

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Optimum breeding schemes for line breeding in wheat

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Many new technologies are available for wheat breeding, for instance genomic selection (GS) or the use of doubled haploids (DHs). However, the question arise whether these technologies are of interest maximizing the selection gain in routine breeding. Particularly, costs in DH technology are still high compared to single seed descent (SSD) programs. Similarly, for GS, costs are still high and prediction abilities across breeding cycles low. I have therefore run computer simulations testing different breeding schemes under a given budget. The input variables are based on a survey between the members of the expert working group “Breeding Methods” of the International Wheat Initiative and orientate at recent elite wheat breeding in Central Europe.

Although DHs are considerably more expensive to produce than SSD lines, breeding schemes with DHs have a higher annual selection gain than breeding schemes with SSDs as long as the DH scheme is faster than the SSD scheme. If DH breeding schemes require several cycles of multiplication of seeds before yield testing, however, they are as slow as SSD schemes and have a lower annual selection gain than SSD schemes. Thus, the choice to use DHs or not should mainly be driven by the feasibility of a fast DH scheme.

Genomic prediction ability of grain yield across different breeding cycles is relatively low for wheat (r ~ 0.3). However, even with this low prediction ability, breeding schemes using one-stage genomic selection followed by one-stage phenotypic selection yield a higher annual selection gain than breeding schemes relying on two stages of phenotypic selection due to the shortened breeding cycle length with GS. The prediction ability, and thus also the selection gain, can be further improved by selective genotyping. This requires the phenotypic test of 5-15% poor lines, but seems to improve the prediction ability across breeding cycles (Zhao et al. 2012). My simulations clearly suggest that already small increases in the prediction ability justifies the phenotyping of a small proportion of poor lines maximizing annual selection gain and increasing further the potential of GS in elite breeding programs.

Related publication:

Zhao, Y., M. Gowda, C.F.H. Longin, T. Würschum, N. Ranc, and J.C. Reif. 2012. Impact of selective genotyping in the training population on accuracy and bias of genomic selection. Theoretical and Applied Genetics 125:707-713.

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A synthesis of crop growth and statistical genetic modelling to evaluate phenotyping strategies in wheat

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Complex target traits like yield show low genomic prediction accuracy because they frequently suffer from low heritability and are regulated by a large number of loci with small effects. Yield can be decomposed into a number of underlying genetically-correlated traits. As genetically-correlated traits are informative with respect to each other, modelling yield simultaneously with its component traits allows to achieve higher prediction accuracy compared to single-trait genomic prediction. Phenotyping additional traits implies an investment that does not always pay off by a larger prediction accuracy. Therefore, it is crucial for breeders to estimate in advance whether their phenotyping strategy for additional traits is likely to increase prediction accuracy of the target trait. This is especially relevant for high throughput phenotyping (HTP). HTP makes the phenotyping of additional traits affordable but may suffer from large measurement error. A large measurement error in the additional trait may lead to no contribution to the accuracy of the target trait. A strategy to evaluate the potential of phenotyping strategies is by combining crop growth models and statistical-genetic models to characterize trait correlations and heritability over time. Simulated data of intermediate and target traits during the growing season is a useful resource to evaluate; i) multi-trait prediction using traits measured during the whole growing season and ii) yield predictions from traits measured early in the growing season. In this presentation, we propose a decision support tool based on the combination of statistical-genetic and crop growth models to design an effective phenotyping schedule across the Australian TPE for wheat. We compare different strategies to integrate traits over time (i.e. penalized splines and nonlinear regression model), using an Australian wheat panel simulated with APSIM to grow over a sample of 40 environments representing water deficit patterns present in the Australian TPE.

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Genomic prediction of genotype by environment interactions by coupling genetic and physiological modellingt

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Genomic prediction (GP) models can be used to predict the performances of unphenotyped individuals thanks to their genotypic information. This approach was shown to be efficient for many species, but its interest in crops is limited by the presence of genotype x environment interactions (GEI): the ranking of the varieties often depends on the environments.

Recent studies proposed to adapt GP models to predict GEI by using environmental covariates. In the same way that molecular information is used to link the genotypes, the environmental covariates are used to link the environments. In these models environments with similar limiting factors are supposed to interact similarly with the varieties.

We propose here to evaluate and improve these approaches by using ecophysiological modelling. The prediction efficiency of different strategies were compared in a dataset comprising 220 elite wheat varieties, phenotyped for yield components in around 40 environments and genotyped with the TaBW420K SNP array within the BreedWheat project. We show that the use of environmental covariates increased prediction accuracy in comparison to additive models, and that crop models were efficient to derive environmental covariates more relevant than those directly obtained with pedoclimatic information.

Acknowledgements: we thank the BreedWheat program (ANR-10-BTBR-0003) for the production of this dataset and the Agreenskills+ program for the stay of R. Rincent in Wageningen University.

We thank the BreedWheat program (ANR-10-BTBR-0003) for the production of this dataset and the Agreenskills + program for the stay of R. Rincent in Wageningen University

Keywords: *Triticum aestivum* L., genomic prediction, genotype by environment interactions, crop modelling

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Rare and wanted! How to breed for favorable male floral traits required for hybrid wheat?

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The exploitation of heterosis in wheat promises to boost grain yield and yield stability. To make the 10 % mid-parent heterosis for grain yield in wheat also economically competitive, however, a number of issues need to be solved. Hybrid seed production of wheat is largely hampered by its low cross-pollination ability resulting in high seed production costs and lines with favorable male floral traits are displaying a severe bottleneck. We employed 31 wheat male lines and evaluated the hybrid seed set on two female tester lines. We observed a large genetic variance and high heritability estimates underscoring the possibility of selecting for increased hybrid seed set. Unfortunately, testing large numbers of candidates for seed set in crossing blocks under field conditions is cumbersome and is highly affected by secondary traits like plant height or heading date. Therefore, we evaluated trait correlations with more easy to screen indirect male floral traits. Visual anther extrusion appeared most promising showing a high association with seed set. We further evaluated genomics-assisted breeding strategies for indirect male floral traits. To this end, we employed a large panel of 209 winter wheat lines assessed for male floral traits, and genotyped with genome-wide markers as well as for Rht-B1 and Rht-D1. We found a complex genetic architecture underlying all observed male floral traits and Rht-D1 was identified as the only major QTL. Consequently, we evaluated genomic prediction approaches and fitting the identified major- and medium-effect QTL as fixed effects yielded the highest prediction accuracies and outperformed marker-assisted selection. Finally, we integrated our findings in an initial concept to breed for favorable male floral traits in wheat. Our results show that the redesign of the wheat floral architecture as required for efficient hybrid wheat breeding is challenging, but a combination of phenotypic and genomics-assisted selection can solve this issue in the following years.

Related publication:
Boeven PHG, Longin CFH, Leiser WL, Kollers S, Ebmeyer E, Würschum T (2016) Genetic architecture of male floral traits required for hybrid wheat breeding. Theor Appl Genet 129:2343–2357. doi:10.1007/s00122-016-2771-6

Keywords: Hybrid wheat, male floral traits, phenotyping, genomics-assisted breeding

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Differentially penalized regression improves genomic prediction of wheat flowering time

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Several models have been proposed for genomic prediction. Of these, ridge regression best linear unbiased prediction (RR-BLUP) is based on the assumption that all genotyped markers make equal genetic contributions to phenotypic variation. However, information from known marker-trait association and candidate genes is generally of greater significance to the phenotype under test and can be used to improve the accuracy of prediction. In this study, Differentially Penalized Regression (DiPR), a modification of standard RR-BLUP was used to make predictions of flowering time in an association mapping panel and a multi-parent advanced generation inter-cross (MAGIC) population. Predictive genetic markers for flowering time from an ongoing FSOV (fonds de soutien à l'obtention végétale) project, candidate adaptive gene markers, and genome-wide molecular markers were genotyped and genomic prediction conducted using DiPR to differentially weight all markers. Each marker set was also used independently and as a combined genotype data matrix to predict flowering time. Overall, the DiPR model performed better than single or combined marker sets for predicting flowering in both the association mapping panel and MAGIC population. This work demonstrates the importance of weighting genetic information to include key candidate gene and QTL markers in order to improve the accuracy of genomic selection.

Keywords: Genomic prediction, candidate genes, differentially penalized regression

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POSTERS

SESSION I. CHARACTERIZATION AND USE OF GENETIC RESOURCES

PI-01 - Hermann BÜERSTMAYR - Comparative genetic analysis of common bunt and dwarf bunt resistance in winter wheat

Comparative genetic analysis of common bunt and dwarf bunt resistance in winter wheat

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Bunts of wheat were a serious threat to wheat before the advent of effective fungicides. With the recent increase in organic wheat production, bunt diseases re-emerge as relevant wheat diseases. The major bunt pathogens in Europe are *Tilletia tritici* and *Tilletia laevis* causing common bunt and *Tilletia controversa* causing dwarf bunt. Infested wheat crops display varying levels of yield losses and quality losses due to the fishy smell of bunt spores caused by trimethylamine. Most current cultivars are bunt susceptible. Resistance to bunt diseases is therefore of increasing relevance, particularly for organic wheat production.

We evaluated three mapping populations for bunt resistance descending from crosses of parents with high resistance to common bunt and dwarf bunt crossed with a susceptible winter wheat cultivar. The resistant parents were Bonneville, Blizzard and PI199333. Bonneville, Blizzard are North American winter wheats with unknown resistance, and PI119333 is a landrace which is described as carrier of the gene Bt12. Susceptible parent was the Austrian cultivar Rainer.

Mapping populations were evaluated in nurseries inoculated with either common bunt or dwarf bunt, using local spore mixes. Bunt severity was evaluated in percent infected spikes per plot. Parents and populations were genotyped using a 15k Illumina SNP array. Phenotypic data were highly informative with broad sense heritability for common bunt severity of 0.94-0.97 and for dwarf bunt severity of 0.83-0.85. Marker data were used for linkage map construction and QTL mapping.

Both large effect and small effect QTL were detected. Blizzard and Bonneville share a large effect QTL for common bunt and dwarf bunt resistance on chromosome 1A (R2 19.5-28), and a large effect QTL for common bunt resistance on 1B (R2 29.6-34). Several smaller effect QTL for dwarf bunt resistance mapped to 2D, 7A and 7B. The Bt12 donor line PI119333 was tested for common bunt resistance only and possesses common bunt resistance QTL on 7D (R2 42.9) and 4B (R2 10.5).

In summary, several large effect alleles for resistance to dwarf bunt and/or common bunt were detected and mapped. These are promising targets for marker assisted selection in order to rapidly introgress bunt resistance into regionally adapted wheat cultivars.

Keywords: common bunt, dwarf bunt, resistance, QTL, mapping, wheat, *Tilletia*

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PI-02 - Nicholas FRADGLEY - The NIAB Diverse MAGIC wheat population: a community resource for QTL mapping

The NIAB Diverse MAGIC wheat population: a community resource for QTL mapping

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Development of experimental mapping populations enables dissection of complex traits in crops and underpins modern plant breeding. Plant Multi-parent Advanced Generation Inter-Cross (MAGIC) populations have now been constructed in many crop species. The advantages of MAGIC include greater genetic diversity captured from the multiple parents as well as multiple rounds of inter-crossing maximising recombination and minimising population structure.

The 'NIAB Diverse' MAGIC wheat population has recently been developed from 16 founders, chosen to capture over 90% of the genetic diversity of UK adapted wheat varieties based on markers. These included both elite and historic varieties originating from several different northern European countries.

596 RILs (Recombinant Inbred Lines) have recently been genotyped at F7, and a genetic map is under development. Preliminary results from the first year of replicated yield trials indicated high power to detect QTLs in heritable traits such as flowering time and height, and further analysis on a larger number of traits including yield and yield components is ongoing. These results confirm the NIAB Diverse MAGIC population as an excellent resource for genetic dissection of complex traits.

The population is being developed as community resource. A replicated yield trial will be available for phenotyping at NIAB-Cambridge in the 2018 season, and the population is available on request. Basic phenotype data and the founder/progeny genotype data will also be available, along with the associated genetic map and QTL analysis pipeline under development at UCL. Please contact nick.fradgley@niab.com for further information.

The MAGIC CARPeT' (MAGIC: Community Access to Resources, Protocols and Training) project is funded by the Biotechnology and Biological Sciences Research Council (BBSRC) under grants BB/M011666/1 (NIAB) and BB/M011585/1 (UCL)

Related publication:

Mackay, I., Powell, W. 2007. Methods for linkage disequilibrium mapping in crops. Trends Plant Sci., 12:57-63.

Allen, A. M., Winfield, M. O., Burridge, A. J., Downie, R. C., Benbow, H. R., Barker, G. L. A., Wilkinson, P. A., Coghill, J., Waterfall, C., Davassi, A., Scopes, G., Pirani, A., Webster, T., Brew, F., Bloor, C., Griffiths, S., Bentley, A. R., Alda, M., Jack, P., Phillips, A. L. Edwards, K. J. 2017. Characterization of a Wheat Breeders' Array suitable for high-throughput SNP genotyping of global accessions of hexaploid bread wheat (*Triticum aestivum*). Plant Biotechnol J. 15:390–401.

Keywords: wheat, multi-founder populations, MAGIC, QTL mapping, community resources

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From heritage to modern germplasm: evaluation of competitive ability, molecular characterization and genome-wide association in common wheat

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Traits such as tillering capacity, biomass accumulation before the stem elongation phase, final plant height, and flag leaf morphology are functional in determining the competitiveness of a crop against weeds. The use of competitive germplasm is a fundamental weed control strategy in organic and low-input agriculture. The trade-off between productivity in weed-free situations and competitive ability is a main obstacle to the release of competitive cultivars (Andrew et al., 2015). Yield potential remains a primary criterion for cultivar choice by farmers, therefore it would be desirable to prioritize traits that confer higher competitive ability without incurring in considerable grain yield reduction in the absence of weed competition.

Genome-wide association studies (GWAS) on competitive ability traits may be important for further advancing the knowledge about the genetic determination of the competition ability, and could be useful for increasing the information about the trade-off for yield and capability to resist to other stresses in common wheat.

In the present work, a *Triticum aestivum* collection summarizing the process of common wheat breeding in Italy during the latest hundred years was genetically characterized and screened for four traits related to competitiveness against weeds (above ground biomass accumulation before stem elongation phase, tillering index, plant height and flag leaf morphology) and two yield-related traits (grain yield and thousand kernel weight). The work was conducted with the aim to: (i) study the trends in four competitive ability traits and select genotypes with high competitive ability potential and with a combination of productivity and competitiveness adequate to organic and low-input systems; (ii) study the structure of the collection at molecular level and identify the main differences between heritage and modern germplasm; (iii) identify marker-trait associations (MTAs) for traits related to crop-weed interference.

The collection showed high phenotypic variability and molecular diversity. Accessions with high value for all competitiveness traits studied were identified, belonging mostly to the heritage germplasm. The accessions with the lowest grain yield to plant height trade-off were also identified. The genetic characterization highlighted a trend of polymorphism loss passing from heritage to modern germplasm and the presence of unique polymorphisms in both groups. This result underlines the importance of studying both heritage and elite germplasm. Marker-trait associations for all the evaluated traits were identified, both confirming associations already reported in the literature and describing new genomic regions that may disclose new breeding perspectives in common wheat.

The research was sponsored by the Italian Ministry of Agriculture in the framework of the Project RGV-FAO

Related publication:

Andrew, I.K.S., Storkey, J., Sparkes, D.L., 2015. A review of the potential for competitive cereal cultivars as a tool in integrated weed management. Weed Res. n/a-n/a. doi:10.1111/wre.12137

Keywords: Competitiveness, Crop-weed interaction, GWA (genome wide association), MTA (marker trait association), QTL (quantitative trait locus), *Triticum aestivum* L.

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Reconstruction of the triticale genome in the progeny of incompatible crosses

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The wild and cultivated relatives of family *Gramineae* are source of useful alleles for wheat improvement. Rye (*Secale cereale* L.) is a close relative of wheat (*Triticum aestivum* L.) that provides a vast genetic variation for commercially important traits such as stress tolerance, biomass, yield and photosynthetic potential not only for triticale, but also for wheat itself. This biodiversity is not yet sufficiently utilized in wheat and triticale breeding programs and only a limited number of the modern cultivars of wheat contain wild species in their pedigree. Postzygotic isolating mechanisms between the *Triticum* and *Secale* genera ensure that not every cross between wheat and rye results in a hybrid plant. Such postzygotic barriers as embryonic lethality and hybrid sterility, which are controlled by a pairs of complementary incompatible alleles of both parents, significantly limit the possibility of obtaining primary triticale and lead to a sharp decrease in the efficiency of hybridization of parental forms. The proposed approach allows to overcome these barriers effectively and to obtain highly fertile forms of primary triticale based on incompatible combinations. The use of GBS analysis of genotypes of the offsprings from such incompatible crosses made it possible to evaluate the role of somaclonal and combinative variability in the stabilization of new forms and to classify the rearrangements of the genomes of both parents in offsprings with different fertility levels.

This study was funded by the German Research Foundation (grant number BO 1423/17-1/603175).

Related publication:

Tikhenko N D, Tsvetkova N V, Lyholay A N, Voylokov A V. Identification of complementary genes of hybrid lethality in crosses of bread wheat and rye. Results and prospects. Russ. J. Genet. Appl. Res. 7 (2017) 153-158. dx.doi.org/10.1134/s2079059717020149

Tikhenko N, Poursarebani N, Rutten T, Schnurbusch T, Börner A Embryo lethality in wheat-rye hybrids: dosage effect and deletion bin mapping of the responsible wheat locus. Biol. Plant. 61 (2017) 342-348. dx.doi.org/10.1007/s10535-016-0691-6

Tikhenko N, Rutten T, Senula A, Rubtsova M, Keller E R J, Börner A The changes in the reproductive barrier between hexaploid wheat (*Triticum aestivum* L.) and rye (*Secale cereale* L.): different states lead to different fates. Planta 246 (2017) 377–388. dx.doi.org/10.1007/s00425-017-2694-8

Tikhenko N, Rutten T, Tsvetkova N, Voylokov A, Börner A (2015) Hybrid dwarfness in crosses between wheat (*Triticum aestivum* L.) and rye (*Secale cereale* L.): a new look at an old phenomenon. Plant Biol 17: 320-326.

Tikhenko N., N. Tsvetkova, S. Priyatkina, A. Voylokov & A. Börner. (2011) Gene mutations in rye causing embryo lethality in hybrids with wheat – allelism test and chromosomal localization. Biol Plantarum 55 (3): 448-452.

Keywords: postzygotic incompatibility, embryo lethality, embryo rescue, chromosome doubling, hybrid sterility, postzygotic incompatibility, wheat-rye hybrids, GBS analysis.

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BreedWheat GWAS data in GnpIS information system

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BreedWheat project (https://breedwheat.fr/) aims to support the competitiveness of the French wheat breeding sector, answering to societal challenges for a sustainable and quality production. Moreover, the BreedWheat project characterize yet poorly exploited genetic resources to expand the diversity of the elite germplasm. Finally, new breeding methods are developed and evaluated for their socioeconomic impact.

The URGI (Research Unit in Genomics Info) is an INRA research unit in genomics and bioinformatics dedicated to plants and their parasites. It develops and maintains an information system in genomics and genetics: GnpIS (Steinbach et al., Database 2013, doi: 10.1093/database/bat058).

BreedWheat data available in GnpIS are genetic resources (collection of 5,232 accessions), polymorphisms (724,020 SNPs from 10 sources), genotyping (Affymetrix Axiom TaBW420K array), phenotyping (48,000 micro-plots in 21 locations) and Genome Wide Association Study (775,621 association results calculated from phenotyping and genotyping values): https://wheat-urgi.versailles.inra.fr/Projects/BreedWheat.

GnpIS interface allows to display association values (with links to metadata, phenotyping and genotyping related values), these data can be filtered according to several criteria (eg p-val) and visualized graphically (QQplot, boxlplot based on genotyping alleles, Manhattan plot mapped to IWGSC RefSeq v1.0).

Related publication:
Steinbach et al., Database 2013, doi: 10.1093/database/bat058

Keywords: BreedWheat, GnpIS, GWAS, Association, Phenotyping, Genotyping, Genetic Resources

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Genomic variability of Spanish Durum wheat landraces

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One of the main goals for the XXI century breeding is the development of cultivars that can maintain current yields under unfavorable conditions. Wheat landraces that have been grown under diverse and extreme local conditions include variability that will be necessary to achieve this objective. Several recent international initiatives are characterizing local wheat germplasm collections. However, Spanish landraces, representing a really wide diversity of eco-climatic conditions, are absent or only represented by a small number of accessions.
The Center of Plant Genetic Resources of the Spanish Institute for Agriculture Research (CRF-INIA) holds a collection with 591 landraces of durum wheat (subsp. *turgidum*, *durum* and *dicoccum*). Based upon agro-ecological distribution, passport data and agro-mophological characterisation a core set up to 190 accessions was selected to represent the diversity in the national collection. This sample includes nine different Spanish durum wheat populations, according to previous molecular data.
We have performed GBS on the set of 190 accessions using the DArTseq technology. We have identified more than 90K DArTs (absence/presence) polymorphisms, and among the sequence obtained more than 50K SNPs. More than 50% of the markers have been mapped in the available bread wheat genome (TGACv1). These markers have been used to explore the genetic diversity present in the collection at genomic level, and the population structure obtained compared with previous analysis. Furthermore, to fully exploit the potential of durum wheat Spanish landraces, the collection is being phenotyped for a wide set of traits in order to conduct GWAs analysis.

Keywords: Durum wheat, DArTseq

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PI-07 - Davide GUERRA - Allele mining for frost tolerance in barley unveiled untapped alleles and copy number variants of CBF and Vrn-H1 genes

Allele mining for frost tolerance in barley unveiled untapped alleles and copy number variants of CBF and *Vrn-H1* genes

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In temperate cereals, a transcriptome reprogramming in response to low-freezing temperature relies on the action of the key family of CBFs (C-Repeat Binding Factors) transcription factors. In barley, more than 13 HvCBFs map to the major QTL FR-H2, making them as the best candidates to explain frost tolerance. A second major QTL controlling phenotypic variation in frost resistance is FR-H1, located 40 Mbp apart from FR-H2 and coincident to *Vrn-H1* vernalization response locus.

To identify new alleles governing frost tolerance, the allelic variation of specific HvCBF and *Vrn-H1* genes was assayed through exome capture and sequencing of a collection of 403 barley accessions spanning a wide geographic range and including modern and old cultivars, landraces and wild relatives. Analysis of the whole exome sequence data outputted more than 64 million genome-wide raw SNPs mainly in barley coding sequences.

This dataset along with curated annotation of the CBF and *Vrn-H1*genes allowed us to uncover SNP variants in HvCBF2, HvCBF3, HvCBF4, HvCBF9, HvCBF10, HvCBF14 and *Vrn-H1* and to predict functional effects at the protein level. Furthermore, whole exome sequencing was exploited to detect copy number variants (CNV) and insertions/deletions at FR-H2andVrn-H1 loci.

To validate this set of high confidence structural variants in silico approaches, qRT-PCR analysis and dataset querying for known gene deletions or duplications in barley genome were used. This work allowed to unveil different patterns of CNV affecting the FR-H2 locus, most of which encompass HvCBF2, HvCBF4 and HvCBF9 genes as well as several novel deletions in the promoter and in the first intron of *Vrn-H1*. Finally, an accurate phenotyping experiment was carried out to verify the impact of the novel CBF loci on frost tolerance.

This work provides a snapshot of the existing genetic diversity for FR-H2 and *Vrn-H1*/FR-H1 loci and paves the way for efficiently exploiting the natural genetic diversity of barley germplasms.

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Hyperspectral assessment of senescence dynamics and its relationship to grain yield and grain protein content in a high-yielding environment

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The stay-green phenotype (i.e. a prolonged maintenance of green leaf area after anthesis) has been linked to increased yield in several crops. In wheat, potential yield is still considered to be predominantly limited by sink strength, which is largely determined up until and including a short period after anthesis. However, individual grain weight is an important yield component which is predominately regulated post-anthesis and is affected by both the onset of senescence and the rate of the process. In particular, the efficiency of remobilization processes in the course of senescence has been proposed as a possible target to improve grain yield (GY) and grain protein content (GPC), as well as their stability across environments.

The effects of senescence dynamics on GY and GPC as well as senescence dynamics per se are strongly influenced by environmental conditions and have not been studied in detail on a large number of genotypes in high-yielding environments such as the Swiss plateau region. For genetic studies and a better understanding of the effects on GY and GPC, senescence dynamics need to be measured accurately on a large number of genotypes in several contrasting environments at high temporal resolution.

Here, hyperspectral reflectance measurements of a set of > 300 wheat genotypes in two years were used to compare the widely used normalized difference vegetation index (NDVI) to passive-sensor derived narrow-band indices and full-spectrum analysis to predict senescence-related traits. Phenotypic correlations between GY, GPC and senescence dynamics were also investigated.

Keywords: Senescence, Hyperspectral reflectance, Vegetation index

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Integrated genetic approaches to modify canopy architecture for improved barley biomass and yield

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In barley, genetic improvement of grain yield traditionally has been based on changes in plant architecture aimed at increasing harvest index and partitioning of biomass from straw to grains. With the increasing demands for renewable energy sources, plant biomass such as cereal straw is becoming more attractive. Barley straw is characterized by the largest content of carbohydrates among cereals, making it a valuable resource for its potential conversion into biofuels and other products. The BarPLUS consortium, a multidisciplinary group of scientists from different countries, is working to find genes, alleles, and candidate lines related to barley architecture and photosynthesis to maximize barley biomass and yield (https://barplus.wordpress.com/). In this framework, the major focus of our research group is optimizing tillering, leaf size and angle through integration of different genetic approaches exploiting both induced and natural allelic variation. Using a forward genetics approach, we screened the HorTillus population-derived from chemical mutagenesis of the barley cultivar "Sebastian"- under both field and controlled conditions and identified mutants with increased tillering and/or erect leaves: these are being targeted for mapping-by-sequencing in order to isolate the underlying genes. In parallel, we are analyzing natural genetic variation in biomass-related traits by genome wide association analysis on a set of 240 diverse barley accessions using exome capture data provided by the WHEALBI consortium (http://www.whealbi.eu/). Based on a preliminary analysis, heritability estimates revealed a substantial genetic component influencing leaf size and angle and several markers were found to be significantly associated with these traits. Through extensive literature search, bioinformatics analysis and comparative genomics, we identified a total of 48 candidate genes potentially involved in biomass-related traits. The sequences of these genes are being explored for mining functional variants in 403 barley accessions taking advantage of WHEALBI exome capture data. A subset of the candidate genes has been also subjected to TILLING to identify lines carrying mutated alleles. These resources will be characterized phenotypically to evaluate their effect on biomass traits. Together, this research will provide valuable knowledge and resources for potential improvement of barley biomass and yield.

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Genome scan to detect loci involved in culm morphological traits in a diverse barley collection

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Barley (*Hordeum vulgare* L.) is ranked fourth in worldwide cereal production and is used for animal feed, as malt and food for humans, and as biomass for biofuel production. Although the introduction of semi-dwarfing genes greatly improved stem sturdiness and harvest index, new plant ideotypes are needed to face the increasingly frequent extreme climatic events that threaten barley production. Morphological and anatomical features of the cereal culm have been linked to lodging resistance and biomass increase in rice and wheat, but information on the genetic basis of such traits is scarce in barley. As part of the ClimBar project, the objective of our studies is to explore and dissect natural genetic variation for culm traits in barley, using a germplasm collection of 233 diverse barley accessions. This germplasm panel was genotyped by exome capture sequencing, which provides an ideal basis for genome-wide association studies (GWAS). Our main focus was the second (basal) internode as a critical site for lodging in barley. Phenotypic data were collected from field-grown plants at both dough stage (Zadok 83) in Italy and at pre-harvest growth stage (Zadok 90) in four European countries with different climatic conditions. Straw culms were dissected and scanned images collected. Green culm sections were stained with Safranin and photographed using a stereomicroscope. ImageJ was used to extract quantitative data for culm diameter, culm wall thickness, and culm circularity. The number of vascular bundles from green culm images were also counted. Statistical analyses indicated the existence of significant genetic variation for the studied traits as supported by high heritability values. For GWAS, 176,000 SNPs from exome sequencing were considered, applying a minor allele frequency ≥0.05. Marker-based Principal Component Analysis (PCoA) indicated the presence of population structure mainly due to spike row type (2-rowed vs. 6-rowed), an important trait associated with domestication in barley. GWAS identified genomic regions associated with the studied traits and potential candidate genes. In addition, trial-dependent associations were detected, possibly due to the existence of GxE interactions for the traits. Samples from a second year of field trials (2017) are being processed and will be used to run integrated analyses and evaluate the stability of identified associations. Regions of special interest will be analysed to further select putative candidate genes and carry out allelic diversity studies.

This work was supported by the FACCE JPI CLIMBAR project. http://plen.ku.dk/english/research/plant_soil/breeding/quality/climbar/

Keywords: *Hordeum vulgare*, GWAS, culm architecture

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7BS.7HL wheat_barley robertsonian translocation line with increased salt tolerance and ß -glucan content

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One of the main challenge of current crop breeding is to improve yield stability and grain quality. Along with food production it is highly influenced by the conservation of biodiversity and responsible use of genetic resources. The modern wheat cultivars have excellent yield potential, and the new breeding programs put emphasis on breeding cultivars withbetter nutritional components (e.g. dietary fibres, essential amino acids), high resistance against diseases and better capacity of adaptation. Barley (*Hordeum vulgare* L., 2n=2x=14, HH) carries genes for abiotic stress tolerance and has good nutritional parameters. Cytologically stable, compensating wheat/alien translocations are promising gene sources for wheat improvement.

A QTL for salinity tolerance during germination was mapped on 7H chromosome. Salt stress analysis during germination of the Asakaze komugi/Manas 7HL ditelosomic addition line suggests that the 7HL chromosome arm is a good candidate for improving salinity tolerance of wheat during germination. In addition, a QTL responsible for ß-glucan content of the seeds was mapped on chromosome 7H.

The present study was initiated to develop compensating Robertsonian translocations involving wheat chromosome 7B and barley chromosome 7H and to analyse its effect on salt tolerance of the genotype developed and β-glucan content of the seeds. A 7B monosomic stock of ′Rannaja′ wheat was crossed with the disomic Asakaze/Manas wheat/barley 7H addition line. Plants carrying monosomic centric fusion were selected from F3 generation and the barley chromosome arm was identified using molecular markers. Fluorescent in situ hybridization using wheat specific repetitive DNA probes (pSc119.2, Afa family and pTa71) were used to identify wheat chromosome arm involved in the Robertsonian translocation. The F4 progenies homozygous for the presence of wheat/barley translocation were investigated from the viewpoint of salt tolerance and β-glucan content. The 7BS.7HL recombinant line exhibited higher salt tolerance both during germination and in the early developmental stages than the wheat parents. The expression of HvCslF6 barley gene in the wheat background and increased (1,3;1,4)-β-D-glucan content of this line proving that the 7BS.7HL translocation line could be of potential importance for improving β-glucan of wheat. The 7BS.7HL line is multiplicated in field trials.

The research leading to these results has received funding from the European Community's Seventh Framework Programme (FP7/ 2007-2013) under the grant agreement no. FP7- 613556, Whealbi project (http://www.whealbi.eu/project/ project), the MTA KEP 5/2016 (Hungarian Academy of Sciences) and by National Research, Development and Innovation Office (K112226). AC gratefully acknowledges support from a Marie Skłodowska-Curie Fellowship Grant (H2020-MSCA-IF-2016-752453)

Related publication:

Darko, E., Janda, T., Majlath, I., Szopko, D., Dulai, S., Molnar, I., Turkosi, E.,, Molnar-Lang, M. (2015): Salt stress response of wheat-barley addition lines carrying chromosomes from the winter barley "Manas". EUPHYTICA 203:(3) pp. 491-504.

Türkösi, E., Cseh, A., Darkó, É., Molnár-Láng, M. (2016): Addition of Manas barley chromosome arms to the hexaploid wheat genome. BMC GENETICS 17: Paper 87.

Darko, E., Gierczik, K., Hudák, O., Forgó, P., Pál, M., Türkösi, E., Kovács, V., Dulai, S., Majláth, I., Molnár, I., Janda, T., Márta Molnár-Láng (2017): Differing metabolic responses to salt stress in wheat-barley addition lines containing different 7H chromosomal fragments PLOS ONE 12:(3) Paper e0174170. 20 p.

Keywords: wheat/barley Robertsonian translocation, monosomic stock, wheat/barley disomic addition line, salt tolerance, ß-glucan content

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Dissecting the QTLome for protein content in durum wheat Mediterranean landraces

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Durum wheat (*Triticum turgidum* L. var. durum) was originated in the Fertile Crescent (10,000 BP) and spread over the Mediterranean Basin developing into local landraces specifically adapted to their growing regions. The wide genetic diversity and high level of polymorphism in quality genes of landraces can be used as a source of genetic variation quality traits in breeding programs (Nazco et al., 2012). Additionally, the decrease of protein content in durum wheat cultivars as a consequence of the release of high yielding cultivars (Subirà et al. 2014) make the use of landraces as a source of protein content.

The aim of this study was the identification by association mapping (AM), of molecular markers linked to protein content (PC, %) in a set of 169 durum wheat Mediterranean landraces. Experiments were carried out during the 2007, 2008 and 2009 harvesting seasons in north-eastern Spain, under rainfed conditions. PC analysis was performed as reported previously by Nazco et al. (2012). Phenotypic data was fitted to a linear mixed model to produce the best linear unbiased predictors (BLUPs). The durum wheat PstI/TaqI array v2.0 was used for genotyping the panel and 872 markers were ordered according to the consensus map of durum wheat developed by Maccaferri et al. (2014). AM was performed using a Mixed Linear Model and marker trait associations (MTA) were considered significant using a threshold of –log10(P) > 3. False discovery rate (FDR) was calculated for each trait.

Four 4 MTAs were significant for the PC in the 3 years and their calculated mean. The marker wPt-2737 was detected in 3 associations including the mean across years with a –10log(P) over the FDR threshold and explaining a high percentage of the phenotypic variance explained (Table 1).

To compare the associations reported in our collection with previously mapped QTLs, a meta-analysis was performed. A total of 70 QTLs for grain protein content, from 18 studies, were projected onto a durum wheat consensus map (Maccaferri et al. 2014).In total 15 meta-QTLs were identified and 23 QTLs remained as singletons. The marker wPt-2737 was located within a MQTL, while wPt-1140, wPt-7355 and tPt-5342 identified new genomic regions controlling PC.

Table 1. Significant marker trait associations (-10log(P)>3) obtained for protein content in 169 durum wheat Mediterranean landraces.

Marker	Year	Chromosome	Position (cM)	p-value	-10logP	Marker R2	Effecta
wPt-2737	**2007**	**7B**	**68.9**	**3.87E-04**	**3.41**	**0.08**	**0.71**
wPt-2737	**2008**	**7B**	**68.9**	**7.99E-04**	**3.10**	**0.07**	**0.55**
wPt-2737	**Mean**	**7B**	**68.9**	**2.98E-05**	**4.53***	**0.11**	**0.53**
wPt-1140	**2007**	**2B**	**133.4**	**7.01E-05**	**4.15**	**0.10**	**1.71**
tPt-5342	**2008**	**4B**	**16.5**	**4.25E-04**	**3.37**	**0.08**	**-0.78**
wPt-7355	**2007**	**4A**	**59.8**	**9.26E-04**	**3.03**	**0.08**	**0.74**

*(False Discovery Rate>4.24); a (allele presence effect).

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Related publication:

Maccaferri, M., et al. (2014). BMC Genomics, 15(1), 873.
Nazco, R., et al. (2012). Euphytica, 185(1), 1–17.
Subirà, J., et al. (2014). Crop & Pasture Science 65, 16–26.

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PI-13 - Roberto TUBEROSA - Development and characterization of a whole-genome radiation hybrid panel from reference tetraploid wheat cultivar Svevo

Development and characterization of a whole-genome radiation hybrid panel from reference tetraploid wheat cultivar Svevo

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The recent release of high-quality sequence information from hexaploid wheat (IWGSC 2014) coupled with the availability of high-density consensus maps for tetraploid wheat (Maccaferri et al. 2014) has accelerated marker and gene discovery in durum wheat (*Triticum durum*), thus facilitating the genetic dissection of agronomic traits. This notwithstanding, the construction of genetic maps remains a bottleneck for the investigation of the durum wheat genome. In this context, the development of physical mapping resources to facilitate the assembly of BAC contigs in future sequencing projects of durum wheat is becoming a primary necessity. Radiation hybrid (RH) mapping is a promising recombination-independent mapping approach, which involves the use of radiation-induced chromosomal breakage and marker segregation to reconstruct marker order (Tiwari et al. 2016). In this study, for the first time, a RH panel for tetraploid wheat was developed for reference durum genotype Svevo: the Svevo-Whole-Genome Radiation Hybrid (Sv-WGRH) panel. The Sv-WGRH panel was developed at Kansas State University (USA), according to the protocol reported by Tiwari et al. (2016). Freshly dehiscing pollen of Svevo was irradiated with γ-rays (10-Gy) and this was used to pollinate ~150 emasculated spikes of Senatore Cappelli (used as the female parent), which produced ~1300 RH1 seeds, each representing an independent RH event. Greenhouse planting of 1000 RH1 seeds resulted in ~730 RH1 plants, each representing a RH line of the Sv-WGRH panel. Initial assessment of the panel was performed on the DNA of 613 RH1 plants by means of 23 SSR markers. Results indicated that average marker retention frequency of the panel is ~87%, with 35% of RH1 lines having a retention frequency between 20 and 90%. Approximately 175 RH1 plants were obtained from planting 269 RH1 seeds in the greenhouse at DipSA UNIBO. DNA of these RH1 lines will be genotyped by means of the GBS technique in order to construct a physical map. This RH panel is an important resource contributing towards the assembly of the genome sequence of durum wheat and other tetraploid wheats and supporting positional cloning of important genes and QTLs in durum wheat.

Keywords: Durum wheat, physical map, radiation hybrid, sequencing, SSR markers

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Ph2 gene mapping through development and phenotyping of deletion lines in bread wheat

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Wheat (*Triticum aestivum* L.) emerged by hybridization of three closely-related species. Thus its genome consists of three highly similar sub-genomes (A, B and D) called homoeologs. The coexistence of similar homoeologous chromosomes led to establishment of a diploid-like system of homologous chromosome pairing mainly genetically controlled by two *Ph* (for pairing homoeologous) genes: *Ph1* and *Ph2*. *Ph2* was mapped to a distal region of 80 Mbp on the short arm of chromosome 3D. Mutants for this gene were developed and it was observed that *Ph2* has only a small effect on homoeologous wheat-chromosome pairing suppression. On the other hand, pairing of wheat and alien chromosomes was witnessed after removal of *Ph2* gene in hybrids derived from crosses between wheat and closely related species such as rye (*Secale cereale*). This discovery suggests a capability of *Ph2* gene to be used as a new breeding tool by introgression of alien genes into wheat gene pool. Positional cloning of *Ph2* would thus be of interest. However, the size of the actual deletion (80 Mb) hampers the identification of any candidate and it would be useful to reduce the deletion to a maximum of a few Mb.

The goal of the project is to scale down the *Ph2* gene region by deletion mapping up to 5 Mbp radius, so more precise mapping by radiation deletion lines can be performed. We have established a new set of deletion lines for a short arm of chromosome 3D, which is being currently extended. We focused on a distal 80 Mb part of a short arm of chromosome 3D, which is the pinpointed area of *Ph2* gene presence. We used the 2C gametocidal chromosome from *Aegilops cylindrica* as a tool for the development of the deletion lines after monosomic introduction into 'Chinese Spring' cultivar of wheat. Some gametocidal chromosomes introduced into wheat are inherited preferably by causing sterility of gametes in which they are absent. The sterility is caused by the ability of these chromosomes to induce genomic rearrangements. In some cases, these changes are not lethal, thus giving the opportunity to transfer aberration into progeny.

The novel deletion lines were characterized by molecular markers. These lines will now be crossed with rye to see if we observe the *ph2* mutant corresponding phenotype.

This work has been supported by the Czech Science Foundation (grant award 17-05341S) and the Ministry of Education, Youth and Sports of the Czech Republic (award LO1204 from the National Program of Sustainability I)

Keywords: *Ph2*, deletion lines, chromosome 3D, phenotyping

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Genetic structure of Mediterranean wheat landraces assessed with SNP markers

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Wheat is one of the most important grain crops in the world, being the hexaploid wheat (*Triticum aestivum* L.) the 95% of world wheat production. Landraces represent a valuable source of genetic diversity and are well adapted to local environmental conditions. Landraces may provide new alleles for the improvement of commercially valuable traits, being very useful especially in breeding for suboptimal environments as the Mediterranean Basin. Knowledge of genetic diversity is essential for understanding the relationships between cultivars and for the use of landraces in breeding programmes.

In this study, the genetic structure of a collection of 170 wheat landraces from 24 Mediterranean countries was ascertained using the wheat 15K SNP array. A total of 10,458 markers resulted polymorphic among the 170 genotypes. A model-based cluster method (Bayesian algorithm) using STRUCTURE software allocated 83.5% of the accessions in three subpopulations related to their geographic origin: North Africa and West of Mediterranean, East of Mediterranean and South Europe and North of Mediterranean.

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Genomic architecture of morphological and phenological traits associated with domestication syndrome in durum × wild emmer wheat RIL population

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Domestication and subsequent evolution under domestication of wheat caused substantial genetic changes, which affected plant morphology, physiology and phenology. Morphological characters, such as compactness of spikes, the number of side shoots, can be mentioned as domestication related traits in cereals. We suggest to consider the angle of side shoots (Ash) as a novel trait associated with the domestication syndrome. The objective of this study is to provide a better understanding of the antagonism between natural and man-made selection of the traits under domestication in order to identify the significant changes in phenology and morphology of wheat during domestication. We used a recombinant inbred line (RIL) population derived from a cross between *Triticum durum* (cv. Langdon) and *Triticum dicoccoides* (acc. G18-16) for mapping of quantitative trait loci (QTL) of five morphological and three phenological traits. A total of 36 QTL effects were identified that were co-located in 21 loci. Eight of these loci showed pleiotropic effects on the studied traits (including phenology). A major QTL effect of Ash, co-located with strong phenological effect, was identified on chromosome 2BL. We found that phenological loci affected the duration of flowering and development of wheat in different manners. The duration of the reproductive stage in cereals affects the development of apical meristem and many other morphological traits, such as the number of spikelets per spike and the number of side shoots. These results shed more light on shaping of wheat plant architecture and development during its evolution under domestication.

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Investigation of genetic diversity patterns and search for selective sweeps in a worldwide collection of rye

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Rye (*Secale cereale* L.) is an important cereal in Poland and other countries of Central and Eastern Europe. There are over 80 rye gene banks worldwide, with over 21,000 accessions. A better knowledge about the genetic variation contained in rye ex situ collections would facilitate both the conservation and management of genetic resources and their exploitation in breeding programs.

We compiled a rye germplasm collection consisting of over 600 accessions, representing major gene banks, different improvement status and various geographic origins. So far, 269 accessions were genotyped using Genotyping by Sequencing (DArTSeq). The plant material included 44 wild accessions, 105 landraces, 83 historical varieties, 14 modern varieties and 25 accessions of unknown improvement status. Based on 7,664 SilicoDArT and 2,559 SNP markers (missing data < 0.05, minor allele freq. >0.05) we conducted hierarchical clustering, principal coordinates analysis, analysis of molecular variance, and assignment tests. Distribution of polymorphism along rye chromosomes was also analyzed.

Obtained results allow us to conclude about the influence of improvement status, accessions source and geographical origin on the genetic diversity structure. Our aim is also an identification and characterization of genome regions targeted by selection during the domestication and breeding of rye.

This research was founded by the Polish National Science Centre grant No. DEC-2014/14/E/NZ9/00285

Keywords: rye, genetic diversity, genotyping by sequencing, DArTSeq, population structure, genetic resources

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PI-18 - Arantxa MONTEAGUDO - Detecting useful genetic diversity from Spanish landraces to improve an elite cultivar

Detecting useful genetic diversity from Spanish landraces to improve an elite cultivar

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Spanish Barley Core Collection (SBCC) has shown genetic variability with potential interest for breeding, particularly aiming at adaptability to Mediterranean environments. Cierzo, a successful elite barley cultivar selected under Spanish Mediterranean conditions, was chosen as recurrent parent of a backcross scheme. The donor parent, SBCC073, was selected among the SBCC lines due to its good yield performance under drought conditions. Under an augmented design with incomplete blocks, yield and drought-related traits and flowering time were evaluated in two field trials in Zaragoza (Spain). Raw data were spatially corrected using a moving average correction approach. Genotyping of 264 lines was carried out using the Barley Illumina iSelect 50k SNPchip. A genetic map was generated with 12,667 SNPs covering the seven linkage groups. Using the results of two years of field trials (2014-2015 and 2015-2016), we identified seven regions with stable QTLs. Among them, SBCC073 contributed favorable alleles for vegetative growth, measures in different moments of development, with a QTL in the region of 5H 138.5 cM, related also to flowering. The candidate of this region could be the response-regulator PRR95, component of the circadian clock. Another important region was found on 6H, at 59.1 cM, with QTLs for plant height, thousand-kernel weight and yield, being the SBCC073 allele responsible of increasing plant height and thousand-kernel weight, and reducing yield. No candidate gene has been found for this region yet. The QTLs found would be useful to develop improved breeding strategies based on exploiting the SBCC genetic diversity.

Keywords: Landraces, Yield, drought, flowering, QTLs

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PI-19 - Hanna Bolibok-Bragoszewska - Highly parallel amplicon sequencing for characterization of genetic diversity in rye germplasm

Highly parallel amplicon sequencing for characterization of genetic diversity in rye germplasm

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Improved plant cultivars will be required to feed the world's growing population, while preserving natural resources, and to face climatic changes. Novel, favorable alleles can be found within the crop - mostly in landraces and also in the wild relatives. The aim of the project was to develop a low-cost amplicon sequencing approach employing high sample pooling suitable for assessment of genetic diversity in heterozygous and heterogenous rye accessions.

We performed a pilot screen involving 95 diverse accessions of different origin and improvement status (landraces, historical and contemporary varieties, wild species). Each accession was represented by a pooled DNA sample representing 96 individuals, thus 9,120 plants were analyzed in total. Six genes: aluminium activated citrate transporter (AACT1), taumatin-like protein (TLP), fructose-biphosphate aldolase (FBA), prolamine binding factor (PBF), secaloindoline-b (SEC-B) and grain softness protein (GSP1), ranging in length from 456 to 4,638 bp, were amplified from the 95 pooled samples. Amplicon sequencing was done using an Illumina MiSeq with 2x300PE reads. Due to small scale of the experiment a high depth of coverage was obtained (above 1000x for the lowest represented amplicon). So far, 869 polymorphic sites with 990 variant alleles were identified using the GATK SNP-discovery tool. Allele frequencies varied from 0.001 to 1.0 and 25% of all values were in the range 0.1 – 0.2. Among detected variants, 28% (243) appeared in only one accession from the set, and the number of polymorphic sites per accession ranged from 111 to 337. In a validation experiment, based on a small number of individual plants (10-20) from selected accessions and information on restriction enzyme site gain/loss provided by PARSESNP, we were able to confirm SNPs occurring with frequencies above 0.1. Currently, evaluation of nucleotide variation is being carried out using SNVer, FreeBayes and CAMBa. Nucleotide changes are further evaluated for their effect on gene function.

We conclude that the proposed experimental design allows for reliable, high-throughput SNP discovery and accurate variant frequency estimation in large rye germplasm sets.

This research was founded by the Polish National Science Centre grant No. DEC-2014/14/E/NZ9/00285

Keywords: rye, NGS, genetic diversity, Ecotilling, amplicon sequencing

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PI-20 - Ian DAWSON - Exploring local adaptation in barley landraces through latitudinal differences in days to heading

Exploring local adaptation in barley landraces through latitudinal differences in days to heading

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Landraces are defined as being adapted to their local production systems, but actual tests of their adaptation are limited, despite the importance of the issue for designing crop-breeding strategies that respond appropriately to anthropogenic climate change. Here, we explore the issue of local adaptation through assessing the heading dates of spring-type barley landraces and formally bred cultivars sampled from across a wide latitudinal range in spring-planted field experiments including those of the current WHEALBI initiative. Our tests provide some evidence for local adaptation in barley landraces that supports conventional – but frequently unproven – wisdom, but reveal a number of caveats and limitations in the applied approach. We discuss more optimal approaches to explore adaptation more widely.

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Coexpression network and phenotypic analysis identify metabolic pathways associated with the effect of warming on grain yield components in wheat

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Wheat grains are an important source of human food but current production amounts cannot meet world needs. Several environmental constraints such as heat could potentially negatively affect wheat production. Therefore, it is important to understand the response of grains during the development to an increase of ambient temperature in a global warming context.

Plants from two wheat genotypes have been submitted to two growth temperature regimes. One set has been grown at optimal mean daily temperature of 19°C while the second set of plants has been submitted to warming at 27°C from 2 days after anthesis (daa) to 13 daa. Gene expression was evaluated using micro-array during nine different seed developmental stages and agronomic traits were measured at harvest.

While warming did not affect mean grain number per spike, it significantly reduced other yield-related indicators such as grain width, length, volume and maximal number in the endosperm. Whole genome expression analysis identified 6,258 and 5,220 genes whose expression was affected by temperature in the two genotypes respectively. The greater difference between low and high temperature was displayed at a key stage 120°C days after anthesis (°Cd) which suggest that significant transcriptional changes occur during the transition that encompasses the 120°Cd stage in response to warming.

Co-expression analysis using WGCNA uncovered modules associated with agronomic traits. In particular, modules enriched in genes related to nutrient reservoir and endopeptidase inhibitor activities were found positively associated with cell number in the endosperm. A hypothetical model pertaining to the effects of warming on gene expression and growth in wheat grain is proposed.

Network analyses suggest a negative effect of the expression of genes related to seed storage proteins and starch biosynthesis upon moderately high temperature on the grain size in wheat.

Keywords: wheat, elevation of temperature, grain size, grain development, transcriptomic, WGCNA

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PI-22 - José Miguel SORIANO - Eastern and Western durum wheat Mediterranean landraces differ in marker alleles responsible for yield components and phenology as revealed by association analysis

Eastern and Western durum wheat Mediterranean landraces differ in marker alleles responsible for yield components and phenology as revealed by association analysis

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A panel of 172 durum wheat landraces from 21 Mediterranean countries were phenotyped during 3 years in northern and southern locations from Spain for 14 traits including phenology, biomass, yield and yield components.

The genetic structure of the collection was ascertained with 44 simple sequence repeat markers allocating 90% of the accessions in four subpopulations related to their geographic origin: eastern Mediterranean, eastern Balkans and Turkey, western Balkans and Egypt, and western Mediterranean.

Association mapping was used to identify genome regions affecting yield formation, crop phenology and crop biomass. The panel was genotyped with 1,149 DArT markers. A total of 245 significant marker trait associations (MTAs) were detected. Eighty-six MTAs corresponded with yield and yield component traits, 70 to phenology and 89 to biomass production. Twelve genomic regions harbouring stable MTAs were identified, while five and two regions showed specific MTAs for northern and southern environments, respectively. Sixty per cent of MTAs were located on the B genome and 29% on the A genome.

QTL meta-analysis was performed to compare MTAs with previously identified QTLs. A total of 477 unique QTLs were projected onto a durum wheat consensus map and were condensed to 71 meta-QTLs and left 13 QTLs as singletons. Sixty-one percent of QTLs explained less than 10% of the phenotypic variance confirming the high genetic complexity of the traits analysed.

Using a subset of the landrace panel 23 marker alleles were identified with a different frequency in landraces from east and west of the Mediterranean Basin, which affected important agronomic traits. Eastern landraces had higher frequencies than the western ones of alleles increasing the number of spikes, grains per m2, and grain filling duration. Eastern landraces had higher frequencies of marker alleles associated with reduced cycle length, and lighter grains than the western ones. Breeders may use the molecular markers identified in the current study for improving yield under specific Mediterranean environments.

Related publication:

Soriano JM, Villegas D, Aranzana MJ, Garcia del Moral LF, Royo C (2016) Genetic structure of modern durum wheat cultivars and Mediterranean landraces matches with their agronomic performance. PLoS One. 11 (8): 0160983.

Soriano JM, Malosetti M, Roselló M, Sorrells ME, Royo C (2017)Dissecting the old Mediterranean durum wheat genetic architecture for phenology, biomass and yield formation by association mapping and QTL meta-analysis. PLoS ONE 12 (5): e0178290

Keywords: Durum wheat, genetic structure, association mapping, meta-QTL, Mediterranean Basin

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PI-23 - Monika RAKOCZY-TROJANOWSKA - New aspects of the genetic background of benzoxazinoid biosynthesis in rye (*Secale cereale* L.)

New aspects of the genetic background of benzoxazinoid biosynthesis in rye (*Secale cereale* L.)

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Benzoxazinoids (BX) are the major protective and allelopathic secondary metabolites that are synthesized in many species of the *Poaceae* and, sporadically, in single species of the dicots. Despite years of research many problems connected with BX remain unsolved. Especially ambiguous is the role of two genes controlling indole synthesis: Bx1 and Igl, both taking part in transformation of indole-3-glycerolphosphate (IGP) to indole. According to the general opinion: (1) Bx1 codes for BX1 enzyme catalyzing the conversion of IGP to indole, which is further used in BX biosynthesis. Igl (its rye orthologue), that has been recently isolated by our team, codes for indole-3-glycerol phosphate lyase (IGL) which, “produces” free indole as a response to the stress agent; (2) the Bx1 gene is under developmental control, and is mainly responsible for BX production, whereas the Igl gene is inducible by stress signals, such as wounding, herbivory, or jasmonates. However, the results of our previous study contradict this concept and show, that the expression of ScBx1 gene increases in the native conditions until the third day after germination and then decreases rapidly, while the BX are still synthesized at a constant level. The situation changed completely after the BSMV infection – the expression of ScBx1 increased nearly 40 fold (!). Quite poorly understood aspect of the genetic background of BX biosynthesis is the regulation of Bx gene expression. The majority of studies were focused on the influence of BX on pests, pathogenic fungi and weeds. The knowledge about the opposite relationship: how different factors affect Bx gene expression is still fragmentary and limited. So far, it has not been investigated whether the induction of BX synthesis in the above ground parts of plants is accompanied by an increase in the production of these compounds in the roots (and vice versa) and what is the mechanism of signalling. Another question concerns the sites of BX synthesis - whether it takes place only in the above ground parts of plants or also in the roots.

With regard to rye, we have been able to clarify some of these doubts. Three rye inbred lines: D33, L318 and D39 characterized, respectively, by high, middle and low content of BX in the above ground parts and roots (measured in the early spring) were used.

The main results of the reported study are presented below.

1. *ScIgL* gene has been isolated from line L318 BAC library and characterized. *ScIgl* is predicted to be composed of seven exons of total length equal 989 bp. Its complete length (including introns) is 1,651 bp. The results of bioinformatic analysis (presence and frequency of specific motifs potentially involved in different abiotic and biotic stress response) suggest that *ScIgL* gene plays role in different defense processes.
2. It was proved that the expression of *ScBx1* and *ScIgL* genes take place both in the above ground parts of rye plants and in roots. It denies the current opinion according to which a step of BX synthesis controlled by *Bx1* is located in chloroplasts.
3. The examination of expression profiles of genes *ScBx1* – *ScBx5* and *ScIgL* in plants affected by allochemicals secreted by berseem clover (*Trifolium alexandrinum* L.) showed that: (i) compared to control (cultivation without berseem clover), their expression usually decreased after 2 weeks, after the next 2 weeks – it increased and in the sixth week, the significantly heightened expression of the majority of genes (except for *ScBx1*) was observed only in line D39; (ii) the expression level of *ScBx1* decreased while in case of *ScIgL* - it increased in the above parts of plants. (iii) line D39 was characterized by the highest enhance of an expression due to allochemicals of the majority of genes up to the fourth week of co-cultivation with berseem clover (iv) among six investigated genes, *ScIgL* and *ScBx5* were most affected by allochemicals in the case of the above ground parts of plants, and *ScBx3*, *ScBx4* and *ScBx5* in the case of roots, especially after 4 weeks of co-cultivation with berseem clover, (v) an increase in the level of DIBOA

synthesis in the above ground parts of plants was observed after 2 weeks of co-culture with berseem clover in case of line D33, in fourth week – for line L318 and in sixth week - for line D39 which not fully correlate with gene expression profiles. Full interpretation of these results will be possible after the completion of biochemical analyses of BX content in roots which are currently in progress.

The obtained results not only broadened the current knowledge about the genetic basis of BX biosynthesis, but they also changed some of the existing dogmas, at least those concerning the places of BX biosynthesis and the regulation of ScBx and ScIgL gene expression by some stresses. Perhaps our findings will change the opinion that only Bx1 takes part in the biosynthesis of BX and is developmentally regulated whereas *Igl* controls the synthesis of free indole emitted by plants as a response to the stress.

The research has been financed by the National Science Centre (Poland) – No UMO-2015/19/B/NZ9/00921

Keywords: rye, *Secale cereale* L., *ScBx* gene, *ScIgL* gene, gene expression, allelolopathy, biotic stress, berseem clover, *Trifolium alexandrinum* L.

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Mapping QTL for drought tolerance in spring wheat genebank accessions using genome wide association approach

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Drought is one of the most important abiotic stresses affects yield production in cereal crops world-wide. As a result of climate changes, drought severity is expected to increase in the future. In order to understand the genetic variation and to detect quantitative trait loci for drought tolerance, a set of 111 spring wheat genebank accessions from 27 countries and genotyped with a 15k chip was grown in the field in two consecutive years. Drought stress was applied by chemical desiccation using potassium iodide (KI, 0.5% w/v) applied 14 days after anthesis to simulate post anthesis drought stress. In addition, a rain-shelter experiment was initiated at the Center for Agricultural Research in Martonvásár, Hungary during one season. The accessions were evaluated for a number of morphological and agronomical traits such as plant height, spike length, grain number and thousand-grain weight. Genome-wide association study (GWAS) was performed to reveal significant marker-trait associations (MTAs) under drought and control conditions. Analysis showed that drought stress had strong impact on yield parameters. Significant differences between genotypes and GWAS analysis revealed major MTAs under control, chemical desiccation and rain-shelter conditions.

Keywords: Drought, Chemical desiccation, Genome wide association

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SESSION II. GENOMICS AND EPIGENOMICS IN THE POST SEQUENCE ERA

PII-25 - Lukas KRONENBERG - Wheat stem elongation dynamics assessed by GWAS: short term growth rate depends on temperature

Wheat stem elongation dynamics assessed by GWAS: short term growth rate depends on temperature

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Stem elongation is a critical phase for yield formation in wheat. During this period, active spike growth takes place as the pike is pushed up the stem. Spike dry weight at anthesis is closely related to grain number per spike, a critical determinant of potential yield in wheat. A prolongation of stem elongation has therefore been proposed to increase dry matter accumulation by the spike, increasing potential yield.

By using terrestrial 3D-Laserscanning mounted on a novel high throughput phenotyping platform, stem elongation dynamics were monitored on a set of > 300 commercial European wheat varieties (GABI wheat panel) in high temporal resolution. Results showed that there is large genotypic variation in duration as well as growing degree-days until start and end of stem elongation and that those traits are partly interdependent. Genome wide association studies revealed stable and highly significant quantitative trait loci (QTL) across three years for start and end of stem elongation. Canopy height measurement in high temporal resolution allowed to investigate short term growth response to temperature during stem elongation. Preliminary results indicate a stable QTL for this trait as well as a connection to final height. Start of stem elongation is a key stage in wheat development and the large genetic variation suggests opportunities for breeding. However, traditional rating of this trait is low in throughput and labor intensive. New phenotyping techniques allow to examine the genetic basis of these difficult-to-phenotype trait and offer opportunities for breeding via marker assisted selection.

Related publication:
Kronenberg, L., Yu, K., Walter, A. et al. Euphytica (2017) 213: 157. https://doi.org/10.1007/s10681-017-1940-2

Keywords: Wheat, Stem elongation, GWAS, Growth, Phenotyping

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Unmasking new intra-species diversity through k-mer count analysis

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High-throughput sequencing is often used to examine intra-species diversity. Most studies are focused on calling and genotyping SNPs. Other kinds of genomic variation, such as copy-number variation (CNV), are more rarely exploited despite literature reports linking them to phenotypic differences.

For some loci, it is difficult to identify reliable SNPs. For instance, reads from closely related sequences (e.g. paralog genes) will often map stacked to the same location if some of those loci are absent from the reference sequence. Such piled up mappings produce abundant fake heterozygous SNPs, and thus have been called apparent heterozygous mappings (AHMs).

To avoid wrong conclusions from false positive calls, SNPs from AHMs are often discarded, either in early (e.g. samples expected to be homozygous), or in downstream steps of the analysis (e.g. when incoherent haplotype blocks are identified). This would lead to information loss at certain loci.

AHMs can be seen as a kind of CNV which is specific to non-identical copies. Unmasking such variation could help to i) assess the completeness of a genome or pan-genome reference, ii) confirm results from other CNV genotyping methods, when the copies originate in non-identical loci, iii) provide hints about the history and behavior of duplicating DNA loci, and iv) reveal novel intra-species genetic diversity.

Here we present a software pipeline, kmeleon, available at https://github.com/eead-csic-compbio/kmeleon, designed to identify regions harboring AHMs. kmeleon is based on mappings, and thus it can be used for both homozygous and heterozygous samples. First, the different k-mers (sequences of length k) mapping to a single locus are identified and counted. Then, loci are classified based on the presence or absence of AHMs. From those intervals, it is straightforward to perform comparisons between genotypes, or to translate existing annotation to the regions with AHMs.

We used exome capture data to detect AHMs in a set of barley accessions. We included the cultivar Morex, the genotype of the genome reference, as a control sample. As expected, it had the lowest number of AHMs, although some were still detectable. For all accessions, AHMs were found both in inter- and intragenic loci. Enrichment analysis showed that NBS-LRR proteins were overrepresented at AHMs, whereas PPRs proteins were depleted. Also, we will show that AHMs can be used to infer phylogenetic trees which are congruent to those produced with SNP-based approaches, supporting the information value, of this hidden variability, to describe genetic relationships.

Keywords: cnv genome diversity bioinformatics genotyping

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Genetic improvement facing climate challenging: integrating classical and novel approaches

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Growing on over six million acres in Oklahoma, production of winter wheat that plays a vital role in both grain production and cattle industry in the US, is subject to the worsening drought condition in the southern Great Plains. A double-haploid (DH) Buster population of 282 DH lines derived from the intercross of Duster and Billings was evaluated under condition with various degrees of water stress. To provide genomic predictors, the release of Buster_hmp_v1.0 contains 213, 940 SNPs from both genotyping-by-sequencing (GBS) and exome capture techniques, after removing non-informative and erroneous SNPs. Genome-wide association mapping (GWAS) identified SNP variants significantly associated both yield and quality phenotypes; however, genetic response to environmental perturbations (G×E) clearly demonstrated impact on individual year's yield potential, with a total of 85 variants significantly showing differential response based on a single locus analysis. A closer look of transcript assemblies revealed five genes in drought tolerant genotype and 82 genes in drought susceptible genotype that, mostly related to photosynthetic activity, exhibit high levels of transcriptional variation in response to drought stress.

Further, significant overinflation in accuracy was found when examining genomic selection (GS) performance across field condition. With a more realistic cross-season validation, our results reckoned a nearly 50% reduction in GS accuracy for grain yield can be expected due to environmental variability; this overinflation was most discernible for wheat protein content amongst the 13 traits examined; showing an overall 45% prediction accuracy, adjusted sedimentation was however the most predictable end-user quality phenotype, despite the difference in water pressure. Finally, the input genotypic information used in GS algorithms can also be a determining factor for GS predictability. Our results indicate that including SNPs derived from functional genomic components, such as the exome capture technique that uses expressed mRNA as templates for probe construction, could lead to more predictable outcomes in well-watered condition; GBS, however, shows slight advantage when predicting grain yield under severe drought.

Keywords: genomic resource, GxE, genomic selection

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SESSION III. YIELD POTENTIAL AND RESOURCE EFFICIENCY

PIII-28 - Elizabeth CHAPMAN - Combining staygreen and altered phenology to improve wheat productivity

Combining staygreen and altered phenology to improve wheat productivity

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Staygreen plants demonstrate delayed, or altered, senescence profiles, but need to remain photosynthetically active for longer to be of interest. In wheat and other grain crops, functional staygreens have demonstrated yield benefits, especially under stress, including; - heat, drought, and low nitrogen environments. Often physiological adaptations are associated with the staygreen trait, including altered stomatal conductance, leaf, and root architecture. However, the underlying genetics of senescence, and staygreen, are largely unknown, especially in wheat, aside from NAM genes and certain NAC transcription factors.

Originally three staygreen EMS mutant *Triticum aestivum*, cv Paragon, lines were identified during a nitrogen use efficiency trial, which concluded the staygreen trait to be environmentally stable over a 3-year period. These three lines differ relative to their parental cv Paragon in both their senescence profiles; subtle to extreme staygreens; and yield; nitrogen insensitive to high yielding.

Two single seed descent (SSD) populations were developed per staygreen, one of which was a segregating population designed for mapping the mutation(s) (Staygreen x Paragon). The second, a population segregating for phenology and senescence, through crossing with a triple photoperiod (Ppd) insensitive Paragon near isogenic line (NIL); carrying Ppd-A1a, Ppd-B1a, Ppd-D1a. Within these populations, there exists variation in heading date of up to 3 weeks under UK field conditions, within an isogenic background.

Early heading and grain maturity traits are typically adapted to avoid heat/drought stress. Unfortunately, this can negatively affect pollen viability and impair grain filling, whilst it is hypothesised staygreens are associated with stress adaptation under such environments. Thus, can combining early flowering and staygreen result in synergism to increase wheat productivity? Alternatively, does this unite conflicting life history strategies, potentially negatively affecting yield or quality?

A selection of ‘early’ and ‘late’ heading segregants, demonstrating staygreen and normal senescence profiles were phenotyped for phenology, yield and yield components during 2015-16 and 2016-17 in UK and France. For 2016 flowering time, rather than senescence, was responsible for significant differences in yield between groups, attributable to low light, but high rainfall, levels. In contrast, 2017 witnessed prolonged periods of drought, prior to late season rainfall, thoroughly testing stress tolerance of these staygreen, especially regarding the early flowering ± staygreen material grown in France.

What effect does staygreen have on grain filling dynamics? Initial grain filling experiments demonstrated a significant extension in grain fill duration for the staygreen parents relative to Paragon, which mirrored their senescence phenotypes. However, in both 2016 and 2017 certain segregants demonstrated phenotypes indicative of sink limitation, and therefore potential indicating benefits of staygreen may be going unrealised.

Exome capture has been undertaken on the three staygreen parents, and Paragon, to identify EMS mutations for the development of molecular markers to begin to map associated region(s) through QTL analysis. Meanwhile 2017-18 field trials focus on testing the staygreen character under different stress condition such as low nitrogen regimes, climates, and soil types. These trials will be conducted by KWS in the UK, France, and Germany.

Keywords: Staygreen, wheat, senescence, NUE, stress-adaption, photoperiod-insensitive

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Different growth dynamics within the wheat spike

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European wheat production is stagnating while its demand is in constant evolution. Indeed, maintain or increase its yield in less favorable conditions is a major objective for the future. Grain yield is built during wheat development mainly through two components: the number of grains per square meter until anthesis and the thousand-grain weight during grain filling. Usually only the average grain weight is considered but grain weight may vary largely depending on the position along the spike and within a spikelet. As study was then carried out to explain why the individual weight grain is not homogenous along the spike.

Two different varieties were grown in a greenhouse at daily temperature of 22.6°C and a photoperiod of 16 hr, with water and nutrient solution supplied twice a day. From anthesis to harvest at least 13 samplings of eight spikes of main tillers were conducted for each variety. The ear was divided into three parts (basal, central and apical) in which the two proximal grains of two spikelets were dissected. The individual grains were then dried and weighed. A three-parameter logistic model was fitted to describe grains growth depending on their position on the spike.

At maturity, no significantly difference of dry matter was shown between the two proximal grains for both varieties. They were pooled in the following analyses. Grains in the central part of the spike were the heavier followed by the basal ones, the lighter ones being in the apical part. The logistic model fit well to the data. Differences in final grain weight are mainly explained by a difference of ovary's dry matter at anthesis and grain growth while the duration of grain filling is the same for each part.

This work brings new information to explain individual grain weight within the spike through different growth dynamics.

Keywords: wheat, spike, grains, dry matter, grain filling, distribution within spike, yield

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Seeding density effects on grain yield in bread wheat

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Seeding rate is considered as one of the cultivation practices that most affect crop grain yield and its components. It is based on planting a predetermined number of live seeds per square metre to achieve a specific plant density optimal for overall health and vigor of the plants. In order to study influence of seeding density on tillering potential and yield performances of bread wheat, a set comprised of 80 elite wheat varieties and 16 breeding lines contrasting for tillering was sown at locality Rimski Sancevi, Novi Sad, Serbia in 2016-17 growing season. A trial was set at randomised block design with three replications for each seeding density (300 and 550 viable seeds per square metre) and size of each plot was five square metres. During the season and after harvest the following traits were recorded: number of tillers, stem height, spike length, grain yield and thousand-kernel weight (TKW). The genetic diversity and population structure of the 96 accessions were assessed by 60 microsatellite markers distributed across all three wheat genomes. The presence of marker-trait associations (MTA) was analysed using mixed linear model. A total of 496 alleles were detected, with average PIC value of 0.613. Within analysed wheat accessions, three subpopulations were determined and the grouping was in accordance with their pedigree data. Analysis of variance showed statistically significant differences for grain yield between two seeding densities, as well as among detected subpopulations. Positive correlations were observed between grain yield and TKW, while tillering was negatively correlated with TKW. The 21 and 22 MTAs ($p < 0.01$) were obtained for trials with 300 and 550 seeds per square metre, respectively. However, only 12 MTAs were common for both. The presence of unique MTAs could indicate to specific genetic mechanisms involved in plant adaptation to different seeding densities in wheat, which should be further studied.

Keywords: sowing rate, *Triticum aestivum* L., tillering

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Stay green traits contributed to recent grain yield improvement in six-rowed winter barley in Serbia

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Stay green phenotype in small grain cereals is characterized by longer maintenance of green leaf area after anthesis, allowing extended photosynthesis and accumulation of assimilates. Moreover, stay-green traits are related to improved yield stability and adaptation, especially to water limited environments. Climate changes, reflected in the mean temperature increase, precipitation decrease and frequent drought stress occurrence at the end of grain filling period, have had major influence on cropping system in Serbia during past decades. Therefore, the objective of this paper was to examine changes in stay green traits related to grain yield improvement in winter barley cultivars that have been grown in Serbia during last four decades. Grain yield and stay green traits (maximum NDVI, thermal time to the midpoint of senescence and thermal time to senescence nearing conclusion of stay green) were determined in a historical series of 8 Serbian and 8 international cultivars from surrounding countries. The experiment was conducted during two growing seasons with two levels of nitrogen application (0 and 100 kg N ha-1). Results from this study showed that in Serbian and international set of cultivars grain yield was positively related to maximum NDVI and the thermal time to midpoint of senescence. Thermal time to senescence nearing conclusion of stay green was positively related with grain yield only in the international set of cultivars. Consistent increase per year of release in maximum NDVI and the time to midpoint of senescence were observed in both sets of cultivars under two levels of nitrogen fertilization, while correlation coefficients between the time to senescence nearing conclusion of stay green and year of release were not significant in either set. Therefore, our study suggests that further winter barley breeding progress should be more focused on increase of maximum NDVI and prolongation of period to midpoint of senescence.

Keywords: *Hordeum vulgare* L., NDVI, thermal time

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Breeding progress in yield and grain properties of winter wheat

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Constant grain yield increase of winter wheat represent one of the major breeding objectives worldwide, due to increase in human population and rapid decrease in the cropping area. Moreover, knowledge of the changes in grain traits associated with genetic gains in grain yield is important for future improvement of wheat breeding programs.

Therefore, the objective of this study was to analyze changes in grain yield and related grain traits in winter wheat cultivars grown in Serbia during the past 50 years. For this study, grain yield, 1,000-grain weight and test weight, were analyzed in historical set of 15 winter wheat cultivars. Trial was conducted during two growing seasons at the Experimental Field of the Institute of Field and Vegetable Crops, southeastern Europe.

Results from this study show that there was positive relation between grain yield and year of cultivars release. The average rate of increase of yield potential per year of release was 93 kg ha-1 year-1. In general, 1,000-grain weight varied between 35.8 and 47.0 g, while average values of test weight ranged from 75.1 to 84.5 kg hl-1. However, there was no significant changes in 1,000-grain and test weight in the last 50 years of wheat breeding at the Institute of Field and Vegetable Crops.

In conclusion, there was a significant yield progress during the examined period, however yield increase was not followed by changes in grain properties (1,000-grain and test weight).

Keywords: breeding, yield, grain, wheat

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SESSION IV. IMPROVING TOLERANCE TO BIOTIC AND ABIOTIC STRESS

Quality preservation in FHB attack due to antioxidant system

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Biotic stress leads to the formation of ROS which can harm the plants. Due to that plants have evolved an antioxidant defense system that includes non-enzymatic compounds, like ascorbate, glutathione, tocopherol, carotenoids, flavonoids and enzymes such as superoxide dismutase (SOD), catalase (CAT), guaiacol peroxidase (POD), ascorbate peroxidase (APX) and polyphenol oxidase (PPO). Six winter wheat varieties, representing commercial Croatian wheat classes with different intrinsic qualities, were subjected to Fusarium Head Blight (FHB) stress. Sampling was done at the 14th day of the FHB stress when the first symptoms occurred. Antioxidant properties of both treatments (control and FHB) were investigated to document the effect of FHB resistance to protein components (albumins, gliadins, ω-, α-, γ- gliadins, glutenins, HMW-GS and LMW-GS) which were detected by RP-HPLC method. The plants of all six varieties under test showed less or no symptoms of FHB till the 14th day of the FHB stress, when two wheat varieties progressively enlarged symptoms up to 90% till 26th dai (days after inoculation) and due to that they were characterized as FHB-susceptible. Very low FHB severity was obtained by 'Kraljica', 'Olimpija' and 'Vulkan' (20% symptoms at 26th dai), while 'Sana' showed moderate susceptibility (35%).

The decline in CAT activity was in correlation with the increase of H2O2 in more resistant varieties ('Olimpija' and 'Vulkan'). The activity of APX increased with the appearance of symptoms in 'Kraljica', 'Golubica', 'Bc Anica', while the activity of APX declined in 'Olimpija' in FHB treatment in comparison to control. However, there was significant increase in FHB treatment in comparison to control, in respect to phenols, POD and H2O2 for all varieties, except for POD in 'Sana' and H2O2 in 'Golubica'. Varieties 'Golubica' and 'Vulkan' exhibited higher PPO activity in FHB treatment in comparison to control despite the presence of enlarged phenol content in FHB treatment in all varieties. The accumulation of MDA content was higher in two susceptible varieties than in tolerant varieties, with one exception.

After harvest, gliadin level in FHB treatment in comparison to control increased in four varieties ('Kraljica', 'Sana', 'Golubica', 'Anica'). There was a significant effect of FHB on percentage of glutenins showing a respective reduction in comparison to control of 5%, 20% and 16% of 'Kraljica', 'Golubica' and' Bc Anica', respectively, while HMW subunits decreased in those three varieties (9.93, 14.61 and 20.33%, respectively) including 'Sana' (8.79%). LMW subunits were not largely affected as HMW, but 'Olimpija', 'Golubica' and 'Bc Anica' showed statistically significant decrease.

It is interesting that in both treatments the most susceptible variety had the lowest amount of phenol content and PPO activity among other varieties. Due to the role of polyphenols as secondary plant metabolites that play a role in the protection of plants against pathogen, they were higher in genotypes with better-preserved protein quality. The protein components of 'Vulkan' and Olimpija' were untouched which could be due to high PPO activity which could participate in detoxification of phenolic acids induced by pathogen attack.

Keywords: FHB, wheat, protein, antioxidant activity

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PIV-34 - Hanane AADELA - Agrobacterium-mediated Transformation of Mature Embryo and immature Tissues of Bread Wheat (*Triticum aestivum* L.) Genotypes

Agrobacterium-mediated Transformation of Mature Embryo and immature Tissues of Bread Wheat (*Triticum aestivum* L.) Genotypes

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Although significant progress has been made on Agrobacterium-mediated wheat transformation, the current methodologies using immature embryos as recipient tissues, is labor intensive, time consuming and expensive. The use mature embryo (ME) as explants is increasingly recognized as a best method for developing regenerable cell lines in wheat transformation. Therefore, we have developed an Agrobacterium-based transformation protocol using mature embryos, adjusting other factors that can influence the yield of transformed plants. The experiment was focused on acetosyringone concentrations, genotypes and different kinds of mature embryos used as explants source for transformation studies. The *A. tumefaciens* strain EHA101 and pTF101.1 plasmid carrying HVA1 gene and bar-selectable marker gene were used. The intact mature embryos and half mature embryos produced a highest number of putative transgenic plantlets on selection medium. However, no plantlets were obtained from fragmented mature embryos. ‘Amal’ and ‘Rajae’ regenerated highest number of putative transgenic plants and 200 M acetosyringone was found to be better concentration for transformation. A total of 47 transgenic plants were selected. 11 plantlets showed resistance to leaf painting. The molecular analysis revealed 1% and 0.66% of T0 regenerated plantlets were successfully transformed and carried the HVA1 gene for the genotypes ‘Amal’ and ‘Rajae’, respectively. Additional analysis showed that the transgene was stably inherited from the T1 generation.

Related publication:
Agrobacterium-mediated Transformation of Mature Embryo Tissues of Bread Wheat (*Triticum aestivum* L.) Genotypes. Published in Cereal Research Communications CRC-D-17-00013R4-Agrobacterium-mediated transformation of immature embryo of bread wheat (*Triticum aestivum* L.) genotypes. Published in biologia

Keywords: bread wheat; mature embryo; explants; *Agrobacterium tumefaciens*; plasmid pTF101.1; HVA1; acetosyringone; genetic transformation

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How may warm spells during winter influence the plant development of cereals?

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The climatic conditions during winter influence multiple processes in cereals. In addition to determining the winter survival and the vegetative-generative phase transitions of the apex, these conditions significantly affect the later plant development and thus yield components. Due to the climate change, the occurrence of unusual weather conditions and anomalies increases which can include shorter or longer warm spells during winter. We wanted to study the effect of a warm temperature period on seedlings with various degrees of vernalization requirement saturation. For this purpose 19 wheat cultivars of various origins were treated with a two-week period of warm temperature (25 °C) right after the end of 30-, 45- and 60-day low temperature vernalization in the juvenile stage.

Based on the results, the two weeks of 25 °C at the beginning of the plant development significantly modified both plant phenology and morphology, the direction of this effect, however was strongly dependent on the saturation level of the vernalization requirement and on the genotypes. After a 30-day vernalization, the plant development was delayed significantly by the warm temperature averaged over the 19 wheat cultivars, which was more pronounced at the later stages. As the saturation of the vernalization requirement increased, there was a dramatic change in the effect of warm temperature. After a 45-day vernalization the difference between the control and the 25 °C-treated plants were minimal, especially at the later stages, while after a 60-day vernalization treatment the 25 °C temperature fastened the plant development significantly. The short period of warm temperature in juvenile stage also influenced significantly the plant height, the final leaf number and the number of reproductive tillers.

In conclusion, under inductive photoperiods the effects of a short, warm ambient temperature spell on the juvenile plants with winter growth habit depends on to what degree their vernalization requirements are saturated. If it is not saturated then the plant development becomes delayed. If it is saturated then the warm temperature spell hastens the plant development, the extent of which is strongly genotype dependent.

This research was funded by the following grants: EU-FP7 ADAPTAWHEAT, and GINOP-2.3.2-15-2016-00029

Keywords: wheat, vernalization, ambient temperature, plant development

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Investigations on the effects of drought stress on the morphology, physiology and yield components of durum wheat in Hungary

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The aim of the work was to identify morphological, physiological, biochemical and yield biological traits exhibiting a close relationship with higher yields even when water supplies are less than satisfactory. For this purpose a total of 188 spring durum wheat genotypes with diverse genetic backgrounds and eight near-isogenic lines of spring durum wheat were sown in field experiments under irrigated and rainfed conditions or under a rain shelter in Martonvásár between 2011 and 2014 in order to investigate which plant traits contribute to an increase in drought tolerance. In the course of the field experiment, the individual genotypes were sown on the same experimental field each year so as to reduce the effect of the environment (soil).

The results proved that the year and the treatment had the greatest effect on the traits tested, even in the case of parameters that have a high heritability (h2) index. The only exception was the thousand-kernel weight, which was shown by the three-year data to have great stability even when exposed to drought stress. It was demonstrated that only plants with a larger number of fertile tillers and with flag-leaves having a longer period of photosynthetic activity were able to produce higher yields even with poor water supplies. The significant positive correlation found between the SPAD and NDVI values recorded in the early stage of waxy ripeness (Z83) and the yield showed that some genotypes have delayed senescence in this phenophase under drought conditions, which is manifested in the form of higher yields. The analyses led to the identification of a cultivar (‘Cham-1’) that was able to maintain stable photosynthetic activity during drought stress in all the years, thus giving better performance.

Association mapping was used to identify loci responsible for drought tolerance. With the help of the SNP map characterising the 188 cultivars, the population structure of the association panel was analysed, and QTL regions linked with yield, heading date and morphological or physiological traits were identified for different water supply levels. Numerous marker-group regions were found to be significant in specific environments, as a function of year or treatment. In the course of the work, two chromosome regions were identified where QTLs for chlorophyll content and heading date were linked (1B, 5A), and in the case of the 5A chromosome the yield QTL was also linked with the identified region.

The results suggest that the phenotyping of morphological, physiological, biochemical and yield biological traits could be an efficient way of identifying genotypes with better resistance to water deficit. The most important criteria for success are a clearly defined trait that can be objectively measured, a satisfactory statistical model and successful validation.

Funding from the EU FP7-244374 DROPS and GINOP-2.3.2-15-2016-00029 projects

Keywords: durum, drought stress, phenology, physiology

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PIV-37 - Krisztina BALLA - Investigation of heat stress tolerance on yield properties in cereals

Investigation of heat stress tolerance on yield properties in cereals

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Heat stress is becoming increasingly frequent and often affect wheat plants at any time during vegetative growth and especially around heading in Hungary. Our studies focused on that the effect of heat stress strongly depends on the timing and duration of the heat period during the growing season and the damage caused by heat is greatly influenced by the developmental stage of the plants. Therefore, heat stress experiments were undertaken to determine the effects of heat stress on the physiological processes, as well as yield components in the stage of booting (Zadoks-49) and emergence of inflorescence completed (Zadoks-59). 100 winter wheat varieties of different origins were involved in phytotron where the treatments lasted for 5 (H5), 10 (H10) and 15 (H15) days under control conditions. The plants were exposed to heat stress of 36oC/20oC for 8 hours. The specific aim of this research project was to identify various heat tolerance sources and to determine the genetic background of the responses given to heat stress.

It was found that the shortest duration of heat stress (H5) had the lowest impact on the examined parameters, while the longest treatment (H15) resulted in the most negative impact on the morphological and yield parameters in both examined phenophases. On the average of the varieties, the plant height (PH) and the last internode length (LIL) were reduced only due to the heat stress treatment applied in Zadoks-49. The heat stress resulted in drastic significant reduction in the grain number (GN), grain yield (GY), harvest index (HI), biomass (B) and the grain number of one spikelet. But the thousand kernel weight (TKW) was increased due to the drastic reduction in the number of grains under heat stress for 5 and 10 days. In Zadoks-59, H5, H10 and H15 treatments caused a significant reduction in the number of productive tillers, LIL, HI, B, GN and GY. The high temperature treatments, however, had no significant effect on the PH, the length of the ear, the grain number of one spikelet and the grain number of the main spike too. TKW was declined only by the H15 treatment significantly. The photosynthetic activity was dropped in response to heat stress in both examined phenophases. The stomatal conductance was declined due to heat stress resulted in lower CO2 binding capacity. The net assimilation (Pn) was reduced by more than 50% due to heat stress treatments (H5) in both stages. The plants tried to survive the heat stress by increasing the transpiration level. The chlorophyll content was changed to the most negative extent when the wheat varieties were exposed to heat stress for 15 days.

The best Pn value was measured in the variety Aura (at Zadoks-49) and in Altay-2000 (Zadoks-59) under the applied longest heat stress. The best GY value was received in the variety Valoris (at Zadoks-49) and Spada (at Zadoks-59) when exposed to H15 treatment.

As a consequence of the changing climate and the higher occurrence of extreme temperatures it is important to breed new wheat varieties with high yield quantity and good quality even under extreme weather conditions. The impact of this analyse was to identify different heat tolerance sources and to produce genetic populations, in which the high heat tolerance properties are incorporated.

This project was supported by a grant from the National Research, Development and Innovation Office (NKFIH-119801) and the János Bolyai Research Scholarship of the HAS.

Keywords: heat stress, yield properties, photosynthetic activity, wheat

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Comparative analysis of stem rust resistance in winter wheat genotypes at Hungarian and East African locations

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Stem rust, caused by the fungus *Puccinia graminis* f. sp. *tritici* Eriks. & E. Henn., is an impending disease for wheat production worldwide hence breeding for stem rust resistance is a very important challenge of wheat breeders all around the world. Although the disease has been under effective control in Hungary through the use of host resistance, there are needs for new sources of resistance because of the threatening new pathogen races in Africa and Middle East. The main objective of this work was to compare the stem rust resistance of winter wheat genotypes under different pathogen pressure in Hungary and Kenya and to identify those genotypes which has acceptable resistance under both conditions.

The stem rust resistance of the 161 collected genotypes were scored in adult plant stage by the modified Cobb scale, evaluating the severity and the host response. From the phenotypical data average coefficient of infection (ACI) values were calculated. The field experiments were set up at two different locations, in the stem rust screening nursery of the Kenya Agriculture and Livestock Research Organization, Njoro in 2017 and in the artificially inoculated stem rust nursery of Agricultural Institute, Centre for Agricultural Research, Martonvásár in 2016 and 2017.

We investigated the frequency of the *Sr1A.1R, Sr2, Sr15, Sr31, Sr38* stem rust resistance genes and the *Lr34* leaf rust resistant gene in the collected material, using the gene specific SSR Xscm9 (for *Sr1A.1R* and *Sr31*), csSr2 CAPS (for *Sr2*), STS638 (for *Sr15*), VENTRIUP-LN2 (for *Sr38*) and STS csLV34 (for *Lr34*) molecular markers. Statistical analysis of the data was performed to determine the connection between the investigated Sr genes and the phenotypic responses of the genotypes.

Among the 161 genotypes 90 (55.9%) carried the *Sr38*, 36 (22.4%) the *Sr31*, 34 (21.1%) the *Lr34*, 33 (20.5%) the *Sr15*, and 4 (2.5%) carried the *Sr2*, respectively. The *Sr1A.1R* were detected in two (1.2%) genotypes. About one third of the genotypes had very good resistance against both the Hungarian and African pathotypes of the stem rust pathogen. From the results it can be concluded that the resistance of the genotypes against the Hungarian races of *Puccinia graminis* f.sp. *tritici* is primarily due to the presence of the *Sr38* or *Sr31* resistance genes meanwhile in the case of the Ug99 lineage the connection between the investigated *Sr* genes and the phenotypic response is not significantly provable.

We would like to thank the excellent help of Dr. Ruth Wanyera and Dr. Sridhar Bhavani for managing the stem rust resistance field tests in Kenya Agriculture and Livestock Research Organization. This research was funded by the following grant: GINOP-2.3.2-15-2016-00029

Keywords: stem rust, resistance breeding, molecular markers, wheat (*T. aestivum* L.)

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PIV-39 - Valérie CADOT - *Tilletia caries*, causal agent of Common Bunt: study of the most prevalent virulences in order to develop a resistance test for the registration to the National List for common wheat in Organic Farming

Tilletia caries, causal agent of Common Bunt: study of the most prevalent virulences in order to develop a resistance test for the registration to the National List for common wheat in Organic Farming

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Common bunt, mainly caused by *Tilletia caries* and *T. foetida*, with a high capacity of dissemination has been a re-emerging seed-borne cereal disease since 2007. In infected ears, bunt balls cause important yield reductions of up to 80% and a deterioration in seed health quality. Most commonly cultivated wheat cultivars are susceptible, due a lack of selection tools for resistance but also because chemical seed treatments can be used to control this problem in conventional farming. In organic farming, only a few seed treatments are available which are not always effective, thus genetic resistance is an important trait to consider for registration to the French List.

The first aim of program ABBLE was to study the variability in populations of common bunt and to identify the main virulences present in France. Samples were collected in 2015 from the main production areas and the identification of *Tilletia* species was performed by GEVES. All 20 collected strains were identified as *T. caries*. For each strain, the characterization of virulence was done on a range of 15 Host Differentials from Metzger. After 7 months of growth, symptom expression (bunt ears) was quantified on each host differential showing that the most frequent virulences were Bt-2, Bt-7 and Bt-15.

The second aim was to develop a miniaturized resistance test to common bunt, by comparison with resistance test at adult stage in field. A test was carried out in controlled conditions with detection of *Tilletia caries* by qPCR at an early stage (2 leaves) and compared with symptom expression for similar varieties at milky stage. Currently, the method allows the differentiation of susceptible and resistant varieties. The qPCR protocol was tested on the range of 15 host differentials infected with a strain representing the most prevalent virulences in France. The same strain has also been inoculated to 10 cultivars mainly used in organic farming. The qPCR method will be validated after comparison of resistance classification between the number of positive plantlets detected by qPCR and the percentage of infected ears observed in field for these same varieties.

Evaluation of cultivar resistance mainly used in organic farming will improve understanding of the current resistance against the main virulences present in France. The development of an early, rapid and reliable resistant test will provide a new tool for screening resistant varieties for breeders and for registration to the National List.

Keywords: *Tilletia caries*, virulence, early resistance test, qPCR, Organic farming, National List

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Flowering pathway genes regulating *Fusarium* head blight resistance in wheat

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Fusarium head blight (FHB) caused by *Fusarium graminearum* and other spp. of genus *Fusarium*, is a most devastating disease of wheat affecting most growing areas in Canada. Infection with Fusarium leads to shriveled seed or tombstones, which are white or pink in appearance and known as *Fusarium* Damaged Kernels (FDK). The high proportion of FDK in a commercial wheat crop results in severe yield and quality losses. The fungus also produces a toxin known as deoxynivalenol (DON) in the infected grain which is a major food safety issue, especially in the export market. The use of resistance cultivars offers the most effective approach to manage FHB epidemics. Therefore, a number of studies have been conducted to identify FHB resistance QTL in wheat around the globe, however, molecular mechanisms underlying FHB resistance have not been fully known yet. In this study, a doubled haploid mapping population segregating for flowering pathway genes and FHB resistance have been screened for a number of flowering related traits and FHB followed by SNP genotyping and QTL analysis. The results of present study will be discussed.

Keywords: *Fusarium* head blight, wheat

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PIV-41 - Aurore BERAL - Is wheat individual grain characteristics variance a source of genetic variability for water stress tolerance?

Is wheat individual grain characteristics variance a source of genetic variability for water stress tolerance?

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Agronomic performance of wheat genotypes is usually evaluated by studying the mean value of pertinent agronomic variables. In particular, grain yield is analyzed through its different average numeric components (number of plants per m-2, number of spikes per plant, number of spikelets per spike, number of grains per spike, number of grains per m-2, individual grain weight). Nevertheless, the variance of these components has not be so far fully explored. Moreover, number of grains per m-2 is often considered as the most important component impacting grain yield. Yet it has been observed that the bigger the number of grains is, the more the number of small grains is, especially those in apical spikelets or in apical position within a spikelet. The variance associated to the grain size and individual grain composition (nitrogen concentration) is linked to both tillering and spike structure. Its impact on agronomic performance, especially in stressful environments (abiotic stress) is not much known.

The aim of this study is to analyze the genetic and ecophysiological determinism of wheat individual grain characteristics variability and evaluate its impacts on agronomic performance in order to quantify the actual genetic variance potential. To this aim, several issues have to be investigated: (i) understand precisely the structure of the variance associated to individual grain characteristics and (ii) understand causal processes of this variance, especially spatial structure of the spike and spikelets.

The present work is based on a field experiment where 228 genotypes were grown under optimal conditions or under a severe water stress. All yield components were recorded. Out of this panel, four genotypes were selected for in depth analysis of grain traits variance structure. For these genotypes, 10 spikes were randomly chosen in each plot and were individually dissected in order to record the exact position of each grain within the spike structure. Grains originating from 2 spikes among these 10 spikes, nitrogen concentrations per grain were also measured.

Our results show that (i) individual grain weight is strongly affected both by its position within the spike as well as by mean spike characteristics and (ii) variance components are under both genetic and environmental control. These results suggest that studying variable components of grain traits can help defining typologies of wheat genotypes differing by their strategies to set grain number (and thus potential yield) at anthesis. Further studies will be needed in order to test whether these strategies have differential effects on wheat tolerance to abiotic stresses during grain filling.

Keywords: Variance, grain weight, water stress, wheat

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PIV-42 - Barbara STEINER - Improving *Fusarium* head blight resistance in durum wheat

Improving *Fusarium* head blight resistance in durum wheat

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Triticum durum is particularly susceptible to infection with pathogenic fungi of the Fusarium genera, which cause Fusarium head blight (FHB), one of the most destructive diseases of wheat worldwide. FHB leads to significant yield losses but the major issue associated with FHB is the contamination of the crop with mycotoxins. Notably, durum wheat is almost exclusively used for human consumption; consequently, the risk of toxin-contaminated grain entering the food chain is especially precarious.

The development of resistant varieties is the most effective approach for managing FHB, but resistance breeding in durum wheat has been hampered by the limited genetic variation in its gene pool and the difficulties in efficiently combining the numerous often small effect resistance conferring alleles in the same breeding line.

To increase genetic diversity for FHB resistance in durum wheat we introgressed resistance alleles from wild and cultivated relatives (*T. aestivum*, *T. dicoccoides*, *T. dicoccum*) and developed a large population of several bi- and multi-parental crosses. The population was phenotyped over several years resulting in a broad variation for FHB resistance including many moderately resistant experimental lines. Markers indicative for the major FHB resistance QTL Fhb1, identified in bread wheat, were clearly associated with FHB severity, confirming its successful implementation into durum wheat.

In addition, we investigated the genetic architecture of FHB resistance in the elite durum wheat gene pool and compared the accuracy and response to selection when using phenotypic and genomic selection strategies. A collection of 228 genotyped durum wheat cultivars was phenotyped for FHB resistance in three years and used for genome-wide association mapping. FHB resistance was strongly influenced by the semi-dwarfing gene RhtB1 as well as one QTL on chromosome 3B, which explained 10% of the phenotypic variance. Interestingly, this QTL was positioned in the same chromosomal interval as reported for Fhb1, but further genomic analysis suggests different genetic control.

Integrating the 3B QTL as fixed effect into genomic prediction models increased the prediction accuracy by 10%; however this extension had merely a minor effect on the actual response to selection. Using a selection index approach, we found that genomic selection had a 50% higher response to selection for FHB resistance than phenotypic selection in two-stage selection, without altering the population mean for plant height i.e. selecting undesirable tall plants. Furthermore, phenotyping intensity could be reduced by testing only 50% of the most promising lines. Genomic selection among all lines in the population in this scenario gave the same response to selection as testing all lines in the disease nursery with subsequent phenotypic selection.

Keywords: *Durum wheat*, *Fusarium* head blight

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PIV-43 - Pawel CZEMBOR - Genome wide association study of seedling leaf rust resistance in European winter wheat cultivars

Genome wide association study of seedling leaf rust resistance in European winter wheat cultivars

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Leaf rust caused by the biotrophic fungus *Puccinia triticina* Eriks. (Pt) is one of the most important fungal disease affecting wheat worldwide. Genetic resistance is the most economical, effective and ecologically sustainable method of controlling the disease. Almost 80 leaf rust resistance genes have been characterized. Efficient utilization of genetic resistance relies on an appropriate knowledge of the leaf rust resistance genes and their effectiveness in different environments. We applied a genome wide association study (GWAS) approach to identify resistance genes in 143 winter wheat cultivars recently registered in Poland, Germany, France, Switzerland, Denmark and United Kingdom. Identified significant marker-trait associations (MTAs) were verified on a panel of 36 Thatcher near-isogenic lines containing different Lr genes (Tc-LrNILs). Wheat cultivars and Tc-LrNILs were genotyped on DArTseq platform and phenotyped with 18 single *Pt* isolates at seedling stage. After filtering, out of 40,035 SNP only 2,983 were used in GWAS using restricted maximum likelihood (REML) approach and information on structure of the analyzed wheat set was included. We identified four resistance genes *Lr3bg* (6B), *Lr9* (6B), *Lr24* (3D) and *Lr26* (translocation T1BL.1RS) and several leaf rust resistance loci not present in Tc-LrNILs set on chromosomes 1B, 2A, 3B, 6B, 7A and 7D. Only one-third of the cultivars contained at least one of the identified resistance genes. For some resistant cultivars we were not able to identify Lr genes that might be attributed to the low resolution of *Pt* isolates used in the study.

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Keywords: Brown rust, DArTseq markers, gene postulation, resistance, wheat cultivars

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Resistance of European winter wheat cultivars to *Zymoseptoria tritici* isolate IPO92006

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Septoria tritici blotch (STB) of wheat (*Triticum aestivum*), caused by the fungal pathogen *Mycosphaerella graminicola* (anamorph: *Zymoseptoria tritici*, syn. *Septoria tritici*), is present in most wheat-growing areas worldwide. Host resistance is the most economical and safest method of controlling the disease and information on resistance loci is crucial for effective breeding for resistance programs. In the study we used a set of 83 wheat cultivars registered in the Descriptive List of Agricultural Plant Varieties (COBORU 2012), 110 cultivars from other European countries and 25 cultivars/lines with identified STB resistance loci. The wheat genotypes were tested on adult plant stage under polytunnel conditions with watering system. Fully expanded flag leaves were sprayed with spore suspension of IPO92006 *Z. tritici* isolate. After incubation period, the percentage leaf area covered by necrosis (NEC) and covered by pycnidia (PYC) were measured on flag leaf of each wheat cultivar/line that were used in agglomerative hierarchical clustering (AHC) analysis with UPGA algorithm (unweighted pair-group average). Three groups of wheat cultivars/lines were identified and the largest group comprised 170 resistant genotypes with NEC 12.4%─82.1% and PYC 1.1%─53.5%. Within this group, set of 43 highly resistant wheat cultivars were identified (NEC 1.1%─41.8%, PYC 1.1%─31.9%): TE9111, Capone, Lear, Intro, Bombus, Fermi, Heros, KWS Erasmus, Oxal, Tabasco, Elixer, Dacanto, RGT Kilimanjaro, Florett, Sukces, Reciproc, Frument, Desamo, Edgar, Colonia, RGT Djoko, Addict, Forum, Famulus, Pionier, Amifor, Mandub, Memory, Terroir, Julius, Joker, Solitar, Kranich, Grapeli, KWS Dacanto, Eron, Marcopolo and Celebration. In addition in the same subgroup two genotypes with identified resistance loci were classified: Florett (QTL-3B, QTL-6D, Stb6 and Stb15) and TE9111 (Stb11, Stb6 and Stb7). This may suggest that resistance to STB in European cultivars is contributed mainly by quantitative loci and those with main effects. Presented work (phenotyping data) is a part of larger project aiming at identification of resistance genes to *Septoria tritici* blotch in winter wheat and will be used in near future in association mapping approach.

This research was supported financially by the Polish Ministry of Agriculture and Rural Development, program Fundamental Research for Biological Progress in Crop Production (years 2014-2020).

Keywords: Resistance, *Septoria tritici* blotch, wheat

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PIV-45 - Éva SZAKÁCS - Development and characterization of a new recombinant 1RS.1BL translocation line carrying leaf and stripe rust resistance from perennial rye (*Secale cereanum*)

Development and characterization of a new recombinant 1RS.1BL translocation line carrying leaf and stripe rust resistance from perennial rye (*Secale cereanum*)

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The most intensively used source of rye chromatin in bread wheat has been the 1RS chromosome arm in the form of 1RS.1BL translocation because it provided a source of resistance genes against leaf rust (*Lr26*), stem rust (*Sr31*), stripe or yellow rust (*Yr9*) and powdery mildew (*Pm8*). Nowadays, the proportion of wheat varieties carrying the 1RS.1BL translocation is decreasing due to the fact that resistance genes *Lr26*, *Pm8* and *Yr9* are no longer effective against new biotypes of diseases. Virulence to the *Sr31* resistance gene was first reported from Uganda in 2010. Appearance of the *Sr31*-virulent pathotype in countries where wheat production is based on cultivars carrying the 1RS.1BL chromosome translocation can cause serious problems for agriculture. The genetic vulnerability of 1RS.1BL cultivars is the consequence of the genetic uniformity of the 'Petkus'-derived 1RS arm. Therefore, it is important to involve new rye breeding materials in resistance breeding programmes in order to improve the genetic base of cultivated wheat.

In order to widen the genetic diversity of this rye chromosome arm, the winter wheat line Martonvásári 9 kr1 (Mv9kr1) was crossed with a Hungarian *S. cereanum* cultivar (Kriszta). The F1 hybrids were propagated in tissue culture, the regenerated plants were backcrossed with the parental wheat genotype Mv9kr1 and selfed. Progenies were propagated in the Martonvásár nursery. Selection of disease resistant lines carrying recombinant 1RS.1BL started in 2012. A wheat line, designated as line 179, was grown together with the highly susceptible parental wheat line Mv9kr1 genotypes in a pesticide-free nursery and in the Breeders nursery in three consecutive seasons (2013-2014, 2014-2015 and 2015-2016). Mv9kr1 suffered severe attack by stripe rust, while the leaves of the line '179' did not show any symptoms. Disease resistance tests under greenhouse conditions by artificial inoculations with *Puccinia triticina* isolates proved that this line was also resistant to leaf rust. Furthermore, this line has elevated arabinoxylan content. Molecular (SSR) marker analyses showed that the 1RS arm present in the line '179' was different from that of Petkus rye. In order to detect altered DNA sequences in the recombinant 1RS chromosome arm, a detailed analysis based on DArTseq and SNP analysis is in progress.

This work was supported by the National Research, Development and Innovation Office – NKFIH, K 119387

Keywords: 1RS.1BL translocation, disease resistance, leaf rust, stripe rust, perennial rye (*Secale cereanum*)

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PIV-46 - Lidia KOWALSKA - Improvement of *Parastagonospora nodorum* blotch resistance in winter triticale by using the in vitro somaclonal and androgenic approaches.

Improvement of *Parastagonospora nodorum* blotch resistance in winter triticale by using the in vitro somaclonal and androgenic approaches.

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Leaf and glume blotch caused by *Parastagonospora nodorum* is a widespread triticale disease. It can cause severe losses in grain yield wherever triticale is grown. Resistance breeding is the most acceptable way to control the disease. Plant breeders and pathologists have worked intensively to incorporate resistance to the pathogen in new cultivars. Conventional methods of breeding for resistance can be supported by using the biotechnological ones, i.e. somatic embryogenesis and androgenesis. Therefore, an effort was undertaken to study and to compare variation in *P. nodorum* resistance among winter triticale somaclones, dihaploids and conventional varieties.
For the purpose we have evaluated under controlled environment conditions eight triticale commercial cultivars, twenty one double haploids and seventeen somaclonal lines produced from cultivars varying in resistance to *P. nodorum*. The first seedling leaves were inoculated with a mixture of *P. nodorum* isolates. Spore concentrations were adjusted to 5 x 106 of viable spores per one millilitre. After 10 days of incubation, the disease severity on seedling leaves was rated on a scale, where 100% – susceptible, 0 – resistant. The differences between seedling leaves for all components were statistically significant. Most of the somaclonal and dihaploid lines produced from commercial triticale cultivars showed significantly improved resistance to the pathogen in question. However, resistant parental cultivars produced primarily more resistant somaclones and dihaploids. Higher resistance to *P. nodorum* was observed more often on leaves of somaclonal lines then on dihaploid ones. On average disease severity reached 10% on leaves of somaclones and 12% on leaves of dihaploids. Some of genotypes were showing low leaf infection, e.g. dihaploid DH63 received from F1 plants of Borwo x Tomko. Similarly, lower infection level was observed on a somaclone SE64 produced from F1 plant of Borwo x Cyrkon. In conclusion, we have demonstrated that somaclonal variation might be used as an additional source of triticale resistance to the pathogen and it could be recommended to use in commercial breeding programs. The reported results broaden our knowledge about triticale resistance to *P. nodorum* and bring closer the use of biotechnological methods in breeding for disease resistance of the small grain species.

Keywords: *Parastagonospora nodorum*, resistance, somaclonal, androgenic variation, triticale, breeding

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Occurrence of *Parastagonospora nodorum* blotch, *Parastagonospora avenae* blotch and *Zymosetoria tritici* blotch on wheat and triticale in Poland

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Septoria diseases belong to the most devastating diseases in central Europe. Common casual agents causing septoria leaf spot in Poland are *Phaeosphaeria nodourm* (E. Müller) (anamorph: *Parastagonospora nodourm* (Berk.)), *Phaeosphaeria avenaria* (G. F. Weber) (anamorph: *Parastagonospora avenae* (A. B. Frank), *Mycosphaerella graminicola* (Fuckel) (anamorph: *Zymoseptoria tritici* (Desm.). Yield losses due to *P. nodorum* wheat infection can reach up to 31%, while *Z. tritici* can cause even 50% yield reduction. *P. avenae* is host-specific to oats, however recently the widening of *P. avenae* host range was observed.

Breeding for resistance is one of key elements of integrated plant protection. It offers the possibility to improve the resistance level to septoria disease complex through phenotyping and selection of resistant cereal types. The aim of this paper is to assess wheat and triticale varieties according to their resistance to septoria leaf spot caused by *P. nodourm*, *P. avenae*, *Z. tritici* and glume blotch caused by *P. nodorum*. Field experiments were set up at height locations in Poland to determine the natural infection of small grain cereal species by the above mentioned pathogens. The so called "septoria disease nursery" in each location consisted of 10 - winter wheat cvs., 10 - winter triticale cvs., 6 - spring wheat cvs. and 6 - spring triticale cvs. After natural infection, disease symptoms development rating was made. The 9-digit scale was used in assessment of disease severity on leaves and glumes. Affected leaves and glumes were collected and mature pycnidia were analysed under binocular 10-45X and inverted microscope at 200X magnification. The length and width of pycnidiospores were measured and plated on solid media to identify pathogen species by using a microscope camera. Dimensions (length by width) of conidia ranged on average for: *P. avenae* f. sp. *tritici* (25-45 x 3-4 µm), *P. nodorum* (15-32 x 2-4 µm) and *Z. tritici* (35-98 x 1-3 µm). The incidence frequency and disease severity on leaves were determined for each species of pathogens. Our investigation has shown that *Z. tritici* was the predominant pathogen of wheat, while *P. nodorum* was the most often encountered on triticale. Our examination revealed that presently *P. avenae* is quite common on triticale and in certain geographical regions it appeared also on wheat. Sources of resistance to examined pathogens among wheat and triticale were identified.

Keywords: *Parastagonospora*, *Zymoseptoria*, resistance, wheat, triticale, breeding

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PIV-48 - Faiza BOUSSORA - Delaying in the spike development and reduction in spikelet primordia numbers decrease the yield in salt-stressed Tunisian barley

Delaying in the spike development and reduction in spikelet primordia numbers decrease the yield in salt-stressed Tunisian barley

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The big challenges which we face in the twenty-first century are producing sufficient food to meet demands of the increasing global population. Abiotic stresses caused by climate change, such as salinity, drought, low or high temperatures, flooding and frost are predicted to become more severe and widespread. The losses of crop productions due to salinity and drought stresses have increased progressively over the last decades. Common cereal members such as wheat (*Triticum* spp.), maize (*Zea mays* L.), rice (*Oryza sativa* L.) and barley (*Hordeum vulgare* L.) are the main important members of most worldwide nations in food and feed system. Barley is one of the oldest cereal crops known to be cultivated since about 10,000 years. It has a natural tolerance to salinity, drought, and fungal diseases, thus making it a model plant in stress biology research. We aim in this study to understand the effect of salinity stress on the spike developmental stages and the final grain yield of five Tunisian barley cultivars. Experiments were performed at IPK-Gatersleben, Germany. Seeds were sown under controlled condition in green house (LD condition, 16/8 h day/night and ~20/~16°C day/night). A saline treatment with 10gNaCl/l was imposed starting 7 days after sowing and was compared to a nonsaline control treatment. Results obtained show that, in addition to its impact on plant growth, salt has a severe and marked effect on spike development and final yield. It caused delaying in inflorescence developmental stages and decreasing in floral units numbers. Anthesis and flowering also occurred later in treated plants compared with control plants. The measurement of ten main-spike components indicated that salt negatively affected number of spikelets primordia/spike, number of grains/spike, spike length and thousand-grain weight, as well as number of tellers, number of leaves and plant height. Cultivars Ardhaoui and Rihane showed to have faster spike developmental stages and more yield than the other three under salt stress conditions.

Keywords: Tunisian barley, salinity, spike development, yield, growth traits

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Examination of the resistance of barley genotypes to *Pyrenophora teres f. teres* in the seedling and adult stage

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One of the aims of the breeding programs is to elevate abiotic and biotic stress resistance, because this is an essential point of adaptation and sustainable agriculture. Therefore, the development and cultivation of disease resistant/tolerant cultivars is of great importance and the deployment of resistant cultivars is the most economic and eco-friendly method to control plant diseases.

Pyrenophora teres Drechsler (PTT) [anamorph *Drechslera teres* (Sacc.) Shoem.] has become an economically important and major fungal pathogen of barley worldwide. The susceptibility of current cultivars and trends in environmental conditions could contribute to the pathogen’s increased importance. In Hungary, *P. teres* had become a very harmful causal agent of spring barley by the 1980’s, and now is considered to be a major pathogen in both spring and winter barleys. Due to the regional and global diversity in fungal virulence it is necessary to know and breed against the local fungal pathotypes. The efficiency of net blotch resistance genes depends on the developmental stage of barley and the fungal pathotypes present in the pathogen’s local population. Screening a wide genetic basis of barley varieties provides a good opportunity to find a resistance or tolerant genotypes which can be the parents of the new cultivars.

Therefore, our aims is to characterise the resistance of the Martonvásár barley collection against the pathogen in seedling and adult plants.

260 barley genotypes were tested under greenhouse and field conditions. The young barley genotypes were inoculated under greenhouse conditions with mycelium suspension of *Pyrenophora teres f. teres* isolate (H-502/1 derived from Hungary).

The barley genotypes were sown in small plots in autumn 2016 in two rows (1.5 m long with 15 cm in-row gap) and two replicates, arranged in randomised blocks. Spreading net blotch-infected straw from previous season. The area under the disease progress curve (AUDPC) was calculated from the lesion types (Tekauz scale) and infected area at various times.

The average seedling infection (AUDPC) was 13.26, ranged from 0,02 of Flagship to 91 of Hundred. In the case of adult resistance the level of infected leaves were also scored. The average AUDPC was 4495 ranged from 1642 of Patriciana to 7443 of Adagio. There were not significant difference among the infection of sensitive control and the 39% of tested varieties. The 27% of the cultivars proved to be less sensitive then the control and further 27% of studied genotypes were moderately tolerant ($p=0.01$). The correlation between the seedling and adult resistance was 0.32, which indicate the quite independent background of the two types of resistance. The majority of tested varieties were more sensitive in adult stage. The reason of that must be that the seedling test was carry out with one isolate while the adult resistance of genotypes were tested with PTT population. The 7.3% of the varieties were resistance in both growing stages.

This information assists the breeder to choose resistant parents in the crossing programs, but the clarification of the background of net blotch resistance of barley needs further detailed study.

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Related publication:

Ficsor A, Bakonyi J, Tóth B, Tomcsányi A, Palágyi A, Csősz M, Károlyi-Cséplő M, Mészaros K, Vida G (2010) First report of spot form of net blotch of barley caused by *Pyrenophora teres f. maculata* in Hungary. Plant Dis. 94 (8), p. 1062.

Ficsor A, Bakonyi J, Csősz M, Tomcsányi A, Varga J, Tóth B (2014a) Occurrence of barley pathogenic *Pyrenophora* species and their mating types in Hungary. Cereal Res. Commun. 42 (4), 612–619.

Ficsor A, Tóth B, Varga J, Csősz M, Tomcsányi A, Mészáros K, Kótai É, Bakonyi J (2014b) Variability of *Pyrenophora teres f. teres* in Hungary as revealed by mating type and RAPD analyses. J. Plant Pathol. 96 (3), 515–523.

Keywords: barley, *Pyrenophora teres f. teres*, seedling and adult resistance, biotic stress

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Using grain carbon isotope composition (d13C) at harvest to assess growing conditions and cultivar drought tolerance

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Althought drought is a frequent limiting factor of cereal production across the world, breeding for drought tolerance remains challenging (Blum, 2005). Possible causes have been frequently debated: diversity of drought scenarios, difficulties to run field trials under suitable stressing conditions, large genotype x environment interactions, numerous traits to study and phenotype for (Chenu et al., 2011).

The use of carbon isotope composition (δ13C) or discrimination (Δ13C) in plant tissues as an estimate of water use efficiency has been demonstrated for many years (Farquhar and Richards, 1984); however, its use as a common tool in cereal breeding program seems limited, despite some successful demonstrations (Rebetzke et al., 2002).

The interest of using grain δ13C in West-European bread wheat breeding programs has been explored during an FSOV research project focused on identifying traits leading to drought tolerance and associated evaluation tools (Deswarte et al.). A set of grain samples have been analyzed from various trials, in order to provide basic and practical information to breeders about the way to implement and analyse such data.

The experimental precision of grain δ13C within trial was assessed by analyzing separately samples from 3-4 blocks. Average values were thus completed with error-types. Across a 15-trials network and two check cultivars (Apache and Bermude), average error-type was 0.37‰ (with a range between 0.12 to 0.99‰), with higher plot-to-plot instability under water-stress conditions.

Genetic diversity (70 cvs) for δ13C in grain was assessed through 25 trials, with a single sample per trial and per cultivar. Difference between the highest and the lowest lines was >2‰, and between the top and bottom 20th percentiles was 0.41‰. Within a set of French or European commercial cultivars, genetic diversity for δ13C largely exceeded experimental precision; it is thus feasible to screen extreme parts of populations and identify lines to keep or discard. It is noticeable that under west-European conditions, high-yielding varieties tend to show low water-use efficiencies.

Tissue δ13C can also be used as an indicator of actual water stress experienced by a crop. Average trial grain yield was regressed against estimated crop water deficit (based on soil description and weather data) and averaged δ13C (measured on several known cultivars) across 35 trials with contrasting water supply and yield levels. Correlation between mean δ13C and grain yield was higher ($r^2=0.58$) than the one between estimated crop water deficit and yield ($r^2=0.38$). In particular, the use of δ13C provided a significant modification of interpretation when soil water supply was difficult to assess (chalky soil, ground water table), or climatic conditions were stressful (high temperature and evaporation demand).

δ13C can thus be used either to select high-yielding lines or to qualify growing conditions without extensive soil description and computation of agro-environmental indices. Using grains instead of leaf tissue allows mechanization compared to leaf tissue, and final output remains precise and informative.

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Keywords: drought, carbon isotope composition, growing conditions, wheat, grain tissue

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PIV-51 - Roberto TUBEROSA - Dissecting the root angle QTLome to enhance durum wheat sustainability

Dissecting the root angle QTLome to enhance durum wheat sustainability

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The optimisation of root system architecture (RSA) is an important objective for the sustainability of durum wheat grown under different water regimes. In the present study, linkage and association mapping (AM) for RSA evaluated at the seedling stage evidenced 20 clusters of quantitative trait loci (QTLs) for root length and number as well as 30 QTLs for root growth angle (RGA). The most divergent RGA phenotypes observed by seminal root screening were validated by root phenotyping of field-grown adult plants. QTL analysis of RSA and grain yield data indicates RGA as a valuable target to enhance grain yield and yield stability across different soil moisture regimes (Maccaferri et al. 2016, JXB, 67, 1161–1178, 2016). Based on their relative additive effects, allelic distribution in the AM panel and co-location with QTLs for yield, eight RGA QTLs have been prioritised in terms of breeding interest and value. These QTLs were investigated for gene content based on the chromosomal pseudomolecules of Chinese Spring *T. aestivum* and the TriAnnot v4.3 gene prediction and annotation pipeline and the Zavitan *T. dicoccoides* genome assembly (Avni et al. 2017). The chromosome regions contained 25 to 242 predicted genes (123 on average). In six RGA QTLs, from one to four gene annotations were involved in auxin pathways. The comparison between the *T. aestivum* and T. *dicoccoides* gene content indicates the high quality of the *T. dicoccoides* assembly and its usefulness to identify candidates to explore the polymorphism and the structural variation present in the A and B wheat genomes.

Keywords: Durum wheat, Root angle, Root system architecture, QTL, Molecular mapping, Candidate gene

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PIV-52 - Roberto TUBEROSA - Fine-mapping of QSBM.UBO-2BS=SBM2, a major QTL for resistance to soil-borne cereal mosaic virus (SBCMV)

Fine-mapping of QSBM.UBO-2BS=SBM2, a major QTL for resistance to soil-borne cereal mosaic virus (SBCMV)

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QSbm.ubo-2BS=Sbm2, a major QTL controlling the response to Soil-Borne Cereal Mosaic Virus (SBCMV) in durum wheat, was characterized in two recombinant inbred line populations, namely Meridiano (resistant, R) x Claudio (moderately susceptible, MS) and Simeto (susceptible, S) x Levante (R). By means of meta-QTL analysis QSbm.ubo-2BS was mapped as a unique QTL within a 2 cM-wide interval (LOD-2) in the distal region of chromosome arm 2BS (Maccaferri et al., 2012). The addition of the Illumina 90K SNPs array to the durum linkage maps allowed to identify 36 transcripts-associated SNPs tightly associated with the Mendelized QTL (Maccaferri et al., 2014). Nine SNPs from the Illumina 90K wheat array were converted to KASP markers, which provided fluorescent high-throughput assays spanning the QTL region. Marker-assisted selection (MAS) was performed on ~2,000 RILs from the Svevo (R) x Ciccio (S) population (Gadaleta et al., 2009) with two KASP markers flanking the QTL interval, KUBO 9 and KUBO 13. MAS identified 303 lines recombinant between the two markers. These lines were characterized for SBCMV response in the 2016 field nursery under severe and uniform SBCMV infection. The lines were scored for symptom severity (SS) on a 0 to 4 scale , where 0 = very resistant and 4 = very susceptible (Vallega and Rubies Autonell, 1985). They were also genotyped with seven KASP markers distributed along the QTL interval (KUBO 1, KUBO 3, KUBO 27, KUBO 29, KUBO 38, KUBO 40 and KUBO 41) and with the DArT marker wPt-2106, the marker most associated with the phenotype in the Meridiano x Claudio RIL population (Maccaferri et al., 2012). The bimodal distribution of SS reflects the segregation of the resistance trait in the QTL interval: frequency peaks were observed around values of 1.7 and 3.8. The fine-mapping allowed to narrow the most probable support interval to 0,2 cM between KUBO 27 and KUBO 1 based on the Svevo x Ciccio RILs population. Work is in progress to analyze other ~1,000 RILs of the same population in order to confirm the results obtained with the first part of the study and to improve the ongoing fine-mapping of the gene responsible for the resistance.

Keywords: durum wheat, SBCMV, QTL, fine-mapping, KASP marker

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Fusarium head blight and *Fusarium* toxins accumulation in grain of winter wheat breeding lines inoculated with *Fusarium culmorum*

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Fusarium head blight (FHB) is a disease of cereals caused by fungi of genus *Fusarium*. These fungi produce toxic metabolites – mycotoxins, with phyto-and zootoxic potential. There are several types of resistance to FHB: type I – to infection, type II – to the spread of *Fusarium* in the ear, type III – to the kernel damage by *Fusarium*. Other types are the tolerance to FHB or toxins (DON) (type IV) and type V – resistance to the accumulation of toxins (trichothecenes) in the grain through their chemical modification or blocking of synthesis.

In order to obtain the forms of winter wheat that combine different types of resistance 77 lines were evaluated in field experiments in two locations. Among them there were five resistant checks: '20828[Fhb1–]', 'A40-19-1-2', 'Arina', 'Fregata', 'UNG 136.6.1.1[Fhb1+]'; five lines with Fhb1 gene from crosses of winter wheat cultivars with 'Sumai 3'; six susceptible checks; three lines with high accumulation of trichothecenes; three high yielding cultivars 'Artist', 'Patras' and 'RGT Kilimanjaro'. Wheat heads were inoculated with the spore suspension of *F. culmorum* isolates producing DON, NIV and ZEN. *Fusarium* head blight index (FHBi) was evaluated. The proportion of *Fusarium* damaged kernels (FDK) was determined visually by dividing the sample on healthy looking kernels and with symptoms of *Fusarium* damage. Using the technique of gas chromatography and immunoenzymatic tests the contents of the DON and acetyl derivatives, NIV and ZEN in the grain were analysed.

Average FHBi was 14.6%; at the range of 3.6 – 453.2%. The least infected were lines 'S43[Fhb1 +]', 'Fregata' cv., '20828 [Fhb1-]', 'S32[Fhb1+]', 'S38[Fhb1-]', 'A40-19-1-2', 'UNG 136.6.1.1', 'S10[Fhb1+]', STH 032, STH 2041, KOH 275 and the most infected were six susceptible checks. The average value of the FDK amounted to 14.4%; at the range of 1.5 – 41.5%. The least damaged kernels had lines 'S 43[Fhb1+]', 'S38[Fhb1-]', 'S10[Fhb1+]', 'POB 679/03', 'S 32[Fhb1+]', 'POB 170/04'. The most damaged were kernels of susceptible checks and three cultivars 'Artist', 'Patras' and 'RGT Kilimanjaro'. There was a significant linear relationship between the FHB index and the proportion of Fusarium kernel damage – $r = 0.771$, $p < 0.001$. Chemical analyses showed the presence of *Fusarium* toxins DON, 3AcDON, NIV and ZEN in the grain samples. Average concentrations of DON, 3AcDON, NIV and ZEN were 6.777 mg/kg (0.750-19.750 mg/kg), 0.303 mg/kg (0.050-0.950 mg/kg), 5.146 mg/kg (0,750-22.600 mg/kg), 1115 mg/kg (147-6875 mg/kg), respectively. Lines differed significantly in terms of content of toxins in the grain. The lowest concentration of toxins was detected in grain of resistant checks 'S32[Fhb1+]', 'S38[Fhb1-]', 'S10[Fhb1+]', 'S30[Fhb1+]', 'S43[Fhb1+]', '20828[Fhb1-] and breeding lines 'POB 0616' 'POB 0111, 'POB 0211', 'POB 6793/03', 'NAD 13015', POB 170/04', POB 457/07', 'NAD 13024'. The highest concentration of toxins was detected in grain of five susceptible checks, cultivar 'Artist' and breeding lines 'DM 2566/11', 'KBP 08 13', 'LAD 463/05', 'C 3779/10'.

It was possible to identify lines combining resistance to FHB (type I + II), to kernel damage (type III) and the low accumulation of *Fusarium* toxins (type V).

Keywords: *Fusarium*, wheat, toxins, trichothecenes, zearalenone

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A fast and reliable method for assessing *Fusarium* damaged kernels and the mycotoxin content by digital picture analysis in wheat and triticale

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Fusarium Head Blight (FHB) is a cereal disease that induces significant yield losses and contaminates the kernels with mycotoxins. It is widely admitted that the use of FHB-resistant plants is the best method for controlling the disease. The ultimate goal of resistance breeding is to identify cultivars with low risk of mycotoxin contamination. However, testing large numbers of lines for toxin content is expensive and remains prohibitive. As infected kernels are smaller, shriveled and white to pale pink in color, another method widely used is to visually estimate the proportion of *Fusarium* Damaged Kernels (FDK) as an approximation for *Fusarium* severity. This method is an easy and efficient way to select for low toxin, but is time consuming and labor intensive. Digital picture analysis could be a very good option to simplify FDK evaluation. Our new method is based on the hypothesis that it is possible to select for low mycotoxin accumulating lines by checking the level of whitening of the kernel sample. Pictures are taken with a digital camera under controlled light conditions. We used RGB criteria to segregate the picture´s pixels in three categories: background pixels, healthy-grain pixels and diseased-grain pixels. The Whitened Kernel Surface (WKS) is then calculated as the proportion of diseased-grain pixels among all grain pixels. We tested this method on two bread wheat panels gathering 228 grains samples in total, and one triticale panel of 37 samples. We showed a high Pearson correlation between FDK and WKS (r=0.79 and r=0.77 for bread wheat and r=0.81 for triticale), and between mycotoxin content and WKS (r=0.86 and r=0.73 for bread wheat and r=0.61 for triticale). This new notation criterion based on pictures analysis is a promising tool for breeders and researchers. It is as efficient as the traditional visual notation and happens to be faster, easier and more stable. A very economical way to evaluate mycotoxin content and to enable the large scale scoring and ranking needed to select resistant cereal varieties for the future.

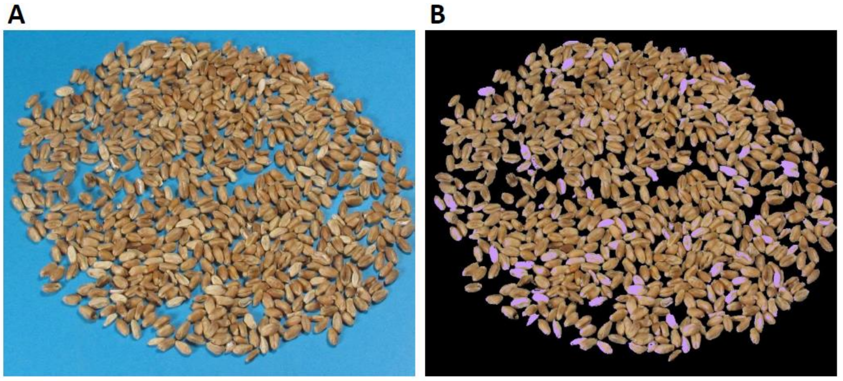

Fig. 4. Infected sample of bread wheat. **A**, Original picture; **B**, Treated picture, with background pixel artificially turned in black, and diseased grain pixels turned in pink.

Keywords: Digital picture analysis, *Fusarium* Head Blight, FHB, Wheat, Triticale, FDK, DON, Mycotoxin, WKS

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PIV-55 - Jose MORENO-AMORES - Assessing the incorporation of covariates in genomic predictions for *Fusarium* head blight in durum wheat

Assessing the incorporation of covariates in genomic predictions for *Fusarium* head blight in durum wheat

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Durum wheat (*Triticum turgidum* var. durum) is an important cereal in Europe especially susceptible to *Fusarium* head blight (FHB) which produces yield and quality reduction due to the grain contamination with mycotoxins. In this study, we used a cross-validation schema to assess the accuracies of the genomic predictions for our trait of interest (FHB mean severity) and its correlation with two covariates: heading date (HD) and plant height (PH). Predictions relied on genomic best linear unbiased prediction GBLUP models using a panel of 184 durum wheat genotypes across five environments alongside with their genome-wide set of 30,611 SNP and DArT markers.

Significant negative correlations were found between the estimations of FHB mean severity and the two covariates. This motived us to evaluate: (i) two methods to correct the FHB estimated values before running the prediction models; (ii) the covariate(s) incorporation as fixed factor(s) on GBLUP models and (iii) the use of restriction indexes in a multi-trait model context.

We corroborated the increase in FHB mean prediction accuracy (0.54) when genotype-environment interaction was included compared to the one obtained within each trial (ranging from 0.38 to 0.52). Also, accuracy differences were mainly found within environments between the adjustment attempts for covariate HD as follows: correcting FHB values before predictions proved to be the best option for at least not producing any significant decrease in accuracy terms. On the other hand, among adjustments methods for PH, there were no accuracy differences. All of the attempts succeed reducing to non-significant levels the correlation between the predicted FHB and both covariates. Our results describe the impact of incorporating covariates in genomic prediction models and possible approaches to addressee it.

Keywords: Genomic prediction, *Fusarium* head blight, durum wheat, covariates, plant height, heading date

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PIV-56 - Mahbubjon RAHMATOV - Development and characterization of new stem rust and stripe rust resistance genes in Swedish wheat-rye introgression lines

Development and characterization of new stem rust and stripe rust resistance genes in Swedish wheat-rye introgression lines

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The emergence of new stem rust (*Puccinia graminis* f. sp. *tritici*) and stripe rust (*Puccinia striiformis* f. sp. *tritici* (Pst)) races poses a major threat to Swedish wheat production. To mitigate the threats of these virulent pathogens, the objectives of this study were to characterize new sources of resistance genes in wheat-*Secale cereale* derivatives.

A new 2DS·2RL wheat-rye translocation was obtained through the breakage-fusion mechanism and found to carry a novel stem rust resistance gene designated *Sr59* from Secale cereale. *Sr59* confers a high level of resistance to many widely virulent races including TTKSK, TTTSK, TTKTT, TKTTF, TKTTF+Sr25, TTRTF, TPMKC, RKQQC, RCRSC and two isolates of race TTTTF from USA and Russia. Research is currently being conducted to transfer *Sr59* into adapted elite lines with high yield potential, agronomic adaptation, and end-use quality characteristics. The characterization of *Sr59* and its introgression through 2RL into wheat provides wheat breeders with an additional genetic resource for breeding for stem rust resistance. Moreover, six 4R (4D), 5R (5D) and 7R (7D) wheat-rye multiple disomic substitution lines were found highly resistant to a suite of broadly virulent Pst races (21 in total). Development of wheat-rye translocations are currently being investigated through induced-homoeologous recombination that may enhance the utility of rye resistance genes, thus contributing to the diversification of both stem rust and stripe rust resistance. Rye has been an important donor of rust resistance genes, in addition to contributing to yield potential, adaptability, and tolerance to various biotic and abiotic stresses in wheat. Rye remains a largely untapped reservoir of genes for wheat improvement and should be investigated more thoroughly.

Related publication:

Rahmatov M, Rouse MN, Steffenson BJ, Andersson SC, Wanyera R, Pretorius ZA, Houben A, Kumarse N, Bhavani S, Johansson E (2016) Sources of stem rust resistance in wheat-alien introgression lines. Plant Disease 100:1101-1109

Rahmatov M, Rouse MN, Nirmala J, Danilova T, Friebe B, Steffenson BJ (2016) A new 2DS·2RL Robertsonian translocation transfers stem rust resistance gene *Sr59* into wheat. Theor Appl Genet 129

Rahmatov M, Hovmøller MS, Nazari K, Andersson SC, Steffenson BJ, Johansson E (2017) Seedling and adult plant stripe rust resistance in diverse wheat-alien introgression lines. Crop Science 57:2032–2042

Keywords: wheat, rye, resistance gene, stem rust, stripe rust, Robertsonian translocation

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Detection of functional and non-functional alleles of the stripe rust resistance gene *Yr15* using diagnostic markers

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Stripe rust, caused by the fungus *Puccinia striiformis f.s. tritici* (Pst), is a destructive disease of wheat globally. Depletion of effective resistance to Pst in cultivated wheat has led to search for new resistance genes in the wild relatives of wheat. One of the most promising genes conferring broad-spectrum resistance to stripe rust is *Yr15*, derived from wild emmer wheat (*Triticum dicoccoides*) accession G25. *Yr15*, mapped on chromosome arm 1BS, has recently been cloned by our consortium and designated as Wheat Tandem Kinase 1 (WTK1). We found wtk1 susceptible alleles in most 274 tested durum, bread, and wild emmer wheat lines. Out of 69 tested durum and bread wheat cultivars and lines, only 33 *Yr15* introgression lines contained the functional allele (Wtk1) from G25 and were resistant to Pst. The remaining 36 susceptible lines carried non-functional alleles (wtk1), which included insertions of large transposable elements that resulted in changes in reading frame. Development of reliable molecular markers can facilitate the introgression of *Yr15* into new varieties via marker-assisted selection. Diagnostic markers designed based on the polymorphism between the WTK1 alleles are preferred in order to avoid negative linkage drag. Therefore, we have designed high-throughput co-dominant KASP markers that can differentiate between the functional (Wtk1) and all known non-functional (wtk1) alleles, and can be used in breeding programs for development of modern cultivars with high resistance to stripe rust.

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Using crop-pathogen modeling to identify plant traits to control *Zymoseptoria tritici* epidemics on wheat

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Diversification in pathogen control methods to reduce the severity of economically important foliar diseases such as *Zymoseptoria tritici* on wheat is needed. One way is to identify plant physiological and architectural traits that influence disease development and that can be selected in the process of crop breeding. Such traits may be used for improving tolerance or disease escape. Traits favoring disease escape, the focus of our work, may significantly decrease crop epidemics (Robert et al., 2018). However, understanding the role of such traits in crop-pathogen interactions is a daunting task because the interactions are multiple and dynamic in time. To characterize and quantify crop-pathogen interactions, an innovative trait-based and resource-based modeling framework was developed (Precigout et al., 2017). In this framework, the pathosystem is assumed to respond dynamically to both architecture and physiological status of the host canopy. A canopy consists of plenty of small patches, i.e. small functional and infectable units of leaf tissue. Production of new patches, for canopy growth and renewal of photosynthetically active plant tissues, is a function of the available resources produced by the other patches. Pathogen spores can contaminate nearby healthy patches. The definition of patch proximity depends on dispersal abilities of the pathogen and canopy architecture. We used and adapted this modeling framework to quantify the effects of several plant traits on *Zymoseptoria tritici* epidemics for varied climate scenarios. The complex infection cycle of *Z. tritici* characterized by a long symptomless incubation period was implemented in the model. We studied plant architectural traits such as leaf size or stem height, and plant physiological traits such as leaf lifespan or leaf metabolite contents. In our simulations, these traits impacted the epidemics dynamics though their effects on pathogen dispersal and on the amount of resources available for the pathogen. Sensitivity analyses showed how disease severity depended on plant traits and pathogen virulence. The importance of several plant and pathogen traits could be linked to the pathogen's ability to manage the race for the colonization of the canopy in the face of canopy growth. Playing on host traits also made it possible to simulate different wheat varieties - with contrasted heights, pathogen resistance or precocity - to characterize the behavior of the pathosystem of interest for different host ideotypes. We argue that this kind of trait-based modeling approach is a valuable tool to identify plant traits promoting more resilient agroecosystems in particular for crop breeding in a context of innovative and sustainable crop protection.

Related publication:

Précigout P-A, Claessen D, Robert C. 2017. Crop fertilization impacts epidemics and optimal latent period of biotrophic fungal pathogens. Phytopathology 107, 1256-1267.

Robert C, Garin G, Abichou M, Houlès V, Pradal C, Fournier C. 2018. Plant architecture and foliar senescence impact the race between wheat growth and *Zymoseptoria tritici* epidemics. Annals of Botany (in press).

Keywords: *Septoria tritici* blotch, *Zymoseptoria tritici*, wheat, fertilization, epidemiology, tolerance, trait-based modeling, virulence, consumer-resource dynamics, disease-escaping traits

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PIV-59 - Claire GUÉRIN - Genome-wide identification, characterization and expression profiling of NAC genes in *Triticum aestivum*

Genome-wide identification, characterization and expression profiling of NAC genes in *Triticum aestivum*

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The bread wheat, *Triticum aestivum*, is an important economic crop worldwide and its yield is continuously affected by the global warming. Insights into wheat adaptability mechanisms of its seed development under different abiotic stresses represent a new challenge to improve wheat yield.

Transcription factors are major actors of molecular and cellular mechanisms involved in the regulation of development under physiological conditions and in response to many biotic and abiotic stresses.

The NACs are a large family of transcription factors, present in the whole plant reign. Some members of this family have been shown to be involved in the development such as flowering, lateral roots division, cell division or seed development; and in response to biotic (i.e. stripe rust) and abiotic (i.e. salinity or drought) stresses in different species. However, the characterization of this family remains largely unknown in *Triticum aestivum*.

Here, we compared the latest sequences available in the database of the International Wheat Genome Sequencing Consortium (https://www.wheatgenome.org/) with the data gathered by Borrill et al. (2017) in order to improve the quality of the NAC family existent set.

Furthermore, a phylogenic structuration of the enriched family was done; particularly by completing the homeologous groups. Its chromosomal repartition was also studied and showed that: i) NACs are equitably distributed within the three subgenomes (A, B and D) of *Triticum aestivum* and ii) NACs have a different distribution within the seven chromosomes, with a higher abundance on chromosomes 2 and 7.

We hypothesized that some duplication events may explain the physical and phylogenical clustering of several NAC genes within the family.

Secondly, we focused on the expression profile of some key members of the NAC family in bread wheat.

We selected 24 NAC genes that were found to be differentially expressed in different tissues (roots, leaves, seeds) or in response to different abiotic stresses in leaves (heat shock and PEG induced drought at a young stage), based on the data available on the www.wheat-expression.com platform (Borrill et al., 2016). Using real-time quantitative PCR, the behavior of these 24 NAC genes was studied on the genome referenced genotype, Chinese Spring (IWGSC) in the same heat shock conditions. The expression of those 24 NAC genes was also quantified in leaves and seeds at 220 and 450°Cd post-anthesis, under severe drought conditions applied during the seed development of two contrasted genotypes in field (Pheno3C platform, INRA Clermont-Ferrand).

This work may lead to increase knowledge about the NAC family in *Triticum aestivum*. We identified genes which are preferentially expressed in leaves and seeds among which some respond to heat and drought stresses.

These data might further give useful information for breeders to develop adapted varieties to global warming.

Keywords: bread wheat, transcription factor, NAC family, phylogenic structuration, gene expression

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The association mapping of short-term heat shock and chronic heat stress tolerance in durum wheat

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Heat stress due to increased temperature is an agricultural problem in many areas of the world. Short-term heat stress (acute heat stress) provokes heat shock response, in contrast, long-term heat exposure (chronic heat stress) induces larger scale adaptations. The present work aims to carry out a comprehensive phenotypic analysis of a durum wheat collection for the response to high-temperature followed by an association mapping study to detect Quantitative Trait Loci (QTLs) associated with heat tolerance. The response to chronic heat stress and short-term heat shock has been studied in controlled environments where specific heat treatments may be applied. The panel was profiled with simple sequence repeat, Diversity Arrays Technology and sequence-tagged site markers (957 markers in total). The genetic relationships among the accessions have been investigated using both a genetic-similarity and a model-based Bayesian clustering method. Significant differences among genotypes were observed for all measured heat response traits and thirty four single marker/QTL regions were located in all chromosomes. It is interesting that a higher number of QTLs were detected for constitutive heat response trait (heat shock applied to detached leaves) as compared to the two traits involving acquired responses, measured on living plants pre-adapted to high temperatures. Molecular markers linked to the heat tolerance trait represented a more reliable tool for selecting heat tolerance genotypes in durum wheat breeding programs.

Related publication:

Maccaferri M, El-Feki W, Nazemi G, Salvi S, Canè M.A, Colalongo M.C, Stefanelli S, Tuberosa R 2016 Prioritizing quantitative trait loci for root system architecture in tetraploid wheat. Journal of Experimental Botany. 67(4):1161-78.

Nazemi G, Valli F, Maccaferri M, Salvi S, Tuberosa R 2015 Genetic variation for aerenchyma and other root anatomical traits in durum wheat. (2015), Genetic Resource Crop Evolution Journal. DOI 10.1007/s10722-015-0279-6.

Keywords: Durum wheat, Heat stress response, association mapping

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Molecular breeding of salt tolerant wheat

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Over the last few decades, on average ca 2,000 ha of irrigated agricultural land has been lost every day due to high salinity levels. The size of high salinity areas has been growing from 45 million ha in the early 1990s to its current volume of 62 million ha [1]. Globally, 3.1% of the total land mass is affected by salinity, with over half of the world's countries afflicted [2]. Presently, about one-fifth of the world's irrigated lands are too salty for agriculture. Clearly, this negative trend has to be broken.

Our aim is to develop wheat (*Triticum aestivum* L.) varieties with increased salinity tolerance. Starting from Gom-25 [3], a commonly grown in Bangladesh variety already fairly well adapted to salt and heat stress, we introduced ca 1 million mutations per genome by EMS (Ethyl Methane Sulphonate), and raised a population of ca 1800 lines. Mutated wheat lines with an increased salinity tolerance are now selected from this population. The aim is to reuse some parts of the 1 million ha area of salt contaminated land in Bangladesh.

Related publication:

[1] Qadir *et al* (2009) Salt-induced land and water degradation in the Aral Sea basin: A challenge to sustainable agriculture in Central Asia. Natural Resources Forum. Wiley Online Library, pp 134-149
[2] Corbishley and Pearce (2007) Growing trees on salt-affected land. Australian Centre for International Agricultural Research
[3] Hossain and Da Silva (2013) Wheat production in Bangladesh: its future in the light of global warming. AoB plants 5: pls042

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SESSION V. HIGH QUALITY AND SAFE CEREALS FOR FOOD, FEED AND INDUSTRY

PV-61 - Catherine RAVEL - Rapid identification of high-molecular-weight glutenin subunits by SNP markers to breed for high quality

Rapid identification of high-molecular-weight glutenin subunits by SNP markers to breed for high quality

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In bread wheat (*Triticum aestivum*), the high-molecular-weight glutenin subunits (HMW-GS) are the most important determinant of technological quality. Known for their high level of diversity, they are encoded by tightly linked genes (*Glu-1-1* & *Glu-1-2*). Their identification is based on elecphoresis (SDS-PAGE). However, molecular markers would facilitate the early selection of high-quality HMW-GS alleles in breeding programmes. In this work, we identified sequence polymorphisms and developed 19 SNP markers thanks to an alignment comprising the nucleotidic sequences of the main HMW-GS alleles publicly available. Genotyping and electrophoresis result obtained in a core collection (n=364 lines), which represents the worldwide diversity, were compared. SNP markers developped at *Glu-A1* and *Glu-D1* and SDS-PAGE gave concordant results. Discrepancies were observed mainly at *Glu-B1* locus. However, genetic association showed that markers at *Glu-B1* genes could be more efficient than SDS-PAGE allele to improve rheological properties of the dough. To conclude, this study provided a set of SNP markers, which could constitute a useful tool-box to breed for the end-used value.

Keywords: Bread wheat (*Triticum aestivum* L.). End used value. High-molecular-weight glutenin subunit. Single Nucleotide Polymorphism

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PV-62 - Emmanuelle BANCEL - Proteomic changes occuring in einkorn (*Triticum monoccocum*) grain during filling stages of development

Proteomic changes occuring in einkorn (*Triticum monoccocum*) grain during filling stages of development

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Albumins and globulins of wheat endosperm represent about 20% of total grain proteins. Nutritionally, the albumins and globulins (non-glutens) have a very good amino acid balance. Many of these proteins are enzymes involved in metabolic activity. Most of these physiologically active proteins can also influence the processing and rheological properties of wheat flour.

Incomplete knowledge of these enzymes and related proteins present in the endosperm of wheat limits our understanding of the biochemical events taking place in the grain during the first stages of its development. So it is necessary to study and evaluate qualitative and quantitative changes occurring during this process. To reach this objective, our study is based on *Triticum monococcum* (einkorn), a diploid wheat species, used as a model species of bread wheat (*T. aestivum*).

We used a proteomic approach to identify the key actors during the filling stages of development in einkorn grain. Five stages were studied (200 to 600°Cd). Large-scale analysis using the gel free proteomic approach allowed us to identify a total of 2,475 proteins. Among these proteins, 512 (20.7%) were accurately quantified, 488 of them presented changes related to stage development, highlighting this dynamic process (Bonnot et al, 2017).

Hierarchical cluster analysis revealed four major expression profiles throughout grain development. The classification of proteins identified by MS/MS in 17 different biochemical functions and nine molecular functions provided a picture of the temporal cellular processes involved. Several proteins differentially expressed and associated with grain development and/or quality were also shown.

A co-accumulation network of all differentially expressed proteins mainly centred on enzymes involved in carbon and protein metabolism was finally established.

Related publication:

Bonnot T, Bancel E, Alvarez D, Davanture M, Boudet J, Pailloux M, Zivy M, Ravel C, Martre P (2017) Grain subproteome responses to nitrogen and sulfur supply in diploid wheat *Triticum monococcum* ssp. *monococcum*. Plant J. doi: 10.1111/tpj.13615

Keywords: wheat, grain, albumins globulins, proteome, co-accumulation network

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PV-63 - Julie BOUDET - SPA Heterodimerizing Protein, a bZIP transcription factor, represses glutenin synthesis in *Triticum aestivum*

SPA Heterodimerizing Protein, a bZIP transcription factor, represses glutenin synthesis in *Triticum aestivum*

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The quality of the wheat grain is determined by the quantity and the composition of grain storage proteins (GSPs). These GSPs consist of the glutenin of low- (LMW-GS) and high- (HMW-GS) molecular-weight and the gliadin. The synthesis of these proteins is essentially regulated at the transcriptional level and by the availability of nitrogen and sulfur. This network of regulation has been extensively studies in barley. BLZ1 and BLZ2, belonging to the basic leucine Zipper (bZIP) family activate the synthesis of hordeins. To date, in wheat, only the orthologue of BLZ2, SPA or Storage Protein Activator, was identified to play a major role in the regulation of GSP synthesis. Here, the involvement of the orthologous of BLZ1 named SPA Heterodimerizing Protein (SHP) was analyzed on the transcriptional regulation of the genes coding for GSPs. By gel mobility shift assays, we have demonstrated that SHP binds with cis-motifs, known to bind to the bZIP family TFs, on the HMW-GS and LMW-GS promoters (*GluB1-1* and *GluD3*, respectively). Moreover, we showed by transient expression assays in wheat endosperms that SHP acts as a repressor on the activity of these gene promoters. These results were supported by transgenic lines overexpressing SHP gene grown with low and high nitrogen supply. The phenotype of SHP overexpressing lines showed a lower proportion of both LMW-GS and HMW-GS, while the proportion of gliadin was unchanged, whatever the availability in nitrogen. Thus, the gliadin/glutenin ratio was increased, which suggest that gliadin genes may be regulated differently than glutenin genes.

Keywords: bZIP transcription factor, gluten, SPA Heterodimerizing Protein, storage proteins, wheat (*Triticum aestivum* L.).

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PV-64 - Pierre BERTHOMIEU - Towards the positional cloning of genes controlling zinc and iron content in durum wheat grain

Towards the positional cloning of genes controlling zinc and iron content in durum wheat grain

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Wheat grain contains low levels of iron (Fe) and zinc (Zn). In addition, most of the phosphorus in the grain is stored as phytate, which is an antinutrient that chelates Fe and Zn, thereby rendering them unavailable in the human diet. Thus, improving both accumulation and bioavailability of Fe and Zn in wheat grains is desirable. A couple of international breeding programs work in that direction. However, there is up to now very little information on the mechanisms and genes controlling the amount and bioavailability of Fe and Zn in the grain.

Our project aims at identifying genetic loci and associated genes controlling Fe, Zn and phytate accumulation in durum wheat grain. We developed a genome wide association study on an original population (1) obtained by intercrossing a large set of durum, emmer and wild wheat accessions. Preliminary results show that two regions (6 Mb- and 4 Mb-long, respectively) were identified as harbouring loci controlling Fe and Zn content in grain. Interestingly, the haplotype associated to high Fe content seems to come from the wild compartment.

Thanks to the recent availability of the sequence of the durum wheat genome (2), positional cloning is under way.

Related publication:

David et al. (2014) Genotyping by sequencing transcriptomes in an evolutionary pre-breeding durum wheat population. Molecular breeding, 34:1531-48

Avni et al. (2017) Wild emmer genome architecture and diversity elucidate wheat evolution and domestication. Science. 357:93-7

Keywords: Wheat grain, biofortification, zinc, iron, GWAS, EPO population

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SESSION VI. FUTURE CHALLENGES AND INNOVATIONS

PVI-65 - Péter MIKÓ - Selection of winter cereals for organic agriculture

Selection of winter cereals for organic agriculture

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Productivity in agriculture has more than tripled in developed countries since the 1950s. Beyond the success of plant breeding, the increased use of inorganic fertilizers, application of pesticides, and spread of irrigation also contributed to this success. However, impressive yield increases started to decline in the 1980s because of the lack of sustainability. One of the most beneficial ways to increase sustainability is organic agriculture. In such systems the prerequisite of successful farming is the availability of crop genotypes that perform well. However, selection of winter cereals for sub-optimal growing conditions is still neglected, and the organic seed market also lacks of information on credibly tested varieties suitable for organic agriculture.

Quality and agronomic performance of a diverse set of 37 winter bread wheat varieties and 14 winter durum varieties were examined in organic and conventional management systems of Austria, France (only durum) and Hungary through 3 years to identify traits sensitive to management systems.

Based on the results of the winter wheat trial, seven traits showed significant management × genotype interaction. Heading date, sensitivity to leaf rust and powdery mildew had high heritability making it reasonable to select them for organic agriculture also in conventional field in early generations. However, it is suggested to select for the other four traits (grain yield, test weight, leaf-inclination and vigorous plant growth) later under organic growing conditions.

In the case of winter durum, heading time, wet gluten content, semolina yield and grain protein content are traits that showed genotype-dependent significant differences between the two management systems examined. Therefore, breeding for these traits could result in specifically adapted genotypes for organic agriculture in the different countries. Based on strong or moderate significant correlations between the winter durum traits, gluten index and plant height could also be specifically selected in an indirect way. The need for environmentally specific selection for grain yield in later generations was also demonstrated.

Our findings provide evidence for the influence of the selection environment, whether it is the management system or the growing region. Based on these findings, at least a partly separate winter cereal selection program is recommended for organic and low input agriculture in each country, where targeted selection could be carried out more effectively.

Knowledge gained from these experiments will be used to design efficient cultivar testing strategies for organic farming in a European network. This will support farmers in their choice of cultivar with stable yield and high quality and thus support the emerging movement towards sustainable farming systems.

This research received funding from the European Community's Seventh Framework Programme (FP7/2007-2013) under Grant Agreement No. 245058-SOLIBAM (supplementary Hungarian project EU_BONUS_12-1-2012-0032) and from the European Union's Horizon 2020 research and innovation programme under grant agreement No 727230 (LIVESEED). The information contained in this communication only reflects the author's view. The Research Executive Agency is not responsible for any use that may be made of the information provided.

Related publication:

Mikó, P., Vida, G., Rakszegi, M., Lafferty, J., Lorentz, B., Longin, C.F.H., Megyeri, M. (2017): Selection of winter durum genotypes grown under conventional and organic conditions in different European regions. Euphytica, 213: 169; DOI 10.1007/s10681-017-1953-x

Mikó, P., Löschenberger, F., Hiltbrunner, J., Aebi, R., Megyeri, M., Kovács G., Molnár-Láng, M., Vida, G., Rakszegi, M. (2014): Comparison of bread wheat varieties with different breeding origin under organic and low input management. Euphytica, 199(1-2): 69-80; DOI: 10.1007/s10681-014-1171-8

Keywords: wheat, durum wheat, organic breeding, sustainability, variety performance

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PVI-66 - Rita ARMONIENE - Identification of novel alleles of starch biosynthesis genes responsible for amylose variation in winter wheat TILLING population

Identification of novel alleles of starch biosynthesis genes responsible for amylose variation in winter wheat TILLING population

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Starch is a main component of wheat grain and consists of two glucan polymers amylose and amylopectin with the ratio ranging between 25 and 75%, respectively. The amylose/amylopectin ratio has a major influence over the physiochemical properties of starch and determines its optimal application in the industry. Recently, amylose-free (Waxy) and high-amylose wheats, consisting up to 100% apylopectin and 70% amylose, respectively, were produced through the development of new biotechnology techniques. The starches of these wheats provide the unique starch functional properties that are desirable for food and non-food industries.

Targeting Induced Local Lesions in Genomes (TILLING) population of the two winter wheat cultivars (‘Kena DS’ and ‘Gaja DS’) were developed using ethylmethane sulfonate (EMS) in order to induce mutations in genes of interest. Mutation density of one mutation per 37.84 Kb of the DNA was observed in this TILLING population while screening freezing tolerance associated genes. In this study, pilot experiment to evaluate amylose content variation in our material was performed. Twenty M3 generation genotypes of TILLING-population and its wild type cultivar ‘Kena DS’ and Waxy wheat breeding line as control were chosen to identify variation for amylose content using traditional Iodine-Potassium Iodide (I2–KI) solution. Low blue colour intensity indicating low amylose content was observed in three M3genotypes. This indicates our TILLING population will serve as a valuable source of novel alleles of starch biosynthesis genes in order to develop winter wheat cultivars with various amylose content. Further work will be: (1) to identify amylose content variation in 756 genotypes of M3 TILLING population using traditional I2–KI dyeing method; (2) to estimate precise amylose content in selected genotypes by spectrometry; (3) to make sequence analysis of key starch biosynthesis genes of selected genotypes in order to identify novel alleles.

Keywords: Starch, TILLING, *Triticum aestivum* L. Waxy wheat

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Multi-trait selection in wheat: contribution of genomic selection tools

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Genomic selection has shown its effectiveness in improving complex traits for livestock and for some crop species. It has been shown to improve genetic gain per unit of time and cost through prediction of performance before or without phenotypic characterization of new candidates. Most of genomic selection studies in plant science have focused on single-trait analyses.

The objective of my PhD program is to explore multi-trait genomic selection methods that were specially designed to exploit the information brought by correlated traits. It has been shown that prediction accuracy of traits with low heritability can be improved by a multi-trait model using correlated traits with higher heritability. In addition, prediction accuracy of difficult or expensive to measure traits can be improved by a multi-trait model using less expensive or easier to measure correlated traits with higher number of phenotypes.

A reference population of 760 lines from the INRA-AO breeding program that was evaluated for many traits between 2000 and 2016 will be analyzed as a proof of concept. It was genotyped for 180,000 SNPs developed within the Breedwheat project. This PhD program integrates with the objectives of metaprogram Selgen INRA (http://www.selgen.inra.fr/).

Keywords: multi-trait model, genomic prediction, wheat

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proWeizen – the German wheat research and breeding alliance

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Wheat is one of the most important crops and Germany is an important wheat producer. In Germany, 16 breeding companies are running independent wheat breeding programmes.

The German Wheat Research and Breeding Alliance was founded in 2012 by the German wheat breeders to combine the scientific excellence in wheat research and breeding expertise in Germany. As a public-private partnership, the proWeizen alliance acts to foster wheat breeding and research on a national and international level as well as a platform for communication and coordination. The proWeizen platform (www.proweizen.de) is equally open to scientists and companies working in wheat breeding and research.

Currently, 14 research projects, funded by the German Federal Ministry of Food and Agriculture (BMEL) as well as the German Federal Ministry of Education and Research (BMBF), are run within the proWeizen alliance and focus on breeding for yield increase and stability, better adaptation to environmental stresses and utilization of heterosis. In these projects, German universities and research institutes are working in close collaboration with wheat breeders who are vital partners and plan to implement project results in their future breeding programmes.

In addition to support in project management and coordination, proWeizen liases with wheat researchers and breeders and participates in wheat research and breeding on national and international levels, respectively. proWeizen also helps with mobilizing funding opportunities and to gain public interest and awareness of wheat research and breeding.

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BreedWheat: Breeding for sustainable wheat varieties, an integrated project from genomics to selection

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The challenge for wheat breeding is to deliver safe, high-quality, and health-promoting food and feed in a sustainable manner across environments affected by global change. In that context, the BreedWheat project gathers 28 public and private partners in research and breeding to ensure that the knowledge, resources, and methods are translated rapidly into products and varieties. BreedWheat will not only participate in new technological developments of markers and phenotyping methods, but will also integrate high throughput genotyping, phenotyping, physiological studies and modeling to decipher the genetic and ecophysiological bases of major traits for Northern Europe. This long-term project (9 years) includes the sequencing of chromosome 1B, detection of new structural genetic polymorphisms, large scale SNP production, genetic and physical mapping of those SNP, and the generation of more than 2 billion genotyping data points. A large experimental network was developed to phenotype association panels for nitrogen use efficiency, grain proteins composition, drought tolerance, Septoria leaf blotch and Fusarium head blight resistance. About 5,000 wheat lines from the INRA genetic resources centre were characterized and used to construct a new association panel. Moreover nine advanced backcross populations were created to introgress stress tolerance diversity in French elite material. Innovative statistical tools, including genomic selection, are developed within a real size selection program, in order to address both technical and socio-economic aspects of the implementation of novel breeding strategies in seed companies. Finally, a robust bioinformatics platform enabling efficient data storage and breeder friendly access is established.

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