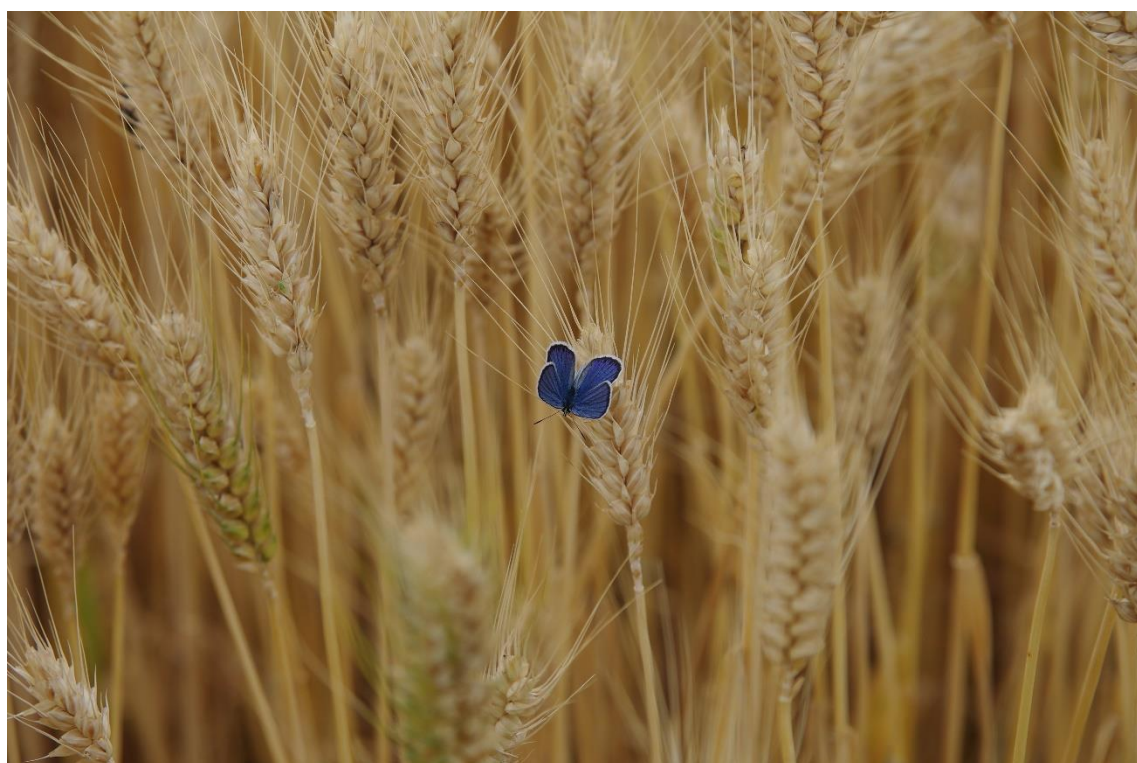


Cereal Breeding - Challenges and Opportunities for Global Improvement

Book of Abstracts of the Eucarpiia Cereals Section Conference



Edited by

Bernadett Mihály-Langó, Lajos Bona, Beáta Tóth, Andreas Börner

Szeged
2023

IMPRESSUM

Cereal Breeding - Challenges and Opportunities for Global Improvement

Book of Abstracts of the Eucarpia Cereals Section Conference

Edited by

B. Mihály-Langó, L. Bona, B. Tóth, A. Börner

Organizers of the Conference

Cereal Research Non-Profit Ltd.

EUCARPIA Cereals Section

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PROGRAM

Opening Day, 15th May Monday

12:00 – 14:00 REGISTRATION

14:00 OPENING CEREMONY, PLENARY SESSION

Session Chairs: Beata Tóth, Lajos Bona

Performance of **Mentés Másként Ensemble** - Ancient Hungarian Folk Music

- **Andreas Börner**, *President of EUCARPIA*
EUCARPIA – The European Association for Research on Plant Breeding
- **Beáta Tóth**, *Co-Chair of the Organizing Committee, Research Director of CR Ltd.*
Traditions and innovations at the 99 years old Cereal Research Non-profit Ltd.
- **Tamás Tarpataki**, *Deputy State Secretary, Ministry of Agriculture*
The importance of cereals and cereal research in the Hungarian agriculture
- **János Pauk**, *President of the Association of Hungarian Plant Breeders*
Anniversary and Moments: Introduction of the 50 years old Cereal Research Communications and the Association of Hungarian Plant Breeders

PRESS CONFERENCE and COFFEE BREAK

16:30 – 17:45 COST ASSEMBLY - Reducing acrylamide formation in cereal products

Chair: Navneet Kaur and Marianna Rakszegi

16:30-16:50 **Nigel Halford**, Naveet Kaur: Acrylamide in food: regulatory developments, progress in producing low acrylamide wheat, and the aims of the acryred cost action (ca21149)

16:50-17:10 **Navneet Kaur**, Martin Urban, Kim E. Hammond-Kosack, Nigel G. Halford: Uncoupling the signalling network linking biotic stress and free asparagine accumulation in wheat and the aims of the acryred cost action (ca21149)

17:10-17:30 **Marianna Rakszegi**, Viktor Korzun, Nigel Halford: Does it make sense to breed low-asparagine cereals?

17:30-17:45 DISCUSSION, CONCLUSIONS OF THE ASSEMBLY OF REDUCING ACRYLAMIDE FORMATION IN CEREAL PRODUCTS

19:00-22:00 WELCOME RECEPTION

16th May Tuesday

09:00 – 13:00 SESSION 1 – Genetic resources and pre-breeding

Chair: Gyula Vida

09:00-09:40 **KEYNOTE Friedrich Longin**, Jochen C. Rief: How to design an efficient prebreeding program in wheat

9:40-10:10 **Magdalena Lunzer**, Maria Buerstmayr, Iris Fallbacher, Almuth E. Müllner, Heinrich Grausgruber, Hermann Buerstmayr: Identification of a major common bunt resistance QTL in mapping populations descending from PI-166910

10:10-10:30 Eszter Gaál, **István Molnár**, András Farkas, László Ivanizs, Balázs Kalapos, Edina Türkösi, Klaudia Kruppa, Éva Szakács, Mahmoud Said, Abraham Korol, Jaroslav Doležel, Miroslav Valárik: Characterization of *Aegilops biuncialis* introgressions in a wheat prebreeding population using DArTseq genotyping

10:30-10:50 **Galyna Chebotar**, Andreas Börner, Manuela Nagel: Genome-wide association study of viability in barley stored under ambient conditions

10:50-11:10 **Eva Johansson**, Lan Yuzhou, Mahboobeh Yazdani, Rimsha Ashraf, Olawale Olalekan, Ramune Kuktaite, Aakash Chawade, Elaine Ceresino, Mahubjon Rahmatov: Diverse wheat lines as sources to breed for resistance and tolerance to emerging climate change related abiotic and biotic stresses

11:10 – 11:30 COFFEE BREAK

11:30-11:50 **Csaba Lantos**, Ferenc Békés, Katalin Ács, Bernadett Mihály-Langó, Róbert Mihály, László Cseuz, Mihály Jancsó, Árpád Székely, János Pauk: Development and Application of in vitro anther culture in cereal breeding of CR LTD.

11:50-12:10 **Matías Schierenbeck**, Ahmad M. Alqudah, Edit Lantos, Evangelina G. Avogadro, María Rosa Simón, Andreas Börner: Green Revolution dwarfing Rht genes affect floral traits related to cross pollination efficiency

12:10-12:30 **László Purnhauser**, Tungalag Munkhbat, Tar Melinda: Identification of stem, leaf and yellow rust resistance genes in Hungarian wheat cultivars

Flash presentations

12:30-12:35 **Ajit Nehe**, Aakash Chawade: Genome-wide association analyses identify key QTLs for seedling root system architectural traits in Nordic-adapted barley collection

12:35-12:40 Eszter Gaál, **Mahmoud Said**, Adoración Cabrera, Éva Szakács, András Farkas, László Ivanizs, Edina Türkösi, Takashi R. Endo, Jaroslav Doležel, István Molnár: Widening the gene pool of wheat by crested wheatgrass using the gametocidal system

E-posters

P-1 Alena Hanzalová, Vojtěch Holubec: Evaluation of the representatives of the genus *Aegilops* in the Czech Republic

P-2 László Ivanizs, András Farkas, Eszter Gaál, Edina Türkösi, Klaudia Kruppa, Éva Szakács, Luigi Cattivelli, Alessandro Tondelli, István Molnár: Detailed exploration of genetic diversity in *Aegilops Biuncialis* using DArTseq genotyping

P-3 Andrea Horváthné Uhrin, Gyula Vida, Mónika Károlyiné Cséplő, Péter Mikó: Testing *Triticum x dasypyrum villosum* hybrids using molecular markers

P-4 Paulina Bolc, Dorota Dziubińska: Phenotyping as part of the implementation of the AGENT project in the Polish gene bank

P-5 Anna Hawliczek, Fatemeh Shoormij, Hilderlith Awuor Abuya, David Chan Rodriguez, **Hanna Bolibok-Brągoszewska**: Sequence diversity of Candidate genes for Selection in rye (*Secale cereale* L.)

P-6 Márton György, Viola Tóth, Ottó Veisz, Péter Mikó, Marianna Rakszegi, Gyula Vida, Balázs Varga: New small grain cereals bred in Martonvásár widening the offered species Sortiment

P-7 Alejandra Cabeza, Beatriz Larruy, Vanesa Martínez-Agudo, Asunción Costar, Ana M. Casas, Ernesto Igartua: A QTL for dry matter partitioning found in seedlings of a local RIL population of barley

P-8 Ana M. Casas, Francesc Montardit-Tarda, Bruno Contreras-Moreira, Davide Guerra, Agostino Fricano, Alessandro Tondelli, Andrea Visioni, Ernesto Igartua: GENDIBAR: a collection of Mediterranean barley landraces for cereal adaptation studies

13:00 – 14:00 LUNCH

14:00 – 17:45 SESSION 2 – Grain yield and quality
Chair: Bernadett Mihály-Langó

14:00-14:40 **KEYNOTE Frank Bekes**: Present situation and future trends in quality related cereal chemistry and technology

14:40-15:00 **Mehmet Tekin**, Aytekin Aksoy, Taner Akar: Diversity trends of agronomic and quality traits in various old and modern wheat varieties cultivated under irrigated conditions of Türkiye

15:00-15:20 **Luca Bonfiglioli**, Ieva Urbanavičiūtė, Mario A. Pagnotta: Breeding durum wheat for organic farming

15:20-15:40 **Marianna Rakszegi**, Viola Tóth, Verica Takac, László Láng, Sanja Mikic, Milan Mirosavljevic, Lovro Sinkovič, Barbara Pipan, Sándor Tömösközi, Gyula Vida, Péter Mikó: Compositional and quality characterization of spelt wheat

15:40 – 16:10 COFFEE BREAK

16:10-16:30 **Lisa Call**, Stefano D'Amico, Heinrich Grausgruber: Neglected wheat as a source of low-ATI food products?

16:30-16:50 **Anna Nowicka**, Zuzana Pobořilová, Kateřina Navratilová, Martin Kovačik, Edina Türkösi, István Molnár, Aleš Pečinka: Parental-dosage-dependent effects on barley seed development

Flash presentations

16:50-16:55 **Lajos Bona**, Szandra Purgel, Róbert Mihály, Bernadett Mihály-Langó: Triticale grain quality traits: are there enough variation for selection?

16:55-17:00 **Salvatore Esposito**, Palombieri Samuela, Vitale Paolo, Taranto Francesca, Sestili Francesco, De Vita Pasquale: A new MYB gene in durum wheat genome as candidate for Pp-B1

17:00-17:05 **Zaiga Jansone**, Māra Bleidere, Andris Lapāns, Vija Strazdiņa, Valentina Fetere, Ligita Šalkovska: Relationship between grain yield and spectral reflectance indices in spring wheat at various growth stages

17:05-17:10 **Katalin Acs**, Csaba Lantos, Annamaria Polgar, Bernadett Mihaly-Lango, Ferenc Bekes, Janos Pauk: Developing spelt genotypes for functional dietetic usage: a possible solution for FODMAP problem

17:10-17:15 **Jolanta Groszyk**, Mateusz Przyborowski, Marta Puchta-Jasińska, Paulina Bolc: Project R-GRAIN - identification of the molecular traits determining the rye yielding and identification of effective molecular markers

17:15-17:20 **Olawale Olalekan**, Mahbubjon Rahmatov, Elaine Ceresino, Eva Johansson: Genetic diversity in old and alien wheat genotypes for nutritionally important traits

17:20-17:25 **Mahboobeh Yazdani**, Mahbubjon Rahmatov, Eva Johansson: Protein quality in 2BS.2BL-2RL wheat-rye translocation lines with resistance to novel stem rust races

E-posters

P-9 **Stanislav Ježek**, Pavel Horčíčka, Ondřej Veškna, Ema Holavová, Irena Bížová: Baking quality of wheat varieties in the Ecobreed project

17:25-17:45 SUMMARY, CONCLUSIONS OF THE SESSIONS OF THE DAY - Session Chairs and Keynotes

17th May, Wednesday

09:00 – 11:45 SESSION 3 – Biotic stress response
Chair: Peter Mikó

09:00-09:40 **KEYNOTE Akos Mesterhazy**: How to supply 11 billion people with food? Reduction of losses in the cereal production chain, where are the hot spots and what can be done?

9:40-10:00 **Jonas Anderegg**, Radek Zenkl, Achim Walter, Andreas Hund, Bruce A. McDonald: Separating disease and senescence in wheat canopies using deep learning and dynamic modelling

10:00-10:20 **Diminika Piaskowska**, Urszula Piechota, Magdalena Radecka-Janusik, Piotr Słowacki, Paweł Czembor: Efficiency of the molecular markers linked to the STB resistance loci in wheat

10:20-10:40 **Attila Berényi**, Balázs Szabó, Tamás Mészleányi, Csaba Vágvölgyi, Beáta Tóth: Fungal infection and toxin contamination of small grain cereals in Hungary

10:40-11:00 **Renata Orłowska**, Janusz Zimny, Jacek Zebrowski, Piotr Androsiuk, Piotr T. Bednarek: Metabolomic changes as one of the underappreciated sources of tissue culture-induced variation in triticale

Flash presentations

11:00-11:05 **Klára Mészáros**, Jeny Jose, Antónia Szakács, Viola Kunos, Mónika Cséplő, Judit Bányai, Magda Pál, József Bakonyi, Csaba Éva: The role of ORA59 transcription factor in the hormone-mediated defence mechanism during *Pyrenophora teres* f. *teres* infection in barley

11:05-11:10 **Agnieszka Niedziela**, Krystyna Rybka, Dariusz Mańkowski, Rafał Marcinkowski, Przemysław Matysik: Selection of winter wheat forms with increased tolerance to periodic water shortages

11:10-11:15 **Dániel Nagy**, János Pauk, Judit Óvári, László Cseuz: Reactions of Szeged Wheat Varieties to Drought Stress

11:15-11:20 **Piotr Słowacki**, Urszula Piechota, Magdalena Radecka-Janusik, Dominika Piaskowska, Paweł Czembor: Assessing stripe rust (*Puccinia striiformis* f. sp. *tritici*) population structure using molecular methods

E-posters

P-10 Valentina Spanic, Ana Vukovic, Monika Cseplo, Rosemary Vukovic, Katalin Puskas, Thomas Roitsch: Physiological changes as early response in infected winter wheat (*Triticum aestivum* L.) seedlings with leaf rust

P-11 Mateusz Matuszkiewicz, Święcicka M, Szewińska J, Wakuliński W, Rakoczy-Trojanowska M: Does ScLr67APR gene confers seedling stage resistance of rye to leaf rust?

P-12 Anett Klaudia Kovács, Eszter Csepregi-Heilmann, Ágnes Áldott-Sipos, Katalin Körösi, Tamás Spitkó, Magda Pál, Csaba Szőke: Changes in polyamine concentration in maize seedlings due to Fusarium infection

P-13 Mateusz Przyborowski, Joanna Chojak-Koźniewska, Sebastian Gasparis, Anna Nadolska-Orczyk: The effect of long-term medium drought on the development of wheat (*Triticum aestivum* L.) grains

P-14 Iryna Zambriborshch, Shestopal O.L., Traskovetskaya V.A., Vasiliev O.A., Babayants L.T.: Obtaining dihaploid lines of soft winter wheat with complex resistance to rust and common bunt by anther culture *in vitro*

P-15 **Lidiya Mishchenko**, Rahmatov M., Mishchenko I., Molodchenkova O., Motsnyi I., Dunich A., Dashchenko A.: Increasing of wheat resiliency to viral and fungal diseases under climate change conditions

P-16 **Ivan Motsnyi**, Rahmatov M., Sokolov V., Molodchenkova O.O., Lytvynenko M., Mishchenko L., Fait V., Halaiev O., Zambriborshch I., Fanin Ya.S.: An evaluation of disease resistance, drought tolerance, and agronomic traits in winter wheat introgression lines

P-17 Saulyak N. I., **Oleksii Vasiliev**, Traskovetska V. A., Saulyak N. I., Rudenko V. A., Bushulyan M. A.: Genetic basis of resistance of winter wheat to major foliar diseases

P-18 **Yevhenii Kirchuk**, Alieksiienko Ye.V.: Efficiency of different genetic systems of resistance to leaf rust in the steppe zone of Ukraine

11:20 – 12:00 CEREALS SECTION ASSEMBLY – Chaired by Andreas Börner

12:15 – 13:15 LUNCH

13:15 – 17:00 SESSION 4 – Environmental adaptation, crop management

Chair: Borislav Kobiljski

13:15-13:55 **KEYNOTE Geert Haesaert**, Veerle Derycke, Szanne Degraeve, Sofie Landschoot, Riccardo Zustovi, Anneleen De Zutter, Kevin Dewitte: Breeding triticale for more sustainability in our cropping systems

13:55-14:15 **Lukas Roth**, Achim Walter: Using field-based phenotyping to explore the temperature responses of growth: From overgeneralising to overfitting of domain knowledge in crop models?

14:15-14:35 **Lucas Kronenberg**, Oscar E. Gonzalez-Navarro, Michelle Leverington, Sarah Collier, Luzie U. Wingen, Simon Griffiths: Dissection of phenology and its effects on yield in a Buster-charger mapping population in the field

14:35-14:55 **Mesut Keser**, Umran Küçüközdemir, Beyhan Akın, Fatih Özdemir: The effect of Vernalization genes on cold tolerance in diverse wheat germplasm

14:55-15:15 **Flavian Tschurr**, Norbert Kirchgessner, Andreas Hund, Lukas Kronenberg, Jonas Anderegg, Achim Walter, Lukas Roth: Frost damage index: The antipode of growing degree days

15:15– 15:45 COFFEE BREAK

15:45-15:05 **Tesfaye Walle Mekonnen**, Angeline van Biljon, Maryke Labuschagne: The impact of different planting dates on protein quality and quantity and grain yield of maize

15:05-16:25 **Péter Borbély**, Zahra Tahmasebi, Tamás Pálmai, Mohamed Ahres, Orsolya Kinga Gondor, Gábor Galiba: Far-red and blue light Enrichment differentially influence the metabolism and frost tolerance of barley

Flash presentations

16:25-16:30 **Aziza Zerrouk**, Ana M. Casas, Ernesto Igartua: Effects of PPDH1 and VRNH2 on yield and development of spring barley

16:30-16:35 Beáta Strejčková, Kateřina Kaduchová, Radim Čegan, Romana Nesnadná, Aleš Pečinka, Jan Bartoš, **Jan Šafář**: Structural variability and function of *VERNALIZATION1* in cereals

16:35-16:40 **Māra Bleidere**, Zaiga Jansone, Andris Lapāns, Vija Strazdiņa, Valentīna Fetere, Ligita Šalkovska: Results of UAV-based phenotyping for spring wheat genotypes across contrasting growing conditions

E-posters

P-19 **Ondřej Veškrna**, Stanislav Ježek, Pavel Horčíčka, Jan Humplík: Evaluation of frost resistance using image analysis

P-20 Asmaa A. M. Ahmed, Mona F. A. Dawood, Ameer Elfarash, Elsayed A. Mohamed, Mohamed Y. Hussein, **Andreas Börner**, and Ahmed Sallam: Genetic variation in important physiological responses and their effects to alleviate drought stress in wheat seedlings

P-21 **Mónika Cséplő**, Katalin Puskás, Gyula Vida, Judit Bányai, Klára Mészáros, Andrea Uhrin, Viola Tóth, Heinrich Grausgruber, Luca Bonfiglioli, Mario Augusto Pagnotta, Péter Mikó: Results of multi-year durum wheat variety test carried out in different management systems

P-22 **Yaroslav Fanin**, Molodchenkova O.O., Lytvynenko M.A.: Influence of genotypes on nitrogen accumulation and reutilization in winter bread wheat

P-23 **Martyna Bodzak**, Michał Kempa, Krzysztof Mikołajczak, Piotr Ogrodowicz, Monika Mokrzycka, Paweł Krajewski, Anetta Kuczyńska: Effect of melatonin on hormonal regulation in barley root under drought stress

P-24 **Gallia Butnaru**, Ioan Sarac, Dorin Tarziu: Adaptation of local and foreign einkorn genotypes on chernozem, solonetz and luvisoil from Banat County

16:45-17:00 SUMMARY, CONCLUSIONS OF THE SESSIONS OF THE DAY - Session Chairs and Keynotes

19:00-20:00 EVENING ORGAN CONCERT

St. Elizabeth Church, Torontál square, Szeged, Artist: Levente Kuzma

18th May, Thursday

09:00 – 11:50 SESSION 5 – Molecular approaches, bioinformatics and genomic selection
Chair: Janos Pauk

09:00-09:40 **KEYNOTE Luigi Cattivelli**: Advanced genomic tools for leveraging durum wheat diversity

9:40-10:00 **Hermann Gregor Dallinger**, Franziska Löschenberger, Naim Azrak, Christian Ametz, Sebastian Michel, Hermann Bürstmayr: Genome-wide association mapping for pre-harvest sprouting in European winter wheat

10:00-10:20 **Mohammed Ali**, Dávid Polgári, Adél Sepsi, Levente Kontra, Ágnes Dalmadi, Zoltán Havelda, László Sági, András Kis: Rapid and cost-effective karyotyping in wheat x barley hybrids by chromosome-specific multiplex PCR

10:20-10:40 **Olivia Zumsteg**, Flavian Tschurr, Steven Yates, Dario Fossati, Flavio Foiada, Lukas Roth, Andreas Hund: Genomic Prediction of traits related to phenology and temperature-response during stem elongation of wheat

10:40-11:00 **Maciej Majka**, Eva Janáková, Irena Jakobson, Kadri Järve, Petr Cápál, Zuzana Korchanová, Adam Lampar, Jakub Juračka, Miroslav Valárik: The chromatin determinants and Ph1 gene effect at wheat sites with contrasting recombination frequency

11:00-11:20 **Johanna Åstrand**, Sylvain Poque, Kristiina Himanen, Aakash Chawade: Evaluating drought tolerance in spring barley by high throughput phenotyping and genome-wide association analysis

Flash presentations

11:20-11:25 **Admas Alemu**, Lorena Batista, Pawan K. Singh, Alf Ceplitis, Aakash Chawade: Haplotype-based genomic prediction for Fusarium head blight resistance and yield-related traits in wheat

11:25-11:30 **Guoliang Li**, Yusheng Zhao, Albert Wilhelm Schulthess, Jochen C. Reif, Yong Jiang: An efficient algorithm for mapping heterotic quantitative trait loci with whole genome sequencing data

11:30-11:35 **Salvatore Esposito**, Vitale P, Taranto F, Saia S, Pecorella I, D'Agostino N, Rodriguez M, Natoli V, De Vita P: Simultaneous improving of durum wheat yield and grain protein content by using association tests and weighted GBLUP

11:35-11:40 **David Chan-Rodriguez**, Aleksandra Paradowska, Magdalena Świącicka, Brian Wakimwayi Koboyi, Anna Hawliczek1, Leszek Bolibok, Maksymilian Królik, Hilderlith Abuya, Hanna Bolibok-Braęoszewska: Identification and expression profiling of rye phosphorus-homeostasis-related genes

E-posters

P-25 Ádám D. Horváth, Tibor Kiss, András Cseh, Ádám Horváth, Zita Berki, Krisztina Balla, Ildikó Karsai: Daily expression patterns of major plant developmental genes in hexaploid wheat under inductive conditions of plant growth

P-26 Péter Kovács, András Farkas, Eszter Gaáll, László Ivanizs, Kitti Szőke-Pázsí, Edina Türkösi, Klaudia Kruppa, Éva Szakács, István Molnár: Gene introgressions into wheat from *Aegilops comosa* and *Aegilops umbellulata*

P-27 Rumana Azad, Bartosz Biernacik, Mateusz Olechowski, Alperen Ozturk, Monika Rakoczy-Trojanowska: Expression of R-genes in response to brown rust caused by Prs strains in rye (*Secale cereale* L.) inbred lines through RT-qPCR

P-28 Tibor Kiss, Ádám D. Horváth, András Cseh, Ádám Horváth, Zita Berki, Krisztina Balla, Ildikó Karsai: Interactions of major flowering pathway genes in hexaploid wheat under inductive conditions of flowering

P-29 Marta Puchta-Jasińska: Profiling of sRNA expression in germinating barley seeds maintained in long-term storage

P-30 Magdalena Świącicka, Gniazdowska D, Rakoczy-Trojanowska M: RT-qPCR verification of gene expression in rye upon leaf rust infection

P-31 Oleksii Halaiev, Halaieva M., Rahmatov M.: Identification and distribution of Ne1 and Ne2 hybrid necrosis gene alleles in Ukrainian bread wheat varieties

P-32 Sabina V. Chebotar, Toporash M.K., Motsnyi I.I., Blagodarova O.M.: Analysis with molecular markers wheat lines with novel types of translocations 1RS.1BL at the prebreeding stage

P-33 Popovych Yu. A., Blagodarova O. M., **Sabina V. Chebotar**: Correspondence between alleles of *Gli-1* loci and the allelic variants of gliadins of *Triticum aestivum* L.

12:00 – 13:00 LUNCH

13:00 – 17:00 SESSION 6 – Future challenges and innovations

Chair: Andreas Börner

13:00-13:40 **KEYNOTE Viktor Korzun**: Bridging Research and Plant Breeding for a Sustainable Agriculture: current achievements and future challenges in cereals

13:40-14:00 **Sebastian Michel**, Franziska Löschenberger, Christian Ametz, Herbert Bistrich, Hermann Bürstmayr: Can we teach A.I. to select like a plant breeder?

14:00-14:20 **Heinrich Grausgruber**, Nadine Bauer, Daniel Cristina, Matilda Ciucă, Pavol Hauptvogel, Bojan Jocković, Cristina-Mihaela Marinciu, Marianna Mayer, Péter Mikó, Primoz Titan, Ondrej Veskrna: Breeding wheat for organic farming – lessons learnt from the ECOBREED project

14:20-14:40 **Szilvia Bencze**, Ferenc Bakos, Péter Mikó, Mihály Földi, Magdaléna Lacko-Bartošová, Nuri Nurlaila Setiawan, Dóra Drexler: Emmer and einkorn reloaded – the pesticide-free crop alternatives

14:40 – 15:00 COFFEE BREAK

15:00-15:20 **Jana Palicova**, Jana Chrpova, Anna Tobolkova, Jaroslava Ovesna, Milena Stranska: Effect of Pulsed electric field on viability of fusarium micromycetes

15:20-15:40 **Borislav Kobiljski**, Gojko Mladenović, Zorica Jestrović, Katarina Kobiljski: Novel wheat breeding approach to counter the climate challenges and biodiversity loss

Flash presentations

15:40-15:45 **Péter Mikó**, Mihály Földi, Mária Megyeri, Vida Gyula, Dóra Drexler: The Hungarian participatory organic wheat breeding program

15:45-15:50 **Maja Boczkowska**: Science beyond jars

15:50-15:55 **Fabio Fania**, Patrizio Spadanuda, Ivano Pecorella, Nicola Pecchioni, Pasquale De Vita: Field-based canopy height estimation using UAV images improves

15:55-16:00 **Mustafa Zakieh**, David S. Gaikpa, Fernanda Leiva Sandoval, Marwan Alamrani, Tina Henriksson, Firuz Odilbekov, Aakash Chawade: Accelerated growth conditions for advancing winter wheat resistance breeding against fusarium head blight

16:00-16:05 **Zsuzsanna Farkas**, Ottó Veisz, Balázs Varga: Can the rising atmospheric CO₂ concentration mitigate the severe drought damages?

E-posters

P-34 **Vishnukiran Thuraga**, Beáta Strejčková, Ajit Nehe, Linda Öhlund, Aakash Chawade: Phenotrolley: Affordable phenotyping system for controlled growth conditions

P-35 **Judit Bányai**, György Balassa, Richárd Turbéli, József Bakonyi, Mónika Cséplő, Klára Mészáros, Ottó Veisz, Péter Mikó: Application of drone-based-high-throughput-phenotyping system in cereal breeding

P-36 **Michał Kempa**, Martyna Bodzak, Krzysztof Mikołajczak, Piotr Ogrodowicz, Anetta Kuczyńska: High-throughput automatic phenotyping of barley under drought stress

P-37 **Krzysztof Michalski**, Sławomir Sowa: Cas9-expressing Cell suspensions for grna evaluation: optimization of construct delivery via electroporation

16:10-17:00 SUMMARY, CONCLUSIONS OF THE SESSIONS OF THE DAY - Session Chairs and Keynotes

CLOSING REMARKS OF SCIENTIFIC PARTS - Andreas Börner

19:00 GALA - DINNER & DANCE-HOUSE - Traditional Hungarian Round Dance
Fortress Museum and Stone Collection, Stefánia Street 2. (Downtown)

19th May, Friday

09:30 EXCURSION: FIELD EXPERIMENTS

Buses start from the Hotel Forrás at 9.30 a.m., lead by Lajos Bona and Laszlo Cseuz

Lunch at the Hódagro Farm

20th May, Saturday

09:30 SATELLITE EXCURSION TO GODOLLO

Bus starts from the Hotel Forrás at 9.30 a.m., lead by Janos Pauk

Lunch in Gödöllő

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PLENARY SESSION

EUCARPIA – THE EUROPEAN ASSOCIATION FOR RESEARCH ON PLANT BREEDING

Abdreas Börner – President of EUCARPIA

Leibniz Institute of Plant Genetics and Crop Plant Research (IPK),
Gatersleben, Germany

EUCARPIA aims to promote scientific and technical co-operation in the field of plant breeding in order to foster its further development. To achieve this purpose, the association arranges and sponsors meetings of members, to discuss general or specific problems. The association was founded in 1956. It has about 600 individual and 60 corporate members with about 400 delegates nominated.

EUCARPIA consists of 11 Sections divided into two categories:

Crop specific sections:

Potatoes
Cereals
Fodder Crops and Amenity Grasses
Maize and Sorghum
Vegetables
Fruits
Ornamentals
Oil and Protein Crops

Thematic sections:

Biometrics in Plant Breeding
Genetic Resources
Organic and Low-Input Agriculture

Members can participate in any/several sections of their interest. They will receive mailings about upcoming events of their interest. Further details about EUCARPIA you can find on the webpage: www.eucarpia.eu.

EUCARPIA offers a one-year free membership for the participants of the Cereals Section Conference in Szeged. Please register using the coupon code: CSC2023.

**TRADITIONS AND INNOVATIONS AT THE 99 YEARS OLD CEREAL RESEARCH
NON-PROFIT LTD.**

Beáta Tóth, Attila D. Mandák

Cereal Research Non-profit Ltd., Szeged, Hungary

Our institution's predecessor, the Hungarian Royal Agricultural Institute of the Great Planes, which later was called the Cereal Research Institute, was founded in 1924 in Szeged, Hungary. The experience in agricultural research accumulated during decades, combined with the recent research efforts opens up the possibility regularly to bring new plant varieties and hybrids to the market. Presently our company is one of the most significant breeding centers of cereals, oil and protein crops in Hungary under the aegis of the Hungarian University of Agriculture and Life Sciences (MATE). We contribute to the Hungarian crop production with more than 180 registered plant varieties and hybrids of 23 crop species. The total growing area of our cultivars and hybrids domestically and abroad covers hundreds of thousands of hectares annually. Field trials and breeder's seed multiplication can be accomplished on 1800 hectares of land at own management. In our R&D activity, wheat, triticale, barley, grain sorghum, soybean, linseed are the main species. We are market leaders and/or significant players in the domestic seed supply chain of these crops. Breeding, marketing, seed production and trade is based on a network of more than 200 domestic partners and 50 foreign companies and institutions embracing the research and market sphere.

The target of the small grain cereal breeders in Szeged is to develop bread wheat, durum wheat, spelt, triticale, barley, oats and rye varieties delivering high and stable yield and exhibiting superior quality parameters and to elaborate their production techniques. Crop production was risked by extreme climatic conditions due to global warming during the past decade. Our ultimate goal is to produce new cultivars with improved level of tolerance to biotic and abiotic stresses. Certain research areas have crucial importance within the activities, like breeding for resistance to diseases, analysis of the relation between qualitative traits, the question of fungicide application and environmental protection, the enhancement of conventional breeding work by the results of biotechnology and molecular genetics. In addition to analyze the quality of flour, protein and oil as usual, an up-to-date laboratory has been equipped to investigate the toxin content of the grains infected with *Fusarium* and other different toxin producing fungi. Patients suffering from celiac disease, PKU or renal failure can rely on the dietetic food invented at the Company. Triticale, buckwheat and soybean breeding has also been motivated by focusing on healthy food.

Our research workers participate both in the domestic and foreign scientific public life, as well as in the work of the Hungarian Academy of Sciences. Colleagues are either consortium leaders or co-operators in joint projects with foreign partners, publish in prominent R&D periodicals worldwide.

Research work at the nearly 100 years old institute is done consequently and concentrating on the constantly changing demands in order to offer the best varieties and hybrids to our customers from more than 20 countries, year by year.

**ANNIVERSARY AND MOMENTS: INTRODUCTION OF THE 50 YEARS OLD
CEREAL RESEARCH COMMUNICATIONS AND THE ASSOCIATION OF
HUNGARIAN PLANT BREEDERS**

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Establishing Cereal Research Communications was the late academician's Zoltán Barabás' idea, and the foundation of the journal happened fifty years ago. Today, we cannot even imagine how innovative this idea was at that time, we can even say it was revolutionary. Perhaps it seemed impossible under the given historical circumstances, but Zoltán succeeded. Right in the middle of the socialist system, he invited excellent researchers from all over the world to be the members of the editorial board. Even today, it would be very difficult to bring together such scientists: J. Bojanowski, E. Borlaug, A. Bozzini, G. Kimber, Á. Kiss, K. Kristev, P.P. Lukyanenko, J.F. Mackey, R.A. McIntosh, M. Okamoto. The Editor-in-Chief was Zoltán Barabás, who at that time still had the title of technical editor.

From the very beginning, the main goal of the Journal was to provide an opportunity to publish the results of genetic, breeding, agrotechnical, physiological and disease-related research on the most important cereals (wheat, corn, barley, oats, triticale, sorghum). The goals have not changed since then. The career of the journal had begun with free distribution, but it has been available by subscription for a long time. In the past, the paper could be seen from afar on the shelves of libraries, today the appearance is not so attractive in the online world, but the number of online subscriptions and downloads is growing every year. The impact factor in 2021 reached the 1.240 value. CRC now belongs to Springer family, which contributes to its growing popularity. Nowadays, it is already one of the most important breeding journals. In the past fifty years, the journal has had only four editors-in-chief: Zoltán Barabás, Zoltán Kertész, János Pauk and currently Andreas Börner from Germany. The Editor-in-Chief's work is supported by two technical editors: Csaba Lantos and Kornélia Hajduné Buza, they still work in founding town Szeged, Hungary. Regarding the journal's appearance, it has been published with three types of front pages so far, but the current one, which is in A4 format, with great technology and colorful illustrations, makes the paper really attractive. We wish everyone good luck publishing in Cereal Research Communications.

The Hungarian Plant Breeders' Association is a non-profit organization devoted to the support of the art, science, career, work and business of plant breeding as a profession via assistance and communication among professionals in the field of breeding, seed industry, agronomy, and other related areas. There are cca. 250 members from the different areas and sectors. We emphasize importance of breeders' rights as well as the cooperation among the concerned disciplines maintaining the ethical standards, thereby we are also open to cooperate with plant breeders from European and other countries as well.

COST ASSEMBLY

Reducing acrylamide formation in cereal products

ACRYLAMIDE IN FOOD: REGULATORY DEVELOPMENTS, PROGRESS IN PRODUCING LOW ACRYLAMIDE WHEAT, AND THE AIMS OF THE ACRYRED COST ACTION (CA21149)

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Acrylamide is a carcinogenic processing contaminant that forms from free (non-protein) asparagine and reducing sugars during high-temperature cooking and processing of grains, tubers, beans, storage roots and other plant products. Food businesses face ever-tightening regulations on acrylamide, particularly in the European Union, with the prospect of Maximum Levels being set for some foods this year. Compliance would be greatly facilitated by the development of crop varieties with reduced acrylamide-forming potential. We have used CRISPR/Cas9 to knock out the asparagine synthetase-1 and -2 (*TaASN1* and *TaASN2*) genes of bread wheat. Wheat lines with mutations in the *TaASN2* gene have also been produced by chemical mutagenesis, and we have also assessed the effect of a natural deletion that means that the B genome *TaASN2* gene (*TaASN-B2*) is missing in some varieties. A field trial of *TaASN2* knockouts produced using CRISPR/Cas9 and chemical mutagenesis (the first field trial of CRISPR wheat in Europe) was harvested in August 2022, with the CRISPR lines showing significant reductions in free asparagine concentration and a concomitant decrease in acrylamide formation in heated flour. A second field trial including *TaASN1/TaASN2* knockouts is underway. These and other genetic approaches are being undertaken alongside experiments on crop management strategies, including fertilisation rates, especially of sulphur, and disease control. Networking between researchers, breeders and food industry experts on the acrylamide issue is being promoted by a European COST Action, Acryred (CA21149), the aims and activities of which will be presented.

Keywords: Acrylamide; Asparagine; CRISPR; Food safety; Acryred COST Action

UNCOUPLING THE SIGNALLING NETWORK LINKING BIOTIC STRESS AND FREE ASPARAGINE ACCUMULATION IN WHEAT, AND THE AIMS OF THE ACRYRED COST ACTION (CA21149)

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In many plant species, asparagine plays a central role in nitrogen storage and transport. However, asparagine also acts as a precursor for acrylamide, a Class 2A carcinogen. Free (soluble, non-protein) asparagine accumulates in high concentrations during seed germination as well as under abiotic/biotic stresses. In wheat, fusarium head blight disease, caused by *F. graminearum* (*Fg*), reduces yield, and contaminates grains with mycotoxins, e.g., DON. DON treatment in wheat increases free asparagine, glutamine, and aspartate concentration (Warth *et al.*, 2015). SnRK1-type protein kinases are key regulators of plant metabolism and defence. During infection, *Fg* secreted orphan protein, OSP24, interacts with SnRK1 and accelerates its degradation. Meanwhile, wheat orphan protein, TaFROG, competes with *Fg*OSP24 to prevent SnRK1 degradation (Jiang *et al.*, 2020). However, further signalling proteins/transcription factors leading to free asparagine accumulation are still unknown. The current study will elucidate this network using *Fg* mutants, RNA-seq analysis, phosphoproteomics and proximity labelling. VIGS will then be used to validate our model of the signalling hub. Networking between researchers, breeders and food industry experts on the acrylamide issue is being promoted by a European COST Action, Acryred (CA21149), the aims and activities of which will be presented.

Keywords: Acrylamide; Biotic stress; *Fusarium graminearum*; Asparagine; Acryred COST Action

DOES IT MAKE SENSE TO BREED LOW-ASPARAGINE CEREALS?

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The concentration of individual free amino acids in cereal grains varies depending on species and fractions, with asparagine being one of the most abundant. Free asparagine reacts with reducing sugars to form the processing contaminant, acrylamide, in cereal products through the Maillard reaction, promoted by high temperature and low moisture content. The acrylamide monomer is both neurotoxic and carcinogenic. Thus, the European Commission issued Benchmark Levels and codes of practice for the food products in 2017. The presence of acrylamide in food is now a difficult regulatory compliance issue for the food industry.

Cereal breeders could contribute to the reduction of acrylamide level in food by reducing the free asparagine level in cereals. A variation of 0.56–11.82 mmol/kg of free asparagine was measured in different studies on 25–270 wheat varieties grown on 3–6 sites. Variation was also found among different cereal species, with rye having the largest values and the widest variation. Although a high variation was found in asparagine concentration, no genetic resources have so far been identified for breeding purposes.

The most frequent heritability value of free asparagine in wheat was 0.6 (0.13–0.94) in different studies, showing the relevance of breeding for this trait. Nine QTLs were identified for free asparagine concentration, but with 5A chromosome being the most determinant in one study and 7B in another. From the enzymes involved in asparagine metabolism, asparagine synthetase was the most widely studied and genes were identified in different species, but so far, no molecular markers suitable for marker-assisted selection have been developed. Efforts have also been carried out to develop fast, cost-effective methods for measuring free asparagine concentration in cereals but without a realistic solution.

Environmental factors and field management practices can also influence the asparagine level in cereals. Asparagine increased with a delayed harvest, more solar radiation and disease pressure, while the sulphur supply, fungicide treatment or organic farming reduced it in cereal grain. The latter might also be related to the smaller kernel size, harder grain texture and lower protein content of the seed. This may also have contributed to the failure of the selection of the appropriate gene resource.

The working group WG2 „Agronomy and cereal breeding” in COST ACRYRED Action (CA21149) aims to assess diversity of free asparagine in cereal species and cultivars and to identify those having low content. The understanding of the pathways for accumulation of free asparagine and identification of efficient ways for reduction of free asparagine concentrations in cereal grains through plant breeding including genetic tools, cereal production and/or field management practices is an important aspect, and developing fast techniques for screening free asparagine level in cereals is also unsolved. Providing recommendations for farmers and food industry is also of interest.

Keywords: ACRYRED, asparagine, breeding, cereals, COST

SESSION 1

Genetic resources and pre-breeding

HOW TO DESIGN AN EFFICIENT PREBREEDING PROGRAM IN WHEAT

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Genetic resources cannot be used directly in elite breeding programs due to their poor performance in at least few traits important for the market. Thus, our aim was to establish a prebreeding program delivering new lines, which perform that good, that an elite breeder can directly use them in elite crosses. Based on theory and adapted throughout 6 years of practical experience, we use the following strategy. Parental choice regarding genetic resources is done by their breeding value in hybrid combinations with elite lines. Elite crossing parents must have a high yield, high disease resistance and good quality. As crossing scheme, a four-way cross is used with three elite lines and one genetic resource to have roughly 25% genetic resource in the offspring. Thereafter, we apply pedigree-based breeding scheme with strict and multigeneration early selection based on disease resistance, plant height and adaptation followed by two years intensive yield testing in multiple locations. All prebreeding lines are genotyped at that stage for major QTL markers commonly used in wheat breeding. Furthermore, the genome contribution of the genetic resource within the prebreeding line is tracked. The first yield tests have shown that most prebreeding lines have lower yield and/or disease resistance and/or quality compared to best elite cultivars. However, few prebreeding lines were at the level of the best elite cultivars in almost all traits carrying > 20% genetic resource. These lines are of high interest for future elite breeding underlining the success of our concept and strategy. These lines are available to all breeders and scientists upon request.

Keywords: genetic resources, wheat, prebreeding, GeneBank 2.0

**IDENTIFICATION OF A MAJOR COMMON BUNT RESISTANCE QTL IN
MAPPING POPULATIONS DESCENDING FROM PI-166910**

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Once one of the most devastating wheat diseases, common bunt lost most of its importance and frightening qualities with the invention of systemic fungicides that could be applied as seed dressings during the first half of the 20th century. As these chemicals are prohibited in organic farming, common bunt has experienced a come-back during the last few decades, causing an urgent need for resistant cultivars. Due to the lack of research attention this disease got over a long time, knowledge about the genetic architecture of bunt resistance and availability of markers to resistance genes are still rather scarce. We therefore aim to unlock a new genetic resource providing full and durable resistance for research and breeding programs by characterizing it through a QTL mapping study. Four mapping populations (MP) descending from the Turkish landrace PI-166910 (postulated to harbour bunt resistance genes *Bt7*, *Bt9* and *Bt11*) and the bunt differential line for the *Bt11*-gene, PI-554119 ('Elgin' X PI-166910), respectively, were phenotyped for common bunt resistance in artificially inoculated field trials over two, three or four seasons (depending on the MP). Genome-wide marker data was obtained from the 25K wheat SNP array and used for construction of linkage maps for each MP separately and a consensus map. QTL analyses identified a major bunt resistance locus on the distal end of chromosome 6DL and an additional QTL near the proximal end of chromosome 4BS. Together, these loci conferred full resistance against common bunt infection. Additional loci associated with common bunt resistance were found on chromosomes 1B and 2A, both in one out of the total four MPs. The amount of phenotypic variation explained by the detected QTL varied between 29.9% and 73.3%, depending on the MP and the environment. Analyses are ongoing and more detailed results will be available until April 2023.

Keywords: common bunt, *Tilletia caries/laevis*, winter wheat, QTL mapping, resistance breeding

CHARACTERIZATION OF AEGILOPS BIUNCIALIS INTROGRESSIONS IN A WHEAT PREBREEDING POPULATION USING DArTseq GENOTYPING

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Tetraploid wild goatgrass, *Aegilops biuncialis* (U^bU^bM^bM^b) represents an attractive and untapped source of genetic diversity that could be exploited in wheat improvement through chromosome mediated introgressions of desirable traits, especially traits related to biotic and abiotic stress resistance and grain quality. However, gene introgression from wild relatives into wheat has been hampered by the lack of genome reference sequence and scarcity of molecular tools. DArTseq is a cost effective and high throughput marker system that doesn't require preliminary sequence information and allows high resolution genome mapping. DArTseq genetic map of the MvGB642 x MvGB382 *Ae. biuncialis* F₂ population revealed that most of the M-genome chromosomes preserved a collinearity with *T. aestivum*, while multiple reorganizations were identified in the U-genome. The wheat-*Aegilops* synteny allowed mapping of the DArTseq markers in a wheat x *Ae. biuncialis* BC₃F₁₋₃ population. The DArTseq genotyping in combination with fluorescence *in situ* hybridization using genomic- and repetitive DNA probes confirmed the presence of *Ae. comosa* chromosomes 1MS, 3M, 4M, 5M and 6M, while majority of *Ae. umbellulata* (U) chromatin was lost and only 1U-, 2U-, 4U- and 7U chromosomal fragments were present in some lines. This approach allowed identification of new lines with deletions, translocations, chromosome additions and substitutions. The new introgression and addition lines together with the chromosome specific markers will facilitate development of new wheat cultivars adapted to changing environmental conditions and will become an ideal base for cloning agronomically important genes.

Keywords: segregating genetic map, introgression breeding of wheat, *Aegilops biuncialis*

This work has been supported by ERDF project 'Plants as a tool for sustainable global development' (No. CZ.02.1.01/0.0/0.0/16_019/0000827), the Hungarian National Research, Development and Innovation Office (K135057; TKP2021-NKTA-06), and the Marie Curie Fellowship Grant award 'AEGILWHEAT' (H2020-MSCA-IF-2016-746253).

GENOME-WIDE ASSOCIATION STUDY OF VIABILITY IN BARLEY STORED UNDER AMBIENT CONDITIONS

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Barley seed viability after storage is crucial for seed multiplication, long-term conservation, food and malting industry. To identify the genetic factors that affect viability of seeds stored under ambient storage conditions the so-called ‘EcoSeed’ panel was investigated. The set consists of 184 spring barley accessions from 23 countries and was genotyped by Illumina 9K. In 2014, seeds were multiplied, mature seeds harvested and stored at 20°C and 15% relative humidity. Percentage of total seed germination (%TG) and of normal seedlings (%NS) were tested according to ISTA rules in 2014, 2018 and 2022. To analyze the ageing behavior of each accession, %NS and %TG were transformed to probit units and on the basis of the viability equation $v = K_i - \sigma^{-1} p$ (where K_i - initial seed germination), the half-viability period (P50) and sigma (σ) were calculated. By using the GAPIT FarmCPU method, genome-wide association (GWAS) analysis, 30 significant ($-\log(p) > 5$) marker trait associations (MTAs) were detected for the tested traits. As candidate genes, we identified the *thioredoxin* (*Horvu_MOREX_1H01G499100*) gene that interact with gibberellic acid (GA), the *ubiquitin-conjugating enzyme E2* gene (*HORVU.MOREX.r2.3HG0185170.1*) affecting the GA-induced DELLA protein degradation and the auxin transporter *WAT1-related protein* gene (*HORVU.MOREX.r2.2HG0178770.1*) that can take part in the auxin GA crosstalk. In summary, MTAs were found in regions of genes associated with transferase activity, transmembrane transport as well as nucleotide and metal ion binding which may play an important role for viability maintenance during ambient dry storage.

Keywords: seed longevity, P50, GWAS, barley

DIVERSE WHEAT LINES AS SOURCES TO BREED FOR RESISTANCE AND TOLERANCE TO EMERGING CLIMATE CHANGE RELATED ABIOTIC AND BIOTIC STRESSES

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Diverse wheat lines such as old and ancient wheat or alien introgression lines containing chromosomal fragments from rye or other grass species are important sources for the introduction of genes that can contribute resistance or tolerance mechanisms against abiotic and biotic stresses that have a significant influence on wheat yield and quality in a climate change situation with an emerging high disease and pest pressure. We have used such a diverse wheat material to hunt genes of specific importance in relation to abiotic and biotic stresses related to the predicted climate change. Thus, we have identified and described genes responsible for resistance to all known stem rust races on wheat-alien introgression lines holding a small segment of the rye chromosome 2R. Furthermore, resistance genes to novel emerging races of stripe rust from the rye chromosomes 4R and 6R have been identified. Additionally, the chromosome 3R was found extremely important for quality characteristics such as baking quality and nutritional quality, as well as for stability for the quality parameters across environments varying in drought conditions. Early root development and plant growth was attributed to genes present in 1R although a high uptake of heavy metals seemed to go along with these characters. Previous studies have indicated 1R as the contributor to several disease resistance genes and to tolerance to environmental stresses, which our results confirm. Currently, we are evaluating the opportunities to use novel genomic tools to transfer the genes that we have hunted, into adapted wheat genotypes. The success story of the transfer of SR59, a resistance gene to all currently known stem rust races, is presented here. Genotyping by sequencing (GBS) was used to identify the adapted character of the breeding lines simultaneously as the presence of SR59 was verified with KASP markers throughout the breeding cycles. By this methodology, the breeding time of adapted lines containing a new resistance line was shortened by ca 50%. As novel genes of importance are identified from divers lines, we will use the same methodology to transfer these genes into adapted wheat material and post it into gene banks, to make lines with novel resistance, tolerance and quality genes available for breeders across the globe.

DEVELOPMENT AND APPLICATION OF *IN VITRO* ANTHOR CULTURE IN CEREAL BREEDING OF CR LTD.

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The increasing importance of doubled haploid (DH) plant production methods is incontrovertible in modern plant breeding and research programs. Homozygous DH lines can be produced within one generation via the application of DH plant production methods based on *in vitro* androgenesis. In some cereal species (wheat, spelt wheat, triticale, barley and rice), the *in vitro* DH plant production methods are well-established methods in several laboratories, crop breeding and research programs.

The method of *in vitro* wheat anther culture, which is applied in our winter wheat breeding and research programs, was adapted for DH plant production of triticale and spelt wheat. More thousands of DH lines are produced year by year for breeding purposes in our laboratory to support and accelerate the breeding programs. After more-year strong selections, ‘GK Déva’, our new DH winter wheat variety was released (2020) and patented (2021) following the earlier registered series of DH varieties. In the last few years, we have made significant efforts to establish the methods of barley *in vitro* anther culture also in our program. Recently, hundreds or thousands of barley DH lines can be produced depending on the requirements of the barley breeding project.

The method of *in vitro* anther culture was improved (pre-treatment, induction and plant regeneration media) to produce rice DH lines for the special conditions of the Carpathian Basin (abiotic- and biotic tolerance). Strong genotype dependency was observed, japonica genotypes produced thousands of green plantlets in the experiments, while the indica genotypes proved more recalcitrant.

However, the induction of *in vitro* androgenesis still remained a scientific challenge in einkorn (*Triticum monococcum* L.). In anther culture of einkorn, androgenesis was induced, the development of numerous ELS was observed, and green and albino plantlets were regenerated. The ploidy level of the regenerated green plantlet (n) was determined by flow cytometric analyses. After significant improvements, it will become a useful tool in einkorn research and breeding programs.

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**GREEN REVOLUTION DWARFING *RHT* GENES AFFECT FLORAL TRAITS
RELATED TO CROSS POLLINATION EFFICIENCY**

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Hybrid wheat breeding is a promising strategy to increase grain yield. Due to the prevalence and usefulness of the *Rht* dwarfing alleles, it is important to gain a better understanding of their effect on traits related to hybrid seed production. Field experiments were performed at the IPK Gatersleben (Germany) and the National University of La Plata (Argentina) during four growing seasons. Traits associated with cross-pollination efficiency were studied using four sets of Near Isogenic Lines (NILs) carrying the alleles *Rht1*, *Rht2*, *Rht3*, *Rht1+2*, *Rht2+3* and *rht* (tall). Results showed that the extreme dwarfing alleles *Rht2+3*, *Rht3* and *Rht1+2* presented the greatest effects in all the traits analyzed ($P < 0.001$). Plant height showed reductions from 21–23% (*Rht1* and *Rht2*), 49% (*Rht1+2*), 56% (*Rht3*), and 64% (*Rht2+3*) compared to *rht* (tall). Spike length was increased up to 9.4% (*Rht1+2*), whereas spikelets/spike were increased up to 5.2% (*Rht2+3*). Floral organs were negatively influenced by *Rht* dwarfing alleles compared to *rht*. Decreases up to 20.2% (*Rht2+3*) in anther length and -33% in anther filament length were observed. Anthers extrusion decreased from 40% (*rht*) to 20% (*Rht1* and *Rht2*), 11% (*Rht3*), 8.3% (*Rht1+2*) and 6.5% (*Rht2+3*). Positive correlations were detected between plant height and anther extrusion, anther and anther filament lengths, suggesting the negative effect of dwarfing alleles in traits with importance for hybrid seed production. Moreover, the magnitude of these negative impact of *Rht* alleles on floral organs depended on the combination of the alleles: *Rht2+3* (extreme dwarf) > *Rht3/Rht1+2* (dwarf) > *Rht2/Rht1* (semi-dwarf) > *rht* (tall). Our results indicate that *Rht* alleles are involved in traits of interest for hybrid production and the need of selecting the best alleles combinations for hybrid breeding programs in order to reduce the cost of seeds production.

Keywords: *Triticum aestivum* L., dwarfing genes, grain yield, hybrid seed, yield potential

IDENTIFICATION OF STEM, LEAF AND YELLOW RUST RESISTANCE GENES IN HUNGARIAN WHEAT CULTIVARS

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In Hungary, all rusts (stem, leaf, and yellow) of wheat are important diseases. As new virulent races can appear, it is important for breeders to know the genetic background of the resistances in their cultivars.

In this study, 65 Hungarian wheat cultivars, registered from 2005 to 2020, were examined using molecular markers to determine the presence or absence and frequency of the stem (*Sr24*, *Sr31* and *Sr36*), leaf (*Lr34*) and yellow (*Yr17*) rust resistance genes. All cultivars, except 2, derived from two main Hungarian breeding programs – Szeged (GK) and Martonvásár (Mv), 26 and 37 cultivars, respectively. The effects of these genes on the rust resistance were also examined.

As for the stem rust resistance, our results showed that 43% of the Hungarian wheats registered in the last 15 years carried at least one of the resistance genes of *Sr31* and *Sr36*. Among these, *Sr31* was more common (29.2%) than *Sr36* (15.4%) - only one variety, GK Rozi, carried both genes. The *Sr24* gene, on the other hand, was not detected in any variety. The two domestic breeding programs typically differed in the use of the *Sr31* and *Sr36* genes. Regarding the *Sr31* gene, the occurrence rate was 37.8 (Mv) and 19.2% (GK) in favour of Martonvásár wheats, however, *Sr36* was only detectable in Szeged varieties (38.5%).

Our results showed that the *Sr31* and *Sr36* genes were widely but biasedly used in the two domestic breeding programs. Due to the emergence of new, virulent pathogen races, there is an urgent need to introduce new, efficient *Sr* genes into new wheat cultivars.

The seedling resistance test of cultivars showed that the *Sr31* and *Sr36* genes were effective against the two stem rust races collected in Hungary, although it is known that new races virulent to these resistance genes are already widespread in large parts of the world.

As for the leaf and yellow rust resistance, the leaf rust resistance gene *Lr34* was found in 7 cultivars (10.8%), and the yellow rust one, *Yr17* was found in 19 cultivars (29.2%) among all cultivars investigated. The two main Hungarian breeding programs also differed in the exploitation of these two genes. Among the Szeged cultivars, *Lr34* was present at a low frequency (7.7%) and in Martonvásár cultivars it occurred at a higher frequency (13.5%). However, the difference in the use of *Yr17* gene was much pronounced in Martonvásár cultivars (43.2%) than in Szeged ones (11.5%).

Based on our pathological field observations and the data of official field trials showed that the *Yr17* gene was effective in Hungary during the epidemic years between 2013 and 2016. However, in later years this gene failed to provide defence against yellow rust in our field trials. Data may help breeders to incorporate effective rust resistance genes into new cultivars.

Keywords: *Sr24*; *Sr31*; *Sr36*; *Lr34*; *Yr17*

EVALUATION OF THE REPRESENTATIVES OF THE GENUS *AEGILOPS* IN THE CZECH REPUBLIC

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An ex situ gene bank collection has been maintained in the Gene Bank Prague since 1985. The whole collection was evaluated for breeding-valuable characters, mainly for resistance to cereal rusts. Six different leaf rust races and five different stem rust races collected from the Czech Republic were applied in the tests. The highest number of accessions resistant to wheat leaf rust and wheat stem rust and powdery mildew was found in *Aegilops speltoides* (90%).

The recent updating of resistant tests to current Czech races of stem and leaf rusts was carried out in greenhouse on 528 accessions of all 21 *Aegilops* species. In these tests the highest proportion of accessions resistant at least to one leaf rust race was in the following species: *Ae. speltoides* (100%), *Ae. biuncialis* (91%) and *Ae. triuncialis* (88%). The same was true for the stem rust in the following species: *Ae. speltoides* (100%), *Ae. cylindrica* (97%), *Ae. triuncialis* (97%), *Ae. neglecta* (95%) and *Ae. geniculata* (95%). A relatively high proportion of the tested accessions was resistant both to leaf- and stem rust. The species *Aegilops speltoides* and *Ae. lorentii* possessed the highest number of accessions resistant to wheat leaf rust was found within the species *Aegilops speltoides* and *Ae. biuncialis* possessed. Species *Ae. speltoides*, *Ae. cylindrica*, *Ae. triuncialis* possessed the highest number of accessions resistant to wheat stem rust. The highest number of accessions resistant both to wheat leaf rust and wheat stem rust was found in the species *Ae. speltoides*.

Keywords: *Aegilops*, resistance, stem rust, leaf rust

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**GENOME-WIDE ASSOCIATION ANALYSES IDENTIFY KEY QTLs FOR
SEEDLING ROOT SYSTEM ARCHITECTURAL TRAITS IN NORDIC-ADAPTED
BARLEY COLLECTION**

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Spring barley (*Hordeum vulgare* L.) is a major crop in Nordic countries contributing to its food security. In recent years climate change has increased the frequency of drought and heat stress threatening food security. Moreover, there are growing concerns about the use of fertilizer in intensive agriculture and its adverse effect on the environment. Root system architecture traits related to early vigor have shown the potential to improve both. In this experiment, we developed a paper-based seedling root phenotyping method along with a root image analysis pipeline using ImageJ. Genome-wide association study using 208 barley genotypes and 13300 SNP markers was performed. The experiment was conducted in a randomized block design with 2 replications (16 seeds per replication). ANOVA, BLUEs and BLUPs were calculated using Meta-R. ANOVA showed a highly significant difference between genotypes and higher heritability ($h^2 > 0.69$) for all the traits. The variation in traits, seminal root number, root surface area, average root area, root angle, and root spreading area were 1.6, 4.1, 4.0, 3.5, and 9.7-fold respectively. Using the FarmCPU model, a total of 26 QTLs were identified (LOD above 3.0). The highest number of significant markers was located on chromosome number 5 related to the seminal root number. The top two markers were on chromosome 6 (LOD 3.9) and 1 (LOD 3.8) for root area per plant and root angle, respectively. We conclude that these associated markers can be used in marker-assisted selection to select genotypes for higher drought tolerance and efficient nutrient uptake.

Keywords: spring barley, root system architecture, genome-wide association study

WIDENING THE GENE POOL OF WHEAT BY CRESTED WHEATGRASS USING THE GAMETOCIDAL SYSTEM

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Gene pool of wheat has narrowed during domestication and cultivation over thousands of years. Wild relatives of wheat are promising source of new genes for wheat improvement. *Agropyron cristatum* (PPPP) represents a rich source of new genes that can be transferred into wheat using the Gametocidal (*Gc*) system, an efficient approach to induce wheat-alien chromosomal rearrangements. The utilization of wild genetic diversity has been hampered by low throughput of selection methods and the poor knowledge on their genomes. Conserved Ortholog Set (COS) markers define orthologous regions, enabling the comparative analysis of wheat and related species.

We crossed wheat Chinese Spring (CS)-*A. cristatum* 5P and 6P chromosome addition lines with CS-*Ae. cylindrica* 2C addition line with *Gc* effect to induce wheat-*Agropyron* translocations. GISH and FISH analysis of BC₁F₂ generation detected seven and two wheat-*Agropyron* translocations for 5P and 6P, respectively. Three 5P-wheat and one wheat-6P translocations were transferred to the next generation. Characterization of selfed progenies indicated that 5P introgressions positively affected the grains number per plant. We also used fourteen wheat-*A. cristatum* disomic and ditelosomic additions and the 1PS·1BL translocation line to map COS markers and investigate wheat-*Agropyron* synteny. Out of 279 COS markers amplified products in *A. cristatum*, 139 were polymorphic relative to wheat. Sixty-nine markers could be mapped on chromosomes 1P–6P in wheat background (6 to 17 markers per chr.).

The results indicated a generally close homeologous relationships between wheat and *A. cristatum* chromosomes.

Keywords: *Agropyron cristatum*, COS marker, genome analysis, translocations, wheat

This research was funded by the Grant Agency of the Czech Republic GAČR Award 16-16992S, MŠMT ČR Grant LO1204, OTKA K135057, TKP 2021-NKTA-06, by RTI2018-093367-B-I00 and PID2021-122530OB-I00 from the Spanish State Research Agency (Ministry of Science and Innovation), co-financed by the European Regional Development Fund (FEDER) from EU.

DETAILED EXPLORATION OF GENETIC DIVERSITY IN *AEGILOPS BIUNCIALIS* USING DArTseq GENOTYPING

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The genetic potential of wild *Aegilops* species can be used to improve the agronomic traits of bread wheat through interspecific hybridization. Accessions of the allotetraploid *Aegilops biuncialis* Vis. ($2n=4x=28$; $U^bU^bM^bM^b$) adapted to different ecological habitats have been considered promising reservoirs of useful allelic variants that would be desirable to transfer into wheat. Understanding its genetic diversity may greatly facilitate chromosome-mediated gene introgression. Diversity Array Technology® (DArT) is a sequence-independent, cost-effective, high-throughput genotyping-by-sequencing platform, which is suitable for scanning the polyploid genomes of wild and cultivated wheats. A collection of 186 *Ae. biuncialis* accessions was previously genotyped by DArTseq approach, generating two types of molecular markers: co-dominant SNP-DArT and dominant silicoDart markers. The population structure of the *Ae. biuncialis* collection was analysed using different and complementary statistical methods - UPGMA clustering, Principal Coordinate Analysis, and the Bayesian model implemented in the software STRUCTURE. The three methods gave a consistent result and revealing that the collection of *Ae. biuncialis* genotypes can be divided into two subpopulations, which are correlated with their geographic distribution: subpopulation A (mainly from Balkans, North Greece and region of Aegean Sea) and subpopulation B (mostly from Turkey, South Greece, Crimea, Caucasus and Near East) with 40 and 118 accessions, respectively. The two major subpopulations can be split into further groups, again according to their origin. Twenty-eight genotypes could not be classified into any of the subpopulations indicating that they have admixed allelic patterns. The population structure with genetically different groups proves the large variability of *Ae. biuncialis*, which could serve as a potential gene source for wheat introgression breeding. Moreover, assessing the genetic relationships between the *Aegilops* accessions is necessary to reduce false associations in GWA studies linking phenotypes and genotypes.

Keywords: genetic diversity, cluster analysis, DArTseq platform, *Aegilops biuncialis*

This work was supported by the Hungarian National Research, Development and Innovation Office (K135057; TKP2021-NKTA-06), and COST European Cooperation in Science and Technology (E-COST-GRANT-CA18101-e7709544).

TESTING *TRITICUM X DASYPYRUM VILLOSUM* HYBRIDS USING MOLECULAR MARKERS

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Dasypyrum villosum (L.) Candargy (syn. *Haynaldia villosa*) is a diploid ($2n = 14, VV$), annual grass-species native to mainly Mediterranean and Caucasian areas. It belongs to the tertiary gene pool of wheat and has been widely used to enhance its resistance to pathogens as well as to improve grain protein quality (De Pace *et al.*, 2001). Our aim was to prove the presence of the 6V chromosome or chromosome segment in hybrid genotypes. Thus, in later generations, these hybrids can be followed and selected via MAS. The tested plants, that were developed earlier at DAFNE (It) and the Centre for Agricultural Research, were the following: {6V disomic substitution lines (CSV32), disomic addition lines (CSV63) and 6V introgression lines (CSV58, CSV59, CSV60)} and a few backcrossed genotypes. Four of the tested markers are located on the short arm of chromosome 6V {(6VS-03, 6VS-08, 6VS-12 (Li *et al.*, 2017) and *Pm* resistance gene marker SCAR1265 (Liu *et al.*, 1999))} while the other 8 tested ESTs or EST derived STS markers are mapped to the long arm of chromosome 6V {BE422631, BE442546, BF262689, BE403818, BE605218, BQ280965 (Sun *et al.*, 2018), BE497099-STS (linked to resistance gene *Sr52*, Qi *et al.*, 2011) and BE518064-STS}. Out of the tested markers, 6 yielded polymorphic fragments that could have been distinguished on agarose gel. According to the results, the presence of the 6V chromosome in the substitution and disomic addition lines were consistent, but it was not detectable in any of the introgression lines. For the reliable MAS of the backcrossed and later generations, more microsatellite markers need to be involved or developed. Therefore, we are planning to compare the phenotypic/disease-resistance data of the hybrid lines with the molecular test results and carry out further studies using *in situ* hybridization techniques to confirm the presence of the 6V fragment.

Keywords: *Dasypyrum villosum*, wheat, hybrids, aneuploid line, introgression line, MAS

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**PHENOTYPING AS PART OF THE IMPLEMENTATION OF THE AGENT
PROJECT IN THE POLISH GENE BANK**

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The National Centre for Plant Gene Resources is implementing the Horizon 2020 project "Activated GEnebank NeTwork - AGENT" between 2020 and 2025.

One of the main objectives of the project is to establish an active gene bank network that will develop protocols and standards for the collection and storage of gene bank data. The FAIR (Findable / Accessible / Interoperable / Reusable) principles will also be implemented to ensure that genetic resources are publicly available and searchable.

Within the Horizon 2020 project 'Activated GEnebank NeTwork - AGENT', unique objects representing the Polish wheat and barley gene pool have been selected. During the project, genotype and phenotype information will be obtained for selected objects from European barley and wheat collections. The obtained data will be compiled and made available to the public. They will be used to develop a strategy for the creation of a global atlas of the genetic diversity of wheat and barley genebank accessions. The compilation of genotypic and phenotypic data obtained during the project, together with the historical data collected over decades by curators in gene banks, will lead to the discovery of new genes and traits, thus stimulating the development of science and breeding.

The following characteristics were evaluated during the two growing seasons: occurrence of certain plant diseases, mowing, flowering, average plant height, yield and thousand kernel weight for the 471 barley SSD lines and the 462 wheat SSD lines. 301 lines from this collection are currently being evaluated for drought tolerance.

Keywords: genetic diversity, barley, wheat, gene bank

**SEQUENCE DIVERSITY OF CANDIDATE GENES FOR SELECTION IN RYE
(*SECALE CEREALE* L.)**

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During domestication and subsequent improvement plants were subjected to intensive positive selection for desirable traits. Identification of selection targets is important with respect to the future targeted broadening of diversity in breeding programs. Rye (*Secale cereale* L.) is closely related to wheat and an important crop in Europe. Recently, we have identified putative candidate genes targeted by selection in cultivated rye germplasm. The aim of this study was to evaluate sequence diversity of selected candidate genes in diverse rye accessions.

Plant material consisted of 94 inbred lines, landraces, historic and modern cultivars, and wild rye species of various geographic origin. Based on low coverage resequencing (DARtreseq) we analysed sequence diversity of 47 putative candidate genes with obvious connection to traits relevant for plant breeding, such as plant growth and development, plant fertility and reproduction, response to various environmental stimuli (including pathogen resistance). In total we identified over 1200 polymorphic sites in the candidate genes and their putative promotor regions. Rate of SNPs per base pair varied among the genes. The highest polymorphism was detected in Leucine-rich repeat receptor-like kinase (SECCE1Rv1G0037030.1) locus and Gibberellin receptor GID1a (SECCE1Rv1G0037070.1).

Keywords: *Secale cereale* L, selective sweeps, sequence diversity, candidate genes, DARtreseq

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NEW SMALL GRAIN CEREALS BRED IN MARTONVÁSÁR WIDENING THE OFFERED SPECIES SORTIMENT

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Mv Télgyöngye is the first hullless winter oat variety bred in Hungary: Former naked spring oat varieties which were introduced or bred in Hungary couldn't compete with the ones with husks, therefore they couldn't spread widely. Oats are traditional forage crops in Hungary, but recently their role is increasing in human consumption. Husk weight could be 25-35% of the whole grain yield which means cost implications beyond peeling. Its grain is naked, the potential productivity can reach the yield of husky spring oats and can also compete with hulled winter oats either in favourable growing seasons along with appropriate agrotechnics. It is even better at tillering than Mv Hópehely. Winter hardiness is suitable, and the ears are not sensitive to stark wind even after the final ripening. The variety has large grains. Thousand kernel weight is usually higher than the unhusked GK Impala controls, and if the environmental conditions are favourable, it competes with Mv Hópehely. The variety is resistant against foliar diseases, however, due to a lack of husks, grains are defenseless against pathogens, therefore, a fungicide treatment is recommended at booting stage. The protein (13-17%) and starch (48-50%) content of the grains are high, but crude fibre content is lower (1,5-2,5) compared to whole grain hulled oats (8-15%). It has a high protein content and a favorable ratio of essential amino acids. Mv Télgyöngye combines applicability on a wide range with efficiency, resistance and high productivity.

Mv Pangolin new baking quality spelt variety: Since 2002 there is spelt breeding in Martonvásár, as a result of which already three varieties got state recognition (Mv Martongold, Mv Vitagold, Mv Pangolin) and the latest of all, Mv Pangolin was just registered early this year. It is a winter-type spelt with awnless ears and a red husk colour. It has no bread wheat in its pedigree; therefore, (as a "pure" spelt) it is a more sought-after variety among eco farmers and manufacturers. Its winter hardiness, tillering and weed suppression ability are outstanding. Strong and well-developed root system and stem provides great lodging resistance. The average height of the plant is 115 cm which classifies it among the short oat varieties. Heading time is in late May. Both crude protein and wet gluten content are high (18,2% and 30%,) while its pharino-graphic classification is B2-C1. The potential productivity is 5-7 tons in husky seeds. As a recent result of the breeding efforts in Martonvásár, Mv Pangolin is a pure spelt which is directly suitable for the baking industry. Its gluten quality exceeds predecessors and competes with Franckenkorns. Great step to meet the expectations of both eco farmers (pure spelt, low input, good resistance and nutrient utilization capacity, stable and abundant harvest) and bakery industry (milling quality, high falling number).

Keywords: naked winter oat, spelt, human diet, milling quality

Project no. TKP2021-NKTA-06 has been implemented with the support provided by the Ministry of Innovation and Technology of Hungary from the National Research, Development and Innovation Fund, financed under the TKP2021-NKTA funding scheme.

A QTL FOR DRY MATTER PARTITIONING FOUND IN SEEDLINGS OF A LOCAL RIL POPULATION OF BARLEY

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The process of allocating resources to different plant organs in the early stage of development can affect their adaptation to drought conditions, by influencing water uptake, transpiration, photosynthesis and carbon storage. A distinct behavior of biomass partitioning between two Spanish barley landraces was observed in a rhizotron experiment (Boudiar et al., 2020, *Agronomy* 10). A RIL population of circa 200 lines, derived from the cross of those lines was developed at the EEAD-CSIC. We devised an experiment to test if seedling biomass partitioning was under genetic control. The experimental set-up consisted of seedlings grown in pots filled with silica sand (0.4-0.8 mm). Five pre-germinated seeds of similar size were transplanted to one pot, as the experimental unit. Four replicates per genotype (RILs and parents) were tested, in a growth chamber under controlled conditions. After one week, plants were washed and dried, shoot and root separated and kept in envelopes, then oven-dried at 70°C for three days, and weighted. Data were analyzed using a model with random replicates and genotypes as a fixed factor. There were genotypic differences for all variables measured. The RILs were genotyped with a commercial 15k SNP chip. A map was constructed with 1353 SNP markers, with 7 linkage groups, and a total of 1485 cM. A QTL analysis, using Genstat.20, revealed no QTL for weight variables. However, a clear single QTL for biomass partitioning (root/shoot ratio) was found, in the long arm of chromosome 5H. This is one of the first reports of QTL for biomass partitioning in barley, and could have relevance for devising barley ideotypes for breeding.

Keywords: barley, biomass allocation, seedling, roots

**GENDIBAR: A COLLECTION OF MEDITERRANEAN BARLEY LANDRACES
FOR CEREAL ADAPTATION STUDIES**

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A collection of 989 barley landraces from the Mediterranean basin has been gathered and multiplied by SSD. This collection, the Mediterranean barley reference set, is the basis for genomics and physiological approaches studying barley adaptation in response to climatic factors. The collection was genotyped by genotyping-by-sequencing. Furthermore, the dataset was enriched with data of another 1779 accessions of the Mediterranean and neighboring regions (Milner et al., 2019), gathered from the IPK Bridge web portal (König et al., 2020), genotyped with the same technology. The combination of both datasets resulted in 10,754 SNPs, with a minimum read depth of 5. Population structure was analyzed with the software sNMF (Frichot et al., 2014), distinguishing fourteen groups, with clear geographic and climatic basis. A set of 25 genes, related to the control of flowering time, was resequenced with a custom-made capture array, with a MiSeq sequencer. The distribution of the main flowering time genes over the genetic groups will be presented, indicating the key role of growth habit and photoperiod sensitivity in the spread and adaptation of barley landraces.

Keywords: barley, landrace, diversity, flowering

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SESSION 2

Grain yield and quality

PRESENT SITUATION AND FUTURE TRENDS IN QUALITY RELATED CEREAL CHEMISTRY AND TECHNOLOGY

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During the last one and half century the attributes for evaluating wheat quality significantly differentiated and expanded.

In recent years the new challenges originated from the demands of the industry and the consumer, altered the traditional priorities of quality related research and development in two directions. In the industry it become essential to implement processes capable to reduce the production cost of the end-products and therefore to use source materials, new genotypes which are suitable for these technological challenges. New requirements such as the need for wheats with less mixing energy requirement but stable, strong dough, or the increased demand for higher water absorption are good examples for this industry-driven trends. The other major revolutionary change in the term 'quality' is caused by the consumer's strong demand to get healthier, more nutritious but low-calorie containing baked goods in a wide product range satisfying even those individuals who are sensitive to gluten containing products.

Current and potential approaches to quality evaluation through the cereal chain from pre-breeding to end-product characterisation will also be discussed.

DIVERSITY TRENDS OF AGRONOMIC AND QUALITY TRAITS IN VARIOUS OLD AND MODERN WHEAT VARIETIES CULTIVATED UNDER IRRIGATED CONDITIONS OF TÜRKIYE

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Plant breeding has contributed to increase grain yield of winter bread wheat (*Triticum aestivum* L.) over decades; however, the rate of this increase has varied for ecological and growing conditions of the countries. Thus, our objectives were to compare old and modern wheat varieties for agronomic and quality traits, and to determine long-term gains for these traits. For this purpose, twenty-two winter wheat varieties registered in Türkiye were grown for two consecutive years (2019 and 2020) at under irrigated conditions at Konya province of the country. Twenty winter bread wheat varieties registered from 1931 to 2020, one einkorn (*T. monococcum* spp. *monococcum*) and one emmer (*T. dicoccum* Schrank) variety were used as genetic material in this study. ANOVA showed a clear trend for many traits from old to modern ones. Improvements in grain yield were based on linear increase in harvest index, while plant height decreased. Grain yield increased at a rate of 26.7 kg/ha/yr and plant height decreased by 0.18 cm/yr. Additionally, some quality traits such as volume weight, Zeleny sedimentation, grain hardness and gluten index were in increasing trend whereas protein ratio and wet gluten ratio remained unchanged. Principal component analysis allowed an overall view of the breeding activity. A clear-cut separation was observed for economically important traits such as grain yield, plant height, Zeleny sedimentation, and gluten index from ancient varieties to modern bread wheat varieties, matched with two-step gradient, before and after green revolution.

Keywords: Wheat, genetic gain, grain yield, plant height, grain quality

This study was financially supported by The Scientific and Technological Research Council of Türkiye (TUBITAK) with the project number "122O333".

BREEDING DURUM WHEAT FOR ORGANIC FARMING

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According to the European Union Farm to Fork strategy, at least 25% of the EU agricultural land shall be under organic farming by 2030. However, the lack of wheat varieties specifically adapted and selected for organic conditions and lower input of pesticides and fertilizers in organic production systems result in lower grain yields.

The aim of the study, in the frame of the ECOBREED project (European Union's Horizon 2020 research and innovation program under grant agreement No 771367), was to select suitable durum wheat accessions for organic farming with high grain quality, production, and tolerance to biotic and abiotic stresses.

The trial started in 2018 at Tuscia University, Central Italy, with a preliminary evaluation of 72 durum wheat genotypes, including old varieties, landraces, and new accessions developed in the Central European and Mediterranean areas. The screening was focused mainly on traits important for organic farming, including crop ground cover/competitiveness with weeds, disease resistance/tolerance, grain yield, protein content, etc. From the first-year trial, 27 genotypes were selected and evaluated in the field trial for three years more. The accessions were also characterized genotypically by SSR markers associated with traits of interest.

The study allows us to identify seven accessions suitable for organic agriculture with wide adaptation to different environmental conditions.

Keywords: durum wheat; organic farming; phenotyping; genotyping

COMPOSITIONAL AND QUALITY CHARACTERIZATION OF SPELT WHEAT

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The use of spelt wheat (*Triticum aestivum* subsp. *spelta* L.) in the baking industry has become fashionable again in recent decades. This is due in part to the fact that it can be grown well in organic farming conditions, and in part to the fact that the number of people following the trend of healthy eating has increased. It is difficult to thresh, however, the presence of the husk protects the seed from abiotic and biotic stress factors, so it requires less chemical protection, on the other hand, the bioactive compounds in the grain are assumed to have unique nutritional value and health effects.

We therefore examined the variability of the physical, compositional and baking quality characteristics of 90 spelt genotypes, which was significant for almost all characteristics. Among them, the protein and gluten content was on average higher than that of wheat, while the strength and stability of the dough was weaker than that. Starch gelation temperature was also higher in spelt, while starch damage was lower than in wheat, resulting in lower water uptake. Baking quality is determined by the amount and composition of storage proteins. The protein composition of five bread and spelt wheat was studied grown in conventional and organic production. We found a significant difference between varieties, wheat species and growing areas. The total protein content was significantly higher in spelt than in bread wheat in both cultivation systems. However, the content of the glutenin- and insoluble polymeric protein was lower in spelt than in wheat, which indicates a weaker breadmaking quality. The proportion of monomeric gliadins differed significantly in conventional and organic systems. Some spelt varieties have proven suitable for cultivation in organic and low-input systems. By examining the bioactive components, we found a higher content of alkylresorcinol, but a lower content of fructan in spelt wheat. Organic spelt had significantly higher starch, fiber and alkylresorcinol content, but lower β -glucan and protein content than conventionally grown spelt. The 'Oberkulmer-Rotkorn' was characterized by the highest values for the majority of the tested properties in both farming systems. Significant differences were also found in the S, Na content of the spelt varieties, as well as in the content of all macroelements, Fe, Mn, Zn and Cu between the fractions.

The properties of the tested spelt genotypes showed great diversity, thus proving their suitability for breeding purposes. We have identified gene bank sources and international varieties suitable for baking purposes, but by using these genotypes, quality suitable for the production of other specific local products can also be achieved.

Keywords: breadmaking, breeding, composition, diversity, spelt

The research was supported by K135211, SA-25/2021, TKP2021-NKTA-06, while the research collaborations were supported by COST Sourdomics (CA 18101) projects.

NEGLECTED WHEAT AS A SOURCE OF LOW-ATI FOOD PRODUCTS?

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Although the genus *Triticum* covers a broad genetic diversity, species and subspecies aside durum and common wheat are rarely used in production, consumption, breeding, and research. However, some neglected wheat species such as einkorn and emmer have been shown to cause less ATI-induced intestinal inflammation compared to other wheat species. ATIs (amylase-trypsin inhibitors) are a group of wheat allergens associated with baker's asthma and non-celiac wheat sensitivity (1,2,3).

As little is known about the extent of variability in ATI levels and bioactivities within the broad *Triticum* spectrum, this study aimed to determine ATI levels, isomeric distribution, and inhibitory activities in a broad collection of underutilized wheat species with rare genome constitutions, in order to identify low-ATI lines for sensitive patients.

The results confirmed the high ATI variability depending on the genotype's ploidy level and genomic constitution. While diploid genotypes showed the highest trypsin inhibitory activity due to the presence of a specific einkorn trypsin inhibitor (ETI), significantly lower activities were observed in tetraploid and hexaploid cultivars. In contrast, α -amylase inhibitory activity followed an opposite trend and showed the highest levels in hexaploid wheat, while no α -amylase inhibitory activity was detected in diploid genotypes.

LC-MS/MS results revealed that the occurrence of ETIs in particular was strongly associated with the presence of A^m and A^t genomes, while it was absent in species with A^u genomes. Understanding the genetic effects on ATIs may help identify and develop low-ATI wheat lines, which is a promising strategy to combat diseases associated with cereal consumption.

Keywords: Allergy, amylase-trypsin inhibitors, LC-MS/MS, *Triticum*, wheat sensitivity

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PARENTAL-DOSAGE-DEPENDENT EFFECTS ON BARLEY SEED DEVELOPMENT

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Cereal grains represent a specific type of seeds where the largest part is formed by endosperm, a specialized triploid tissue for embryo protection and nourishment. The endosperm combines both parental genomes in an unusual ratio of two maternal and one paternal copies (2m:1p), and this proportion is crucial for normal seed development. As a model to study parental-dosage-dependent seed development in cereals, we use elite varieties and landraces of cultivated barley (*Hordeum vulgare* subsp. *vulgare*) and wild progenitor of cultivated barley (*H. vulgare* subsp. *spontaneum*). We developed and characterized barley synthetic autotetraploid lines, and performed several combinations of inter-ploidy reciprocal crosses. We found that the viability of inter-ploidy/inter-accession hybrid seeds was impaired in both directions of hybridization, indicating a strong triploid block in the tested varieties. However, the block had a different strength depending on the direction of the cross. Pollination of 2x mothers with pollen from 4x fathers resulted in a maternal genotype-dependent failed endosperm cellularization. In contrast, the endosperm of 4x mothers pollinated by 2x fathers cellularized precociously and the seeds were aborted on different days after pollination depending on the father's genotype. This finding sets a new long-term direction for our study to understand the molecular mechanisms of parental conflict and triploid block in intra- and interspecific hybridization in cereals.

Keywords: parental conflict, triploid block, tetraploid barley

TRITICALE GRAIN QUALITY TRAITS: ARE THERE ENOUGH VARIATION FOR SELECTION?

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Due to its high adaptation and cost-effective production, hexaploid triticale (*X Triticosecale* Wittm.) is widely spread in marginal soils in Europe. The grain has satisfactory yield in low input environments with valuable and healthy feeding quality. Beside the flour –particularly the whole meal – is a very useful natural additive to achieve high nutritional quality flour blends for bakeries and other food purposes. Triticale whole meal is rich in mineral nutrients and arabinoxylanes, fibers, and alpha amylase enzymes. On the field of the various quality traits, however, quite limited information available on the selection possibilities. For breeders, the information based on large number of test-dates are particularly valuable. The present study focused on establishing the extent of some grain quality traits in a European triticale *gene-pool*. Triticale entries (n=480; parental lines, advanced breeding stocks) produced in the experimental station near to Szeged, Hungary in 2022 were used in the laboratory tests. Grain physical and chemical parameters were dignified by SKCS Instrument Model 4100, the MinInfra, and Perten DA7250 NIR analyzers. For Hagberg falling number (FN) –due to absence valid method for triticale - we followed the standard procedure developed for wheat measurements.

Grain hardness (HI) values of the tested triticale genotypes ranged between 8 (minimum) and 90 (maximum) with a median of 34. Thousand kernel weight (TKW) showed also huge variability. The mean value was 43.2 g with the minimum of 16,2 g, and maximum of 60g. The extent in grain diameter (d) was between 2.45 and 3.36 mm – with a median of 2,86 mm. TKW and d proved to be in positive correlation ($r=0,89$) in the study. A huge variation was detected in protein content of the whole meals; minimum: 7.9%, maximum: 16.3% with a median of 12.8%. FN showed an average of 224 s with extremely large individual variation between 17 - 366 s. A smaller extent in the starch content was proved in the study: median was 74.2% with minimum of 71.4% and maximum of 77,3%. We detected wider variation for crude fiber content with limit values of 2.05% and 3.45% (median: 2.7%). Values in all the tested parameters showed good fit to normal distribution ($p=0.05$). At the same time, the starch and fiber contents showed two-peak normal distributions that should be considered in selection mode and efficiency. The present study and our parent –offspring regression data from our past studies allow us to draw the conclusion that triticale breeders may carry out tandem and/or rotation selection work for quality. Most of the parameters studied in this experiment proved mid- or large extent providing good base for successful breeding.

Keywords: triticale, quality, protein, fiber, grain physical properties

**A NEW *MYB* GENE IN DURUM WHEAT GENOME AS CANDIDATE FOR
*PP-B1***

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Anthocyanins constitute the largest and the most important group of water-soluble natural pigments with health-promoting effects. In wheat, anthocyanins are mainly localized in different bran layers, conferring black, purple, blue, or red tone to the grains.

From the genetic of view, three main loci have been identified in wheat. The *Purple pericarp 3* (*Pp3*) has been mapped to chromosome 2A both in bread (*Triticum aestivum* L.) and durum (*T. durum* Desf.) wheat, whereas *Pp1* has been positioned on chromosomes 7B (*Pp-B1*) in durum and on 7D (*Pp-D1*) in bread wheat. In durum wheat, although these loci have been mapped through molecular markers, the causative genes are still unknown. To uncover this aspect, an F7:8 recombinant inbred line population (RIL) derived from parents with different anthocyanin content was used for QTL mapping. Three regions were identified on chromosomes 2A, 3A and 7B and explained the highest phenotypic variation (> 50%). Taking advantage of Svevo (v1.0) reference genome and the latest insights regarding the functional role of *Pp3* and *Pp1*, a new *MYB* gene was *de-novo* annotated on the short arm of chromosome 7B and reported as candidate for *Pp-B1*. Non purple varieties harbor an insertion of ~ 1.6 kb within the first exon which led to a non-functional allele. Structural genes of the anthocyanin pathway such as *Dihydroflavonol-4-reductase* (*DFR*), and *Cinnamoyl-CoA reductase* (*CCR*) were also identified. Further studies are on-going to validate both the candidate *MYB* gene and the other genes involved in the accumulation of anthocyanins in grain pericarp.

Keywords: purple durum wheat, health-promoting effects, transcription factors, *MYB*, grain colors

RELATIONSHIP BETWEEN GRAIN YIELD AND SPECTRAL REFLECTANCE INDICES IN SPRING WHEAT AT VARIOUS GROWTH STAGES

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Spectral reflectance indices may be a useful method to evaluate crop development and grain yield. The aim of this study was to evaluate the relationships between grain yield (GY) and vegetation indices (VIs) in spring wheat (*Triticum aestivum* L.) at various growth stages. Research was carried out at the Institute of Agricultural Resources and Economics (AREI), Stende Research Centre located in the North West part of Latvia. Field trials was established in 2021 and 2022 years including 300 wheat genotypes, with plot size 5 m² in 2 replications. Canopy spectral images were collected in three growing stages - tillering begins (GS 21), flowering halfway (GS65) and early milk (GS73). Wheat genotypes were evaluated for grain yield and various vegetation indices. Unmanned aerial vehicle (UAV) Phantom 4 Pro was equipped with a multi-spectral camera used for the Normalized Difference Vegetation Index (NDVI) indice calculation. Phenomobile equipped with Red-Green-Blue (RGB) camera applied for Green Area (GA), Greener Area (GGA), Crop Senescence Index (CSI) and Early Vigor (EV) determination. Statistical analyses were conducted using the open-source software, RStudio 4.2.2. Data were evaluated by analysis of variance (ANOVA) and Pearson correlation to test the spectral reflectance indices relationship with grain yield. In this study, principal components analysis (PCA) was used to reduce the number of variables to be analysed. The mean grain yield per plot among the wheat genotypes ranged from 249.48 to 689.78 g m⁻², average grain yield was 503.73 g m⁻². All vegetation indices and grain yields showed significant variations between different wheat genotypes (p<0.01). Comparatively the highest positive correlation with grain yield between all vegetation indices was detected at early milk growth stage. In 2021 and 2022 correlation coefficients between GY and NDVI were r=0.64 and r=0.55, respectively. In both years significant (p<0.001) positive correlation coefficients were also observed for GA (r=0.52) and EV (r=0.52), and a negative correlation for CSI (r=-0.40 and -0.44). PCA was used for data analysis where the first three components accounted for 77% in 2021 and 83% in 2022 of the total variation. The first principal component, which accounted for 31% of the variation in both research years, was strongly associated with GY and all VIs obtained in GS73. The results showed that the multi-spectral and RGB imaging tools have potential to be used in wheat breeding programmes for plant phenotyping, especially in large-scale field trials. This study was carried out in the frame of EEA and Norway Grants project “NOBALwheat – breeding toolbox for sustainable food system of the NOrdic BALtic region” (NOBALwheat).

Keywords: wheat breeding, UAV multi-spectral imaging, RGB image, vegetation indices, correlation

DEVELOPING SPELT GENOTYPES FOR FUNCTIONAL DIETETIC USAGE: A POSSIBLE SOLUTION FOR FODMAP PROBLEM

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Wheat flour sensitivity is an emerging problem in human population, especially in the West-cultured countries. The number of IBS patients multiplied in the last decade, and for most of them the only cure is keeping a low FODMAP diet which excludes several gluten-bearing cereals. Breeding low FODMAP spelt variety can give a solution if the flour is processed by the adequate cooking/baking technology. The aim of our work is to develop low FODMAP spelt genotypes with good baking quality. We combined the *in vitro* DH production and tradition selection in Cereal Research Ltd's breeding program. More than 400 DH lines of spelt wheat were produced and selected during 5 years. FODMAP content, wheat quality and protein composition were analysed to select/improve the best ones for baking purposes. White flour was produced to make sourdough bread. Fructan content is measured primarily to follow the changes of FODMAP content through the process from harvest to end-product. According to our results, there are spelt genotypes that has low fructan (<1%) content which is independent by seasons. Milling and baking technology reduces further the fructan content. Sourdough bread which is made of low FODMAP spelt variety is tested recently in a small scale dietetic study. The results of this study verifies so far the efficiency of the low FODMAP spelt wheat variety.

Keywords: spelt, FODMAP, quality

**PROJECT R-GRAIN - IDENTIFICATION OF THE MOLECULAR TRAITS
DETERMINING THE RYE YIELDING AND IDENTIFICATION OF EFFECTIVE
MOLECULAR MARKERS**

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Common rye (*Secale cereale* L.) is one of the youngest cultivated crop species. Due to its tolerance to unfavorable environmental conditions during plant development, this cereal can be grown in many regions of world. However, the adaptation to unfavorable conditions often results in reduced quantity and quality of the yield. One of the breeding goals is to produce rye varieties with high yielding potential, tolerating biotic and abiotic stresses. Due to the open-pollination of this species, the early stages of selection require both enormous expenditure in terms of physical work involving observation and selection in the field as well as high financial outlays. The goal of R-GRAIN project is understanding the regulation of grain development in rye plants and then, identify the molecular markers that will allow the effective selection of breeding lines with high yielding potential at an early stage of plant development. The identification of transcripts and their splicing variants possibly controlling grain development was performed using the Oxford Nanopore Technologies and Illumina sequencing. At the second stage of the project, sequencing of cDNA and gDNA, as well as expression analysis of selected genes, will be used to identify functional polymorphisms that correlate with exact grain phenotype.

GENETIC DIVERSITY IN OLD AND ALIEN WHEAT GENOTYPES FOR NUTRITIONALLY IMPORTANT TRAITS

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Wheat is an important staple crop that is grown and consumed around the world. It is a major source of calories, protein, vitamins, and minerals in many homes, also contributing micronutrients to the diets. Breeding micronutrient-dense staple crops is a powerful strategy to address global undernutrition and malnutrition. Wild crop relatives is an important source that can be used to introduce valuable genetic diversity to elite wheat cultivars to improve their nutritional value. This study assesses the variation in protein and mineral content in a genetically diverse set of wheat consisting of among 173 winter and 166 spring genotypes. Analysis of variance (ANOVA) revealed a significant effect ($P < 0.01$) of the genotypes on all analysed traits protein related traits Furthermore, ANOVA revealed a significant impact of the wheat genotypes on the content of all analysed minerals (Fe, Zn, Se, Ca, Mn, K, Cu, and Cd). High content of iron and zinc, combined with low cadmium content, were recorded in many genotypes, with the spring wheat lines SW58 and SW358, and the winter wheat lines Kr 08-150 and Kr 98 108-3B as the top performers. Genotype performance analysis based on high %UPP and TOTE values resulted in Kr 08-78 and Kr 08-10 as the best performers among winter genotypes, while SW20 and SW256 were the highest performers among spring genotypes. The study demonstrates ample genetic variability in old and wild relatives of wheat for protein content, gluten strength, and minerals, which can be harnessed for improving wheat nutrition and protein functionality through breeding programs. The findings of this study will result in a search for the genes of nutritional traits of interest to introduce into elite cultivars to enhance the nutritional benefits and combat nutritional deficiency.

Keywords: spring and winter wheat, crop wild relatives, undernutrition, protein content, gluten strength

**PROTEIN QUALITY IN 2BS.2BL-2RL WHEAT-RYE TRANSLOCATION LINES
WITH RESISTANCE TO NOVEL STEM RUST RACES**

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Wheat is the primary source of daily calories and protein in many diets worldwide. Since its early domestication to today's modern plant breeding, this crop has been subjected to strong selection pressures, resulting in high yielding cultivars with good resistance to the major diseases. However, novel races of the major diseases such as stem rust, have resulted in an increased need to develop new varieties with durable resistance. The introduction of desirable genes from wild relatives into cultivated wheat is one opportunity to develop cultivars with sustainable and durable resistance. At the Swedish University of Agricultural Sciences wheat-rye introgression lines have been developed through a life-long work by the late Professor Arnulf Merker. These lines have demonstrated promising resistance and tolerance mechanisms towards several biotic and abiotic stresses. One line is a 2BS.2BL-2RL translocation that contains genes for disease resistance against all currently known stem rust races. Thus, this line is extremely interesting to introduce into breeding programs to develop commercial varieties with resistance to the emerging races of stem rust. However, the current form of the line does not hold agronomic performance to be grown as a commercial high yielding variety. Therefore, this 2BS.2BL-2RL translocation line was crossed and backcrossed with three commercial varieties to enhance its agronomic performance. KASP markers were used in subsequent generations to demonstrate the inheritance of the 2BS.2BL-2RL gene to produce another generation until BC₂F₇. These lines were evaluated using SE-HPLC on BC₂F₇ and BC₂F₈ generations for their protein quality. The results demonstrated that a transfer to commercial wheat varieties of a short segment of the 2R to the distal region of chromosome 2BL, does not adversely affect the protein content and composition of bread wheat. There was no significant difference between BC₂F₇ and BC₂F₈ and the commercial varieties regarding the amount of SDS-extractable proteins (TOTE), SDS-unextractable proteins (TOTU), and the percentage of unextractable polymeric proteins (%UPP). Thus, the results from the present clearly showed the opportunity to use the 2BS.2BL-2RL translocation line in breeding for high quality wheat with improved disease resistance to stem rust.

Keywords: molecular marker, protein, *Secale cereale*, wheat

BAKING QUALITY OF WHEAT VARIETIES IN THE ECOBREED PROJECT

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Selgen, a.s.

The purpose of organic farming is to produce healthy and high-quality food in a sustainable manner. Organic wheat cultivation tries to use some traditional agrotechnical practices in order to cope with the limited fertilization use and pesticide exclusion. Only farm fertilizers usage significantly limits the achieved baking quality of wheat in the protein content parameter. It is necessary to involve the most modern scientific knowledge from breeding and phytopathology to breed new wheat varieties suitable for organic farming. The presented work summarizes the multi-year results of the baking quality in set of selected varieties (Ecobreed project) from experiments managed in organic conditions. Further, compares these results in a paired test resulted from conventional experiments.

Keywords: wheat, organic farming, quality

SESSION 3

Biotic stress response

**HOW TO SUPPLY 11 BILLION PEOPLE WITH FOOD? REDUCTION OF LOSSES
IN THE CEREAL PRODUCTION CHAIN, WHERE ARE THE HOT SPOTS AND
WHAT CAN BE DONE?**

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Nearly half of the harvested grain 2100 million metric tons (MMT) is lost, by adding the preharvest losses at about half of the harvested grain mass, more than 65% of the production capacity disappears, 2028 from the possibly total of 3152 MMT Diseases and abiotic stresses on the field are responsible for 350 MMT. The mycotoxin contamination is 10 % (210 MMT), so the plant breeding is responsible for 560 MMT we need multiple resistance to the most important diseases and abiotic stresses. However, without updating harvest logistics and storage loss 440 MMT) the investment into the breeding cannot be profitable, also here precision technology is needed. This is valid also for the abiotic stresses where agronomy and irrigation effective water supply is inevitable. The whole breeding process should be renewed, the increased biotic and abiotic resistance will be a cornerstone supported by a much more variety registration process. By plant breeding a 250 MMT reduction from 560 MMT loss can be achieved within 5-10 years, and 95 % reduction of the storage losses is possible. The mixing of precision technology combined with the stone age solutions is not sustainable. Reduction of total loss within ten years by 506 MMT (25%) that is enough for additional 3-4 billion people and a further 400 MMT can be spared by a more sophisticated storing technology. This way we can resist to much of the problems of the climate change and spreading toxins like aflatoxins and fumonisins.

Keywords: breeding for resistance, reduction of grain losses, variety registration to toxigenic fungi, production chain, resistance to climate change

SEPARATING DISEASE AND SENESCENCE IN WHEAT CANOPIES USING DEEP LEARNING AND DYNAMIC MODELLING

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The capability of wheat to capture resources from the environment and convert them to harvestable products during grain filling is strongly affected by canopy longevity and healthiness. Age-related physiological senescence and various biotic and abiotic stressors drive overall greenness decay dynamics under field conditions. Separating their effects on greenness decay dynamics is necessary for efficient selection of optimized senescence and disease resistance under field conditions. We developed methods that enable image-based monitoring of chlorosis and necrosis separately for ears and shoots (stems + leaves) based deep learning models for semantic segmentation and color properties of vegetation. Application of the models to image time-series revealed temporal patterns of greenness decay as well as the relative contributions of chlorosis and necrosis. Image-based estimation of greenness decay dynamics was highly correlated with scoring-based estimations ($r \approx 0.9$). Contrasting patterns were observed for plots with different levels of foliar diseases, particularly septoria tritici blotch. Our results suggest that tracking the chlorotic and necrotic fractions separately may enable (i) a separate quantification of the contribution of biotic stress and physiological senescence on overall green leaf area dynamics and (ii) investigation of the elusive interaction between biotic stress and physiological senescence. The high-throughput nature of our methodology paves the way to conducting genetic studies of disease resistance and tolerance.

Keywords: deep learning, semantic segmentation, high-throughput field phenotyping, Septoria Tritici Blotch, disease tolerance

**EFFICIENCY OF THE MOLECULAR MARKERS LINKED TO THE STB
RESISTANCE LOCI IN WHEAT**

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Septoria tritici blotch (STB) is one of the most devastating foliar diseases of wheat, that accounts for approximately 70% of the annual usage of cereal fungicides in Europe. Nevertheless, the policy on the use of chemical plant protection products has been tightening recently and therefore much greater attention has to be put on finding fast and effective methods of identification of resistance genes and their introduction into breeding materials. One of the available solutions is Marker Assisted Backcrossing (MAB) method which essentially involves two steps: selecting individuals that carry the target allele (referred as foreground selection, FS) and next selecting individuals that carry the highest number of the recipient parent alleles outside the target gene (referred as background selection, BS).

In our study, we conducted the haploblock-based GWAS on a panel of wheat, comprising of lines/cultivars demonstrating diverse resistance to STB. The analysis led to choosing two highly resistant genotypes (Mandub and M3 Synthetic) for crossing with two susceptible cultivars (Arkadia and Patras). To investigate the presence of introduced loci and genetic background within offspring generations the DArTseq genotyping as well as SSR and AS-PCR markers assays were performed. The presentation will show the efficiency of various genotyping systems in foreground and background selection.

Keywords: association and linkage mapping, DArTseq markers, Septoria tritici blotch, SSR, *Zymoseptoria tritici*

FUNGAL INFECTION AND TOXIN CONTAMINATION OF SMALL GRAIN CEREALS IN HUNGARY

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Due to the effects of climate change, the appearance of filamentous fungi that prefer warmer weather conditions became more frequent, and many of them produce toxic secondary metabolic products. Therefore, it is important to constantly examine the occurrence of fungal species and monitor their toxin production.

In this study a total of 66 winter wheat samples from three different regions (South, East and Middle Hungary) were examined in 2021 and 2022. Sequence-based molecular method was used for the identification of the fungal species responsible for infection. A new multitoxin analytical method was developed for the measurement of mycotoxin contamination. By this method we determined fourteen toxins (e.g. trichothecenes, aflatoxins and fumonisins) simultaneously (using an Agilent 1260 Infinity II HPLC coupled with an Ultivo QQQ MS system).

The results revealed, that the average internal fungal infection level was 39.6% and 39.9%, in 2021 and 2022, respectively. However, the infection level was similar, significant differences were discovered in the distribution of fungal species. In South Hungary the average infection level was 56.2% and 46.2% during the two years survey. At the east part of the country the average fungal infection level was 39.3% and 42.7%, in 2021 and 2022, respectively. In both years the lowest infection level (25.8% in 2021 and 30.8% in 2022) was detected in the middle region of the country. We revealed a high frequency and the dominance of *Alternaria* species on winter wheat samples. The sequence-based identification results showed that these fungal species were responsible for the 73% of total infection level in 2021 and more than 88% in 2022. Moreover, we found *Penicillium*, *Cladosporium*, *Stemphylium*, *Rhizopus*, and other fungal species with relatively low frequency. *Fusarium* and *Aspergillus* species were found just in traces at all of the three locations and two years.

The measured grain toxin contamination levels did not reach the EU limit values of the great majority of the examined samples. However, in 2021, the detected average aflatoxin B1 level (3.6 µg/kg) was over the limit (2 µg/kg) in some samples derived from the south region. The trichothecenes were responsible for a significant part of the toxin contamination in these two years. We could not find significant correlations between toxin contamination and the level of fungal infection, so for maintaining the food safety the toxin contamination per se must be checked.

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METABOLOMIC CHANGES AS ONE OF THE UNDERAPPRECIATED SOURCES OF TISSUE CULTURE-INDUCED VARIATION IN TRITICALE

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The development of the plant *in vitro* techniques has brought about the variation identified in regenerants known as somaclonal or tissue culture-induced variation (TCIV). Studies in barley and partly in triticale have shown that S-adenosyl-L-methionine (SAM), glutathione (GSH), β -glucans, pectins, and Cu(II) ions may be involved in green plant regeneration efficiency (GPRE) and TCIV. These metabolites have been linked through structural equation models (SEM) to the biochemical pathways (Krebs and Yang cycles, glycolysis, transsulfuration), but not for triticale. Therefore, the study aimed to elaborate the statistical model of triticale regeneration efficiency utilizing metabolomic and (epi)genetic data.

Different concentrations of Cu(II) and Ag(I) ions were added to the induction medium of anther cultures for regeneration. We have also varied the time of plant regeneration. MetAFLP was used to study TCIV regarding DNA in symmetric (CG, CHG) and asymmetric (CHH) sequence contexts in the anther-derived regenerants and the donor plant. The metabolomic data concerning β -glucans/pectins, SAM, and GSH was obtained via Attenuated Total Reflectance–Fourier Transfer Infrared (ATR-FTIR) spectroscopy. A structural equation model was used to frame the data.

MetAFLP analysis showed that the average sequence change in the CHH context was 8.65%, and the amount of new methylation in the CHH context was 0.58%. The FTIR analysis delivered spectra for pectins (0.5512), SAM (3.9617), and GSH (0.00515). The GPRE mean value was 2.55 green regenerants obtained per 100 plated anthers. The model explains GPRE by linking the levels of pectin demethylation, SAM, *de novo* methylation, and GSH. Triticale's GPRE can be raised by changing the concentration of Cu(II) ions in the medium, which affects the amount of pectin.

Keywords: anther culture, triticale, pectins, GSH, SAM

THE ROLE OF *ORA59* TRANSCRIPTION FACTOR IN THE HORMONE-MEDIATED DEFENCE MECHANISM DURING *PYRENOPHORA TERES F. TERES* INFECTION IN BARLEY

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Barley (*Hordeum vulgare* L.) is in the four most important cereals in the world. Improvement of resistant cultivars is the most economic and eco-friendly method to control plant diseases. *Pyrenophora teres f. teres* (PTT), the causal agent of net form of net blotch disease of barley, is one of the most important fungal pathogens of barley.

Plant hormones have a key role in defence mechanism against plant pathogens. The defence reaction induced by salicylic acid (SA) and jasmonic acid (JA) are the two most important pathways, which enable the plant to respond to the pathogenic attack properly. Their interaction provides an opportunity to the fine-tuning of the response.

The APETALA2/Ethylene-Responsive Factor (AP2/ERF) superfamily of transcription factors (TFs) are involved in the responses to both biotic and abiotic stress. *ORA59* is one of the member of this family, which has been shown to increase its expression as a result of infection in the model plant *Arabidopsis*. The production of *ORA59* is stimulated by JA and repressed by SA. Therefore, we investigate the role of *ORA59* gene in defence mechanism against *Pyrenophora teres f. teres* (PTT) infection in barley, where it has not been studied, yet.

The resistance of an *ORA59* mutant *Arabidopsis* and a control (Columbia) against *Botrytis cinerea* were tested at first. The mutant proved to be more sensitive than the control, which confirm the role of *ORA59* in biotic stress response.

Two barley genotypes cv. Golden Promise and cv. Mv Initium was involved in the next experiment. The barley *ORA59* gene (HORVU4Hr1G000700.2 in Plant Ensembl) was identified based on Blast searches, it showed 69% AA identity to the *Arabidopsis* protein.

The PTT infection caused significant change in both hormones (p=0,01) in barley genotypes. The concentration of SA was the highest in the susceptible varieties and the lowest in the most resistant variety. While, the concentrations of JA were higher in the resistant genotype and lower in susceptible ones.

In our experiment, significant increase in the barley *ORA59* gene expression was observed after PTT infection. The connection between *ORA59* expression and PTT resistance of barley genotypes was studied by gene knock-out using CRISPR/Cas9 system. *ORA59* edited barley plants were produced by Agrobacterium-mediated transformation of immature embryos of Golden Promise. One of the transformed plant proved to be *ORA59* edited. The resistance of gene edited barley plants against PTT was characterized by detached leaves assay. As a result of PTT infection, necrosis appeared in the detached leaf test on the leaves, which were recorded according to the Tekauz scale. Based on the lesion type, the AUDPC curve of *ORA59* shoots was calculated. The three samples, unable to produce the transcription factor *ORA59*, showed significantly higher infection than the wild type.

Keywords: barley, *Pyrenophora teres f. teres*, plant hormones

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SELECTION OF WINTER WHEAT FORMS WITH INCREASED TOLERANCE TO PERIODIC WATER SHORTAGES

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The tolerance of crops to environmental stresses (both biotic and abiotic) determines the economic efficiency of farm production. Among the abiotic stresses, periodic water shortages are the main factor limiting plant production. In Central Europe, the leading causes of agricultural drought are groundwater deficiencies, exacerbated by periodic rainfall shortages, and the change in rainfall patterns from moderate drizzle to heavy rainfall, providing large amounts of water in a short time. Therefore, climate forecasts force us to serious engagement in efforts to improve the tolerance of plants to periodic water shortages. We will achieve this goal by developing and verifying screening methods for forms with more extensive roots and stable photosynthesis in field conditions.

One hundred fifty wheat cultivars and breeding lines consist of material for studies. The electrical capacity of the roots was measured with a capacitance meter at a voltage of 1 V between the ground electrode, inserted into the soil and the electrode covering the plant shoots, according to the method described by Cseresnyés (2018; <https://doi.org/10.3389/fpls.2018.00093>). The advantages of this method are simplicity and low price on the one hand and on the other hand, the potential resulting from the properties of the electric current (flowing almost exclusively through the absorbing and not suberized parts of the roots) to assess the functional size (activity) of the root system, including the hairs. An extensive root system is not only a factor determining water uptake but also better uptake of minerals from the soil and, in fact, better yield. Chl *a* fluorescence was measured to detect plant physiological state using PocketPEA portable fluorimeter (Hansatech Instruments, King's Lynn, Norfolk, UK). The results of the first season measurements in three replications per plot will be presented.

Keywords: drought, root, electrical capacitance, photosynthesis, *Triticum aestivum*

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REACTIONS OF SZEGED WHEAT VARIETIES TO DROUGHT STRESS

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Global temperature rise, an increase in the number of hot days, and extreme precipitation distribution cause significant crop losses in wheat production due to drought stress. In Hungary, the harvested wheat yield in 2022 was nearly a quarter less than the average of the previous five years. Water scarcity is especially critical during the tillering and flowering stages, which results in decreased yield and deterioration of its quality. Understanding the response reactions to heat and drought stress and identifying tolerant genotypes are key to breeding stress-tolerant varieties and thereby increasing crop stability.

In our experiments, we sought to answer which of the varieties derived from our breeding program have favorable drought stress resistance properties. The experiments were carried out between 2013 and 2022 under field conditions in our institute's automated rainout shelter, as well as in adjacent control treatments. In the study, we compared the drought stress response of 12 wheat varieties and one drought-tolerant control variety, *Plainsman V*, based on four parameters (heading date, plant height, thousand kernel weight and yield). In both treatments, each variety was represented by 2x3 one-meter-long, two-row (0.3 m²) plots. The heading date of the varieties was expressed in days from January 1 until the main stems of the spikelets had left more than half of the leaf sheath. Based on our experimental results, water-deprived plants headed on average two days earlier, which supports the literature data that plants respond to drought stress with earlier heading. As a result of the stress treatment, the plants reached an average height of 3 centimeters less than the control plots, but not all varieties showed a decrease in plant height. The thousand kernel weight was determined based on the measurement of 2x100 seeds. This trait showed a high degree of stability under water withholding, with varying responses among the varieties, but on average, we only measured a 1.38 g difference between the two treatments. The grain yield decreased for all examined varieties under unirrigated conditions, with an average decrease of 31% (10.51% - 52.61%). However, significant differences were observed among the varieties regarding the stress tolerance index (STI) and the yield loss. The drought-tolerant control variety reacted to stress conditions with a 25.79% yield loss, but 5 of the examined varieties performed better in the experiments (*GK Pilis* - 10.51%, *GK Hortobágy* - 15.77%, *GK Szilárd* - 16.47%, *GK Bagó* - 17.99% és *GK Megyer* - 22.04%).

Based on the results, it can be determined which varieties are suitable for cultivation in drought-prone areas, can be used for further detailed research on the effects of drought stress, and can serve as a basis for further breeding for drought tolerance.

Keywords: wheat, drought, rainout shelter, phenotyping, tolerance

This presented work was supported by VP4-10.2.2.-20 and TKP2020-NKA-21 projects.

**ASSESSING STRIPE RUST (*Puccinia striiformis* f. sp. *tritici*)
POPULATION STRUCTURE USING MOLECULAR METHODS**

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Yellow rust caused by the fungus *Puccinia striiformis* f. sp. *tritici* (Pst), may induce severe yield losses under favorable conditions. It is considered one of the most important pathogens of wheat. The risk of the decreasing quantity of yield depends on the frequency and speed of spreading of new fungus races. Knowledge of the pathogen population structure is one of the key elements of resistance breeding and sustainable agriculture.

In this study 48 single uredinium Pst isolates obtained from wheat, triticale and rye from six localizations in Poland in year 2022 were assessed. DNA from Pst isolates was extracted using CTAB method. Assessment of the isolates was carried out using two methods: analysis of SSR profiles for 19 loci (Ali et al. 2011) and MARPLE (Mobile And Real-time PLant disEase; Radhakrishnan et al. 2019).

The SSR method can be easily implemented in most laboratories using existing equipment and is a robust tool used to characterize isolate races. MARPLE allowed us to obtain much more data and information about the isolates, however it takes up more resources, both in consumable reagents as well as computing power and data disc storage.

The *P. striiformis* population in Poland was diverse. Both methods allow us for rapid identification of known races or new variants. Monitoring of the pathogen population and knowledge of its genetic structure is very important for crop protection and resistance breeding.

Keywords: MARPLE, SSR, yellow rust, pathogen

PHYSIOLOGICAL CHANGES AS EARLY RESPONSE IN INFECTED WINTER WHEAT (*TRITICUM AESTIVUM* L.) SEEDLINGS WITH LEAF RUST

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Leaf rust, also known as brown rust, is caused by the fungus *Puccinia triticina* Eriks., that can result in a significant threat to the grain yield and quality of wheat. Elevated disease severity, known as increased biotic stress pressure, correlates with increased accumulation of reactive oxygen species (ROS), where plants activate defence mechanisms against oxidative damage to regulate toxic levels of ROS. To improve our understanding of the physiological mechanism of wheat resistance to leaf rust, we set up the experiment with wheat seedlings of six genotypes differing in leaf rust resistance to elucidate interactions of this pathogen with carbohydrate and antioxidant metabolism of wheat genotypes. Obtained results showed that genotype with the highest severity of leaf rust symptoms decreased cytoplasmic invertase (cytInv) very early, 8 hours after inoculation (hai) with leaf rust. The downregulation of cytInv in susceptible plants may facilitate the maintenance of elevated apoplastic sucrose availability serving as nutrients for pathogen growth thus allowing spreading of symptoms more rapidly. The significant role in wheat seedling resistance to leaf rust can be attributed to vacuolar invertase (vacInv) that was supported by the fact that moderately resistant genotypes to leaf rust significantly increased vacInv when symptoms were fully developed. Also, vacInv were previously reported to assist in pathogen defence and scavenging of ROS. Simultaneously, glucose-6-phosphate dehydrogenase (G6PDH) and UDP-glucose pyrophosphorylase (UGPase) in moderately resistant genotypes decreased and further might restrict normal growth and development of pathogen due to reduced sugar content. The main contributors of leaf rust resistance in antioxidative metabolism were ascorbate peroxidase (APX) and catalase (CAT) activity resulting in less oxidative damage in the early stage of leaf rust development and thus maintaining a higher antioxidant capacity resulting in lower oxidative damage. Thereby, carbohydrate and antioxidant mechanisms acted mutually in defence of wheat seedlings during leaf rust stress. In addition to well-known antioxidative defence systems, carbohydrate metabolism can now be recognized as a crucial mechanism in coordinating plant developmental responses under leaf rust occurrence in wheat.

Keywords: leaf rust, physiology, wheat seedlings

DOES SCLR67APR GENE CONFERS SEEDLING STAGE RESISTANCE OF RYE TO LEAF RUST?

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One of the most important rye disease in Europe is brown rust (LR), caused by *Puccinia recondita f. sp. secalis* (Prs). Despite many years of extensive studies, the genetic background of the immune response of rye to LR, both determined by R genes at seedling stage and APR (Adult Plant Resistance) genes at adult stage, is still not well understood. The polymorphisms in two genes – *Lr34* and *Lr67* have been shown to be associated with APR in wheat and barley. Moreover, both genes also confer the resistance to LR at seedling stage.

The aim of this work was to find in rye genome the orthologue of *Lr67* and to verify its function as the APR conferring gene at seedling stage. Firstly, we tried to find associations between *ScLr67* gene structure and APR/non-APR phenotype in a large population of rye inbred lines selected based on precise phenotyping done at two locations in two subsequent vegetation seasons. Using as a criterion index of severity, we selected several APR and non-APR lines for structural analysis. In the first stage of research, we identified as many as 31 variants of *Lr67* gene (named *ScLr67-1* ÷ *ScLr67-31*) in the reference genome of rye inbred line Lo7. To select the most interesting variants for further analysis, we used the transcriptomic data from parallel experiments whose goal was to find genes differentially expressed after Prs infection. The selection was also supported by additional bioinformatics analysis. Finally we have chosen 5 variants for detailed structural analysis and transcriptional (qRT-PCR) analysis. We have shown, that the expression levels and profiles of *ScLr67* variants are affected by LR infection at the seedling stage. Those changes depend on several factors, such as plant genotype and phenotype, *Prs* strain and time post treatment. Our study indicates that the gene *ScLr67* gene may be involved in the response of rye to LR at the seedling stage. The research was financed by the National Science Centre, Poland, project No. 2018/31/B/NZ9/00439.

Keywords: rye, brown rust, Adult Plant Resistance, *ScLr67*, seedling stage resistance

CHANGES IN POLYAMINE CONCENTRATION IN MAIZE SEEDLINGS DUE TO *FUSARIUM* INFECTION

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The different *Fusarium* species are pests of many of our cultivated crops and are of major importance from both human and animal health points of view due to mycotoxin production. It is estimated that these microscopic fungi are plant pathogenic organisms causing diseases in 80% of our cultivated crops, including maize. The two most important pathogens damaging maize are *Fusarium graminearum* Schwabe [teleomorph = *Gibberella zeae* (Schwabe)] and *Fusarium verticillioides* [teleomorph = *Gibberella moniliformis* (Wineland)].

Previous research has shown that polyamines found in all living cells play a crucial role in responses to various biotic stresses. However, the biosynthesis of polyamines is of paramount importance not only for plants but also for their pathogens in promoting stress tolerance and pathogenicity.

In our experiments, we investigated the changes in polyamine content induced by isolates of two maize-damaging *Fusarium* species (*F. verticillioides* and *F. graminearum*) in maize seedlings of different susceptibility genotypes. In addition, we investigated how infection efficiency and changes in polyamine content are modified by distilled water, salicylic acid or putrescine pretreatments.

Our results confirmed that initial- and stress-induced changes in polyamine content are not directly related to tolerance in either maize coleoptile or radicle. However, there are differences in the life cycles of the two pathogens: *F. verticillioides* coexists with maize as an endophytic pathogen in most cases, whereas *F. graminearum* behaves as a necrotrophic microorganism during its life cycle. As a consequence, the two *Fusarium* species induced different changes in polyamine concentrations. The effect of salicylic acid and putrescine treatments depended on both the pathogen and the plant resistance: against *F. verticillioides*, both salicylic acid and putrescine seed treatments produced positive results, whereas in the case of *F. graminearum* infection, seed soaking in distilled water alone positively affected biomass parameters in the tolerant genotype. The accumulation of putrescine in parallel with the decrease in spermidine/spermine content and, concomitantly, the reduced catabolism of the latter could inhibit the necrotrophic pathogen *F. graminearum*. At the same time, the accumulation and metabolism of polyamine leads to an increase in H₂O₂ content, which prevents the spread of the endophytic pathogen *F. verticillioides* in plants.

Our results suggest that the role of salicylic acid and the polyamines tested may be important against *Fusarium* spp. infections, but further information and studies are needed to understand the polyamine-related resistance and changes in polyamine metabolism.

Keywords: polyamine, fusarium, maize, putrescine, salicylic acid

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THE EFFECT OF LONG-TERM MEDIUM DROUGHT ON THE DEVELOPMENT OF WHEAT (*TRITICUM AESTIVUM* L.) GRAINS

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Wheat is one of the most important cereal crops in the world. In Europe common wheat is cultivated on more than 60 million hectares. The demand for wheat production is still increasing due to the global population growth, however, climate change is becoming a major cause of yield reduction. During growth and development, plants are exposed to various kinds of biotic and abiotic stresses, and drought is the most important from the agricultural point of view because the drought stress causes significant losses in the quantity and quality of grain yield. In most studies, the drought is intense, but is applied in a short period of time. However, under agronomic conditions, plants are often exposed to the milder, uninterrupted stress of drought.

The main goal of the project was to identify genes and microRNAs that contribute to plant's response to medium, long-term drought stress in developing grains of two bread wheat (*Triticum aestivum* L.) cultivars – drought-sensitive and drought-tolerant. For this purpose, at the end of leaf development (BBCH 29), the plants were adapted to drought stress by gradually reducing the irrigation from optimal soil moisture for the plants to 40% soil hydration. This drought stress was applied continuously until harvesting. The control plants were grown under optimal soil moisture conditions. RNAseq and smallRNAseq analysis was performed at two time points during grain development, 7 DAP and 21 DAP. The results highlight the wide variation in gene expression between drought-tolerant and drought-sensitive cultivars.

Keywords: RNAseq, drought, miRNA, transcriptomic analyzes

OBTAINING DIHAPLOID LINES OF SOFT WINTER WHEAT WITH COMPLEX RESISTANCE TO RUST AND COMMON BUNT BY ANTHHER CULTURE *IN VITRO*

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To develop pre-breeding material for winter wheat, nine lines (F₄-F₅) with different genetic backgrounds (complex hybrids) resistant to major diseases were developed at the Department of Phytopathology and Entomology of the SGI-NCNS. The aim of this research was to produce homozygous dihaploid lines that contain a complex of effective disease-resistance genes (types of rust and common bunt) through *in vitro* androgenesis.

The culture of isolated wheat anthers was used to produce double haploids. For each genotype, a percentage of callus formation and green plant regeneration was calculated based on the number of anthers planted. Following the adaptation to soil conditions, wheat regenerants were vernalized and grown in artificial climates. The percentage of fertile plants (DH) was determined by dividing the number of germinated plants by the total number of plants. The following dihaploid lines were obtained in 2021: hybrids 2/20, 3/20, 120/20, 132/20, and 352/20. A field nursery was evaluated in 2022 for its resistance to three rusts (leaf, stem, and yellow) and common bunt infections induced by artificial inoculation. There were differences in the frequency of callus formation (from 0.99 to 16.26 percent of planted anthers) and in the ability to regenerate plants (from 0 to 3.05 percent of planted anthers) in the process of androgenesis *in vitro* of soft winter wheat. The anthers culture produced fertile regenerants in 5 of the 9 hybrids evaluated. Based on the received regenerants, the effectiveness of the last process varied from 12.5 to 60.0% (an average of 29.41%). The obtained dihaploids demonstrated high resistance (8-9 points) to a complex of diseases. Microspores in soft winter wheat exhibited genotype-specific morphogenetic reactions during androgenesis *in vitro*. Among the samples 120/20 and 132/20, the greatest amount of callus formation was observed. We obtained fifteen dihaploid lines with complex resistance to rust and common bunt.

Keywords: dihaploid, wheat, androgenesis *in vitro*, resistant, rust

INCREASING OF WHEAT RESILIENCY TO VIRAL AND FUNGAL DISEASES UNDER CLIMATE CHANGE CONDITIONS

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Ukraine is one of the top five grain exporters in the world. However, the ongoing war waged by Russia against Ukraine disrupted the global supply chains of food products from Ukraine in 2022. Several difficulties have resulted from these wartime realities, including a decrease in sown areas due to active hostilities and the occupation of territories. As well as this, some minefields are not cultivable and are a source (reservoirs) of harmful organisms (insects – viruses vectors) such as insects, viral vectors, and plants infected with viral and fungal diseases. In recent years, wheat in Ukraine has been damaged by *Wheat streak mosaic virus* (WSMV), *Barley yellow dwarf virus* (BYDV), and *Wheat dwarf virus* (WDV). These viruses have a high epidemic potential, leading to a 30-50% decrease in harvest and quality. Virological monitoring has shown that the ‘Smuglyanka’ and ‘Donska semi-dwarf’ wheat varieties were usually affected by WSMV at moderate moisture. In later years, with the increase in drought frequency after 2013, these varieties were also infected with BYDV. In 2020, WDV appeared mainly on new varieties such as ‘Patras,’ ‘Producent,’ ‘Matrix,’ and lines from the V.M. Remeslo Myronivka Institute of Wheat. We further observed that wheat exhibited an adaptive response to environmental factors, such as temperature differences between May and June, similar to the pathologies caused by phytoviruses – this is the appearance of "crimson" leaves. Mutations in the nucleotide sequences of phytoviruses can alter the harmfulness of the virus. For example, the unique amino acid substitutions of the sequences of the Ukrainian isolates of WSMV and the higher divergence of some of them are different from those of isolates from other countries. Therefore, developing and using resistant wheat varieties is almost the only option for increasing food security under climate change and war conditions in Ukraine.

Keywords: *Triticum aestivum* L., viral and fungal diseases, resiliency

AN EVALUATION OF DISEASE RESISTANCE, DROUGHT TOLERANCE, AND AGRONOMIC TRAITS IN WINTER WHEAT INTROGRESSION LINES

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Global climate change facilitates the spread of diseases in winter wheat (*Triticum aestivum* L.) and increases the yield losses caused by a combination of these diseases and drought. Preventing these losses depends on identifying and introducing resistance genes into wheat cultivars. To create wheat lines carrying valuable alien genetic material, a number of advanced introgression lines have been developed from complex interspecies crosses. This study aimed to evaluate resistance to diseases, drought, and agricultural traits among these lines.

Field trials were conducted during the crop seasons of 2018-2022. Depending on the year, the winter wheat diversity panel consisted of 24 to 153 introgression lines and two check cultivars (Kuyal'nyk and Nasnaga) suitable for the arid climate zone of southern Ukraine. Data on eight agronomic traits (quantitative scores) and five pathogen response traits (point scores) were collected and subjected to variance and correlation analysis to determine the contribution of individual genetic backgrounds toward plant resilience.

The majority of the lines showed resistance to rust species. The distribution of responses in the analyzed lines was bimodal in response to rusts and unimodal in response to powdery mildew or Septoria leaf blotch. The resistance traits displayed no correlation with heading date, plant height, or protein content. The yield traits did not correlate with the resistance to diseases except for yellow rust ($R_{sp} = 0.19^* \dots 0.26^{***}$), stem rust ($R_{sp} = -0.03 \dots 0.34^{**}$), and Septoria blight ($R_{sp} = 0.15 \dots 0.27^{***}$), which are possibly the most harmful diseases in the southern part of Ukraine. Under drought conditions, the yield is reduced by 32.5% to approximately 70%, depending on the year. The drought resistance index showed a direct correlation with the yield in arid conditions ($r = 0.69^{***}$) and correlated with the yield under favorable conditions ($r = -0.26^{**}$). A negative correlation was observed between quality traits and grain yield. Specifically, the correlation between protein content and grain yield varied from $r = 0.01$ to $r = -0.45^{***}$, depending on the year, and the negative correlation between these traits increased under drought. However, the quality traits mainly correlated positively with each other.

The high disease resistance trait correlated with low yield, reduced protein content, or small grain size. The frequency of lines combining pathogen resistance was low, as measured by total yield with grain quality. However, the introgression lines provide a source of genes for improving winter wheat disease and drought resistance.

Keywords: *Triticum aestivum*, introgression lines, resistance, agronomic traits

GENETIC BASIS OF RESISTANCE OF WINTER WHEAT TO MAJOR FOLIAR DISEASES

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To select wheat for immunity, it is necessary to have access to source materials, such as resistant varieties, lines, and forms of the crop, which act as donors of resistance with highly efficient genes. A primary focus should be placed on identifying donors of resistance to the most prevalent and harmful disease agents, including rust (leaf, stem, and yellow), loose smut, and powdery mildew, which are the most detrimental diseases to wheat in Ukraine.

A total of 69 *Lr*-genes to leaf rust, 83 *Yr*-genes to yellow rust, 62 *Sr*-genes to stem rust, 21 *Bt*-genes to common bunt, 7 *Ut*-genes to loose smut, and 56 *Pm*-genes to powdery mildew have been identified. Most of these genes have been rendered ineffective by virulent and aggressive races and biotypes of the above-mentioned diseases.

Based on previous studies, we have shown that efficiency is still low in Ukraine. The Ukrainian population retains 11 – *Lr*, 1 – *Yr*, 8 – *Sr*, 6 – *PM*, 8 – *Bt*, and 2 – *Ut* genes. Among them are those genes that we first identified.

Some genes have been identified as effective against leaf rust races, including *Lr9*, *Lr19*, *Lr24*, *Lr37*, *Lr42*, *Lr47*, *Lr51*, *Lr52*, *Lr53*, *Lr56*, *Lr64*, and the combination of *Lr26* and *Lr34*. It has been found that the *Yr9* gene showed resistance against yellow rust races, whereas the *Sr21*, *Sr24*, *Sr26*, *Sr27*, *Sr31*, *Sr38*, and *Sr39* genes were effective against stem rust races. Several genes have also been identified as effective against powdery mildew, such as *Pm3c*, *Pm4a*, *Pm4b*, *Pm17*, *Pm20*, *PmPI 170911*, and their combinations *Pm3a* with *Pm3c* and *Pm17* with *Pm38* and *Pm39*. Our wheat lines contain a number of the above-mentioned genes (*Lr24*, *Lr26* + *Lr 37*; *Sr24*, *Sr31*; *Pm17*, *PmPI 170911*, *Pm17* + *Pm38* + *Pm39*).

The lines were developed by us with the addition of wild wheat relatives (*Aegilops* species, *Tr. erebuni*, etc.), and they were shown to be resistant to leaf, stem, and yellow rusts, powdery mildew, and loose smut.

Line 4/16 carrying *Aegilops cylindrical* introgression carried *UtAc1* and *UtA2* resistance genes for loose smut and *BTAc1* and *BtAs2* resistance genes for the common bunt. Moreover, line 13/16 with *Aegilops variabilis* introgression was identified with *UtAv1* and *UtAv2* resistance genes to loose smut and *BtAv1* resistance gene to common bunt.

Plant pathologists and breeders are primarily responsible for searching for and identifying effective resistance genes. Wild wheat relatives are the main source of these genes. Pre-breeding material containing their additional efficient genes of resistance against winter wheat diseases forms the basis of modern selection for stability.

Keywords: *Aegilops* spp., disease resistance, rusts, wild relatives, wheat improvement

EFFICIENCY OF DIFFERENT GENETIC SYSTEMS OF RESISTANCE TO LEAF RUST IN THE STEPPE ZONE OF UKRAINE

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Creating varieties with combined (pyramidal) resistance to various pathogens is one of the main challenges facing modern breeding science in intensive grain growing technologies. According to the FAO (Food and Agriculture Organization of the United Nations), crop losses caused by diseases can reach 30%.

This work presents a study of the possibility of expanding the resistance of winter bread wheat (*Triticum aestivum* L.) to the most common rust disease - leaf rust (*Puccinia triticina*) using the method of gene pyramiding from different genetic resistance systems in order to increase the level of economically valuable parameters of winter bread wheat.

The research was conducted in 2021-2022. The evaluation of the breeding material consisted of two stages: 1) in winter, the resistance of soft winter wheat to brown rust in the juvenile period of development (on seedlings) was assessed under artificially created infection background; 2) in the summer, in the phase of an adult plant on an artificially created hard infectious background in the field. The source material was 349 lines of competitions (I-II) from different crossing schemes, which were grouped into genetic systems depending on the ecological and geographical origin and the combination of resistance genes in one genotype, such as: "Serbia-Odesa", "Lr34", "Western Europe", "CIMMIT-ICARDA-Turkey", "Translocation (1BL/1RS)+Lr34+Western Europe", "*Aegilops tauschii*".

According to the results of evaluations at different stages of development in the laboratory and field, it was found that the most effective systems were "Lr34" and "Western Europe", as well as the combined genetic system: "Translocation (1BL/1RS) + Lr34 + Western Europe". The lines included in the mentioned systems showed a quite high level of resistance up to 6-7 points and yields of 5-7 t/ha, regardless of the year of cultivation and the impact of abiotic and biotic factors.

Keywords: bread winter wheat, brown rust, genetic systems resistance, pyramiding, yield

SESSION 4

Environmental adaptation, crop management

BREEDING TRITICALE FOR MORE SUSTAINABILITY IN OUR CROPPING SYSTEMS

Geert Haesaert, Veerle Derycke, Szanne Degraeve, Sofie Landschoot, Riccardo Zustovi, Anneleen De Zutter, Kevin Dewitte

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Triticale (\times *Triticosecale* Wittmack), the intergeneric hybrid of wheat (*Triticum* spp.) and rye (*Secale* spp.) gets more and more attention because the effects of climate change and the drive for more sustainability in cropping systems. Indeed, triticale's properties such as a good tolerance to abiotic and biotic stress, an excellent kernel and forage yield-level and a good fit in intercropping systems will play an important role to mitigate our cropping systems in future. This keynote lecture highlights the followings traits:

Triticale as a high-potential forage crop

With the expectation of more frequent dry summers in temperate climate zones, winter cereals as triticale becomes an attractive alternative to maize, a well-established forage crop on dairy farms. Contrary to maize, winter cereals such as winter triticale are hardly affected by summer dryness because they rely mainly on winter soil moisture to produce biomass during spring. However, triticale forage quality is generally inferior to that of maize forage and research to improve its forage quality is lagging behind that of other forage crops. The results of a study that investigated the genetic control of the *in vitro* digestibility of organic matter and related traits in soft dough triticale forage using a GWAS approach will be presented. Phenotypic data of a panel of 118 winter triticale genotypes were used in combination with a set of DArTseq SNP markers.

Breeding triticale for intercropping with faba beans

There is a growing interest for growing winter cereals with winter legumes in intercropping systems. Until today the focus laid on varieties, sowing density, N and P fertilization, dry matter yield and LER-values but no breeding strategies to become genotypes which are better adapted to intercropping were developed. A first attempt to characterise the G x G interactions and the underlying properties e.g., the effect of phyllotaxis, plant length, sprouting, plant root interactions will be presented.

Vulnerability to drought-induced xylem embolism as a proxy for drought resistance

Multiple techniques are combined to apprehend the different aspects of xylem vulnerability to drought and for making possible to link the results to the different genotypes tested. Through continuous measurements of AEs during bench-top dehydration, AE₅₀ values are quantified and vulnerability to drought-induced xylem embolism assessed. Further, hydraulic capacitance and capacitive water release during dehydration are derived from desorption curves after continuously weighing of samples. To corroborate these results, xylem anatomical traits linked with a species' individual trade-off between hydraulic safety and efficiency are determined. This set of techniques is used to assess vulnerability of wheat, triticale and rye to drought-induced xylem embolism and to study the possibility as a proxy for drought resistance in breeding programs.

USING FIELD-BASED PHENOTYPING TO EXPLORE THE TEMPERATURE RESPONSES OF GROWTH: FROM OVERGENERALISING TO OVERFITTING OF DOMAIN KNOWLEDGE IN CROP MODELS?

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The ability to predict plant development is a long-desired but nontrivial objective in crop physiology.

The challenges posed by global climate change reinforce the need for reliable estimations. Plant development modeling involves predicting two main, interwoven components: (i) Growth, and (ii) Phenology. While temperature is undoubtedly the main driver of growth, evidence of cultivar specific differences in growth response to temperature is so far sparse. Still, for phenology, severe genotyp-by-environment interactions are commonly observed. It is uncertain if parts of these interactions are due to genotyp-specific responses to temperature. In this work, we evaluated a large outdoor winter wheat and soybean data set and extracted both growth rates and phenology stages. The data was collected with temporally resolved plant organ tracking devices as well as high-throughput field phenotyping devices in multiple years. The results provide indications that genotyp-specific differences in growth rates are very subtle, but essential to understand genotype-by-environment interactions. We found genotyp-specific temperature responses for the monocotyledonous species winter wheat as well as the dicotyledonous species soybean. Nevertheless, when applying these trained models to less related test sets—i.e., to data of other growth periods or plant organ levels—their performance dropped. Species level models and the baseline model 'thermal time' showed much better generalization abilities. Still, large genotyp-by-environment effects were observed when expressing the timing of phenology for winter wheat with thermal time. Replacing thermal time with genotyp-level temperature responses could reduce the observed genotype-by-environment interactions in phenology by up to 75%. We conclude that while models with high generalization ability such as thermal time can predict growth with high precision, the incomplete modeling of cultivar specific temperature response introduces artifacts for downstream genotype-by-environment analysis. With the quality of field phenotyping data available today, cultivar-specific temperature responses can and should be integrated in advance, thus eliminating confounding effects when observing responses to other environmental conditions such as for example drought stress.

Keywords: temperature response, phenology, plant growth modeling, GxE

DISSECTION OF PHENOLOGY AND ITS EFFECTS ON YIELD IN A BUSTER-CHARGER MAPPING POPULATION IN THE FIELD

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Extending the duration of the late reproductive phase, i.e. the stem elongation (SE) between terminal spikelet stage (TS) and anthesis, has often been proposed as an avenue to increase yield in wheat. However, accurate determination of the SE duration is difficult and labour intensive, as precise timing of the TS stage requires the dissection of the shoot apex meristem. Existing evidence on advantageous yield effects of an extended SE phase is thus often based on a limited number of genotypes observed under controlled conditions. Studies under field conditions are rare and often resort to proxy-measures to determine the start of SE.

In this study, a Buster x Charger population comprising 108 doubled haploid lines was grown across four year-sites in the field. TS was recorded through meristem dissection and SE duration was measured as the time between TS and ear emergence (EE). Mixed model analysis across year-sites revealed high heritabilities ($H^2_{\text{yield}} = 0.83$, $H^2_{\text{TS}} = 0.97$, $H^2_{\text{SE}} = 0.95$, $H^2_{\text{EE}} = 0.98$) and strong genetic correlations between yield and the phenology traits SE and EE ($r_g = 0.53$ and $r_g = 0.6$, respectively). Furthermore, SE duration appeared to be driven by EE ($r_g = 0.86$) rather than TS ($r_g = 0.32$), even though a strong correlation was found between EE and TS ($r_g = 0.76$). The genetic correlations were reflected in the results of a QTL analysis. One common QTL each was found for yield and EE (Chr 2D), yield and SE (Chr 7A), as well as for EE and SE (Chr 4A). Apart from that, two independent QTL each were detected for TS (Chr 3A and 7A) and SE (Chr 1D and 2B).

Together, these results confirm the proposed positive relationship between SE duration and yield under field conditions based on accurate SE duration measurements. Furthermore, independent QTL for TS and SE suggest that extending SE without affecting heading time is possible. Even though further research is warranted to confirm the QTL discovered here in more diverse germplasm, the QTL for TS and SE may serve breeders to select for TS and SE via marker assisted selection.

Keywords: terminal spikelet, stem elongation, yield, QTL mapping, genetic correlations

THE EFFECT OF VERNALIZATION GENES ON COLD TOLERANCE IN DIVERSE WHEAT GERMPLASM

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Vernalization is a crucial process for winter wheat to transition from vegetative stage to reproductive stage. This process is regulated by Vernalization (Vrn) genes, which have played a pivotal role in wheat adaptation. The primary aim of this study was to investigate the effect of Vrn genes on cold tolerance. To determine this, 175 winter wheat genotypes of diverse origins, including materials from the International Winter Wheat Improvement Program (IWWIP), Turkish, Eastern European countries and USA cultivars, and others were tested in temperature-light controlled cold chamber. The experiment was conducted twice, wherein the genotypes were exposed to cold treatment at -15, -17, and -19 °C, with three replications in each trial. The cold treatment implemented with modified Tischner et al. (1999) method, and cold hardiness was evaluated by measuring survival rates of each genotype in each cold treatment. The study evaluated the effect of individual Vrn genes and combinations of them on cold tolerance. The results showed statistical differences in cold tolerance among the genotypes, with the most cold-tolerant genotypes found among Eastern European and USA cultivars. However, these differences were not significant among the groups at -15 °C while they were more prevalent at -17 and -19. When Vrn genes combinations were considered, the differences among the Vrn gene combination were not significant at -15, the highest cold tolerance was observed in Vrn-A1_springClaire-typeJagger-typevrn-B1_winterVrn-D1a followed by vrn-A1_winterHereward-typeJagger-type:2147-typevrn-B1_wintervrn-D1 and vrn-A1_winterHereward-typeJagger-type:2147-typevrn-B1_winterVrn-D1a at -19 °C, while the lowest survival rate was in vrn-A1_winterClaire-typeJagger-typeVrn-B1_springVrn-D1a combination.

Keywords: winter wheat, vernalization, cold tolerance

FROST DAMAGE INDEX: THE ANTIPODE OF GROWING DEGREE DAYS

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Winter crops are sown in autumn, withstand even freezing temperatures at early developmental stages and start into the vegetation period with a considerable biomass. It is obvious that frost induces senescence of vegetative organs, but the timing and severity of such processes are largely unknown. High-Throughput Field Phenotyping methods have recently opened the possibility for quantitative analysis of such processes. For crops such as winter wheat it is highly relevant to analyse the biomass with which they start into the new vegetation period in spring. The results of this study show that there is a linear correlation between the product of freezing temperatures and the period of endurance and the loss of canopy surface. We harnessed this relationship to construct an index termed „forst damage index” (FDI) that quantifies frost damage. Similar to the concept of growing degree days (GDD), it is the product of exposure time and amplitude of temperature beyond a base temperature that determines the physiological effects. In contrast to GDD, however, a certain delay time has to be taken into account, after which the senescence process becomes visible via bleaching of the leaf tissue. The FDI may be applied to determine winter hardiness of wheat cultivars and therefore is of immediate relevance for breeding. The concept of FDI could be adapted to other factors such as drought or heat stress. While commonly not considered in plant growth modelling, integrating such degradation processes may be key to improving the prediction of plant performance for future climate scenarios.

Keywords: abiotic stress, breeding, crop growth modelling, high-throughput field phenotyping, winter wheat

THE IMPACT OF DIFFERENT PLANTING DATES ON PROTEIN QUALITY AND QUANTITY AND GRAIN YIELD OF MAIZE

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Maize is the primary source of protein for low-income populations who depend exclusively on maize-based diets. Due to different factors, farmers are often forced to plant later than the ideal planting date, but how this affects the grain quality and yield needs to be clarified. This study aimed to determine the grain yield, protein content, and zein composition of six maize hybrids at three different planting dates over two seasons. The experiment was conducted at Bloemfontein, South Africa, using a randomized complete block design with three replications. The effect of the planting date was highly significant for all measured traits. The hybrid effect was highly significant for starch, α -, β -, γ - and δ -zein, but not for protein content and grain yield. A significantly lower yield at planting date three compared to planting date two led to a significant decrease in starch content and an increase in protein content at planting date three, with an associated decrease in γ -zein and increase in α -zein. The first and second planting dates gave good yields, with an actual increase in yield at planting date two, but with a sharp decline in yield at planting date three. To conclude, farmers could plant at either the first or second planting date, but the third planting date should be avoided at all costs. The second planting date had similar protein content as the first planting date, but protein content was increased in the third planting date compared to the second, with significantly higher α -zein content, and lower γ -zein content.

Keywords: maize, planting windows, protein, starch, zein fractions

FAR-RED AND BLUE LIGHT ENRICHMENT DIFFERENTIALLY INFLUENCE THE METABOLISM AND FROST TOLERANCE OF BARLEY

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During cold acclimation (typically 2-7 weeks), plants undergo an intensive transcriptional and physiological remodelling to tolerate freezing temperatures. Before the full development of the process, plants are more susceptible to freezing; however, a shorter cold period could also trigger a pre-hardening effect. The alteration of the light spectrum, e. g. by the decrease of Red : Far-red (R:FR) ratio of the incident light also induce cold acclimation even at warmer temperatures. More recently, it was reported that the addition of blue light into the incident FR enriched white light (W) further improve the frost tolerance of barley. The increment of blue light proportion significantly altered both the membrane lipid composition and metabolite content of the treated leaves. Both soluble carbohydrates and free amino acids participates in cold acclimation, however, the available information about the impact of the modified light spectrum on some key metabolites of cold acclimation in cereals is sparse. Thus, in this work, the effects of W supplemented with FR (WFR) or FR + blue light (WFRB) (with different emission peaks, 410 nm [B₄₁₀] and 450 nm [B₄₅₀]) at different temperatures (15 °C and 5 °C), for 6h 10 (15 °C) or 7 days (5 °C), on 2-week-old winter barley plants (cv. ‘Nure’) were investigated. All plants were grown in plant growth chambers equipped with controllable LED light ceilings. The freezing tolerance of barley plants was evaluated by measuring electrolyte leakage of detached leaves after freezing. The concentrations of some key metabolites contribute to frost tolerance and the expression of related genes was also investigated. As expected, FR enrichment increased the frost tolerance of barley leaves, and WFRB treatment was enhanced it further. Interestingly, the effect of blue light supplementation showed spectral and temperature dependence even in this mixed light environment. At 15 °C, which is considered as non-cold acclimation inducing temperature, enrichment of WFR with B₄₁₀ provided significant enhancement for frost tolerance, but at 5°C, B₄₅₀ supplementation also triggered this effect, although, B₄₁₀ was more effective. Interestingly, at 15 °C, WFRB light modified the levels of more metabolites, than in the case of WFR, while at 5 °C, the FR effect showed to be more pronounced after 6 h. Furthermore, the addition B₄₅₀ light to the WFR mixture negated the enhancing effect of FR on some soluble carbohydrates after 7 days of cold treatment. This negative effect was not observed in the case of B₄₁₀ light. In summary, FR and FR+B light enrichment of the incident W light exerted a fine-tuning effect on the concentrations of these metabolites and the expression of related genes. Interestingly, in the case of some metabolites, B enrichment of the WFR mixture negated the effect of FR or induced an opposite effect.

Keywords: far-red light, blue light, metabolism, cold acclimation, frost tolerance

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EFFECTS OF PPDH1 AND VRNH2 ON YIELD AND DEVELOPMENT OF SPRING BARLEY

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Previous research in our group indicated a possible agronomic advantage conferred by genes involved in the control of phenological development of barley (*Hordeum vulgare* L.). These findings led to suggest some hypotheses that will be experimentally tested in several sets of near-isogenic lines (NILs). Here, we present the results of experiments carried out with one of these sets, four sister lines that were developed at CSIRO Agriculture and Food (Canberra, Australia), obtained by repeated backcrossing of donors carrying a spring VRNH1 allele (VRNH1-7), an active VRNH2 allele, and a recessive ppdH1 allele onto a recipient line *vrnH1/vrnH2/PPDH1*, creating 8 NILs nearly identical, except for a small chromosomal segment surrounding the three genes of interest. Four of these isolines (C05-C08) are spring type, with no vernalization requirement due to the presence of the VRNH1-7 allele, and differ in VRNH2, a long-day expressed repressor of flowering that normally blocks flowering before overwintering, and in PPDH1, the major determinant of long photoperiod response. These lines were grown over three seasons at two different locations in Spain to evaluate the combined effects of diverse environmental conditions, and presence/absence of genes of interest on phenology, development and yield components. In addition, these lines are currently grown in controlled conditions at 16h light, 20°C/8h dark, 16°C, to describe plant morphology, and the developmental progression of the main shoot apex (MSA). This work will shed light on the effects of variations at VRNH2 and PPDH1 alleles on plant and shoot apices development, and their effect on agronomic responses, providing insights for future research on the complex relationship between reproductive development and yield in barley.

Keywords: barley, development, photoperiod, vernalization, yield

STRUCTURAL VARIABILITY AND FUNCTION OF *VERNALIZATION1* IN CEREALS

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The flowering time plasticity in wheat and barley is secured by different interconnected genetic pathways, including the cold-induced vernalization pathway. *VERNALIZATION1* (*VRN1*), which determines the growth habit and duration of the vernalization requirement, belongs to the key players of the pathway. Ancestral alleles without mutations confer winter growth habit and require vernalization to accelerate flowering. Independent insertions and deletions in the promoter or intron 1 of the *VRN1* gene gave rise to spring genotypes, in which *VRN1* is transcribed without vernalization. In such genotypes, the putative repressor binding site is disturbed by mutations. We inspected the natural variability of *VRN1* in domesticated cultivars and their wild progenitor species to study the evolutionary aspects of vernalization. We analyzed the expression profile of novel alleles and examined the heading time of these genotypes. Moreover, we developed barley reporter lines to study the function of the VRN1 protein: (i) VRN-H1 fused with the green fluorescent protein (GFP) to monitor tissue-specific expression using confocal microscopy, and (ii) VRN-H1 fused with the β -Glucuronidase (GUS) reporter gene to detect the signal in the chlorophyll-containing tissues. Such reporter lines could be used for generating various deletion to analyze *VRN-H1* cis-regulatory elements and setting up a new forward-directed genetic screen to identify additional VRN-H1 regulators. The study of vernalization and characterization of different alleles of the *VRN1* gene can be used in the breeding process of wheat and barley.

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RESULTS OF UAV-BASED PHENOTYPING FOR SPRING WHEAT GENOTYPES ACCROSS CONTRASTIG GROWING CONDITIONS

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Phenotyping nitrogen use (NU) traits is labor intensive, time consuming, and requires destructive biomass harvest and the destruction of experimental plots. The unmanned aerial vehicle (UAV) based non-destructive phenotyping has given a great opportunity to assess plants growth by capturing different bands (blue, NIR, red, green, and red-edge) of light spectrum and have been validated as complementary platforms for several traits like green cover, biomass, nutrient status in plants, chlorophyll level, and photosynthesis rate. The objective of this study is to assess the UAV-derived multispectral vegetative indices (VIs) most useful for estimating grain yield and NU traits in the context of a spring wheat breeding program. This study evaluated 5 VIs (RNDVI, GNDVI, NDRE, RECI, NGRDI) to test the variation and their relationships with grain yield (GY) and NU traits (NU_pE, NU_tE, NUE). The field trial was located at the Stende Research Centre, Institute of Agricultural Resources and Economics, Latvia (57.18927N 22.56187E). Sixteen spring wheat cultivars were evaluated under two N fertilization levels over two consecutive seasons (2021 and 2022). The trial was conducted as a split plot factorial design with two randomized replicates with N fertilization levels applied before sowing [low N (LN); 75 kg N ha⁻¹; high N (HN); 150 kg N ha⁻¹] as main plots and 16 wheat genotypes as subplots with size of 10 m². Total N in soil (0-20 cm depth) in early spring and mature plant samples were determined by Kjeldahl method. DJI Phantom 4 Multispectral was used for imaging at 20 m altitude maintaining 85% frontal and 75-80% side overlaps among images. UAV-based multispectral data were collected at three growth stages (GS21, GS65, GS73). Pix4D mapper was used for orthomosaic generation, QGIS was used for image segmentation to extract the useful information of each plot. Meteorological conditions differed in the two years of the trial, rainfall from sowing until ripening in 2021 and 2022 was 200 and 254 mm, average daily temperature 16.7 and 14.6°C, respectively. Means of evaluated traits showed significant differences between the LN and HN, accross testing years and genotypes tested, but only in GS65 and GS73. Correlation coefficients between GY and VIs varied depending on growth conditions and GS; the highest values were observed in flight of 2021 (comparatively dry and warm) under HN conditions (r=0.67-072) in GS73. In both years in GS73 and only under HN conditions the highest correlations were observed between NUE and VIs, with the highest values for NDRE and RECI. Our findings revealed that productive genotypes with heightened nitrogen use efficiency can be selected based on UAV multispectral phenotyping data. Study was carried out in the frame of EEA and Norway Grants project “NOBALwheat – breeding toolbox for sustainable food system of the NOrdic BALtic region”.

Keywords: DJI Phantom 4 Multispectral (P4M) camera, vegetation indices (VIs), grain yield, NUE

EVALUATION OF FROST RESISTANCE USING IMAGE ANALYSIS

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Frost resistance is the main factor of wheat winter hardiness. It describes the ability of plants to withstand freezing temperatures. Frost resistance is traditionally monitored by the special developed frost tests, which resulted as the proportion (%) of surviving plants. However, survival rates are determined using subjective assessment, which can be imprecise, time- and labour - intensive, and does not "mine" enough information from relatively expensive tests. Image analysis can be perspective method of the frost test evaluation. This procedure is non-destructive and can rapidly evaluate a large number of genotypes. This work presents partial results of methodological procedures suitable for quick determining the degree of winter wheat frost resistance.

Keywords: wheat, frost resistance, image analysis

GENETIC VARIATION IN IMPORTANT PHYSIOLOGICAL RESPONSES AND THEIR EFFECTS TO ALLEVIATE DROUGHT STRESS IN WHEAT SEEDLINGS

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Water deficit is one of the major environmental factors that affect wheat growth and development. The seedling stage is a sensitive growth stage to drought stress. Therefore, studying physio-morphological traits is very important to understand the different mechanisms associated with drought tolerance. In this study, 10 drought-tolerant and 10 drought-susceptible genotypes were selected based on the drought tolerance. At the seedling stage (one-leave stage), all genotypes were exposed to natural drought stress for 13 days. Six physiological components (protein (P), proline (PRO), glucose (G), fructose (F), amino acids (AM), and total soluble carbohydrates (TSC)) both were analyzed from the leaves of genotypes under control and drought conditions. The ANOVA analysis revealed high genetic differences among the tested genotypes in all traits. An observable increase was found in P, G, PRO, AM, and TSC in the drought-tolerant genotypes, while, s, while, PRO and AM were increased only in the susceptible genotypes.. The most drought-tolerant genotypes were screened with nine specific drought genes (*TaDREB1*). Out of nine *TaDREB1* genes used, six primers were polymorphic among the tested genotypes. It was observed that the largest number of *TaDREB1* genes was present in the two Egyptian genotypes MISR1 and SAKHA93 (six genes), while the lowest number of these genes was present in the American genotype Huch (one gene). The most adapted genotypes among the tolerant plants were identified and will be included in the future breeding program.

Keywords: drought tolerance, *Triticum aestivum* L., seedling stage, physiological traits, *TaDREB1* genes

RESULTS OF MULTI-YEAR DURUM WHEAT VARIETY TESTS CARRIED OUT IN DIFFERENT MANAGEMENT SYSTEMS

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Durum wheat (*Triticum turgidum* ssp. *durum*) is the second most cultivated *Triticum* species after bread wheat (*T. aestivum* L.) in the world, 33.8 million tons of durum grain were produced worldwide in 2021. In 2022, durum wheat was cultivated on around 35000 ha in Hungary. The acreage under organic farming is constantly growing and the consumers' demands for organic products are also increasing. The objective of this study was to identify phenotypic parameters and yield components of durum wheat varieties of different geographic origin under Hungarian organic and conventional management in three growing seasons. Between 2019 and 2022, unlikely the previous years' average, extreme weather conditions were observed. Durum wheat germplasm was examined in organic, common conventional and low-input breeding trials with 3 replicates and 6 m² plot size. No fungicides were applied in any trial, however, conventional plots were treated with herbicide. During the growing season, early spring ground cover, heading time, plant height, disease severity and lodging were recorded. After harvest, grain yield, grain size (width, length), test weight (TW) and thousand kernel weight (TKW) were determined. Analysis of variance revealed statistically significant effects for the year, management, genotype and their interactions for heading, lodging, yield, grain width and length, TW and TKW. In the case of plant height and ground cover, the management×genotype interaction was not significant. In 2020 and 2022, our latest heading variety, 'Mv Makaróni' headed on 20 and 21 May, respectively, while in 2021, due to the rainy and cold spring weather, 85% of the varieties started to head only after 20 May. Lodging was observed only on the conventional sites in two years. The average yield of the experiment was between 5.79 t/ha and 6.02 t/ha. The varieties 'NS-Zad', 'Sambadur' and 'Mv Vékadur' performed in each trial in each year significantly superior compared to the average of the respective experiment. TW was between 78 kg/hL and 81 kg/hL on average, TKW varied between 42 g and 48 g considering the average of the three field trials. Varieties 'Simeto' and 'Senatore Cappelli' had significantly longer and wider grains compared to the average of the field trials in all three years. Among the naturally occurring pathogens, only powdery mildew was present at each site of each year.

Keywords: *Triticum turgidum* ssp. *durum*, organic, conventional, low-input

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INFLUENCE OF GENOTYPES ON NITROGEN ACCUMULATION AND REUTILIZATION IN WINTER BREAD WHEAT

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The primary biochemical component of wheat's nutritional value is protein. There are several methods for increasing wheat protein content, but two are the most common. The first is to ensure optimal nutrition by using fertilizers that contain nitrogen. The second method of increasing protein content in grain is to use varieties capable of producing grain with high protein content. Our research aimed to identify genetic systems that could enhance fertilizer uptake and increase the overall protein content of grains.

The research was conducted during the 2019-20 and 2020-21 growing seasons. The varieties were categorized based on the following principles: Odeska 16, an early-stage breeding variety with tall stems; Kuyalnyk carries the short-stemmedness genes *Rht8*, *Rht-B1*, *Rht-D1*; Shchedritsa, a wheat-rye translocation (1BL.1RS); a line from crosses with *Aegilops tauschii* Coss (Er 1598/12); a line with the GPC-B1 gene (Er 9520 GPC-B1+) and one without (Er 9099 GPC-B1-). Nitrogen content was measured after each phase of flag leaf emergence, heading, flowering, and grain ripeness. The experiments were conducted using two variants of fertilizer application: the first involved the application of ammonium nitrate at a rate of 60 kg/ha, while the second involved an application rate of 120 kg/ha.

Two years of research indicate that the leaves and grain filling contain more nitrogen than the stems during the growing season. The most significant nitrogen remobilization occurs during the heading and milk development stages. Intensive varieties, such as Kuyalnyk and Shchedrist, contain significant amounts of nitrogen in their leaves and stems, as compared to Odeska 16. Kuyalnyk and Shchedrist varieties outperform other genotypes in terms of yield, however they are inferior in terms of protein in grain and protein yield per hectare to Odeska 16 and Er 9520, and Er 1598/12 varieties. There is a slight decrease in nitrogen content in leaves and stems of the Er 9520 line with the GPC-B1 gene compared to the sister line Er 9099 without the gene and intensive varieties. Despite this, nitrogen recycling (reuse) is much higher, especially with an increasing agro background, indicating that GPC-B1 may affect nitrogen assimilation and reuse processes. Compared to intensive varieties, there are no fundamental differences in nitrogen accumulation and utilization in line with high protein genes from *Aegilops tauschii* Coss.

Keywords: *Triticum aestivum*, fertilizers, nitrogen, recycling, grain quality

EFFECT OF MELATONIN ON HORMONAL REGULATION IN BARLEY ROOT UNDER DROUGHT STRESS

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Melatonin is involved in many crucial processes of plants' functioning including the generation of new lateral and adventitious roots. It is also suggested that melatonin can manage cross-regulation between brassinosteroids (BRs) and other phytohormones in roots organogenesis in barley, contributing to acclimation to environmental stresses, as a consequence. Moreover, the exogenous melatonin application may decrease the oxidative damage caused by reactive oxygen species via directly scavenging hydrogen peroxide and enhancing antioxidant enzyme activities.

The objective of this study was to investigate the performance of barley plants, differed in BRs synthesis/signalling, under various water regimes through phenotypic and physiological measurements. Secondly, the effect of melatonin application on shaping the root system and on hormones content was explored under optimal and drought conditions.

Plant material consisted of cv. Bowman (BW, wild type) and its near isogenic lines (NILs): BW091 (BR-deficient) and BW885 (BR-insensitive). Plants were treated with 100 μ M melatonin solution, one week before the start of drought for three following days. Drought stress was imposed at three leaf stage (early drought) and at tillering stage (late drought) and it was maintained for two weeks. Biological samples were collected for each treatment at 7th and 14th day of stress, and one week after the re-watering. Phenotyping included the observation of above-ground part of plant (plant structure and yield components) along with major phenological stages and root architecture evaluation (in total 19 traits). SC-1 Leaf Porometer and FluorPen FP 110 were employed to measure the stomatal conductance and chlorophyll fluorescence parameters, respectively. Additionally, the nitrogen-flavonol index and the content of chlorophyll and flavonols were measured using Multi Pigment Meter MPM-100. Chromatographic and mass spectrometry techniques were used for quantification of selected hormones in the roots.

We proved, that the positive effect of melatonin on the barley root development required no BRs disorders since melatonin-treated wild type showed a significant increase in root weight in late drought in contrast to both mutants. In the following steps of the project, we will investigate early and late barley root transcriptomic reprogramming induced by drought and melatonin treatment, and next RNA-seq data will be integrated with hormones quantification that will enrich the knowledge on plant adaptations to stressful growing conditions.

Keywords: abiotic stress, brassinosteroids, drought, *Hordeum vulgare*, phytohormones

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**ADAPTATION OF LOCAL AND FOREIGN EINKORN GENOTYPES ON
CHERNOZEM, SOLONETZ AND LUVISOIL FROM BANAT COUNTY**

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Beside the two major forms of wheat (*T. aestivum* and *T. durum*) in the breeders attention is Einkorn with valuable genes for increasing tolerance to abiotic (salty soils and drought) and biotic stress (stem rust) conditions, for grain quality (Vitamin A, beta-carotene, lutein) and for organic farming. The paper highlights the results regarding the spike structure index (SSI) of 28 genotypes from Romania and other Countries to point out some traits related to tolerance on different soil conditions. The experiments were organized under 3 different ecosystems: on Chernozem and Solonetz & Luvisoil from Timisoara and Cenad respectively. The results emphasized the strong correlation between ecosystem and spike traits. On Chernozem, SSI was superior for 50% of the varieties, while 25% of the genotypes grew better on Solonetz. The spike structure index (SSI) pointed out the largest amplitude in Luvisoil conditions from 7.25 for DE62 to 10.01 for AMj8. On the Luvisoil 3-5 basal leafs dried up and the sibling's organogenesis took place late, while heading was repressed. The best spike phenotype was pointed out by the genotype USA1 with SSI values 8.25, 8.77 and 9.40 under the cultivation on Chernozem, Solonetz and Luvisoil, respectively. From this point of view the SK68 genotype on chernozem revealed the best SSI. Among the Romanian genotypes, those of AMj emphasize the highest homeostasis with an average SSI of 8.43 ± 0.19 , 8.08 ± 0.18 and 8.94 ± 0.33 under all soil types.

Keywords: Spike Structure Index, einkorn, Chernozem, Solonetz, Luvisoil

SESSION 5

Molecular approaches, bioinformatics and genomic selection

ADVANCED GENOMIC TOOLS FOR LEVERAGING DURUM WHEAT DIVERSITY

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Although durum wheat represents only 8% of the worldwide wheat production its relevance is much higher since the tetraploid wheats represent a large part of the wheat haplotypic diversity. During the last ten years we have developed several tools for leveraging the tetraploid wheat diversity.

In 2019 the first reference genome of durum wheat (cv. Svevo) was completed by an international consortium (Maccaferri et al., 2019). The Svevo v1 genome consisted of $\approx 129,000$ scaffolds spanning 10.45 Gb, although the longest 2,019 scaffolds represented 90% of the genome. To further improve the quality of the reference genome we have recently sequenced and assembled the Svevo genome using long reads generated by PacBio HiFi and optical maps. The new assembly (Svevo v2), provided in kind by CORTEVA, consists of 259 scaffolds only. An effort to complete the pangenome of tetraploid wheat by sequencing about 25 accessions representing most of the genetic diversity is in progress.

Through a large international collaboration, we have assembled a comprehensive global durum wheat resource comprising the Tetraploid wheat Global Collection (TGC, 1,856 accessions), including wild and domesticated emmer and local varieties of durum wheat and related subspecies and the Global Durum Panel (GDP; 1,033 accessions, Mazzucotelli et al., 2020) including cultivated and locally grown durum wheat varieties. The *iSelect* 90K SNP molecular marker anchored to the Svevo genome sequence were used to study population structure and perform association mapping studies. Passport information and genotypic data of the collections are available in GrainGenes (https://wheat.pw.usda.gov/GG3/global_durum_genomic_resources).

Keywords: durum wheat, genetic diversity, genome sequencing

GENOME-WIDE ASSOCIATION MAPPING FOR PRE-HARVEST SPROUTING IN EUROPEAN WINTER WHEAT

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Pre-harvest sprouting (PHS), the germination of seeds before harvest, is a major problem in global wheat production, and leads to reduced bread-making quality in affected grain. Breeding for PHS-resistance can prevent losses under adverse conditions. Selecting resistant lines in years lacking pre-harvest rain, requires challenging of plants in the field or in the laboratory or using genetic markers. To identify such markers in a breeding context, genome-wide association studies (GWAS) are required. Therefore, we challenged a population of European wheat varieties and breeding lines with PHS conditions and phenotyped them for PHS-traits, grain quality, phenological and agronomic traits to conduct GWAS. Furthermore, we compared these marker-trait associations to previously reported PHS-loci and evaluated their usefulness for breeding. We found markers associated with PHS on all chromosomes, with evidence for novel QTL on chromosome 1A and 5B. The QTL on chromosome 1A lacks pleiotropic effect, for the QTL on 5B we detected pleiotropic effects on phenology and grain quality. Multiple peaks on chromosome 4A co-located with the major resistance locus *Phs-A1*, for which two causal genes, *TaPM19* and *TaMKK3*, have been proposed. Mapping markers and genes to the pan-genome and chromosomal alignments provide evidence for structural variation around this major PHS-resistance locus. Although PHS is controlled by many loci distributed across the wheat genome, *Phs-A1* on chromosome 4A seems to be the most effective and widely deployed source of resistance, in European wheat varieties.

Keywords: pan-genome, structural variation, sprouting, wheat, SNP

**RAPID AND COST-EFFECTIVE KARYOTYPING IN WHEAT X BARLEY
HYBRIDS BY CHROMOSOME-SPECIFIC MULTIPLEX PCR**

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Inter-specific hybridization is a powerful tool for increasing genetic diversity during breeding programs. Wheat (*Triticum aestivum* L.) x barley (*Hordeum vulgare* L.) inter-specific hybridization provides the transfer of agronomically useful genes to wheat. The determination of the karyotype of hybrid progenies possessing altered chromosome distribution is therefore essential for the subsequent breeding steps. The most widely used technique for chromosome identification, Genomic in Situ Hybridization (GISH) is labour intensive. Therefore, a routinely performable highly specific, off-target free and technically less demanding method would be beneficial. To provide a reliable and easier alternative to GISH we developed Multiplex Polymerase Chain Reaction (MPCR) methods highly specific to the individual wheat and barley chromosomes. Primer pairs suitable for MPCRs were designed utilising the available genome sequences of *Triticum aestivum* and *Hordeum vulgare*. Potential primer pairs specific for individual barley and wheat chromosomes were selected according to different amplicon sizes to easily distinguish individual chromosomes. The primer pairs were curated and sorted into 4 sub-groups composing MPCR sets (plex-A wheat A subgenome; plex-B wheat B subgenome; plex-D wheat D subgenome; plex-H barley genome). Analyses of wheat x barley progeny lines demonstrated that MPCR primer sets are suitable for the easy and cost-effective detection of various wheat and barley chromosomes. The established detection system was also validated with GISH technique. This method is suitable for high-throughput analysis and, in contrast to GISH, can be performed from any tissue types.

Keywords: Inter-specific hybridization, Karyotype, GISH, MPCR

GENOMIC PREDICTION OF TRAITS RELATED TO PHENOLOGY AND TEMPERATURE-RESPONSE DURING STEM ELONGATION OF WHEAT

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Breeding for an increased resilience to the negative impact of climate-change is an important but difficult task. High-throughput phenotyping holds great promise to deliver information about traits related to yield. To evaluate the suitability of a phenotyping platform approach, we grew 254 and 242 genotypes in 2021 and 2022, respectively, of the F8 material of the DSP-Agroscope breeding program as training set in the field phenotyping platform (FIP) of ETH Zurich. After canopy height extraction, crop growth was modelled using P-splines. The targeted traits were start and end of stem elongation, final canopy height and temperature-response during height development. Correlations of genomic predictions models were in the range of 0.05 to 0.73 in 2021, and 0.12 to 0.78 in 2022, lowest for the steepness of the temperature response and highest for final canopy height. Furthermore, models were trained using genotypes measured in 2021 and predictions were made for a new set of genotypes cultivated in 2022, and vice versa. Correlations between 0.34 and 0.70 were achieved for stop of stem elongation, maximum growth rate and final height, while the remaining traits showed genotype-by-environment interactions, lowering prediction accuracy. Correlation for start of stem elongation, a trait whose evaluation in fields is time-consuming, increased to 0.44 in 2022, and therefore has potential to speed up the breeding process. However, predictions with the GABI wheat panel covering a wide range of varieties from all over Europe revealed that including multiple years in a training set with higher genetic diversity increases model accuracy.

Keywords: genomic prediction, high throughput phenotyping, temperature response, dynamic modelling, abiotic stress

THE CHROMATIN DETERMINANTS AND PH1 GENE EFFECT AT WHEAT SITES WITH CONTRASTING RECOMBINATION FREQUENCY

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This study aims to investigate a wheat recombination hotspot (*HI*) in comparison with a “regular” recombination site (*Rec7*) on the sequence and epigenetic level in conditions with functional and non-functional *Ph1* locus.

The DNA sequence, methylation pattern, and recombination frequency were analysed for the *HI* and *Rec7* in three mapping populations derived by crossing introgressive wheat line 8.1 with cv. Chinese Spring (with *Ph1* and *ph1* alleles) and cv. Tähti.

The *HI* and *Rec7* loci are 1.586 kb and 2.538 kb long, respectively. High-density mapping allowed to delimit the *Rec7* and *HI* to 19 and 574 bp and 593 and 571 bp CO sites, respectively. A new method (ddPing) allowed screening recombination frequency in almost 66 thousand gametes. The screening revealed a 5.94-fold higher recombination frequency at the *HI* compared to the *Rec7*. The *HI* was also found out of the *Ph1* control, similarly as gamete distortion. The recombination was strongly affected by larger genomic rearrangements but not by the SNP proximity. Moreover, chromatin markers for open chromatin and DNA hypomethylation were found associated with crossover occurrence.

Our results, for the first time, allowed study of wheat recombination directly on sequence, shed new light on chromatin landmarks associated with particular recombination sites, and deepened knowledge about role of the *Ph1* locus in control of wheat recombination processes. Understanding this phenomenon may become a base for more efficient wheat genome manipulation, gene pool enrichment and breeding.

Keywords: Hotspot, Crossovers, Recombination, *Ph1* locus, Wheat

This work was supported by the Czech Republic Ministry of Agriculture (award QK22010293).

EVALUATING DROUGHT TOLERANCE IN SPRING BARLEY BY HIGH THROUGHPUT PHENOTYPING AND GENOME-WIDE ASSOCIATION ANALYSIS

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Drought stress causes the most yield loss of all abiotic stresses and affects 10-40% of arable land worldwide. Stress during flowering can be particularly detrimental on yield parameters and poses a global challenge for breeding of abiotic stress resilient crops. Barley has a high tolerance to abiotic stresses which can be used for breeding of drought resilience varieties and for studies on water deficiency response. In this study a diverse barley population of breeding lines and genebank accessions were subjected to controlled drought conditions in an automated high-throughput phenotyping facility. Through non-destructive image-based methods the above-ground biomass and photosynthesis parameters was monitored by analyzing changes in digital biomass and photosystem quenching throughout the growth cycle. At early flowering a severe drought was applied for three weeks to identify genotype effects on plant response to drought in terms of yield, grain physiology, and biomass. The effect of the drought on yield is estimated by comparing seed number and thousand grain weight as well as image-based analysis of grain physiology for each genotype in drought and well-watered conditions. Potential QTLs involved with these traits are investigated through genome-wide association studies to aid future breeding for abiotic stress tolerant crops.

Keywords: Drought, high-throughput phenotyping, GWAS, image-based analysis

**HAPLOTYPE-BASED GENOMIC PREDICTION FOR FUSARIUM HEAD BLIGHT
RESISTANCE AND YIELD-RELATED TRAITS IN WHEAT**

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Genomic prediction is a powerful tool to enhance genetic gain in plant breeding. However, the method is accompanied by various complications that lead to low prediction accuracy. One of the major challenges arises from the complex dimensionality and multicollinearity of marker data. To overcome this issue, we applied two pre-selection methods for SNP markers viz. LD-based haplotype-tagging and GWAS-based trait-linked marker identification. Six different models were tested with pre-selected SNPs to predict the genomic estimated breeding values (GEBVs) of four traits measured in 419 winter wheat genotypes with two distinct populations comprising breeding lines and old cultivars. Ten different sets of haplotype-tagged SNPs were selected by adjusting the level of LD thresholds. In addition, various sets of trait-linked SNPs were identified with different scenarios from the training-validation combined and only from the training populations. The BRR and RR-BLUP models developed from haplotype-tagged SNPs had a higher prediction accuracy for FHB and SPW by 22.6 and 25.1%, respectively, compared to their corresponding models developed without marker pre-selection. The highest prediction accuracy for SPW and FHB was achieved with tagged SNPs pruned at weak LD thresholds ($r^2 < 0.5$) while stringent LD was required for spike length (SPL) and flag leaf area (FLA). Trait-linked SNPs identified only from training populations failed to improve the prediction accuracy of the four studied traits. Prediction across the two independent populations was very low and the two SNP pre-selection methods didn't improve the accuracy. Pre-selection of SNPs via LD-based haplotype-tagging could play a vital role in optimizing genomic selection and reducing genotyping costs. Furthermore, the method could pave the way for developing low-cost genotyping methods through customized genotyping platforms targeting key SNP markers tagged to essential haplotype blocks.

AN EFFICIENT ALGORITHM FOR MAPPING HETEROTIC QUANTITATIVE TRAIT LOCI WITH WHOLE GENOME SEQUENCING DATA

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Heterosis refers to the superior performance of hybrids over their parents and it has been exploited systematically in breeding of crops and is considered a major asset to meeting world food needs. However, its genetic and molecular basis are complex and have been extensively studied. Inspired by a pioneering study on bi-parental populations (Melchinger et al. 2007), a tailored quantitative genetic framework to study the genetic basis of heterosis in diverse hybrid populations has been developed and applied to elucidate the genetic architecture of heterosis for grain yield in bread wheat (Jiang et al. 2017). In this framework, the heterotic effect of a certain locus is defined as a linear combination of its dominance effect and its digenic interaction effects with the entire genetic background. A multi-step approach was designed to test the heterotic effect (Hqtl_MSS). Despite its successful applications, a clear disadvantage is that the GWAS was two-dimensional, namely the interaction effect between each pair of marker has to be tested. Hence, the required time for conducting the analysis increases quadratically as the increase of the number of markers. Performing two-dimensional GWAS with a high-density marker panel such as whole genome sequencing (WGS) is computationally not feasible.

To address this issue, we developed a new efficient algorithm to detect heterotic QTL. Instead of a multi-step procedure involving a two-dimensional scan for component effects, it directly tests the heterotic effects in a one-dimensional scan (Hqtl_ODS). Thus, it is much more efficient than Hqtl_MSS as the time for computation increases only linearly with the increase of the number of markers. Applying both approaches to a data set consisting of 1,557 hybrids with 57,846 SNPs, we found that Hqtl_ODS is 20 times faster than Hqtl_MSS. Hqtl_ODS is also capable of handling the WGS data with millions of markers.

Keywords: heterosis, heterotic QTL, one-dimensional scan, WGS

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SIMULTANEOUS IMPROVING OF DURUM WHEAT YIELD AND GRAIN PROTEIN CONTENT BY USING ASSOCIATION TESTS AND WEIGHTED GBLUP

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Despite the importance of grain protein content (GPC) in determining wheat quality, its negative correlation with grain yield (GY) is still one of the major challenges for breeders and farmers. Here, a durum wheat panel of 200 genotypes was evaluated for GY, GPC, and their derived indices (GPD and GYD), under different agronomic management for two consecutive years. A genome-wide association study using two statistical models revealed dozens of marker-trait associations (MTAs), each explaining up to 30% phenotypic variance. Two SNP markers on chromosomes 2A and 6B were consistently identified by both models and were found to be significantly associated with GY and GPC. MTAs identified for phenological traits co-mapped to well-known adaptive genes (i.e., *Ppd-1*, *Vrn-1*). The significance value (p-values) that measure the strength of the association of each SNP marker with the target traits were used to perform genomic prediction (GP) with a weighted genomic best linear unbiased prediction (WGPLUP) model. This statistical model outperformed conventional GBLUP for all traits (prediction accuracy increase up to 70%). The trained models were ultimately used to predict the agronomic performances of an independent durum wheat panel, confirming the utility of GP, although environmental conditions and genetic backgrounds may still be a challenge to overcome. The results generated through our study confirmed the utility of GPD and GYD to mitigate the inverse GY and GPC relationship in wheat, provided novel markers for marker-assisted selection, and opened new ways to develop new cultivars through GP approaches.

Keywords: single nucleotide polymorphisms, GWAS, genomic selection, WGPLUP, grain protein deviation

IDENTIFICATION AND EXPRESSION PROFILING OF RYE PHOSPHORUS-HOMEOSTASIS-RELATED GENES

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Low-phosphorus availability in soils negatively affects plant growth, hampering crop production, especially in acidic soils. Rye (*Secale cereale* L.) is a cereal with high tolerance to abiotic stresses, including phosphorus deficiency. Thus, studying the mechanism controlling rye phosphorus homeostasis will provide cues to discover loci contributing to phosphorus-deficiency resilience. Phosphate transporters are the core of the phosphorus homeostasis machinery maintaining adequate phosphorus levels in plant tissues and within cellular compartments. However, the function and genetic control of rye phosphate transporters remain unknown.

In this study, we performed a genome-wide analysis to identify putative members of the phosphate transporter in the *Secale cereale* Lo7 and Weining reference genomes. We also performed a phylogenetic, gene structure, and conserved motive comparative analysis to determine the evolutionary relationships of rye phosphate transporters to other grasses. We identified 1 PHT2, 2 PHT4, and 4 PHT5 putative transporter in both Lo7 and Weining rye genomes. Interestingly, the Lo7 contains 16 PHT1 and 6 PHT3 while weining contains 19 PHT1 and 9 PHT3 putative transporters. Some *ScPht* family members contain the P1BS *cis*-elements in the promoter region, suggesting phosphorus deficiency as the expression driving force. Expression profiling in K3 and L310 inbred lines shows that *ScPht1;2*, *ScPht1;13*, and *ScPht1;15* transcript levels increase in both shoots and roots under phosphorus-limiting conditions.

Our results will provide the ground base for the functional characterization of PHT transporters and sequence diversity screens in germplasm from different cultivation environments.

Keywords: *Secale cereale*, phosphorus-deficiency resilience, phosphate transporters

This study was funded by grant No. 2020/37/B/NZ9/00738 from the National Science Centre, Poland.

DAILY EXPRESSION PATTERNS OF MAJOR PLANT DEVELOPMENTAL GENES IN HEXAPLOID WHEAT UNDER INDUCTIVE CONDITIONS OF PLANT GROWTH

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Of the flowering regulatory pathways, the main components of the vernalization (*VRN*) and photoperiod (*PPD*) regulatory genes have been characterized in details in cereals, but there is little information available on the diurnal rhythm of these genes. Therefore in this work, we determined the daily expression patterns of three major vernalization response genes (*VRN1*, *VRN2* and *VRN3*) and the major photoperiod sensitivity gene (*PPD1*) including diverse genetic and phenotypic backgrounds. Three winter wheat cultivars ('Mv Toborzó' /AT1 with early heading/, from Hungary, 'Tommi' /AT3 with late heading/, from Germany and 'Charger' /AT20 with late heading/, from Great Britain) were examined under inductive controlled environmental condition (constant temperature: 18 °C, and long photoperiod: 16h). The leaf sampling started at 6:00 a.m. (1 h after the start of the light period - ASLP), and it was carried out every three hours, for two consecutive days (48h) using two-week old plants after vernalization (60 d). The qrt-PCR was carried out with the syber-green technology and the relative concentration was normalized with housekeeping genes.

The daily rhythm in all *VRN* and *PPD* genes expression could be detected but this was highly specific to the genotype. In the case of *VRN1*, the differences appeared not only in the daily timing of the peak expression, but also in its magnitude and the amplitude of the rhythm. In general, the early cultivar (AT1) showed the highest peak expression (4h ASLP), followed by AT3 (7h ASLP), and then AT20 (7h ASLP), demonstrating a strong positive coincidence with the heading time. The daily absolute values of *VRN2* negatively associated with the earliness. Thus the peak expression was the lowest in the early AT1 (no visible distinct peak), followed by AT3 (two peaks at 1h and 10h ASLP), and remained the highest in AT20 (7h ASLP). The activity of *VRN3* was increased in parallel with earliness, thus AT1 showed the highest peak expression (19h ASLP). The daily rhythm of *PPD1* in the early vs late genotypes were characterised with distinctly different patterns from each other, where the magnitude of this gene activity showed negative coincidence with the earliness. In the case of early genotype (AT1), the highest peak of early morning (1h ASLP) was followed by a smaller peak in early afternoon (13h ASLP). On the contrary, three peaks of similar magnitude were characterised the late genotypes (AT3 and AT20), an early morning (1h ASLP), a midday (7h ASLP), and a late night peak (19h ASLP).

Keywords: developmental, circadian and photoreceptor genes, daily gene expression, hexaploid wheat

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**GENE INTROGRESSIONS INTO WHEAT FROM AEGILOPS COMOSA AND
AEGILOPS UMBELLULATA**

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The genetic diversity of wheat narrowed down due to domestication and eight thousands years of cultivation, a problem hampering to find optimal allele combinations under changing environment. The goatgrasses *Aegilops umbellulata* (UU) and *Ae. comosa* (MM), the diploid progenitors of U and M genomes, respectively, are important gene sources to improve disease resistance and grain quality of wheat. The wheat-*Aegilops* comparative analysis highlighted six chromosomal rearrangements in *Ae. umbellulata*, while *Ae. comosa* chromosomes were more collinear with wheat. Altered structure of U genome may hamper the gene transfer into wheat through preferred chromosome elimination.

With the aim to test this hypothesis, amphiploids of durum wheat x *Ae. umbellulata* or x *Ae. comosa* were backcrossed with *T. aestivum* (Mv9kr1). Sequential GISH (with U/M and D genomic probes) and FISH with probes pSc119.2, pTa71 and Afa family confirmed the theoretical chromosome composition of the amphiploids and BC₁ progenies.

All of the M chromosomes with nearly similar frequency were also detected in the *Ae. comosa* BC₂ population together with the formation of 7D.7M robertsonian translocation and an 1DS-MS.ML-1DL interstitial translocation. However, only the chromosomes 1U, 3U and 7U were identified in the *Ae. umbellulata* BC₂ progenies with disomic 2US.2UL-6D and monosomic 6DS.6DL-2UL translocations.

The results indicate that wheat-*Aegilops* cross-genome homology may affect to the transfer of alien chromatin into wheat. The produced wheat-*Aegilops* chromosome additions and translocations will represent important genetic stocks for increasing the alelic diversity of wheat.

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EXPRESSION OF *R*-GENES IN RESPONSE TO BROWN RUST CAUSED BY *PRs* STRAINS IN RYE (*SECALE CEREALE L.*) INBRED LINES THROUGH RT-QPCR

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Brown rust caused by *Puccinia recondita* f. sp. *secalis* (*Prs*) is one of the most damaging diseases of rye. Nevertheless, knowledge about the genetic basis of immune response to this disease, in particular to the expression of genes conferring resistance, is very limited. The aim of the present study was to investigate the expression (by RT-qPCR method) of ten different genes (*ScLr1_ChrUn2*, *ScLr1_ChrUn3*, *ScLr1_Chr7R6*, *ScLr1_Chr7R7*, *ScLr1_Chr7R9*, *ScLr1_Chr7R10*, 1-deoxy-D-xylulose 5-phosphate synthase, Pathogenesis-Related Protein 1, Beta-1,3-glucanase, Glycosyltransferase; the first six selected on the basis of comparative analyses of the Lo7 rye genome with the wheat genome, the next four genes on the basis of RNA-seq analysis) in two rye inbred lines (L310 and SE31) inoculated by two different *Prs* isolates (i1.1.6 and i83) at 20 and 36 hours post-treatment (hpt). The relative expression of *ScLr1_ChrUn3* gene was significantly up-regulated at 20 hpi in both inbred lines infected by i83. Whereas, this gene was significantly up-regulated in L310 but down-regulated in SE31 line at 36 hpi by i1.1.6 isolate. Whilst, *ScLr1_Chr7R7* showed up-regulation at 20hpi only in inbred line SE31 inoculated with i83 isolate. The gene Pathogenesis-Related Protein 1 (*PR1*) was significantly up-regulated at both time points in L310 line infected by only i83 isolate and in SE31 line inoculated by both *Prs* isolates. Also, the expression of Beta-1,3-glucanase (*Glu*) gene was significantly up-regulated in SE31 inbred line at both time points inoculated by i83 and at 36 hpi inoculated by i1.1.6 isolate. Additionally, the expression of *ScLr1_Chr7R6* and *ScLr1_Chr7R10* were up-regulated at 20 hpi but down-regulated at 36 hpi in line SE31 inoculated by i1.1.6 isolate. Moreover, *ScLr1_Chr7R10* was down-regulated at 20 hpi in the same inbred line infected by i83 isolate. The genes *ScLr1_ChrUn2* and 1-deoxy-D-xylulose 5-phosphate synthase (*DXS*) were up-regulated only at 20 hpi and 36 hpi respectively in L310 line infected with i83 isolate. Also, in line L310, Glycosyltransferase (*GT*) gene was significantly up-regulated after inoculation with i83 *Prs* isolate at both time points. Therefore, *ScLr1_ChrUn3*, *ScLr1_Chr7R7*, *ScLr1_Chr7R9*, *PR1*, *Glu* and *GT* genes presumably play a significant role in conferring resistance to brown rust in rye.

Keywords: Rye, Brown rust, *Prs* isolates, resistance genes, RT-qPCR

INTERACTIONS OF MAJOR FLOWERING PATHWAY GENES IN HEXAPLOID WHEAT UNDER INDUCTIVE CONDITIONS OF FLOWERING

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The processes regulating flowering in cereals has not been completely dissected yet: So far, only the main components of the vernalization and photoperiod regulatory pathways have been identified in details, for which either diploid *Triticum monococcum* and tetraploid species with a smaller genome size or special genetic stocks (RIL, NIL, mutant and transgenic lines) were used. This hinders the dissection of the environmental flexibility in the complex flowering regulation networks. Therefore in the present study, we determined the system of relationships between three major vernalization response genes (*VRN1*, *VRN2* and *VRN3*), the major photoperiod sensitivity gene (*PPD1*), seven major circadian clock genes (*CCA1*, *PRR95*, *TOC1*, *LUX*, *ELF3*, *GI* and *CO1*) and five major photoreceptor genes (*PHYA*, *PHYB*, *PHYC*, *CRY1* and *CRY2*) in a diverse genetic and phenotypic backgrounds (wheat cultivars). Three winter wheat cultivars ('Mv Toborzó' /AT1/, from Hungary, 'Tommi' /AT3/, from Germany and 'Charger' /AT20/, from Great Britain) were examined under inductive controlled environmental condition (constant temperature: 18°C and long photoperiod: 16h). The leaf sampling started 1 h after the start of the light period and it was carried out every three hours, for two consecutive days (48h) using two-week old plants after vernalization (60 d). The qrt-PCR was carried out with the syber-green technology and the relative concentration was normalized to the geometric averages of three housekeeping genes.

A negative association between *VRN2* and the other two *VRN* genes (*VRN1* and *VRN3*) was observed ($r = -0.39$ and $r = -0.34$), but the connection between *VRN1* and *VRN3* genes was opposite ($r = 0.45$). A strong positive association between *VRN1* and *CCA1* was detected ($r = 0.54$), whereas a strong negative correlation was showed between *VRN3* and *GI* ($r = -0.36$). A strong positive relation was present between *VRN2* vs *PPD1* ($r = 0.50$) and a positive network was described between *VRN2* and two central circadian genes (*GI* and *CO1*, $r = 0.52$ and $r = 0.42$). Positive correlation could be detected between *TOC1* and *LUX* ($r = 0.53$), while a negative relation was observed between *TOC1* vs *CCA1* ($r = -0.42$) and *CCA1* vs *LUX* ($r = -0.37$). A strong positive correlation was present between *GI* and *LUX* ($r = 0.64$). In addition, a positive correlation was described between *GI* and *CO1* ($r = 0.52$) as well. A strong association was showed between *PHYA* and *PHYB* ($r = 0.68$) genes. In addition, a positive network was observed between all the phytochrome genes examined (*PHYA*, *PHYB* and *PHYC*) and *VRN3* ($r = 0.51$, $r = 0.43$ and $r = 0.45$).

Keywords: developmental, circadian and photoreceptor genes, gene network system, hexaploid wheat

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**PROFILING OF sRNA EXPRESSION IN GERMINATING BARLEY SEEDS
MAINTAINED IN LONG-TERM STORAGE**

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Seed storage and viability is an important aspect of meeting all human food needs, feeding animals, and maintaining biodiversity. Recently, much research has focused on seed senescence, dormancy, and germination. However, the mechanisms responsible for seed senescence and germination during long-term storage still remain unknown.

The main objective of this study was to determine whether, and to what extent, changes in sRNAs are associated with the aging of seeds stored in a dry state. Although miRNAs are involved in many developmental processes, their role in seed germination after long-term storage is unknown. The results obtained will be important for understanding the role of miRNAs and other sRNAs in regulating seed germination after long-term storage in gene banks. Plant material unique in the world was used in the study. It was barley seeds from a single lot from the 1972 harvest. The seeds were stored dry in hermetically sealed containers. Three groups of seeds were used for comparative studies: seeds with low germination capacity, seeds with high germination capacity, and reproduced seeds in which the aging process had not yet occurred.

The analyses identified 54 known miRNAs belonging to 12 families. The highest expression levels were found in four conserved miRNA families, namely miR159, miR156, miR166 and miR168. However, the most surprising result was the lack of significant differences in the levels of all miRNAs in seed samples with significantly different viability. This finding demonstrates the extreme stability of miRNAs in dry seeds. It is also the first RNA fraction identified that does not degrade with loss of seed viability.

To verify the target genes for miRNAs, degradome-seq analysis was performed and 133 targets were identified. To determine the involvement of miRNAs in the germination process, the functions of the targets corresponding to miRNAs with DEGs were analyzed.

RT-qPCR VERIFICATION OF GENE EXPRESSION IN RYE UPON LEAF RUST INFECTION

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Leaf rust (LR), caused by *Puccinia recondita* f. sp. *secalis* (Roberge ex Desmaz) (*Prs*), is one of the most economically significant fungal diseases of rye in Central and Eastern Europe, including Poland. Despite many years of research, knowledge about the genetic basis of resistance to LR is still limited. As a part of the project implemented in our department, an RNA-seq analysis was performed to identify differentially expressed genes (DEGs) in plants of three rye inbred lines – D33, D39 and L318 infected with compatible (c) and non-compatible (nc) *Prs* isolates. Some of these genes, including *ScNLR1*, *ScNLR6*, and *ScLr34_3* may play a really important role in immune response to leaf rust and have never been assigned to the immune response to this pathogen before. The aim of the presented work was to verify their possible role in response to LR determined by RNA-seq approach performing quantitative reverse transcriptase PCR (RT-qPCR) analysis in plants infected with *Prs* isolates 36 hours after treatment. This analysis showed that expression levels of selected genes changed significantly in infected comparing with mock treated plants, indicating their significant participation in the immune response to LR. In general, the results obtained by RT-qPCR confirm the RNA-seq studies, which indicates the legitimacy of such verification.

Keywords: leaf rust, transcriptome, gene expression, *Secale cereale*, resistance response

Research funded by the National Science Centre, Poland, project No. 2018/31/B/NZ9/00439.

IDENTIFICATION AND DISTRIBUTION OF *NE1* AND *NE2* HYBRID NECROSIS GENE ALLELES IN UKRAINIAN BREAD WHEAT VARIETIES

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Hybrid necrosis in wheat is characterized by progressive chlorosis and necrosis of plant leaves, tillers or whole plants in F₁₋₂ hybrids resulting from certain crossing combinations. This condition is caused by the interaction of two complementary dominant genes, *Ne1* and *Ne2*, which are located on chromosome arms 5BL and 2BS, respectively. The degree of hybrid necrosis manifestation depends on the strength of the interacting dominant alleles, including three alleles of the *Ne1* gene (*Ne1^w*, *Ne1^m*, *Ne1^s*) and five alleles of the *Ne2* gene (*Ne2^w*, *Ne2^{mw}*, *Ne2^m*, *Ne2^{ms}*, *Ne2^s*).

Hybrid necrosis poses a significant challenge in combining desirable traits into a single genotype or transferring genes from wild species to commercial cultivars. Furthermore, it can complicate the results of genetic analysis for some traits by causing the loss of certain genotypes in segregating populations. As a result, when selecting parental pairs for crossing, breeders, and geneticists should avoid pairing strong alleles of hybrid necrosis genes.

The literature shows that microsatellite loci *Xbarc74-5B* and *Xbarc55-2B* are linked to *Ne1* and *Ne2* genes, respectively. However, there is no information on how the alleles (amplification products) of these loci correspond to the alleles of *Ne1* and *Ne2* genes, which have different strengths. Therefore, our study aims to identify the relationship between the alleles of *Xbarc74-5B* and *Xbarc55-2B* loci and the distribution of *Ne1* and *Ne2* gene alleles by strength, search for new loci linked to hybrid necrosis genes, and investigate the distribution of *Ne1* and *Ne2* gene alleles among Ukrainian breeding varieties.

We developed a system of molecular genetic markers to identify *Ne1* and *Ne2* hybrid necrosis gene alleles in bread (*Triticum aestivum* L.) and durum (*Triticum durum* L.) wheat varieties, lines, and populations. We also determined the advantages of specific alleles for cultivars in different regions, which may indicate their value for selection and adaptation.

Keywords: *Triticum aestivum* L., hybride necrosis genes *Ne1* and *Ne2*, microsatellite analysis

ANALYSIS WITH MOLECULAR MARKERS WHEAT LINES WITH NOVEL TYPES OF TRANSLOCATIONS 1RS.1BL AT THE PREBREEDING STAGE

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To find new useful for breeding combinations of loci/genes occurred from homoeologous recombination between chromosome arms 1RS of rye and 1BS of wheat the crossing between the introgression wheat strain Erythrosperrum 125/03 (that has 1RS.1BL translocation from cv. Aurora) and Chinese Spring *ph1b*-mutant was made. Lines BC₁F₈ with different types of translocations and recombinations were developed and analyzed at the pre-breeding stage with molecular markers: microsatellites, SNP, NOR, KASP and protein markers – gliadins and glutenins.

Among 63 wheat pre-breeding lines (for each we analyzed 5-12 seeds) the lines with intact translocation 1RS.1BL, recombinant 1RS.1BS translocation with pericentromeric part of 1BS, recombinant 1BS with deletion of terminal part of the chromosome arm, 1RS.1AL and 1RS.1DL translocations were found. That occurred as a result of relocation of 1RS in the recombination process and recombination among wheat homoeological chromosome. In the 28.3% of lines, null alleles at *Gli1*-loci were revealed: 4 lines with *Gli-B1null*, 9 lines with *Gli-D1null*, 2 lines with *Gli-A1null*. The application of Zip4 marker revealed *ph1b* mutation in 17.3% of lines. The perspective lines according to combination of allelic variants of gliadins and molecular markers for 1RS were found, the some of them showed resistance (5-8 points) to leaf, stem, stripe rusts and powdery mildew in the field. Three lines resistant to rust diseases and without *Sec-1* gene were crossed with modern cultivars. The lines with different recombinations were used for further crosses between each other. The lines with 1RS.1AL and 1RS.1DL, as well as the derivatives from crosses between lines with different recombinations were crossed to breeding lines having morphological markers of 1AS, 1BS and 1DS chromosomes. The developed and controlled with molecular markers lines are on the quality control step and will be used in breeding program.

Keywords: bread wheat, recombinant lines, 1RS.1BL translocation, *ph1b*-mutant, molecular markers, prebreeding

CORRESPONDENCE BETWEEN ALLELES OF *GLI-1* LOCI AND THE ALLELIC VARIANTS OF GLIADINS OF *TRITICUM AESTIVUM* L.

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Gliadins are important monomeric storage proteins of wheat endosperm that interact with glutenin subunits forming gluten complex, determining flour breadmaking properties. Highly polymorphic gliadin genes provide a set of suitable genetic markers for the identification and comparison of wheat genotypes.

Different variations of acid PAGE method are usually used for fractionation of gliadin proteins expressed by at list six main gliadin-coding loci. Each of these loci produce a set of polypeptides inherited together as one Mendelian trait called allelic variant of gliadins. There are two classifications of allelic variants of gliadins: by Sozinov and Poperelya [1980] and by Metakovsky et al. [1991]. There is a number of peptides encoded by all *Gli* loci, on one track. Allelic variants of gliadins of the same wheat variety may differ in two catalogues due to the differences of fractionalization protein bands caused by different variations of acid PAGE methods that allow clearer separation of the one or the other zone on the electrophoregram. So, the users of these methods are encountered with difficulties.

Thereby, our research is devoted to the investigation of gliadin polymorphism of *Gli-1* loci by PCR method and evaluation of molecular markers suitable for identification of allelic variants of gliadin proteins. The clear correspondence between protein allelic variants of gliadins and alleles detected with allele-specific primers developed by Zhang et al. [2003] and *Taglgap* microsatellite alleles were found for *Gli-B1* [Popovych et al., 2020, 2021; Metakovsky et al., 2021] and *Gli-A1* loci, but not for *Gli-D1*.

Keywords: *Gli-1* loci, gliadin polymorphism, molecular markers

SESSION 6

Future challenges and innovations

**BRIDGING RESEARCH AND PLANT BREEDING FOR A SUSTAINABLE
AGRICULTURE: CURRENT ACHIEVEMENTS AND FUTURE CHALLENGES IN
CEREALS**

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Feeding a global population set to reach 9 billion by 2050, in the face of climate change and pressure on finite natural resources, will require a 70% increase in global agricultural output while reducing input use and GHG emissions. At the same time, there are growing opportunities and demands for the use of plant-derived biomass to provide livestock feed and for other uses, including renewable materials to support the bioeconomy, thereby decreasing dependency on imports and reducing regional and global environmental impact.

Securing a sustainable global supply of high-quality food and feed and supplying renewable biomass for the production of bio-based products and energy are key priorities for both European and global societies. In the coming decades, the European plant sector will play a central and essential role in meeting this challenge and achieving this will depend strongly on the generation and transfer of new knowledge, and greater innovation in the plant sector.

In this context, genomics and especially related molecular genetic technologies play an important role in the creation of new plant varieties that optimally combine high and stable yields with resistance to abiotic stresses and biotic factors of the cultivation environment. Over the past decade, molecular marker technology has provided a wide range of innovative approaches to improve the efficiency of modern breeding strategies and methods. The availability of new molecular tools and technologies has a significant impact on the planning and development of the critical elements of breeding required to accelerate this time-consuming and laborious process. Monitoring of genetic diversity associated with successful breeding, targeted use of plant genetic resources, examples of specific applications of molecular markers in cereal breeding, the potential of genomic selection and the use of genomics and gene edits in cereal breeding will be presented and discussed using the example of cereals. Future perspectives for public-private partnership in cereals research and breeding will be highlighted.

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CAN WE TEACH A.I. TO SELECT LIKE A PLANT BREEDER?

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Plant breeding is considered to be the science and art of genetically improving plants according to human needs. Scientific breeding is thereby driven by big data from various omics disciplines, while the art of breeding is determined by breeder's intuition and experience that leads to selection decisions by the so-called breeder's eye. For the purpose of this study, we investigated the possibility of predicting both data-driven and knowledge-driven selection decisions of a breeder by artificial intelligence utilizing methods of statistical learning. The target trait was thereby the retrospective binary classification of selected versus non-selected breeding lines during a period of six years in an applied bread wheat breeding program. Genome-wide predictions of this classification were conducted within each cohort as well as across cohorts by employing, amongst others random forests and neural networks, whereas the heritability of these selection decisions was estimated by Bayesian logistic models. The heritability of selection decisions was moderate, while the prediction ability within and across cohort suggested that the breeder's selection decisions were to some extent predictable. It is furthermore shown that probabilities based on the binary classification into selected versus non-selected breeding lines can be employed as a comprehensive breeder's index, which comprises the entirety of data-driven and knowledge-driven inputs that are available for decision-making in a breeding program. Although this breeder's index was in line with the designated breeding goals, our results show that artificial intelligence cannot replace actual breeder's decisions, but have a large potential to aid breeders in their decision-making processes.

Keywords: breeder's eye; selection theory; genome-wide prediction; machine learning; artificial intelligence

BREEDING WHEAT FOR ORGANIC FARMING – LESSONS LEARNT FROM THE ECOBREED PROJECT

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The ECOBREED project aims to strengthen and/or initiate organic wheat breeding in Europe, especially in countries where the organic sector is still underrepresented. Within the project, organic multi-environment trials (METs) with two winter wheat diversity panels (i.e. early and late maturity) were carried out from 2020-2022 in seven European countries. The METs revealed an extremely high variability in grain yield caused on the one hand by severe drought in some environments, on the other hand by a significant legacy effect of previous mineral fertilization in one test site. Spatial modelling of individual field experiments usually improved trial efficiency significantly, however, in some trials the used field design did not allow an efficient recovering of natural or extraneous variation in the field. Grain yield was generally highest in modern cultivars derived from conventional breeding programmes and lowest for landraces. Tested organic heterogenous material (OHM) showed medium yields as well as cultivars derived from organic breeding programmes. It is also worth mentioning that in the early maturity panel which was sourced from eastern Europe no genotype derived from a breeding programme devoted to organic farming. The availability of a specific organic VCU test as in Austria, Germany and Czechia seems to be a key driver for the establishment of specific organic breeding programmes. Essential for organic wheat breeding seems to be also a satisfying resistance level against common bunt as severe infection levels were observed at single sites after three seed multiplication cycles. Molecular markers for rust resistance, dwarfing and photoperiod insensitivity genes, as well as genes associated with grain yield under drought conditions did not reveal a specific association pattern with respect to the geographic or breeding programme origin of the germplasm.

Keywords: Disease resistance, marker assisted selection, organic traits, *Triticum aestivum*

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EMMER AND EINKORN RELOADED – THE PESTICIDE-FREE CROP ALTERNATIVES

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Ancient wheats have long been abandoned due to their low performance under high-input farming. With them, we have lost not only an eye-catching diversity but also health-supporting foods. Especially einkorn and emmer are hardly known, although they are assumed to have good weed and disease resistance, making them good candidates for pesticide-free cropping. To help gain ground, we investigated their possible re-introduction into organic farming. Our experiments confirmed that einkorn had excellent disease resistance (without any incidence of leaf fungi), while emmer was occasionally moderately susceptible to rusts. Both species showed good *Fusarium* resistance, including a low tendency for deoxynivalenol accumulation. Weed suppression ability was acceptably good for both organic wheats and ancient wheats. Although cereal leaf beetle (*Oulema melanopus*) caused severe damage to durum, winter wheat and spelt in 2022, at Szár site, Hungary, einkorn and emmer exhibited an apparent repellent effect. Although both species had on average 2.6% higher grain protein content compared to winter wheat, they could only produce hulled grain yield of about half of the grain yield of wheat on the most productive soils. Under low-input or marginal conditions, however, they could achieve similar results but could rarely go above wheat. Although they cannot substitute wheat, it was proved here that they could safely be grown without the use of pesticides while they both can provide tasty and healthy alternatives for our foods.

Keywords: einkorn, emmer, grain protein content, pesticide-free agriculture, yield

EFFECT OF PULSED ELECTRIC FIELD ON VIABILITY OF *FUSARIUM* MICROMYCETES

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Fusarium species are one of the most studied fungal pathogens of cereals. They cause Fusarium head blight or stem base diseases that are risky not only in terms of yield losses but also due to mycotoxins production. Several strategies are used to prevent *Fusarium* colonisation of plants, including fungicide treatment. However, according to the EU Biodiversity Strategy for 2030, the use of fungicides needs to be reduced. Therefore, alternative seed treatments should be sought. One possible method is the use of a pulsed electric field (PEF) that could kill or remove the fungi from the surface. We first investigated the sensitivity of fungi to PEF treatment. Spore suspensions of four *Fusarium* species (*F. culmorum*, *F. graminearum*, *F. poae*, *F. sporotrichioides*) were treated in continuous PEF-system (OMNIPEF; VITAVE, Czech republic) (10 kV/cm, 450-900 Hz). Spore viability was assessed on an artificial medium. Different species of the genus *Fusarium* showed statistically significant differences in their response to PEF. As the frequency increased from 450 to 900 Hz, the number of viable spores gradually decreased for all *Fusarium* species tested. *Fusarium poae* responded already at the lowest frequency of 450 Hz with a strong reduction in spore viability (only 9 % of spores survived). At the same frequency, 53 % of *F. culmorum* spores, 29 % of *F. sporotrichioides* spores and 15 % of *F. graminearum* spores remained viable. PEF treatment was found to be an effective tool and the impact on the whole system (seeds and fungi) is currently evaluated.

Keywords: pulsed electric field, *Fusarium culmorum*, *F. graminearum*, *F. poae*, *F. sporotrichioides*

This work was supported by the Czech Science Foundation (project No 20-14649S).

NOVEL WHEAT BREEDING APPROACH TO COUNTER THE CLIMATE CHALLENGES AND BIODIVERSITY LOSS

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Global climate change is forcing us to adapt. Among several possible solutions, enrichment of biodiversity is one of the most prominent options. Instead of trying to pinpoint the “best” combination of parents to create a “perfect” variety, for almost 10 years we have focused on creating insurmountable number of crosses to generate and discover new and valuable genetic variability with inherent potential to counter the imposed negative climate effects, on both global and regional levels.

Our approach is fundamentally based on crossing of material with highly diverse, but desirable genetic backgrounds. For example, European wheat genotypes generally harbour excellent yield potential, tolerance to prevalent diseases and lodging, while Eurasian genepools typically hold genes for excellent technological quality, yield adaptability and stability, and are often more tolerant to water, cold, heat, salt and other stresses.

So far, our work resulted in creation of 9581 wheat crosses using 432 varieties, 158 advanced lines and 82 DH lines. Parental material originates from both private and public breeding entities and over 50 countries.

Our entire breeding concept will be presented including obtained results.

Keywords: wheat, breeding, biodiversity, climate change

THE HUNGARIAN PARTICIPATORY ORGANIC WHEAT BREEDING PROGRAM

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To increase the number of cultivars suitable for organic farming, one of the effective and low-cost concepts is to perform the selection work on the target farm in close cooperation with the farmer. On the basis of this, ATK had sent its organic-suited winter wheat varieties (Mv) and two populations (Mv Elit CCP, Mv Bio2020 Pop) to organic farmers with the aim to start participatory testing (PVS: participatory variety selection) and breeding (PPB: participatory plant breeding) on their farms. These on-farm trials were coordinated by ÖMKi and were run on 3 farms in 2021 completed with 2 additional farms from the following year. Cultivars were evaluated by farmers during the vegetation period, while breeders of ATK visited the farms and they discussed the steps of observation, selection and harvest of trials with the farmers.

In general, Mv varieties had higher grain yield than the populations at each site. Based on the results of the on-farm trials, Mv Elit CCP showed mostly the highest protein and gluten contents and best Zeleny sedimentation value, while the Mv varieties were superior at the farm of Biocentrum (Želiezovce, SK). As only 3-4 Mv varieties could be examined on-farm, replicated small plot trial with 15 entries was also established on one of the farms (Szár, HU). Grain yield of the small-plot trial showed similar rank, showing the populations below the trial average (7.58 t/ha). The variety, Mv Szilke had the highest protein and gluten contents, so as Zeleny sedimentation volume, followed by the two populations. Unlike the other population, test weight and thousand grain weight of Mv Elit CCP were found to be above the trial average.

The two diverse populations are also part of a PPB program, where 3 farmers have made their own positive spike selection in the populations and, after threshing them into bulks, the resulted 6 new subpopulations were examined at two sites (Szár, Martonvásár). Average heading dates of the trial entries and their respective mother-populations were similar, except for the earlier heading subpopulation of Mv Bio2020 Pop selected by Biocentrum. Based on the mean plant height data, the farmer preference could be also revealed: the farmer of Biocentrum had selected taller plants, while the farmer in Szár had selected shorter plants from the populations. Only the subpopulation of Mv Elit CCP selected in Szár could yield more grains than the original population. Grain yield of Mv Bio2020 Pop could be also improved in Szár by 8.6%. The subpopulations selected in Szár and tested in Szár showed better performance than their mother-populations regarding each quality traits (protein, gluten, Zeleny). All subpopulations had higher average test weight and thousand grain weight than their mother-populations, except the Mv Elit CCP subpopulation selected in Füzesgyarmat (HU).

Keywords: organic, participatory breeding, wheat, population

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SCIENCE BEYOND JARS

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Plant genetic resources are key to crop adaptation to a changing climate. However, their actual use in crop improvement is limited and in stark contrast to their potential and value. The Polish gene bank, stores seeds of over 70,000 accessions.

Our main tasks are to collect, keep alive and provide access to germplasm of plant genetic resources. In addition, we describe accessions for morphological, phenological and agronomic traits and maintain the database. For some time now, molecular studies have complemented the activities of our gene bank.

We are interested both in the material collected and stored, and in the changes related to the aging process of seeds stored for many years in a dry state.

We are carrying out molecular characterization of the stored collections. Based on the results of genotyping by sequencing, we assess intra- and inter-accession variation, identify duplicates and outliers.

We also focus on natural seed aging. This is a complex and multi-component process that complicates the work of all seed banks. Loss of viability is inextricably linked to loss of biodiversity. We have focused on identifying changes in the transcriptome and miRNAome that have occurred in barley seeds during 45 years of dry storage.

We plan to expand our research into cryobiotechnology in the near future. We are interested in the response of biological systems to low temperatures, especially tolerance to deep freezing.

So the Polish gene bank is more than just priceless seeds sealed in jars.

Keywords: max. gene bank, molecular characterisation, seed aging, miRNA

**FIELD-BASED CANOPY HEIGHT ESTIMATION USING UAV IMAGES
IMPROVES AFTER SPATIAL AND TEMPORAL ADJUSTMENT**

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The present study aims to evaluate the accuracy of canopy height (CH) data derived from the digital canopy model using plot-level information in a large panel of wheat varieties, and the improvements in precision and the predictions of genotypic values produced by using a P-spline mixed model.

Multi-temporal images were collected with an unmanned aerial vehicle (UAV) platform on a field trial conducted in Italy on 354 wheat genotypes grown in replicate 10 square meters plots during 2020-21. CH data from UAV (CH_{htp}) and direct ground (CH_{field}) measurements were taken almost weekly throughout the growing season. P-spline statistical approach was used for spatial and temporal adjustment. Data fitting was evaluated both before and after the statistical correction, using coefficient of determination (R^2) and root mean square error (RMSE).

Spatial trend adjustment for CH showed a significant increase in R^2 . Comparing individual dates for CH_{field} and CH_{htp}, the R^2 value also increased significantly, particularly in the early stage of crop development, thanks to statistical spatial modelling, from a range of 0.26-0.82 to 0.47 to 0.83, respectively. A further increase in R^2 values resulted from P-spline temporal interpolation for the entire panel, ranging from 0.86 to 0.95, for CH_{htp} and CH_{field}, respectively, resulting in a RMSE values ranging from 0.04 to 0.18 meters.

This study provided evidence on the goodness of fit of time-series CH data acquired from UAV for breeding purposes.

Keywords: Wheat, HTP, digital canopy model, P-spline, UAV

ACCELERATED GROWTH CONDITIONS FOR ADVANCING WINTER WHEAT RESISTANCE BREEDING AGAINST FUSARIUM HEAD BLIGHT

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Winter wheat disease resistance breeding is a time-consuming process due to the long life cycle of the plants. To overcome this, the goal of the winter wheat project was to improve existing methods using speed breeding to advance the genetic gain of the plant and development of resistant varieties. A novel speed breeding protocol for winter wheat with resistance selection for Fusarium head blight (FHB) was developed and validated using 432 genotypes from a breeding program and a genebank. Several agronomic traits were also measured. The protocol reduced the time required for plants to begin heading while avoiding any visible symptoms of stress on plants. Enabling disease resistance evaluation of four generations per year. The AUDPC for FHB severity was higher in the breeding set compared to the genebank set, indicating that the germplasm from the genebank set had higher resistance to FHB. An association study using genome-wide association analysis identified ten quantitative trait loci (QTL) for FHB resistance. Nine QTL were identified in the combined set, which included both the genebank and breeding sets, while two QTL were identified in the breeding and genebank sets separately, with some QTL overlapping across the three datasets. The results demonstrate that the FHB evaluation protocol incorporating accelerated growth conditions is an effective approach for FHB resistance breeding in winter wheat and can be applied to spring wheat with minor modifications. This study provides an important step towards enhancing the efforts to improve winter wheat resistance breeding.

Keywords: winter wheat, genebank, speed breeding, fusarium head blight resistance, quantitative trait loci (QTL)

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CAN THE RISING ATMOSPHERIC CO₂ CONCENTRATION MITIGATE THE SEVERE DROUGHT DAMAGES?

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Although carbon dioxide is a natural component of our atmosphere and is necessary for the assimilation of plants, due to its almost doubling level over the last centuries, it has become one of the most significant greenhouse gases. The increased CO₂ level has positive effects on photosynthesis, reduces water uptake, and improves the growth and production of plants. The positive effect of CO₂ on C₃ plants is a well-known phenomenon, but this effect is highly environmental effects-dependent, such as air temperature or the nutrient and water content of the soil. According to the forecasts, we can expect more and more intense drought periods in many agricultural areas in the coming decades. Drought stress is one of the most important abiotic stresses, which significantly reduces plant production and affects up to 40-60% of cultivated areas. Water-use efficiency (WUE) is an important metric, as it sheds light on the relationship between carbon and water turnover and is therefore an indicator of drought tolerance. Determining the WUE is important in the study of plant responses to climatic change, which metric shows significant differences between species and influences many environmental factors. To investigate the effects of the elevated carbon dioxide level and the drought we examined four winter wheat varieties: 'Mv Ikva', 'Mv Nádor', 'Mv Nemere', and 'Mv Kolompos'; one winter barley variety: 'Mv Initium' and one winter oat variety: 'Mv Hópehely'. The tested varieties were bred in Martonvásár and have different ripening-type; 'Mv Ikva' and 'Mv Initium' are early-ripening, 'Mv Nádor' and 'Mv Nemere' are mid-ripening, and 'Mv Kolompos' and 'Mv Hópehely' are late-ripening varieties. In our climate-controlled greenhouse experiment, the experimental design included three replications, three treatments, three carbon dioxide levels, and six varieties. The drought was simulated at the stem elongation or at the heading developmental stage. The treatments were repeated at three different CO₂ levels: ~400 ppm, 700 ppm and 1000 ppm. Our tested varieties showed different reactions to the applied drought stress and CO₂ concentrations. The most sensitive varieties to the applied drought stress treatments were the 'Mv Hópehely' and the 'Mv Ikva', and in these varieties, the negative effects of drought stress could not be offset by the elevated CO₂ levels. The most favourable drought tolerance was observed in 'Mv Kolompos' wheat and 'Mv Initium' barley variety. The CO₂ reaction of these varieties was the most favourable, the increased carbon dioxide concentration alleviated the negative effects of the drought treatments. Compared to the current atmospheric level, the artificial increase of the carbon dioxide concentration had a positive effect only at 1000 ppm CO₂ level, among the examined carbon dioxide concentrations; with regard to the water requirements of the winter wheat varieties, the water uptake of the tested varieties was significantly reduced even in the case of optimal water supply and simulated drought.

Keywords: rising carbon dioxide concentration, drought stress, cereals, WUE

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PHENOTROLLEY: AFFORDABLE PHENOTYPING SYSTEM FOR CONTROLLED GROWTH CONDITIONS

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In plant science research, a controlled growth environment is used for inducing abiotic and biotic stresses to facilitate germplasm evaluation and comparison. Challenge, however, is that under controlled growth facilities where space is limited, there is a need to screen maximum numbers of genotypes in a short time, repetitively and non-destructively. Moreover, high-throughput phenotyping is challenging mainly because of the need for suitable equipment, infrastructure, and software skills. Although commercial phenotyping platforms are being developed, there is a need for simple-to-use, custom-made, affordable phenotyping systems for the local growth infrastructure. To address this, a simple, manually controlled, affordable system called “Phenotrolley” is proposed. The phenotrolley system and image analysis pipeline will be presented using two case studies - barley (*Hordeum vulgare*) and forage grasses. Forage species of tall fescue (*Festuca arundinacea*) and white clover (*Trifolium repens*) were subjected to one or two drought situations over time. Different growth parameters like early vigor, the reduction in dry matter (DM), stress recovery, and area under the growth progression curve were estimated for all genotypes. There was a significant association between DM and conventional biomass for barley ($R^2=0.74$), white clover ($R^2=0.42$), and tall fescue ($R^2=0.87$). Phenotrolley is custom built and such affordable systems enable better use of existing growth infrastructure for phenotypic evaluation of germplasm.

Keywords: affordable, phenotyping, barley (*Hordeum vulgare*)

APPLICATION OF A DRONE-BASED -HIGH-THROUGHPUT- PHENOTYPING SYSTEM IN CEREAL BREEDING

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Multi- and hyperspectral imaging systems enable the uniform examination of a large number of genotypes even in multi-environment experiments, as a result of which we have a detailed phenotypic data set. Unmanned Aerial Vehicle (UAV) or Drone flight is a "high-throughput" phenotyping methodology, during which we cover the area with photos, to create orthomosaic and reflectance maps.

In 2021, an expanded reference variety trial was established, made of 11 winter wheat, barley, durum wheat and triticale genotypes which are sensitive and tolerant to disease, drought, and heat stress. The trial was inoculated with leaf rust and *Pyrenophora teres* f. *teres* (only on barley), and 4 out of 8 randomly arranged replications were treated with fungicide. From the spring of the 2022 growing season, the drone flew weekly over the plots at a height of 100 meters. The drone forwarded the recorded data to the AGRONMaps platform, where they were processed, and the vegetation indices were calculated from this database (AGRON Analytics Ltd.). During the season, 17 phenotypic parameters were recorded, 47 spectral data were measured by the drone on 13 occasions, and all plots were harvested in the summer of 2022.

The differences between treatments and varieties were calculated using linear discriminant analysis. With the analysis, we performed the correlation analysis and grouping between the categorical dependent variable (genotype) and quantitative independent variables (phenotypic data, vegetation indices). Using the entire database, groups or clusters linked to varieties were created (heat map).

Using the heat map, we depicted the spectral composition of the reflectance of varieties and its changes during the growing season. The difference between the fungicide-treated and artificially infected groups clearly shows the sensitive and tolerant varieties, so we can get a detailed picture of the spectral composition of the two types and the value of the derived vegetation indices. In the case of the 1/N668 spectrum, opposite changes in the values of three sensitive varieties can be seen starting from the second half of April.

We continue to collect the phenotypic and UAV data. Our goal is to map the multispectral fingerprint of the genotypes over several years, and to connect the data obtained in this way with the phenotypic values influenced by biotic and abiotic stress factors.

Keywords: UAV, high-throughput phenotyping, multispectral fingerprint

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HIGH-THROUGHPUT AUTOMATIC PHENOTYPING OF BARLEY UNDER DROUGHT STRESS

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Current methods of automatic plant phenotyping are providing a new tool for plant biology studies. Commercial phenotyping platforms established recently employ non-invasive methods and measurements on a high-throughput, large scale. Dynamic features are measured in a non-destructive way in real time. Thanks to this, the imaged plants are not damaged and are able to reach full maturity hence the yield potential can be assessed. Moreover, real-time phenotyping made it possible to observe the dynamics of plant growth, during the entire vegetation period, not only at a single point in time, as is the case in conventional phenotyping. Dynamic assessment of plant traits is particularly important for understanding plant adaptation and response to environmental factors such as drought, one of the main abiotic factors limiting plant productivity worldwide.

The aim of the study was the continuous and non-invasive phenotypic evaluation of the tested pool of 120 spring barley genotypes in real-time on a platform for automatic plant phenotyping. They were analyzed in control (80% FC) as well as in drought stress (20% FC) conditions. The experiment was carried out using the LemnaTec Scanalyzer3D system equipped with RGB, UV and NIR cameras. This made it possible to obtain information on the growth dynamics of the plant, its shape, the expected area and biomass of shoots, the content of photosynthetic pigments and the water use efficiency (WUE) of the tested barley genotypes. After the imaging was completed, the plants were moved to the greenhouse to reach full maturity, and then the spikes were harvested to assess the yield components characteristics.

Observations carried out in the presented experiment made it possible to assess the diversity of barley genotypes in terms of the speed of perception of the stress factor and the size of its negative effects on the plant. The correlation of yield forming traits with the dynamic features from the platform allowed to obtain information on the behavior of the tested genotypes under drought stress.

Integration of the results from the experiment conducted in water shortage conditions from the platform for high-throughput plant phenotyping and data obtained from plant phenotyping in natural conditions will allow for a deeper understanding of the response of barley plants to abiotic stress.

Keywords: abiotic stress, high-throughput phenotyping, *Hordeum vulgare* L., LemnaTec Scanalyzer3D, non-invasive phenotyping, real-time phenotypic platform

Project is funded by Polish Ministry of Agriculture and Rural Development (MRiRW, Biological Progress 2021-2025, Task 14).

**CAS9-EXPRESING CELL SUSPENSIONS FOR gRNA EVALUATION:
OPTIMIZATION OF CONSTRUCT DELIVERY VIA ELECTROPORATION**

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CRISPR/Cas9-based genome editing became the most important tool of molecular genetics throughout the last decade. Despite huge scientific effort, there are still some limitations to its widespread use in crop improvement. One of the biggest obstacles for routine use of Cas9 in plants is low efficiency of editing, especially in polyploid species. Due to that fact, it is necessary to precisely evaluate the fitness of each newly designed gRNA sequence before its use *in planta*. Standard protocol for gRNA evaluation involves protoplast isolation from plant leaves and their transfection to achieve transient expression of gRNA/Cas9 construct. From our experience, such approach often leads to much higher editing frequencies compared to that observed after stable transformation of plants. We believe that a possible explanation for such observations might be a dosage effect, as numerous copies of gRNA/Cas9-coding vectors are delivered to protoplast during transfection.

To counteract such dosage effect we obtained stably transformed wheat cell suspensions cultures expressing single-copy Cas9 gene. Those cell lines are now re-established in suspension cultures and ready to be used as a source of protoplasts. Here, we present one of the optimization steps in development of our evaluation system – plasmid delivery via electroporation using novel pipette electroporator device.

Keywords: CRISPR/Cas9, cell-suspensions, grains, electroporation, gRNA-evaluation