



1ST INTERNATIONAL WORKSHOP ON BARLEY LEAF DISEASES

“Healthy Barley for Healthy Feed and Food for the Future”



Hotel Valentini, Salsomaggiore Terme, Italy, 03-06 June 2014

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Welcome to the 1st International Workshop on Barley Leaf Diseases

The workshop will provide a forum for the exchange of information and ideas relating to understanding and controlling leaf diseases of barley. The goal of the International Scientific Committee (ISC) is to bring together international experts and young students interested in recent advanced studies in all aspects (theoretical and applied) of barley leaf diseases.

HEALTHY BARLEY means vigorous growth and development, which ultimately produces superior grain for various end uses.

*The 2014 SalsomaggioreTerme event represents the debut of the IWBLD, which replaces the “International Workshop on Barley Leaf Blights” – last held in Dundee, Scotland in 2011. This name change reflects the ISC’s goal to include a wider group of researchers investigating **all barley leaf diseases**, instead of only leaf blights.*

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Chair of the Local Organizing Committee

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Nadia Faccini, Renzo Alberici, Fabio Reggiani, Marcello Baravelli, Donata Pagani, Marina Baronchelli, Stefano Delbono, Roberta Ghizzoni, Ivana Tagliaferri

Scientific Program

Tuesday 03 June 2014

From 12.00 *Registration of delegates and poster set up*

14.00 – 14.20 *Welcome address and Workshop opening by Adrian Newton, Chairman of IWBLD*

14.20 – 15.00 *“Lectio Magistralis” – Brian Steffenson: Combating cereal rusts over the Millennia: from the Robigalia to African stem rust*

15.00 – 17.00 *Session 1: Pathogen populations and variation*
Chair: Adrian Newton

15.00-15.20 Alireza Akhavan, T. Kelly Turkington, Berisso Kebede, Andy Tekauz, Kequan Xi, H. Randy Kutcher, James R. Tucker, Colleen Kirkham, Krishan Kumar, Stephen. E. Strelkov (Lacombe, Winnipeg, Saskatoon, Brandon, Melfort - **Canada**):

Comparative genetic diversity of *Pyrenophora teres* f. *teres* and *P. teres* f. *maculata* populations on the Canadian Prairies

15.20-15.40 Ryan Fowler, Jerome Franckowiak, Gregory Platz, Lee Hickey (St Lucia - **Australia**):
Pathotypic variation of the *Pyrenophora teres* f. *teres* population in Australia

15.40-16.00 Celeste Linde (Canberra - **Australia**):
Effect of a weedy host on pathogen evolution

16.00-16.20 Leona Leisova Svobodova (Prague - **Czech Republic**):
Population study of *Pyrenophora teres*

16.20-16.40 Mark S. McLean, T.K. Turkington, M. Jalli, F. Smit, G.J. Platz (Horsham, Warwick - **Australia**, Lacombe - **Canada**; **South Africa**; Jokioinen - **Finland**):
A new international differential set for testing *Pyrenophora teres* f. *maculata*

16.40-17.00 Mark W. Sutherland, G Platz, R. Fowler, A. Martin (Toowoomba, Warwick - **Australia**):
Assembly of candidate barleys for inclusion in an international differential set to assess virulence and pathotype of *Bipolaris sorokiniana* isolates

17.00 – 17.30 *Coffee break and poster viewing (with authors)*

17.30 – 18.50 Session 2: Epidemiology and disease management: conventional and bio-based products for barley health
Chair: Hugh Wallwork

17.30-17.50 T. Kelly Turkington, K. Xi, G. Peng, R.A. Martin, K.N. Harker, J.T. O'Donovan (Lacombe, Saskatoon, Charlottetown – **Canada**):

The impact of seed treatment, foliar fungicide, and variety resistance on barley leaf disease severity and productivity

17.50-18.10 Matthew Cromey (Christchurch - **New Zealand**):

Factors associated with changes to the barley leaf disease spectrum in New Zealand from 1899 to 2014

18.10-18.30 Adrian C. Newton, David C Guy (Dundee – **United kingdom**):

Effects of component proportions in barley cultivar mixtures and cultivation on rhynchosporium symptoms

18.30-18.50 Inger Åhman (Alnarp - **Sweden**):

Induction of barley responses by saccharin giving contrasting effects on biotrophic and necrotrophic pathogens

Wednesday 04 June 2014

09.00 – 10.30 Session 3: Structural and functional genomics of leaf barley pathogens
Chair: Brian Steffenson

09.00-09.30 Pietro Spanu (London - **United Kingdom**):

The genomics of *Blumeria graminis* f.sp. *hordei*

09.30-09.50 Wolfgang Knogge, Daniel Penselin, Claudia Wenzel, Martin Münsterkötter, Stefan Taudien, Marius Felder, Matthias Platzer, Anna Avrova, David Hughes, Kevin King, Bruce Fitt, Thomas Wolf, Ekaterina Shelest (Halle, Munich, Jena – **Germany**; Dundee, Hatfield, Harpenden – **United Kingdom**):

Host specialization in the fungal genus *Rhynchosporium*

09.50-10.10 Anna Avrova, Louise Gamble, Lucie Griffe, Barbara Franco Orozco, Olaya Ruiz, Martin Münsterkötter, M Looseley, Wolfgang Knogge, Adrian Newton, Paul Birch, Kim Hammond-Kosack, Kostya Kanyuka, (Dundee, Harpenden – **United Kingdom**; Munich, Halle – **Germany**):

Rhynchosporium commune effectors identification and exploitation

10.10-10.30 Anke Martin, Ryan Fowler, Terry Usher, Greg Platz, A. Kilian (Toowoomba, Warwick, Yarralumla – **Australia**):

DARtseq markers associated with phenotype in *Pyrenophora teres* f. *teres* genetic diversity study and using DARt markers in mapping of hybrid population

10.30 – 11.00 Coffee break and poster viewing (with authors)

11.00 – 12.50 Session 4: Molecular plant-pathogen interactions

Chair: Frank Ordon

- 11.00-11.30 Patrick Schweizer, Dimitar Douchkov, Jeyaraman Rajaraman, Hassan Razzak, Wenjing Hu, Tingting Zhang, Rajiv Sharma, Benjamin Kilian, Nils Stein, Wubei Dong (Gatersleben - **Germany**):

Broad-spectrum resistance of barley to powdery mildew - one trait, many genes

- 11.30-11.50 Timothy L. Friesen, Rachel A. Shjerve, Vaidehi Koladia, Robert S. Brueggeman, Justin D. Faris (Fargo - **USA**):

Evaluation of a *Pyrenophora teres* f. *teres* mapping population shows multiple independent interactions with the barley 6H chromosome region

- 11.50-12.10 Mark Looseley, Adrian Newton, Peter Werner, David Harrap, Bruce Fitt (Dundee, Hertfordshire – **United Kingdom**):

Image analysis methods for studying resistance during early infection of barley by *Rhynchosporium commune*

- 12.10-12.30 Chiara Biselli, Alessandro Tondelli, Davide Bulgarelli, Nicholas C. Collins, Gabriella Consonni, Paul Schultze-Lefert, Nils Stein, A. Michele Stanca, Luigi Cattivelli, Giampiero Valè (Fiorenzuola d'Arda, Vercelli, Milano– **Italy**; Köln, Gatersleben – **Germany**; Adelaide - **Australia**):

Characterization and discovery of barley leaf stripe resistance genes

- 12.30-12.50 Luca Pozzana (Milan - **Italy**):

Promega: Development of the Maxwell® 16 LEV Plant DNA Kit, and its application to plant leaf tissues

13.00 – 14.30 Lunch

14.30 – 16.10 Session 5: Incidence and control of barley leaf diseases in the CWANA region

Chair: Sajid Rehman

- 14.30-14.50 Aziz Karakaya, Zafer Mert, Arzu Çelik Oğuz, M. Reza Azamparsa, Esra Çelik, Kadir Akan, Lütfi Çetin (Ankara, Eskişehir – **Turkey**):

Current status of scald and net blotch diseases of barley in Turkey

- 14.50-15.10 Fatiha Bentata, Labhilili M., Essouaadi N., Benchaachoua M., Maafa I., El Aissami A., El Jaouadi A., Ibijbijen J. (Rabat, Kénitra, Meknés – **Morocco**):

Emergence and importance of *Rhynchosporium secalis* in Morocco

15.10-15.30 Rajan Selvakumar, R.P.S. Verma, S.S. Vaish, S.P. Singh, V.K. Goyal, A.S. Kharub, I. Sharma (Karnal, Varanasi, Faizabad – India; Rabat – **Morocco**):

Characterization of ICARDA barley germplasm lines for leaf blight resistance in India

15.30-15.50 Sajid Rehman, A. Visioni, M. El Hadi Maatougui, S. Gyawali, R.P.S. Verma (Rabat – **Morocco**):

Mitigating barley foliar diseases at global scale through germplasm enhancement at ICARDA

15.50-16.10 Fernanda M. Gamba, Mónica Ziminov, Amor Yahyaoui (Paysandú – **Uruguay**; Texcoco – **Mexico**):

Phenotypic diversity of *Pyrenophora teres* f. sp. *teres* in Uruguay, Morocco and Syria

16.10-16.30 Beyene Bitew (Debre Berhan, **Ethiopia**):

Status of barley leaf diseases in North Shewa Highlands, Ethiopia

16.30 – 17.00 Coffee break and poster viewing (with authors)

17.00 – 18.20 Session 6: Durable resistance in Barley

Chair: Giampiero Valè

17.00-17.20 Hugh Wallwork, Milica Grcic, Diane Mather (Adelaide – **Australia**):

Towards durable resistance to scald in barley

17.20-17.40 Marja Jalli, Outi Manninen, Teija Tenhola-Roininen, Lauri Jauhiainen, Tuomo Puroola, Ari Rajala, Pirjo Peltonen-Sainio (Jokioinen - **Finland**):

Achievements and challenges in breeding for durable net blotch resistance

17.40-18.00 Jerome D. Franckowiak, Ryan .A. Fowler, Gregory J. Platz and Lee T. Hickey (Warwick, St Lucia – **Australia**):

Insights on durable resistance to the net form of net blotch in barley

18.00-18.20 Sanjiv Gupta (South Perth – **Australia**):

Barley disease resistance in Western Australia - Issues and solutions

From 21.00 Choir concert “La Corale di Fiorenzuola” conducted by maestro Cassi at the main hall of the Thermae Palace

Thursday 05 June 2014

09.00 – 12.40 Session 7: Resistance breeding: the case of *Pyrenophora teres* and *Puccinia hordei* **Chair: Maria Jalli**

09.00-09.30 Frank Ordon, Janine König, Doris Kopahnke, Dragan Perovic (Quedlinburg – **Germany**):

Mapping and exploitation of new sources of resistance to the net form of net blotch (*Pyrenophora teres* f. *teres*) in barley

09.30-09.50 Olga Afanasenko, Elena Potokina, Anton Koziakov, Pete Hedlay, Nina Lashina, Anna Anisimova, Outi Manninen, Marja Jalli (Saint Petersburg – **Russia**; Dundee – **United Kingdom**; Jokioinen – **Finland**):

Mapping of qualitative and isolate specific quantitative trait loci associated with resistance to *Pyrenophora teres* f. *teres* and *Cochliobolus sativus* in two double haploid barley populations

09.50-10.10 Anke Martin, Francois Smit, Francois G. Potgieter, Greg Platz, Renée Prins (Toowoomba, Warwick – **Australia**; Caledon, Worcester, Bloemfontein – **South Africa**):

Identification of *Pyrenophora teres* f. *teres* resistance in South African barley line UVC8

10.10-10.30 Timothy L. Friesen, Vaidehi Koladia, Robert S. Brueggeman, and Justin D. Faris (Fargo – **USA**):

A CI5791 × Tifang RI population shows major dominant net form net blotch resistance located on barley chromosomes 6H and 3H in CI5791 and Tifang, respectively

10.30 – 11.00 Coffee break and poster viewing (with authors)

09.00 – 12.40 Session 7: Resistance breeding: the case of *Pyrenophora teres* and *Puccinia hordei* **Chair: Jerome D. Franckowiak**

11.00-11.20 Liliana Vasilescu, Alionte Eliana, Cană Lidia, Bude Alexandru (Fundulea – **Romania**):

Effect of net blotch disease (*Pyrenophora teres* f. *teres*) on yield and grain quality of winter barley

11.20-11.40 Morten Lillemo, Ronja Wonneberger, Timothy L. Friesen, Andrea Ficke (As - **Norway**, Fargo – **USA**):

Can susceptibility to net blotch in barley be explained by sensitivity to necrotrophic effectors?

11.40-12.00 Robert F. Park, P.G.Golegaonkar, L. Derevnina, K. Sandhu, H. Elmansour, P. Dracatos, D. Singh (Narellan – **Australia**; Bangalore – **India**; Davis – **USA**):

Adult plant resistance to leaf rust in barley: the story so far

12.00-12.20 Peter Dracatos, D. Singh, L. Derevnina, M. Zhou, R.F. Park (Narellan, Tasmania – **Australia**; Davis – **USA**):

Novel additive adult plant resistance sources to leaf rust in barley

12.20-12.40 Laura Ziemis, Colleen Hunt, Emma Mace, Gregory Platz, Robert Park, Jerome Franckowiak, David Jordan, Lee Hickey (St Lucia, Warwick; Narellan – **Australia**):

Association mapping of resistance to *Puccinia hordei* in Australian barley breeding germplasm

12.40 - 14.00 Lunch

14.00 – 14.40 Session 8: Resistance breeding

Chair: Anna Avrova

14.00-14.20 Greg Platz, Francis Ogbonnaya (Warwick, Barton – **Australia**):

National barley foliar pathogens variety improvement program - a coordinated approach to disease control

14.20-14.40 Lee Hickey, Silvia Germán, Silvia Pereyra, Juan Díaz, Laura Ziemis, Ryan Fowler, Greg Platz, Jerome Franckowiak, Mark Dieters (St Lucia, Warwick, Brisbane – **Australia**; Colonia – **Uruguay**):

Validation of rapid gene transfer methodology in barley

15.00 – 18.30 Visit to field trials – CRA-Genomics Research Centre, Fiorenzuola d'Arda

From 20.00 Social dinner at the Hotel Valentini

Friday 06 June 2014

09.00 – 10.20 Session 9: Barley diseases and climate changes

Chair: T. Kelly Turkington

09.00-09.20 Franz W. Badeck (Fiorenzuola d'Arda - **Italy**):

...For the times they are a changing...hence the diseases they are a changing.
Climate change and barley diseases

09.20-09.40 Michael Lyngkjær, Bolette L. Mikkelsen (Copenhagen – **Denmark**):

Impact of climate change on susceptibility to powdery mildew and spot blotch disease in barley

09.40-10.00 Sonia Mansouri, Leila Radhouane, Inès Abidi (Ariana - **Tunisia**):

Climate change impacts on barley diseases in the Béja region of north-western Tunisia

10.00-10.20 Hajer Ben Ghanem, Mouldi El Felah (Ariana – **Tunisia**):

Agro-meteorological evolution of barley leaf diseases in Tunisia

10.20 – 11.00 Coffee break and poster viewing (with authors)

11.00 – 12.30 Round Table “from Salsomaggiore to next meeting in 2018” organized by Adrian Newton, Hugh Wallwork and Brian Steffenson

From 12.30 Lunch, poster take down and departure of delegates

Oral Communications

Combating cereal rusts over the Millennia: from the *Robigalia* to African stem rust

Brian J. Steffenson

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Civilizations have battled cereal rust diseases since the dawn of agriculture. The ancient Greeks and Romans chronicled maladies of wheat such as “mildewing” and “blasting” that were likely due in many instances to rust diseases. The cereal rusts were so important to the early Romans that they held a festival called the *Robigalia* each year on April 25. During this festival, red dogs were sacrificed to appease the rust god *Robigus* from sending the rust scourge onto their crops. In 1766, Italy suffered a devastating stem rust epidemic of wheat that became the driving force for the seminal investigations by Felice Fontana and Giovanni Targioni-Tozzetti, who fostered the idea that fungi were the cause of plant diseases. Anton De Bary’s elegant experiments unequivocally proved that fungi were independent life forms that can cause plant disease and that *Puccinia graminis* could cycle as a heteroecious rust on both cereals and common barberry. Rust diseases were feared in many wheat-producing cultures because they caused repeated famines and ruined economies. Their potential as a destroyer of crops was well earned because the rusts can reproduce quickly to gargantuan-sized populations, spread readily over long distances, and destroy large plantings of cereals within just a few weeks. The rediscovery of Gregor Mendel’s laws of genetics, together with Rowland Biffen’s work on the inheritance of stripe rust resistance in wheat gave great promise for controlling rust diseases through breeding. Indeed, the introgression of single resistance genes into new cereal cultivars led to many early successes in rust control. However, these successes were often short-lived due to virulence changes in the pathogen population. The concept of physiological specialization (i.e. pathogenic races) by E. C. Stakman contributed greatly to our understanding of this phenomenon in plant breeding. Spectacular boom and bust cycles of single-gene-based resistance breeding occurred in the first half of the 20th century and led to a dramatic change in resistance gene deployment. One such strategy included the incorporation or “pyramiding” of multiple resistance genes into new cultivars. This approach has been highly successful in mitigating stem rust losses for more than 50 years in many wheat-producing countries around the world. However, the wheat community now faces a new challenge in the form of highly virulent stem rust races from Africa. In an unprecedented effort, a consortium of international scientists is now engaged in combatting this new rust threat.

Comparative genetic diversity of *Pyrenophora teres* f. *teres* and *P. teres* f. *maculata* populations on the Canadian Prairies.

Alireza. Akhavan¹, T. Kelly Turkington², Berisso Kebede¹, Andy Tekauz³, Kequan Xi⁴, H. Randy Kutcher⁵, James R. Tucker⁶, Colleen Kirkham⁷, Krishan Kumar⁴, Stephen. E. Strelkov¹.

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⁵Crop Development Centre, University of Saskatchewan, Saskatoon, SK, S7N 5A8, Canada;

⁶Agriculture and Agri-Food Canada, Brandon Research Centre, Brandon, MB, R7A 5Y3, Canada;

⁷Agriculture and Agri-Food Canada, Melfort Research Farm, Melfort, SK, S0E 1A0, Canada

The genetic structure of three *Pyrenophora teres* f. *teres* (Ptt) and three *P. teres* f. *maculata* (Ptm) populations from Alberta, Saskatchewan, and Manitoba, Canada, was assessed with microsatellite markers. All 13 microsatellite loci examined were polymorphic within both Ptt and Ptm populations. In total, 110 distinct alleles were identified, of which 19 were shared between Ptt and Ptm, 75 were specific to Ptt and 16 were specific to Ptm. Among the Ptt isolates, 94 alleles were detected with an average of 7.2 alleles per locus and a range of 3 to 11 alleles per locus, while for the Ptm isolates 35 alleles were detected with an average of 2.7 alleles per locus and a range of 1 to 5. High levels of genotypic diversity were found among isolates of both forms, with a clonal fraction of approximately 12% and 11% within Ptt and Ptm populations, respectively. Following clone-correction, significant genetic differentiation ($\Phi_{iPT} = 0.230$, $P = 0.001$) was detected among all populations, with 77% of the genetic variation occurring within populations and 23% between populations. Lower but still significant genetic differentiation ($\Phi_{iPT} = 0.038$, $P = 0.001$) was detected among Ptt populations, with 96% of the genetic variation occurring within populations and only 4% between populations. No significant genetic differentiation ($\Phi_{iPT} = 0.010$, $P = 0.177$) was observed among Ptm populations, with 99% of the total genetic diversity found within populations and only 1% between populations. Analysis of the microsatellite data by the unweighted pair group method with the arithmetic mean procedure and Jaccard's similarity coefficient revealed that isolates clustered in two distinct groups conforming to Ptt or Ptm. The Ptt isolates further clustered in two main clades, with 69% of isolates collected from Alberta in the first clade and 67% of isolates collected from Saskatchewan in the second clade. No clear clustering based on geographical origin was observed within Ptm populations. The high number of haplotypes observed within the Ptt (88%) and Ptm (89%) populations and an equal mating type ratio for both forms suggests that *P. teres* goes through regular cycles of sexual recombination on the Prairies.

Pathotypic variation of the *Pyrenophora teres* f. *teres* population in Australia

Ryan Fowler^{1,2}, Jerome Franckowiak¹, Gregory Platz¹, Lee Hickey²

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²The University of Queensland, Queensland Alliance for Agriculture and Food Innovation, St Lucia, QLD 4072, Australia.

Net form of net blotch (NFNB), caused by *Pyrenophora teres* f. *Teres* (*Ptt*), is an economically important disease of barley (*Hordeum vulgare*) in Australia and internationally. Pathotypic variation of the Australian *Ptt* population has been documented previously with the most recent study published in 2001. Since then changes in the cultivars grown have introduced and/or conversely removed genetic resistances to which the pathogen population is exposed. In this study we performed a comprehensive examination of the current pathotypic variation and virulence frequencies of the Australian *Ptt* population. Phenotypic data were taken for 118 isolates collected between 2007 and 2012 along with seven historic reference isolates collected between 1985 and 2003, on 31 barley accessions (20 Australian cultivars and 11 international lines). Standardised monoconidial spore suspensions of individual isolates were inoculated onto seedlings and infection types were scored adhering to the Tekauz (1985) rating scale. The *Ptt* isolates segregated into three distinct groups based on differential virulence to Beecher, Skiff and Prior, with virulence frequencies of 6.7%, 52.5% and 29.6% respectively and accounted for 83.9% of surveyed isolates. Of these three groups, 6.06% of the isolates showed virulence on two of the three defining differentials, while no isolate had virulence to all three differentials. Pathotypic analysis of the Australian *Ptt* population differentiated 41 pathotypes, of which 17 pathotypes were represented by two or more isolates. *Ptt* isolates with Beecher, Skiff and Prior virulence were represented by 5, 13 and 19 pathotypes respectively. Simple, Gleason, Shannon and Simpson complexities of the population were calculated at 0.35, 8.38, 3.02 and 0.91 respectively. None of the *Ptt* isolates displayed virulence to CIho 5791, which remains a useful source of NFNB resistance in Australia. Up to date knowledge of the virulences present in the Australian *Ptt* population will aid barley breeding organisations to implement effective screening for NFNB which should lead to the development of cultivars with durable genetic resistance Australia wide.

Effect of a weedy host on pathogen evolution

Celeste Linde

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Scald caused by *Rhynchosporium commune* is commonly found on barley as well as barley grass (*Hordeum leporinum*), a common weed in Australia. Wild hosts of diseases could play a significant role in the epidemiology and evolution of diseases. Barley grass is genetically more diverse than cultivated barley and likely harbour more resistance genes than barley. This heterogeneity could select for a pathogen population that is also genetically diverse with virulences that could render newly introduced resistance genes in barley ineffective. To investigate the effect of barley grass on the evolution of scald, pathogenicity of scald isolates from barley and barley grass was assessed on both hosts. No significant difference in leaf area affected on barley was observed for isolates from barley and barley grass, thus isolates from barley grass are equally likely to affect barley than barley grass. In contrast, isolates from barley resulted in a significantly smaller percentage of leaf area infected on barley grass lines. This suggests that scald populations from barley rarely infects barley grass, however scald populations from barley grass has a high potential for gene flow to barley populations. Barley grass therefore successfully acts as an ancillary host to scald harbouring highly virulent scald populations.

Population study of *Pyrenophora teres*

Leona Leisova Svobodova

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The population structure of the fungal pathogen *Pyrenophora teres*, collected mainly from different regions of the Czech and Slovak Republics, was examined using a microsatellite analyses (SSR). Among 305 *P. teres* f. *teres* (PTT) and 82 *P. teres* f. *maculata* (PTM) isolates that were collected, the overall gene diversity was similar ($\hat{h} = 0.12$ and $\hat{h} = 0.13$, respectively). A high level of genetic differentiation ($F_{ST} = 0.46$; $P < 0.001$) indicated the existence of population structure. Nine clusters that were found using a Bayesian approach represent the genetic structure of the studied *P. teres* populations. Two clusters consisted of PTM populations; PTT populations formed another seven clusters. An exact test of population differentiation confirmed the results that were generated by Structure. There was no difference between naturally infected populations over time, and genetic distance did not correlate with geographical distance. The facts that all individuals had unique multilocus genotypes and that the hypothesis of random mating could not be rejected in several populations or subpopulations serve as evidence that a mixed mating system plays a role in the *P. teres* life cycle. Despite the fact that the genetic differentiation value between PTT and PTM ($F_{ST} = 0.30$; $P < 0.001$) is lower than it is between the populations within each form ($F_{ST} = 0.40$ (PTT); $F_{ST} = 0.35$ (PTM); $P < 0.001$) and that individuals with mixed PTT and PTM genomes were found, the two forms of *P. teres* form genetically separate populations. Therefore, it can be assumed that these populations have most likely undergone speciation.

A new international differential set for testing *Pyrenophora teres* f. *maculata*

M.S. McLean¹, T.K. Turkington², M. Jalli³, F. Smit⁴, G.J. Platz⁵

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³ MTT, FI31600, Jokioinen, Finland

⁴ The South African Brewers,

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Spot form of net blotch (SFNB), caused by *Pyrenophora teres* f. *maculata*, is a major foliar disease of barley worldwide, causing grain yield and quality loss when severe. Growing resistant barley varieties is the best method for control of SFNB, however, *P. teres* f. *maculata* is pathogenically diverse and frequently undergoes sexual recombination. This can result in resistances becoming ineffective where virulent pathotypes are present and selected for in a population. A differential set has been developed for pathogenic characterisation of *P. teres* f. *maculata* populations, to identify effective resistance sources and monitor for significant changes in pathogenicity. The differential set was developed through extensive testing in Australia, South Africa, Finland and Canada during 2008-13 in which it has demonstrated the potential to differentiate virulences within and between populations as seedlings and adults. The set consists of 26 barley lines in total, and include; Arimont, Baudin, Beecher, Cape, Chebec, CI11458, CI3576, CI5286, CI5791, CI7584, CI9214, CI9776, CI9819, CI9831, CII6150, Galleon, Haruna Nijo, Keel, Kombar, Skiff, Steptoe, Stirling, Summitt, Torrens, TR250 and Yagan. In total, nine of these lines have been genetically characterised previously, 19 have been described as differential lines in previous studies and five were identified as new differential lines in this study. One resistant and one susceptible control have also been included. Recent testing of this differential set toward 26 isolates as seedlings and as adults at five field locations in Australia has revealed abundant pathogenic diversity with regionally based virulences, while testing in Canada, Finland and South Africa has identified a far greater diversity of virulences internationally.

Assembly of candidate barleys for inclusion in an international differential set to assess virulence and pathotype of *Bipolaris sorokiniana* isolates

M. W. Sutherland¹, G Platz², R. Fowler², A. Martin¹

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Foliar spot blotch of barley caused by the fungal pathogen *Bipolaris sorokiniana* (teleomorph: *Cochliobolus sativus*) can cause significant yield losses under warm, humid, late season growing conditions in a majority of regions where barley is grown. Long term control of this disease is sought in many countries through selection of resistant germplasm, however our understanding of the nature and uniqueness of the various sources of resistance that have been successfully identified is incomplete. Furthermore the virulence of *B. sorokiniana* isolates from different regions of the globe has not been determined on most of these putative resistance sources. To improve our understanding of these issues it was agreed at a meeting held during the 4th International Workshop on Barley Leaf Blights in 2011 to assemble an international set of resistant barley lines that could be made available to research groups world-wide for analysis of local pathogen populations. A common differential set of barley genotypes will allow researchers to compare the performance of resistance sources across different geographic regions, to determine whether distinct fungal pathotypes can be identified and to compare the apparent pathotypes present among barley growing environments world-wide. A set of 40 spring barley lines showing differential responses to spot blotch infections in studies from Australia, Europe, North America, Russia, Scandinavia, Syria, and Uruguay has since been assembled in Australia from overseas and Australian sources. Following a seed increase these lines have been analysed for genetic purity using a range of microsatellite DNA markers. Where several sources of the same named variety were available, these were compared using this same set of markers. Single seeds of some of these candidate lines were also subjected to diversity array analysis (DART). As a result of these genetic screens a number of lines were removed and the remaining candidate lines have now been despatched to participating international research groups. Phenotypic screening over the next twelve months will provide data that will allow us to consolidate this international set of lines and to begin to assess the effectiveness of the various resistance sources towards local disease isolates across barley growing regions worldwide.

The impact of seed treatment, foliar fungicide, and variety resistance on barley leaf disease severity and productivity

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At Lacombe, AB, Melfort, SK, and Charlottetown, PEI, the impact of seed treatment and foliar fungicide (treated versus untreated), and variety resistance (susceptible, intermediate and resistant) on barley leaf disease severity and crop productivity was assessed. The focus at Lacombe was scald, while at Melfort and Charlottetown net-form net blotch was the main disease issue. InsureTM (triticonazole + pyraclostrobin + metalaxyl) seed treatment was used at two times the recommended rate, while TwinlineTM (metconazole + pyraclostrobin) fungicide was applied at the recommended rate at flag leaf emergence. Data for Charlottetown are currently being analyzed. At Lacombe and Melfort, early and mid-season leaf disease severity was very low and not affected by any of the treatments. However, final disease severity on flag -1 and flag -2 leaves collected at late milk/early dough was significantly affected by the treatments. For example, scald and net blotch severity were significantly higher for the susceptible versus intermediate or resistant varieties at Lacombe and Melfort, respectively. In general at both sites, seed treatment and fungicide application reduced disease severity, but only for susceptible varieties. Also the response to foliar fungicide was similar with or without seed treatment. Variety had a significant effect on yield at Melfort, where the intermediate and resistant varieties had similar yields, but significantly higher than the susceptible. Seed treatment resulted in an increase in yield at Lacombe and Melfort; however, at both sites fungicide application resulted in the greatest yield increase. At Melfort there were also interactions of variety by seed treatment, variety by fungicide, seed treatment by fungicide, and seed treatment by fungicide by variety. Yields were increased by seed treatment for the susceptible variety, but not the intermediate and resistant varieties. In contrast, yields for both the susceptible and intermediate variety were increased by fungicide application, but not for the resistant variety. Seed treatment increased yield when no fungicide was applied, while yields were higher and similar when a fungicide was applied with or without a seed treatment. Overall at Melfort, only the susceptible variety significantly responded to seed treatment when no fungicide was applied. Furthermore, when a fungicide was applied with or without a seed treatment, yields increased, but only for the susceptible variety.

Factors associated with changes to the barley leaf disease spectrum in New Zealand 1899 to 2014

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Barley production in New Zealand dates back to the 1840s, with the arrival of European settlers. The first record of a barley disease was leaf rust, in 1899. By 2014, 18 foliar pathogens have been recorded in New Zealand barley crops: eight are common, two have been common in the past, three are uncommon, and five primarily cause disease on wheat or grasses. The spectrum of barley leaf diseases and their relative importance have varied over the years because of factors such as: incursions from overseas, cultivar change, pathotype change, fungicide resistance, changes in fungicides and their use, changes in crop management practices, and short- and long-term fluctuations in climate. The importance of net blotch has fluctuated since it was first recorded in 1969. Widespread outbreaks occurred after the withdrawal of organo-mercury seed treatments in the 1970s and again in the 1980s with the spread of triadimenol-resistant strains of the pathogen. Leaf rust was considered a minor disease in New Zealand barley crops until the 1980s. An upsurge in the disease was linked to a number of factors, such as the ‘green bridge’ provided by the introduction of winter barley, a new pathotype virulent on the popular cultivar ‘Triumph’ and a series of warm growing seasons. Scald is another disease linked with the uptake of winter barley in New Zealand, in crops of which it is difficult to control. Ramularia leaf spot was first identified in New Zealand in 1983, although records of the symptoms date back to 1977. Grower fungicide programmes have been adjusted to manage this disease following its diagnosis, although it is still sometimes found late in the growing season. Widespread use of effective seed treatments has all but eliminated some diseases, such as barley leaf stripe and barley stripe mosaic virus.

Effects of component proportions in barley cultivar mixtures and cultivation on rhynchosporium symptoms

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Barley cultivar mixtures range in their efficacy from no interaction to around 80% disease reduction and 17% increase in yield compared with the mean of their component monocultures grown separately (in monoculture). For rhynchosporium the epidemic progress in monocultures is influenced by soil cultivation method, probably due to residual inoculum of crop debris, and in mixtures it is strongly affected by contrasting epidemiologically functional traits such as resistance and canopy structure or plant habit. The greater the number of components the more disease is reduced. Furthermore, the spatial arrangement, patch size and distribution can affect their efficacy. However, resistance components can contribute disproportionately, a 10% proportion of a resistant component accounting for up to 50% disease reduction. This behaviour is in line with the outcomes seen in some mixture model predictions.

Induction of barley responses by saccharin giving contrasting effects on biotrophic and necrotrophic pathogens

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With the ultimate purpose to breed barley for increased response to an artificial resistance inducer, barley plants treated with the sweetener saccharin were inoculated with three different barley pathogens; *Pyrenophora teres* (net form), *Cochliobolus sativus* and *Blumeria graminis* f.sp. *hordei*. Resistance to the biotrophic *B.g.* f.sp. *hordei* has previously been shown inducible by saccharin (Boyle & Walters 2006, Plant Pathol. 55: 82-91) and this was confirmed in the present greenhouse study. However, on the contrary, saccharin induction resulted in higher levels of infection by the former two fungal species, that have a necrotrophic lifestyle. Possibly, the previously shown saccharin-induced increase in peroxidase activity leads to cell death that favours these types of diseases. In line with this, mechanical damage by fine pins before fungal inoculation resulted in larger blotches of *P. teres*. A lesson learned from the present study is to make sure that resistance-inducing agents do not result in contrasting effects depending on the lifestyles of the pathogens that may occur in the field.

The genomics of *Blumeria graminis* f. sp. *hordei*

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Reading the sequence and interpreting the annotation of the barley powdery mildew genome surprised us and taught us some valuable lessons that have affected our understanding of these important agents of disease. It has also influenced the direction of our current research (Spanu et al. 2010, *Science* 330:1543-1546). The first surprise was that the overall size of the powdery mildew genomes, in excess of 130 Mb, is far greater than originally predicted for an obligate parasite. This increase is due to a remarkable proliferation of retro-transposons that has resulted in the accumulation of repetitive DNA that makes up more than 90% of the *Blumeria* genome. The second surprise was that this increase in genome size is accompanied by a decrease in the number of protein coding genes, due to a general loss of paralogs (i.e. smaller gene families) and the loss of some pathways in primary and secondary metabolism. Similar genes are also missing in other obligate biotrophic pathogens of plants (rusts and downy mildews); this is a remarkable convergence in the evolution of pathogens with similar life-style. In the light of these losses, the expansion of effector-like genes that now comprise >7% of the protein coding capacity underscores the important of these elements in the biology of the powdery mildews (Pedersen et al. 2012, *BMC Genomics* 13:694). The most prominent group of effector-like genes encodes small proteins that appear to be derived from an ancestral RNase. We have shown that full expression and function of some of these RNase-like effectors is necessary for full disease development (Pliego et al. 2013 *Molecular Plant-Microbe Interactions* 26:633-642). We are currently investigating these effectors and I will propose here a model of how they may contribute to establishing disease, by interfering with host immunity.

Host specialization in the fungal genus *Rhynchosporium*

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The fungal genus *Rhynchosporium* (order: Leotiales) contains haploid fungi that are pathogenic to grass species (*Poaceae*) including cereal crops. Five morphologically very similar fungal species have been described to date that are characterized by their host specificity. Based on spore shapes two groups of species can be discerned. The beaked conidia group comprises the narrowly related species *R. commune*, *R. secalis* and *R. agropyri*, which are pathogenic to barley spp., to rye and triticale and to couch grass (*Elymus repens*, syn. *Agropyron repens*), respectively. The cylindrical conidia group includes the related species *R. orthosporum*, a pathogen of orchard grass, and *R. lolii*, which grows on perennial ryegrass. To unravel the molecular base of host specialization, the genomes of isolates from the five species were sequenced using a whole-genome shotgun sequencing approach. The genome of *R. commune* isolate UK7 was established as the reference genome harboring nearly 12,000 genes in a total size of about 55 Mb. A comparative genomics approach is being followed to identify genes or genomic regions that are unique to the different fungal species. For candidate genes from *R. commune* and *R. secalis* functional analysis is carried out using deletion and RNAi-based strategies to verify the impact of the genes on the specific interactions. In addition, the possible role of secondary metabolites in disease development is studied. Surprisingly, deletion of a polyketide synthase gene led to a substantially stronger growth of the mutant as compared to the *R. commune* wild-type strain on barley. Structure elucidation of the corresponding compound is in progress.

***Rhynchosporium commune* effectors identification and exploitation**

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One of the most destructive pathogens of barley, *Rhynchosporium commune*, has remained a threat to barley production for over a century. It has spread around the world to all barley-growing areas from Europe to the Middle East, Central Asia, North and South Africa, the Americas, Australia and New Zealand. Current control strategies heavily rely on the use of fungicides. Despite regular fungicide applications *R. commune* is costing the UK economy around £7.2 million per year (assuming a current average barley price of £150 per tonne). Sustainable disease management for this pathogen involves the introduction and maintenance of effective cultivar resistance. Identification of new sources of resistance requires a deeper understanding of the biology of *R. commune* and its interaction with barley. Sequencing of the *R. commune* genome and transcriptomes from germinated conidia and an early time point during infection allowed identification of the putative effector population mediating interactions with the host plant barley. Comparative genomics involving 9 *R. commune* strains with different race specificities allowed rapid prediction of candidate effectors specific to some of the strains as well as the ones less variable in *R. commune* populations. Less variable effectors are more likely to be essential for pathogenicity. Recognition of any of these effectors by barley resistance genes can lead to potentially more durable resistance compared to that provided by Rrs1, recognising a non-essential virulence protein Nip1. Transcription levels of candidate effectors have been assessed during first 2 weeks of barley colonisation using qRT-PCR. Association genomics combined with screening of genetically diverse barley accessions for *R*-gene mediated recognition of individual effectors expressed using a *Barley stripe mosaic virus* (BSMV) mediated system are being used to identify *R. commune* virulence genes and novel sources of potentially durable resistance to this pathogen. Targeted gene disruption of candidate effectors, highly abundant early during the infection, will help to determine their importance for pathogenicity.

DArTseq markers associated with phenotype in *Pyrenophora teres* f. *teres* genetic diversity study and using DArT markers in mapping of hybrid population

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In Australia, net form of net blotch caused by the fungus *Pyrenophora teres* f. *teres* (*Ptt*) has been recorded as an important disease of barley (*Hordeum vulgare* L.) since the 1960s. This disease is present in all barley-growing areas in Australia and can cause a 25% yield loss in years suitable for disease development. Changes in virulence can be devastating to the barley industry especially if a limited number of barley varieties are grown. Continual studies of the occurrence and distribution of different virulence types of this fungus are vital for the production of resistant barley varieties. Here we have investigated the genetic diversity and virulence structure of 86 *Ptt* isolates recently collected across different states in Australia. This study is the first to make use of Diversity Arrays Technology sequence (DArTseqTM) markers in *Ptt*. Data for more than 4000 DArTseqTM markers was used in distance and model-based clustering analyses to determine the genetic diversity and population structure of the isolates. The pathogenic diversity of all isolates was determined and the virulence profiles correlated with the genetic clusters produced with the DArTseqTM markers. DArTseq markers were also used to produce a map of a cross between Beecher virulent *Ptt* isolate NB29 and Prior virulent *Ptt* isolate NB85. The 89 hybrids from this cross were phenotyped using a detached leaf assay. A number of different pathotypes were observed in this population of which seven occurred frequently. Four isolates were virulent on seven out of the eight hosts used in the differential set. QTL mapping was undertaken and a virulence gene located on the largest chromosome.

Broad-spectrum resistance of barley to powdery mildew – one trait, many genes

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Broad-spectrum (race-nonspecific) pathogen resistance is of high importance to plant breeders due to its durability. However, it is usually controlled by multiple quantitative trait loci and therefore, challenging to handle in breeding practice. Knowing about the underlying genes would allow its more targeted utilization by allele introgressions. With the available omics tools and data of barley and one of its major fungal pathogens, the powdery mildew fungus *Blumeria graminis* f.sp. *hordei*, at hand we are now enabled to functionally address genes for defense and attack on both sides of this plant-pathogen interaction at a genome-wide scale. To identify genes that mediate race-nonspecific resistance of barley to *B. graminis* we combined a functional-genomics approach based on genomewide transcript profiling and transient-induced gene silencing (TIGS, 1400 genes) with a genetic approach consisting of association- and *Meta-QTL* mapping plus analysis of copy-number variation. This guided us to a shortlist of approximately 50 candidates with converging evidence for an important role in race-nonspecific resistance of barley. In *B. graminis* we can combine gene-expression studies with host-induced gene silencing (HIGS) to address potentially important genes for fungal attack and accommodation. This is revealing another list of candidates for further studies including protein-protein interactions between host and pathogen. In conclusion, the integration of functional-genomic with genetic approaches may accelerate the discovery of genes underlying complex, quantitative traits in barley and in other crop plants, with the prospect to utilize and combine favourable alleles in a knowledge-based approach.

Evaluation of a *Pyrenophora teres* f. *teres* mapping population shows multiple independent interactions with the barley 6H chromosome region

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The necrotrophic fungal pathogen *Pyrenophora teres* f. *teres* causes the foliar disease net form net blotch (NFNB) on barley (*Hordeum vulgare*). To investigate the genetics of virulence in the barley- *P. teres* f. *teres* pathosystem, we used 118 progeny derived from a cross between the California isolates 15A and 6A. The barley lines Rika and Kombar were chosen based on their differential reactions to 15A and 6A and were evaluated for NFNB disease caused by the 15A × 6A progeny. Genetic maps generated with SNP, SSR, and AFLP markers in the fungal population were scanned for quantitative trait loci (QTL) associated with virulence in *P. teres* f. *teres*. Genes underlying two major QTL, VR1 and VR2 were associated with virulence on Rika barley, accounting for 35 and 20% of the disease variation, respectively. Two different genes, VK1 and VK2, were shown to underlie two major QTL associated with virulence on Kombar barley accounting for 26 and 19% of the disease variation, respectively. A multiple regression model using markers associated with each of the virulence QTL associated with disease on Rika and Kombar accounted for 48% and 41% of the disease phenotype, respectively, showing that the genes associated with each barley line were largely additive. Progeny isolates harboring VK1, VK2, or VR2 alone were inoculated onto the Rika × Kombar recombinant inbred line mapping population and the susceptibility induced by each pathogen genotype corresponded to the same region on barley chromosome 6H as that identified for the parental isolates 15A and 6A. Data presented here indicate that the *P. teres* f. *teres* – barley interaction can at least partially be explained by a necrotrophic effector triggered susceptibility (NETS) model where pathogen produced necrotrophic effectors (NEs) interact with dominant barley susceptibility genes to induce disease.

Image analysis methods for studying resistance during early infection of barley by *Rhynchosporium commune*.

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Rhynchosporium commune is a highly destructive fungal pathogen of barley. However, *R. commune* can grow extensively and even sporulate without producing visible diseasesymptoms. Little is known about what causes the switch between asymptomatic and symptomatic infection and how this phase interacts with host resistance, butsymptomless infection may have important consequences for subsequent epidemic development.

Recently developed GFP transformed *R. commune* isolates, in conjunction with confocal microscopy, have been used to make detailed examinations of fungal growth during the asymptomatic phase, but the time required to process the large number of images generated in such studies, and the difficulty of deriving quantitative traits limit their effectiveness as a tool for genetic studies.

To complement these microscopic methods, image analysis techniques and tools were developed to automate the processing of images and identify quantitative traits related to resistance. These methods were used to identify a number of detailed morphological traits associated with fungal growth up to 9 days post inoculation.

Two uncorrelated morphological traits were identified which accounted for most of the variation in growth patterns between images. These traits (along with spore germination rates) were tested in 5 cultivars with differing resistance levels. Significant cultivar effects were identified in all three traits, with patterns of variation between cultivars, differing for each of the three traits. This suggests that these methods are able to distinguish between the effects of specific resistances on early interaction between *R.commune* and barley.

Characterization and Discovery of Barley Leaf Stripe Resistance Genes

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Barley leaf stripe is caused by the seed-transmitted hemi-biotrophic fungus *Pyrenophora graminea*. Two resistance genes are known to confer race-specific resistance to leaf stripe: *Rdg1a* and *Rdg2a*. *Rdg2a*, effective against isolate *Dg2*, was isolated by positional cloning and functionally characterized. The *Rdg2a* locus carries three sequence-related CC-NB-LRR encoding genes. Sequence comparisons suggested that the three paralogs evolved through a recent gene duplication. Transformation of the susceptible cv. Golden Promise with two *Rdg2a*-candidates identified a member of the CC-NB-LRR gene family that conferred resistance against *Dg2*. Histological analyses demonstrated that the *Rdg2a*-resistance involves cell wall reinforcement preventing pathogen colonization without any detectable hypersensitive cell death response. The genome evolution and diversity at the *Rdg2a* locus was studied comparing this locus in two barley cvs., Thibaut (resistant) and Morex (susceptible). An overall high level of sequence conservation interrupted by several rearrangements, including three main deletions, was observed in Morex. The largest 13,692-bp deletion was most likely derived from an unequal crossing over between *Rdg2a* paralogs generating a chimeric *rdg2a* gene, not associated to detectable levels of resistance toward leaf stripe. Haplotypic variability at the *Rdg2a* locus was studied in 29 *H. vulgare* lines and one *H. vulgare* ssp. *spontaneum* and highlighted large variability in the cultivated barley gene pool suggesting a rapid and recent divergence at this locus. Allele mining of *Rdg2a* for five barley *Dg2*-resistant genotypes identified two lines with non conservative mutations in the CC-NB and LRR encoding domains, respectively. The polymorphic sites were subjected to positive selection, indicating that both positive selection and divergence at homologous loci possibly represent the molecular mechanisms for the generation of high diversity at the *Rdg2a* locus. A Genome-Wide Association Scan experiment was carried out using a low-structured collection of about 210 spring 2-rowed European barley cvs. tested for resistance to isolate *Dg5*, against which *Rdg2a* is not effective. The cvs. were genotyped with 7,864 gene-based SNPs incorporated into a single Illumina Infinium™ iSelect assay. SNPs mapping on the short arm of barley chromosome 6H (8.6–13.6cM) showed a significant association with *Dg5*-resistance. This genomic region is syntenic with a 240-Kb region of rice chromosome 2, containing 42 annotated genes that could serve for identifying candidate resistance genes or developing new markers.

Development of the Maxwell[®] 16 LEV Plant DNA Kit, and its application to plant leaf tissues.

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Promega is a leader in providing innovative solutions and technical support to the life science research fields. Promega portfolio includes many products for nucleic acid extraction, including Maxwell[®] 16 Instrument, a high quality automated system that combines compact instrumentation, prefilled cartridges and optimized automated methods to maximize performance and flexibility while minimizing hands-on time required for DNA and RNA purification. Plant molecular studies have become increasingly complex, requiring high concentration DNA isolated from plant tissue samples. These studies frequently require extraction and purification of microgram-amounts of DNA from plant leaf tissues which employ laborious methods to extract and isolate genomic DNA, such as organic extraction with CTAB (hexadecyltrimethylammonium bromide). Alternatively, DNA can be isolated through binding to the surface of a paramagnetic particle, which has the advantage of being adaptable to automation, as has been done with the Maxwell[®] 16 instrument, a benchtop instrument designed for this purpose. We have recently developed a DNA purification method from plant lysates built around a novel cellulose-based paramagnetic particle, called the MagnaCel[®] particle. This method consists of two key features: after a brief extraction process using mechanical disruption, the resulting crude lysate is added to a disposable Maxwell[®] 16 cartridge. This cartridge was designed to allow the Maxwell[®] 16 instrument to perform this chemistry to isolate microgram amounts of DNA from plant lysates in about 45 minutes. Second, the chemistry was designed to place the resulting DNA into a low elution volume (50µl). The resulting automated system results in a high concentration DNA isolates from crude tissue lysates. We can demonstrate the features of this chemistry, as it applies to problems in automating DNA isolation from plant tissue lysates. While the formal development of this chemistry has centered around extraction of leaf tissue from three species used as research models (*Zea mays*, *Glycine max*, and *Arabidopsis thaliana*), we can demonstrate initial success of application of this chemistry to other species and tissue types.

Current status of scald and net blotch diseases of barley in Turkey

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Scald caused by *Rhynchosporium secalis* and net blotch caused by *Drechslera teres* are two main diseases affecting barley in Turkey. Surveys were carried out in important barley growing areas of Turkey in 2012 and 2013. Two hundred seventy-four and 105 barley fields were surveyed in 2012 and 2013, respectively. In 2012, net blotch was found in 219 fields (79.9%) and scald was found in 215 fields (78.5%). In 2013, net blotch was found in 76 fields (72.4%) and scald was found in 73 fields (69.5%). Net and spot forms of *Drechslera teres* were found, however, spot form of net blotch was more common. The incidence values (number of plants infected of those examined) of net blotch and scald were between 1-100%. Disease severity values of both *Rhynchosporium secalis* and *Drechslera teres* ranged over all values of the 1-9 scale. It appears that these diseases are widespread and they are the most important leaf diseases of barley in Turkey.

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Emergence and importance of *Rhynchosporium secalis* in Morocco

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Leaf scald of barley is caused by an imperfect fungus *Rhynchosporium secalis* (Oudem.) J.J. Davis, and is one of the most economically important diseases of barley worldwide (Williams et al., 2003). In Morocco, Leaf scald is immerging since 1975 (Boulif, 1975) and the prevalence is less than net blotch and Powdery mildew. In fact, during the last 10 years of prospection, the prevalence was increasing and largely exceeds the two above mentioned diseases (Bentata, 2013). The prospection realized during Mars 2014 in Chaouia, Doukkala, Abda, Zaer, Zemmour, Gharb, North and the Mountain, showed that on 62 fields visited, 50% are infected by the scald. Indeed, the rate of infection reached in some fields 90%. The fields whose stage phenologic arrived at flowering, the rate did not exceed 25%. The high rates were observed especially in Gharb and North with an average of 70%.

Characterization of ICARDA barley germplasm lines for leaf blight resistance in India

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The identification of resistance sources to barley leaf blight is an area of intensive research in India. *Bipolaris sorokiniana* is the major pathogen associated with leaf blight diseases in India. Three hundred sixty ICARDA barley germplasm were evaluated during 2012-13 crop season for leaf blight plant resistance under artificial field epiphytotic conditions at hot spot locations Faizabad and Varanasi. For phenotyping, disease severity was recorded at three different growth stages using double-digit scale. The average of three stages for each genotype was calculated by taking mean of first and second digit separately and the infection responses were categorized as Immune, Resistant, Moderately resistant, Moderately susceptible, Susceptible and Highly susceptible. More than 30 lines were showing moderately resistance to leaf blight at both locations. The Area Under Disease Progress Curve (AUDPC) was calculated and the lines were showing range between 393 and 1404. The lowest AUDPC was recorded for entries viz., Isaria, Defra, Diamant, C 9173, C9609, FNC 6-1, Berolina, ND 19957, MN 599, CLE 203 INYA AROMO, L94, PAPELILLO, CABUYA, Bella Union, Hiproly, Weihestephan Meltharesistente I, Sebastian, Ana, ZIGZIG/ BLLU// PETUNIA 1P.STO/3/LBIRAN/ UNA80//LIGNEE640/4/ BLLU/5/ PETUNIA 1, C97073, AYAROSA/ BLLU//CALI92, Peruvian, Haisa and BLLU/ MINN DESC 2//PETUNIA 1. The highly susceptible line RD 2503 showed the highest AUDPC value of 1404. The identified resistant germplasm lines are being utilized in barley breeding programme under CRP –ICARDA Barley project as donors for leaf blight resistance.

Mitigating barley foliar diseases at global scale through germplasm enhancement at ICARDA

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The International Center for Agriculture Research in the Dry Areas (ICARDA) promotes the livelihoods of smallholder farmers in the Dryland regions of Africa and Asia through innovative research and development platforms in its six focal countries (Ethiopia, Kazakhstan, India, Iran, Morocco and Turkey). Barley (*Hordeum vulgare* L.) is one of the crops for which ICARDA has a global mandate to preserve its genetic diversity, to develop and distribute improved and better adapted barley germplasm for sustainable production in dry areas. Amongst various biotic yield limiting factors, the economic losses inflicted by the foliar blights are substantial. For example, alone in Morocco the annual yield losses caused by net & spot form of net blotch (*Pyrenophora teres*) accounts for 29 (El-Yousfi and Ezzahiri, 2002). The main activities of barley crop protection at its Rabat (Morocco) platform include surveillance and disease monitoring, dynamics of the pathogen population and forecast disease risk in a region. One of the survey objectives is to develop a risk map for important diseases of barley based on the survey data, weather data and geographical information. Study of pathogen population dynamics is monitored through trap nurseries representing the cultivars with known sources of resistance (differential set) and representative cultivars of the respective country (local checks). These trap nurseries are being organized at National Agriculture Research Stations (NARS) in many countries including the disease hot spots to monitor the variations in the pathogen population dynamics. The evaluation of available germplasm with various pathogenic isolates deciphers the importance of resistance genes which could be used in the breeding programs to mitigate the new virulent races. The barley germplasm is being evaluated with Focused Identification of Germplasm Strategy (FIGS) for screening with emerging races of fungal pathogens such as Net blotch, powdery mildew, scald and stripe rust. This exercise will result in the identification of novel sources of resistance which will be incorporated in the elite barley lines for distribution to target regions.

Phenotypic diversity of *Pyrenophorateres* f. sp. *teres* in Uruguay, Morocco and Syria.

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A collection of one seventy nine single conidia isolates from different regions was investigated in 2011-2014 in a proposed international set of twenty one barley genotypes. The studies were conducted at seedling stage and under controlled conditions. The frequency of virulent isolates to each barley genotype, pathotypes composition, and share of identical phenotypes in different geographic populations were studied. Considerable variability appears to exist in the current Uruguayan, Moroccan and Syrian populations of *Pyrenophorateres* f. sp. *teres*. No similarity was found among these populations. Testing of additional isolates is needed to obtain more comprehensive information on the diversity of virulence, determine if current resistance sources are adequate and validate the usefulness of the international differential set to characterize the local *P. teres* f. sp. *teres* populations. A comprehensive and continuous evaluation of *P. teres* in Uruguay, and elsewhere is warranted to assess the current presence, distribution, and virulence of the pathogen.

Status of barley leaf diseases in North Shewa Highlands, Ethiopia

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Barley (*Hordeum Vulgare* L.) is one of the leading food crops in Ethiopia. However, the production of the crop is threatened by a number of biotic and abiotic factors, among which leaf diseases are the major constraints for its production in North Shewa highlands as well as other areas of Amhara region. Field survey was conducted in 2011 main cropping season from booting to heading crop growth stage. The main objective of the survey was to see the severity, incidence and prevalence of barley leaf diseases in major barley growing areas. Scald (*Rhynchosporium secalis*), Net blotch (*Pyrenophora teres*), spot blotch (*Chochilobolus stativus*), Loose smut (*Ustilago nuda*), Covered smut (*Ustilago hordei*), leaf rust were the major diseases recorded in many fields. Among the above diseases scald, loose smut and Net blotch were the most important diseases respectively. About 80 fields were observed in the season. Five varieties were encountered in farmer's fields, namely farmers local, Basso, Miserach, Agegnehu, & mixed unknown varieties. No variety was found immune to scald. Scald was the most widely occurring and devastating diseases in all barley growing fields. In some fields 100% incidence and about 50% severity was recorded due to scald disease.

Towards durable resistance to scald in barley

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Testing of scald isolates reveals a wide diversity of pathotypes with virulence detectable on all known major seedling resistance genes and most minor adult plant resistance genes. Durable resistance has not been reported although Schooner has been observed to maintain a low level of resistance in field trials despite the variety being grown over large areas for 30 years. Durable resistance might be achieved through the pyramiding of major seedling genes or minor (adult plant) resistance genes. The former will require molecular markers and/or the use of multiple screens using selected differential isolates that have been identified in SARDI. Minor genes can be combined through phenotypic selection of transgressive segregants using virulent isolates although the use of molecular markers would certainly increase selection efficiency. Tests using a diversity of differential scald isolates have revealed several new donors of major seedling resistance genes. These have been developed into single seed /doubled haploid populations and are being phenotyped to identify possible new resistance loci. Resistant lines from these initial populations that display improved agronomic characteristics are being crossed back onto elite parents for delivery of novel resistances in well-adapted backgrounds. These can then be inter-crossed to generate effective gene pyramids in lines immediately useful to barley breeding programs. Minor genes have been identified through mapping of three doubled haploid populations, Schooner/WI3580, Mundah/Keel and Vlamingh*Buloke. Phenotyping of these populations and similar populations has been made more accurate using specific avirulent isolates that have been selected for their ability to detect specific minor genes in plants at the seedling stage. Transgressive segregants from the first two of these crosses are being used to transfer multiple minor gene resistance into elite varieties and to combine together to create new lines with potentially higher levels of transgressive and hopefully durable resistance. Molecular mapping in these populations has identified QTL on 4H and 7H in Schooner, 5H and 6H in WI3580 and 3H, 6H and 7H in Keel. Crosses between the two transgressive segregant lines have the potential to pyramid 6 minor genes in reasonably well adapted germplasm.

Achievements and challenges in breeding for durable net blotch resistance

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The frequency of barley pathogen infected fields has significantly increased during the past 40 years in Finland. Based on the long term field trials, the main barley pathogens *Pyrenophora teres*, *Rhynchosporium secalis* and *Blumeria graminis* cause annual yield losses averaging 600 kg/ha on a susceptible barley cultivar. The challenges will even increase in the near future when changing climate couples with low-input farming systems aiming to reduce environmental footprint. Disease resistance is an environmentally and economically sustainable means to manage plant pathogens. Genetic resources may carry the needed traits to minimize cultivation risks. In our project ‘Genetic resources and genomics for adapting barley to climate change’ we studied the resistance of 985 barley cultivars and landraces with different geographical origin for their net type (*Pyrenophora teres* f. *teres*) and spot type (*Pyrenophora teres* f. *maculata*) resistance. Also, part of the material was studied for genes controlling resistance traits by association genetics. A significant improvement in the net blotch resistance level was found in the European barley cultivars released during the last 40 years. The frequency of resistant genotypes against net type net blotch was highest among the European barley cultivars and Syrian landraces. For the spot type net blotch, the average resistance level in the studied material was clearly weaker than for the net type net blotch. Most promising material was found in the landraces originating from Jordania. The *P. teres* virulence surveys made in 2010-2012 showed increased frequency of *P. teres* f. *maculata* in the Finnish barley fields. The changes in the *P. teres* species and virulence spectrum were rapid especially in the barley monoculture under minimum tillage.

Insights on Durable Resistance to the Net Form of Net Blotch in Barley

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Development of barley (*Hordeum vulgare*) cultivars with durable resistance to the net form of net blotch (*Pyrenophora teres* f. *teres*, *Ptt*) has proven difficult. Accessions CIho 9819 and CIho 5791 from Ethiopia were identified in the 1950s and 1960s as resistant to all *Ptt* isolates tested. Recent surveys worldwide have shown that these accessions are still resistant to most *Ptt* isolates; however, this stability in response to *Ptt* isolates was not fully recovered from CIho 5791 in cultivars bred for Canada. To evaluate the inheritance of resistance, differentials for response to *Ptt*, cultivars, breeding lines and a doubled-haploid population were evaluated in Queensland for reactions to *Ptt* isolates and characterized for molecular markers using the DArTseq genotyping by sequence (GBS) platform. Based on similar marker haplotypes in a 5 to 8 cM segment of chromosome 6HL, the reaction to *Pyrenophora teres* 5 (*Rpt5*) locus from CIho 5791 is present in both resistant and susceptible cultivars. Published inheritance studies using other barley accessions and other *Ptt* isolates have demonstrated that the *Rpt5* region of 6HL contains a series of isolate specific genes. Since the mapping studies of Bockelman et al. (1977) did not detect a resistant allele at the *Rpt5* locus in CIho 9819, *Rpt* genes at other loci, as reported by Manninen et al. (2006), are involved in full expression resistance associated with the *Rpt5.f* allele of CIho 9819 and CIho 5791. Dissection of marker haplotype patterns in the *Rpt5* region of 6HL of breeding lines and cultivars has identified a number of recombinants. The recombinants show a range of reactions to *Ptt* isolates from susceptible to highly resistant and some have isolate specific reactions. Thus, breeding cultivars with durable resistance to *Ptt* could involve selection for a specific recombinant at the *Rpt5* locus, which provides relatively isolate non-specific resistance, plus adding two or more *Rpt* genes at other loci.

Barley Disease Resistance in Western Australia – Issues and Solutions

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Disease threats are ubiquitous in barley grown regions of Western Australia where crops are affected by a range of leaf and root diseases. It constitutes a significant limitation to sustainable barley production. Reducing impacts from diseases is important for reliable production of high quality barley. The lack of resistance to diseases like net blotches, scald, leaf rust and powdery mildew can impact on yield and grain quality mainly through reduced grain size. Reduced grain yields and low malting quality reduce returns to growers and affect domestic and overseas exports for its market ability. An integrated approach to disease control is recommended, yet genetic resistance underpins effective management strategies in barley production systems to develop varieties with adequate levels of resistance to the range of important biotic stresses. A major objective in barley breeding programs around Australia is to develop varieties with improved combinations of disease resistances without compromising its yield, quality and other agronomic traits. Disease epidemics are variable in incidence and severity across regions and seasons. This has been due to the identification of variability and virulence patterns in various leaf diseases. The new variability and virulences have been identified using differential sets for powdery mildew, net and spot form of net blotches and leaf rust, which were unidentified before in Western Australia. Current studies indicate resistance genes for powdery mildew, net blotches and leaf rust have broken down with the incursion of new pathotypes. This has further impacted the barley varieties making them susceptible to the various diseases. Efforts are underway to screen barley varieties, advanced lines and germplasm to fully understand their current disease status and also identify novel resistance genes operative at adult plant stage for various diseases. Incorporating and combining such adult plant resistance genes with seedling resistances in the development of new barley varieties will help to reduce disease impacts.

Mapping and exploitation of new sources of resistance to the net form of net blotch (*Pyrenophora teres f. teres*) in barley

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Breeding for resistance against *Pyrenophora teres f. teres* (*Ptt*) in barley is difficult due to the high physiological variability of the pathogen and the fact that in field-trials often a simultaneous infection with other fungal pathogens occurs. To avoid this, a so called summer-hill trial was developed in which winter barley is sown at the beginning of August at optimum conditions for *Ptt* infection. Using this approach a rather high heritability for *Ptt*- resistance was estimated in the DH-populations analysed, i.e. $h^2=0.80$ for the DH-population 'Uschi x HHOR3073' and $h^2=0.62$ for '(Post x Viresa) x HHOR9484'. In parallel a detached leaf assay was conducted with a set of 11 single conidial lines (SCLs) of *Ptt* to characterize the genotypes additionally at the seedling stage. Genetic maps with 705.7cM for 'Uschi x HHOR3073' and 1035.8 cM for '(PxV) x HHOR9484' were constructed. Based on the results of the summer-hill trial in the DH-population 'Uschi x HHOR3073' QTL were detected on chromosomes 2H and 3H, each and two QTL on chromosome 5H. With respect to resistance to the SCL 'QLB' a single major gene was located on chromosome 7H in this population, two QTL were detected on chromosome 3H and 7H for resistance against SCL 'WvB' and for resistance against SCL 'd8_4' two loci were mapped on chromosome 3H. In the population '(PxV) x HHOR9484' three QTL were detected on chromosome 5H and one on 7H in the summer-hill trial. Based on the detached leaf assay one locus conferring resistance to the SCL 'AR' was located on chromosome 3H and against SCL 'net1840' two QTL were mapped on chromosome 4H and 5H. The QTL located on 5H detected based on the results of the summer-hill trial and the one obtained with SCL 'net1840' are only 2 cM apart giving hint that this locus may be effective in different developmental stages (seedling/adult). Obtained results elucidate that the summer-hill trial design is best suited to get reliable phenotypic data for *Ptt* resistance under field conditions and that the donors of resistance are differentiated by their different reactions to the SCLs tested reflecting the high variability of *Ptt*.

Mapping of qualitative and isolate specific quantitative trait loci associated with resistance to *Pyrenophora teres* f. *teres* and *Cochliobolus sativus* in two double haploid barley populations

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Net blotch of barley (*Hordeum vulgare* L.) caused by *Pyrenophora teres* f. *teres* and spot blotch caused by *Cochliobolus sativus* are the most widespread and harmful diseases in Russia. Two double haploid (DH) mapping populations were developed by use of barley anther culture: "A" from cross of the Ethiopian highly resistant to *P. teres* f. *teres* accession c-23874, with susceptible cultivar Pirkka and "B" from cross of cultivars Zernogradsky 813 (MR to *C. sativus*) and Ranny 1 (MR to *P. teres* f. *teres*). Genotyping carried out with set of 384 SNP markers at the Laboratory of Genetics of the James Hutton Institute by use of the technologies of the BeadXpress reader (Illumina Inc.). Seedling evaluation to three *P. teres* f. *teres* isolates of population "A" was done in greenhouse conditions and on detached leaves in controlled conditions. Phenotyping of "B" population to 12 *P. teres* f. *teres* and 12 *C. sativus* isolates was conducted in laboratory conditions on detached leaves. In mapping population "A" a major seedling resistance QTL (SNP-marker 11_11067, 58cM) was detected on chromosome 6H for *P. teres* f. *teres* isolates V278 (Finland), PL5 (Leningrad Region, Russia), and PN10 (Novgorod, Russia), explaining 27- 33% of the phenotypic variation. Four novel QTLs controlled resistance to *P. teres* f. *teres* isolates were found: two on chromosome 1HL, one on 4HS and 5HL, explaining 20%, 20%, 11% and 12% of the phenotypic variation, accordingly, and seven novel QTLs contributed resistance to nine *C. sativus* isolates explaining 12 – 22% of the phenotypic variation in depend of isolate using.

Identification of *Pyrenophora teres* f. *teres* resistance in South African barley line UVC8

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Net form of net blotch (NFNB) caused by *Pyrenophora teres* f. *teres* (PTT) can cause high yield losses on barley in South Africa. Even though this foliar disease has been increasing in incidence, the South African barley industry has focused on breeding for malting quality and breeding for disease resistance has been secondary. The aim of this study is to introduce NFNB disease resistance into South African barley varieties, whilst retaining malting quality (MQ) traits.

In Australia the screening of germplasm for this disease is conducted on a routine basis. This provided us with an opportunity to rapidly advance our understanding of the availability of resistant South African germplasm. Thus in 2008, 29 South African barley breeding lines were screened in the field in Australia with three characterized Australian NFNB isolates and line UVC8 was identified as showing good resistance against NFNB. This line was re-tested in South Africa in three consecutive seasons and disease resistance scores similar to those obtained under the Australian environment, were obtained in South Africa. To determine which genetic loci are associated with this NFNB resistance, UVC8 was crossed with Erica (a good malting quality parent) and the resulting doubled haploid population phenotyped in the field in South Africa in 2013. QTL were identified on chromosomes 3H and 6H in the same region as other studies have identified NFNB QTL. Molecular markers are now available for use in a marker-assisted selection to speed up the process of introducing resistance into other current South African malting quality varieties.

A CI5791 × TifangRI population shows major dominant net form net blotch resistance located on barley chromosomes 6H and 3H in CI5791 and Tifang, respectively

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CI5791 is a barley line that harbors high levels of resistance to *Pyrenophora teres* f. *teres*, causal agent of net form net blotch (NFNB), with only a few rare examples of isolates overcoming CI5791 resistance. Tifang barley is a line that has been used in the majority of pathotype diversity studies completed in the last few decades. Tifang also harbors resistance that was previously mapped to barley chromosome 3H. We have advanced and generated a saturated SNP map for an F₆ recombinant inbred line (RIL) population from a cross of CI5791 × Tifang to evaluate the genetics of resistance in these two lines. Inoculations were done using a global collection of ten *P. teres* f. *teres* isolates, including isolates collected from barley growing regions of the US, Canada, Europe, Asia, and South America. Resistance in Tifang was effective against five of the isolates used whereas resistance in CI5791 was effective against all ten isolates tested. Resistance conferred by CI5791 mapped to chromosome 6H and as expected resistance conferred by Tifang mapped to chromosome 3H as previously reported. F₂ analysis of a CI5791 × Tifang cross indicated that both the 3H and 6H resistance genes were dominant based on R:S ratios of 15:1 and 3:1 when inoculated with isolates avirulent on both Tifang and CI5791 and those isolates avirulent on only CI5791, respectively. RILs harboring both the 3H and 6H resistance along with the associated flanking markers will be useful in efforts to breed for resistance to NFNB.

Effect of net blotch disease (*Pyrenophora teres* f. *teres*) on yield and grain quality of winter barley

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Winter barley (*Hordeum vulgare* L.) is one of important species, grown in Romania especially for malt, beer, feed and lately for food products. The most damaging disease, which occurs every year in the south-east of the country and affects yield and quality of grain, is net blotch disease (*Pyrenophora teres* f. *teres* Smedeg.). The most efficient way to alleviate the problem is to breed new winter barley varieties with genetic resistance to disease. The use of diverse combinations of resistance genes is an economic method to minimize yield losses and to improve the quality. Winter genotypes released at NARDI Fundulea have not been sufficiently tolerant to *P. teres*. As a consequence, the main objective of winter barley breeding program is to identifying useful backgrounds into which genes from highly resistant sources can be introgressed.

In order to study the losses of yield and quality associated with specific levels of tolerance and the impact of this disease on the yield and grain quality, two separate experiments were conducted during 2011-2012 and 2012-2013. Thirty varieties and lines (six rows and two rows) obtained by pedigree method or by *bulbosum* method varying in yield potential and grain quality characteristics were tested under natural infection conditions (non-treated) and treated with fungicide at stem elongation stage. Analysis of variance indicated differences among genotypes in grain yield, TKW, protein content, starch content and size of seeds (six and two rows winter barley varieties and lines). The genotypes varied in response and few of them had a reasonable behavior under both conditions, regarding yield (F 8-41-2006, F 8-19-2010, DH 220-5) and at least one grain quality parameter (Ametist, DH 334-8). Many of them were very susceptible to this disease as indicated by the inadequate values of grain quality indices (lower than accepted parameters for malting and brewing). In 2012, yields were reduced 14 to 21%, TKW from 6 to 11% and in 2013, yield losses were 17 to 38% and TKW from 10 to 18%. Protein content, starch content and size of kernel were affected also by this disease. Analysis of variance indicated differences among genotypes in grain yield, TKW, protein content, starch content and size of seeds (six and two rows winter barley varieties and lines). The disease had a significant influence on yield and TKW and no significant for protein and starch content. Interaction year x disease was no significant for yield and protein content in the case of two rows winter barley. In addition, no significant interactions ($P=5\%$) between genotypes x disease and year x genotypes x disease were observed for all studied characteristics. According to the results of these experiments, few variety and lines possess adequate levels of tolerance to *P. teres* and will be useful parents in future crossing programs, one of the main aim of winter barley breeding program being improvement of net blotch resistance in Romanian winter barley.

Can susceptibility to net blotch in barley be explained by sensitivity to necrotrophic effectors?

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Net blotch is a major barley disease in Norway caused by the necrotrophic pathogen *Pyrenophora teres* leading to yield losses of up to 40%. The closely related wheat pathogens *Parastagonospora nodorum* and *Pyrenophora tritici-repentis* secrete necrotrophic effectors (NEs) which act as virulence factors in order to gain entry and nutrient in the host. NEs cause cell death in the presence of corresponding dominant host susceptibility factors. The main objective of this study is to examine the potential role of NEs and corresponding host receptors in explaining susceptibility to net blotch in barley. This knowledge together with an understanding of the genetic background of the Norwegian net blotch population will be utilized to speed up resistance breeding. By collecting naturally infected barley samples from different regions, a representative isolate collection is being established which will be sequenced in order to assess genetic diversity and population structure of the Norwegian net blotch population. Selected isolates and their culture filtrates will be screened for specific reactions against a) differential barley lines to investigate the role of NEs, b) a collection of ca. 200 barley lines for association mapping and c) segregating mapping populations to characterize novel NE-host susceptibility interactions and to map the corresponding sensitivity loci. Effector protein candidates will be purified and further analysed to verify their effect on disease development. Preliminary screenings with a small number of isolates revealed a resistance/susceptibility QTL on barley chromosome 3H in the Hector x NBD112 mapping population and two resistance QTL on 3H and 6H in the CI5791 x Tifang mapping population under greenhouse conditions. Field testing of the association mapping panel in 2013 revealed large differences in net blotch resistance among Norwegian barley cultivars and breeding lines. The field data also indicated recent changes in the pathogen population. For instance the current cv. 'Heder' was the most susceptible of all entries, while it previously had been considered to have good resistance. 'Arve' was widely grown in the 1990's and considered highly susceptible, but was rated moderately resistant during the field testing in 2013.

Adult plant resistance to leaf rust in barley: the story so far

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Barley leaf rust (caused by *Puccinia hordei*) is a major disease of barley (*Hordeum vulgare*) crops worldwide. In Australia, the disease is estimated to cause average annual losses of \$21 million (Murray and Brennan 2009). It often causes yield losses up to 32% in Australia and North America (Park and Karakousis 2002, Plant Breeding 121:232–236), with losses as high as 60% in very susceptible varieties (Cotterill et al. 1995, Aust. J. Agric. Res. 46:127–134; Castro et al. 2002, Crop Science 42:1701–1708). Since the late 1990s, it has emerged as a significant national limitation to barley production in most Australian barley production areas. Research initiated at the University of Sydney in 1989 has led to a sound understanding of the pathogenicity of the causal agent *P. hordei* and of major gene resistance in Australian barley cultivars. Research we initiated in 1998 identified a number of sources of adult plant resistance (APR) to leaf rust in barley, including the Dutch cultivar Vada. Subsequent genetic analyses resolved the resistance to leaf rust in Vada and a range of other European and Australian barley genotypes to a locus on chromosome 5HS, which was subsequently designated *Rph20*. Extensive greenhouse and field phenotyping over the past 16 years has led to the identification of more than 50 diverse barley genotypes that carry APR to leaf rust. We have shown in tests of allelism that at least six of these have genes conferring APR that are independent of *Rph20*, and that act in an additive manner to provide high to very high levels of resistance to leaf rust. In very recent work, we characterised a second gene conferring APR to leaf rust in barley, *Rph23*, located in chromosome 7HS and very closely (< 0.1cM) associated with the marker Ebmac0603. *Rph23* acts in an additive manner with *Rph20* and a third uncatalogued gene conferring APR on chromosome 3H. Future research will focus on resolving the identities of the APR in the remaining genotypes we have identified, assessing the potential of each in contributing additive resistance, and also looking for potential beneficial pleiotropic effects on resistance to other biotic stresses.

Novel Additive Adult Plant Resistance Sources to Leaf Rust in Barley

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Leaf rust, caused by *Puccinia hordei* is one of the most destructive foliar pathogens of cultivated barley, causing significant yield losses in all growing regions throughout the world. Identifying new sources of APR in barley to leaf rust is essential to decrease the overuse and reliance on all-stage resistance and *Rph20*. We report on a new adult plant resistance (APR) gene *Rph23* conferring resistance to leaf rust in barley. The gene was identified and characterized from a double-haploid population derived from an inter-cross between the Australian barley varieties Yerong (Y) and Franklin (F) that were both seedling susceptible to the *P. hordei* pathotype 5457P+. Genetic analysis of the adult plant field leaf rust scores of the Y/F population collected over three successive years indicated the involvement of two highly additive genes controlling APR, one of which was named *Rph23*. The gene was mapped to chromosome 7HS positioned at a genetic distance 36.6 cM. *Rph23* is closely linked to marker Ebmac0603 at a recombination value ~ 0.8cM. A PCR-based marker was optimized for marker assisted selection (MAS) of *Rph23* and on the basis of this marker, the gene was postulated as being common in Australian and global barley germplasm. Pedigree and molecular marker analyses indicated that the six-rowed black Russian landrace 'LV-Taganrog' is the likely origin of *Rph23*. International barley germplasm from countries including Australia, China, Germany, Spain, and Uruguay were further assessed for APR response in field nurseries over three successive years and then screened with markers linked to *Rph23* and the previously identified APR (*Rph20*). There was high variability for the levels of APR within specific international germplasm collections and in many cases, uncharacterized sources of APR were postulated based on the absence of markers for *Rph20* and *Rph23*. The presence of *Rph20* was strongly correlated with high levels of APR in the field, while lines that were positive for the marker for *Rph23* only often carried lower levels of APR. Both *Rph20* and *Rph23* were found in combination only in three lines from Germany and in a single Australian cultivar, all of which were highly resistant under field conditions. The phenotypic assessment of international germplasm collections for APR and subsequent marker screening with closely linked markers to *Rph20* and now *Rph23* provide a valuable genetic resource for MAS and enable the identification of uncharacterised APR.

Association mapping of resistance to *Puccinia hordei* in Australian barley breeding germplasm

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Elite barley (*Hordeum vulgare* L.) breeding lines from the Northern Region Barley Breeding Program in Australia were evaluated for both seedling and adult plant resistance (APR) to leaf rust (*Puccinia hordei* Otth), and association mapping (AM) was performed to identify genomic regions harbouring resistance genes. The germplasm was initially derived from different geographic origins or hubs of international barley breeding ventures and represented Stage 2 material (F3:5 lines) spanning 4 years of trials (2009, 2011, 2012 and 2013). The 2009 and 2011 breeding populations (i.e. BP1 and BP2) comprised a total of 360 lines and were genotyped with 3,244 polymorphic diversity arrays technology (DArT) markers, whereas the 2012 and 2013 breeding populations (i.e. BP3 and BP4) comprised a total of 390 lines genotyped with the DArT genotype-by-sequencing array (DArTseq), which provided 14,855 high quality polymorphic markers with map positions based on the DArTseq consensus map and physical positions based on the barley reference genome. AM was performed using mean scores for disease reaction, accounting for family effects using the eigenvalues from a matrix of genotype correlations. AM of BP1 (two environments) and BP2 (four environments) detected 15 QTL; 5 QTL co-located with catalogued leaf rust resistance genes (*Rph1*, *Rph3/19*, *Rph8/14/15*, *Rph20*, *Rph21*), 6 QTL aligned with previously reported genomic regions and 4 QTL (3 on chromosome 1H and 1 on 7H) were new. The APR gene *Rph20* was identified most environments and pathotypes. The supplementary data provided for DArT and DArTseq allowed alignment of markers across marker platforms and the integration of mapping results for leaf rust across the four breeding populations (13 environments), which were visualised on a single map. Genomic regions associated with leaf rust reactions across multiple breeding populations and environments were shortlisted and will be recommended for pyramiding in barley breeding programs to achieve more durable resistance to leaf rust.

National Barley Foliar Pathogens Variety Improvement Program – a coordinated approach to disease control

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Until recently, Australian research into the control of foliar diseases of barley other than rusts has been conducted by individual State public research providers with the support of the national Grains Research and Development Corporation (GRDC). This approach was reasonably successful and served the State-based breeding programs quite well; however it did not adequately address the host/pathogen differences among states and regions, nor did it adequately service Australia's entry into commercial barley breeding. Furthermore the increasing cost of agricultural research demanded a more efficient approach to addressing like problems across multiple states. In 2013, a National Research Strategy was implemented where research on a common theme is implemented under a national program. This led to the aggregation of several research projects on the control of foliar diseases of barley into a single entity, The National Barley Foliar Pathogens Variety Improvement Program (NBFPVIP), with the emphasis on genetic solutions. Research providers from six state departments of agriculture and five universities now collaborate in the NBFPVIP coordinated by the Department of Agriculture Fisheries and Forestry Queensland and with a total budget of \$11.1million over 5 years. This project covers research into pathogen population surveys of scald (*Rhynchosporium commune*), net and spot forms of net blotch (*Pyrenophora teres* f. *teres* and *maculata*), spot blotch (*Cochliobolus sativus*) and powdery mildew (*Blumeria graminis* f.sp. *hordei*). It coordinates national screening of diverse germplasm for 9 diseases, identifying superior sources of resistance with the aim of understanding the genetics of inheritance, developing molecular markers and resistant germplasm for use by breeding companies. The NBFPVIP is also investigating the evolution of new virulences in some pathogens and the role of naturalised grasses in epidemic development. This paper explains the background and structure of the NBFPVIP to deliver superior sources of resistance in adapted lines to private breeding companies, free of charge and highlights the benefits of a national approach to disease control.

Validation of rapid gene transfer methodology in barley

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New methodology for rapid trait introgression has been developed for spring bread wheat (*Triticum aestivum*). The strategy combines rapid generation advance under controlled environmental conditions, high-throughput phenotypic screening for multiple traits and utilisation of high-throughput DNA markers. We validated this approach in a second crop species using the European barley (*Hordeum vulgare*) cultivar Scarlett. Scarlett is well-adapted for production in Argentina and is preferred for malting and brewing; however it lacks adequate foliar disease resistance needed to minimise production losses. In 2011, four donor lines with multiple disease resistance from the Northern Region Barley Breeding program in Australia were crossed to Scarlett. Large F₂ populations were screened as seedlings for resistance to four foliar diseases: net and spot forms of net blotch (*Pyrenophorateres* f. *teres* and *P. teres* f. *maculata*), spot blotch (*Bipolaris sorokiniana*), and leaf rust (*Puccinia hordei*). Selected F₃ plants were backcrossed to Scarlett. Phenotypic screening of seedlings was repeated in the BC₁F₂ and BC₁F₃ generations and selected plants were grown to maturity to generate 94 BC₁F₃-derived Scarlett introgression lines (SILs). The SILs were genotyped with DArTseq genotype-by-sequencing (GBS) markers and marker haplotypes associated with previously characterised quantitative trait loci (QTL) for resistance to the four foliar pathogens were used to confirm the transfer of critical genomic regions into the Scarlett background. In 2013, the SILs were assessed in disease screening nurseries conducted in Australia and Uruguay and association mapping was performed to confirm donor gene effects. Using phenotypic data (disease profiles, agronomic and micro-malting data), an elite subset of 10 SILs was selected for yield and quality evaluation in Argentina. This methodology could be applied to incorporate multiple disease resistances into other barley cultivars or rapidly combine desirable traits via top crossing or parent building in breeding programs.

**...For the times they are a changing...hence the diseases they are a changing.
Climate change and barley diseases**

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Ongoing anthropogenic climate change (see the latest synthesis report: IPCC, 2013; Stocker et al. Cambridge University Press, available for download at www.ipcc.ch) has profound effects on the interactions within the disease triangle. Climate change comprises shifts in averages and extremes of climate elements as well as eventually changes in atmospheric circulation patterns. Furthermore, not only the physical climate is changing but also the chemical climate, i.e. the atmospheric gas composition. Changes in the atmospheric CO₂ mixing ratio is one of the most prominent of these alterations and at the same time the most important cause of current anthropogenic climate change. Thus, environment as one of the elements in the triad of the disease triangle is subject to manifold changes in its climatic facets, that in turn modify the performance of hosts and pathogens as well as their interaction on all relevant temporal and spatial scales.

Effects concerned are shifts in:

- * the regions suitable for cultivation of diverse crop species,
- * the geographical distribution ranges of pathogens,
- * crop potential production and biochemical composition of crop tissues including resources available for defence,
- * pathogen cycles
- * the composition of soil and rhizosphere biota
- * match or mismatch of host and pathogen phenology.

Studies of climate change impacts on crop diseases are still in their infancy. The talk will address:

- * climate elements subject to change
- * data availability for climate impact studies
- * examples of studies on climate change and crop diseases with a special focus on emerging approaches for the study of climate impacts and on barley diseases.

Impact of climate change on susceptibility to powdery mildew and spot blotch disease in barley

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The predicted changes in the world's climate are believed to affect the physiology of plants, and their interaction with pathogens. It is generally hypothesized that crop plants may become more prone to diseases in the future, but it is difficult to generalize, and not much information is available from experiments using multi climatic factors. Using a climate phytotron, we have examined how susceptibility to powdery mildew and spot blotch disease were affected when barley was grown in different climatic environments with elevated [CO₂] (700 vs. 385 ppm), [O₃] (60/90 vs. 20 ppb) and temperature (24/19 vs. 19/12°C day/night) as single factors and in combinations. When growing under elevated temperature or [O₃], infection by the biotrophic powdery mildew fungus decreased, whereas disease symptoms and growth of the toxin secreting hemibiotrophic spot blotch fungus increased compared to ambient conditions, implying that climate induced changes in disease severity could be linked to the trophic lifestyle of the pathogens. Elevated [CO₂] decreased powdery mildew infection but had no effect on spot blotch disease compared to ambient condition. However, the effect of elevated [CO₂], [O₃] and temperature did not act in an additive manner when combined. This led to a surprising disease development in the combination treatments, where powdery mildew infection increased despite the individual reducing effect of the climatic factors, and spot blotch disease decreased despite the individual promoting effect of temperature and ozone, emphasizing the importance of conducting multifactorial experiments when evaluating the potential effects of climate change. Abundance of diseases were also evaluated under field conditions in a Free Air Carbon Enrichment facility and the enrich CO₂ atmosphere in the field decreased severity of powdery mildew supporting the phytotron results.

Climate change impacts on barley diseases in the Béja region of north-western Tunisia

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In a world where more than one billion people do not have enough to eat and our future food security is threatened by climate change and an ever-growing population, it is essential to improve the control of crop diseases around the globe. The weather plays a big role in the development of the disease on plant crops and increases the risk of severe epidemics. In fact, increased frequency of heat stress, droughts and floods negatively affect crop yields. Changes to any of these climatic factors may influence the distribution and biology of plant pathogens with positive, negative or neutral effects. Even if research in the effects of climate change on plant disease continues to be limited, some striking progress has been made in Tunisia. In fact, researches suggest that climate change will increase the risk of serious diseases on cereals in Northern Tunisia by the middle of this century where climate change models are predicting warmer, wetter winters for the country. The purpose of our study was to establish the influence of climate change on some barley diseases (Stripe and Scald) based on data gathered from field varieties/accessions trials conducted in the Béja region of north western Tunisia under rainfed conditions. Data were extracted for the years 2009 to 2013. Every year, there were seven local accessions, one introduced line and two standard varieties (the most cultivated in this region). The options other than varietals deployment are to identify and incorporate new genetic diversity in barley, produce stable resistant genetic stocks and eventually transfer these new genes into some leading, presently resistant or even susceptible varieties. Results showed that in this region, global minimum temperature and precipitation have increased and summers rains have become more frequent and stormy. Results revealed also that year by year, there is resurgence of Stripe and Scald barley diseases but intensity of pathogenicity depends upon the resistance level of the germplasm and the concerned year. Intensity of disease infection during the year 2011/2012 was higher as compared to the year of 2010/2011. This severity may be attributed to wet weather, which prevailed during this year. More rainfalls during 2011/2012 favoured the intensity of disease in almost materials. Plant material tested demonstrated that varieties/line/accessions are infected and most of them are susceptible but one autochthonous accession and introduced line are partially resistant and had a great potential to be used as a resistant germplasm source against Stripe. The degree of damage suffered by a crop will depend on the synchrony between pest abundance and the most susceptible developmental stage of the crop. For this purpose, Tunisian researchers must be prepared to future climate changes locally by creating varieties that are adapted to climatic zones and that have high resistance to extreme events. Process may be continued with more opportunities for new cultivars to be introduced, but effective disease screening systems must be in place to prevent pathogens from being introduced with these new lines. Impacts of a changing climate on plant disease intensity and yield loss can be combated by mixing several current recommended varieties of barley to partially control pathogens. With increased component number and trait diversity of barley varieties, infection may decrease.

Agro-meteorological evolution of barley leaf diseases in Tunisia

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The new Tunisia still remained welded to its marked by its openness to each other and the spirit of tolerance embedded in olive groves and fields of grain over a thousand years history. In Tunisia, mainly rain-fed cultivation of cereals is still subject to the challenge of climate, random and erratic governance of natural resources and confusion in the transfer of technologies. Despite the quality of genetic material developed during the last decade, new varieties of cereals (wheat and barley), remained subject to the challenge of diseases and pests. Data recorded in semi-arid western location of Boulifa, Kef during the last ten cropping seasons (2004 to 2013) were runned, based on cumulative frequencies, and biased and adjusted to climatic data (cumulative rainfall and temperatures). The hierarchical data analysis of the main foliar diseases of barley in semi-arid Tunisia (powdery mildew, net blotch and scald) showed that barley leaf diseases epidemics are still under-studied and not well reported regarding to climate change in the region. Evaluation of pests using hierarchical cumulative methods could be of interest to face climate changes and to brainstorm efficient strategies for insect and pest management.

Poster Abstracts

P1 Pathogenic diversity in *Pyrenophorateres f. teres* (net form net blotch of barley) populations from the Canadian Prairies.

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Cluster analysis using simple sequence repeat DNA markers was performed on a collection of 126 *Pyrenophorateres f. teres* (Ptt) isolates from the Canadian Prairies (Alberta, Saskatchewan and Manitoba). Subsequently, 40 isolates were selected as representatives of different clades to assess pathogenic variation among pathogen populations collected from this region. The isolates were evaluated against seedlings of nine barley differentials (two-rowed Norbert, TR 473, CI 5791, and CI 9820; and six-rowed Heartland, OAC21, Steptoe, BT201, and CI 9214) with known reactions to western Canadian Ptt isolates. One week following inoculation, the second and third leaves of each plant were rated according to the 1–10 scale developed by Tekauz (1985). Plants with scores of 1–5 and >5–10 were scored as resistant and susceptible, respectively. The entire experiment was repeated twice. When the results were inconsistent between the two experiments, inoculations were repeated as necessary to provide a consistent susceptible or resistant reaction. Data were then transformed into a 0-1 matrix with 0 as resistant and 1 as susceptible. Cluster analysis using the unweighted pair group method with arithmetic mean procedure and simple similarity coefficient revealed 16 pathotype groups among the 40 representative isolates. Two pathotype groups, which comprised 43% of the isolates, were found to be predominant on the Prairies. Moreover, relatively more aggressive pathotypes were found to form separate groups with one or two isolates. Variation in virulence ranged from that of an isolate (MB14) which was virulent on eight of the nine differentials with an average rating score of 6.5, to an avirulent isolate (MB11) with an average rating score of 2.8. Among the genotypes tested, BT 201 and OAC21 were the most susceptible with average rating scores of almost 8.0 and 7.5, respectively. By contrast, excluding isolate MB14, the differentials CI 5791 and CI 9820 were resistant to all other isolates with average rating scores of 2.6 and 2.7, respectively. Therefore, these differential lines and breeding material derived from them can be considered as potential sources of resistance to net form net blotch in barley breeding programs on the Canadian Prairies.

P2 Comparison of the effectiveness of two fungicides on the development of *P.teres* in Morocco

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The chemical treatment on the barley misses almost in Morocco. Thus, the results concerning the products used on wheat are useful to know in the case of possibility of the chemical treatment of the barley. In addition, the study was undertaken to evaluate the effectiveness of two chemicals to knowing the azoxystrobin pertaining to the family of strobilurines and the epoxiconazole gathered within triazoles, on the speed of mycelial growth of *P.teres* and the germination of the spores. The azoxystrobin proved very effective as well on the germination of the spores on the mycelial growth. Indeed, it generated the inhibition of the mycelial growth of *P.teres* to a concentration of 0,05 ppm. Also, it completely slowed down the mycelial growth of *P.teres* in general to concentrations of 5 ppm. As for the action on the germination of the spores, we noted easily that the germination of the spores of the isolate 22 took place after 24h incubation for concentrations 0,01; 0,02 and 0,04 ppm. For the epoxiconazole, its effectiveness marked on the mycelial growth was also noted but its effectiveness on the germination of the spores to amounts lower than 1 ppm did not give satisfaction.

P3 The *Rrs1* locus and resistance against scald in barley

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Scald, caused by *Rhynchosporium commune* (formerly *R. secalis*), is one of the most prevalent barley diseases worldwide. To date, four major scald resistance genes have been mapped in cultivated barley (*Hordeum vulgare* ssp. *vulgare*), and another four in wild barley (*Hv. spontaneum*) or *Hv. bulbosum*. The most abundant and effective one is the *Rrs1* resistance locus, formerly known as *Rh-Rh3Rh4* locus. It was mapped to the centromeric region of chromosome 3H. However, it is still not clear whether *Rrs1* is a collection of several *R*-genes close to each other or several alleles of the same gene. A search for new resistance sources revealed that Spanish landrace-derived lines SBCC145 and SBCC154 showed outstanding resistance to scald. To analyze the genetic basis in more detail two large DH mapping populations were developed crossing each donor line with cv. Beatrix. A large QTL in the centromeric region of chromosome 3H was found in both populations phenotyped for scald resistance in a well-established greenhouse test, therefore, confirmed this locus as the only resistance locus in both populations. To confirm and enclose this locus, the “*Rrs1* region” has been saturated with all available SSR and SNP-markers and a consensus map was constructed. New markers for this region are developed based on the Illumina iSelect custom 9K barley chip, the barley genome zipper and a BSA analysis with AFLP. The genome zipper identified several candidate genes. Because of a low gene/cM density, an enrichment of candidate sequences in the region of *Rrs1* is done by a BSTA (bulked segregant transcriptome analysis) with four normalized cDNA libraries and Illumina HiSeq in combination with a high resolution mapping program. For fine mapping and haplotyping of all the genes around the *Rrs1* loci a mapping population comprising >10,000 F₂ from the cross SBCC145 x Beatrix has been constructed. F₂ screening of about 11,000 lines to select recombinant lines between two flanking markers has identified around 442 verified recombinant plants. The effective *Rrs1* allele found and the closely linked markers developed are already useful tools for molecular breeding programs.

P4 Morphological and pathological variation detected among Turkish isolates of *Drechslera teres*

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Drechslera teres is an important pathogen affecting barley plants worldwide. The disease is also common in Turkey. We obtained 400 *Drechslera teres* isolates, both spot and net forms, from important barley growing areas of Turkey. Isolates differed in their growth rate, colony morphology and color in Potato Dextrose Agar. Isolates also differed in their pathogenicity. Some isolates obtained from Aegean region of Turkey were less virulent on differential cultivars and on susceptible Turkish cultivar Bülbül 89. Generally, Bülbül 89 was more susceptible to *Drechslera teres* isolates than the cultivars Hector and Lacey and could be used as the universal susceptible check. Some isolates were highly pathogenic. Four out of 16 *D. teres* f. *teres* isolates exhibited a moderately susceptible-moderately resistant reaction on the genotype CI 5791. Three out of 23 *D. teres* f. *maculata* isolates exhibited moderately susceptible to moderately susceptible-susceptible reaction on CI 5791. One *D. teres* f. *teres* isolate was virulent on differential host Tifang. Two *D. teres* f. *teres* isolates and two *D. teres* f. *maculata* isolates were virulent on genotype NDB112. It appears that pathogenic variation is high among the Turkish *D. teres* isolates.

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P5 Virulence spectrum of *Pyrenophorateres f. sp. maculata* in Morocco.

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Spot form of *P. teres* has become a major disease of barley in many regions worldwide and it was the most prevalent blight in Morocco in 2012. Thirty six isolates obtained from infected leaf tissue samples obtained from farm fields were inoculated on a set of nineteen barley genotypes. The studies were conducted at seedling stage, under controlled conditions and following standard procedures regarding inoculum concentration. The barley proposed differentials were scored for infection phenotype according to Tekauz's scale of 1-9 reaction scale; scores ≥ 5 were considered susceptible reactions, those < 5 resistant. In this preliminary work, considerable variability and high virulence appear to exist in the current Moroccan population of *P. t. f. sp. maculata*. Twelve isolates (33.33%) exhibited completely different virulence profiles and ten isolates (27.78%) differed only on the infection phenotype on or two barley differentials. The remaining 14 isolates (38.89%) induced a wide range of infection phenotypes being the susceptible interaction as the more frequent one. Further studies of additional isolates is needed to obtain more comprehensive information on the diversity of virulence, and validate the usefulness of the international differential set to characterize the Moroccan *P. t. f. sp. maculate* population. This information is essential to breed for varieties with a more durable resistance.

P6 Histochemical characterization of early response to *Cochliobolus sativus* infection in selected barley genotypes.

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Spot blotch is an important disease in barley, caused by *Cochliobolus sativus*. Many efforts are being made to breed barley with durable resistance to this disease. We hypothesized that resistance and susceptibility traits exhibited by diverse barley genotypes might have resulted from diverse pattern of defense responses operating at the cellular level. In this paper, we provided a quantitative description of the interaction microphenotypes of 11 barley genotypes infected with *C. sativus* by histochemical analysis of the oxidative responses occurring in epidermis and mesophyll leaf tissues during early stages of infection. Early defense responses including a build-up of cell wall appositions (CWA) in epidermal cells as well as hypersensitive response (HR) of epidermis/mesophyll tissues were observed in both resistant and susceptible genotypes. However, different frequencies for these responses were detected among and between resistant and susceptible genotypes. Moreover, unlike previously reported results on biotrophic fungi, we found that HR responses in epidermal cells occurring at post-penetration stages would be rather indicative of compatibility. This study revealed microphenotypic diversity in resistant and susceptible genotypes suggesting that diverse functional mechanisms of resistance and susceptibility might be present in barley.

P7 Identification and characterisation of effectors from barley pathogen *Rhynchosporium commune*

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Rhynchosporium commune is a hemibiotrophic haploid fungus causing scald, one of the most economically important and destructive diseases of barley (*Hordeum spp*). It can lead to yield losses and decrease in grain quality by infecting leaves, leaf sheaths and ears. *R. commune* infection can remain asymptomatic, with the disease threat hidden till conditions favour the pathogen. Agronomic practices in combination with chemicals like foliar fungicides can provide a high level of disease control but their continuous use induces pathogens to develop resistance to them. The most eco-friendly method of protecting barley from *Rhynchosporium* is the use of resistant cultivars. A good strategy to identify host resistance involves better understanding of effector proteins activating barley resistance. Only 3 *R. commune* effectors have been characterised so far. NIP1, NIP2 and NIP3 belong to a family of small secreted cysteine-rich necrosis-inducing proteins. NIP1 protein induces defence responses in barley cultivar carrying the resistance gene *Rrs1*. Furthermore, NIP1 was absent in 45% of isolates worldwide and several SNPs were detected in different strains of *R. commune*. Through this deletion or modification, the fungus avoided the recognition by the resistance gene preventing the disease control by *Rrs1*. Recent sequencing of *R. commune* genome allowed identification of the putative effectors. The aim of the project is to characterise *R. commune* effectors and use them for identification of novel sources of resistance to *R. commune*. Expression of 26 potential effectors with homologs in other fungi have been analysed during barley infection and allowed to prioritise 11 candidates. 2 promising candidates showed matches to known effectors, a protease inhibitor characterised in *Phytophthora infestans* and a secreted chorismate mutase, which might serve as a tool for host manipulation by plant-associated microbes. Selected candidates are being individually expressed in barley cultivars and landraces using BSMV to see if they are recognised by the plant. Selected genes will be deleted in the fungus to elucidate their role in pathogenesis. Tagging of *R. commune* effectors with a fluorescent protein will be used to follow their localisation *in planta*. Essential effectors will be used to identify their virulence targets and barley resistance genes.

P8 Distribution of barley stripe disease in Central Anatolia, Turkey

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Barley stripe disease caused by *Drechslera graminea* is an important disease of barley worldwide. The use of clean seed and seed treatment fungicides effectively control this disease. However, the disease can be a threat if clean seed is not used or no seed treatment is employed. During 2012, 205 barley fields were surveyed for the presence of this disease in Central Anatolia. Eighty-two fields (40 %) were found to be infected with *Drechslera graminea*. However, the disease incidence was low ranging between 1-70 %, usually less than 10%. Large differences were observed among provinces for the proportion of crops infected.

P9 Field evaluation of some Turkish barley landraces to scald and net blotch of barley

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Barley landraces are good source of disease resistance. Two hundred barley landraces were evaluated under field conditions to scald and net blotch diseases of barley in 2013. No artificial inoculations were employed. The incidence values (number of plants infected of those examined) of scald and net blotch were between 0-90% and 0-30%, respectively. Scald incidence was high. Only five genotypes, 4 six-rowed and 1 two-rowed barley, were scald free. Net blotch was observed on 44 genotypes. Net blotch was not observed on the remaining 156 genotypes. A 1-9 scale was used to assess the disease severity. Disease severity values of *Rhynchosporium secalis* and *Drechslera teres* ranged between 1-9 and 1-7, respectively. Genotypes with no diseases could be used in breeding programs for resistance to scald and net blotch.

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P10 Evaluation of leaf stripe disease severity and reaction type in landraces and cultivars of barley (*Hordeum vulgare* L.) from Turkey

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Disease severity and reaction type of 20 landraces and three cultivars of barley to leaf stripe disease were evaluated in greenhouse conditions. Ten *Drechslera graminea* isolates collected in ten locations of Turkey were used for inoculation using the “sandwich method”. Phenotypic variation to leaf stripe disease was observed in the responses of landraces and cultivars of barley with the same and different strains of the fungus. Barley landraces #3 and #5 exhibited resistance and susceptibility to eight isolates of the fungus, respectively. Barley cultivar Çumra 2001 showed a resistant reaction to all isolates. Cultivars Atılır and Larende were susceptible to 9 isolates. The *D. graminea* Konya (Bozkır) isolate was the most virulent while Ankara (Haymana) isolate was the least virulent. This research shows that barley landraces and cultivars could be a rich source of variability against current isolates of *D. graminea* found in barley production areas of Turkey.

P11 New sources of resistance to *Pyrenophora teres* f. *maculata* in barley

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Six barley lines with resistance toward the Australian pathogen population of *Pyrenophora teres* f. *maculata*, the cause of spot form of net blotch (SFNB), have been identified. These include the two row barley types BYDV-018, Dairokkaku, Esperance Orge 289, MXB.468 and Yangsimai 3 and the six row type CI5286. These barley lines were found to be resistant as seedlings and adults in Australia and as adults in Canada and South Africa, indicating that they may be useful internationally. These lines are currently being used in the development of mapping populations and genetic characterisation in collaboration with the Australian Wheat and Barley Molecular Marker Program. These lines are also being tested alongside the international SFNB differential set at various locations in Australia, Canada and Finland to monitor in virulence in the *P. teres* f. *maculata* population internationally.

P12 Occurrence of toxin-producing fungi in barley and other small grain cereals

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Climate change accompanied by global warming affects food safety at different levels. Higher temperatures, elevated humidity or drought will increase the infection of crops by different fungi and therefore increase the probability of mycotoxin occurrence. Fungi have optimum temperature ranges within they can infect agricultural crops more severely. Increasing average temperatures could lead to changes in the range of latitudes at which certain fungi are able to compete. A shift has recently been observed in the occurrence of aflatoxin producers in Europe, with consequent aflatoxin contamination in agricultural commodities in several European countries not facing with this problem before. Although aflatoxin contamination of agricultural products is not treated as a serious threat to Hungarian agriculture due to climatic conditions, these observations led us to examine the mycobiota of different cereals including barley, oat, triticale and rye collected from a south-east location in Hungary. The occurrence of *Aspergillus*, *Penicillium* and *Fusarium* species were investigated after harvest in three consecutive years. Surface-sterilized cereal seeds were placed on selective media, and the isolated fungal strains were identified using morphological methods. Identification of selected isolates was carried out using sequence-based methods. In 2011, 1% of the barley samples, 4% of the triticale and rye samples were found to be contaminated with potentially aflatoxigenic *Aspergillus flavus* isolates. Besides *A. flavus* isolates, other mycotoxin producing species were also isolated. In 2011 *Aspergillus niger*, which potentially produce ochratoxins and fumonisins, and in 2013 *A. ochraceus*, which produces ochratoxins were detected. Regarding *Fusarium* species, in 2011 *F. graminearum* dominated, while in 2013 *F. verticillioides* was the predominant species identified. In 2012 a large number of *Alternaria* species occurred in the samples and we could not detect any *Aspergillus* or *Penicillium* contamination. Other genera (*Nigrospora*, *Epicoccum*, *Cladosporium*) were found in smaller proportions only.

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P13 Control of leaf rust in barley – fungicide or resistance

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Leaf rust (*Puccinia hordei*) is a major disease of barley (*Hordeum vulgare*) world wide. In north-eastern Australia epidemics are infrequent; yet can be severe. In 2010, a combination of over-seasoning inoculum, an extended sowing window, widespread sowings of the very susceptible variety Grout and a particularly favourable environment resulted in severe losses in yield and quality in many crops. In a number of cases the application of fungicide failed to arrest the epidemic. A trial was conducted to examine the value of different levels of resistance in reducing yield losses to leaf rust and the application of fungicide for disease control. Under a late but heavy epidemic, a moderately resistant variety incurred only a 6% reduction in yield while the very susceptible variety lost 32%. The application of fungicide to the moderately resistant cultivar was not economical; however with the very susceptible cultivar, the best fungicide treatment gave a net return of over \$400/ha.

P14 Tea Tree Oil for the powdery mildew control in barley

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In recent years, the use of chemicals is becoming more restrictive and alternative "green" measures have been proposed for crop protection, including mineral salts, biological agents and plant extracts. Among these last natural products, Tea Tree Oil (TTO) is an essential oil consistently present in leaves and terminal branches of *Melaleuca alternifolia*, a tree belonging to *Myrtaceae* family that occurs in Australia, on the north coast and adjacent ranges of New South Wales. TTO has a long history of use as topical microbicide in human pharmacology and is now used in wide range of applications, that include cosmetics, toiletries, house hold products and veterinary/pet care. In agriculture, TTO and its component, terpinen-4-ol has been demonstrated to have contact and fumigant insecticidal action against economically important pests (Isman, 2000). Moreover, TTO and its components have been demonstrated to inhibit the growth of cereal pathogens, like *Aspergillus*, *Fusaria*, *Pyrenophora* (Terzi et al, 2007; Morcia et al, 2011; Morcia et al, 2012).

In this work the effects of Tea Tree Oil has been evaluated for the control of powdery mildew infection in barley. The following activities have been carried out:

- *in vitro* evaluation of antifungal properties of TTO against *Blumeria graminis*;
- *in vivo* validation of the results obtained through greenhouse and open field trials;
- whole transcriptome characterization of the plant infected with *Blumeria* and TTO treated in comparison with the controls.

The rationale behind this approach is that a better knowledge of the molecular targets and mechanisms of actions of plant metabolites at cellular level can optimize their utilization, alone or in combination, and maximize the antifungal efficacy.

P15 A genome-wide survey of leaf stripe resistance in a low-structured barley association panel

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In barley (*Hordeum vulgare*), the application of marker platforms that provide dense genome-wide coverage of molecular polymorphism allows to elucidate the evolutionary history of natural populations and use their biodiversity to dissect traits of agronomic interest, through genome-wide association scan (GWAS) approaches. Here we describe the evaluation of a low-structured collection of ~210 spring 2-rowed European barley cultivars for their resistance to leaf stripe, a seed-borne disease caused by the fungal pathogen *Pyrenophora graminea*. For each line, sixty seeds have been surface-sterilized and incubated in Petri dishes between two Potato Dextrose Agar layers colonized by an actively growing mycelium of the *P. graminea* isolate Dg5. After 20 days of incubation in the dark at 6 °C, the emerged seedlings have been transplanted to pots and grown in the greenhouse (20°C, 14 h light and 12°C, 10 h night). Resistance has been assessed as the percentage of plants showing leaf stripes symptoms, and the whole experiment has been repeated during three consecutive years. The same barley collection has been genotyped with a novel set of 7864 gene-based SNPs incorporated into a single Illumina Infinium™ iSelect assay, in order to investigate: i) trends in the patterning of genetic diversity in European spring barley cultivars in time and space; ii) the utility of a low-structured population for discovering significant associations between genetically mapped markers and important agronomic traits. SNP markers mapping on the short arm of barley chromosome 6H (8.6 – 13.6 cM) showed a significant association with leaf stripe resistance. This genomic region is syntenic with a ~240 Kb of rice chromosome 2, where 42 genes were annotated, that could serve for the identification of candidate genes involved in barley resistance to pathogens or for the development of new SNP markers, in order to increase the resolution of the GWAS.

P16 The impact of fungicide and herbicide timing on barley leaf disease severity, weed management and crop productivity

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Over the last decade interest in tank mixing herbicides and half rate fungicides for malt barley has been increasing in western Canada. At six sites located across the western Canadian prairies combinations of the herbicide Axial[®] (pinoxaden)/Frontline[®] (florasulam) and the fungicide Tilt[®] (propiconazole) were applied to barley at the 2-3 leaf stage (herbicide and half rate fungicide), 5-6 leaf stage (herbicide and half rate fungicide), and/or the flag leaf stage (full or half rate fungicide only). Prior to seeding plot areas, each site was cross-seeded with tame oat as a model weed. At approximately early dough, penultimate and flag leaf – 2 samples were collected for assessment of leaf disease, while later season model weed biomass were assessed. Plots were harvested and grain yield and kernel quality assessed. A mixed model analysis was conducted over all years and sites with site-year and rep as random variables. Overall, total leaf disease severity was higher for the 2-3 or 5-6 leaf stage no fungicide-herbicide only treatments and the combination herbicide and half rate fungicide treatments compared to treatments with fungicide applied at the flag leaf stage. Yield, thousand kernel weight, plumpness and test weight tended to be highest and percentage thins were lowest for those treatments with a flag leaf stage fungicide application. Split applications of fungicide at herbicide timing and the flag leaf emergence stage generally did not improve disease management and crop productivity compared to a single full rate fungicide application at the flag leaf stage. Model weed biomass was very low and generally not influenced by the treatments due to effective herbicide applications at each of the sites. However, yield tended to be lower when herbicide was applied at the 5-6 versus 2-3 leaf stage. Overall results from 2010 to 2012 indicate for malt barley, that fungicide application should include a flag leaf stage timing for adequate protection of upper canopy leaves, thus contributing to enhanced yield and grain filling. In addition, delaying herbicide application to the 5-6 leaf stage may contribute to a reduction in yield due to early season weed competition for water and nutrients.

P17 The impact of barley variety monoculture, rotation and mixtures, and intercropping on leaf disease and silage production

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At two locations in Alberta, Canada the impact of barley variety monocultures and rotation, barley variety mixtures, and intercropping of barley, oat and triticale on leaf disease and silage productivity. A three year rotational sequence was established in 2008 with comparisons made during the third year of the sequence in 2010 and 2013. Treatments included: continuous barley, same variety; a mixture of the same three barley varieties each year; a mixture of three different barley varieties each year; an intercrop of barley, oat, and spring triticale with the same or different crop varieties each year; and an intercrop of barley, oat, and winter triticale with the same or different crop varieties each year. In the third year of the rotational sequence, all treatments had the barley variety Sundre. At Lacombe in 2010, leaf disease on Sundre (primarily net-form net blotch) was highest for continuous Sundre, slightly lower, but similar for the barley variety rotation, and mixtures and intercrops with the same three varieties each year, and lowest for mixtures or intercrops with different varieties each year. Continuous Sundre at Lethbridge in 2010 had the most severe net blotch, with barley variety rotation having intermediate disease levels, while the remaining treatments had lower, but similar levels of disease. In 2013 at Lacombe, net blotch levels were high for most treatments, but tended to be highest for the continuous Sundre and barley variety rotation treatments and lowest for the variety mixture treatments, while the remaining treatments had intermediate disease levels. At Lethbridge in 2013, continuous Sundre had the highest disease, with the remaining treatments having lower, but generally similar levels of disease. At Lacombe in 2010 and 2013, silage yields on a wet or dry weight basis were lowest for the continuous Sundre, highest for the intercropping treatments with the same or different varieties, and intermediate for the barley mixtures. At Lethbridge in 2010 and 2013, the continuous Sundre treatment tended to have the lowest silage yield, although the intercrop treatments with winter triticale also had lower yields, especially on dry weight basis. Barley variety mixtures and intercropping with spring triticale tended to have higher, but similar yields. Results indicate that adding diversity in crop types and barley genetics may reduce leaf disease and improve silage productivity.

P18 Identification of QTL for resistance to spot blotch caused by *Bipolaris sorokiniana* in Kazakhstan

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Spot blotch caused by *Bipolaris sorokiniana* is widespread disease of barley in Kazakhstan. It was reported that the disease causing losses of grain yield up to 20-30% (Rsaliev, 2012). Therefore, identification of the genetic factors related to resistance of barley to *B. sorokiniana* is very important for local breeding programs. The collection of spring barley consisted from 96 cultivars and perspective lines from Kazakhstan and was genotyped by 384 SNP markers based on Illumina Golden Gate array. The collection was grown in the field (2013), which was infected by local *B. sorokiniana* pathotypes population. Plants were inoculated at tillering stage and resistance was estimated at heading and seed maturation stages. The results of plant resistance were used for the association mapping analysis by using TASSEL 2.0 version and MLM method. Genetic analysis is allowed to identify two highly significant QTLs located on chromosomes 2H (62.8 cM) and 7H (129.9 cM). The results can be used in barley breeding projects based on marker-assisted selection.

P19 Resistance to stem rust in spring barley from Kazakhstan

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Stem rust caused by *Puccinia graminis* f. sp. *tritici* is one of the most harmful diseases of barley in Kazakhstan. It is estimated that in certain years it downgrades grain yield up to 40% (Rsaliev, 2012). Therefore, search for resistant forms of cultivated barley to *P. graminis* is very important for local breeding programs. Ninety six two-rowed spring barley cultivars and perspective lines of Kazakhstan were grown in five breeding stations across the country and in one controlled artificially environment, which was infected by local *P. graminis* pathotypes population. The accessions were grown in triplicate randomized blocks in all testing sites and studied for grain yield, yield components and disease resistance to stem rust. In controlled infected environment all accessions were inoculated at tillering stage and resistance was estimated at heading and seed maturation stages. The results suggested that at seed maturation stage 31 were resistant, 28 were moderately resistance and 37 were susceptible accessions. In addition all 96 accessions were tested for genetic variation of *Rpg1* gene conferring resistance to stem rust (Brueggeman et al., 2002). It is identified that seven cultivars and one perspective line were carrying resistant allele of *Rpg1*. Statistical analysis (*t-test*) is suggesting that grain yield in those 8 identified accessions was higher than in other accessions ($P < 0.05$) grown in most of natural environments of 5 breeding stations of Kazakhstan in 2009-2013. The result may indicate that *Rpg1* can be also a contributing factor to better grain yield in various barley growing regions of Kazakhstan.

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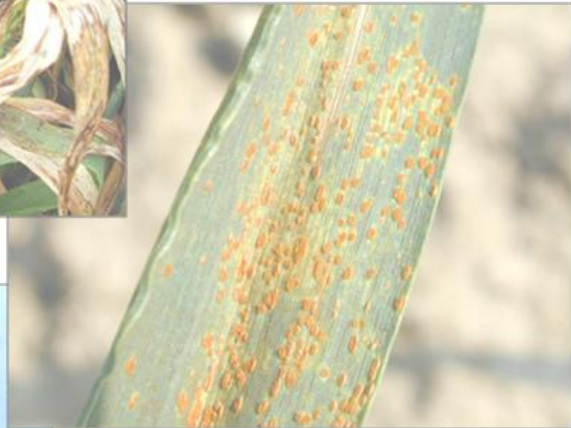
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Notes

Notes

See you at the next International Workshop on Barley Leaf Diseases

Michele, Valeria, Alessandro



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