

12<sup>th</sup> INTERNATIONAL  
CEREAL RUSTS and  
POWDERY MILDEWS  
CONFERENCE

Abstract Book

October 13-16, 2009  
Antalya – Turkey



# ICRPMC 2009 SCIENTIFIC PROGRAM

| <b>Monday 12<sup>th</sup> October</b>        |   |
|--|---|
| 4:30pm – 7:30pm                              | Registration  |
| 8:00pm – 9:00pm                              | Board meeting of the Cereal Rusts Foundation (private)  |
| <b>Tuesday 13<sup>th</sup> October</b>       |   |
| 8:00am – 9:00am                              | Registration  |
| Chair: <b>James Brown</b>                    |   |
| 9:00am – 9:10am                              | <b>Mahinur S. Akkaya</b> (Middle East Technical University, Turkey)<br>and <b>James Brown</b> (on behalf of the Scientific Programme Committee)<br><i>Welcome</i>   |
| 9:10am – 9:45am                              | Plenary: <b>Ravi Singh</b> (CIMMYT, Mexico)<br><i>Building international cooperation: Crucial to mitigate the threat from Ug99 race of stem rust and other rust pathogens</i>   |
| Offered papers: <b>Wheat Stem Rust</b>       |   |
| 09:45am – 10:00am                            | <b>Davinder Singh</b> (CIMMYT, Kenya)<br><i>Stem rust screening and breeding for Ug99 resistance in East Africa</i>   |
| 10:00am – 10:15am                            | <b>Zafer Mert</b> (Central Research Institute for Field Crops, Turkey)<br><i>Global initiatives for management of Ug99 stem rust race and reactions of some winter wheat genotypes to Ug99 in 2008</i>  |
| 10:15am – 10:30am                            | <b>Matthew Rouse</b> (University of Minnesota, USA)<br><i>Aggressiveness of races TTKSK and QFCSC of Puccinia graminis f.sp. tritici at various temperatures</i>  |
| 10:30am – 10:45am                            | <b>Cornel Bender</b> (University of the Free State, South Africa)<br><i>Development of a greenhouse screening method for adult plant response in wheat to stem rust</i>   |
| 10:45am – 11:15am                            | COFFEE BREAK  |
| Chair: <b>Robert Park</b>                    |   |
| 11:15am – 11:50am                            | Plenary: <b>Mogens Hovmøller</b> (University of Aarhus, Denmark)<br><i>Rates of evolution in Puccinia striiformis</i>   |
| Offered papers: <b>Yellow Rust Pathogens</b> |   |
| 11:50am – 12:05pm                            | <b>Eugene Milus</b> (University of Arkansas, USA)<br><i>Comparison of old and new strains of Puccinia striiformis f.sp. tritici for ability to initiate epidemics from overwintering infections and for ability to infect at high temperature</i> |
| 12:05pm – 12:20pm                            | <b>Alexander Loladze</b> (University of Sydney, Australia)<br><i>Differential adaptation of Australian and New Zealand stripe rust isolates to high temperature</i>   |
| 12:20pm – 12:35pm                            | <b>Claude de Vallavieille-Pope</b> (INRA UMR BIOGER-CPP, France)<br><i>Adaptation of the clonal pathogen Puccinia striiformis f.sp. tritici to temperature and resistance genes in North-West Europe and Mediterranean area</i>                   |
| 12:35pm – 12:50pm                            | <b>Colin Wellings</b> (NSW Department of Primary Industries, Australia)<br><i>Potential vulnerability of barley to an undescribed form of Puccinia striiformis in Australia</i>   |
| 12:50pm – 2:00pm                             | LUNCH   |

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|---|---|
| Chair: <b>Colin Wellings</b>                    |   |
| 2:00pm – 2:35pm                                 | Plenary: <b>Wanquan Chen</b> (Institute of Plant Protection, China)<br><i>Regional epidemics and management of wheat stripe rust in China</i>   |
| Offered papers: <b>Rust Pathogen Variation</b>  |   |
| 2:35pm – 2:50pm                                 | <b>Amor Yahyaoui</b> (ICARDA, Syria)<br><i>Wheat yellow rust (<i>Puccinia graminis f.sp. tritici</i>) in Central West Asia and North Africa</i>   |
| 2:50pm – 3:05pm                                 | <b>Xianming Chen</b> (USDA-ARS and Washington State University, USA)<br><i>Race changes of <i>Puccinia striiformis f.sp. tritici</i> and <i>Puccinia striiformis f.sp. hordei</i> in the United States</i>  |
| 3:05pm – 3:20pm                                 | <b>Johanna E. Snyman</b> (University of Sydney, Australia)<br><i>A new pathotype of stripe rust affecting triticale in Australia</i>  |
| 3:20pm – 3:35pm                                 | <b>Julio Huerta-Espino</b> (INIFAP-CEVAMEX, Mexico)<br><i>Phenotypic variation among leaf rust isolates from durum wheat in northwestern Mexico</i>   |
| 3:35pm – 3:50pm                                 | <b>Jochen Prochnow</b> (BASF SE)<br><i>Wheat brown (leaf) rust in Europe: Studies on disease sensitivity towards azoles and strobilurins and their fungicidal efficacy</i>  |
| 3:50pm – 4:20pm                                 | COFFEE BREAK  |
| Chair: <b>Claude de Vallavieille-Pope</b>       |   |
| 4:20pm – 4:55pm                                 | Plenary: <b>Robert Park</b> (Plant Breeding Institute, University of Sydney, Australia)<br><i>Achieving durable rust resistance in agriculture: from gene to continent and beyond</i>   |
| Offered papers: <b>Rust Resistance Genetics</b> |   |
| 4:55pm – 5:10pm                                 | <b>Tom Fetch</b> (Agriculture & Agri-Food Canada, Canada)<br><i>Inheritance of resistance to Ug99 in wheat line Tr129 with an introgression of <i>Aegilops triuncialis</i> chromatin</i>  |
| 5:10pm – 5:25pm                                 | <b>Colin Hiebert</b> (Agriculture and Agri-Food Canada, Canada)<br><i>Genetics and mapping of stem rust resistance genes conferring resistance to race Ug99 (TTKSK) in the wheat cultivars Webster, Peace and AC Cadillac</i>   |
| 5:25pm – 5:40pm                                 | <b>Mike Bonman</b> (USDA-ARS, USA)<br><i>Wheat landraces from the USDA-ARS National Small Grains Collection with resistance to new races of <i>Puccinia graminis f.sp. tritici</i></i>  |
| 5:40pm – 5:55pm                                 | <b>Roger Wise</b> (Iowa State University, USA)<br><i>Genetical genomics of stem rust infection identifies master regulators of defense in barley</i>  |
| 5:55pm – 6:10pm                                 | <b>Henriette Goyeau</b> (INRA UMR BIOGER-CPP, France)<br><i>Efficiency of specific and partial resistance to wheat leaf rust in bread wheat cultivars grown in France</i>   |
| 6:10pm – 6:25pm                                 | <b>Javier Sánchez-Martín</b> (CSIC Institute of Sustainable Agriculture, Spain)<br><i>Screening and characterization of resistance to crown rust (<i>Puccinia coronata f.sp. avenae</i>) and powdery mildew (<i>Blumeria graminis f.sp. avenae</i>) in an oat germoplasm collection</i> |
| 6:30pm – 8:30pm                                 | WELCOME RECEPTION & POSTER PRESENTATIONS  |
| 8:30pm – 10:00pm                                | DINNER (at your hotel)  |

## Wednesday 14<sup>th</sup> October

Chair: **Ralph Hückelhoven**

9:00am – 9:35am Plenary: **Roger Wise** (USDA-ARS, Iowa State University)  
*Regulation of innate immunity in barley-powdery mildew interactions*

Offered papers: **Non-Host and Basal Resistance**

9:35am – 9:50am **Rients Niks** (Wageningen University, Netherlands)  
*The barley-rusts and mildews: Two models to study the molecular basis of host-status of plants to specialized pathogens*

9:50am – 10:05am **Thierry Marcel** (Wageningen University, Netherlands)  
*Efficient targeting of barley genes for basal resistance to *Puccinia hordei**

10:05am – 10:20am **Hossein Jafary** (Zanjan Agricultural & Natural Resource Research Center, Iran)  
*Non-host immunity in barley to *Puccinia hordei-bulbosi* is a polygenic trait with multiple components*

10:20am – 10:35am **Reza Aghnoum** (Wageningen University, Netherlands)  
*SusBgt: Experimental barley lines with susceptibility to wheat powdery mildew as a tool to study non-host resistance*

10:35am – 11:05am COFFEE BREAK

Chair: **Soledad Sacristán Benayas**

11:05am – 11:40am Plenary: **Ralph Panstruga** (Max-Planck Institute for Plant Breeding Research, Germany)  
*The molecular basis of broad-spectrum powdery mildew resistance*

Offered papers: **Molecular Biology and Physiology of Rusts**

11:40am – 11:55am **Mahinur S. Akkaya** (Middle East Technical University, Turkey)  
*Antagonistic behavior of the two players of ubiquitinylation process in disease resistance*

11:55am – 12:10pm **Özge Karakaş** (Istanbul University, Turkey)  
*EST-Based multiplex gene expression in yellow rust infected wheat using GenomeLab GeXP genetic analysis system*

12:10pm – 12:25pm **Javier Sánchez-Martín** (CSIC Institute of Sustainable Agriculture, Spain)  
*Effect of different resistance mechanisms to crown rust (*Puccinia coronata* f.sp. *avenae*) and powdery mildew (*Blumeria graminis* f.sp. *avenae*) on oat stomatal conductance*

12:25pm – 12:40pm **Nicholas Lauter** (Iowa State University, USA)  
*Mlsp confers semi-dominant, developmentally-dependent resistance to barley powdery mildew*

12:40pm – 2:00pm LUNCH

Chair: **Mogens Hovmøller**

2:00pm – 2:35pm Plenary: **Jérôme Enjalbert** (INRA UMR BIOGER-CPP and UMR Plant Genetics, France)  
*Clonality and recombination footprints in wheat yellow rust genetic structure*

Offered papers: **Yellow Rust Genetics**

2:35pm – 2:50pm **Donal O'Sullivan** (National Institute of Agricultural Botany, England)  
*Dissecting the contributions of specific and partial resistance to yellow rust in UK wheat germplasm*

2:50pm – 3:05pm **Eugene Milus** (University of Arkansas, USA)  
*Characterization of adult-plant resistance in soft red winter wheat to stripe rust*

3:05pm – 3:20pm **Xianming Chen** (USDA-ARS and Washington State University, USA)  
*Molecular mapping of a new gene for resistance to stripe rust in durum wheat PI 480148 and transfer the gene into common wheat*

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|-------------------------------------|--|
| 3:20pm – 3:35pm                     | <b>Ahu Uncuoğlu</b> (TUBITAK, Turkey)<br><i>Identification and potential use of DNA markers for yellow rust disease resistance in wheat (Triticum aestivum)</i>                        |
| 3:35pm – 3:50pm                     | <b>Zhensheng Kang</b> (Northwest A&F University, China)<br><i>Molecular mechanism of wheat and stripe rust interaction and functional characterization of resistance-related genes</i> |
| 3:50pm – 4:50pm                     | COFFEE BREAK + POSTER PRESENTATIONS  |
| Chair: <b>Xianming Chen</b>         |  |
| Offered papers: <b>Epidemiology</b> |  |
| 4:50pm – 5:05pm                     | <b>David Hodson</b> (FAO, ITALY)<br><i>Global Cereal Rust Monitoring System – prospects and progress</i>   |
| 5:05pm – 5:20pm                     | <b>Colin Wellings</b> (NSW Department of Primary Industries, Australia)<br><i>Stripe rust epidemics in Australia: Implementing national integrated disease control strategies</i>      |
| 5:20pm – 5:35pm                     | <b>Amarilis Barcellos</b> (Universidade de Passo Fundo, Brazil)<br><i>Sensitivity of Puccinia triticina races to fungicides</i>  |
| 5:35pm – 5:50pm                     | <b>M. Nabil A. Omar</b> (Soils, Water and Environment Research Institute, Egypt)<br><i>Biological control of wheat leaf rust using Pseudomonas fluorescens and Bacillus spp</i>        |
| 5:50pm – 6:05pm                     | <b>Evsey Kosman</b> (Tel-Aviv University, Israel)<br><i>New tools for comprehensive evaluation of virulence and resistance data</i>  |
| 6:05pm – 6:20pm                     | <b>Yehoshua Anikster</b> (Institute for Cereal Crops Improvement, Israel)<br><i>Estimation of nuclear DNA and other methods for identification of wheat leaf rusts</i>                 |
| 7:30pm-11:00pm                      | GALA DINNER  |

## Thursday 15<sup>th</sup> October

Chair: **Ralph Panstruga**

9:00am – 9:35am Plenary: **Beat Keller** (University of Zurich, Switzerland)  
*Molecular basis of durable rust resistance in wheat*

Offered papers: **Lr34 and Lr46**

9:35am – 9:50am **Brent McCallum** (Agriculture and Agri-Food Canada)  
*Leaf tip necrosis co-segregates with seedling leaf rust resistance conditioned by Lr34 at low temperatures*

9:50am – 10:05am **Liselotte Setler** (University of Zürich, Switzerland)  
*Functional characterization of the durable disease resistance gene Lr34*

10:05am – 10:20am **Hassan Soltanloo** (Gorgan University of Agricultural Sciences & Natural Resources, Iran)  
*Identification and characterization of expressed sequences involved in differential host response of wheat carrying the Lr34/Yr18 genes challenged with Puccinia recondita using cDNA-AFLP*

10:20am – 10:35am **Noorali Abad Dadkhodaie** (Shiraz University, Iran)  
*Studies of the interactive effects of Lr16 in combinations involving Lr13 and/or Lr34 and its effectiveness against Australian isolates of Puccinia triticina*

10:35am – 11:25am COFFEE BREAK + POSTER PRESENTATIONS

Chair: **Roger Wise**

11:25am – 12:00 Plenary: **Tzion Fahima** (University of Haifa, Israel)  
*Wild emmer wheat as a source for disease resistance genes: from genetic diversity to gene cloning*

Offered papers: **Genetics of Rust Resistance**

12:00 – 12:15pm **Hanan Sela** (University of Haifa, Israel)  
*Intra specific ancient diversity of splicing motifs and protein surface at the wild wheat Lr10 CC and LRR domains*

12:15pm – 12:30pm **Freddy Yeo Kuok San** (Wageningen University, Netherlands)  
*In the neighborhood of resistance to Puccinia hordei QTL2*

12:30pm – 12:45pm **Silvia Barcellos Rosa** (Cereal Research Center, Canada)  
*Race non-specific adult plant gene involved with the quantitative resistance of Toropi*

12:45pm – 1:40pm LUNCH

2:00pm – 7:00pm EXCURSION: Aspendos & Side

7:00pm – 8:00pm TRAVEL TO KONYAALTI BEACH PARK – (Bars & Coffees)

8:10pm – 11:00pm Free time at Konyaalti Beach Park

## Friday 16<sup>th</sup> October

Chair: **Rients Niks**

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|-----------------|---|
| 9:00am – 9:35am | Plenary: <b>Patrick Schweizer</b> (Leibniz Institute of Plant Genetics and Crop Plant Research, Germany)<br><i>Convergent information for genes underlying quantitative powdery mildew resistance in barley</i> |
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Offered papers: **Molecular Biology of Mildew Resistance**

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|-------------------|---|
| 9:35am – 9:50am   | <b>Ralph Hückelhoven</b> (Technical University of Munich, Germany)<br><i>RAC/ROP binding proteins of barley influence the interaction with <i>Blumeria graminis</i> f.sp. hordei</i>  |
| 9:50am – 10:05am  | <b>Ruth Eichmann</b> (Technical University of Munich, Germany)<br><i>BAX INHIBITOR-1-like cell death inhibitor proteins of plants regulate interactions of barley and <i>Arabidopsis thaliana</i> with powdery mildew fungi</i> |
| 10:05am – 10:40am | Plenary: <b>Pietro Spanu</b> (Imperial College, UK and the BluGen consortium)<br><i>The genome of <i>Blumeria graminis</i> f.sp. hordei</i>   |
| 10:40am – 11:10am | COFFEE BREAK  |

Chair: **Jerome Enjalbert**

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| 11:10am – 11:45am | Plenary: <b>Soledad Sacristán Benayas</b> (Universidad Politécnica de Madrid, Spain)<br><i>Virulence effectors and retrotransposons in <i>Blumeria graminis</i></i> |
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Offered papers: **Pathogen Genomes**

|                   |  |
|-------------------|--|
| 11:45am – 12:00   | <b>Simone Oberhaensli</b> (Institute of Plant Biology, Switzerland)<br><i>Brothers in arms: Two cereal powdery mildews in the spotlight</i>  |
| 12:00 – 12:15pm   | <b>Francis Parlange</b> (University of Zurich, Switzerland)<br><i>Map-based cloning of the avirulence gene <i>Avrpm3</i> in the wheat powdery mildew <i>Blumeria graminis</i> f.sp. tritici</i>          |
| 12:15pm – 12:30pm | <b>Roi Ben David</b> (University of Haifa, Israel)<br><i>Asymmetric reciprocal virulence pattern among <i>Blumeria graminis</i> isolates originating from domesticated wheat and its wild progenitor</i> |
| 12:30pm – 12:45pm | <b>James Brown</b> (John Innes Centre, England)<br><i>Molecular analysis of the resistance of powdery mildew fungi to triazole fungicides conferred by mutations in <i>CYP51</i></i>                     |
| 12:45pm – 2:00pm  | LUNCH  |

Chair: **Amos Dinooor**

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| 2:00pm – 2:15pm | BUSINESS MEETING |
|-----------------|------------------|

Chair: **Brent McCallum**

Offered papers: **Pathogen Proteomics**

|                 |   |
|-----------------|---|
| 2:15pm – 2:30pm | <b>Christof Rampitsch</b> (Agriculture & Agri-Food Canada, Canada)<br><i>A protein profile from in planta infection structures of the wheat leaf rust fungus, <i>Puccinia triticina</i></i> |
| 2:30pm – 2:45pm | <b>Nese Ozgazi</b> (Middle East Technical University, Turkey)<br><i>Proteome analysis of differentially expressed proteins of barley by virulent and avirulent infections</i>               |

| Offered papers: <b>Breeding for Disease Resistance</b> |   |
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| 2:45pm – 3:00pm  | <b>Xiayu Duan</b> (Institute of Plant Protection, China)<br><i>Microsatellite mapping of the powdery mildew resistance gene in two Chinese landraces of wheat (<i>Triticum aestivum</i>) Mazhamai and Xiaobaidong</i> |
| 3:00pm – 3:15pm  | <b>Laszlo Purnhauser</b> (Cereal Research Non-Profit Company, Hungary)<br><i>Detection of resistance genes by molecular markers in wheat cultivars registered in Hungary</i>  |
| 3:15pm – 3:30pm  | <b>Vladimir Shamanin</b> (Omsk State Agricultural University, Russia)<br><i>Spring wheat breeding in western Siberia for resistance to leaf and stem rust</i>   |
| 3:30pm – 3:45pm  | <b>New chairman of Cereal Rust Foundation</b><br><i>Close of conference</i>   |

# LIST OF POSTER PRESENTATIONS

## Rust Pathogen Variation

- P1. Alena Hanzalová** (Research Institute of Crop Production, Czech Republic)  
*Physiologic specialization of wheat leaf rust (*Puccinia triticina* Eriks.) in the Czech Republic in 2006-2008*
- P2. Anna Berlin** (Swedish University of Agricultural Sciences, Uppsala, Sweden)  
*Population structure of *Puccinia graminis* f.sp. *avenae* in Sweden*
- P3. Annemarie F. Justesen** (Aarhus University, Denmark)  
*AFLP marker conversion into single locus markers in *Puccinia striiformis* f.sp. *tritici**
- P4. Berit Samils** (Swedish University Of Agricultural Sciences (Slu), Sweden)  
*Evidence of asexual overwintering of the willow leaf rust fungus *melampsora Larici-Epitea**
- P5. Elena Gulyaeva** (All-Russian Institute for Plant Protection (VIZR), Russia)  
*Occurrence of leaf rust resistance genes in russian wheat varieties and their influence on virulence frequencies in the pathogen population*
- P6. Galina Volkova** (All-Russian Research Institute of Biological Plant Protection of Russian Academy of Agricultural Sciences, Russia)  
*Variability and virulence of *Puccinia triticina* in North Caucasus, Russia*
- P7. Galina Volkova** (All-Russian Research Institute of Biological Plant Protection of Russian Academy of Agricultural Sciences, Russia)  
*Distribution of the stem rust pathogen (*Puccinia graminis* f. sp. *tritici*) and effectiveness of wheat resistance genes in North Caucasus, Russia*
- P8. Mahmut Can Hız** (Bogazici University, Turkey)  
*Molecular marker screening of Turkish wild wheat species for stem rust resistance to ug99*
- P9. Olga Babayants** (PBGI, Ukraine)  
*Yellow rust in the South of Ukraine and resistance of wheat varieties to it in the region*
- P10. Radivoje Jevtic** (Institute of Field and Vegetable Crops, Serbia)  
*Virulence of *Blumeria graminis tritici* in Serbia (2000-2009)*
- P11. Tyrshkin L.G.** (Vavilov All-Russian Institute of Plant Industry, Russia)  
*Seasonal variation in genetic structure of leaf rust pathogen on cereals*
- P12. Zoran Jerković** (Institute of field and vegetable crops, Serbia)  
*Growth ratio differences as leaf rust forecasting factor in semiarid region*
- P13. Zafer Mert** (The Central Research Institute for Field Crops, Turkey)  
*An overview of the network for important cereal diseases management research in Turkey between 2003 and 2007*

## Genetics of Rust Resistance

- P14. Ahmad Arzani** (Isfahan University of Technology, Iran)  
*Study of adult-plant resistance genes for leaf rust in Marvdasht cultivar*
- P15. Alexey Morgounov** (CIMMYT, Turkey)  
*Global genetic diversity of winter wheat germplasm for resistance to leaf, yellow and stem rust*

**P16. Pawel Czembor** (Plant Breeding and Acclimatization Institute, Poland)  
*Mapping new resistance gene to Puccinia hordei orth. in barley landrace ph944-3*

**P17. Galina Volkova** (All-Russian Research Institute of Biological Plant Protection of Russian Academy of Agricultural Sciences, Russia)  
*Resistance of wheat cultivars to Puccinia spp. in Southern Russia and Uzbekistan*

**P18. Hamid Dehghani** (Tarbiat Modares University, Iran)  
*Biplot analysis of diallel data in strip rust of wheat*

**P19. Henriette Goyeau** (INRA UMR 1290, France)  
*Setting up of a differential set to analyse the evolution of durum wheat leaf rust populations in France in 1999-2007.*

**P20. Inna Lapochkina** (Agriculture Research Institute of the Non-Chernozem Zone, Russia)  
*Identification of homeological genes of resistance to leaf rust and powdery mildew in Ae.speltoides, Ae.triuncialis and in common wheat lines obtained with their use*

**P21. Tyrshkin L.G.** (Vavilov All-Russian Institute of Plant Industry, Russia)  
*Induction of leaf rust resistance in cereals' leaf segments by the benzimidazole*

**P22. Tyrshkin L.G.** (Vavilov All-Russian Institute of Plant Industry, Russia)  
*Genetics of adult leaf rust resistance in wheat local samples from VIR collection*

**P23. Ebrahim Ghasemzadeh** (Islamic Azad University, Karaj-Branch)  
*Identification of Puccinia triticina resistance genes in seedlings of Iranian wheat advanced lines*

**P24. Figen Yildirim Ersoy** (Uludağ University, Turkey)  
*Two-hybrid-based analysis of protein-protein interactions of the wheat RAD6 protein*

**P25. Yasin F Dagdas** (Middle East Technical University, Turkey)  
*Detection of micro RNAs putatively involving in disease resistance responses by screening and computational means*

### **Molecular Biology and Physiology of Rusts**

**P26. Elena Gulyaeva** (All-Russian Institute for Plant Protection (VIZR), Russia)  
*Characterization of Puccinia triticina population from Russia in 2007 for virulence and DNA markers*

**P27. Evsey Kosman** (Institute for Cereal Crops Improvement, Israel)  
*Virulence diversity of Israeli population of Puccinia triticina on wheat in 1993-2008*

**P28. Galina Volkova** (All-Russian Research Institute of Biological Plant Protection of Russian Academy of Agricultural Sciences, Russia)  
*Studying the effects of wheat cultivars genotypes on the virulence variability of Puccinia striiformis*

**P29. Sabine Fehser** (Westphalian Wilhelms-University Münster, Germany)  
*"Ug99", a Sr31-breaking race of the wheat stem rust fungus - An initial histological and molecular analysis*

**P30. Sambasivam Kuppusamy Periyannan** (The University of Sydney, Australia)  
*Detection of the shortest Sr22-carrying segment in common wheat*

**P31. Sarah Hambleton** (Eastern Cereal and Oilseed Research Centre, Canada)  
*Development of molecular detection assays for environmental monitoring of cereal rust pathogens*

**P32. Zhensheng Kang** (Northwestern A&F University, P.R. China)  
*Histochemical and cytochemical studies on the accumulation of reactive oxygen species (O<sub>2</sub><sup>-</sup> and H<sub>2</sub>O<sub>2</sub>) in the incompatible and compatible interaction of wheat - Puccinia striiformis f.sp. tritici*

**P33. Dina Raats** (University of Haifa, Israel)

*Fine mapping of the stripe rust resistance gene, yrh52, based on comparative analysis with rice, barley and Brachypodium genomes*

**P34. Tolga Osman Bozkurt** (Middle East Technical University, Turkey)

*Histopathology and transcriptome analysis of wheat responses during compatible and incompatible race-specific interactions with Puccinia striiformis f. sp. tritici*

**P35. Kreshnik Bozhanaj** (Middle East Technical University, Turkey)

*Effect of silenced FAS-Associated Factor 1 upon Blumeria graminis infection in barley*

### **Epidemiology**

**P36. Felicyta Walczak** (National Research Institute Poznań, Poland)

*The influence of climate changes on the powdery mildew (Blumeria graminis) occurrence and harmfulness in last years in Poland*

**P37. Klára Manninger** (Plant Protection Institute Hungarian Academy of Sciences, Hungary)

*Occurrence and virulence changes of leaf rust in Hungary during 1999-2008*

**P38. Lubomir Vechet** (Crop Research Institute, Czech Republic)

*Incidence and development of powdery mildew (Blumeria graminis f.sp. tritici) epidemic in the Prague region in single years of the decade 1999 – 2008*

**P39. Lütfi Çetin** (Central Research Institute for Field Crops, Turkey)

*Determination of reactions some of wheat genotypes against wheat stem rust (Puccinia graminis f.sp. tritici) in Kastamonu epidemic conditions in 2007 and 2008*

**P40. Mahubjon Rahmatov** (Tajik Agrarian University, Tajikistan)

*Epidemiology of wheat powdery mildew in Tajikistan*

### **Mildew Variation and Resistance**

**P41. Anna Tratwal** (National Research Institute Poznań, Poland)

*Variety and species mixtures as the possibility of powdery mildew (Blumeria graminis) incidence reduction in cereals*

**P42. Antonin Dreiseitl** (Agricultural Research Institute Kromeriz, Czech Republic)

*Evolutionary forces and emerging pathotypes of Blumeria graminis f.sp. hordei*

**P43. Jerzy H. Czembor** (Plant Breeding and Acclimatization Institute – IHAR, Poland)

*Resistance to powdery mildew in selections from barley landraces collected in Georgia, Azerbaijan, Iraq and Iran*

**P44. Jerzy H. Czembor** (Plant Breeding and Acclimatization Institute – IHAR, Poland)

*Barley landraces from Israel as sources of resistance to European isolates of Blumeria graminis f.sp. hordei*

**P45. Klára Krizanová** (HORDEUM Ltd., Slovak Republic)

*Powdery mildew resistance - progress and results of spring barley breeding in the Hordeum Ltd., plant breeding station*

**P46. Mohamed Bechir Allagui** (National Institute for Agricultural Research (INRAT), Tunisia)

*Effectiveness of oat resistance sources to powdery mildew and crown rust in Tunisia's conditions*

**P47. Olga Domeradzka** (Plant Breeding and Acclimatization Institute (IHAR), Poland)

*Pathogenicity of powdery mildew (Blumeria graminis (DC.) Speer) on triticale (x Triticosecale Wittm.) in Poland*

**P48. Veronique Troch** (University College Ghent (Ghent University Association), Belgium)  
*Virulence of the powdery mildew (*blumeria graminis*) population on Triticale in Belgium (flanders)*

**P49. Asude Callak-Kirisozu** (Middle East Technical University, Turkey)  
*Molecular characterization of *Blumeria graminis* f. sp. hordei using AFLP markers*

#### **Molecular Biology of Mildew Resistance**

**P50. Balazs Barna** (Plant Protection Institute, Hungary)  
*Induction of resistance to leaf rust in wheat by barley powdery mildew*

**P51. Gina Brown-Guedira** (USDA-ARS, USA)  
*High-resolution mapping of the wheat *Lr46* pleiotropic rust resistance locus*

**P52. Irena Jakobson** (Tallinn University of Technology, Estonia)  
*QTL analysis of powdery mildew resistance in an introgressive line of bread wheat*

**P53. Lubomir Vechet** (Crop Research Institute, Drnovska 507, 161 06 Prgue-Ruzyne, Czech Republic)  
*Effectiveness of resistance specific genes wheat lines to population of powdery mildew (*Blumeria graminis* f.sp. tritici)*

**P54. Yasin F. Dagdas** (Middle East Technical University, Turkey)  
*VIGS of an *F-box* protein decreases powdery mildew resistance in barley*

#### **Breeding for Disease Resistance**

**P55. Alma Kokhmetova** (Institute of Plant Biology and Biotechnology, Almaty)  
*Identification of donors and breeding material resistant to wheat stripe rust*

**P56. Aleksandra Pietrusińska** (Plant Breeding and Acclimatization Institute at Radzików PL)  
*Introduction of two resistance genes against powdery mildew (*Blumeria graminis* f. sp. tritici) and leaf rust (*Puccinia recondita* f. sp. tritici) to winter wheat (*Triticum aestivum*)*

**P57. Beyhan Akin** (CIMMYT, Turkey)  
*Leaf rust (*Puccinia triticina*) resistance genes in *IWWIP* winter-facultative wheat (*triticum aestivum* l.) genotypes*

**P58. Elizaveta Kovalenko** (All Russian Research Institute of Phytopathology, Russia)  
*Selection for resistance sources of wheat to the most harmful diseases for creation durable resistant cultivars*

**P59. Elizaveta Kovalenko** (All Russian Research Institute of Phytopathology, Russia)  
*Evaluation of species from genus *Aegilops* for resistance to leaf rust and septoriosis*

**P60. Mira Dzhunusova** (Kyrgyz National University, Kyrgyzstan)  
*Breeding for resistance to rust diseases of wheat in Kyrgyzstan*

**P61. Muzaffer Tosun** (Ege University, Turkey)  
*Combining ability for plant height, spike length and thousand kernel weight in crosses of powdery mildew resistant wheat lines*

**P62. Olga Domeradzka** (Plant Breeding and Acclimatization Institute (IHAR), Poland)  
*Introduction of powdery mildew (*Blumeria graminis* (DC.)Speer) resistant gene *Pm21* into winter triticale (*x Triticosecale* Wittm.)*

**P63. Siroos Mahfoozi** (Seed and Plant Improvement Institute (SPII), Iran)  
*Improvement of drought tolerant winter and facultative wheat promising lines resistant to yellow rust and stem rusts (*Ug99*)*

**P64. Yuriy Zelenskiy** (CIMMYT, Kazakhstan)

*Improvement of leaf rust resistance of spring bread wheat in the North Kazakhstan*

**P65. Zafer Mert** (The Central Research Institute for Field Crops, Turkey)

*Research on inheritance of yellow rust resistance in Izgi-2001 wheat cultivar*

### **Pathogen Genomes**

**P66. Nick Lauter** (Iowa State University, U.S.A)

*Model genome interrogator: a PLEXdb module that leverages sequenced genomes for motif discovery via meta-promoter extraction and analysis*

**P67. Roger P Wise** (Corn Insects and Crop Genetic Research, USDA-ARS, USA)

*PLEXdb: Plant and pathogen expression database and tools for comparative and functional genomic analysis*

**P68. Stephanie Walter** (University of Aarhus, Slagelse)

*Omics approaches to understand the nature of virulence in Puccinia striiformis f.sp. tritici*

**P69. Aslihan Gunel** (Middle East Technical University, Turkey)

*Identification of proteins differentially expressed upon BTH treatment of Triticum aestivum (wheat) by proteomics approach*

**P70. Manoocheher Khodarahmi** (Seed and Plant Improvement Institute, Iran)

*Identification of Resistance to Yellow Rust in Wheat Germplasm in Iran*

**P71. Manoocheher Khodarahmi** (Seed and Plant Improvement Institute, Iran)

*Diallel Analysis of Yellow Rust Resistance Components in Wheat Genotypes*

**P72. Manoocheher Khodarahmi** (Seed and Plant Improvement Institute, Iran)

*Inheritance and Gene Action for Resistance to Stripe Rust in Bread Wheat*

# **PLENARY PRESENTATIONS**

## **Building international cooperation: crucial to mitigate the threat from Ug99 race of stem rust and other rust pathogens**

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Race Ug99, or TTKSK, of fungus *Puccinia graminis* f. sp. *tritici*, causing stem or black rust disease on wheat (*Triticum aestivum*) has been recognized as a major threat to wheat production. First detected in Uganda in 1998 and now spread throughout East Africa, Yemen, Sudan and Iran, with further predicted spread towards North Africa, Middle East, Asia and beyond, has raised serious concerns of major epidemics that could destroy the wheat crop in various areas. Detection of two new Ug99 variants, TTKST and TTTSK, in Kenya in 2006 and 2007 with virulence to genes *Sr24* and *Sr36*, respectively, also show that Ug99 is evolving. The TTKST variant caused severe epidemics in 2007 in the southern region of Kenya on the *Sr24* carrying variety Kenya Mwamba and rendered about half of the previously known Ug99-resistant global wheat materials susceptible. This has further increased the vulnerability of wheat globally. Rigorous screening since 2005 in Kenya and Ethiopia of wheat materials from 22 countries and International Centers has identified resistant materials, although in low frequency, that have the potential to replace susceptible cultivars. Diverse sources of adequate resistance, both race-specific and adult-plant type, are now available in improved wheat backgrounds and are being used in breeding worldwide. Ug99 threat in most countries can be reduced to low levels by urgently identifying, releasing and providing to growers seed of new high yielding, resistant varieties. The Borlaug Global Rust Initiative (BGRI) was launched in 2005 to build international partnership, raise awareness and financial resources, develop and implement research and developmental priorities to mitigate the threat from Ug99. The consortium has been very successful in achieving these objectives and provides a model for tackling global issues including the threat posed by the yellow rust pathogen. Strong partnerships among and between institutions in developed and developing countries, and international centers will be necessary to bring long-term control of rust diseases in innovative ways through a better pathogen surveillance, development of critical screening sites and facilities, higher emphasis on the use of multiple minor genes in breeding varieties with near-immune levels of resistance, identifying new sources of race-specific resistance genes and their deployment in combinations aided by marker-assisted breeding, and simplifying breeding by developing multiple resistance genes carried in cassettes following their cloning.

## Rates of evolution in *Puccinia striiformis*

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Populations of plant pathogens are shaped by the evolutionary forces operating on these in time and space. The main forces operating on *Puccinia striiformis* (*PSt*) fungus, which is dicaryotic, biotrophic and asexually reproduced, is selection induced by the host crop, migration reflected by frequent and potential long range spore dispersal, and genetic drift due to large fluctuations in pathogen population size across the host crop growing season. Human involvement by plant breeding and cropping system may greatly enhance the effects and speed of these evolutionary processes in *PSt* as compared to organisms in wild ecosystems. Recombination may create new genotypic variability but mutation is the ultimate source of new allelic variation. However, the impact of asexual recombination and the rates of mutation are poorly understood in *PSt*. We analysed the rate of evolution in three lineages of a northwest European *PSt* population. Pathogen samples were collected between 1975 and 2002 in the UK and Denmark, and assayed for 14 individual avirulence/virulence alleles and up to 234 AFLP primer pairs producing approximately 17,000 AFLP fragments. The large number of fragments and a targeted sampling of isolates allowed a reconstruction of detailed phylogenies, i.e., limited homoplasy and a pathogen sample representation of sequential, evolutionary steps. Recent phenotypic loss of avirulence was observed at least once for loci corresponding to *PSt* resistance *Yr2*, *Yr3*, *Yr4*, *Yr7*, *Yr9*, and *Yr15*, whereas *Avr6* and *Avr17* were lost independently in all three lineages, corresponding to 16 events of loss of avirulence (emergence of virulence). The opposite process, restoration of avirulence, was observed for *Yr9* and *Yr32*. An interpretation of phenotypic changes within lineages as independent mutation events resulted in mutation frequencies from  $1.4 \times 10^{-6}$  to  $4.1 \times 10^{-6}$  per AFLP fragment (locus) per generation, whereas the effective rate by which a mutation from avirulence to virulence was established in the pathogen population, when subject to selection by host resistance genes, was approximately three orders of magnitude faster. Rates of evolution of virulence in the absence of host induced selection may be within the same order of magnitude, although these estimates were less accurate due to fewer observations and a potential sample bias. The staggering rates of phenotypic change from avirulence to virulence were confirmed in field experiments where spontaneous virulence mutants emerged at a field scale. Although asexual recombination in *PSt* may occur, recombination rates were low in the NW-European population on the basis of observed lengths of phylogenetic trees compared to expected tree lengths assuming recombination.

## Regional Epidemics and Management of Wheat Stripe Rust in China

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China is one of the biggest agricultural countries as well as the largest wheat producer in the world. Wheat is grown on near 24 million hectares and the total wheat production exceeds 100 billion kg (MOA, 2007). Stripe (or yellow) rust caused by *Puccinia striiformis* f. sp. *tritici* is the most destructive foliar diseases of wheat in many areas around China. Epidemics of the disease have occurred annually since 1950 and the losses of wheat yields due to stripe rust have totalized about 60 billion kg. A great deal of effort has been spent on the survey of regional epidemics, pathogenicity of wheat stripe rust, and strategies for its control in China over the years. It has been found that South of Gansu and Northwest of Sichuan are the most important over-summer areas of *P. striiformis* that act as major sources of inoculum for the autumn-sown wheat in the eastern areas and as a variable zone of rust virulence and wheat cultivar resistance to stripe rust. And a total of 33 formally nominated races (CYR1 to CYR33) and their frequencies have been determined from the stripe rust samples collected throughout the country, of which CYR32 and CYR33 was detected to be the dominant races with the frequency of 30.4% and 12.3% respectively in a total of 5445 isolates tested during 2001-2006. Ecological control of wheat stripe rust in the over-summer areas of the pathogen sources has been considered as the major strategy of sustainable disease control. Effective measures have been put forward and developed as follow in recent years: (1) Improving cultivar resistance and reasonably deploying resistance genes to enhance genetic diversity of wheat rust resistance. A series of improved disease-resistant cultivars (or lines) has been developed by incorporating the resistance genes *Yr3b*, *Yr4b*, *Yr5*, *Yr10*, *Yr11*, *Yr12*, *Yr13*, *Yr14*, *Yr15*, *Yr16*, *Yr17*, *Yr18*, *YrC591*, *YrSp* etc., as well as the durable resistance cultivars and the other resistance materials. DNA molecular markers closely linked or co-segregating with the resistance genes *Yr2*, *Yr5*, *Yr7*, *Yr8*, *Yr9*, *Yr10*, *YrSp*, *YrJu4*, *YrKy2*, *YrC591* and two sets of wheat near isogenic lines with the resistance genes *Yr1*, *Yr2*, *Yr5*, *Yr7*, *Yr9*, *Yr10*, *YrV23*, *YrSp*, *YrKy2*, *YrJu4*, based on the recurrent parents of Mingxian 169 (winter habit) and Taichung 29 (spring habit) respectively, have been successfully developed. (2) Changing cultural practices to raise crop diversity. Maize, upland rice, oil sunflower and other vegetable and forage crops with highly economic value have been introduced and extended, to cut down the wheat plantation in the over-summering areas. Now plastic-mulched maize has been extended to a total acreage of more than 70,000 ha with the economic benefits 2-3 times greater than wheat, leading to a reduced wheat acreage of 40% in the 'hotspots'. (3) Eradicating volunteer seedlings of wheat to cut off the 'green bridge' of inoculum from late-matured wheat to early-sown seedlings. Furrowing deeply more than two times could ultimately control the volunteer wheat after harvest. (4) Regulating wheat planting date and seed dressing with fungicides to reduce the amount of inoculum in the areas of the pathogen sources. Conclusively, establishment of a new agro-ecosystem in the areas of inoculum sources of *P. striiformis* is commended as a more economical and practical approach to the sustainable control of wheat stripe rust epidemics in the whole country of China.

## Achieving durable rust resistance in agriculture: from gene to continent and beyond

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Browder (1985) defined resistance as “any genetically determined characteristic of a host plant that in any way limits damage produced by disease”. The most important features of resistance are the level of protection from economic damage afforded, and durability. Durable resistance is “resistance that remains effective when a cultivar is grown widely in environments favouring disease development” (Johnson 1978). There are examples of simply inherited durable resistances (*eg. Lr34/Yr18, Sr2, mlo*) and of polygenic durable resistances (*eg. leaf rust and stripe rust resistance in Pavon 76; William et al. 2007*).

The durability of polygenic resistance is often attributed to the increasingly lower probability of plant pathogens to simultaneously acquire virulences matching increasing numbers of pyramided resistance genes. In contrast, the basis for the durability of some single gene resistances is less clear but could involve different molecular mechanisms, as suggested for genes like *Lr34/Yr18* and *Yr36*.

While there are many examples of non-durable major resistance genes (*eg. seedling resistance gene Lr26, adult plant resistance gene Lr22b*), there are also examples of genes that “remained effective when present in a cultivar that was grown widely in environments favouring disease development”, and yet eventually succumbed. Stem rust resistance genes *Rpg1* in barley and *Sr31* in wheat were regarded as durable; however, in both cases pathotypes with matching virulence were eventually detected (races QCC and Ug99, respectively). These examples serve as a warning that there can be no guarantee of durability for any resistance source.

It could be generalised that all major gene resistances will eventually prove to be non-durable and should be avoided, and conversely that minor gene resistances are durable. However, Qi et al. (1999) obtained evidence of race specificity for partial resistance to leaf rust in barley, and Rouse et al. (1980) provided evidence for the erosion of slow-mildewing in wheat. The latter authors stated that “the interaction between components of slow-mildewing resistance and parasitic fitness indicate that the resistance could at least to some extent erode over time”. The recent introduction of an exotic pathotype of *P. striiformis* f. sp. *tritici* (*Pst*) into Australia resulted in a rapid and major shift in the Australian *Pst* flora (Wellings 2007). This pathotype is very similar to, or the same as, one occurring in North America and now regarded as being more aggressive (Milus et al. 2006). It has had a significant effect on Australian wheat germplasm that is not related to major gene resistance – and so the question is raised, “has there been an erosion of minor gene resistance?”

Combinations of major resistance genes have been used successfully to control stem rust in both Australia (Park 2007) and North America (Martens and Dyck 1989). In the latter case, Martens and Dyck (1989) concluded that for more than 30 years, relatively few major genes in various combinations, from four genetic sources (Iumillio durum, Yaroslav emmer, Hope and McMurachy), provided control of stem rust in an epidemiological system comprising hundreds of millions of hectares and thousands of billions of rust propagules. Significantly, they also stated that the genetic basis of this widespread control of stem rust was not fully understood. Clearly, the factors that determine the durability of resistance genes when deployed over large areas are complex. There is evidence that genetic “background” can influence durability (*eg. the pvr2<sup>3</sup> gene in pepper, Palloix et al. 2009; Lr24 in wheat in Australia, Park et al. 2000*), lending credence to the strategy of using reputedly durable sources of rust resistance as “background” resistance, to which other resistance genes are added (McIntosh 1988).

In view of the complexity of host : pathogen interactions, genetic diversity must be seen as a key ingredient in large scale sustained control of plant diseases. It has been argued that even where specific or major resistance genes are used, genetic diversity can be used as insurance against lack of durability and hence as a means of reducing genetic vulnerability (McIntosh 1988). Above all, an understanding of the resistance genes present in cultivars and breeding populations, and monitoring pathogen populations with respect to deployed resistances, are crucial in ensuring that the genetic bases of resistances are not narrowed.

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## Regulation of innate immunity in barley-powdery mildew interactions

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Plants have evolved complex regulatory mechanisms to control the defense response against microbial attack. Both temporal and spatial gene expression are tightly regulated in response to pathogen ingress, modulating both positive and negative control of defense. BLUFENSINs, small peptides in barley, wheat, and rice, are highly induced by attack from the obligate biotrophic fungus, *Blumeria graminis* f. sp. *hordei*, causal agent of powdery mildew disease. BLN1 negatively impacts plant defense, is predicted to be secreted, and contains both structural and sequence similarities to knottins, small disulfide-rich proteins characterized by a unique disulfide through disulfide knot. To discern regulatory targets of BLN1, we conducted Barley1 GeneChip analysis of *Bln1*-silenced plants via *Barley stripe mosaic virus*-induced gene silencing (BSMV-VIGS). Sixty GeneChip hybridizations were performed, based on 5 replications of 12 BSMV-VIGS/host-pathogen interactions. Mixed linear model analysis revealed 98 significant new genes ( $p < 0.0001$ ; FDR < 2%) that are suppressed together with *Bln1* (Contig12219\_at;  $p=6.28^{E-06}$ ), or induced when we compare BSMV:Bln1<sub>248</sub> silenced plants to the BSMV:00 control. These candidates appear to have key roles in *R*-gene mediated and innate immunity networks, thus, the functional identification of their precise roles will be a significant step in understanding plant defense.

## **The molecular basis of broad-spectrum powdery mildew resistance**

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Loss-of-function mutant alleles of the barley *Mlo* locus are known to confer durable, broad spectrum resistance against the powdery mildew disease caused by the Ascomycete *Blumeria graminis* f.sp. *hordei*. This type of antifungal immunity has been discovered 65 years ago and has been widely used in European agriculture for more than 25 years. We recently showed that powdery mildew resistance conferred by *mlo* alleles is not restricted to barley, but also occurs in Arabidopsis, tomato and pea. The molecular basis of this unusual type of disease resistance remains, however, mysterious. We exploit the genetic and molecular tools available for the dicot reference species, *Arabidopsis thaliana*, to get insights into the molecular mechanisms leading to *mlo* resistance.

## **Clonality and recombination footprints in wheat yellow rust genetic structure**

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Parasitic species can exhibit sophisticated life cycles, displaying complex transmission strategies based on the use of specific propagules and the infection of different host species. Uredinales are characterised by one of the most complex life-cycles in fungi, with the differentiation of five kinds of spores, and with the sexual and asexual cycles occurring on two distinct and unrelated hosts: an aecidial and a telial host, respectively. In wheat, three uredinale species are of major agronomic importance due to the epidemics caused by their asexual cycle: *Puccinia striiformis* fsp *tritici* (PST), *P. triticina* and *P. graminis* (respectively yellow, brown and black rust). Aecidial hosts and sexual cycles are characterised for brown and black rusts, while the alternate host of yellow rust is still unknown. This lack of a known aecidial host has been used to explain the clonal evolution of some PST populations. Recent molecular studies performed at different scales on PST have confirmed the clonality of populations in some areas, while recombination has been detected in other parts of the world. Using different results we obtained on the genetic structure of PST populations and recent experiments performed on the diversity of teliosore production, we defend the hypothesis of the presence of sexual recombination in PST, against the alternate hypothesis of parasexual recombination.

## Molecular basis of durable rust resistance in wheat

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Improved resistance to fungal rust diseases in cereals is critical for world food security and can only be achieved through breeding varieties with durable rust resistance. Durable resistance is not caused by single genes, but by a combination of usually 3-5 individual partial genes which are combined by classical breeding in a single wheat line to achieve near immunity. The wheat gene *Lr34* is such a quantitatively acting, partial resistance gene which is involved in durable resistance. *Lr34* is associated with resistance to two different rust diseases of wheat, leaf rust (caused by *Puccinia triticina*) and stripe rust (*P. striiformis*). Interestingly, it also confers partial resistance to the powdery mildew pathogen. *Lr34* is expressed in adult plants during the critical grain filling stage and is most effective in the flag leaf. Flag leaves of many wheat cultivars containing *Lr34* develop a necrotic leaf tip, a morphological marker described as leaf tip necrosis. The expression of this trait, however, is strongly dependent on the environment. The *Lr34* gene was first documented in Canada although *Lr34*-containing germplasm has been a part of wheat improvement since the early part of the 20<sup>th</sup> century and seems to have originated in Italian and Chinese material. Wheat cultivars containing *Lr34* occupy more than 26 million hectares in various developing countries alone and contribute substantially to yield savings in epidemic years.

The *Lr34* gene has remained durable and no evolution of increased virulence towards *Lr34* has been observed for more than 50 years of large scale use in resistant wheat lines. Despite the importance of partially acting plant resistance genes involved in durable resistance, very little is known on them at the molecular level. Understanding the molecular nature of this class of resistance has important implications for long term control of rust diseases. Previous studies have localized the co-dominant gene *Lr34* on the short arm of wheat chromosome 7D between the two markers gwm1220 and SWM10. We further reduced the target interval in a map-based cloning approach based on three high-resolution populations. High-resolution mapping revealed a 0.15 cM target interval for *Lr34*. The 363 kb physical interval containing both flanking markers was fully sequenced in the *Lr34* containing hexaploid wheat cultivar 'Chinese Spring'. A PDR/ABC transporter coding gene was finally identified as *Lr34* by the molecular analysis of 8 independent mutants. The mutations were identified as missense mutations, splice site mutations and short deletions. Interestingly, an allele of the *Lr34* gene is also found in all wheat lines lacking *Lr34* activity. This susceptible allele differed from the resistant form in only two amino acid changes. Based on these polymorphisms, highly specific molecular markers have recently been developed. They allow diagnostic detection of *Lr34* in tested germplasm and will be highly useful in combining *Lr34* with other partial resistance genes to achieve near immunity. Recent data on *Lr34* orthologs in several Triticeae and other grass genomes will be presented. In addition, strategies for molecular understanding of *Lr34* function and possible applications in agriculture will be discussed.

## Wild emmer wheat as a source for disease resistance genes: from genetic diversity to gene cloning

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The studies conducted by our group are focused on unraveling the genetic basis of several qualitative and quantitative agronomic traits derived from wild emmer wheat, *Triticum dicoccoides*. Wild emmer wheat, the tetraploid ancestor of domesticated wheat, was discovered in 1906 by A. Aaronsohn in Israel. Aaronsohn had the pioneering vision that wild wheat would become a source of germplasm for crop improvement. Nevertheless, traditional approaches for utilization of wild alleles are usually very slow. The advanced genomic technology available today may help to increase the efficiency of utilization of wild germplasm.

Stripe rust, caused by the fungus *Puccinia striiformis* f. sp. *tritici* is a devastating fungal disease in many wheat-growing regions of the world. New strategies to reduce stripe rust yield losses are required to satisfy the increasing world demand for cereals. Wild emmer wheat, is a promising source of resistance to stripe rust (e.g. *Yr15*, *YrH52* and *Yr36*). *Yr15* and *YrH52* are major dominant genes that confer particularly high resistance; *Yr36* confers high temperature, partial (quantitative) resistance. The availability of wheat genetic maps, wheat ESTs mapped to deletion bins, wheat BAC libraries, and the complete rice and *Brachypodium distachyon* genome sequences enabled us to conduct positional cloning studies aimed at cloning of the stripe rust resistance genes derived from wild emmer wheat.

A primary genetic map of *Yr15* was developed using a cross of a BC<sub>3</sub>F<sub>9</sub> line, which contains a 1BS chromosome segment of *T. dicoccoides* accession G-25 carrying *Yr15*, with the recurrent parent *T. durum* cv. D447. SSR and RFLP markers were used to assign *Yr15* to 1BS chromosome deletion bin Sat0.31. ESTs assigned to 1BS Sat0.31 enabled us to establish colinearity with a 740 kb contig located on *Oryza sativa* chromosome 5 and a 840 kb region located on *B. distachyon* chromosome 2. Further comparative genomic study enabled to narrow down the region carrying *Yr15* to 0.3 cM colinear with a ~28 kb sequence in *B. distachyon*. The sub-centiMorgan map of *Yr15* was used to identify *T. aestivum* BAC clones spanning the target region and for constructing a non-gridded BAC library from the donor line of *Yr15*. Further chromosome walking is underway to assemble a BAC contig containing *Yr15*.

Using a similar comparative genomic approach we have recently completed the positional cloning of the slow rusting gene *Yr36* (Science 323:1357-60), derived from wild emmer wheat. *Yr36* is effective only under relatively high temperatures and provides partial resistance to stripe rust. A gene designated *WKS1* was found to be completely linked to *Yr36* in a large mapping population. *WKS1* has a novel architecture with a kinase and a START lipid-binding domain. Six independent mutations and transgenic complementation confirmed that *WKS1* is *Yr36*. This gene, which was lost during the domestication of pasta and bread wheat, provides a new tool to control this disease and has the potential to contribute to the improvement of stripe rust resistance in a wide range of germplasm.

These studies demonstrate the potential of wild emmer wheat gene pool for improvement of cultivated tetraploid and hexaploid wheats, and emphasize the contribution of the recently developed genomic tools for the utilization of wild wheat germplasm.

## Convergent Evidence for Genes Underlying Quantitative Powdery Mildew Resistance in Barley

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Quantitative pathogen resistance is of high importance to plant breeders due to its durability. However, it usually is polygenic in nature and controlled by quantitative trait loci, which makes it difficult to handle in practice. Therefore, knowing the genes that underlie quantitative resistance would allow its exploitation in a more targeted manner. In order to identify genes that mediate quantitative resistance of barley to the powdery mildew fungus *Blumeria graminis* f.sp. *hordei* (*Bgh*) we have combined a functional-genomics approach based on transcript profiling and transient-induced gene silencing (TIGS) with an association-genetic and a QTL-mapping approach. Approximately 500 differentially regulated candidate genes of barley were silenced by RNAi followed by scoring of *Bgh* infection. Silencing or over-expression of 63 of those candidates resulted in a significantly altered interaction phenotype of attacked epidermal cells with *Bgh*. These plus a number of candidate genes based on publicly available data were selected for re-sequencing in a worldwide selection of barley accessions from the IPK genebank that differed in their quantitative resistance to *Bgh*. This approach revealed a number of genes that exhibited significant association with race-nonspecific seedling resistance. Two candidate genes mapped to chromosome 5H, within a QTL interval for seedling resistance to *Bgh* identified in the OregonWolfe Barley population. Re-sequencing of 20 genes within 20 cM at this locus revealed low linkage disequilibrium and identified additional associated gene candidates including the cell-death regulator protein HvLsd1, which was found nearest to the peak QTL marker. In conclusion, the integration of functional-genomic with association-genetic approaches allow us to rapidly zoom into candidate-gene lists and genetic intervals of interest and hold the promise to accelerate the discovery of genes underlying complex, quantitative traits in crop plants.

## **The genome of *Blumeria graminis* f. sp. hordei.**

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We are sequencing and annotating the barley powdery mildew genome using a combination of Sanger and diverse high-throughput sequencing methodologies. Our aim is to provide a reference genome for powdery mildews as a tool to advance our understanding of these challenging obligate biotrophs. One of the early discoveries is that the powdery mildew genomes are ~120Mb, i.e. much larger than closely related ascomycetes. This massive genome expansion is due to the presence of highly repetitive DNA made up of (retro)transposable elements. These are distributed throughout the genome and constitute ~70% of the genome. We propose that this expansion was the result of the loss of RIPing: one of the genetic pathways controlling immunity against genomic parasites. Other data emerging from the project will also be presented.

## Virulence effectors and retrotransposons in *Blumeria graminis*

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AVR<sub>k1</sub> and AVR<sub>a10</sub> are effector proteins that contribute to the successful establishment of haustoria in *Blumeria graminis* f sp *hordei* (*Bgh*), the causal agent of barley powdery mildew. They belong to a large gene family in the genome of *Bgh*, with homologous sequences in other *formae speciales* (*ff. spp*) infecting other grasses. Members of the AVR<sub>k1</sub> family are found in the proximity of TE1a LINE-1 retrotransposons and both can be expressed as a single transcript. We have studied the extensive proliferation of the AVR<sub>k1</sub> gene family throughout the genome of *B. graminis*, with sequences diverging in *ff. spp* adapted to infect different hosts. The frequency with which members of the AVR<sub>k1</sub> and TE1a retrotransposon lineages occur together in the genome is highly significant, and phylogenetic analysis show that both classes of sequences have coevolved. This is the first direct evidence that a parasite effector gene family and a particular retrotransposon lineage are consistently associated and have coevolved. The coevolution of these two entities indicates a mutual benefit to the association, which could ultimately contribute to parasite adaptation and success. We propose that the association would benefit 1) the powdery mildew fungus, by providing a mechanism for amplifying and diversifying effectors and 2) the associated retrotransposons, by providing a basis for their maintenance through selection in the fungal genome.

# **ORAL PRESENTATIONS**

## **Building international cooperation: crucial to mitigate the threat from Ug99 race of stem rust and other rust pathogens**

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Race Ug99, or TTKSK, of fungus *Puccinia graminis* f. sp. *tritici*, causing stem or black rust disease on wheat (*Triticum aestivum*) has been recognized as a major threat to wheat production. First detected in Uganda in 1998 and now spread throughout East Africa, Yemen, Sudan and Iran, with further predicted spread towards North Africa, Middle East, Asia and beyond, has raised serious concerns of major epidemics that could destroy the wheat crop in various areas. Detection of two new Ug99 variants, TTKST and TTTSK, in Kenya in 2006 and 2007 with virulence to genes *Sr24* and *Sr36*, respectively, also show that Ug99 is evolving. The TTKST variant caused severe epidemics in 2007 in the southern region of Kenya on the *Sr24* carrying variety Kenya Mwamba and rendered about half of the previously known Ug99-resistant global wheat materials susceptible. This has further increased the vulnerability of wheat globally. Rigorous screening since 2005 in Kenya and Ethiopia of wheat materials from 22 countries and International Centers has identified resistant materials, although in low frequency, that have the potential to replace susceptible cultivars. Diverse sources of adequate resistance, both race-specific and adult-plant type, are now available in improved wheat backgrounds and are being used in breeding worldwide. Ug99 threat in most countries can be reduced to low levels by urgently identifying, releasing and providing to growers seed of new high yielding, resistant varieties. The Borlaug Global Rust Initiative (BGRI) was launched in 2005 to build international partnership, raise awareness and financial resources, develop and implement research and developmental priorities to mitigate the threat from Ug99. The consortium has been very successful in achieving these objectives and provides a model for tackling global issues including the threat posed by the yellow rust pathogen. Strong partnerships among and between institutions in developed and developing countries, and international centers will be necessary to bring long-term control of rust diseases in innovative ways through a better pathogen surveillance, development of critical screening sites and facilities, higher emphasis on the use of multiple minor genes in breeding varieties with near-immune levels of resistance, identifying new sources of race-specific resistance genes and their deployment in combinations aided by marker-assisted breeding, and simplifying breeding by developing multiple resistance genes carried in cassettes following their cloning.

## **Stem rust screening and breeding for Ug99 resistance in east-Africa**

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East Africa has always acted as a significant region in relation to stem rust epidemiology and is considered to be hot-spot for evolution of new virulent races. In 1998, a new stem rust race (Ug99) originated from Uganda has posed a potential global threat on wheat production as this race is very aggressive in terms of its broad virulence spectrum and adaptability. The east-Africa (Kenya and Ethiopia) stem rust screening and breeding program under the Borlaug Global Rust Initiative (BGRI) was initiated to systematically reduce threat of Ug99 lineage on national wheat production and the world's vulnerability to evolving stem rust races of this lineage. Since its launch in 2005, the stem rust screening programs of Kenya and Ethiopia have screened over 100, 000 advance breeding lines, identified diverse sources of resistance including adult plant resistance based on minor genes, and initiated comprehensive shuttle breeding and seed multiplication in collaboration with CIMMYT and ICARDA. This paper reviews on importance and achievements of stem rust research activities/strategies currently being used in Kenya and Ethiopia programs and there contribution to global stem rust control along with opportunities and future perspectives, and the challenges faced by this highly significant global program.

## Global Initiatives For Management of UG99 Stem Rust Race and Reactions of Some Winter Wheat Genotypes to UG99 in 2008 Year

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Wheat (*Triticum* spp.) is among the most strategic products of Turkey and interest large number of producers in the country. The rust diseases are very important as they are among the most significant biotic factors affecting wheat yield and quality. Stem rust (*Puccinia graminis* f.sp. *tritici*) is very important, which is one of the most significant biotic factors affecting wheat yield and quality and might cause yield production decrease by 70% in Turkey. Through sexual production and mutation, stem rust can produce often more virulent pathotypes that can infect different wheat genotypes which may be resistant previously, resulting in new epidemics. For example, new yellow rust race which originated in Kenya in 1986 caused 0,5 million tones of yield losses in Cukurova region in 1995. Race/pathotype Ug99, first identified in Uganda during 1999 (therefore called Ug99), is known race of stem rust that has virulence for gene Sr31. Later this race was designated as TTKS using the North American nomenclature system. The fact that 90% of international wheat germplasm has been found susceptible to this race has forced many important wheat producing countries to search for solutions. A global initiative (BGRI Borlaug Global Rust Initiative) has been established to set global management strategies by under the new race threat countries and international organization. Turkey is one of the active member countries of this BGRI and carries out following activities.

In this study, we focused on 1) Monitoring the spread of race Ug99 with the collaboration of international institutes. 2) Determination of resistant germplasm against this race. A total of 169 winter wheat genotypes was sent to Kenya in which UG 99 is very effective for screening to UG99 in 2008. In these genotypes, 20 genotypes were determined as a resistant to Ug 99.

According to this result, we can use these genotypes as resistant germplasm for released or crossing programme. On the whole, the study has a great value in view of producing the necessary information facilitating the control of stem rust through utilization of genetic resistance which is the most environmentally friendly method. Finally, its contribution to establishing the research capacity which the country needs in this field has been realized.

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**Key words:** Stem rust (*Puccinia graminis* f.sp. *tritici*), Wheat (*Triticum* spp), Ug99, Genetic Resistance

## Aggressiveness of races TTKSK and QFCSC of *Puccinia graminis* f. sp. *tritici* at various temperatures

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The broadly virulent race TTKSK of *Puccinia graminis* f. sp. *tritici* has caused epidemics on susceptible varieties of wheat throughout Kenya. Infection as early as the seedling stage has been observed in areas of high altitude with relatively cool temperatures. These observations prompted us to determine whether TTKSK is more adapted to infection at cooler temperatures than other races of *P. graminis* f. sp. *tritici*. In order to test the hypothesis that race TTKSK is adapted to infect wheat at cooler temperatures, we measured two components of aggressiveness at various temperatures for races TTKSK and QFCSC, the latter is the predominant race occurring in North America. Urediniospores of the same age for each race were inoculated at a low concentration onto the primary leaves of six susceptible wheat lines: Federation, Line E, Morocco, Roane, Winmaster, and Yellowstone. Leaves were monitored daily with a hand lens to determine latent period. Single pustule length and width were measured with digital calipers on the 13th day following inoculation and used to estimate pustule area. In order to test the effect of temperature during the incubation period, plants were placed in growth chambers at 20°C, 16°C, or 12°C following standard infection at 18°C. To test the effects of temperature during the infection period, plants were placed in dew chambers at 20°C, 16°C, or 12°C for 18 hours followed by incubation in a growth chamber at 20°C. Mean latent period and pustule area were determined for 10 single pustules each on a different leaf for each combination of wheat line, rust race, and temperature treatment. These means were compared to determine differences in aggressiveness between race QFCSC and race TTKSK at the different temperature treatments. Pustule area data were log-transformed to approximate a normal distribution. Pairwise *t*-tests were used to compare temperature treatment means between the two races across wheat lines. No differences in latent period were found between the two races for any of the treatments. For pustule area, no differences were found between the two races when the infection temperature was 18°C, regardless of the incubation temperature (20°C  $p=0.66$ , 16°C  $p=0.36$ , and 12°C  $p=0.87$ ). TTKSK had 87% and 98% larger mean pustule areas compared to QFCSC after infection at lower temperatures (16°C  $p=0.011$  and 12°C  $p=0.041$ , respectively). These data indicate that race TTKSK is more aggressive than QFCSC at cooler infection temperatures as measured by mean sporulating area 13 days after inoculation.

## Development of a greenhouse screening method for adult plant response in wheat to stem rust

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The detection, adaptation and migration of race Ug99 of *Puccinia graminis* f. sp. *tritici* have sparked a global interest amongst wheat scientists to counter a potential stem rust threat. Amongst these efforts a need for effective screening methods, especially in adult plants, was highlighted. At present, screening for adult plant resistance to stem rust occurs in field plots. Although such evaluations are successful if managed properly, field ratings are time consuming, expensive, dependent on the weather and open to inoculum of unwanted races or other confounding diseases. The objective of this study was to develop a dependable screening system for stem rust on adult plants in the greenhouse. Similar systems have been optimized for stripe and leaf rust infection of adult wheat plants. In addition to host and pathogen genotype, rust infection under controlled conditions are influenced by the amount and duration of moisture, light, temperature, spore viability and concentration, and method of application. An experiment consisting of the cultivars Inia 66 (*Sr2*), Hartog (*Sr2*), Kariega (susceptible) and Morocco (susceptible control), three inoculation methods, and two incubation treatments and periods, was conducted. The best results were obtained when plants were inoculated with urediniospores suspended in water, followed by a 24 h dew period in a chamber constructed in the greenhouse. Temperature within the chamber, which consisted of a water tray covered by plastic sheeting, ranged between 30°C (day) and 22°C (night). In a second experiment, a resistant line carrying *Sr31* was added to the above entries and three spore concentrations were compared in two incubation environments. When reaction type, and stem rust severity and frequency were taken into consideration, the best infection was obtained when plants sprayed with 1.25 mg urediniospores per ml water were incubated in a plastic dew chamber in the greenhouse. A comparison of the stem rust reaction type of 20 South African spring wheat cultivars showed that most entries responded similarly in the field and greenhouse.

## Rates of evolution in *Puccinia striiformis*

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Populations of plant pathogens are shaped by the evolutionary forces operating on these in time and space. The main forces operating on *Puccinia striiformis* (*PSt*) fungus, which is dicaryotic, biotrophic and asexually reproduced, is selection induced by the host crop, migration reflected by frequent and potential long range spore dispersal, and genetic drift due to large fluctuations in pathogen population size across the host crop growing season. Human involvement by plant breeding and cropping system may greatly enhance the effects and speed of these evolutionary processes in *PSt* as compared to organisms in wild ecosystems. Recombination may create new genotypic variability but mutation is the ultimate source of new allelic variation. However, the impact of asexual recombination and the rates of mutation are poorly understood in *PSt*. We analysed the rate of evolution in three lineages of a northwest European *PSt* population. Pathogen samples were collected between 1975 and 2002 in the UK and Denmark, and assayed for 14 individual avirulence/virulence alleles and up to 234 AFLP primer pairs producing approximately 17,000 AFLP fragments. The large number of fragments and a targeted sampling of isolates allowed a reconstruction of detailed phylogenies, i.e., limited homoplasy and a pathogen sample representation of sequential, evolutionary steps. Recent phenotypic loss of avirulence was observed at least once for loci corresponding to *PSt* resistance *Yr2*, *Yr3*, *Yr4*, *Yr7*, *Yr9*, and *Yr15*, whereas *Avr6* and *Avr17* were lost independently in all three lineages, corresponding to 16 events of loss of avirulence (emergence of virulence). The opposite process, restoration of avirulence, was observed for *Yr9* and *Yr32*. An interpretation of phenotypic changes within lineages as independent mutation events resulted in mutation frequencies from  $1.4 \times 10^{-6}$  to  $4.1 \times 10^{-6}$  per AFLP fragment (locus) per generation, whereas the effective rate by which a mutation from avirulence to virulence was established in the pathogen population, when subject to selection by host resistance genes, was approximately three orders of magnitude faster. Rates of evolution of virulence in the absence of host induced selection may be within the same order of magnitude, although these estimates were less accurate due to fewer observations and a potential sample bias. The staggering rates of phenotypic change from avirulence to virulence were confirmed in field experiments where spontaneous virulence mutants emerged at a field scale. Although asexual recombination in *PSt* may occur, recombination rates were low in the NW-European population on the basis of observed lengths of phylogenetic trees compared to expected tree lengths assuming recombination.

## Comparison of old and new strains of *Puccinia striiformis* f. sp. *tritici* for ability to initiate epidemics from overwintering infections

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Since 2000, a new strain of *Puccinia striiformis* f. sp. *tritici* has replaced the old strain in eastern United States, and stripe rust (yellow rust) on winter wheat has been more severe than in years before 2000. Previous research using adult plants showed that isolates of the new strain were more aggressive and better adapted to warmer temperature than isolates of the old strain. Epidemics begin from overwintering infections that develop by spring into discrete foci of diseased plants (“hot spots”). The objective of this research was to determine if isolates of the new strain were better adapted to initiate epidemics from overwintering infections. Plants of a susceptible winter wheat cultivar (Croplan Genetics 514W) were grown in 10-cm pots outdoors during October and November. For the 2007-08 season, there were four plants per pot, and plants were inoculated (0.5 mg urediniospores per ml Soltrol 170 mineral oil) on 28 November 2007 with isolates AR90-01 and AR03-33 that are representative of the old and new strains, respectively. Inoculated plants were incubated in a dew chamber for 24 hours at 12°C to promote infection. Eleven random pots per isolate were incubated in a growth chamber to determine the levels of infection, and ten pots per isolate were transplanted 1.5 m apart into strips of the same cultivar in the field at Fayetteville and Kibler, AR on 29 November to initiate foci. The experimental design was similar in the 2008-09 season except that there was only one plant per pot, six pots per isolate were evaluated to determine the levels of infection, and two isolates (AR90-01 and AR97-01) representative of the old strain and two isolates (AR00-05 and AR03-33) representative of the new strain were used. Plants were inoculated on 24 November 2008 and transplanted to the field on 26 November. To quantify the amount of disease in the spring surrounding each inoculated transplant, all of the wheat stems in 0.5-m lengths of the two rows adjacent to each transplant were cut just above the soil, and each stem was rated for stripe rust severity (percentage of leaf area with stripe rust). The average severity across all stems in the sample was calculated to estimate the amount of stripe rust caused by infections on the transplants. Data were analyzed using analysis of variance. In the 2007-08 season, plants inoculated with AR90-01 averaged 19.5 initial infections per pot (range 14 to 28), and plants inoculated with AR03-33 averaged 6.2 infections per pot (range 1 to 16). Isolate AR03-33 caused significantly more disease ( $P \leq 0.001$ ) than AR90-01 at both locations. At Kibler, isolate AR03-33 averaged 21.8% severity compared to 9.5% severity for isolate AR90-01. At Fayetteville, isolate AR03-33 averaged 16.8% severity compared to 3.4% severity for isolate AR90-01.

In the 2008-09 season, the average (and range) of initial infections per plant for each isolate were AR90-01 = 1.3 (1 to 2); AR97-01 = 1.5 (1 to 3); AR00-05 = 2.8 (1 to 4); and AR03-33 = 1.8 (1 to 3). At Kibler, isolates AR03-33 and AR00-05 averaged 7.7% and 9.5% severity, respectively, and caused significantly ( $P \leq 0.001$ ) more disease than isolates AR90-01 and AR97-01 that averaged 1.5% and 1.9% severity, respectively. No useful results were obtained from the Fayetteville location because little stripe rust developed.

The results of this study indicate that isolates representative of the new strain are more aggressive than isolates representative of the old strain for initiating epidemics from overwintering infections and help explain why stripe rust has been more severe since 2000.

## Differential Adaptation of Australian and New Zealand Stripe Rust Isolates to High Temperature

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Stripe rust or yellow rust, caused by *Puccinia striiformis* Westend. f. sp. *tritici* Eriks., is one of the major diseases of wheat in regions with temperate climates including the wheat growing regions of Australia. A new stripe rust pathotype 134E16A+ was introduced to Australia in 2002 and had a virulence pattern distinctly different from the existing Australian stripe rust pathotype population but closely resembled the US race PST78. Since 2002, four new pathotypes have been detected in Australia that are presumed to be single mutant derivatives of 134E16A+. Severe stripe rust epidemics in the United States since 2000 were associated with a new pathogen population that included PST78 and its derivatives. Published work has shown that a feature of the adaptation of this new population was the comparative advantage for growth at high temperature compared to the previous pathogen population. The adaptation was expressed as shorter latent periods and faster urediniospore germination rates. Since the introduction of pathotype 134E16A+ to Australia in 2002, several serious stripe rust epidemics have occurred and it has been proposed that this may in part reflect an adaptation to high temperature in a manner similar to that found for the putative source population in the United States. This study was conducted to investigate the adaptation of stripe rust pathotypes occurring in Australia since 2002 to high temperatures. Isolates of four new pathotypes occurring in Australia after 2002 were compared with isolates of three New Zealand pathotypes, and nine Australian pre-2002 pathotypes. Four susceptible wheat cultivars were used for studying disease development with twenty plants used as replications for each isolate. Latent period duration (in days) and the final percentage of plants with symptoms were measured for assessment of the effect of two different temperatures (17 and 23°C) during post-incubation period (Experiment 1) and two different temperatures (10 and 15°C) during incubation period (Experiment 2). The spore germination rates were assessed on agar media on Petri dishes (experiment 3). Each experiment was repeated twice. An ANOVA performed on the data from Experiment 1 indicated significant interactions ( $P < 0.05$ ) between temperature, pathogen isolate and host plant. Comparison of the least squares means for each isolate across all four susceptible wheat cultivars indicated that the development of all isolates across all cultivars was delayed at the high temperature regime by at least two days ( $P < 0.05$ ). However, the latent period duration of post 2002 isolates was significantly ( $P < 0.05$ ) shorter at high temperature in comparison with that of the pre-2002 isolates. No significant difference in latent period duration ( $P < 0.05$ ) between new vs. old pathotypes was observed at the low temperature regime. The final number of plants showing disease symptoms at high temperature was significantly higher ( $P < 0.05$ ) for the new pathotypes, while no difference was observed between isolates at the low temperature regime. These results suggest that the post 2002 stripe rust population in Australia may be potentially better adapted to higher temperatures and so possess a superior environmental fitness. Confirmation of the results and further statistical analysis for other experiments of the study is currently underway.

## **Adaptation of the clonal pathogen *Puccinia striiformis* f.sp. *tritici* to temperature and resistance genes in North-West Europe and Mediterranean area**

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The genetic diversity of *Puccinia striiformis* f.sp. *tritici* (PST) is strongly structured by host resistance genes with the successive overcome of *Yr* specific resistance genes. Based on molecular markers and pathotypes, a strong and steady spatial structure between northern and southern populations in France was shown during a 20-yr period. If the northern isolates belong to a North-West European PST populations, little is known on population structure in the Mediterranean area. In order to explain the intriguing N/S structure in a species known for its high dispersal ability, we studied the adaptation for temperature of the two major clonal lineages, as well as the genetic structure of PST in Mediterranean region and its relation to specific resistance genes present in the area. The southern isolates were found to be more adapted to high temperature than the northern ones, both in controlled and field conditions. Then the study of virulence and SSR diversity demonstrated that the Mediterranean area is colonised by a clonal lineage related to a more polymorphic population resident in Middle East. In addition, a specific resistance gene (*Yr8*) found in the North African wheat cultivars but not in the North-West European cultivars is also explaining the prevalence of corresponding virulence in the South. Both selective forces, resistance genes and temperature, are driving the evolution of yellow rust populations in Europe. Here we showed that clones adapted to high temperature were present at least 20 years ago, and were characteristic of the Mediterranean population. Given the higher diversity in the Middle East for pathotypes and genotypes, it is suggested that the West Mediterranean population originated from Middle East populations. We provided evidence for differential response of pathogen genotypes to temperature, in accordance with their region of collection. Wheat yellow populations in Europe and Mediterranean area demonstrates how strong is the influence of host and environment interaction for the distribution of clonal pathogens.

## Potential Vulnerability of Barley to an Undescribed Form of *Puccinia striiformis* in Australia

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The pathogens causing stripe rust in barley (*P. striiformis* f. sp. *hordei*; *Psh*) and wheat (*P. striiformis* f. sp. *tritici*; *Pst*) are regarded as separate *formae speciales* although the host ranges of both overlap. In Australia, *Pst* was first detected in 1979 and it continues to be a serious disease of wheat. *Pst* is not an economically important disease of barley in this region because most Australian barley cultivars are highly resistant to locally occurring isolates of *Pst*. *Psh* is not present in Australia, although a majority of barley cultivars from this region are susceptible to race 24 of *Psh* when field screened at CIMMYT, Mexico. However, a new and undescribed form of *P. striiformis* was detected in Australia in 1978. This form is highly avirulent on wheat, is adapted to weedy barley grass communities (*Hordeum* spp.) and can cause significant disease development on certain commercial barley cultivars under greenhouse and field conditions. Pathogenic and molecular characteristics suggest that this form, temporarily designated 'barley grass stripe rust' (BGYR), is a new *formae specialis* within *P. striiformis*.

Genetic analyses of resistance to BGYR using conventionally derived pedigree lines from resistant/susceptible hybrids and selected doubled haploid populations revealed the presence of several resistance genes. Greenhouse seedling tests indicated the presence of dominant and recessive genes, either alone or in simple combinations. Allelism tests revealed common genes among several cultivars. A doubled haploid population provided the opportunity to map a seedling gene for resistance to BGYR in cultivar Sahara 3771 to the long arm of barley chromosome 7H. Field assessment of the same population revealed the presence of an additional independent gene operating in the adult plant stage, which was located on the long arm of chromosome 3H. These studies suggest that the resistance of certain Australian barley cultivars to BGYR could be based on a relatively small set of genes, with further work required to assess the diversity of these genes between cultivars. In the worst case scenario, it could be predicted that relatively few mutational events in the BGYR population could result in this form of *P. striiformis* becoming a significant problem for barley producers in Australia. For this reason, genetic studies will continue, and the pathogen population will be carefully monitored for potentially important pathogenic change.

## Regional Epidemics and Management of Wheat Stripe Rust in China

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China is one of the biggest agricultural countries as well as the largest wheat producer in the world. Wheat is grown on near 24 million hectares and the total wheat production exceeds 100 billion kg (MOA, 2007). Stripe (or yellow) rust caused by *Puccinia striiformis* f. sp. *tritici* is the most destructive foliar diseases of wheat in many areas around China. Epidemics of the disease have occurred annually since 1950 and the losses of wheat yields due to stripe rust have totalized about 60 billion kg. A great deal of effort has been spent on the survey of regional epidemics, pathogenicity of wheat stripe rust, and strategies for its control in China over the years. It has been found that South of Gansu and Northwest of Sichuan are the most important over-summer areas of *P. striiformis* that act as major sources of inoculum for the autumn-sown wheat in the eastern areas and as a variable zone of rust virulence and wheat cultivar resistance to stripe rust. And a total of 33 formally nominated races (CYR1 to CYR33) and their frequencies have been determined from the stripe rust samples collected throughout the country, of which CYR32 and CYR33 was detected to be the dominant races with the frequency of 30.4% and 12.3% respectively in a total of 5445 isolates tested during 2001-2006. Ecological control of wheat stripe rust in the over-summer areas of the pathogen sources has been considered as the major strategy of sustainable disease control. Effective measures have been put forward and developed as follow in recent years: (1) Improving cultivar resistance and reasonably deploying resistance genes to enhance genetic diversity of wheat rust resistance. A series of improved disease-resistant cultivars (or lines) has been developed by incorporating the resistance genes *Yr3b*, *Yr4b*, *Yr5*, *Yr10*, *Yr11*, *Yr12*, *Yr13*, *Yr14*, *Yr15*, *Yr16*, *Yr17*, *Yr18*, *YrC591*, *YrSp* etc., as well as the durable resistance cultivars and the other resistance materials. DNA molecular markers closely linked or co-segregating with the resistance genes *Yr2*, *Yr5*, *Yr7*, *Yr8*, *Yr9*, *Yr10*, *YrSp*, *YrJu4*, *YrKy2*, *YrC591* and two sets of wheat near isogenic lines with the resistance genes *Yr1*, *Yr2*, *Yr5*, *Yr7*, *Yr9*, *Yr10*, *YrV23*, *YrSp*, *YrKy2*, *YrJu4*, based on the recurrent parents of Mingxian 169 (winter habit) and Taichung 29 (spring habit) respectively, have been successfully developed. (2) Changing cultural practices to raise crop diversity. Maize, upland rice, oil sunflower and other vegetable and forage crops with highly economic value have been introduced and extended, to cut down the wheat plantation in the over-summering areas. Now plastic-mulched maize has been extended to a total acreage of more than 70,000 ha with the economic benefits 2-3 times greater than wheat, leading to a reduced wheat acreage of 40% in the 'hotspots'. (3) Eradicating volunteer seedlings of wheat to cut off the 'green bridge' of inoculum from late-matured wheat to early-sown seedlings. Furrowing deeply more than two times could ultimately control the volunteer wheat after harvest. (4) Regulating wheat planting date and seed dressing with fungicides to reduce the amount of inoculum in the areas of the pathogen sources. Conclusively, establishment of a new agro-ecosystem in the areas of inoculum sources of *P. striiformis* is commended as a more economical and practical approach to the sustainable control of wheat stripe rust epidemics in the whole country of China.

## Wheat yellow rust (*Puccinia graminis* fsp. *tritici*) in Central West Asia and North Africa

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In west Asia, Central Asia and the Caucasus (CAC), and North African (CWANA) countries, wheat stands out for high value of production under irrigation, in higher rainfall and moderate rainfall agro-ecological areas. Monoculture of high yielding varieties is rapidly replacing the traditional lower yielding landrace cultivars. In CWANA countries, wheat yellow rust (*Puccinia graminis* fsp. *tritici*) is among the diseases that present a clear danger to wheat production under irrigation and in the high rainfall areas. Annual yield losses have been recorded in one or more countries and in certain areas have reached epidemic levels. Yield losses of 20 to 80% have been recorded almost annually. Application of fungicides has become a common practice in many countries. Since 1970's yellow has become sporadic due the exploitation of effective resistance genes in different forms and combination. Durable resistance has been linked to a number of genes such Yr18 and some very effective major effective genes such as Yr 9, Yr27; Yr1 that were associated with good parental lines and hence were extensively used by breeding programs globally. As Borlaug said: "rust does not sleep" early 1980's Virulent race affecting the Yr9 resistance gene occurred in East Africa and spread eastward through. Resistance genes Yr27, Yr18, and other Yr-Genes have effectively reduced the impact of yellow rust for over a decade. Recent virulence on Yr27 has been detected in rust trap nurseries. The spread of this new virulence is a typical example of potential risk of wheat rusts. The potential of wheat rust epidemics from new emerging virulent rust races remains a real threat to most wheat producing countries in the world.

## Race changes of *Puccinia striiformis* f. sp. *tritici* and *P. striiformis* f. sp. *hordei* in the United States

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Stripe rust of wheat, caused by *Puccinia striiformis* f. sp. *tritici* (PST), is most frequently destructive on wheat in the western United States and has become more frequently epidemic in the Great Plains and southeastern U.S. states since 2000. Stripe rust of barley, caused by *P. striiformis* f. sp. *hordei* (PSH), has caused damage mainly in the western U.S. since its first report in Texas in 1991. Races of the pathogens have been determined every year from infected leaf samples of wheat and grasses, collected throughout the U.S. PST races were identified on a set of 20 wheat differential genotypes and PSH races were identified on 12 barley genotypes. From 2000 to 2008, a total of 117 races were detected, of which 79 were first detected during this period. The predominant races, which were first detected in 2000, were the group with basic virulences to Lemhi (*Yr21*), Heines VII (*Yr2*, *YrHVII*), Lee (*Yr7*, *Yr22*, *Yr23*), Fielder (*Yr6*, *Yr20*), Express (*YrExp1*, *YrExp2*), AVS/6\**Yr8* (*Yr8*), AVS/6\**Yr9* (*Yr9*), Clement (*Yr9*, *YrCle*), and Compair (*Yr8*, *Yr19*). This race group continues to evolve into new races with additional virulences to differential genotypes, including Chinese 166 (*Yr1*), Moro (*Yr10*, *YrMor*), Paha (*YrPa1*, *YrPa2*, *YrPa3*), Druchamp (*Yr3a*, *YrDru*, *YrDru2*), Produra (*YrPr1*, *YrPr2*), Yamhill (*Yr2*, *Yr4a*, *YrYam*), Tye (*YrTye*), Tres (*YrTr1*, *YrTr2*), and/or Hyak (*Yr17*, *YrTye*). From 2000 to 2003, the predominant races were PST-78 (virulent on wheat differential genotypes Lemhi, Heines VII, Lee, Fielder, Express, AVS/6\**Yr8*, AVS/6\**Yr9*, Clement and Compaie) and PST-80 (the same virulences plus virulence on Produra). In 2004 to 2006, the most predominant race throughout the U.S. was PST-100 (the same virulences as PST-80 plus virulences on Yamhill and Stephens). Starting in 2006, races with the virulences of PST-100 or similar races plus virulence to *Yr1* became predominant in California, and PST-114 with combined virulences of PST-100 and virulence to *Yr10* became predominant in the Pacific Northwest. Over the nine-year period, races with more virulences became increasingly predominant, indicating that races with more virulences have advantages over those with fewer virulences. A total of 82 PSH races have been identified since 1991, of which 30 were first identified after 2000. In contrast to PST, PSH races with few virulences have been predominant. Preliminary evidence has been obtained for somatic hybridization between PST and PSH isolates using virulence tests of stripe rust collections on both sets of differential genotypes and microsatellite markers.

## A new pathotype of stripe rust affecting triticale in Australia

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Stripe rust, caused by *Puccinia striiformis* f. sp. *tritici*, is one of the most important wheat diseases in Australia and can cause extensive yield loss. Low annual yields and erratic rainfall, renders resistance breeding the most cost-effective means of controlling stripe rust. The identification and use of new resistance genes is important as existing genes become ineffective with the occurrence of new virulent pathotypes. Triticale grows superbly in a range of environments normally unsuited for traditional cereals and thus aid in utilizing marginal environments. Disease resistance in triticale has been considered one of its most important features. This advantage however seems to be disappearing with a surge in rust pathotypes attacking triticale varieties worldwide. In 2007 stripe rust occurred on previously resistant triticale varieties in Australia. A new pathotype 134 E16 A+ J+ was identified, resulting in a need for new sources of resistance. This pathotype dominated samples received by the Australian Cereal Rust Survey during the 2008 wheat growing season. It was retrieved from both triticale and wheat varieties. Multipathotype seedling tests and field screening in a collection of varieties identified resistance at both the seedling – and adult plant stage that could be utilized in a breeding program. The presence of seedling resistance genes *Yr9* and/or *YrJ* in some of the varieties have been confirmed. Phenotyping results of F3 resistant x susceptible populations indicated the gene that has been overcome was an uncharacterised single dominant gene. This needs to be confirmed with further population studies. Molecular marker analysis is underway to determine the chromosomal location of the gene involved and to determine linkage relationships associated with other characters of interest.

## Phenotypic Variation Among Leaf Rust Isolates From Durum Wheat In Northwestern Mexico

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Durum wheat in Northwestern Mexico became important when the shift from bread wheat cultivation to durum occurred due to a higher market price as well as its higher level of resistance to Karnal bunt. By 2000, 90% of the wheat area was sown to durum wheat and a single cultivar ‘Altar C84’ occupied 85% of this area. Since the release of Altar C84 in 1984, the most common race until 2001 of *Puccinia triticina* identified from durum wheat was BBB/BN (1). However, in 2001 a new race, BBG/BN (2), was identified and caused susceptibility Altar C84 as well as ‘Atil C2001’, the highest yield potential variety ever released in the region, and almost 80% of the CIMMYT durum wheat germplasm. A second race with unnecessary virulence for *Lr26* was identified in low frequency the same year and designated as BCG/BN (2). ‘Jupare C2001’ which carries the complementary rust resistance genes *Lr27+Lr31* was released in 2001 under emergency and two additional cultivars ‘Banamichi C2004’ and ‘Samayoa C2004’, with resistance genes *Lr27+Lr31* and *Lr14a*, respectively, were released. During 2008, a new variant race that acquired virulence for the adult plant resistance gene *Lr12* and the seedling effective resistance genes *Lr27+Lr31* was identified. Consequently Jupare C2001 and Banamichi C2004 became susceptible. This race was designated as BBG/BP (3). A variant of race BBG/BP with virulence to *Lr3*, designated as CBG/BP, was also identified the same year causing susceptibility ‘Storlorm’ and its derivatives that carried resistance gene *Lr3* (4). Although virulence to *Lr3*, *12*, *26*, *27+31* is known to be present among leaf rust isolates of hexaploid bread wheat, this is the first time that phenotypic variation and evolution towards virulence for these genes among the durum leaf rust population has occurred. Since detection in 2001, race BBG/BN is continually evolving and defeating resistance genes present in both durum and bread wheats.

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## **Wheat brown (leaf) rust in Europe: Studies on disease sensitivity towards azoles and strobilurins and their fungicidal efficacy**

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Wheat brown rust, *Puccinia triticina*, is one of the major diseases threatening successful wheat production in Europe. Whereas *Septoria tritici* is the most relevant pathogen during the early development phases of the crop, *P. triticina* becomes more important in the later stages when environmental conditions are warmer and less humid. Climate change with trends to more humid winters and dryer summers in Northern Europe could increase the relevance of this disease even further. Also the overcoming of the important resistance gene Lr37 in many wheat varieties contributes to an increased threat caused by *P. triticina* as varietal resistance may be less effective. All parameters contributed to a severe rust epidemic in 2007 and caused yield damages of up to 2.8 t/ha. Therefore, to enable farmers to produce on high yield levels under uncertain growing conditions, the availability of resistant wheat varieties and high performing fungicides is essential.

Currently the chemical classes of azoles and QoI fungicides are the backbone of reliable rust control. To be able to predict the future performance of fungicides belonging to these two chemical classes, intensive *in vivo* and molecular biological studies were carried out in order to determine the sensitivity of the European rust population towards fungicides with these modes of action.

As far as azoles are concerned a collection of 180 isolates of *P. triticina* from different European wheat growing regions from 2007 and 2008 were investigated with respect to their sensitivity to the demethylation inhibitor (DMI) fungicide epoxiconazole and alterations in the target gene, *cyp51*, respectively. The *cyp51* gene was highly conserved across Europe. Only in 14 isolates a mutation at codon 134 (Y134F) was identified. The mutation had only a limited impact on the sensitivity of *P. triticina* towards epoxiconazole, if at all. Isolates of *P. triticina* with the highest ED50 values or with the Y134F mutation in the *cyp51* gene were equally well controlled *in vivo* by registered field rates of epoxiconazole as compared to isolates with the lowest ED50 values.

*In vivo* tests were also performed with more than 1000 isolates in 2007 and 2008 for sensitivity monitoring to the QoI F 500®. No QoI resistant isolate was detected in this survey and this was confirmed by genetic analysis of the cytochrome b gene, the target of QoIs, in 180 isolates. Mutations at amino acid positions 129 and 143, which are known to affect QoI sensitivity, were not found. Based on these results a reliable control of European *P. triticina* populations by epoxiconazole and F 500® can be expected at least for the near to mid term future.

# Achieving durable rust resistance in agriculture: from gene to continent and beyond

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Browder (1985) defined resistance as “any genetically determined characteristic of a host plant that in any way limits damage produced by disease”. The most important features of resistance are the level of protection from economic damage afforded, and durability. Durable resistance is “resistance that remains effective when a cultivar is grown widely in environments favouring disease development” (Johnson 1978). There are examples of simply inherited durable resistances (eg. *Lr34/Yr18*, *Sr2*, *mlo*) and of polygenic durable resistances (eg. leaf rust and stripe rust resistance in Pavon 76; William et al. 2007).

The durability of polygenic resistance is often attributed to the increasingly lower probability of plant pathogens to simultaneously acquire virulences matching increasing numbers of pyramided resistance genes. In contrast, the basis for the durability of some single gene resistances is less clear but could involve different molecular mechanisms, as suggested for genes like *Lr34/Yr18* and *Yr36*.

While there are many examples of non-durable major resistance genes (eg. seedling resistance gene *Lr26*, adult plant resistance gene *Lr22b*), there are also examples of genes that “remained effective when present in a cultivar that was grown widely in environments favouring disease development”, and yet eventually succumbed. Stem rust resistance genes *Rpg1* in barley and *Sr31* in wheat were regarded as durable; however, in both cases pathotypes with matching virulence were eventually detected (races QCC and Ug99, respectively). These examples serve as a warning that there can be no guarantee of durability for any resistance source.

It could be generalised that all major gene resistances will eventually prove to be non-durable and should be avoided, and conversely that minor gene resistances are durable. However, Qi et al. (1999) obtained evidence of race specificity for partial resistance to leaf rust in barley, and Rouse et al. (1980) provided evidence for the erosion of slow-mildewing in wheat. The latter authors stated that “the interaction between components of slow-mildewing resistance and parasitic fitness indicate that the resistance could at least to some extent erode over time”. The recent introduction of an exotic pathotype of *P. striiformis* f. sp. *tritici* (*Pst*) into Australia resulted in a rapid and major shift in the Australian *Pst* flora (Wellings 2007). This pathotype is very similar to, or the same as, one occurring in North America and now regarded as being more aggressive (Milus et al. 2006). It has had a significant effect on Australian wheat germplasm that is not related to major gene resistance – and so the question is raised, “has there been an erosion of minor gene resistance?”

Combinations of major resistance genes have been used successfully to control stem rust in both Australia (Park 2007) and North America (Martens and Dyck 1989). In the latter case, Martens and Dyck (1989) concluded that for more than 30 years, relatively few major genes in various combinations, from four genetic sources (Iumillio durum, Yaroslav emmer, Hope and McMurachy), provided control of stem rust in an epidemiological system comprising hundreds of millions of hectares and thousands of billions of rust propagules. Significantly, they also stated that the genetic basis of this widespread control of stem rust was not fully understood. Clearly, the factors that determine the durability of resistance genes when deployed over large areas are complex. There is evidence that genetic “background” can influence durability (eg. the *pvr2<sup>3</sup>* gene in pepper, Palloix et al. 2009; *Lr24* in wheat in Australia, Park et al. 2000), lending credence to the strategy of using reputedly durable sources of rust resistance as “background” resistance, to which other resistance genes are added (McIntosh 1988).

In view of the complexity of host : pathogen interactions, genetic diversity must be seen as a key ingredient in large scale sustained control of plant diseases. It has been argued that even where specific or major resistance genes are used, genetic diversity can be used as insurance against lack of durability and hence as a means of reducing genetic vulnerability (McIntosh 1988). Above all, an understanding of the resistance genes present in cultivars and breeding populations, and monitoring pathogen populations with respect to deployed resistances, are crucial in ensuring that the genetic bases of resistances are not narrowed.

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## **Inheritance of resistance to Ug99 in wheat line Tr129 with an introgression of *Aegilops triuncialis* chromatin.**

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Stem rust of wheat (*Triticum aestivum*), caused by *Puccinia graminis* f. sp. *tritici*, is a major disease worldwide that has mainly been controlled through the use of resistant cultivars. Recently, a new strain (Ug99 or race TTKSK) arose in east Africa with virulence to most *Sr* genes and also most Canadian spring wheat cultivars. Thus, a search was started in 2005 to find new sources of resistance to this highly virulent race. Wheat accessions and cultivars were planted in Njoro, Kenya stem rust nurseries in 2006 and 2007. One line (Tr129) with an introgression from *Aegilops triuncialis* was noted with a high level of resistance (5 to 10% severity, resistant reaction) to Ug99. Thus, a cross was made (RL6071/Tr129) to study the inheritance of resistance in the Tr129 line containing the *A. triuncialis* introgression. The F<sub>2</sub> progeny lines were evaluated at the seedling stage with race TPMK in the greenhouse. The F<sub>2</sub> progeny lines fit a 3:1 (resistant:susceptible) ratio ( $\chi^2=2.1$ , P=0.15), suggesting a single dominant gene conditions resistance in Tr129. The F<sub>2</sub> lines were increased to the F<sub>3</sub> generation and will be evaluated for reaction to TTKSK. The resistance in the Tr129 line is from *A. triuncialis*, which is tetraploid with the CCUU genome and has not previously been used due to the difficulty in transferring disease resistance into the hexaploid wheat genome. The Tr129 line has been tested to numerous North American isolates of *Puccinia graminis* f. sp. *tritici* and found to express high levels of seedling resistance to all isolates. Thus, this line is effective against many races of stem rust and could be very useful in wheat breeding programs as a new source of stem rust resistance. This gene is tentatively designated as *SrAt-1* until genetic and molecular studies are completed to determine the inheritance and chromosomal location of this resistance gene.

## Genetics and mapping of stem rust resistance genes conferring resistance to race Ug99 (TTKSK) in the wheat cultivars Webster, Peace and AC Cadillac

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Stem rust (*Puccinia graminis* Pers. f.sp. *tritici*) of wheat (*Triticum aestivum* L.) has re-emerged as a threat following the evolution of stem rust races with critical virulence, namely Ug99 and its variants. The incorporation of stem rust resistance (Sr) genes into wheat cultivars has historically provided long term disease control. With epidemics of Ug99-type races in Africa and the threat of introduction of these races to other parts of the world, there is a need to identify useful Sr genes to protect wheat crops. Two related Canadian wheat cultivars, Peace and AC Cadillac, are highly resistant to stem rust race TTKSK (Ug99). The accession of the wheat variety Webster maintained at the Cereal Research Centre (Winnipeg, MB, Canada) is also highly resistant to TTKSK. The objectives of this study were to 1) study the inheritance of seedling resistance to stem rust race TTKSK in Webster, Peace and AC Cadillac and 2) genetically map the Sr genes with DNA markers. A population from the cross of the stem rust susceptible line RL6071 by Webster was studied at the F<sub>2</sub> and F<sub>3</sub> generations. The population was inoculated in the F<sub>2</sub> generation with a race native to North America (TPMK) and the F<sub>3</sub> was tested with TPMK and TTKSK. A single recessive gene conferred resistance to TPMK in the F<sub>2</sub> and fit a resistant to susceptible ratio of 1:3 ( $p = 0.66$ ). Webster is known to carry *Sr30*, which is located on chromosome 5DL and is a recessive gene. Genetic mapping with microsatellite (SSR) markers confirmed that *Sr30* conferred resistance to TPMK in Webster. A second dominant gene in Webster conferred resistance to TTKSK but was ineffective against TPMK. This gene, temporarily designated as *SrWeb*, segregated independently of *Sr30* ( $p = 0.33$ ). SSR markers were used to map *SrWeb* to a 14 cM interval on the long arm of chromosome 2B. Based on differential response to races of *P. graminis* and chromosome location, *SrWeb* appears to be different from other Sr genes that confer resistance to TTKSK. However, the possible allelic relationship with other Sr genes on chromosome 2BL is currently unknown. Peace was crossed to RL6071 and the F<sub>1</sub> progeny were used to make a doubled haploid (DH) population. The DH population segregated for a single seedling Sr gene that conditioned resistance to stem rust race TTKSK ( $p = 0.07$ ) and the same gene could be detected with race RTQSC, which is native to North America. AC Cadillac was crossed to the stem rust susceptible line LMPG and was inoculated with TTKSK at the F<sub>2</sub> and F<sub>3</sub> generations. Segregation in the F<sub>2</sub> fit a 3:1 ratio ( $p = 0.19$ ) and the F<sub>3</sub> fit a 1:2:1 ratio ( $p = 0.14$ ). In both the RL6071/Peace and LMPG/AC Cadillac populations, the single seedling Sr gene was mapped to the short arm of chromosome 6D using SSR markers. Furthermore, ALFSD\_RSA, a marker that is tightly linked to the common bunt resistance gene *Bt10*, showed close linkage to the Sr gene on 6DS with a genetic distance less than 2 cM. Peace and AC Cadillac both inherited the Sr gene on 6DS from BW553. BW553 has been used as a parent in several Canadian cultivars as a source of *Bt10* and the incorporation of the Sr gene on 6DS into several of these cultivars was unintentional. The DNA markers identified for *SrWeb* and the Sr gene on 6DS will allow both genes to be selected and included in gene stacks to provide resistance to Ug99 and other stem rust races.

## **Wheat landraces from the USDA-ARS National Small Grains Collection with resistance to new races of *Puccinia graminis* f. sp. *tritici***

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New races of the stem rust pathogen, *Puccinia graminis* f. sp. *tritici*, including TTKSK and its variants, threaten wheat production worldwide. Spring wheat landrace accessions from the USDA-ARS National Small Grains Collection (NSGC) are being screened against the new races at the seedling stage at the ARS Cereal Disease Laboratory (CDL) in St. Paul MN and in the field at the Kenya Agricultural Research Institute at Njoro, Kenya. To guide selection of accessions for Kenya testing, accessions are also being screened with local race QFCS in the greenhouse in Aberdeen ID. This work is part of a coordinated effort to discover and deploy resistance to the new races. There are nearly 10,000 spring landrace accessions in the NSGC and currently about 6000 have been screened against race QFCS and 1761 have been screened in the field in Kenya. Selecting accessions resistant to QFCS in Aberdeen increased the chance of finding field resistance in Kenya fivefold compared to randomly selected accessions. Of those screened in Kenya, twenty-one accessions (1.2%) showed moderate resistance (MR) and another 52 accessions (3.2%) showed a discernable level of resistance with lower infection responses and lower severity compared to susceptible checks. Four accessions showed a higher degree of resistance in repeated field tests: PI 480278, PI 480280, and PI 559962, originating from Ethiopia and PI 436493 originating from Chile. Data for PI 480278 and PI 480280 were very similar in three field experiments in Kenya, ranging from trace R to 15MR-R. In seedling tests, the two lines were highly resistant to TTKSK and TTKST (with infection types 0; to 0) but susceptible to races TTTSK and TRTT. The resistance to TTKSK and TTKST is likely due to *Sr36*. Both accessions were provided to NSGC in the same year by the same donating agency and both are similar for other agronomic descriptors. PI 559962 was resistant to all isolates tested in the seedling trials and showed a high level of resistance in two of three Kenya field tests (25MS; 10R; 1R). PI 436493 was resistant in two Kenya tests (30MR; 10R) and was resistant to all US races tested except TTTT. PI 436493 has not yet been tested against TTKSK and its variants at the seedling stage. There was a tendency for a higher proportion of accessions with some resistance (R, MR, M) originating from Ethiopia (7.2%) and from some countries in Southern Europe (7.5%), including Bosnia and Herzegovina, Greece, Macedonia, Montenegro, Serbia, Portugal, compared to accessions from South America (1.1%) and Western Asia (2.4%). However, these differences were non-significant based on overlap of the 95% binomial confidence intervals due to the relatively small sample sizes. Work continues to screen the remainder of the NSGC spring landrace accessions. Since many landrace accessions are heterogeneous, lines are being derived from resistant accessions and are being re-tested and hybridized for genetic studies to determine if their resistance is due to new genes that have not yet been deployed in modern wheat cultivars.

## Genetical Genomics of Stem Rust Infection Identifies Master Regulators of Defense in Barley

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Quantitative trait locus (QTL) mapping finds statistical associations between genotypes and phenotypes, allowing regions of the genome harboring allelic differences that *cause* variation in the phenotype to be identified; these regions are called QTLs. Transcript abundance of a single gene is a quantitative trait and its regulation can be genetically interrogated. This is often called genetical genomics, or eQTL mapping because the phenotypes in question are the expression of individual genes. In order to characterize the transcriptomic response to stem rust invasion in a genetic context, Barley1 GeneChips were used to measure gene expression in each member of the Q21861 x SM89010 doubled haploid population (QxSM) after treatment with *Puccinia graminis* f. sp. *tritici* isolate TTKS (*Pgt* TTKS), commonly referred to as Ug99. For comparison, global gene expression in the QxSM lines was also measured after mock inoculation as part of the same experiment. By analyzing the changes in genomic distributions of expression Quantitative Trait Loci (eQTL) between *Pgt* TTKS -inoculated and mock-inoculated treatments, major alterations in the regulation of steady-state and inoculation-responsive mRNA levels were uncovered. Notably, five *trans*-eQTL hotspots were identified and appear to regulate the expression of hundreds of inoculation responsive genes scattered around the genome. Interestingly, none of them are associated with the *Rpg5/rpg4* locus at which Q21861 carries an allele that recognizes *Pgt* TTKS. However, one of these *trans*-eQTL hotspots is coincidentally located with an enhancer of *R* gene-mediated resistance that was discovered through a parallel effort to identify QTL alleles that confer resistance to *Pgt* TTKS. The positional identification of *trans*-eQTL hotspots demonstrates that transcriptome-wide induction and suppression of defense genes is tightly coordinated by regulators that are genetically tractable.

## Efficiency Of Specific And Partial Resistance To Wheat Leaf Rust In Bread Wheat Cultivars Grown In France.

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Inoculum of *Puccinia triticina* is regularly present in France, thus maintaining a high level of epidemiological risk. Both fungicides and genetic resistance are currently used to try to control this risk. In particular breeders have maintained a constant effort to breed for resistance to the main wheat diseases, including wheat leaf rust. From an annual survey of pathogen populations, and a postulation of specific resistance genes in the registered cultivars, we have shown strong changes in pathogen population structure, connected to the evolution of resistance genes in the host population. We have shown that this evolution occurs in a clonal pathogen population with a strong correspondence between pathotypes and microsatellite genotypes. However the specific host-pathogen interactions were not sufficient to explain the frequencies of pathotypes : although the widely grown cultivar Soissons (*Lr14a*) was potentially infected by more than 40 pathotypes, a single pathotype largely dominated the leaf rust population on that cultivar. Regardless of the aggressiveness component measured (latent period, spore production per lesion, uredinium size, spore production capacity, lesion life span), this pathotype was repeatedly found more aggressive than at least one of two other pathotypes tested.

This is an example where pathogen population quantitatively adapted to its compatible host, and thus warns against a possible erosion of quantitative resistance. Our objective was then both trying to evaluate this risk, and minimizing it by diversifying sources of resistance. We screened lines and cultivars in the field for sources of quantitative resistance. We evaluated the components of some of these resistance sources in the greenhouse, against the main pathotypes present in France. We have investigated possible specific interactions between pathotypes and our sources of quantitative resistance, and thus evaluated the risk of an erosion of these sources. Identification of the components of our sources of quantitative resistance will allow to propose diversified material to breeders.

## **Screening and characterization of resistance to crown rust (*Puccinia coronata* f.sp. *avenae*) and powdery mildew (*Blumeria graminis* f.sp. *avenae*) in an oat germoplasm collection.**

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An oat germoplasm collection consisting on 141 accessions of *A. sativa* and *A. byzantina* from Spanish origin, together with 32 oat cultivars (*A. sativa*) was screened for resistance against crown rust (*P. coronata* f.sp. *avenae*) and powdery mildew (*B. graminis* f. sp. *avenae*). The study was carried out at seedling stage under controlled conditions. A first macroscopic characterization was performed assessing infection type (IT), infection frequency (IF) and latency period (LP) for rust, and IT and disease severity (DS) for powdery mildew. Accessions showing low levels of macroscopic symptoms were studied histologically to determine the mechanisms responsible for resistance.

All accessions except cv. Kankan displayed a fully compatible reaction (high IT) to rust. Several genotypes showed a reduction in IF and an increase of LP. Histological studies showed a range of defense mechanism, acting alone or combined, at different stages of the infection process. No differences among accessions were found for spore germination. Most of the selected accessions showed a high percentage of early aborted germlings due to penetration resistance through cell wall reinforcement and papilla deposition in the mesophyll cells. In some resistant accessions, papillae were overcome by the fungal penetration peg that successfully reached the cell lumen. However, the hypersensitive response (HR) stopped the fungal development. Finally, in other accessions the HR was not triggered immediately after cell penetration and the fungal pathogen formed a haustorium but a late HR associated with established colonies stopped colony growth and prevented sporulation.

All accessions studied were highly susceptible to the powdery mildew isolate used with high IT and moderate to high DS. Only 23% of accessions showed a moderate reduction in DS. Histologically, a slight reduction in spore germination was observed in some accessions. Penetration resistance of the epidermal cells due to papilla formation was very common among accessions. Also, epidermal cells of several accessions were penetrated by the fungus but then HR leading to cell death hampered fungal development. In all genotypes characterized by HR, this developed fast and no haustorium could be observed in the epidermal cells.

Interestingly, several accessions showed a combination of resistance mechanisms and stopped fungal development at several stages. In addition a few genotypes showed resistance to both, rust and powdery mildew. Coupling resistances affecting different stages of plant-pathogen interaction and/or under genetically complex control would present a series of barriers to pathogenesis. Such resistance should prove more durable than single gene controlled race-specific resistances that, although easily manipulated in plant breeding programs, have repeatedly proved to be ephemeral. We propose these genotypes as possible genotypes for plant breeding programs.

## Regulation of innate immunity in barley-powdery mildew interactions

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Plants have evolved complex regulatory mechanisms to control the defense response against microbial attack. Both temporal and spatial gene expression are tightly regulated in response to pathogen ingress, modulating both positive and negative control of defense. BLUFENSINs, small peptides in barley, wheat, and rice, are highly induced by attack from the obligate biotrophic fungus, *Blumeria graminis* f. sp. *hordei*, causal agent of powdery mildew disease. BLN1 negatively impacts plant defense, is predicted to be secreted, and contains both structural and sequence similarities to knottins, small disulfide-rich proteins characterized by a unique disulfide through disulfide knot. To discern regulatory targets of BLN1, we conducted Barley1 GeneChip analysis of *Bln1*-silenced plants via *Barley stripe mosaic virus*-induced gene silencing (BSMV-VIGS). Sixty GeneChip hybridizations were performed, based on 5 replications of 12 BSMV-VIGS/host-pathogen interactions. Mixed linear model analysis revealed 98 significant new genes ( $p < 0.0001$ ; FDR < 2%) that are suppressed together with *Bln1* (Contig12219\_at;  $p=6.28^{E-06}$ ), or induced when we compare BSMV:Bln1<sub>248</sub> silenced plants to the BSMV:00 control. These candidates appear to have key roles in *R*-gene mediated and innate immunity networks, thus, the functional identification of their precise roles will be a significant step in understanding plant defense.

## The Barley-Rusts and Mildews: Two Models to Study the Molecular Basis of Host-Status of Plants to Specialized Pathogens

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Full nonhost resistance can be defined as immunity, displayed by an entire plant species against all genotypes of a plant pathogen. The genetic basis of (non)host-status of plants is hard to study, since identification of the responsible genes would require interspecific crosses that suffer from sterility and abnormal segregation.

There are some plant/potential pathogen combinations where only 10% or less of the accessions are at most moderately susceptible. Such plant species may be regarded as marginal hosts or near-nonhosts to the considered pathogen, and can provide insights into the genes that determine whether a plant species is a host or a nonhost to a would-be pathogen.

Barley (*Hordeum vulgare*) is an example of a near-nonhost to several rusts (*Puccinia*) of cereals and grasses. By crosses and selection we accumulated susceptibility and developed an experimental line, SusPtrit, with high susceptibility to at least nine different non-adapted rust taxa such as the wheat and *Agropyron* leaf rusts (*P. triticina* and *P. persistens*, respectively). Barley is a full nonhost to the wheat powdery mildew (*Blumeria graminis* f.sp. *tritici*; *Bgt*), although some barley lines have been reported that allow occasional formation of haustoria. We identified such lines, and by intercrosses and selection we developed experimental lines from different parentages, SusBgt-SC and SusBgt-DC, that are susceptible enough to allow development of macroscopically visible colonies by *Bgt*.

We developed barley mapping populations by crossing SusPtrit with two regular, fully resistant barley accessions and are developing such populations for SusBgt-SC and SusBgt-DC as well. These populations allow us to study the inheritance of nonhost resistance of barley to non-adapted rust and powdery mildew fungi. Our observations and conclusions so far include:

- SusPtrit is also susceptible to non-adapted rusts that were not applied to select for increased susceptibility. The SusBgt lines were not particularly more susceptible to other mildew forms than the *Bgt* to which they were selected.
- SusPtrit is still immune to several non-adapted rusts, and SusBgt lines are still immune to nearly all non-adapted mildews, suggesting that the non-host defence mechanisms in these lines are not completely compromised.
- The immunity of barley to non-adapted rusts is based on a large number of QTLs, with overlapping specificities for rust fungal species. Also the immunity to non-adapted powdery mildew fungi seems to inherit polygenically.
- Two cultivars that are immune to non-adapted rusts shared hardly any QTLs for resistance to the same non-adapted rust fungal species, implying the involvement of a high diversity of genes.
- Immunity to non-adapted mildews is mildew form- and mildew-development stage specific.
- There is statistically significant co-localization of QTLs for resistance to non-adapted rust fungi with QTLs for quantitative basal resistance to the adapted barley leaf rust pathogen, *P. hordei*.
- The QTLs for quantitative basal resistance to *P. hordei* and non-host resistance to the non-adapted rust fungi collocate significantly with genes that are implicated in the basal resistance gene network.

Both barley pathosystems are excellent tools to study the genetic basis of natural variation in basal resistance leading to non-host resistance, and so far indicate similar principles for rusts (Basidiomycetes) and mildews (Ascomycetes) that have similar life styles, but are not closely related.

## Efficient targeting of barley genes for basal resistance to *Puccinia hordei*

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Basal resistance has been suggested to be a weak form of non-host resistance, resulting from the partial failure of the microbe to deal effectively with the defence that plant species mount against maladapted microbial intruders. With rust and mildew fungi, basal resistance hampers the formation of fungal haustoria and is due to genes with relatively small, quantitative effects, located on so called quantitative trait loci (QTL).

The barley populations Steptoe/Morex and Oregon Wolfe Barleys (i.e. Dom/Rec) vary quantitatively, without hypersensitive reaction, in their level of resistance to the biotrophic leaf rust fungus *Puccinia hordei*. Each population segregates for a different set of QTLs. The most effective QTL-alleles for resistance that have been detected in seedlings are *Rphq11* in Steptoe and *Rphq16* in Dom. *Rphq11* and *-16* are not effective in adult plants grown in greenhouse or in the field. Steptoe and Dom were crossed and backcrossed with a susceptible barley line and individual F<sub>3</sub> or BC<sub>1</sub> plants were selected that contain the resistance allele of *Rphq11* or *-16* but none of the other resistance QTLs that the donors possess. The effect of each QTL was confirmed in F<sub>4</sub> or BC<sub>1</sub>S<sub>1</sub> families. Large progeny populations were screened resulting in the identification of 114 and 164 recombinants around *Rphq11* and *Rphq16*, respectively. Careful phenotyping of the recombinants allowed refining the position of *Rphq11* to a 0.5 cM genetic interval and the position of *Rphq16* to a 7.4 cM genetic interval. Conserved synteny between barley, rice and *Brachypodium distachyon* at both loci permitted quick saturation of the two QTL intervals with molecular markers.

This strategy allowed a quick fine-mapping of the genes underlying two resistance QTLs without the need to develop near-isogenic lines and required only a handful of molecular markers flanking the QTLs. Subsequently, following a synteny-based approach, markers tightly linked to the QTL-genes were efficiently developed and used to screen the cv. Morex BAC library to identify the physical location of those two QTL-genes on the barley genome. Two of the genes that co-segregate with *Rphq11* are pathogen induced, and their expression levels 18 hours after leaf rust infection are correlated with the level of partial resistance in the Steptoe/Morex population. Those two genes are primary candidates to explain the *Rphq11* effect.

## Nonhost immunity in barley to *Puccinia hordei-bulbosi* is a polygenic trait with multiple components

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When a pathogen challenges a plant to which it is not adapted, what is now considered a heterologous pathogen on a nonhost plant, early and intensive defence responses in the plant are triggered. This leads to a complete type of immunity, so-called "nonhost immunity" which is durable at a time scale that can be observed by humans. Nonhost immunity responses are likely to involve many genes, from both host and pathogen. However, it is hard to investigate the components and inheritance of nonhost immunity in most pathosystems. We developed the barley-*Puccinia* rust model system to study the inheritance and mechanisms of nonhost immunity in barley to heterologous rust fungi. We screened 105 barley accessions in the seedling stage for resistance to *Puccinia hordei-bulbosi* (*Phb*), the leaf rust fungus of the wild barley relative *Hordeum bulbosum*. Our data indicated that only 16% of barley accessions (mainly exotic barleys, landraces and wild barleys) are at the seedling stage somewhat susceptible to *Phb* implying that barley is not a regular host for this rust species. Resistance to *Phb* inherited quantitatively in three mapping populations: Vada x SusPtrit (VxS; Vada is immune), Cebada Capa x SusPtrit (CxS; Cebada Capa is immune) and Oregon Wolf Barley (OWB; Dom x Rec, Dom is immune). QTL analysis based on macroscopic visible infection sites showed that in each mapping population a different set of loci is involved in resistance to *Phb* and only one coincident QTL was found between VxS and CxS populations. QTL analysis in the VxS population using microscopic traits, including colony size, early abortion without necrosis and the percentage of infection units associated with necrotic plant cell(s), showed that each microscopic trait is under the control of a different set of genes, implying that nonhost resistance to *Phb* is a multi-component trait based on a polygenic inheritance. The data presented and previous data collected for others grass rusts – barley interactions did not support the hypothesis that phylogenetic distance between rust fungi may be associated with the host status (or effectiveness of QTLs) of barley to a certain potential pathogen.

**Keywords:** Nonhost resistance. *Hordeum vulgare*. Heterologous rust. *Puccinia hordei*.

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## **SusBgt: Experimental barley lines with susceptibility to wheat powdery mildew as a tool to study non-host resistance**

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The genetic basis of non-host resistance of barley to non-adapted *formae speciales* of *Blumeria graminis* is not known, since there is no susceptible barley line to, for example, the wheat powdery mildew pathogen, *Blumeria graminis* f.sp. *tritici* (*Bgt*). A large collection of barley germplasm was screened and some rare barley accessions were identified with rudimentary susceptibility to the wheat powdery mildew, *Bgt*. Those accessions were intercrossed in two cycles, and resulted in two lines, called SusBgt<sub>SC</sub> and SusBgt<sub>DC</sub>, with exceptional susceptibility to *Bgt* at seedling stage. The quantitative variation among barley accessions and in the progenies after convergent crossing suggest a polygenic basis of this non-host resistance. Both lines show a remarkable level of haustorium formation and colony development by the non-adapted target mildew (*Bgt*). The SusBgt lines and their ancestor lines also allowed haustorium formation and conidiation by four out of seven tested non-adapted *Blumeria* forms. Component analysis of the infection process suggested that non-host resistance factors are *Blumeria*-form and fungal development stage specific. Resistance to establishment (first haustorium), colonization and conidiation are not clearly associated. In general, in 30 to 60% of the haustorium forming infection units the pre-haustorial resistance was backed-up by a hypersensitive reaction. One *forma specialis*, from *Hordeum murinum*, induced extensive hypersensitivity upon haustorium formation in some tested lines. The developed lines will serve to elucidate the genetic basis of non-host resistance in barley to wheat powdery mildew, and are useful tools in gene expression and complementation studies on non-host resistance.

## The molecular basis of broad-spectrum powdery mildew resistance

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Loss-of-function mutant alleles of the barley *Mlo* locus are known to confer durable, broad spectrum resistance against the powdery mildew disease caused by the Ascomycete *Blumeria graminis* f.sp. *hordei*. This type of antifungal immunity has been discovered 65 years ago and has been widely used in European agriculture for more than 25 years. We recently showed that powdery mildew resistance conferred by *mlo* alleles is not restricted to barley, but also occurs in Arabidopsis, tomato and pea. The molecular basis of this unusual type of disease resistance remains, however, mysterious. We exploit the genetic and molecular tools available for the dicot reference species, *Arabidopsis thaliana*, to get insights into the molecular mechanisms leading to *mlo* resistance.

## Antagonistic behavior of the two players of ubiquitylation process in disease resistance

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Ubiquitin/26S proteasome controlled protein degradation machinery is responsible for the regulation of many cellular activities in plants, such as developmental processes. In recent years, its importance in biotic stress responses is better realized, due to the findings of the involvement of E3 ubiquitin ligases in disease resistance mechanism. Ubiquitylation process is initiated with the activation of ubiquitin and a chain of multi-ubiquitin is transferred to the protein which is destined to 26S proteasome for degradation *via* the consecutive functions of ubiquitin-conjugating enzyme (E2 complex) and ubiquitin-ligase (E3-complex). E3 ubiquitin ligase selects appropriate candidate proteins for degradation, thus In our study, previously detected differentially expressed *Rad6* (E2) and ZTL type *F-box* (E3) genes upon yellow rust inoculations of wheat by DDRT-PCR analysis were silenced using virus induced gene silencing method with Barley Stripe Rust Virus to assess the functions of genes in plant resistance or susceptibility processes. We have observed HR and no development of fungal hyphae on the *Rad6* silenced susceptible barley line when inoculated with a virulent powdery mildew race. On the other hand, when *F-box* gene was silenced on the resistant barley line, the hyphal formation occurred upon avirulent mildew inoculations. In conclusion, *Rad6* is found to be a negative regulator and conversely, *F-box* is a positive regulator in barley and powdery mildew interactions. It can be hypothesized that this specific *F-box* protein is needed for the degradation of an unknown plant or pathogen target for resistance. The identification of this target protein would be important to fully understand these underlying processes.

## EST-Based Multiplex Gene Expression in Yellow Rust Infected Wheat Using GenomeLab GeXP Genetic Analysis System

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Wheat yellow rust caused by *Puccinia striiformis* f. sp. *tritici* is an important disease of wheat in many parts of the world. In Turkey, yellow rust could affect wheat (*Triticum aestivum* L.) production on approximately 7.4 million hectares acreage (%80) of Turkey's 9.3 mha, especially in the cooler, humid regions of Central and Eastern Anatolia. The analysis of Expressed Sequence Tags (ESTs) is one of the most important techniques used to study gene expression. Vast amounts of EST data are now available, and the volume is growing rapidly. A total of 1.080.266 ESTs from different libraries are present in the EST database for wheat ([http://www.ncbi.nlm.nih.gov/dbEST/dbEST\\_summary.html](http://www.ncbi.nlm.nih.gov/dbEST/dbEST_summary.html)). Analysis of these EST data, holds tremendous value for the study of gene expression. In the study, the GenomeLab GeXP Genetic Analysis System was used for EST-based multiplex gene expression analysis of yellow rust infected susceptible and resistant wheat varieties. Previously, 1549 ESTs from yellow rust infested *Triticum aestivum* cDNA library (TA117G1X) were chosen from [http://wheat.pw.usda.gov/cgi-bin/westsql/est\\_lib.cgi](http://wheat.pw.usda.gov/cgi-bin/westsql/est_lib.cgi) website. 136 contigs from ESTs were assembled. Using blastX algorithms in the NCBI, 39 contigs and 96 singletons matched with *Triticum aestivum* proteins which were assigned to 8 functional groups namely protein synthesis, photosynthesis, metabolism & energy, stress proteins, transporter proteins, protein breakdown & recycling, cell growth & division and reactive oxygen scavengers. Only stress and stress related contigs and singletons were selected for multiplex gene expression analyses and subjected to GenomeLab GeXP Genetic Analysis System for primer designing. We designed 9 contig and 20 singleton GeXP primers and they were assembled in a three multiplex. Applied gene expression profiling analysis on RNA samples from yellow rust infected and control plants, conditions that promote disease resistance mechanisms performed between 4 time points (0h, 8h, 12h and 48h). Six bread wheat genotypes (*Triticum aestivum* L. cv. PI178383, Izgi01, Sönmez2001- known yellow rust resistant cultivars and *Triticum aestivum* L. cv. Harmankaya99, ES14, Aytın98- known yellow rust susceptible cultivars) were used for the gene expression analyses. Identification of significant expression profiles of EST-derived contig and singletons in resistant and susceptible genotypes are currently under investigation. In conclusion, the EST approach combination with multiplex quantitative PCR is a powerful strategy for discovering new genes and examining gene expression profiles between control and treated organisms.

## **Effect of different resistance mechanisms to crown rust (*Puccinia coronata* f.sp. *avenae*) and powdery mildew (*Blumeria graminis* f.sp. *avenae*) on oat stomatal conductance.**

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In a previous work, we observed in barley genotypes infected by *Blumeria graminis* f. sp. *hordei* that the expression of resistance affected stomatal behaviour and hence plant physiology (1). The highly effective papilla-based resistance had only a temporary effect on stomatal function while HR caused a dramatic and permanent stomatal dysfunction resulting in the plant inability to respond to drought stress by stomatal closure. In this work, we studied the relationship between stomatal behaviour and crown rust and powdery mildew development in several oat accessions susceptible or bearing different resistance mechanisms acting at several stages of the infection process.

In relation to rust infection, in those genotypes able to arrest the infection process before stoma penetration, limiting germ tube elongation and appressorium or substomatal vesicle formation no changes were observed in stomatal conductance. However, genotypes with mesophyll penetration resistance against *P. coronata* showed a decrease in diurnal stomatal conductance similarly as we observed following epidermal penetration resistance to powdery mildew. This effect was more pronounced in genotypes with higher percentages of early abortion of colonies. In these genotypes, stomata were able to open and close according to circadian rhythms. Genotypes with HR showed a stomatal dysfunction, with an increase of stomatal conductance during day and night although no lock-open was observed during the time course assessed.

Genotypes with epidermal papillae-based penetration resistance against *B. graminis* f.sp. *avenae* showed a stomatal dysfunction. These genotypes were characterized by a lower diurnal stomatal conductance as observed in barley, but no changes were observed in conductance during the dark periods when compared to uninoculated plants. This dysfunction appeared transient since from 101 hours after inoculation differences with controls were not significant. However, dysfunction lasted longer than the observed in barley genotypes. An increase of stomatal conductance in darkness was observed in genotypes characterized with HR, but no changes were observed in diurnal stomatal conductance. The data suggest stomata became locked open as observed in barley.

Lost of stomatal control has obvious deleterious consequences for the control of carbohydrate synthesis and metabolism and the ability of plants to withstand drought. This work shows the importance of studying the effect of the different resistance mechanisms in different pathosystems in order to improve the performance of the different cultivars in the field.

(1) Prats *et al.*, 2006. *J. Exp. Bot.* 57: 2211-2226

## ***Mlsp* confers semi-dominant, developmentally-dependent resistance to barley powdery mildew**

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*Blumeria graminis* f. sp. *hordei* (*Bgh*) is the causal agent of powdery mildew on barley, whose resistance is mediated by several resistance (*R*) genes distributed at loci throughout the genome (e.g. *Mla*, *Mlg*, *MILa*). To date, 102 distinct resistances have been identified, mapping to at least 15 independent loci, with no map position known for 32 of them. Aside from the susceptibility factor *Mlo*, only six recessive resistance genes have been identified in barley. Nearly all of these were identified prior to the advent of molecular markers, and their map positions still remain unknown. We have identified a semi-dominant, developmentally-dependent resistance gene in the barley cultivar Steptoe and have named it *Mlsp*. Interestingly, *Mlsp* is located near *mlt* on chromosome 7HS, the only other recessive resistance gene that has been mapped.

For this work, we used barley cultivars Steptoe, Morex and their doubled haploid progeny. Our early phenotyping of Steptoe and Morex had yielded little to no resistance to *Bgh* isolates 5874 and CC148 when they were inoculated at the seedling stage (7 days after sowing, glasshouse grown). This was in stark contrast to results from *Bgh* CC148 inoculation of adult plants that showed a strongly resistant infection phenotype for Steptoe with no visible signs of infection other than considerable amounts of necrosis. Under controlled conditions in a growth room, a time-course gradient of seedlings was generated by planting every day for five days, with inoculation performed six days after the final planting, such that plants were inoculated at 6, 7, 8, 9, and 10 days after sowing (DAS). This series captures the developmental series of the first seedling leaf, from emergence (PO:0007049) to fully unfolded, but just prior to second leaf emergence (PO:0007094). Seedlings of Steptoe progressively became more resistant in this series, with a decreased variability in their reaction to powdery mildew. Across this series of conditions, Morex showed consistent susceptibility, with little variation in its response. From this parental time-course analysis, we selected the developmental time point of 8 DAS for *Bgh* CC148 inoculation of the SxM doubled haploid (DH) population for QTL mapping.

QTL analysis was performed on four replications of infection type data collected from the SxM DH population at 7 days after inoculation (15 DAS) with *Bgh* CC148. We observed bimodal phenotypic distributions among the 139 DH progeny used, with infection phenotypes ranging from 0 to 3.5 on a scale of 0 to 4. A genetic map of 512 non-redundant transcript-derived markers with a total length of 1017 cM was used for mapping. The bimodality of the phenotypic data is squarely accounted for by *Mlsp*, which behaves as a Mendelian factor for resistance. However, three minor-effect QTL were also detected, explaining additional phenotypic variation. The resistant class of lines appeared more phenotypically variable, so we performed QTL analyses on this subpopulation. The minor-effect QTL were confirmed, and have stronger effects in this analysis, indicating that their full contributions epistatically depend on the presence of the *Mlsp* allele from Steptoe, which specifies resistance.

Analysis of SxM F2 plants revealed that the Steptoe allele of *Mlsp* acts semi-dominantly, with some variation in its action among experimental trials. Previously, Honecker found environmental dependence in field trials: under low light and temperatures 15 – 25°C, Nigrate (*mlni*) showed recessive resistance, while in high light and temperatures 25 – 35°C, its resistance acted semi-dominantly. The specific effects of light and temperature variation are currently under investigation for *Mlsp*.

## Clonality and recombination footprints in wheat yellow rust genetic structure

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Parasitic species can exhibit sophisticated life cycles, displaying complex transmission strategies based on the use of specific propagules and the infection of different host species. Uredinales are characterised by one of the most complex life-cycles in fungi, with the differentiation of five kinds of spores, and with the sexual and asexual cycles occurring on two distinct and unrelated hosts: an aecidial and a telial host, respectively. In wheat, three uredinale species are of major agronomic importance due to the epidemics caused by their asexual cycle: *Puccinia striiformis* fsp *tritici* (PST), *P. triticina* and *P. graminis* (respectively yellow, brown and black rust). Aecidial hosts and sexual cycles are characterised for brown and black rusts, while the alternate host of yellow rust is still unknown. This lack of a known aecidial host has been used to explain the clonal evolution of some PST populations. Recent molecular studies performed at different scales on PST have confirmed the clonality of populations in some areas, while recombination has been detected in other parts of the world. Using different results we obtained on the genetic structure of PST populations and recent experiments performed on the diversity of teliosore production, we defend the hypothesis of the presence of sexual recombination in PST, against the alternate hypothesis of parasexual recombination.

## **Dissecting the contributions of specific and partial resistance to yellow rust in UK wheat germplasm**

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Yellow rust (YR) virulence monitoring in the UK has been carried out continuously since the late 1960s by the UK Cereal Pathogen Virulence Survey by collection and pathotyping of field isolates from disease observation plots, untreated field trials and disease samples sent by growers. Representative isolates are used in inoculated field trials to measure the Adult Plant Resistance (APR) of candidate Recommended List cultivars. In its 40 years of existence, the survey has accumulated detailed resistance data on every major variety of wheat grown in the UK at both seedling and adult plant stages, recorded the emergence of previously unseen virulences on newly introduced specific resistances, and been a source of timely information and isolates to growers, agronomists and breeders.

We aim to utilise this unique historic data resource to analyse the genetic architecture of resistance to yellow rust using an association genetics approach. To this end, we have assembled APR and pathotype data relating to a total 311 varieties tested since 1990 into a new relational database and genotyped the full panel with DArT markers. Preliminary analyses on a subset of 100 lines suggest that levels of LD in the panel are sufficient to allow detection of associations to major loci with markers 1-5 cM distant. Analysis of associations with APR to YR is ongoing and we will present an update on results from a genome-wide association scan.

In a complementary approach to the association genetics study, we are undertaking genetic analysis of the as yet undefeated YR resistance found in cultivar 'Cadenza' in the 'Avalon' x 'Cadenza' doubled haploid (DH) population. A single major QTL for resistance at the seedling stage was observed and tests are underway to confirm that this major seedling resistance locus does indeed explain the APR phenotype. Lastly, a new population has been developed recently to permit genetic analysis of the unmapped, but recently defeated resistances found in cultivars 'Robigus' and 'Solstice'. The results from this series of experiments are expected to provide helpful insights and vital tools to wheat breeders to enable them to design rational genetic strategies for protection of their wheat varieties against YR.

## Characterization of adult-plant resistance in soft red winter wheat to stripe rust

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Stripe rust (yellow rust), caused by *Puccinia striiformis* Westend. f. sp. *tritici* Eriks., has been an important disease of soft red winter wheat in eastern United States since 2000 when a new strain of stripe rust with virulence on resistance gene *Yr9* and enhanced aggressiveness and adaptation to warmer temperature replaced the old strain. Adult-plant resistance appears to be important for protecting cultivars from stripe rust. The objective of this research was to characterize the adult-plant resistance to stripe rust in contemporary wheat cultivars and breeding lines.

Seedlings of 50 lines with low to moderate levels of stripe rust in fields infected with the new strain were evaluated for resistance to races PST-3 (isolate AR90-01) and PST-100 (isolate AR00-05) that are representative of the old and new strains, respectively. Nineteen lines that were susceptible to both races and one line with *Yr9* that was susceptible to PST-100 but resistant to PST-3 in seedling stage were selected for adult-plant experiments. To determine the effect of race and post-inoculation temperature of the expression of adult-plant resistance, the 20 lines were inoculated with each race at heading stage and incubated in growth chambers at low (10 to 18°C) and high (12 to 28°C) temperature regimes. To characterize resistance, the time from inoculation to the first sporulation was recorded on flag and flag-1 leaves to estimate latent period, and the infection type (0 to 9 scale) and percentage of leaf area diseased were recorded 21 days after inoculation on flag and flag-1 leaves. To characterize resistance in the field, the same 20 lines were planted in two fields at University Farm in Fayetteville during October 2008. The experimental design in each field was a randomized complete block with six replications. At jointing stage, one field was inoculated with PST-3, and the other field was inoculated with PST-100. Lines were rated for infection type and percentage of total leaf area diseased.

Preliminary analysis of the results showed that the line x race interaction was significant ( $P < 0.05$ ) for all variables, indicating that the adult-plant resistance in some of the lines is race specific. Lines with race specific resistance were more resistant to PST-100, indicating that the new strain lacks virulence on at least one adult-plant resistance gene for which the old strain is virulent. The temperature x race interaction was significant only for latent period and percentage of leaf area diseased on flag leaves. Both races caused approximately twice as much disease at low temperature than at high temperature, and the significant interaction appeared to be due to a difference in magnitude between the two races. The latent period for race PST-3 was similar at both temperatures, but the latent period for race PST-100 was significantly shorter at high temperature. A complete analysis of the data will be presented at the conference.

## Molecular mapping of a new gene for resistance to stripe rust in durum wheat PI 480148 and transfer the gene into common wheat

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Stripe rust, caused by *Puccinia striiformis* f. sp. *tritici* (*Pst*) is one of the most damaging diseases of wheat worldwide. It is essential to identify new genes for effective resistance against the disease. Durum wheat germplasm has excellent resistance to stripe rust, but not many genes for resistance have been identified from durum wheat genotypes. Durum wheat PI 480148, originally from Ethiopia, was resistant in all seedling tests with several US races under controlled greenhouse conditions and at multiple locations under natural infection of the pathogen for four years. To map the resistance gene(s) in the genotype and transfer it into common wheat, a cross was made between PI 480148 and susceptible common wheat genotype Avocet Susceptible (AVS). Resistant F<sub>3</sub> plants with 42 chromosomes were selected through Feulgen-staining of root tip cells and testing with *Pst* races. When tested with PST-100, the most predominant race in the US for the last six years, 157 F<sub>4</sub> plants from a single F<sub>3</sub> hexaploid plant segregated into a 3:1 ratio for resistant to susceptible plants, which identified a single dominant gene from PI 480148. Using the F<sub>3,4</sub> population and the resistance gene-analog polymorphism (RGAP) and simple sequence repeat (SSR) markers, the gene was mapped to the long arm of chromosome 2B. An RGAP marker and a SSR marker (*Xwmc441*) were closely linked to the resistance gene with a genetic distance of 2.7 and 5.6 cM, respectively. The effective resistance of the gene to an Australian isolate virulent to *Yr5*, which is located on 2BL and resistant to all *Pst* races in the US so far, indicated that the gene is different from *Yr5* and should be a new and useful gene for resistance to stripe rust. Resistant common wheat lines with plant types similar to AVS were selected for use in breeding programs to develop common wheat cultivars with the resistance gene. This approach is currently used to identify and transfer different genes for effective resistance from a number of durum wheat genotypes to a common wheat background.

## Identification and Potential Use of DNA Markers for Yellow Rust Disease Resistance in Wheat (*Triticum aestivum* L.)

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Yellow rust disease caused by *Puccinia striiformis* f. sp. *tritici* is a serious problem for wheat production in Turkey as well as in many parts of the world. DNA-based molecular markers are powerful tools in Marker-Assisted Selection (MAS) and provides plant breeders to practise indirect selection for the resistance genes in a disease free environment and pyramid the resistance genes in the absence of distinguish virulences in wheat. In our ongoing project, Bulk Segregant Analysis (BSA) was used to identify molecular markers associated with yellow rust disease resistance. Resistant and susceptible DNA pools, both for seedling and adult plant stage, were constructed from the selected F<sub>2</sub> individuals from the crosses of İzgi2001 (resistant parent) x ES14 (susceptible parent), PI178383 (resistant parent) x Harmankaya99 (susceptible parent), Sönmez2001 (resistant parent) x Ayın98 (susceptible parent). Polymorphism was tested with 890 primers (SSR, EST-SSR, RGAP, RAPD, STS, AFLP, ISSR, EST-derived contig and singleton). Among these, one SSR marker (*Xgwm 382*) and one EST-SSR marker (*Pk54*) which were identified in F<sub>2</sub> populations of İzgi2001 x ES14 and PI178383 x Harmankaya99 crosses. All F<sub>2</sub> individuals contributing to the resistant and susceptible bulks were tested separately. To investigate the usefulness of the markers *Xgwm382* and *Pk54* for MAS, DNA of 108 additional wheat genotypes were amplified using primer pairs of these two markers. 81% of the wheat genotypes known to be yellow rust resistant had the 118 bp fragment revealed by the XGWM382 primers, 68% of resistant wheat genotypes had the 125 bp fragment revealed by the PK54 primers suggesting that the presence of this marker correlates with yellow rust resistance in diverse wheat germplasm. Interestingly, *Xgwm382* was also shown to be linked to the loci conditioning leaf rust (*Lr50*) and powdery mildew (*Pm4b*) resistances in wheat. Therefore, these markers could readily be used for marker assisted selection to identify multiple fungal resistances genotypes in wheat breeding programs.

## **Molecular mechanism of wheat and stripe rust interaction and functional characterization of resistance-related genes**

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Stripe rust is a devastating disease on wheat worldwide. The stripe rust fungus, *Puccinia striiformis* f. sp. *tritici* (Pst), does not have known sexual reproduction, which makes it impossible to study the mechanisms of the host-pathogen interactions through the classic genetic approach. The objectives of this study were to elucidate the expression profiling of wheat-Pst interactions at the transcriptome level, along with identification and characterization of the resistance-related genes.

Expression profiles of wheat cv. 'Suwon 11' infected by Pst pathotypes CY31 and CY23 that produced compatible and incompatible reactions, respectively, were analyzed using the cDNA-AFLP technique, followed by isolation of the differentially expressed transcript derived fragments (TDFs). A total of 54,912 and 52,992 TDFs were detected using 64 primer combinations for compatible and incompatible interactions, respectively, of which 2,306 TDFs (1,340 up-regulated, 966 down-regulated) in the compatible interaction and 2,437 TDFs (1,787 up-regulated, 650 down-regulated) in the incompatible interaction were differentially expressed. We cloned 186 TDFs from the compatible interaction and 255 TDFs from the incompatible interaction. Based on the GO classification system, the majority of the TDFs with known functions in each interaction were found to be mainly involved in basal metabolism, signal transduction and disease/defense. About 60% of the TDFs from the compatible and 56% from the incompatible interactions were found to encode proteins with unclear functions or do not have significant homologies with genes in the GenBank database.

Taking unigenes from the incompatible interaction as the query, BLASTN analyses were used to search genes with similar functions in the compatible interaction database. The comparison showed that 161 of the 255 sequences from the incompatible interaction were overlapped with TDFs from the compatible interaction. Proteins encoded by these common genes were involved in energy and metabolism, signal transduction and disease/defense, protein metabolism, etc. Expression patterns of more than 80 TDFs revealed by the qRT-PCR analysis were similar to the results obtained from the cDNA-AFLP experiments. Moreover, the most of the genes were induced in both compatible and incompatible interactions, but the expression levels and timing during the infection process were different. Therefore, we can conclude that differentially expressed genes with various functions are mostly similar in both compatible and incompatible interactions, and the expression level and timing of these genes determine the resistance phenotype.

We selected 16 kinase-encoding TDFs for further functional characterization using the BSMV-VIGS technique. Positive silencing for each of the genes could be detected using the qRT-PCR analysis. Wheat plants with two genes silenced had altered phenotypes. Histological observations confirmed the results.

The results of this study greatly enhance our understanding of molecular mechanism of wheat-Pst interactions. The techniques proven in this study will be useful for determining functions of wheat and Pst genes involved in the host plant and pathogen interactions.

## **Global Cereal Rust Monitoring System – prospects and progress**

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The identification of the Ug99 lineage of stem rust in Uganda in 1999 has challenged the assumption that stem rust was a conquered disease. Screening of global wheat germplasm has revealed a very low frequency of resistant varieties, and 80% or more of the world's wheat is now considered stem rust susceptible. Ug99 has sparked a global effort by wheat scientists to counter the threat and has highlighted the need for effective surveillance and monitoring systems. Outside of a few developed countries, monitoring efforts are often irregular or even non-existent and no coordinated global surveillance effort currently exists. Ug99 has provided the impetus to implement a global surveillance and monitoring system that provides relevant and timely information as a global public good.

The Global Cereal Rust Monitoring System is being implemented by the UN Food and Agriculture Organization (FAO) in collaboration with International Agricultural Research Centers (CIMMYT and ICARDA), advanced research institutes and national agricultural program partners. The existing, long-running and successful FAO Desert Locust monitoring and early warning system is being used as a conceptual model system for the Global Cereal Rust Monitoring System. In the initial phase, rust monitoring efforts are focused largely on stem rust and the Ug99 lineage in particular. Given the immediate concern of the threat posed by this stem rust lineage, such an approach is entirely justified. However, the other rusts, i.e. leaf rust and stripe rust, cannot, nor should, be excluded from a Global Cereal Rust Monitoring System at a later stage. The current status of the different components of the emerging Global Cereal Rust Monitoring System will be described, including an update on factors such as; current status of Ug99 or derivatives, survey networks and capacity of partners, data flows, tools and information products. Advances and emerging issues in the development of the Global Cereal Rust Monitoring System will be highlighted. In addition, future prospects, directions and key activities will be outlined.

## Stripe Rust Epidemics in Australia: Implementing National Integrated Disease Control Strategies

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Wheat stripe rust (caused by *Puccinia striiformis* f. sp. *tritici*, *Pst*) was first recorded in Australia in 1979 and became endemic to the eastern Australian wheat zone causing serious losses in the mid eighties. Concerted pathology and breeding research and development combined with industry adoption of resistant varieties resulted in minimal losses for nearly 20 years. The first report of stripe rust in Western Australia in 2002 was the result of a new foreign pathotype incursion. This aggressive pathotype widened its distribution in following years to encompass the entire Australian wheat production zone, and caused serious losses including increased annual fungicide expenditure ranging from \$AUD40-90million. The stripe epidemic in eastern Australia in 2008 was the most intensive in the 30 year history of the disease in Australia.

The dynamics of host resistance and pathogen variability requires a close connection between extension and research staff in order to maximize the available resources of host resistance and fungicide availability. This paper presents details of epidemic development of *Pst* under Australian conditions, the interplay of variety resistance and pathogen population dynamics, and the strategic use of fungicides. These strategies need to be developed and applied in an Australian dryland wheat production context that is characterised by relatively low yield, driven by concerns for minimising risks associate with variable input costs, and the need to capture maximum yields in favourable seasons.

## Sensitivity of *Puccinia triticina* races to fungicides

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Several diseases attack wheat (*Triticum aestivum* L.) crop resulting in yield reduction. Among them is leaf rust caused by *Puccinia triticina* Eriks. considered one of the main diseases causing damage up to 63%. Preferential control measures of this disease include the use of resistant cultivars and chemical control. From 2004/2005 growing season wheat growers and farm advisers notice lack of control of leaf rust by some fungicides traditionally effective against the disease. In experiments carried out in greenhouse, four races of *P. triticina* (MCG-MN, MDT-MR, MFP-CT and MDP-MR) isolated from wheat farms with lack of control were tested for their sensitivity to different fungicides. Triazole and strobilurin fungicides and their respective mixtures were tested. Chemicals in recommended rates—were applied preventive, curative and eradivative in relation to time uredospores were inoculated on wheat plants. Race MCG-MN was sensitive to triazoles, while races MDT-MR, MFP-CT and MDP-MR showed sensitivity reduction. The three less sensitive races to triazoles were sensitive to strobilurins and to the triazole-strobilurin mixtures at the different times of fungicide application. To assess disease progress under field conditions and fungicide effects, the chemicals were applied at the end of tillering, flowering and milky kernel stages of the wheat cultivar Safira. The lowest progress rate of leaf rust was obtained for the triazole-strobilurin mixtures, strobilurins and the highest for the triazoles applied alone. Fungicides EC<sub>50</sub> and EC<sub>90</sub> were determined in greenhouse for five races (MCH-MN, MDT-MR, MDK-MR, MDP-MR e MFH-HT) highly frequents in the 2007 growing season. Triazole fungicides were tested in the following concentrations in mg.L<sup>-1</sup> of water: 0.0; 0.02; 0.2; 2.0; 20.0; 100.0 and 200.0 and for the strobilurins concentrations of 0.0; 0.0001; 0.001; 0.01; 0.1; 1.0 e 10.0 mg.L<sup>-1</sup> preventively sprayed. The lowest EC<sub>50</sub> values for triazole fungicides, 0.33 to 0.91, were determined for race MCG-MN (sensitivity) and the highest, 9.63 to 85.64, for races MDP-MR, MDT-MR, MDK-MR, MFH-HT (lower sensitivity). EC<sub>50</sub> for strobilurin fungicides varied from 0.0018 to 0.14. It may be inferred that the control efficacy of wheat leaf rust may be due to strobilurins fungitoxicity which resulted in better control and higher grain yield. It is suggested that the triazole fungicides should not be used alone in susceptible cultivars to races with reduced sensitivity.

## Biological control of wheat leaf rust using *Pseudomonas fluorescence* and *Bacillus* spp

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The efficiency of *Pseudomonas fluorescent*, *Bacillus polymyxa* and *Bacillus circulans* and their mixture were evaluated as bioagents on leaf rust disease of wheat cultivar Giza-139 caused by *Puccinia triticina* f.sp *tritici*. In greenhouse the bioagents were used as suspension for soaking wheat grains (24h before sowing) and for spraying seedlings (seven days old) with each bioagents singly or mixture of them. Experiments were carried out on both seedling and adult stages. In greenhouse experiments, the incubation period was increased from 8 days (in control plant) to 10.3 days when wheat seedlings were sprayed with *Pseudomonas fluorescence*, 72 hours after inoculation. Biocontrol agents used in this study caused significant decrease in No. of pustules/leaf. The inhibition effect of bioagents in reducing disease components extended to the significant reduction in infection type, compared with the control. The highest percentage of disease reduction (54.02%) was obtained by soaking seeds in *Bacillus circulans* followed by *Pseudomonas fluorescence* (48%). However, in field experiments, *Pseudomonas fluorescence* significantly reduced rust severity leading to an increase in weight of 1000 kernel as well as yield plot, compared with the control.

Keywords: *Puccinia triticina*, LEAF RUST, PGPR, wheat .

## New Tools for Comprehensive Evaluation of Virulence and Resistance Data

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VAT (Virulence Analysis Tool) is a user-friendly software package for diversity analysis of binary data. It facilitates a comprehensive, effective and logically consistent evaluation and presentation of virulence and resistance data. VAT is also applicable to molecular marker data according to Kosman and Leonard (2005), and allows for the analysis of sexually and asexually reproducing populations. The package offers the following features:

-Basic routine steps of data entry, their transformation and dichotomization, identification and listing of phenotypes in various common race nomenclatures.

-Characterization of pathogen/host samples by phenotype distribution, virulence/resistance frequencies and complexities, associations, diversities and distances etc.

-Re-sampling based estimates of means and variances for different population parameters.

-Compatible output for additional analyses (e.g. PCA, clustering, or dendrograms), and transfer of information to and from Excel.

The VAT provides the following applications: “Data entry”, “Resampling and Coding”, “Descriptive Statistics”, “Inferential Statistics” and “Miscellaneous”.

**Data entry.** This application allows creating/modifying differential sets; entering new data in different formats (binary, any numerical assessment scale, and octal, hexadecimal and binary-decimal race codes); access to existing data files (system-generated “.vat” or conventional “.txt” files); and data editing and validation.

**Resampling and Coding.** The “coding” part deals with dichotomization of original data (conversion of non-binary data into binary vectors of zeros and ones), which is a necessary step for further analysis. It also handles translation to and from the octal, hexadecimal, and binary-decimal race codes. The “re-sampling” part generates random samples from the original (dichotomized) data set, which is an indispensable precondition for any inferential statistics in VAT. Number and size of the generated pseudo-samples are set by the user.

Two statistical applications of VAT (“Descriptive Statistics” and “Inferential Statistics”) provide a broad range of over 50 approaches for data analysis. Relevant parameters can be calculated based on three commonly used dissimilarity measures (Simple mismatch, Jaccard, Dice).

**Descriptive Statistics.** Individual and population parameters are derived from the dichotomized form of the original data.

**Inferential Statistics.** Calculations are performed on computer-generated pseudo-random samples drawn independently with replacement from the dichotomized original observations. This renders the possibility of inferential conclusions (e.g. statistical significance or confidence intervals to sample parameters).

New and commonly used concepts of diversity analysis (Kosman and Leonard 2007) are provided in both statistical modules, which are subdivided into “within” and “between” population sections.

**Within** is comprised of analyses that are applied separately to each population, allowing characterization of its phenotypes (frequency, virulence complexity and pair-wise dissimilarity), its virulences (frequency and pair-wise associations), and within-population diversity parameters (14 different indices).

**Between** provides analyses among populations comparing phenotypes of any two populations (pair-wise dissimilarities), displaying common phenotypes and virulence frequencies for all included populations, and calculating between-populations distances (11 different indices).

**Miscellaneous** (under development). This application will include a number of supplementary features for diversity analysis.

VAT provides a user, at any step, with automated support and guidance by the program (e.g. warning and error messages, explicatory tag labeling). The VAT manual is an introductory tutorial as well as a quick reference book. VAT and its manual can be downloaded at

[http://www.tau.ac.il/lifesci/departments/plant\\_s/members/kosman/VAT.html](http://www.tau.ac.il/lifesci/departments/plant_s/members/kosman/VAT.html).

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Kosman E and Leonard KJ (2007) *New Phytologist* 174: 683-696.

## Estimation of nuclear DNA and other methods for identification of wheat leaf rusts

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Leaf rust caused by *Puccinia triticina* is the most common rust disease of bread wheat. In the last twenty years severe leaf rust epidemics have been described annually on durum wheat also. Ten years ago a different *forma specialis* of *P. triticina* was found in Israel. This form attacks *Aegilops speltoides* in nature and proved to be compatible with wild emmer wheat (*Triticum dicoccoides*) and *T. urartu* in artificial inoculations. In addition, in Morocco, Spain and Portugal, durum wheat is being attacked by a different leaf rust species: *P. recondita*.

The host range tests used for the identification of these wheat pathogens don't separate between some overlapping isolates. As a result, a wide range of methods were developed and applied for the identification of each of these rusts.

- Flow cytometry of urediniospore and pycniospore nuclei stained by propidium - iodide, was used for DNA content estimation
- Studying the shape of the urediniospore substomatal vesicle
- Defining the specific alternate host
- Crossing between the tested forms and species
- Inducing telia formation and measuring teliospore dimensions (length, width, area), by the use of image analysis
- Studies of the main host range

### Conclusions:

Estimation of DNA content of urediniospore nuclei is a quick, cheap and reliable method for distinguishing the bread wheat leaf rust from the durum wheat leaf rust.

*Ae. speltoides* form may be distinguished by a very narrow range of its main hosts.

*P. recondita* originated on wheat in Morocco, differs in all parameters tested from the three forms of *P. triticina*.

## Molecular basis of durable rust resistance in wheat

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Improved resistance to fungal rust diseases in cereals is critical for world food security and can only be achieved through breeding varieties with durable rust resistance. Durable resistance is not caused by single genes, but by a combination of usually 3-5 individual partial genes which are combined by classical breeding in a single wheat line to achieve near immunity. The wheat gene *Lr34* is such a quantitatively acting, partial resistance gene which is involved in durable resistance. *Lr34* is associated with resistance to two different rust diseases of wheat, leaf rust (caused by *Puccinia triticina*) and stripe rust (*P. striiformis*). Interestingly, it also confers partial resistance to the powdery mildew pathogen. *Lr34* is expressed in adult plants during the critical grain filling stage and is most effective in the flag leaf. Flag leaves of many wheat cultivars containing *Lr34* develop a necrotic leaf tip, a morphological marker described as leaf tip necrosis. The expression of this trait, however, is strongly dependent on the environment. The *Lr34* gene was first documented in Canada although *Lr34*-containing germplasm has been a part of wheat improvement since the early part of the 20<sup>th</sup> century and seems to have originated in Italian and Chinese material. Wheat cultivars containing *Lr34* occupy more than 26 million hectares in various developing countries alone and contribute substantially to yield savings in epidemic years.

The *Lr34* gene has remained durable and no evolution of increased virulence towards *Lr34* has been observed for more than 50 years of large scale use in resistant wheat lines. Despite the importance of partially acting plant resistance genes involved in durable resistance, very little is known on them at the molecular level. Understanding the molecular nature of this class of resistance has important implications for long term control of rust diseases. Previous studies have localized the co-dominant gene *Lr34* on the short arm of wheat chromosome 7D between the two markers gwm1220 and SWM10. We further reduced the target interval in a map-based cloning approach based on three high-resolution populations. High-resolution mapping revealed a 0.15 cM target interval for *Lr34*. The 363 kb physical interval containing both flanking markers was fully sequenced in the *Lr34* containing hexaploid wheat cultivar 'Chinese Spring'. A PDR/ABC transporter coding gene was finally identified as *Lr34* by the molecular analysis of 8 independent mutants. The mutations were identified as missense mutations, splice site mutations and short deletions. Interestingly, an allele of the *Lr34* gene is also found in all wheat lines lacking *Lr34* activity. This susceptible allele differed from the resistant form in only two amino acid changes. Based on these polymorphisms, highly specific molecular markers have recently been developed. They allow diagnostic detection of *Lr34* in tested germplasm and will be highly useful in combining *Lr34* with other partial resistance genes to achieve near immunity. Recent data on *Lr34* orthologs in several Triticeae and other grass genomes will be presented. In addition, strategies for molecular understanding of *Lr34* function and possible applications in agriculture will be discussed.

## **Leaf tip necrosis co-segregates with seedling leaf rust resistance conditioned by *Lr34* at low temperatures**

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The leaf rust resistance gene *Lr34* is typically described as an adult plant resistance gene, which is not expressed at the seedling stage at normal greenhouse temperatures. However, if plants with *Lr34* are grown at low temperatures, approximately 8 C, then *Lr34* is expressed at the seedling stage. Different lines and cultivars with *Lr34* demonstrate a range of responses from near immunity to susceptibility on the primary leaf. The Thatcher-*Lr34* near isogenic line RL6058 (Thatcher\*6/PI58548) was very resistant to a number of virulence phenotypes at the seedling stage, when grown under these cool conditions, but was susceptible at typical greenhouse temperatures. A cross was made between Thatcher and RL6058 and a doubled haploid population was developed. This population was screened for leaf rust resistance in multiple field trials and at the seedling stage at approximately 8 C. Molecular marker analysis at the *Lr34* locus was also done on each progeny line. In the seedling test the population segregated for a single resistance gene. The seedling data corresponded to the field resistance data and the molecular marker analysis. After extended incubation at cold temperatures seedling progeny lines with *Lr34* also developed distinct leaf tip necrosis, whereas it was absent in susceptible plants, similar to their reaction as adult plants. Leaf tip necrosis was completely linked to *Lr34* in this experiment.

## Functional characterization of the durable disease resistance gene *Lr34*

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*Lr34* is one of the most relevant disease resistance genes in agriculture providing durable disease resistance since several decades in breeding programs worldwide. In combination with other minor disease resistance genes it acts effectively against the devastating biotrophic leaf rust (*Lr34*), stripe rust (*Yr18*) and powdery mildew (*Pm38*) pathogens. *Lr34* is expressed in adult plants during the critical grain filling stage, giving partial resistance by delaying the progression of pathogen development. The gene is further associated with the morphological trait leaf tip necrosis.

The *Lr34* target interval was isolated on wheat chromosome 7DS using a map-based cloning approach and revealed six candidate genes within a 363 kb sequence. Among these candidate genes, sequence analysis of eight independent mutants identified a gene encoding an ATP-binding cassette (ABC) transporter belonging to the pleiotropic-drug resistance (PDR) subfamily as the genetic factor underlying *Lr34* based resistance. Each mutant displayed a sequence alteration leading to either splice site mutations, premature stop codon or an amino acid exchange, whereas the other candidate genes did not provide any sequence polymorphisms. Phenotypic analysis revealed that all mutants were more susceptible to leaf rust, stripe rust and powdery mildew, besides having lost the morphological trait leaf tip necrosis.

In order to functionally characterize the wheat PDR-like ABC transporter *Lr34* different heterologous systems, easier to study and more amenable than hexaploid wheat, will be tested for their applicability. *Lr34* has a close homolog in rice, *OsPDR23*, displaying 84% identity at the amino acid level. Arabidopsis contains *AtPDR5*, *AtPDR9* and *AtPDR8*, sharing 56% similarity with the wheat protein. Mutants of these *Lr34* homologs are available for both diploid systems. Their complementation with the wheat PDR-transporter might help to elucidate the molecular mechanism underlying *Lr34* based durable disease resistance. In order to identify putative transporter substrates, approaches using metabolomics and transport assays in yeast or isolated protoplasts provide promising systems to test candidate compounds.

## **Identification and characterization of expressed sequences involved in differential host response of wheat carry the *Lr34/Yr18* genes challenged with *Puccinia recondita* using cDNA-AFLP.**

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Leaf and stripe rusts are major biotic constraints to wheat production throughout the world. The most promising long-term control strategy is to breed and deploy cultivars carrying durable resistance based on minor, slow rusting genes with additive effects. *Lr34* and *Yr18* genes in wheat are tightly linked together and induce adult plant resistance to leaf and stripe rusts, respectively. Differential gene expression analysis was carried out on the near-isogenic lines Thatcher and Thatcher-*Lr34/Yr18* following inoculation with leaf rust, using the cDNA-AFLP technique. Fluorescent labelled primers permitted the identification of polymorphic DNA fragments by PAGE and fluorescence imaging. Over one hundred polymorphic DNA fragments were identified, isolated and cloned. Based on Blastx and Blastn results, a majority of polymorphic fragments corresponded to sequences with known metabolic functions, however some of the isolated fragments encode disease resistance domain such as leucine rich repeat (LRR) and Nucleotide binding site (NBS). Meanwhile, several pathogenesis related proteins were identified such as cysteine protease, ABC1 family protein, thioredoxin peroxidase. Real-time PCR analysis of isolated transcripts showed that expression level of some transcripts altered in Thatcher-*Lr34/Yr18* inoculated with leaf and stripe rust. Parallel fluorescent histological study was conducted to evaluate defence reaction in both susceptible and resistance lines treated by leaf and stripe rust pathogens. Result showed that accumulation of mRNA related to known isolated genes initiate after appressorium formation however expression level was higher in Thatcher-*Lr34/Yr18* comparing to Thatcher. In seedling stage, expression pattern of isolated transcript in response to leaf and stripe rust pathogens, was slightly different to adult stage and defence response was accompany with delay in expression of isolated transcript. Meanwhile, there was no significant difference between Thatcher-*Lr34/Yr18* and Thatcher at seedling stage regarding to alternation in gene expression.

Key word: leaf rust, thatcherLr34, differential display, *Triticum aestivum*

## **Studies of the interactive effects of *Lr16* in combinations involving *Lr13* and/or *Lr34* and its effectiveness against Australian isolates of *Puccinia triticina***

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While virulence for the leaf rust resistance gene *Lr16* has been reported from some regions, its effectiveness in Australia singly, and in combination with other leaf rust resistance genes, is poorly understood. To study potential interactions between this gene and the genes *Lr13* and *Lr34*, a line carrying all three genes was generated by intercrossing the two Thatcher (Tc) near isogenic lines Tc+*Lr13*+*Lr34* and Tc+*Lr16*+*Lr34*. A genetic analysis of the seedling response to *P. triticina* pathotype 26-0, avirulent for both *Lr13* and *Lr16*, of F<sub>3</sub> lines generated from the intercross indicated digenic inheritance. F<sub>3</sub> lines from this cross that produced a very low infection type, lower than that of both parents, were presumed to carry *Lr13*, *Lr16* and *Lr34*. This was supported by genetic analyses based on a model in which two genes segregated independently. Several of the lines regarded as carrying all three genes, along with cultivar Thatcher and lines near isogenic to Thatcher and carrying *Lr13*, *Lr16* and *Lr34* singly or in combination (*Lr13*+*Lr34*, *Lr16*+*Lr34* and *Lr13*+*Lr16*), were tested for response to five Australian *P. triticina* pathotypes with contrasting pathogenicities for *Lr13* and *Lr16*. The resistance genes *Lr13* and *Lr16* were found to interact to give higher levels of resistance regardless of the pathogenicity of the pathotype used, while all three genes interacted to pathotypes avirulent for *Lr13* and *Lr16* under certain conditions only. Additional studies using these lines and the *Lr16* carrying cultivars Selkirk, Etoile de Choisy and Exchange showed that three Australian *P. triticina* pathotypes regarded previously as virulent on *Lr16* were in fact partially virulent only. These studies also demonstrated that the expression of *Lr16* varied between pathotype and cultivar, with cultivar Exchange giving the clearest response across all pathotypes.

## Wild emmer wheat as a source for disease resistance genes: from genetic diversity to gene cloning

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The studies conducted by our group are focused on unraveling the genetic basis of several qualitative and quantitative agronomic traits derived from wild emmer wheat, *Triticum dicoccoides*. Wild emmer wheat, the tetraploid ancestor of domesticated wheat, was discovered in 1906 by A. Aaronsohn in Israel. Aaronsohn had the pioneering vision that wild wheat would become a source of germplasm for crop improvement. Nevertheless, traditional approaches for utilization of wild alleles are usually very slow. The advanced genomic technology available today may help to increase the efficiency of utilization of wild germplasm.

Stripe rust, caused by the fungus *Puccinia striiformis* f. sp. *tritici* is a devastating fungal disease in many wheat-growing regions of the world. New strategies to reduce stripe rust yield losses are required to satisfy the increasing world demand for cereals. Wild emmer wheat, is a promising source of resistance to stripe rust (e.g. *Yr15*, *YrH52* and *Yr36*). *Yr15* and *YrH52* are major dominant genes that confer particularly high resistance; *Yr36* confers high temperature, partial (quantitative) resistance. The availability of wheat genetic maps, wheat ESTs mapped to deletion bins, wheat BAC libraries, and the complete rice and *Brachypodium distachyon* genome sequences enabled us to conduct positional cloning studies aimed at cloning of the stripe rust resistance genes derived from wild emmer wheat.

A primary genetic map of *Yr15* was developed using a cross of a BC<sub>3</sub>F<sub>9</sub> line, which contains a 1BS chromosome segment of *T. dicoccoides* accession G-25 carrying *Yr15*, with the recurrent parent *T. durum* cv. D447. SSR and RFLP markers were used to assign *Yr15* to 1BS chromosome deletion bin Sat0.31. ESTs assigned to 1BS Sat0.31 enabled us to establish colinearity with a 740 kb contig located on *Oryza sativa* chromosome 5 and a 840 kb region located on *B. distachyon* chromosome 2. Further comparative genomic study enabled to narrow down the region carrying *Yr15* to 0.3 cM colinear with a ~28 kb sequence in *B. distachyon*. The sub-centiMorgan map of *Yr15* was used to identify *T. aestivum* BAC clones spanning the target region and for constructing a non-gridded BAC library from the donor line of *Yr15*. Further chromosome walking is underway to assemble a BAC contig containing *Yr15*.

Using a similar comparative genomic approach we have recently completed the positional cloning of the slow rusting gene *Yr36* (Science 323:1357-60), derived from wild emmer wheat. *Yr36* is effective only under relatively high temperatures and provides partial resistance to stripe rust. A gene designated *WKS1* was found to be completely linked to *Yr36* in a large mapping population. *WKS1* has a novel architecture with a kinase and a START lipid-binding domain. Six independent mutations and transgenic complementation confirmed that *WKS1* is *Yr36*. This gene, which was lost during the domestication of pasta and bread wheat, provides a new tool to control this disease and has the potential to contribute to the improvement of stripe rust resistance in a wide range of germplasm.

These studies demonstrate the potential of wild emmer wheat gene pool for improvement of cultivated tetraploid and hexaploid wheats, and emphasize the contribution of the recently developed genomic tools for the utilization of wild wheat germplasm.

## **Intra specific ancient diversity of splicing motifs and protein surface at the wild wheat *Lr10* CC and LRR domains**

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The gene pools of wild relatives of crop plant have been proposed as a source of new disease resistance genes to broaden the basis of genetic resistance. Wild emmer wheat (*Triticum dicoccoides*) is a promising source of disease resistance. To estimate the potential of the wild emmer gene pool, there is a need to study the diversity of the resistance genes at the molecular level and to evaluate the effect of this diversity on the gene function. Here, we have studied the allelic diversity of the *Lr10* leaf rust resistance gene, encoding a CC-NBS-LRR protein originally identified in cultivated bread wheat, in 60 genotypes of wild emmer wheat representing 12 populations collected in Israel. The nucleotide diversity of *Lr10* was high relative to other wild emmer wheat genes ( $\pi=0.029$ ). Diversity and selection pressures were measured along the gene by calculating the nucleotide diversity and the difference in synonymous vs. non-synonymous mutations for each codon. In contrast to most R-genes, the CC domain was the most diverse domain, with a high rate of non-synonymous mutations, suggesting that it is subject to positive selection. A 3D prediction of the CC domain (one of the first 3D predictions of CC domains in R-genes) has revealed that the diversity in the CC domain is located at a solvent-exposed helix structure where it might interact with pathogen effectors. The LRR domain was relatively conserved, but had a hot spot of amino acid variation between the two haplotypes in the ninth repeat. This repeat was longer than the other repeats and its 3D structure was unique, forming an extensive alpha helix. The two haplotypes differed in the splice junction and in their splicing regulatory region. One haplotype was alternatively spliced, leading to a deletion of five repeats from the LRR domain. The two haplotypes differed in 18 out of 70 amino acids (26%), suggesting a long evolutionary process. Tajima neutrality tests revealed that the region is under balancing selection. Those two haplotypes were found also in *T. monococcum* and *T. urartu*, which also carry the A genome. This diversity, maintained for about 3 million years, supports the "trench warfare" model of plant - pathogen interactions.

## In the neighborhood of resistance to *Puccinia hordei* QTL2

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Partial resistance of barley to leaf rust (*Puccinia hordei* Otth) can probably be considered as a basal defense of the plant to attacking microbes. Such a resistance is based on a prehaustorial mechanism, associated with the formation of papillae, that retards epidemic development in the field, although plants show a compatible infection type. The pathogen has a longer latent period, a lower infection frequency and a lower sporulation rate on partially resistant barleys. In previous studies, we found that typically 3 to 5 QTLs segregated for partial resistance to leaf rust in biparental barley mapping populations. More than 20 such QTLs have been identified and placed on a high-density integrated map of barley. We selected one QTL, *Rphq2*, as a target for map-based cloning because it was highly effective in seedlings and because it mapped in a region with high recombination near the telomere of Chromosome 2HL. *Rphq2* was identified in the recombinant inbred line populations L94 x Vada and Vada x SusPtrit, the resistance allele coming from Vada. Interestingly, a QTL for resistance to the heterologous rust species *P. persistens* co-localized with *Rphq2* in the latter population, suggesting that *Rphq2* may reveal a direct link between the host and the non-host components of basal resistance. The substitution mapping of *Rphq2* using 39 sub-NILs pinned the QTL into a genetic window of about 0.1 cM between markers WBE114 and WBE115. This interval displays synteny with a 70 Kb stretch of rice Chromosome 4 and with a 50 Kb stretch of *Brachypodium distachyon* contig super\_0.

The gene underlying *Rphq2* could be responsible for an increased resistance (resistance factor, in Vada) or for an increased susceptibility (susceptibility factor, in SusPtrit) to leaf rust. For this reason, we constructed two pooled BAC libraries, one from 'Vada' that contains a resistance allele at *Rphq2* and one from 'SusPtrit' that contains a susceptibility allele. The Vada BAC library consists of approximately 161,000 clones with an average insert size of 82 Kbp per clone, corresponding to 3 genome-equivalents. The SusPtrit BAC library consists of 173,000 clones with an average insert size of 118 Kbp per clone, corresponding to 4 genome-equivalents. A contig of three BAC clones from Vada spanning the QTL region was successfully constructed. Two BAC clones were isolated from SusPtrit that were positive each for a marker flanking *Rphq2* but that did not overlap with each other. The three BAC clones from Vada contig and one clone from SusPtrit were sequenced. In Vada, the region between flanking markers is a stretch of 184 Kb that carries at least 7 predicted genes. The 72 Kb sequence obtained from SusPtrit poorly aligned with the sequence from Vada suggesting a highly divergent region between the two genotypes. Indeed, while 3 genes were common to Vada and SusPtrit, a putative NADH-ubiquinone oxidoreductase was only found in the Vada contig and a serine/threonine protein kinase was only found in the SusPtrit BAC clone. From all predicted genes, six have homology to genes previously involved in disease resistance. Those six genes are candidates to explain *Rphq2*. Gene expression experiments are being performed to confirm that the candidate genes are indeed expressed and to possibly detect association of (a) gene(s) expression with leaf rust infection.

The identification of the gene underlying *Rphq2* will provide new insights into the genetic basis of partial resistance of plants to fungal pathogens and may help to provide a direct link between host basal resistance and non-host resistance.

## Race non-specific adult plant gene involved with the quantitative resistance of Toropi

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One of the major wheat diseases in most growing regions of the world is leaf rust, caused by *Puccinia triticina* Erikss. Incorporation of resistance genes in wheat cultivars is the preferred method to control the disease. Most leaf rust resistance genes described to date are race-specific seedling genes which act by complementary gene-for-gene interaction imposing a strong selection pressure on the pathogens to overcome the resistance. The rapid development of new pathotypes of *P. triticina* makes the deployment of more durable resistance a better strategy to protect against leaf rust. Durable resistance has been conferred by race non-specific adult plant resistance genes, however just two genes *Lr34* and *Lr46* have been described to date which confer this type of resistance. Toropi, a Brazilian variety, which has maintained its resistance for more than 40 years, has two non-described recessive adult plant resistance genes on 1AS and 4DS wheat chromosomes. With the aim of identifying, characterizing and fine mapping the source of resistance present in Toropi, crosses between Toropi and IAC13-Lorena (susceptible cultivar in Brazil) and both parents are being analyzed. Eleven pathotypes with different virulence and a mixture of Canadian races were used in seedling tests allowing the identification of at least two race-specific seedling resistance genes. By gene postulation, analysing 28 near isogenic lines, *Lr14b*, *Lr15* and *Lr28* are some of the possible leaf rust resistance genes present in Toropi, however at least one additional seedling gene should confer the resistance. At adult stage, the plants were challenged against six different pathotypes and a mixture of races. To date, one race-specific and probably two race non-specific adult plant resistance genes were identified in Toropi. IAC13-Lorena, which is completely susceptible to Brazilian races, seems to have at least one seedling and one adult plant race-specific gene. The results to date indicated that the resistance in Toropi is conferred by multiple genes. The presence of probably two race non-specific adult plant resistance genes makes Toropi a valuable source of leaf rust resistance.

## Convergent Evidence for Genes Underlying Quantitative Powdery Mildew Resistance in Barley

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Quantitative pathogen resistance is of high importance to plant breeders due to its durability. However, it usually is polygenic in nature and controlled by quantitative trait loci, which makes it difficult to handle in practice. Therefore, knowing the genes that underlie quantitative resistance would allow its exploitation in a more targeted manner. In order to identify genes that mediate quantitative resistance of barley to the powdery mildew fungus *Blumeria graminis* f.sp. *hordei* (*Bgh*) we have combined a functional-genomics approach based on transcript profiling and transient-induced gene silencing (TIGS) with an association-genetic and a QTL-mapping approach. Approximately 500 differentially regulated candidate genes of barley were silenced by RNAi followed by scoring of *Bgh* infection. Silencing or over-expression of 63 of those candidates resulted in a significantly altered interaction phenotype of attacked epidermal cells with *Bgh*. These plus a number of candidate genes based on publicly available data were selected for re-sequencing in a worldwide selection of barley accessions from the IPK genebank that differed in their quantitative resistance to *Bgh*. This approach revealed a number of genes that exhibited significant association with race-nonspecific seedling resistance. Two candidate genes mapped to chromosome 5H, within a QTL interval for seedling resistance to *Bgh* identified in the OregonWolfe Barley population. Re-sequencing of 20 genes within 20 cM at this locus revealed low linkage disequilibrium and identified additional associated gene candidates including the cell-death regulator protein HvLsd1, which was found nearest to the peak QTL marker. In conclusion, the integration of functional-genomic with association-genetic approaches allow us to rapidly zoom into candidate-gene lists and genetic intervals of interest and hold the promise to accelerate the discovery of genes underlying complex, quantitative traits in crop plants.

## **RAC/ROP binding proteins of barley influence the interaction with *Blumeria graminis* f.sp. *hordei***

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RHO-like small monomeric G-proteins are a conserved protein superfamily. The plant specific RHO-subgroup proteins, Rho of plants (RAC or ROP), play a central regulatory role in many cellular processes such as cytoskeleton dynamics and vesicle trafficking. Stable over expression of the constitutive active (CA) forms of three different HvRAC/ROPs in barley results in an increased susceptibility of the plants to the powdery mildew fungus *Blumeria graminis* f. sp. *hordei* (*Bgh*), whereas knockdown of *HvRACB* limits fungal penetration success (1,2). Additionally, data support a function of RAC/ROPs in polar growth processes and in actin cytoskeleton organisation. By deep screening in yeast, RAC/ROP interacting barley proteins were found. This involved a RAC/ROP interactive binding CRIB motif protein RIC171 (3) and a RAC/ROP binding cytoskeleton associated protein, both of which influenced penetration success by *Bgh* when mis-expressed in epidermal cells of barley. In planta protein-protein interactions studies and observation of dynamic localization of fusions of ROPs and interacting proteins revealed functions of ROPs in modelling of the cytoskeleton during accommodation of haustoria in intact cells. Data suggest that barley ROPs act as susceptibility factors by supporting accommodation of haustoria in intact plant cells.

- (1) Schultheiss et al. 2002, Plant Physiol 128: 1447-1454
- (2) Pathuri et al. 2008, Plant Cell Rep 27: 1877–1887
- (3) Schultheiss et al. 2008, Cell. Microbiol. 10:1815-1826

## **Bax Inhibitor-1-like cell death inhibitor proteins of plants regulate interactions of barley and *Arabidopsis thaliana* with powdery mildew fungi**

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Plant – powdery mildew pathosystems are models for biotrophic host – parasite interactions. Whether the interaction results in resistance or susceptibility depends at least in part on the timely execution or suppression of host cell death reactions, i.e. the hypersensitive response. Based on this assumption, it can be expected that the (mis)expression of plant cell death regulators might be used to modulate plant colonization by powdery mildew fungi.

Using a transient transformation assay, we found that transient over-expression of the conserved cell death inhibitor of barley (*Hordeum vulgare* L.), *BAX INHIBITOR-1* (*HvBI-1*), supported establishment of haustoria of adapted and non-adapted *formae speciales* of the grass powdery mildew fungus (*Blumeria graminis*) in barley epidermal cells and delayed pathogen-dependent cell death responses (Hüchelhoven et al., 2003; Eichmann et al., 2004, 2006). In contrast, knock-down of *BI-1* expression seems to reduce penetration success of the virulent barley powdery mildew fungus (*B. graminis* f.sp. *hordei*, *Bgh*), indicating that *HvBI-1* functions as susceptibility factor in the barley-*Bgh* interaction.

The *Arabidopsis thaliana* genome harbours several *BI-1*-like genes. *AtBI-1* promoter activity can be observed at sites of infection by adapted (*Golovinomyces orontii*) and non-adapted (*Bgh*) powdery mildew fungi and seems to be connected to host cell penetration. As in barley, *AtBI-1* over-expression supports development of powdery mildew fungi on *Arabidopsis*. However, the knock-out of *AtBI-1* does not dramatically alter the powdery mildew infection phenotype. This might be due to functional redundancy of *BI-1*-like genes. Nevertheless, heterologous over-expression of *BI-1*-like *Arabidopsis* cDNAs supported *Bgh* entry into barley epidermal cells. In addition, the investigation of *Arabidopsis BI-1*-like T-DNA insertion mutants revealed one, which displayed strongly suppressed conidiophore production by *G. orontii* along with enhanced cellular defense responses. Together, the data suggest an important function of *BI-1* family proteins as regulators of susceptibility in plant-powdery mildew interactions.

Hüchelhoven, R., Dechert, C., and Kogel, K.H. (2003). Overexpression of barley BAX inhibitor 1 induces breakdown of *mlo*-mediated penetration resistance to *Blumeria graminis*. Proc. Natl. Acad. Sci. U.S.A. 100: 5555-5560.

Eichmann, R., Schultheiss, H., Kogel, K.-H., Hüchelhoven, R. (2004) The barley apoptosis suppressor homologue Bax Inhibitor-1 compromises nonhost penetration resistance of barley to the inappropriate pathogen *Blumeria graminis* f.sp. *tritici*. Mol. Plant-Microbe Interact. 17: 484-490.

Eichmann, R., Dechert, C., Kogel, K.-H., Hüchelhoven, R. (2006) Transient over-expression of barley BAX Inhibitor-1 weakens oxidative defence and MLA12-mediated resistance to *Blumeria graminis* f.sp. *hordei*. Mol. Plant Pathol. 7: 543-552.

## **The genome of *Blumeria graminis f. sp. hordei*.**

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We are sequencing and annotating the barley powdery mildew genome using a combination of Sanger and diverse high-throughput sequencing methodologies. Our aim is to provide a reference genome for powdery mildews as a tool to advance our understanding of these challenging obligate biotrophs. One of the early discoveries is that the powdery mildew genomes are ~120Mb, i.e. much larger than closely related ascomycetes. This massive genome expansion is due to the presence of highly repetitive DNA made up of (retro)transposable elements. These are distributed throughout the genome and constitute ~70% of the genome. We propose that this expansion was the result of the loss of RIPing: one of the genetic pathways controlling immunity against genomic parasites. Other data emerging from the project will also be presented.

## Virulence effectors and retrotransposons in *Blumeria graminis*

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AVR<sub>k1</sub> and AVR<sub>a10</sub> are effector proteins that contribute to the successful establishment of haustoria in *Blumeria graminis* f sp *hordei* (*Bgh*), the causal agent of barley powdery mildew. They belong to a large gene family in the genome of *Bgh*, with homologous sequences in other *formae speciales* (*ff. spp*) infecting other grasses. Members of the AVR<sub>k1</sub> family are found in the proximity of TE1a LINE-1 retrotransposons and both can be expressed as a single transcript. We have studied the extensive proliferation of the AVR<sub>k1</sub> gene family throughout the genome of *B. graminis*, with sequences diverging in *ff. spp* adapted to infect different hosts. The frequency with which members of the AVR<sub>k1</sub> and TE1a retrotransposon lineages occur together in the genome is highly significant, and phylogenetic analysis show that both classes of sequences have coevolved. This is the first direct evidence that a parasite effector gene family and a particular retrotransposon lineage are consistently associated and have coevolved. The coevolution of these two entities indicates a mutual benefit to the association, which could ultimately contribute to parasite adaptation and success. We propose that the association would benefit 1) the powdery mildew fungus, by providing a mechanism for amplifying and diversifying effectors and 2) the associated retrotransposons, by providing a basis for their maintenance through selection in the fungal genome.

## Brothers in arms: Two cereal powdery mildews in the spotlight

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*Blumeria graminis* f. sp. *tritici* (*Bgt*), the causal agent of powdery mildew in wheat, and the barley pathogen *Blumeria graminis* f.sp. *hordei* (*Bgh*) are believed to be very similar on the molecular level as they infect the closely related cereals wheat and barley. To better understand their phylogenetic relationship and to estimate their divergence time, we analysed tree BACs of *Bgt* and compared them with available orthologous regions of the *Bgh* draft genome in order to get a first insight on genome structure and conservation between the two species.

The tree BACs have a combined size of 274kb. A large repeat database developed from a *Bgt* 454 whole genome sample and a few publicly available *Bgh* elements were used for annotation and characterisation of repetitive elements. The genome sequences of yeast and *Magnaporthe grisea* were used as references for gene prediction. Comparison of the annotated sequences showed that the coding regions of genes are well conserved between the two species. Gene number and their linear order as well as the individual orientation were found to be identical. The intergenic regions, by contrast, are very diverse in complexity and dimension. Filled with repetitive sequences of various kinds, we found LINE and SINE elements to be the most prevalent. The portion of repetitive DNA in the analysed regions was estimated to be at least 42%-54%.

While the high amount of repetitive DNA in general displays a great challenge, the high similarity of gene sequences between *Bgt* and *Bgh* will be very helpful for whole genome sequencing projects of the two fungi. The divergence time estimation of 10 MYA based on the available data indicates that the two mildews have separated more recently than their hosts (11.6 MYA). From this we conclude that *Bgt* and *Bgh* have co-evolved with wheat and barley after an initial few million years of host-jumping.

## Map-Based Cloning Of The Avirulence Gene *AvrPm3* In The Wheat Powdery Mildew *B. Graminis* F. Sp. *Tritici*

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The ascomycete *Blumeria graminis* f.sp. *tritici* is an obligate biotroph and the causal agent of the disease powdery mildew on wheat. The pathogen-host interaction is controlled by specific resistance genes in the plant, matching fungal avirulence genes according to the gene-for-gene model. Our group has cloned the *Pm3* resistance gene in wheat, which was shown to confer race specific resistance to different isolates of *B. graminis* f.sp. *tritici* in an allelic manner, and seven *Pm3* alleles were identified (Plant Physiol 139: 885-895, 2005; Plant J 47: 85-98, 2006). Isolation of the fungal gene specifying the *Pm3*-mediated resistance, i.e the avirulence gene *AvrPm3*, would now facilitate elucidation of the molecular basis of recognition by *Pm3* (direct or indirect recognition of *AvrPm3*) and of induction of defence processes. It would also provide valuable information in terms of co-evolution between both partners of the interaction, in particular to investigate whether the different *Pm3* alleles recognize different *AvrPm3* genes, or allelic series of the same gene.

Our aim is to clone *AvrPm3* through a map-based cloning strategy. This project takes place in the frame of a recently initiated broad study of *Blumeria graminis* f.sp. *tritici* at the genomic level, for which several tools were developed: first, a *B. graminis* genomic BAC library was constructed. The library consists in 12,288 clones with an average inserts size of 115 kb, and covers 12x the genome of *B. graminis*, assuming a genome size of 120 Mb. Three-dimensional DNA pools and high-density filters are available, making this library a powerful resource, essential for our future works. Second, we developed a whole genome sequencing project. Genome sequences were produced using Solexa and 454 GS FLX Standard technologies, generating 600 Mbp and 120 Mbp, respectively. This will be completed with 5 new runs of 454 Titanium technology (20x genome-coverage expected). Sequencing of BAC-ends performed on 10,752 clones of our library also generated additional 17 Mbp of data. All together, those resources should help us to construct a reasonable assembly of the genome which will be valuable in genome studies and comparative genomic projects. Finally, directly related to the cloning of *AvrPm3*, a *B. graminis* segregating population (150 individuals) was generated from a cross between isolates differing in their capacity to induce a resistance response on wheat lines harbouring *Pm3c* and *Pm3f* alleles. Ninety-four individuals were used to produce a genetic map based on SSR (12), CAPS (5), and AFLP (83) molecular markers.

Three AFLP markers were shown to be linked to *AvrPm3* at two, four and twenty-five centimorgans respectively, defining a genetic interval containing the gene. One marker located 2 cM from *AvrPm3* was cloned and exploited in a screening of our BAC library. A chromosome-walk was performed, resulting in a contig of approximately 300 kb, and three new BAC-ends-derived polymorphic markers flanking *AvrPm3* permitted us to narrow down the interval to a 150 – 200 kb genomic region. BAC clones are now under sequencing process and will be annotated in order to identify candidates of *AvrPm3*.

## Asymmetric Reciprocal Virulence Pattern Among *Blumeria Graminis* Isolates Originating From Domesticated Wheat And Its Wild Progenitor

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Powdery mildew (*Pm*) caused by the biotrophic pathogen, *Blumeria graminis* (DC.) E.O. Speer, f. sp. *tritici* Em. Marchal (*Bgt* hereafter), is a foliar wheat disease causing severe yield losses worldwide. Considerable efforts are needed to enrich the reservoir of resistance genes by extending the knowledge of this interactive pathosystem. In the current study, a diverse collection of wheat germplasm, consisting of 60 wild (*Triticum turgidum* ssp. *dicocoides*, the progenitor of domesticated wheat) and domesticated (*T. turgidum* ssp. *durum*, *T. turgidum* ssp. *dicoccon* and *T. aestivum*) wheat genotypes, were screened for powdery mildew resistance at the seedling stage by measuring pustules density. Screening was conducted with four local *Bgt* isolates, originally collected from contrasting environments and hosts: (i) *Bgt*#15 was collected from a field of domesticated durum wheat; (ii) *Bgt*#70 was collected from a field of domesticated bread wheat; and (iii) *Bgt*#58 and *Bgt*#66 were collected from a wild emmer natural site. Each isolate showed a different reaction pattern in the tested collection. *Bgt*#15 is a highly virulent isolate virulent on 71% of the genotypes in the collection. Nevertheless, the proportion of resistant genotypes differed significantly between the domesticated varieties (12.5%) and the wild accessions (44.4%). *Bgt*#70 collected from a *T. aestivum* plant showed high virulence on *T. aestivum* genotypes (its original domesticated host) and a moderate and low virulence on domesticated *T. turgidum* and wild *T. turgidum*, respectively. Isolates *Bgt*#66 and *Bgt*#58 generated reciprocal reaction, showing reduced virulence on domesticated germplasm and higher virulence to the wild material. *Bgt*#58 is avirulent on only 33% of the genotypes in the collection. This group of genotypes which shows complete or partial resistance to this isolate is governed by domesticated genotypes. *Bgt*#66 showed the lowest level of virulence and was hardly capable of attacking the domesticated group. Analysis of variance of the phenotypic response, revealed significant isolates × host species interaction ( $P(F) < 0.0001$ ). This reciprocal pattern is probably the result of long term plant-pathogen co-evolution. Asymmetry is a main characteristic of this interaction expressed by reduced ability of *Bgt* isolates evolved on wild emmer populations to attack domesticated wheat germplasm.

The genetic basis of powdery mildew responses to *Bgt*#15 and *Bgt*#66 isolates, at the seedling stage, was further dissected in a population of 152 F<sub>7</sub> recombinant inbred lines, derived from a cross between durum wheat (*cv.* Langdon, LDN hereafter) and wild emmer (acc. G18-16). This analysis revealed two different resistance mechanisms. The first strategy is **monogenic**: segregation ratio of the RIL population in reaction to *Bgt*#15 showed that the resistance in the wild parent (G18-16) is controlled by a single dominant gene that was mapped to the distal end of the 7AL chromosome arm. This genomic location harbors a cluster of powdery mildew R-genes, including the known *Pm1* locus. Further mapping and phenotypic tests are needed to determine whether the new gene detected in our tests is allelic to *Pm1* or it might be a novel R-gene derived from wild emmer wheat gene pool. The second strategy is **polygenic**: a total of five significant QTLs were associated with phenotypic reactions to inoculation with *Bgt*#66 explaining totally 53.6% of the trait variance. In all QTLs the higher resistance to *Bgt*#66 was contributed by the domesticated parent (LDN). The QTLs detected in the current study provide isolate-specific quantitative resistance against the *Bgt*#66 isolate. This finding challenges the common dogma which presumes that quantitative resistance is not race-specific. Furthermore, the identification of new resistance alleles from durum wheat could contribute to wheat breeding for *Bgt* resistance by precise exploitation of the available and well studied LDN genetic platform.

## Molecular analysis of the resistance of powdery mildew fungi to triazole fungicides conferred by mutations in *CYP51*

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The enzyme CYP51 catalyses C14 $\alpha$ -demethylation of eburicol, an essential step in the biosynthetic pathway of ergosterol, the predominant sterol in plasma membranes of powdery mildew and other filamentous fungi. It is the target site of sterol demethylation inhibitor (DMI) fungicides including the triazoles, which are widely-used for broad-spectrum control of diseases caused by Dikarya. Resistance of fungi to triazoles may lead to reduced disease control and crop losses. In *Erysiphe necator* (grapevine powdery mildew)<sup>a</sup> and *Blumeria graminis* ff.spp. *hordei* and *tritici* (barley and wheat powdery mildew)<sup>b,c</sup>, mutations in the open reading frame of the *CYP51* gene were associated with decreased sensitivity to triazoles. We sequenced the *CYP51* gene from isolates of *Podosphaera fusca* (syn. *P. xanthii*), the powdery mildew pathogen of cucurbits, which had varying degrees of sensitivity to the triazoles myclobutanil and triadimenol. Isolates with reduced triazole-sensitivity had either three or four mutations in *CYP51*, of which one was only present in the most resistant isolates. In contrast to the known mutations associated with triazole-resistance in *E. necator* and *B. graminis*, those in *P. fusca* clustered in the C-terminal half of the CYP51 protein. Homology modelling of the protein showed that, nonetheless, all the mutations in *P. fusca* and *B. graminis* lay on the surface of the catalytic pocket of the enzyme containing a heme group. Variation in the promoter region of *CYP51* was also associated with resistance to triazoles. In *B. graminis*, an insertion of 309 base-pairs (bp) in the promoter region was associated with reduced sensitivity. Comparison of isolates with and without this insertion suggested it might account for variation in responses to triazoles which cannot be explained by variation in the amino-acid sequence of the CYP51 protein alone. In *P. fusca*, a isolate which was hypersensitive to triazoles had a deletion of 377 bp in its promoter compared to the sequences of other isolates, while a triazole-resistant isolate had 11 nucleotide mutations in its promoter, of which most lay within the 377 bp sequence. However, variation in the promoter was not associated with significantly altered expression of *CYP51* in *P. fusca*. Together, these data suggest a model in which decreased sensitivity of powdery mildews to triazole fungicides may be generated either by mutations around the catalytic site of CYP51 or by mutations in the gene's promoter while resistance may be further enhanced by combinations of both classes of mutation.

<sup>a</sup> Délye C, Laigret F, Corio-Costet MF, 1997. Cloning and sequence analysis of the eburicol 14 $\alpha$ -demethylase gene of the obligate biotrophic grape powdery mildew fungus. *Gene* 195: 29-33.

<sup>b</sup> Délye C, Bousset L, Corio-Costet MF, 1998. PCR cloning and detection of point mutations in the eburicol 14 $\alpha$ -demethylase (*CYP51*) gene from *Erysiphe graminis* f.sp. *hordei*, a 'recalcitrant' fungus. *Current Genetics* 34: 399-403.

<sup>c</sup> Wyand RA, Brown JKM, 2005. Sequence variation in the *CYP51* gene of *Blumeria graminis* associated with resistance to sterol demethylase inhibiting fungicides. *Fungal Genetics and Biology* 42: 726-735.

## **A protein profile from *in planta* infection structures of the wheat leaf rust fungus, *Puccinia triticina*.**

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Haustoria occupy a central position in the exchange of nutrients between host and rust pathogen. They develop inside host cells upon infection and are thought to secrete ‘effectors’ which perturb host cells and divert nutrients to the haustoria ultimately to sustain the fungus and to suppress hostile host responses. In other systems, ‘effectors’ have recently been shown to possess avirulence functions, eliciting host resistance responses. The haustorial proteome and ‘secretome’ are therefore of great interest, however the difficulty of obtaining secretome and proteome components from haustoria has prevented progress in this area. Haustoria were enriched from infected leaves by repeated sucrose density gradient centrifugation and analyzed using several proteomics techniques. Initially two-dimensional gel electrophoresis (IEF x SDS-PAGE and SDS-PAGE x SDS-PAGE) was used but this technique is poorly suited for resolving proteins from highly membraneous structures such as haustoria. Multidimensional protein separation with analysis by liquid chromatography-mass spectrometry (GeLC-MS) was shown to be more suitable, since the first dimension of separation occurs in 2% SDS, in which almost all proteins are soluble. To date 133 proteins have been tentatively identified in several GeLC-MS experiments. The Mascot search engine was used to query a combined database consisting of a *P. triticina* (Pt) EST unigene set (over 12,000 unigenes), *P. graminis* (Pgt) proteins predicted from the published genome (over 22,000 proteins), the NCBI non-redundant database limited to fungal entries and common contaminants. Forty proteins were identified in both Pt and Pgt while 50 seem Pt-specific. For validation, RT-qPCR analysis was performed on 8 of the protein-coding genes. A high number of proteins involved in general and energy metabolism, as well as many ribosomal proteins were identified suggesting their upregulation and importance in this fungal structure. This correlates positively with the provisional analysis of the transcriptome based on EST occurrence in the database, and work on other biotrophic fungi. A number of translation initiation and elongation factors, as well as ubiquitins was apparent and noteworthy are several chaperones (heat shock proteins). Fifteen could not be identified of which 11 were Pt-specific; some of these were homologs of gene products identified in haustoria or “secretomes” of other fungal biotrophs. Experiments to resolve the smaller protein fractions are in progress and a re-analysis of our protein profiles using predicted proteins from the anticipated Pt genome are eagerly awaited.

## Proteome analysis of virulent and avirulent *Blumeria graminis* f. sp. *hordei* inoculated barley

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*Blumeria graminis* f. sp. *hordei* is a biotroph pathogen that causes powdery mildew disease in barley. In this study, Pallas01 and Pallas03 barley lines having *Mla1*, *Ml (Al2)* and *Mla6*, *Mla14 R-genes* were inoculated with Bgh103 race of the *Blumeria graminis* f. sp. *hordei* having avirulence and virulence to Pallas01 and Pallas03, respectively. The proteins were isolated from the three biological replicates of 12, 24, and 48 hr post inoculated samples following the method in Rampitsch *et al.*, 2006. These three biological replicates of three time points together with the mock inoculated plant proteins were separated on 2D-PAGE using IPG strips of 4-7 pH values as three technical replicates. The gels were analyzed on PdQuest 8.0.1 (BioRad). The protein spots were confirmed using the students T-test and principle component analysis in order to detect the up- or down-regulated ones by comparing against the controls (mock inoculated samples) and the samples having resistance or susceptible responses. The protein spots were manually excised and subjected to the nano-LC-ESI-MS/MS analysis (Proteome Factory, Germany). The MASCOT algorithm was used for the identification of the differentially expressed proteins. The proteins determined as differentially expressed will be presented including sub-cellular localization and possible signal peptide containing proteins.

Rampitsch, C., Bykova, N.V., McCallum, B.D., Beimcik, E., and Ens, W. (2006), "Analysis of the wheat and *Puccinia triticina* (leaf rust) proteomes during a susceptible host-pathogen interaction." *Proteomics*, 6(6):1897-1907.

## Microsatellite mapping of the powdery mildew resistance gene in two Chinese landraces of wheat (*Triticum aestivum* L. em. Thell.) Mazhamai and Xiaobaidong

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Powdery mildew, caused by *Blumeria graminis* (DC) Speer f. sp. *tritici* (*Bgt*), is one of the most devastating diseases of common wheat world-wide. Growing resistant cultivars is one of the most effective, economical and environmentally safe approaches for disease control. A significant problem in wheat breeding is the loss of function for powdery mildew resistance genes which can be frequently caused by emergence of new virulent races. Therefore, search for new powdery mildew resistance genes is necessary to wheat breeders. The most widely used resistance sources are the alien relatives of wheat. However, landraces of wheat are also good sources of resistance, especially for their ease of use in breeding programmes. Up to now, 57 powdery mildew resistance genes/alleles mapped to 43 loci (*Pm1-Pm43*) have been characterized in wheat and its wild relatives. Five of these loci (*Pm1a-Pm1e*, *Pm3a-Pm3k*, *Pm4a-Pm4c*, *Pm5a-Pm5e* and *Pm8*) have multiple alleles. Three of these genes (*Pm5d*, *Pm5e* and *Pm24*) come from Chinese landraces.

We identified dozens of resistant cultivars from more than 2000 landraces since 1980s. Some of them displayed good resistance to powdery mildew of wheat. The gene carried by Fuzhuang 30, for example, has been designated as *Pm5e*. We inoculated two landraces Mazhamai and Xiaobaidong with 21 *Bgt* isolates and found out that they exhibited response patterns different from that of the cultivars/lines possessing documented *Pm* genes and still show a wide spectrum of resistance in nearly 30 years after identification. To characterize the resistance gene(s) in Mazhamai and Xiaobaidong, we crossed Mazhamai and Xiaobaidong, respectively, to the susceptible cultivar Chancellor. Seedlings of F<sub>1</sub> and 197 F<sub>2</sub> plants were obtained in a cross between Mazhamai and Chancellor and the population was inoculated with isolate E30 of *Bgt*. Genetic analysis indicated that a single recessive gene control the powdery mildew resistance at the seedling stage. Analyses of the population with SSR markers show that *mlmz* was flanked by Xgwm577 and Xgwm1267 at a genetic distance of 2.9 cM and 2.3 cM, respectively. This gene is considered a new gene and designated temporarily as *mlmz*. Seedlings of reciprocal crossed populations of F<sub>2</sub> plants between Xiaobaidong and Chancellor were inoculated with isolate E09 of *Bgt*. A total of 125 and 107 F<sub>2</sub> plants were used for linkage analysis. *mlxbd* was also flanked by the SSR makers Xgwm577 and Xgwm1267. Genetic and deletion-line maps show that the locations of *mlmz* and *mlxbd* are both on wheat chromosome 7BL, distal to breakpoint 0.78 of deletion line 7BL-10, which is the same region that *Pm5* locus locates.

Up to now, eleven powdery mildew resistance genes near or at the *Pm5* locus have been described (*Pm5a*, *Pm5b*, *Pm5c*, *Pm5d*, *Pm5e*, *mlxbd*, *PmH*, *mljy*, *mlsy*, *PmTm4*, *mlmz*). Powdery mildew resistance gene in Xiaobaidong was previously designated as *mlxbd* and considered as a closely linked gene with *Pm5* (Huang et al. 2000). There maybe also an allele relationship between *mlxbd* and *mlmz* (Duan et al. 2001). Because seed was not made available for allelism test between *Pm5* and *mlmz*, we were unable to determine if the resistance gene in Mazhamai was a novel resistance gene or an allele of the *Pm5* locus. The results indicate that there may be more *Pm5* alleles or a gene cluster near the *Pm5* locus.

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## Detection of resistance genes by molecular markers in wheat cultivars registered in Hungary

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Stem and leaf rust can attack wheat and cause significant yield reduction worldwide. In Hungary, stem rust caused large losses at the end of the 19th and the first part of the 20th centuries. For farmers, application of resistant cultivars provides efficient solution to avoid yield losses. According to annual observations by artificial infections on near-isogenic lines in Hungary, stem rust resistance genes *Sr36* (derived from *Triticum timopheevii*), *Sr27* and *Sr31* (derived from *Secale cereale*) provide effective resistances in the field. In our study we determined the frequency of *Sr31* and *Sr36* stem rust resistance genes in 220 bread wheat cultivars registered in Hungary in a period of 35 years, from 1970 to 2005, by molecular markers. Among the 156 Hungarian wheats a significant part (33%) had the 1BL.1RS wheat-rye chromosome translocation, the source of *Sr31* gene, or the *Sr36* gene (17%). In the 64 foreign cultivars, deriving from 12 countries, only 5% had the *Sr31* and 11% carried the *Sr36*. Comparing the two main Hungarian wheat breeding programs the occurrence of *Sr31* resistance gene in cultivars of Martonvasar institute had reached 92-95% (between 1993 and 1999) with the marginal use of *Sr36*. Conversely, in the Szeged-institute cultivars, *Sr36* reached the frequency of 40-45% (between 2001 and 2003) with a low share of *Sr31*. Analyzing the data of official artificial stem rust infection trials in Hungary (16 years data between 1985 and 2003) cultivars with *Sr31* and *Sr36* had significantly lower infection than those which did not have any of these two genes. While the average infection of cultivars without *Sr31* or *Sr36* proved 48%, the group cultivars with *Sr31* or *Sr36* had only 15% or 5% infection, respectively.

We also studied the frequency of leaf rust resistance genes *Lr20*, *Lr37*, *Lr34* and *Lr52* in the Hungarian gene pool using molecular markers. New molecular markers closely linked to leaf resistance genes *Lr20*, *Lr29* and *Lr52* were also developed. Marker assisted selection program to transfer these and other rust resistance genes to adapted cultivars developed at our institute is progress.

## Spring Wheat Breeding In Western Siberia For Resistance To Leaf And Stem Rust

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Spring wheat is a major crop in Russian Federation and in Western Siberia in particular. It occupies more than 5 mln ha and provides grain for the industrial cities of the region. Scientific breeding was established in 1920s and 1930s in several key research institutes in the cities of Kurgan, Omsk, Novosibirsk and Barnaul. The research and breeding programs in Omsk (Siberian Research Institute of Agriculture and Omsk State Agricultural University) were especially successful in developing varieties which were adapted not only to Western Siberia but also to Northern Kazakhstan and European part of Russia. However, almost all the varieties developed in the 1980s and early 1990s were susceptible (чувствительны) to leaf rust. The importance of the pathogene increased in 2000s when higher precipitations (осаждение) caused significant annual yield losses. Utilization, the diverse gene pool in the crosses resulted in the development of resistant germplasm which also combined good adaptation to local conditions and excellent grain quality. The varieties Tertsiya, Sonata, Kvinta, Duet demonstrated good resistance. It appears that the resistance is most likely controlled by unidentified major genes. The Stem Rust is also present in the region with the latest significant occurrence in summer 2008. This and the potential threat of Ug99 resulted in establishment of a wide collection of spring wheat germplasm both from Western Siberia and from outside of the region for screening both in Omsk and in Kenya in 2009. The preliminary data suggests that there are several genotypes combining resistance to local population of Stem Rust with resistance in Kenya. These genotypes have been used in the crossing program. The methodology of breeding for rust resistance is discussed.

# **POSTER PRESENTATIONS**

## **P1. Physiologic specialization of wheat leaf rust (*Puccinia triticina* eriks.) in the Czech Republic in 2006-2008**

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In 2006–2008 virulence of the wheat leaf rust population in the Czech Republic was studied on Thatcher near-isogenic lines with *Lr1*, *Lr2a*, *Lr2b*, *Lr2c*, *Lr3a*, *Lr9*, *Lr11*, *Lr13*, *Lr15*, *Lr17*, *Lr19*, *Lr21*, *Lr23*, *Lr24*, *Lr26* and *Lr28*. Samples of leaf rust were obtained from different parts of the Czech Republic. Resistance gene *Lr9* was effective to all tested isolates like in the previous years. In 2008 two isolates from one sample were virulent to *Lr19*. Unlike in the previous years sporadic virulence on *Lr24* was observed. Relatively effective were also *Lr1*, *Lr2a*, *Lr28* and *Lr2b*. Other *Lr* genes were defeated by the majority of the tested samples. Our results transformed to the numbers of physiologic races indicate that race 61SaBa prevailed like in the previous years, followed by races 61, 2, 12SaBa, 2SaBa, 14, 77SaBa, 12, 57, 6, 53, 53SaBa, 77, and 14SaBa. Twenty five winter wheat cultivars (registered 2006-2008) were tested with 7 leaf rust isolates in the greenhouse. Out of them 15 showed resistance at least to one rust isolate. The same winter wheat cultivars were tested in the field trials where natural infection was supported by artificial infection. High resistance was recorded in the field for cvs Baletka, Biscay, Helmut, Mulan, Orlando. These cultivars were also resistant to all or most isolates applied in the greenhouse tests. Cultivars Eurofit, Anduril and Buteo, susceptible to all or to all but one rust isolate in the greenhouse showed resistance in the field which demonstrates the importance of field (partial) resistance.

## P2. Population structure of *Puccinia graminis* f.sp. *avenae* in Sweden

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Stem rust, caused by *Puccinia graminis*, is a serious disease of cereal crops and in the past this pathogen has caused severe epidemics worldwide. Incidence of oat stem rust in Sweden has increased recently. One possible explanation could be an increase in the occurrence of barberry (*Berberis* spp.), on which *P. graminis* sexual cycle can be completed, due to the repeal of the barberry eradication law in 1994. The epidemiology of *P. graminis* is not well known in Sweden. In this project, the population biology and epidemiology of *P. graminis* f. sp. *avenae* will be investigated in order to develop new strategies for the control of stem rust in oat. This information will also be relevant for controlling stem rust when new races of wheat stem rust pathogen, such as Ug99 (TTKSK), reaches Sweden.

Multiple samples of *P. graminis* f. sp. *avenae* were collected from 33 different oat fields during the summer of 2008. Samples were taken from both commercial fields and monitoring field plots used by the Swedish Board of Agriculture. Single pustule isolates were obtained from the field collections and increased in a greenhouse. DNA from these isolates was extracted from urediniospores and infected leaf tissue and was used to screen Simple Sequence Repeat (SSR) markers developed for *P. graminis* f.sp. *tritici* (Szabo, 2007; Jin et al, 2009; Zhong et al, 2009). Some of the markers successfully produced amplicons indicating that they will be useful for genotyping isolates of *P. graminis* f. sp. *avenae*. Allele sizes for *P. graminis* f. sp. *avenae* were different than those from a U.S. reference isolate of *P. graminis* f.sp. *tritici*. A selected set of North American isolates of *P. graminis* f.sp. *avenae* are also included in the study which will enable comparison between the populations from both continents. The results from this study will indicate if the oat stem rust pathogen in Sweden is a sexual population and the role of barberry in the epidemiology of *P. graminis* f. sp. *avenae*.

### **P3. AFLP marker conversion into single locus markers in *Puccinia striiformis* f.sp. *tritici***

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AFLP is a multilocus fingerprinting technique which has shown to be very powerful in detecting polymorphism among *Puccinia striiformis* f.sp. *tritici* (*Pst*) isolates from pathogen populations displaying low genetic diversity. The advantage of the AFLP technique is that no prior sequence knowledge is needed. However, the procedure is laborious and provides only dominant markers. Our aim is to convert AFLP markers into simple allele specific PCR-based markers for studying genetics and evolution in *Pst*. Polymorphic AFLP fragments were excised from <sup>33</sup>P labeled gels and sequence characterized. Internal PCR primers were designed on the basis of the sequence data in order to obtain SCAR markers. Some of these markers displayed the same polymorphism as seen with the original AFLP fragment and provided a dominant SCAR marker. For the remaining AFLP fragments the internal PCR primers displayed fragments of identical size from all isolates. We are sequencing these fragments in order to identify internal SNPs. For identification of SNPs or indels that originally caused the AFLP markers, the flanking regions of the AFLP fragments will be identified.

The single locus markers will be applied on wild type and putative mutant and recombinant isolates in order to obtain further insight into the mechanisms of pathogen evolution.

## **P4. Evidence of asexual overwintering of the willow leaf rust fungus *Melampsora Larici-Epitea***

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Leaf rust, caused by the fungus *Melampsora larici-epitea*, is an economically important disease in biomass willow plantations for renewable energy. *M. larici-epitea* has five spore stages and is heterocious, with willow (*Salix* spp.) as the uredinial, telial and basidial host, and larch (*Larix* spp.) as the spermatogonial and aecial host. During summer the polycyclic uredinial phase occurs on willows, and the fungus overwinters as telia on fallen willow leaves. In spring, basidiospores are produced and infect larch where sexual reproduction takes place and results in recombined aecidiospores, which anew can infect willows. The complete life cycle, including the sexual phase on larch, has been presumed to be crucial for winter survival of *M. larici-epitea* in northwestern Europe, since evidence of overwintering in the uredinial form has been lacking. Asexual overwintering of urediniospores - were it possible for *M. larici-epitea* - would enable well-adapted genotypes to persist across years and increase to high frequencies, having then important epidemiological consequences.

To investigate the possibility of asexual overwintering, 586 urediniospore samples were collected from three locations, one in Northern Ireland, one in Germany and one in Sweden in two consecutive years (2000 and 2001). For the locations in Northern Ireland and Sweden, a comparison was also made to a collection made in 1997. To distinguish between genotypes, the DNA fingerprinting technique AFLP was used.

In the population in Northern Ireland, identical AFLP phenotypes were detected between years in 2000 and 2001 in six cases. Two of these six phenotypes also occurred already in 1997, and another two were detected between years in 1997 and 2000. Identical AFLP phenotypes were considered to be of the same genotype, since the probability that two isolates should have identical phenotypes for all the 83 analyzed AFLP loci by chance is negligible when the actual band frequencies and the sample size is taken into account. In the populations in Sweden and Germany, the same genotypes were not found between years. The results show that *M. larici-epitea* has a mixed reproduction system, with the possibility of overwintering both through sexual reproduction and as clonal lineages. Asexual overwintering is suggested to be of relatively great importance in the Northern Ireland population, while it is infrequent or absent in the Swedish and German populations in this study.

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## **P5. Occurrence of leaf rust resistance genes in Russian wheat varieties and their influence on virulence frequencies in the pathogen population**

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Leaf rust is the one of the destructive diseases in Russia although breeding for the disease resistance have been performed in Russia for more than 50 years. To develop strategies of genes for the trait deployment and to forecast changes in the pathogen population structure it is necessary to reveal genetic diversity of grown varieties.

STS, SCAR and SSR markers were used to identify 11 *Lr* genes (*Lr9*, 10, 19, 20, 21, 24, 25, 26, 29, 37, 41) in 250 spring and winter wheat varieties grown in Russian Federation. Using SNP marker for *Lr1* gene a limited set of these samples (45 varieties) was screened.

As a result, genes *Lr10*, *Lr26*, *Lr19*, *Lr9*, *Lr20* and *Lr1* were identified in the varieties and *Lr21*, 25, 29, 37, 41 were not detected.

Gene *Lr10* was the most frequent and was identified in 15% of winter and 40% of spring forms. This gene was among the first ones used at breeding stations in USSR. Wide distribution of varieties with the gene resulted in accumulation of virulent clones in the pathogen population. At the present time frequencies of virulence to the gene *Lr10* in Russian populations make up 85-100%.

Gene *Lr26* was identified in 10% of the varieties. The most of them are grown in North Caucasus, Ural, West Siberia and Volga regions. Effectiveness of this gene was overcome in 70<sup>th</sup> of the last century as a result of wide growing of cvs. Aurora and Kavkaz. Significant temporal and regional differences in frequencies of virulence to the gene *Lr26* (from 0 up to 100%) were found in Russian populations, reflecting different spatial share of varieties with *Lr26*.

DNA marker for *Lr19* was found for 5% of spring varieties (L503, L505, Volgouralskaia, Dobrunya, Ecada 6, Yulya, Kin. Niva). This gene was for the first time used in wheat breeding in Volga region; the first commercial variety L503 was released in 1993. Due to wide growing of these varieties in Ural, and Volga regions the gene lost its effectiveness. Extension of commercial use of the varieties with this gene and the pathogen migration resulted in appearance of virulent clones in other regions.

Gene *Lr9* is highly effective all around the world and is being involved in breeding programs of many countries. 5% varieties were found to have amplification fragments characteristic for *Lr9* carriers. Firstly, varieties with the gene were grown in Russia in Ural and West Siberia regions in early 2000<sup>th</sup>. Now they are recommended for cultivation in all Russia (Udacha, Duet, Pamyaty Ruba, Chelyaba 2 in Western Siberia and Ural regions; Nemchinovskaya 24 in Central and Volga ones; Splav in North-Western one). This distribution of genetically uniform varieties is assumed to cause sharp change in population structure in the nearest future.

Genes *Lr24* and *Lr20* were identified in 0.5% of varieties. Gene *Lr24* was high effective in Russia but since 2007 single virulent isolates in central part of Russia was determined. The frequency of virulence to *Lr20* was high in all Russian population during last 5 years.

Gene *Lr1* was revealed in varieties Moskovskaya 39, Deya, TAU. Now it practically lost its effectiveness in Russian Federation. From the early 2000<sup>th</sup> clear tendency to increase frequency of virulent clones is observed. In previous period (1980-1995) frequencies of virulence were significantly lower in European part of Russia and Caucasus region comparing with these in Western Siberia (Mikhailova, 2006). This tendency is caused for the first time by wide growing of varieties with this gene in European region of Russia (for example, Moskovskaya 39).

The data of population studies confirmed the important role of host genotype in dynamics and changes of the pathogen population structure for virulence to certain genes for resistance. Molecular marker systems are an alternative means of examining populations of plant pathogens providing a potentially greater number of markers which are presumably not subject to direct host selection. These aspects are now under study.

### **Acknowledgments**

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## P6. Variability and virulence of *Puccinia triticina* in North Caucasus, Russia

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Leaf rust (casual agent: *Puccinia triticina* = *Puccinia recondita* Rob.ex Desm.f.sp.tritici Erikss. et Henn.) belongs to the most wide spread and harmful wheat diseases. The protection of wheat from this disease remains of great concern, especially in southern regions of Russia. To perform a successful work on wheat breeding it is necessary to have a clear understanding of the fungus evolution, to pursue its intrapopulation structure and track down a trend of its variations.

In 2000-2007, according to the response of 46 wheat lines (cultivars) virulence of 980 *P.triticina* clones was described by us and frequency of occurrence of complementary virulence genes in the fungus population was determined. Based on the average values of this parameter the virulence genes were subdivided as follows: **Group 1:** p9, p19, p43 – not identified within the population (their share was 7.9 %); **Group 2 :** p24, p29, p41, p42, p45, pW – occurred with the frequency of 0.4 – 5.0 % (15.9 %); **Group 3:** p32, p38, p39 – frequency of occurrence: 6.0 – 10.0 % (7.9 %); **Group 4:** p2a, p15, p20, p21, p25, p36, p44 – frequency of occurrence: 11-25 % (18.4 %); **Group 5 (pp):** 1, 2c, 3, 3bg, 3ka, 10, 11, 14a, 14b, 16, 17, 18, 23, 26, 28, 30, 33, 40, B – frequency of occurrence: over 25 % (50.0 %).

For the research period, some trends of intrapopulation variations in the fungus structure caused by biotic and abiotic environmental factors were detected: increasing frequency of occurrence of the following virulence genes (pp): 2a, 2c, 3ka, 18, 21, 26, 32, 33, 38, 39, B; reduction in the frequency of occurrence for the following virulence genes (pp): 3bg, 15, 24, 25, 29; nearly the same level of the genes (pp): 1, 11, 20, 36, 40, 44, 45, W; elimination of the p41 and p42 genes for the last three years; reduction in the frequencies of occurrence with their subsequent increase for the genes (pp): 3, 10, 14a, 14b, 16, 17, 23, 28, 30.

The virulence gene pool monitoring in the North Caucasus *P.triticina* population showed its high heterogeneity due to active morphogenesis processes of the fungus. So, during the 8-year research period a significant number of phenotypes differing in their qualitative and quantitative composition of virulence genes were detected while identifying 980 single pustule isolates of this plant pathogen. The phenotypes with average virulence (10 to 22 genes) have the highest percentage (66.2) in the leaf rust population.

The special and temporal changes of the leaf rust fungus gene pool are caused by numerous reasons, but the selection pressure plays a key role here leading to the accumulation of those clones that are adapted to ecological and climatic factors of the region. The representativeness of one or another clone in a cultivar depends on the relationship between the plant pathogen genotype and that one of the host plant while the latter is a determining factor.

The work was done under a partial support of the ISTC Grant # 3036.

## **P7. Distribution of the stem rust pathogen (*Puccinia graminis f. sp. tritici*) and effectiveness of wheat resistance genes in North Caucasus, Russia**

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For the recent years an ever growing frequency of occurrence of this dangerous disease has been observed in the North Caucasus wheat fields. This fact may be explained, in our opinion, by an expanding area of barberry which is an intermediate host of *Puccinia graminis*, changes in the fungus race composition, loss of resistance by wheat cultivars *etc.* North Caucasus is known to be a zone where the pathogen completes a whole cycle of its development by colonizing barberry, grass and wheat. There is a source of a continuous recurrence of the infection here. Urediniospores may be transferred by air flows at long distances and under a favorable combination of climatic factors and presence of susceptible wheat cultivars they may cause the disease epidemics both in this and neighbouring regions of Russia.

During route surveys conducted by us in the wheat growing seasons of 2006-2008 the stem rust pathogen was detected each year both in different wheat cultivars and barberry and grass in Kochubeevsky, Kirovsky and other Districts of the Stavropol Region situated in the South Submountain Zone of North Caucasus. Separate outbreaks of the disease were also recorded in individual wheat fields in the Krasnodar and Rostov Regions with about 5% of severity. In spite of the limited stem rust manifestation in commercial fields the pathogen monitoring in different host plants and assessment of resistance genes effectiveness are important because of a great damaging effect of the pathogen and urediniospores of the fungus borne with air currents, which may cause high severity of stem rust in wheat under the conditions favorable for the disease development.

The results generated by two-year tests of known wheat resistance genes that were conducted at the milk-wax ripeness stage by determining a type of reaction (score) and severity of the disease (%) under the natural infection conditions showed that a wheat line containing the Sr9e gene was of high effectiveness (its type of reaction was scored as 0). Effectiveness (type of reaction: 1, 1(2); severity: 1 to 5%) was demonstrated by the Sr genes: 8, 9b, 11, 13, 14, 16, 17, 21, 25, 27, 31, 32, 33, 35, 36, 37, Tt1.

The highly effective and effective genes may be recommended for breeding rust-resistant wheat cultivars in the south of Russia by using their constant rotation, involvement of nonspecific resistance genes and taking into account the possibility of transmitting infections from adjacent territories.

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## **P8. Molecular marker screening of Turkish wild wheat species for stem rust resistance to Ug99**

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Wheat production is threatened by a constantly changing population of pathogen races. Given the rapid ability of many pathogens to overcome genetic resistance, the identification and practical implementation of new sources of resistance is essential. Recent outbreak of Ug99 is a typical example of stem rust pathogen rapid changing ability causing drastic yield loss on all major wheat varieties grown throughout the world. Although more than fifty Sr genes which were available to wheat cultivars were defeated by this new race, there are still few major resistance genes that are effective which were originally derived from the landraces and wild relatives. Therefore, as always been the case during thousands of years of domestication of modern wheat, the combat against this new race will require, pyramiding of these few available resistance genes as well as search of new ones, once again, in the rich diversity of landraces and wheat wild relatives. Using limited number of available PCR-based molecular markers to still effective few resistance Sr genes, thirteen wild wheat species (covering 80 accessions) collected from Blacksea, Central-East-Southeast Anatolia and Southeast Mediterranean regions of Turkey were screened for Sr2, 22, 24, 26, 36, 38 and 39 loci. Among 13 wild species, almost all of them were observed to have Sr 22, 26, 36, and 38 genes. Except those accessions belonging to 4 of the species (*Ae. caudata*, *Ae. ovata*, *Ae. triuncialis* and *T. urartu*), Sr2 loci were also present in all of them. None of the accessions belonging to 12 species contained the Sr24 and Sr39 genes. Only those accessions of *Ae. Speltoides var. ligustica* that were collected from Urfa region contained Sr39 gene. Although recent studies have shown that the Ug99 race and its derivatives have already defeated the resistance deployed by Sr24, 26 and 38 and thus they are no longer recommended for use as resistance source against Ug99, our results indicated that almost all of the wild species in Turkey still contained those loci. However, it was very encouraging that those accessions unique to Hatay region (*Ae. kotschyi*) and Central Anatolia region (*Ae. mutica*) of Turkey additionally contained Sr 2, 22, and 36 genes which are still considered highly effective genes as slow rusting and moderate to highly resistant to Ug99 respectively. Therefore, *Ae. Mutica*, *Ae. Kotschyi* and *Ae. Speltoides var. Ligustica* as potential donor for Sr39 gene from Urfa region might be valuable candidates for future combination breeding programmes to battle with this new stem rust epidemic.

## **P9. Yellow rust in the South of Ukraine and resistance of wheat varieties to it in the region**

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Yellow rust in the Steppe region of Ukraine is a highly harmful disease. In the pathogen population the races OEO, 6EO, 6E16 are prevailing and accompanied by the races 6E4, 6E17, 6E20, 7EO and 7E16. In 2009 the resistance to the pathogen of 660 domestic and foreign wheat varieties was studied in the artificially infected field nursery. A good number of varieties revealed high resistance to the disease. The rate of the disease affection on highly susceptible varieties was within 90 – 100%. The varieties with the Yr3c, Yr5, Yr9, Yr10, Yr15 and Yr17-genes were highly resistant.

## P10. Virulence of *Blumeria graminis tritici* in Serbia (2000-2009)

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*Blumeria graminis tritici*, the causal agent of powdery mildew is a regular and economically important disease of wheat in Serbia. Infection intensity varies depending on wheat variety, climate conditions and agricultural practices. Although there have been no epiphytotics in Serbia yet, this disease can play a significant role in the formation of yield. Successful wheat breeding for resistance to causal agent of powdery mildew is based on the identification of virulence and changes in the pathogen population. In Serbia, continual population surveys started in 1961, when physiological races were identified, and they continued up to date. In the early 1970s, conventional investigations of physiological races were improved by introducing a survey of the virulence of pathogen populations, based on the interaction between resistance alleles and virulence according to the “gene-for-gene” theory.

The sexual generation of the population was studied for ten years (2000-2009). Wheat samples containing cleistothecia of the fungus were collected in 46 locations in Serbia. Fungal cultures were produced by the standard method. Pathogenicity of 288 isolates (cultures) of *Blumeria graminis tritici* was determined by comparing them with wheat varieties and lines that possess resistance genes: Axminster/8\*CC (Pm1), Ulka/8\*CC (Pm2), Idaed 59b/8\*CC (Pm2+), Asosan/8\*CC (Pm3a), Chul/8\*CC (Pm3b), Sonora/8\*CC (Pm3c), Khapli/8\*CC (Pm4a), Weihenstephan M-1 (Pm4b), Hope/8\*CC (pm5), Michigan Amber/8\*CC (Pm6), Transec (Pm7), Kavkaz (Pm8), Amigo (Pm17), Normandie (Pm1+Pm2+Pm9), CI 12633 (Pm2+Pm6), Coker 983 (Pm5+Pm6), Halle Stamm 13471 (Mld), Granada (Pm5+Pm8), Dolomit (Mli), C-39 (Pm2+Pm4b+Pm6). (\* - eight-times backcrossed to Chancellor). Reaction of seedlings was assessed 8-10 days after inoculation, on the 0-9 scale.

The 288 isolates analyzed rendered 259 virulence formulae. The results showed that numerous genotypes had V7, V6, V5+8, V8, V3c, v5, Vi and V4a virulence alleles. Most efficient among the sexual populations of the parasite was the gene combination Pm5+6 from the variety Coker 983. A large number of the pathotypes produced by sexual reproduction cannot exist in nature, or their frequencies in the population are very low.

## **P11. Seasonal variation in genetic structure of leaf rust pathogen on cereals**

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In many studies population structure of cereal rust casual agents was investigated with analysis of different volume one-time samplings of the pathogens. At this methodological approach some aspects of population genetics are obviously lost, for instance the possibility of rapid changes of genetic structure of populations during one season due to selective reproduction of more adapted pathogen genotypes. The aim of the work was to verify hypothesis on possibility of changes in *Puccinia recondita* f. sp. *tritici* populations during one season of the host vegetation.

Monopustule isolates were sampled several times in 2007 season on wheat varieties in 4 regions of Russia and analyzed for virulence to 6 near-isogenic Thatcher lines. To describe intrapopulation variability and significance of differences between samples the criteria proposed by Givotovskii (1982) were used.

To elucidate the minimal number of isolates to be studied for comprehensive analyses of the pathogen population structure and differences between populations, subsamples of different volumes from 3 subpopulations were compared. Student's test revealed significant differences between subsamples from evidently one subpopulation at volume of sampling less than 100 isolates. Depending on quantity of analyzed isolates in each subpopulation the different number of phenotypes for virulence, different dominating phenotypes or their frequencies were identified. According to criterion of identity significant differences between subsamples from the same subpopulations existed if to analyze 25 or 50 clones. Thus, at comparison of *P. recondita* populations, the volume of sample should not be less than 100 isolates.

Among 939 isolates in 9 samples studied 56 phenotypes for virulence have been revealed from 78 possible. The average number of phenotypes varied from 13.65 to 29.91, share of rare phenotypes - from 0.06 to 0.26. In all populations change of a dominating phenotype during vegetation of the host was observed. Average number of phenotypes and in most cases shares of rare phenotypes differed significantly for samples from 1 region indicating to the lability of genetic structure of the pathogen populations during one season of wheat vegetation.

In three regions criterion of similarity between the subpopulations sampled at the same variety at different data differed from 1. Statistically significant differences between samples from Volgo-Vyatskii region as from North Caucasus could be explained by long interval between samplings (2 months). Significant differences between subpopulations from Krasnodarskii krai clearly proved the possibility of genetic structure of *P. recondita* f.sp. *tritici* populations to change in a very brief time period (8 days). The results could be explained by either influence of abiotic environmental factors on differential fitness of the pathogen genotypes or by migration of clones from nearby growing wheat samples.

*P. recondita* f.sp. *tritici* was sampled in Derbent (North Caucasus) from leaves of semiwinter triticale k-85 in 2008 when cereals in the region were harvested and wild hosts of the pathogen also completed their vegetation. Analysis of 2 samples (July 23 and August 3) revealed change of dominating phenotype and significant differences between samples frequencies of phenotypes and of alleles for virulences to 5 near-isogenic lines. In this case only influence of abiotic environmental factors can explain these differences. So, the important role of abiotic factors in seasonal dynamics of *P. recondita* f.sp. *tritici* population genetic structure was shown in this study.

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## **P12. Growth ratio differences as leaf rust forecasting factor in semiarid region**

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The relation of the large group of wheat genotypes resistance characters at seedling stage was not satisfactory established with adult symptom severities in the semiarid region (around 600mm of annual rainfall). The point was defining the water and soluble carbohydrate speed as missing linkage with obligate parasite development.

Focused were six genotypes with high reaction types and short latency periods according to first appeared pustules. The sum of the stem and first leaf lengths of seedlings ten days after inoculation grown at 20°C, were divided with second leaf ones and defined as infected growth ratio (IGR). The values below 1,3 were connected with lower symptom severities. Simultaneous was estimated the growth ratio (GR). At 25°C the differences between genotypes were related to previous but followed by lower values. Beside tested varieties according to GR most distanced were Rapsodija and Sonata as of IGR-GR Rapsodija and Novosadska rana 2. The correlation between first mentioned character and field leaf rust cause maximal severities at last two leafs was 0,67 as of second -0,94 ,when *Pyrenophora tritici repentis* and *Septoria spp.* did not appeared severely. Mentioned facultative parasites were such related opposite -0,793, 0,972 and -0,905, 0,951. *Puccinia triticina* decreaseable after 20<sup>th</sup> May were approximately 1:2 and 1: 0,5 according to last decade data for calculation.

### **P13. An overview of the network for important cereal diseases management research in Turkey between 2003 and 2007**

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Wheat (*Triticum* spp.) and barley (*Hordeum* spp.) are among the most strategic products of Turkey and interest large number of producers in the country. Wheat and barley are grown under diverse geographical regions in Turkey and various diseases which can cause significant yield and quality losses can occur different climatic conditions. The cereal diseases research network has been established to link disease research activities of the research institutes to strengthen the integrated management efforts. Specific objectives of the study include monitoring disease occurrence, multilocation screening of the joint germplasm for resistance to important diseases, and facilitation of exchange of disease resistance sources among the research institutes. Various research institutes run research activities and breeding programs to control important diseases of wheat and barley. This study is conducted to establish a network to integrate the activities of different research institutes located in different regions for management of the important diseases of wheat and barley.

Primary target diseases are yellow (stripe) rust (*Puccinia striiformis* f.sp. *tritici*), Leaf rust (*Puccinia triticina*), Stem rust (*Puccinia graminis* f.sp. *tritici*), common bunt (*Tilletia caries*, *Tilletia foetida*), loose smut (*Ustilago tirtici*), powdery mildew (*Erysiphe graminis*) for wheat; scald (*Rhynchosporium secalis*) and leaf stripe (*Pyrenophora graminea*) for barley, and root and foot rots, caused by *Fusarium* spp. and *Drechslera sorokiniana* for both crops. Viral diseases (*Soil-borne mosaic virus* (SBWMV) and *Barley stripe mosaic hordeivirus* (BSMV)) are also included in the study where they occur naturally.

In the network, a total of 100 wheat and 35 barley nurseries was established with 12.121 and 2.274 entries respectively with contributions from the 11 research institutes located in different regions in 2003-2007. Of these, 24 nurseries (2.639 genotypes) were durum wheat, 76 nurseries (9.482 genotypes) were bread wheat. The nurseries were sown in 11 locations for screening against the target diseases. Disease development was promoted with appropriate measures, including irrigation and artificial inoculation in some locations for screening purposes.

In 2003-2007 leaf rust, stem rust and powdery mildew occurred naturally in some coastal locations and yellow rust screening could be done efficiently under artificial inoculation in Ankara. The results of the activities indicated that total 4.422, 1.849, 327, and 878 wheat genotypes were identified to have good level of resistance to yellow rust, leaf rust, stem rust and powdery mildew respectively. The data on yellow rust trap nursery indicated that Yr1, Yr3V, Yr 4+, Yr5, Yr15, YrSP and YrCV were still resistant while other differentials were scored susceptible, intermediate or variable. The data on stem rust trap nursery indicated that Sr 24, Sr 26, Sr 27, Sr 31 were still resistant.

The study facilitated exchange of the germplasm with disease resistance properties among the breeding programmes.

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**Key words:** Wheat (*Triticum* spp), Rusts (*Puccinia* spp.), Resistance

## P14. Study of adult-plant resistance genes for leaf rust in marvdasht cultivar

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Leaf rust, caused by *Puccinia triticina* Eriks., is one of the most common and widespread disease of wheat worldwide. This study was conducted to identify the adult plant *Lr* genes carrying by "Marvdasht", a highly resistant cultivar to leaf and yellow rust diseases in Iran using field survey and molecular analyses. The results of field studies in a F<sub>2,3</sub> population derived from Marvdasht (R) × Bolani (S) indicated that there were two genes which conferred resistance against leaf rust isolate 140 in Marvdasht cultivar. Chi-square analysis on data collected for presence/absence of leaf tip necrosis (LTN) as a morphological trait revealed the monogenic inheritance for this trait in the F<sub>3</sub> population. Due to tight linkage between LTN and *Lr34/Yr18* or *Lr46/Yr29*, these genes were considered as two adult plant resistance candidates carrying by Marvdasht cultivar. Molecular tests using STS markers *CSLV34* and *CSHM46* designed for detection of *Lr34/Yr18* and *Lr46/Yr29* respectively, confirmed the presence of *Lr46/Yr29* as one of the two adult plant resistance genes in Marvdasht cultivar. Microsatellite analysis using *Xbarc160* marker located at 1cM distance of *Lr13*, resulted in confirmation of *Lr13* as the second resistance gene at the adult plant stage in Marvdasht. The F<sub>3</sub> families were grouped according to presence/absence *Lr46* and *Lr13* genes in homozygous condition. The results of analysis of variance on field data showed that the family groups were not significantly different for days to flowering, days to pollination, days to maturity and plant height. However, they varied significantly in terms of yield per plant, area under disease progress curve (AUDPC), LTN and coefficient of infection (C.I.). The results of correlation coefficient analysis indicated the negative and significant correlations of AUDPC with both yield per plant and LTN. Based on regression analysis, a linear relationship with negative slope was found between yield per plant and AUDPC using percentage leaf area affected by leaf rust on F<sub>3</sub> homozygous family groups derived from Bolani×Marvdasht cross.

## **P15. Global genetic diversity of winter wheat germplasm for resistance to leaf, yellow and stem rust**

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Winter and facultative wheat covers around 15 mln ha in the region of Central and West Asia and is grown in two distinct environments: irrigated wheat across the region normally in the valleys and lowlands; semi-arid rainfed wheat is primarily grown in the higher elevations and moisture stress presents a major challenge. Both environments are subjected to frequent and devastating attacks by rusts. Yellow Rust is a major pathogen across the region. Leaf Rust is more limited to irrigated lowlands or high rainfall areas. Stem Rust is observed in pockets across the region normally at higher altitudes. Resistance breeding for Yellow and Leaf Rust is very high priority while breeding for resistance to Stem Rust is primarily driven by the potential danger of *Ug99* spread. International Winter Wheat Improvement Program ([www.iwwip.org](http://www.iwwip.org)) is a joint cooperative program between the Ministry of Agriculture and Rural Affairs of Turkey, CIMMYT and ICARDA established more than 20 years ago. IWWIP aims to develop the germplasm suitable for Central and West Asia combining broad adaptation with resistance to prevailing biotic stresses, primarily rusts. IWWIP also plays a key role in facilitating the global germplasm exchange among the winter wheat breeding programs. The germplasm developed by IWWIP as well as the material received from cooperators is distributed to more than 130 cooperators in 50 countries through Facultative and Winter Wheat Observation Nursery (FAWWON) and International Winter Wheat Yield Trial (IWWYT). High turnaround of the winter and facultative wheat germplasm through IWWIP allowed detailed evaluation and screening for disease resistance of a large set of germplasm. Annually 250-300 entries are received from the main winter wheat breeding programs in America, Europe and Asia. The current study summarized the field data from Turkey and other countries of the region for the last three years (2007-2009) for the diverse set of 1000 entries representing both the commonly grown varieties, newly released varieties and advanced breeding lines from more than 30 countries. It appears that the distribution of the resistant germplasm depends on the prevailing rust in the target area of a particular breeding program. The material from the mid-west of USA is normally resistant to Leaf Rust but susceptible to Yellow Rust. The germplasm from Turkey, Iran and neighboring countries has high frequency of resistance to Yellow Rust. Testing of the diverse set of germplasm in Kenya for Stem Rust *Ug99* population in the field revealed that around 15% of the germplasm is resistant but there is no clear geographic source of resistance as it occurred equally from different regions. Winter/facultative lines and varieties with multiple resistances to two or three rusts have been identified.

## **P16. Mapping new resistance gene to *Puccinia hordei* Otth. in barley landrace ph944-3**

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Leaf rust of barley caused by *Puccinia hordei* Otth., is important disease in many barley growing areas. New virulent isolates as well as combinations of virulent genes are able to overcome resistance expressed by modern barley cultivars. However, only the leaf rust resistance gene *Rph7* is still effective in Europe. Since limited number of effective resistance genes are available it is necessary to identify new sources of resistance. The line Ph944-3 selected from barley landraces originated from ICARDA (International Center for Agricultural Research in the Dry Areas, Aleppo, Syria) carries single resistance gene to leaf rust and it is resistant to isolates virulent on lines containing resistance genes *Rph1* – *Rph6* and *Rph8* – *Rph12*. The allelism test excluded that the resistance is conditioned by gene *Rph7*. Eighty nine F<sub>2,3</sub> families were developed from the cross Ph944-3 × L94 for mapping experiments. Bulked segregant analysis with SSRs revealed linkage of the resistance locus with polymorphic microsatellites GMS021, Bmac0213, Bmag0872, EBmac0405, Bmag0347, Bmac0090 – specific to chromosome 1H. Saturation chromosome region of interest with other molecular markers is under way. So far, on chromosome 1H only one resistance gene *Rph4* was identified, thus we postulate new resistance gene on this chromosome.

## **P17. Resistance of wheat cultivars to *Puccinia* spp. in Southern Russia and Uzbekistan**

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One of the key reasons of high wheat grain losses is in mass outbreaks of fungus diseases among which leaf rust (casual agent: *Puccinia triticina* Erikss.) and stripe rust (casual agent: *Puccinia striiformis* West.) are most widespread and harmful. Leaf rust occurs in the south of Russia practically each year; the stripe rust occurrence is ever increasing. In Uzbekistan stripe rust has become a common disease in the recent years by reducing wheat yield significantly – down to 40% in some regions.

Resistant cultivars form the most important element within integrated control of rust diseases in wheat. An immunological assessment of 39 winter wheat cultivars against *Puccinia striiformis* and 13 cultivars against *Puccinia triticina* was undertaken in the All-Russian Research Institute of Biological Plant Protection under the artificial inoculation conditions. 8 cultivars (Rostovchanka 5, Amazonka, Vostorg and others) showed resistance to stripe rust, 15 cultivars (Dea, Don 107, Vita and others) demonstrated moderate resistance, and the rest 16 cultivars tested were susceptible to the pathogen. Against leaf rust, the cultivars Don 105, Rostovchanka 5, Axinit, Granit, Terra (38.5% of 13 tested) combined race-specific resistance (differential responses of seedlings to the fungus phenotypes) with the adult plant resistance; cvs. PalPich, Pamyat, Voyage, Garant, Esaul, Visa, Nota, Devise (61.5%) had a retarded type of the disease progress (slow rusting). These cultivars, in case of a susceptible type of reaction have an area under the disease progress curve that is about 50% less than for the control cultivar and a significant lesser reduction in 1000 kernel weight.

Among 36 wheat cultivars of local breeding that were tested in Uzbekistan cvs. Mira, Zamin-1, Saikhun, Emir, and Line-3 appeared to be moderately resistant to stripe rust. The rest cultivars were rated as the susceptible ones. None of 24 soft wheat cultivars tested was resistant to leaf rust.

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## P18. Biplot analysis of diallel data in strip rust of wheat

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Five bread wheat cultivars (Kokart, Domino, Ruapuna, Pool, and Tiritea), showing a wide response to yellow rust, were intercrossed in all combinations to provide a F<sub>1</sub> half-diallel for assessing disease infection type in Iran. The diallel dataset of the five cultivars was carried out by diallel GGE biplot (genotype main effect and interaction) analyze. The parents and 10 F<sub>1</sub> progenies were evaluated in the greenhouse by three pathotypes 7E18A<sup>-</sup>, 38E0A<sup>+</sup>, and 134E134A<sup>+</sup>. Infection types were recorded on day 14 and 19 after inoculation using the 0-9 scale Johnson et al. For each pathotype a randomized complete block design was used and data were analyzed by ANOVA. The first two principal components of biplot explained 95, 94 and 85% of the variation for pathotypes 7E18A<sup>-</sup>, 38E0A<sup>+</sup>, and 134E134A<sup>+</sup>, respectively. Ruapuna for the pathotypes 7E18A<sup>-</sup> and 134E134A<sup>+</sup>, Kokart for the pathotype 38E0A<sup>+</sup> had negative general combining ability (GCA) (more resistance) for infection type. Parent Tiritea was the best mating partner with the other parents for the pathotypes 7E18A<sup>-</sup> and 38E0A<sup>+</sup> while this parent was the best mating partner only with testers KOKART and POOL for the pathotype 134E134A<sup>+</sup>. In this pathotype (134E134A<sup>+</sup>) parent Kokart was the best mating partner with testers DOMINO, RUAPUNA, and TIRITEA. The results showed that parent Ruapuna was good in three combinations of pathotypes (7E18A<sup>-</sup> + 38E0A<sup>+</sup> + 134E134A<sup>+</sup>, 7E18A<sup>-</sup> and 38E0A<sup>+</sup> + 134E134A<sup>+</sup>) and so had good ability to show resistance by low infection type. Additive genetic component indicate the possibility of improving for lower infection type of stripe rust in breeding programs.

**Keywords:** Wheat; Disease resistance; Diallel; Biplot

## **P19. Setting up of a differential set to analyse the evolution of durum wheat leaf rust populations in France in 1999-2007.**

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Although the same species *Puccinia triticina* is responsible for leaf rust epidemics on bread and durum wheats in most areas, populations of the fungus have been shown to be different between bread and durum wheats, in several countries including France. Consequently the set of Thatcher isolines with *Lr* genes used to characterize *P. triticina* populations from bread wheat appeared not adapted for a good discrimination between durum wheat leaf rust pathotypes. Our first objective was thus to set up a differential set.

A set of 103 isolates collected in France, chosen to maximize geographical and varietal origin, was inoculated onto lines to be screened for their differential capacity. The set of lines tested comprised 19 international cultivars proposed as differential in the literature, 27 durum wheat cultivars from the French germplasm, in addition to a set of 31 Thatcher isolines. This led to the selection of a differential set comprising 18 french cultivars, Thatcher lines *Lr14a*, *Lr20* and *Lr23*, and cvs Altar and Gaza. All the other international cvs were susceptible to all isolates.

Using our differential set, a total of 10 pathotypes was identified. A strong evolution of the populations occurred from 1999 to 2007, e.g. the emergence of a pathotype in 2001 (avirulent on Altar but virulent on *Lr14a*) responsible for the susceptibility of the previously resistant cv Nefer. Virulence for Altar, already present from the beginning of our survey but at a very low frequency, increased as it was present in 2007 in several new pathotypes. New pathotypes virulent on most of the differentials (14 out of 19), appeared in 2007. The population was strongly differentiated according to cultivars.

The differential set appeared useful to characterize populations according to their virulence for resistance genes present in the French durum wheat germplasm. The limit is that this set, based mainly on local cvs, and well adapted to local epidemiological situation, brings at the moment no information on the genetic basis of virulence and resistance. Thus it is not directly and fully transferable elsewhere especially where a very different host germplasm is used, but could be a basis evolving towards a more international tool while integrating new information and replacing/adding differential hosts. Nevertheless our results remain very useful to provide breeders with informative data about pathotypes in the population, as they are directly characterized on their material. It also allows to provide them with well characterized inoculum for the testing of their material.

## **P20. Identification of homeological genes of resistance to leaf rust and powdery mildew in *Ae.speltoides*, *Ae.triuncialis* and in common wheat lines obtained with their use**

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The frequency of genes of resistance to fungal diseases is not high in the common wheat cultivars. The Laboratory of Genetics and Cytology of the Agriculture Research Institute of the Non-Chernozem Zone developed the lines of spring and winter wheat highly resistant to leaf rust and powdery mildew (Lapochkina et al., 1994, 2001) with the use of wild relatives (*Ae. speltoides*, *Ae. triuncialis*). The investigations were carried out for identification of genes of resistance to these diseases with the use of STS and SSR markers linked with *Lr1*, *Lr9*, *Lr10*, *Lr19*, *Lr21*, *Lr24*, *Lr34*, *Lr35*, *Lr37*, *Lr39*, *Lr46*, *Lr50* genes and *Pm2*, *Pm3c*, *Pm4b*, *Pm13*, *Pm16* genes. The amplification was conducted according to recommended protocols. Amplification products were separated by electrophoresis in 2% agarose gel in tris-borat buffer.

In the sample *Ae. speltoides* ( $2n=14$ , genome S) (k-389) several genes of resistance were identified to diseases that had been found in other relatives of wheat, in particular gene *Lr9* (*Ae. umbellulata*), *Lr19* (*Ae. elongatum*), *Lr37* (*T. ventricosum*) and *Lr50* (*T. timopheevii*), as well as genes of resistance to powdery mildew *Pm2* (*Ae. squarrosa*) and *Pm13* (*Ae. longissima*). The presence of these genes in *Ae. speltoides* can testify to phylogenetic relationship of the species and wheat, longevity of these genes of resistance and stability of their locuses in time and shows the significance of these genes for immunity development in the species of tribe *Triticeae* L. Genes *Lr39* (*Ae. tauschii*), *Lr37* and *Pm2* were identified in the sample *Ae. triuncialis* ( $2n=28$ , genome CU) (k-6452).

In the samples with genetic material *Ae. speltoides* the genotypes (141/00-1, DA line 4/00<sup>i</sup>) resistant to powdery mildew were found with the presence of gene *Pm13*, effective for the Non-Chernozem Zone, and effective gene *Pm2* (79/00<sup>i</sup>), as well as genotypes with resistance to leaf rust 100 rw (*Lr21*, *Lr35*, *Lr50*), 79/00<sup>i</sup> (*Lr19*, *Lr21*, *Lr34*). This makes it possible to use them universally in breeding programs for immunity.

In the samples with genetic material *Ae. triuncialis* the genotypes were found with group resistance to leaf rust and powdery mildew that is controlled by several effective genes *Lr* and *Pm* genes: 113/00<sup>i</sup>-1 (*Lr37*, *Lr39*, *Lr46*, *Pm2*, *Pm13*), 114/00<sup>i</sup>-1 (*Lr37*, *Lr39*, *Pm2*, *Pm13*). These samples are recommended as donors of resistance to leaf rust and powdery mildew in breeding for immunity.

## P21. Induction of leaf rust resistance in cereals' leaf segments by the benzimidazole

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To study juvenile resistance in cereals to leaf rust (*Puccinia recondita* f. sp. *tritici*) method of inoculation of leaf segments placed on cotton wool wetted with benzimidazole solution is widely used especially in countries of former USSR. To date there is a little information on coincidence of intact plant reaction to inoculation and that of detached leaves. This information is of interest because in the case of complete coincidence it is possible to study easily resistance of one plant in segregating populations to several clones of the pathogen, to one clone under different conditions etc. We studied resistance of cereal species samples from VIR World Collection to the disease at use of intact plants and leaf segments (0.5 cm in length) in benzimidazole (40 ppm) for inoculation with complex population of *P. recondita*. Inoculated plants and leaf segments were incubated at 22°C under constant illumination and types of reaction were recorded 7-8 days after inoculation according to Mines&Jackson (1926) scale.

From 338 studied samples of bread wheat four were resistant in intact state to leaf rust. At inoculation of detached leaves resistant types of reaction were recorded for 64 entries, 22 having type 0 (absence of the disease symptoms).

At inoculation of intact plants high level of juvenile resistance to the rust was found in 29 forms of triticale from 491 studied. At inoculation of leaves in benzimidazole 125 additional resistant genotypes were found, 37 having type of reaction 0.

Among 183 samples of 7 *Aegilops* L. species 9 (2 of *Ae. cylindrica*, 2 – *Ae. tauschii*, 1 – *Ae. longissima* and 4 of *Ae. heldreichii*) were resistant to leaf rust at both methods of evaluation. For 30 samples of above mentioned species and 5 of *Ae. bicornis*, susceptible at inoculation of intact plants, resistant types 0 and 1 were recorded on detached leaves.

In *Triticum durum* Desf. we studied juvenile resistance to the rust in 1527 samples, 76 being classified as possessing types of reaction lower than 3 after inoculation of leaf segments and only 5 were resistant in intact state. So, for 12 systems of cereal – *P. recondita* interaction high frequency of genotype-dependent induction of resistance by benzimidazole was found. It means the method of inoculation with the pathogen of leaf segments in benzimidazole can not be used for reliable evaluation of effective juvenile resistance.

In another experiment intact plants of near-isogenic lines of cv. Thatcher (*T. aestivum*) with genes for wheat leaf rust resistance *Lr12*, *Lr13*, *Lr34* were inoculated with 30 monopustule isolates of the pathogen from North-West region of Russia: all lines were susceptible to all isolates. Moreover susceptible reactions were recorded after inoculations of leaf segments in benzimidazole (60 ppm) with the isolates. But resistance to several isolates was found in leaf segments placed in benzimidazole solution (100 ppm) at high (25°C) temperature. The clones avirulent to the certain line under this specific condition were used to inoculate leaf segments of varieties known to have the gene of the line under the same condition. In all cases we observed resistant reaction to inoculation. Distinct gene-specific induction of the resistance indicated that this induction under complex of abiotic factors is not affected by the whole genotypes but by certain «major» gene for adult resistance. The method of leaf segments inoculation with isolates of *P. recondita* can be recommended for rapid preliminary postulation of the genes for adult resistance in wheat samples susceptible to the rust in juvenile stage.

## P22. Genetics of adult leaf rust resistance in wheat local samples from VIR collection

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Genetic diversity of wheat (*Triticum aestivum* L.) for seedling resistance to leaf rust (*Puccinia recondita* f. sp. *tritici* Erikss) is very narrow: samples with high level of the trait expression from World Collection of Vavilov All-Russian Institute of Plant Industry (VIR) possess only genes *Lr9*, 19, 24, 41, 45 and 47. To note virulence to *Lr9*, 19, 24 has been detected in several populations of the causal agent from Russia.

Most samples described as resistant in adult stage in Russian literature were shown to be susceptible to current populations of the pathogen from North-western region of our country. So, it is urgently necessary to find new sources of effective wheat resistance to leaf rust. One of the possible approaches to solve this problem is regarded to be evaluation of local samples.

We studied seedling and adult resistance to leaf rust in 2100 spring local samples from VIR Collection comprising 63 coun. No one form was resistant to the disease in juvenile stage.

There was no relation between disease development in the field and a sample origin. High level of flag-leaf resistance was found only in 24 samples from Kazakhstan (disease severity less 5%).

According to modified phythopathological test the samples can not possess known genes for adult resistance *Lr11*, 12, 27+31, 34, 46, 48 and 49. Gene *Lr13* was shown to be present in each sample. All the accessions have identical gliadin patterns and the same products of amplification after PCR with primers to SSR loci *Xbarc352*, *Xgwm630* and *Xgwm130* indicating to their genetic identity or very close origin.

Analysis of resistance of  $F_1$  plants from crosses with susceptible varieties proved recessive character of the resistance inheritance. In  $F_2$  segregation for resistance fits to theoretical 1 : 15 (two complementary recessive genes).

In  $F_2$  of crosses with Thatcher near-isogenic line carrying *Lr13* highly susceptible plants were not observed; segregation corresponded to expected one 1 : 3 (highly resistant : moderately resistant).

No susceptible plants were found in  $F_2$  of cross combinations between resistant samples.

As a result the adult resistance in local wheat samples from Kazakhstan is controlled by 2 complementary recessive genes one of them being *Lr13*. This gene combination could be useful for breeding for leaf rust resistance as the samples having the genes have proven high level of the resistance during 4 years under artificial epiphytotic conditions.

## **P23. Identification of *Puccinia triticina* resistance genes in seedlings of Iranian wheat advanced lines**

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Wheat leaf rust (Brown rust) caused by (*Puccinia triticina*) is a, worldwide major disease of wheat . In this research, three leaf rust isolates from Gorgan , Ahvaz, Hamedan were collected and determined their Avirulence and virulence formula 39 isogenic lines .The results showed that these three isolates were avirulence for plants with *Lr1*, *Lr14a*, *Lr19*, *Lr25*, *Lr23+ Lr28,Lr10+* genes while these isolates had virulence on plant with *Lr 22b\**, *Lr 3*, *Lr 3ka*, *Lr 3bg*, *Lr 9*, *Lr 11*, *Lr 12*, *Lr 13*, *Lr 14b*, *Lr 15*, *Lr 18*, *Lr 30*, *Lr 32*, *Lr 35\**, *Lr 36*, *Lr 37\**, *Lr b* genes. In order to evaluate of resistance to leaf rust, 122 advanced lines from Iranian regional program were tested at seedling stages with three races in greenhouse condition. The experiment was conducted in randomized complete block design (RCBD) with three replications. The latent period (number of days from inoculation to appearance of the first uredia) and infection type were recorded by McIntosh *et al*; 1995 method. Analysis of variance showed significant difference between 122 genotypes for infection type and latent period in Gorgan, Ahvaz and Hamedan races conditions which indicates. that there are more diversity between genotypes. The results showed that 28.5% of these genotypes, are resistance against three isolates , 22.5 percent of these are moderately resistance and 49 percent were completely susceptible.

Key words: wheat, seedling resistance genes and leaf rust

## **P24. Two-hybrid-based analysis of protein-protein interactions of the wheat RAD6 protein**

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*Rad6* is an ubiquitin-conjugating enzyme (E2) and it is shown that *Rad6* gene has differentially expressed in early infections of yellow rust (Bozkurt *et al.*, 2008). Our aim was to find the proteins that interact with wheat *Rad6* protein to understand the its function in disease resistance. The Yeast Two- Hybrid system was used to search for the interactors in yeast.

The yeast proteins that interact with wheat *Rad6* protein are found to be PRY3 (Pathogen Related in Yeast 3; Protein of unknown function which has similarity to the plant PR-1 class of pathogen related proteins), OXR1 (Oxidation Resistance 1; Protein of unknown function required for normal levels of resistance to oxidative damage, null mutants are sensitive to hydrogen peroxide), MAF1 (Negative regulator of RNA polymerase III; component of several signaling pathways that repress polymerase III transcription in response to changes in cellular environment), AIM10 (Altered inheritance rate of mitochondria protein 10, Protein with similarity to tRNA synthetases), FAR1 (Factor Arrest 1; A Cyclin-dependent kinase inhibitor that mediates cell cycle arrest in response to pheromone), and an unknown protein Y11066c (Putative protein of unknown function with similarity to helicases).

## **P25. Detection of micro RNAs putatively involving in disease resistance responses by screening and computational means**

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Micro RNAs are emerging control elements of plant physiology and their roles in biotic stress responses are extending daily. In this study the roles of miRNAs in powdery mildew-barley pathosystem and yellow rust wheat pathosystem were investigated via microarray analysis. Since plant miRNAs are highly conserved elements of cellular machinery, expression level determination of known plant miRNAs in infected wheat and barley enabled, identification of miRNAs that are changing in resistance and susceptibility. Moreover the target of miR159 was shown to be GAMyb in barley, in which no miRNAs are annotated yet. These data are the first showing the differential expression level of miRNAs upon race specific pathogen inoculation.

## **P26. Characterization of *Puccinia triticina* population from Russia in 2007 for virulence and DNA markers**

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Leaf rust (*Puccinia triticina* Erikss) is an important disease of wheat in many agroecological regions of Russia. Virulence surveys, conducted by Laboratory of Mycology and Plant Pathology annually throughout wheat regions, detect new pathotypes and monitor shifts of pathotype frequencies in the pathogen populations. Leaf rust survey data in 2007 have been used to characterize virulence and pathotypes diversity within and between wheat growing regions and the molecular polymorphism in isolate collection.

A leaf rust causal agent samplings were made at commercial fields and breeding nurseries in eight regions of Russia. Overall 468 single pustule isolates were analyzed on the set of 31 NILs with *Lr*-genes: 1, 2a, 2b, 2c, 3a, 3bg, 3ka, 9, 10, 11, 14a, 15, 16, 17, 18, 19, 20, 21, 23, 24, 25, 26, 28, 29, 30, 44, B, W, 27+31, 41 and 42. Race identification was done according to Long & Kolmer (1989). Index of similarity and criterion of identity (Givotovskii, 1982) were used to determine differences between samplings

Genes for resistance *Lr9*, *Lr28*, *Lr29*, *Lr41*, *Lr42* were revealed to be highly effective against all populations under study. Isolates virulent to *Lr19* were found in Ural region with frequency 7%. In 2007 isolates virulent to *Lr24* were identified in Central chernozem region (frequency 2%), Central region (3%) and West Siberia (4%).

Frequencies of isolates virulent to genes *Lr2a*, *Lr15*, *Lr20*, *Lr23*, *Lr44*, *LrW*, *Lr27+31*, vary in different region and were approximately the same as in previous years. Frequency of isolates virulent to gene *Lr1* increased as we observed since early 2000<sup>th</sup>. We found interregional differences in frequency of isolates virulent to gene *Lr26*, and the frequencies in North Caucasus, North-West region and West Siberia were the same as in previous years.

Frequency of monopustule *P. triticina* isolates virulent to all other genes for resistance were up to 80-100%.

Among 468 isolates 29 phenotypes for virulence were identified dominated ones being THTT and TGTT. As compared with previous years frequency of FGTT and FHTT phenotypes decreased significantly in all European regions. According to criterion of identity population from West Siberia differed significantly from North Caucasus, Central chernozem and Central region populations. Sampling from North-West region differed from all others under study. Differences between populations from West Siberia and Ural region were not significant, as between population from Central chernozem region and these from Ural, North Caucasus and the Lower Volga regions.

For molecular studies 5 UP-PCR primers (L45, 3-2, 15-19, L45 inv, AS4) (Mironenko, Bulat, 2004) and 20 RAPD primers (Park et al., 2000; Kolmer et al, 1995) were tested. At present we found differences between isolates for amplification profiles at use of 3 UP-PCR and 11 RAPD primers. There were not coincidence of isolate phenotyping at use of virulence and molecular markers. Differences between some populations from Russia were found to be significant for molecular phenotypes.

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## **P27. Virulence diversity of Israeli population of *Puccinia triticina* on wheat in 1993-2008**

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Leaf rust, caused by the fungus *Puccinia triticina* Eriks., is the most common rust disease of wheat that occurs in regions of wheat production worldwide. Monitoring Israeli populations of wheat leaf rust has been consistently performed since 1993. A total of 831 single urediniospore isolates was analyzed during 1993 -2008. Virulence phenotypes of 20 to 84 isolates were annually determined on 17 Thatcher isogenic lines that possess *Lr1*, *Lr2a*, *Lr2c*, *Lr3*, *Lr3ka*, *Lr9*, *Lr10*, *Lr11*, *Lr15*, *Lr16*, *Lr17*, *Lr18*, *Lr21*, *Lr23*, *Lr24*, *Lr26* and *Lr30* resistance genes. The number of detected pathotypes changed from 11 to 38 in 2004 and 1994, respectively, which were years of the least and highest richness of the pathogen annual collections. Structure of the pathogen populations has changed to a large extent since 1993. Clear separation of the annual collections of wheat leaf rust into two distinct groups of the 1993 – 1999 and 2000 – 2008 populations was established by descriptive tools like race composition and frequency of virulences as well as using methods of diversity analysis and clustering. This differentiation among the annual pathogen populations can be mainly attributed to the following forces: (i) the possible massive migration of leaf rust urediniospores from the neighboring regions in 1994 (in parallel with those of yellow rust with *Yr9* virulence firstly recorded in Israel in the same year); (ii) selection pressure of new wheat cultivars resistant to yellow rust that were introduced in Israel since 1997. Diversity within the annual collections of *P. triticina* isolates was highest in 1994 when many new pathotypes and linkages between virulences were observed. Single-step mutants of the new pathotypes were naturally selected and became predominant since 2000. Significant changes of virulence frequency on a number of *Lr*-genes for resistance (e.g. *Lr2a*, *Lr15*, *Lr17*, *Lr21*, *Lr26*) were also registered in 2000 – 2008. Patterns of genotypic, gene and genetic diversity within the annual populations of wheat leaf rust were not congruent.

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## **P28. Studying the effects of wheat cultivars genotypes on the virulence variability of *Puccinia striiformis***

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Constant variations in the varietal structure of areas under wheat find their reflection in the genetic structure of pathogenic populations including the stripe rust populations. Within agroecosystems, the motive selection pressure strengthens if genetically homogeneous cultivars and hybrids are used. As a response, mass multiplication and establishment of new pathogenic phenotypes take place. Therefore, it is necessary to control constantly the directional selection of phenotypes with different virulence and virulence genes by wheat cultivars to plan the varietal structure of wheat fields correctly.

Under the climatic chamber conditions we studied the seedling responses of 39 promising wheat cultivars to 37 *P. Striiformis* phenotypes differing in their virulence. Depending on their genotype the cultivars showed susceptibility to a different number of the phenotypes: from 3 (cvs. Veda, Yubileinaya 100) to 36 (cv. Don 105). The most phenotypes had a wide specialization in regard to the cultivars tested. Phenotype 19 infected the maximum amount of the cultivars (pp: 6,7,18,32,A) being virulent to 35 out of 39 tested cultivars (89.7 %). High virulence was shown by Phenotype 4 (pp: 6,7,18) that infected 34 cultivars and Phenotypes 13 (pp: 6,7,8,9,18), 16 (pp: 6,7,8,18,A), 31 (pp: 6,7,9,18), 33 (pp: 1,6,7,18,A) – each of them infected 31 cultivars. Weak virulence was recorded for Phenotypes 9 (pp: 6,7,27,32,A) and 10 (1,6,7,A) which infected 7 and 6 cultivars, respectively.

The examined cultivars showed good capability to select many virulence genes (up to 14 out of 16 tested). Depending on a cultivar genotype the pp genes 1, 6, 7, 8, 9, 17, 18, 27, 32, A were subjected to the selection to a greater degree; the genes p5, p10, p24, p26 – to a lesser degree. None of the tested 39 cultivars selected the virulence gene pSP.

## **P29. "Ug99", a Sr31-breaking race of the wheat stem rust fungus - An initial histological and molecular analysis**

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In 1999, a new race of the wheat stem rust fungus, that has overcome the resistance gene Sr31, was found in Uganda. This gene, introgressed from rye, had been effective for more than 30 years, and it is present in most mega-wheat cultivars today, except for Australia. Therefore, race "Ug99" poses a severe threat to the world's wheat production. It has already spread via Kenya and Ethiopia to Yemen in 2007, and it has reached Iran in 2008. "Ug99" has probably evolved from the progenitor race UVPgt55, a race identified in South Africa, as the only difference between both races seems to be the absence or ineffectiveness of Avr31 in "Ug99".

In an attempt to support the efforts of the Global Rust Initiative, which has been initiated to combat this threat, we have begun a study to try and identify molecular differences between "Ug99" and UVPgt55. As a first step, we successfully grew mycelia of both races in axenic culture, from which pure rust DNA was isolated and cloned. Both races were also grown on near-isogenic wheat lines with and without the Sr31-gene, fungal growth and host responses were carefully studied to pinpoint the timing of resistance responses. Two SSH-libraries were constructed, one containing mainly genes of "Ug99" that are active during infection, the other containing those of UVPgt55. Macroarrays revealed the strong homology between the two races, but some genes were identified which were differentially expressed. Out of 67 clones more strongly expressed in "Ug99" and 96 clones from UVPgt55, we selected those with a secretory signal peptide, as potential candidates for avirulence genes in UVPgt55. These genes are now being expressed heterologously to assess their Sr31-specific elicitor activities.

### **P30. Detection of the shortest Sr22-carrying segment in common wheat**

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Stem rust resistance gene *Sr22* transferred to common wheat from *Triticum boeoticum* and *T. monococcum* remains effective against commercially prevalent pathotypes of *Puccinia graminis* f. sp. *tritici*, including Ug99 and its derivatives. It was located on the long arm of chromosome 7A. Several backcross derivatives (hexaploid) possessing variable sized *Sr22*-carrying segments were produced. ESTs belonging to the deletion bin 7AL-0.74-0.86, corresponding to the genomic location of *Sr22* were screened for polymorphism. In addition, RFLP markers that mapped to this region were targeted to develop PCR based markers. Initial screening was performed on the resistant and susceptible DNA bulks obtained from three backcross derivatives identified to carry shorter *Sr22* segments. The RFLP probe csIH81, showing clear difference between the resistant and susceptible bulks, was converted into a sequence tagged site (STS) marker. Validation was carried out on backcross-derivatives in 13 genetic backgrounds. The STS marker distinguished all backcross-derivatives from their respective recurrent parents and co-segregated with *Sr22* in a Yarralinka (-*Sr22*)/Schomburgk (+*Sr22*)-derived recombinant inbred line (RIL) population. In addition, recombinants from a cross between *Sr22*TB and Lakin showing dissociation of *Sr22* with previously reported flanking microsatellites, were all found to contain the *Sr22* associated csIH81 STS marker. Several accessions of *T. boeoticum*, *T. monococcum* and *T. urartu* were screened using the csIH81 STS marker to predict the possible presence of *Sr22* in these genotypes.

### **P31. Development of molecular detection assays for environmental monitoring of cereal rust pathogens**

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Sentinel plot networks can function as effective early warning systems for crop disease outbreaks. An integrated and multi-faceted approach to air-borne pathogen surveillance combines field surveys enhanced by scouting of early-seeded sentinel plots for disease symptoms, networks of rainfall and air sampling equipment, and microbiological and molecular diagnostic screening. In Canada, such a program has been in operation for the surveillance of Asian Soybean Rust (ASR), caused by *Phakopsora pachyrhizi*, for 3 years. As a result, in 2007 the first detection in Canadian history occurred of an air-borne plant pathogen before a diseased plant was found, and was followed afterwards by confirmation of the first infected soybean plant in Canada. This result highlights the capability of such an early warning system to accurately forecast locations potentially at risk (or indicate where risk is low), and contribute to informed pest-management solutions. The efficacy of an environmental monitoring program depends on well-validated DNA-based assays specific at the diagnostic level of interest. To this end, we have been developing and testing a set of molecular assays for three cereal rust fungi, *Puccinia graminis*, *P. striiformis* and *P. coronata*. Sequence data for a number of genome regions including ITS,  $\beta$ -tubulin and RPB2, were analyzed for a broad sampling of collections from diverse hosts and geographic locations to assess phylogenetic relationships and characterize the population structure of each species complex. Based on our analyses, *Puccinia graminis* and *P. striiformis* each comprised 4 sub-specific lineages, while *P. coronata* comprised 12. Real-time PCR assays were developed with specificity for the lineages corresponding to the agriculturally important hosts. Assays were validated against DNA samples of reference specimens and the environmental samples collected for the ASR surveillance program. The ultimate goal is to develop a hierarchical and internally redundant set of molecular tests with specificity for the target species at the level of resolution required (e.g. varieties, pathotypes, genotypes) but also including markers at higher taxonomic levels to ensure detection of previously overlooked (e.g. due to sampling bias during the assay development phase) or newly evolving lineages within the target species. Our preliminary efforts to target population-level detection have focused on analyses of molecular markers derived from microsatellite flanking regions for *P. graminis* f. sp. *tritici*. Pyrosequencing of selected environmental samples has been undertaken to accumulate baseline data for the genetic diversity of air-borne organisms in our region. DNA array hybridization technology could be developed for multiple species detection in a single assay using a range of DNA markers that have specificity for various taxonomic levels, including populations.

### **P32. Histochemical and cytochemical studies on the accumulation of reactive oxygen species (O<sub>2</sub><sup>-</sup> and H<sub>2</sub>O<sub>2</sub>) in the incompatible and compatible interaction of wheat - *Puccinia striiformis* f.sp. *tritici***

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Wheat stripe rust, caused by *Puccinia striiformis* Westend f. sp. *tritici* Erikss., occurs world-wide and is considered a major disease in temperate regions, particularly in China. During the process of plants against pathogen infection, plants can induce a series of defence responses including rapid generations of reactive oxygen species (ROS), which are known as oxidative burst. It has been demonstrated that ROS play several important roles in defense responses during plant-pathogen interactions. The generation and accumulation of O<sub>2</sub><sup>-</sup> and H<sub>2</sub>O<sub>2</sub> were examined in the interactions between wheat cv. 'Suwon 11' and two races of *P. striiformis* f.sp. *tritici* (avirulent and virulent) by histochemical and cytochemical methods. At the pre-penetration stage during appressorium formation both stripe rust races induced H<sub>2</sub>O<sub>2</sub> accumulation in guard cells. In the incompatible interaction a rapid increase of O<sub>2</sub><sup>-</sup> and H<sub>2</sub>O<sub>2</sub> generation at infection sites was detected. The percentage of infection sites showing O<sub>2</sub><sup>-</sup> and H<sub>2</sub>O<sub>2</sub> generation was 36.1 % and 40.0 %, respectively, 12 h after inoculation (hai). At extended incubation time until 24 hai, percentage of infection sites showing H<sub>2</sub>O<sub>2</sub> accumulation further increased, whereas those exhibiting O<sub>2</sub><sup>-</sup> accumulation declined. The early infection stage from 12 hai to 24 hai coincided with primary haustoria formation in mesophyll cells. In contrast, in the compatible interaction O<sub>2</sub><sup>-</sup> and H<sub>2</sub>O<sub>2</sub> generation could not be detected in most of the infection sites. In the incompatible interaction, intensive DAB staining for H<sub>2</sub>O<sub>2</sub> was also determined in mesophyll cells, especially in cell walls, surrounding the infected cells 16-24 hai; thereafter, these cells contained fluorescing compounds and underwent hypersensitive response (HR). The number of necrotic host cells surrounding the infection sites increased continuously from 20 hai till 96 hai. It might be concluded that H<sub>2</sub>O<sub>2</sub> accumulation during the early infection stage is associated with the occurrence of hypersensitive cell death and that resistance response is leading to arrest the avirulent race of the obligate stripe rust pathogen. In the compatible interaction at 96 hai, H<sub>2</sub>O<sub>2</sub> accumulation was observed in mesophyll cells surrounding the rust lesion. At the subcellular level, O<sub>2</sub><sup>-</sup> accumulation could be detected in the tonoplast, the plasma membrane and the cell wall of mesophyll cells adjacent to hyphae and necrotic host cells, and main distribution of O<sub>2</sub><sup>-</sup> production was in the tonoplast of host cells. The accumulation of H<sub>2</sub>O<sub>2</sub> was observed mainly in the cell wall and the plasma membrane of mesophyll cells adjacent to hyphae and necrotic host cells, as well as in the tonoplast of some host cells and in the intercellular space. Accumulation of H<sub>2</sub>O<sub>2</sub> was also observed in the cell wall of hyphae. The result indicated that the subcellular localization of O<sub>2</sub><sup>-</sup> accumulation and H<sub>2</sub>O<sub>2</sub> accumulation in the compatible interaction were similar to the incompatible one, but there were striking differences in the content of ROS accumulation in different interactions.

### **P33. Fine mapping of the stripe rust resistance gene, *yrh52*, based on comparative analysis with rice, barley and *Brachypodium* genomes**

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Stripe rust of wheat, caused by the fungus *Puccinia striiformis*, is one of the most destructive diseases of wheat. Yield losses caused by stripe rust have ranged from 10% to 70% in susceptible varieties and a total yield loss has been reported when severe epidemics occur. Wild emmer wheat, *Triticum dicoccoides*, has been recognized as a particularly promising source for yellow rust resistance. The wheat stripe rust resistance gene *YrH52*, derived from wild emmer wheat, *T. dicoccoides* accession H52, confers resistance to a broad spectrum of stripe rust races. The gene *YrH52* was previously identified and mapped on the short arm of the chromosome 1B. A primary genetic map was constructed on the basis of a F6 recombinant inbred line (RIL) mapping population, developed by crossing *T. durum* cv Langdon with *T. dicoccoides* accession H52. Using *Nor* as RFLP probe, *YrH52* gene was located on the deletion bin 1BS.sat-0.31, distal to *XNorB1* region. Markers flanking a 23.5 cM chromosome segment around *YrH52* were identified. To further delimit the location of *YrH52*, we have exploited the colinearity between rice, *Brachypodium distachyon*, barley and wheat genomes and developed new EST based cleavage amplified polymorphic sequence (CAPS) markers. Our preliminary results, based on screening of RIL mapping population indicate that our newly developed markers are located in close vicinity to the gene *YrH52*. We are currently screening a large F<sub>2</sub> population, with the closest markers flanking *YrH52* in order to refine the genetic map of *YrH52* and develop sub-centiMorgan map suitable for physical mapping and positional cloning of *YrH52*.

The research leading to these results has received funding from the European Community's Seventh Framework Programme (FP7/ 2007-2013) under grant agreement FP7-212019 (CA 9.2).

### **P34. Histopathology and transcriptome analysis of wheat responses during compatible and incompatible race-specific interactions with *Puccinia striiformis* f. sp. *tritici***

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Race-specific resistance against wheat yellow rust (*Puccinia striiformis* f. sp. *tritici*) mediated by *Yr1* results in a strong clean phenotype with visible necrotic flecking but no fungal colonisation. We have used confocal microscopy and transcriptome analysis tools to characterise the biological and transcriptional responses of wheat during compatible and *Yr1*-mediated incompatible interactions. The initial stages of wheat yellow rust infection triggered a hypersensitive cell death (HCD) response in both compatible and *Yr1*-mediated incompatible interactions, although the response was earlier and more extensive during the incompatible interaction. Later stages of fungal development were only associated with a HCD response in the incompatible interaction, the HCD response being effectively suppressed in the compatible interactions. Mesophyll cell autofluorescence was observed in both cells in direct contact with fungal infection hyphae, primary HCD, and in adjacent mesophyll cells, secondary HCD, indicating the activation of cell-to-cell signalling. Transcription profiling highlighted a number of defence-related transcripts implicated in *Yr1*-mediated resistance, including classical pathogenesis-related transcripts and genes involved in plant defence. qRT-PCR time course analysis showed significant differences in *Yr1*-specific defence-transcript levels that were associated with *Yr1*-mediated resistance responses over time. Furthermore, this analysis indicated that the *Yr1*-avirulent isolate 232E137 more strongly induced the defence-related transcripts compared to the *Yr1*-virulent isolates even in the absence of the *Yr1* resistance gene. These data indicate that *Yr1*-mediated resistance against wheat yellow rust involves a two component system initiated by rapid HCD of cells in close proximity to invasive hyphae and supported by a second wave of HCD in cells associated with runner hyphae that had escaped the first layer of defence.

### **P35. Effect of silenced FAS-Associated Factor 1 upon *Blumeria graminis* infection in barley**

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FAS-Associated Factor 1 (FAF1) is a CC-NB-LRR protein involvement in apoptosis in addition to other roles. Previous research in our lab showed an augmentation in mRNA levels of FAF1 gene in fungus infected wheat, suggesting a role of this gene in the resistance mechanism. We hypothesized that the apoptotic role of FAF1 protein in metazoan is conserved in plants by including FAF1 as a factor in hypersensitive response. Barley lines Pallas01 and Pallas03 which are respectively resistant and susceptible against fungus *Blumeria graminis hordei* 103 (*Bgh103*) were used for silencing of FAF1 by Barley Stripe Mosaic Virus induced gene silencing method (VIGS) to test if the hypersensitive response or disease formation against *Bgh103* was affected. In this aspect, the formation of death lesions on the Pallas01 leaves due to fungal resistance was not prevented demonstrating that FAF1 silencing with VIGS in the resistant Pallas01 line of barley is not sufficient to stop apoptosis. On the other hand, the FAF1-silenced barley susceptible line Pallas03 became more sensitive to fungal stress; based on the slightly increased conidia observation after trypan blue staining of the infected leaves. The UBA and UBX domains of FAF1 might be involved in the susceptibility based on the reported data obtained about these domains' functions in other UBX family proteins. These results suggest that FAF1 is a catalyst in the hypersensitive response and its loss of function makes barley more susceptible to fungal stress.

### **P36. The influence of climate changes on the powdery mildew (*Blumeria graminis*) occurrence and harmfulness in last years in Poland**

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The agrophages economic importance is determined by various environmental factors, namely: intensified farming, simplified agro technology, varieties with different level of disease and pest resistance and the global warming effect within the last few years.

The global climate changes give rise to new challenges for plant protection. Temperature, humidity of the air and soil are very important factors affecting agrophages developmental rate, quantity, population dynamics, range and intensity of occurrence, feeding intensity and harmfulness.

In Poland every year Plant Protection and Seed Health Inspection Service provide detailed field observations in order to get the information about phytosanitary state of agricultural plants. Obtained results from pests/diseases monitoring in connection with observations provide at the Plant Protection Institute at the Department of Forecasting and Registration Pest and Diseases, are the base of “Phytosanitary state of agricultural plants in Poland with prognosis to the next year” which is issued every year.

In Poland the harmfulness and occurrence of the agrophages has been monitored from 1950. Such information are the base of the evaluation the tendency of pests and diseases spread as well as their economic value affected among other factors by climate changes.

Powdery mildew caused by fungi *Blumeria graminis* is one of the most important and dangerous disease occurred every year in Poland, especially on wheat and barley.

Analysis for average month air temperature and sum of month rainfall in autumn (September, October, November) and spring (March, April, May) months for places (Koszalin, Poznań, Warszawa, Wrocław, Rzeszów) for four last sun 11-years cycles was done and its influence on powdery mildew occurrence and harmfulness in Poland.

## **P37. Occurrence and virulence changes of leaf rust in Hungary during 1999-2008**

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The rusts of wheat are important pathogens that can be spread across continents by wind. Rusts are obligate parasites that interact with resistance genes in wheat in a gene-for-gene manner.

Survey of wheat rusts fungi confirmed that all the three rusts of wheat occur in Hungary, but their importance varies year by year. Ten years ago stem rust completely disappeared because Hungarian wheat varieties have excellent resistance to stem rust. Most of cultivated varieties contain resistance gene *Sr31* or *Sr36*, but many cases the genetic background of the resistance is still unknown.

Leaf rust (brown rust) caused *Puccinia triticina* currently is the most important disease of wheat in Hungary. This pathogen emerges every year; its spread and the extent of damage are influenced by the susceptibility of the cultivated varieties and environmental conditions.

Isolates of leaf rust were obtained from wheat varieties in various wheat growing regions of Hungary every year during 1999-2008. Pathotypes were identified on 15 near-isogenic Thatcher backcross lines with resistance genes *Lr1*, *Lr2a*, *Lr2b*, *Lr2c*, *Lr3*, *Lr9*, *Lr11*, *Lr15*, *Lr17*, *Lr19*, *Lr21*, *Lr23*, *Lr24*, *Lr26*, *Lr28*. Ten-fifteen different pathotypes were identified every year. The most widespread pathotypes were as follows: 43722, 43702, 53522, 73722, they were found in all wheat growing regions of Hungary. The wheat leaf rust populations were virulent on *Lr1*, *Lr2b*, *Lr11*, *Lr15*, *Lr17*, *Lr21*, *Lr23* and *Lr26*. The resistance genes *Lr2a*, *Lr9*, *Lr19*, *Lr24* and *Lr28* were highly effective among the tested *Lr* genes till 2006. However, in the last two years rust isolates were identified with virulence to resistance genes *Lr24* and *Lr28*. The number of virulent pathotypes to *Lr1* and *Lr2b* increased; but declined to other *Lr* genes from 1999 to nowadays.

During the past ten years, lots of susceptible varieties were removed from cultivation and resistant cultivars with unknown resistance genes were introduced. The introduction of new highly resistant varieties into cultivation changed the diversity of virulence patterns in Hungarian wheat rust populations.

### **P38. Incidence and development of powdery mildew (*Blumeria graminis* f.sp. *tritici*) epidemic in the Prague region in single years of the decade 1999 – 2008**

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Incidence and development of powdery mildew was studied in small-plot experiments during ten years in the period from the second half of May to the end of July on the cultivar Kanzler, susceptible standard to this disease. Incidence of powdery mildew was evaluated as disease severity and development of the disease by measuring disease severity in several time intervals. Disease severity was expressed as cumulative percentage of leaf area disease - CPLAD. The highest disease severity was in 2004 and the lowest in 2003. The highest disease severity in the first evaluation was in the year with the highest values of powdery mildew (2004). Low disease severity was in the years 2000, 2007 and 2005. Higher disease severity was in 2001 but it was lower than in 2004. Development of the disease in some years, such as 1999, 2001, 2002, 2004 and 2007, was gradual. On the contrary, in the years 2000, 2003 and 2005 in some time intervals development of the disease has stopped. But in the years 2006 and 2008 were two measurements made, only. From of weather factors measured for ten days before date of evaluation average temperature had high influence on progress of epidemic. For example the lowest temperature for followed period was in the years with the highest disease severity, it means in 2004 and 2001. Vice versa the highest temperature was the year 2003 when disease severity was the lowest.

### **P39. Determination of reactions some of wheat genotypes against wheat stem rust (*Puccinia graminis* f.sp. *tritici*) in Kastamonu epidemic conditions in 2007 and 2008**

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Wheat (*Triticum* spp.) is the world's one of the most important and strategic crop. Stem rust (*Puccinia graminis* f.sp. *tritici*) is very important which is one of the most significant biotic factors affecting wheat yield and quality and might cause yield production decrease by 70% under epidemic condition.. Among all the control measures of this disease, genetic resistance is the only economic and practical control measure, causing no additional cost to the farmer. Stem rust was effective every year in highlands of Kastamonu (Seydiler) region.

In this study, reactions of some registered cultivars/advanced lines and effective resistance genes against to local stem rust population were determined in 2007 and 2008. The genotypes were planted and tested in the farmer field in Seydiler (1.100 m) under the natural stem rust epidemic condition. 1.239 and 720 wheat genotypes were assessment to Stem Rust (*Puccinia graminis* f.sp. *tritici*) in 2007 and 2008 years, respectively. In addition to this, trap nursery which includes most of the resistant genes was planted. High disease pressure was observed in two years. The susceptible checks Little Club and Michigan Amber gave readings of over 80 S in general. According to the result, 144 and 139 wheat genotypes were found to be resistant to stem rust in 2007 and 2008 respectively. Sr 24, Sr 27, Sr 31 stem rust resistance genes were still effective to stem rust races in Kastamonu under the natural epidemic condition in 2007 and 2008 years.

Most of the older Turkish bread wheat varieties included in the study were susceptible. Of the newly some registered varieties, Demir 2000, Mızrak, Tosunbey and Zencirci 2002 were susceptible with a score of 60 S-80 S, while Bayraktar 2000, Seval, Karacabey, Tahirova 2001 were found to be resistant in this study.

**Key words:** Stem rust (*Puccinia graminis* f.sp. *tritici*), Wheat (*Triticum* spp), Resistance, Kastamonu

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## P40. Epidemiology of wheat powdery mildew in Tajikistan

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Wheat is a staple food crop in Tajikistan and has significant contribution to food security. Wheat area in Tajikistan has expanded during the last 10 years, but due to a number of biotic and abiotic stress factors as well as inferior seed quality, the grain yield still remains relatively low, with an average in 2008 of 1,9 tons/ha . Among the diseases, powdery mildew caused by *Blumeria graminis* is becoming one of the most important fungal diseases of wheat in Tajikistan. Powdery mildew can damage wheat at any growing stage from seedling to ripening of seed, and the disease can lead to losses of grain yield and quality. In Tajikistan powdery mildew epidemics are commonly observed on winter wheat. High humidity caused by high level of precipitation and cold spring as well as a crop with high plant density intensifies pathogen development. Monitoring of diseases was conducted by our research group in three agro ecological zones of Tajikistan in breeding nurseries and farmers’ fields. In the wet years (2003 and 2009) frequent outbreaks of powdery mildew were observed in the Northern and Central Tajikistan but not in the South, leading to declining wheat yields.

In 2003 in the North of Tajikistan (Isfara) 30% of entries in the breeding nurseries were damaged by powdery mildew and the most susceptible variety was the local variety Navruz with infestation levels up to 50% in multilocation yield trial. In 2009 powdery mildew started spreading in susceptible varieties around the booting stage causing reduced kernel size and low grain yield. According to the evaluation made in the spring of 2009 there is a clear varietal difference in resistance to powdery mildew, with certain varieties being moderately susceptible like Gelibolu (30%), Sadokat (40%) and Krasnodar 99 (40%), and others being susceptible like Navruz (80%), Alex (60%) and Iqbol (70%). The variety Konya-2002 was highly susceptible to powdery mildew and was damaged almost 100% causing complete crop failure. These results demonstrate the value of evaluating germplasm for genetic resistance to powdery mildew providing information that can facilitate the recommendation of resistant varieties to farmers. Furthermore the information of resistant germplasm can be utilized in a crossing program to increase the frequency of genes resistant to powdery mildew in future varieties.

## **P41. Variety and species mixtures as the possibility of powdery mildew (*Blumeria graminis*) incidence reduction in cereals**

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In Poland monocultures in cereal crops are popular due to the technical and organizational reasons. They are easier in crop husbandry, quality and product use, but on the other hand they are more susceptible to diseases, pests and sometimes weed infestation. In order to keep high and stable grain yields and quality in monoculture one has to use high inputs. Experimentally and practically it has been proved that variety and species mixtures can constitute an alternative to cultivar growing in pure stands. It has been found that in mixtures operate different epidemiological and ecological factors, which lead to considerable disease reduction, pest and weed control, which finally result in higher and more stable grain yields than in cereals grown in pure stands.

In last years in Poland about 17% of cereals growing area was mixtures. The mixtures are designed particularly for the control of powdery mildew, but more general recommendations for their use are:

- broader genetic variation, a more resistant variety can be used as “physical barrier” for pathogens,
- yields of the mixtures are usually higher and more stable compared with the individual pure stands of the components,
- better overall disease performance resulting in reduced need for fungicides (lower costs and better environment impact),
- variety mixtures can be cultivated in the same agronomic and husbandry way as pure stands [Gacek et al. 1996].

Appropriate variety and species mixtures can considerably restrict the development of powdery mildew (*Blumeria graminis*) and to some extent other airborne diseases [Gacek i in. 1996]. Cultivar mixtures can provide functional diversity that limits pathogen and pest expansion by making use of knowledge known about interactions between hosts and their pests and pathogens to direct pathogen evolution. Indeed, one of the most powerful ways to reduce risk of resistance break-down and to still make use of defeated resistance genes is to use cereal variety and species mixtures [Finckh et al. 1999, 2000, Newton et al. 2002].

The results of field experiments designed to evaluate epidemiological and economical effects of variety and species mixtures are presented. The aim of the studies was to evaluate the possibility of reduction of powdery mildew (*Blumeria graminis* f.sp. *tritici*).

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## **P42. Evolutionary forces and emerging pathotypes of *Blumeria graminis* f.sp. *hordei***

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Powdery mildew, frequently the most damaging disease of barley in the Czech Republic as well as in other parts of Central and North West Europe, is caused by the highly adaptable and ubiquitous obligatory parasite *Blumeria graminis* f.sp. *hordei* (*Bgh*). To limit the damage to barley, genetic resistance is an effective, economically sound and safe alternative to fungicide application. However, the pathogen can adapt to specific resistance genes (and also to active ingredients of fungicides), reflect the corresponding host genetic structure and overcome its resistance in the field due to operation of all known evolutionary forces.

Current changes in the Czech population of *Bgh* can be considered gradual. It is mostly due to the prevailing area of cultivars that do not arouse a directional selection, but as well as the absence of dominant cultivars carrying specific resistance genes. It results in slow decrease of the virulence complexity for most of 12 earlier significant *Ml* resistance genes (*a1*, *a3*, *a6*, *a7*, *a9*, *a12*, *a13*, *kl*, *La*, *g*, *at* and *Bw*). The virulence complexity to these resistance genes was about 0.90 in 1971, culminated (9.27) in 2000, and since that time it has been decreasing (to 7.32 in 2007). However, it takes place along with increasing the population diversity because most of the virulences, at their frequency decline, approach to the level of 50% (optimal virulence frequency with respect to maximizing the population diversity). At the same time the virulence frequency for newer resistance genes (whose frequency is below 50%) has been gradually rising. Therefore, the increase in these virulence frequencies induces not only the growth of the virulence complexity but also the growth of the population diversity.

The present Czech (Central European) population of *Bgh* is characteristic of both gradual extending the spectrum of virulences (increasing the virulence complexity due to a larger number of present virulences) and optimization of their frequencies (approaching the frequencies of individual virulences to 50%). It leads towards the increase in the population diversity as a whole, i.e., the increase in the number of existing pathotypes as well as their even proportions (decreasing the frequency of the most abundant pathotypes and increasing the value of evenness). A result is the increase in adaptation potential of the current pathogen population to resistance genes in grown cultivars. It can decrease the efficiency of intraspecific diversification of barley with regard to protection against powdery mildew. It can be assumed that the suggested trend in the development of Czech population will continue also in next years.

### **P43. Resistance to powdery mildew in selections from barley landraces collected in Georgia, Azerbaijan, Iraq and Iran**

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Barley powdery mildew is caused by the pathogen *Blumeria graminis* f. sp. *hordei*. It is one of the most destructive foliar diseases of barley in regions with a maritime climate. This pathogen is showing the high level of pathogenic variability. Consequently, geneticists, plant pathologists, and breeders working with barley are constantly looking for gene pools from which new resistance genes to powdery mildew can be introduced into existing cultivars. Many investigations showed that barley landraces are a valuable source of resistance to powdery mildew for cultivated barley (Czembor 1976, 1996, 2005).

*Seed samples of 126 barley landraces were used for screening for resistance to powdery mildew. These landraces originated from 4 countries: Georgia (28), Azerbaijan (39), Iraq (24) and Iran (35). All landraces originated from International Center for Agricultural Research in the Dry Areas – ICARDA, Aleppo, Syria.. After preliminary testing 16 single plant lines resistant to powdery mildew were obtained. These lines originated from landraces collected in: Georgia (9), Azerbaijan (1), Iraq (3) and Iran (3). These lines were increased in field and during winter were tested in greenhouse in seedling stage with 20 differential isolates of powdery mildew. These isolates had virulences corresponding to all major resistance genes used in Europe. These lines showed high level of resistance to most of isolates used. The most common infection type observed was 2 - which in most cases was represented as different types of chlorosis.*

*The results presented here come from the tests performed on the seedlings and investigation of field performance of the selected resistant lines is needed. Also, presence of partial resistance in tested lines may have influence on conclusions concerning postulation of presence of specific resistance genes. Final confirmation of resistance composition of tested lines should be established through crosses and backcrosses among appropriate hosts (Czembor 1996, 2005).*

*This study confirmed findings of other investigators that many barley landraces possess mildew resistance genes different from genes present in cultivated varieties. New sources of resistance identified in this investigation may increase the diversity of the powdery mildew resistance genes present in barley cultivars in Europe.*

Czembor H.J. 1976. Sources of resistance to barley powdery mildew *Erysiphe graminis* f. sp. *hordei*. Hod. Rośl. Aklim. Nasien. 20: 467-490.

Czembor J.H. 1996. Presence and expression of resistance genes to powdery mildew of barley in selections from Tunisian barley landraces. Ph.D. thesis, Montana State University, Bozeman, USA, p 1-144.

Czembor J.H. 2005. Powdery mildew [*Blumeria graminis* (DC.) E. O. Speer f. sp. *hordei*] resistance in landraces of barley (*Hordeum vulgare* L.) – habilitation monography. Monographies and Dissertations of IHAR no. 2005(24), p 1-164.

## **P44. Barley landraces from Israel as sources of resistance to European isolates of *Blumeria graminis* f.sp. *hordei***

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Barley (*Hordeum vulgare* L.) is an economically important crop. In many regions barley is grown in marginal agricultural areas. Landraces of barley are important in many mountain regions, including Ethiopia, because they are often the only crop possible to be cultivated on slopes at high elevations (Czembor 1996, 2005).

Powdery mildew, caused by the pathogen *Blumeria graminis* f. sp. *hordei*, is one of the most destructive foliar diseases of barley in Central and Western Europe. In countries where mildew is a problem, yield losses in experimental tests often exceed 25%, although average losses in barley production are smaller and about 10%.

*Barley landraces constitutes a rich genetic resource, and many examples of their successful use have been reported. However only for less than 2 percent of barley landraces the attempts were made to identify powdery mildew resistance genes using differential lines and isolates. These types of studies were mostly conducted in Germany, Denmark and Sweden and on smaller scale in other countries such as Czech Republic, The Netherlands, USA and Poland (Czembor 1976, 1996, 2005).*

*Seed samples of 22 barley landraces were used for screening for resistance to powdery mildew. All landraces were collected in Israel and originated from Centre for Genetic Resources the Netherlands (CGN). In preliminary study, about 30 plants per landrace were evaluated in greenhouse with isolate 33. Isolate 33 represented the most avirulent isolate available allowing the expression of maximum number of resistance genes.*

From 14 resistant landraces single plant lines were selected. These lines were tested in seedling stage with 21 differential isolates of powdery mildew. The isolates were chosen according to their virulence spectra on the Pallas isolines differential set and 7 additional differential cultivars. These isolates had virulences corresponding to all major resistance genes used in Europe. Twelve tested lines were resistant to all isolates used.

This investigation identified new sources of resistance to barley powdery mildew in lines selected from barley landraces collected in Israel. These new sources may contribute significantly to the diversity of the powdery mildew resistance gene pool available for barley breeders.

Czembor H.J. 1976. Sources of resistance to barley powdery mildew *Erysiphe graminis* f. sp. *hordei*. *Hod. Rośl. Aklim. Nasien.* 20: 467-490.

Czembor J.H. 1996. Presence and expression of resistance genes to powdery mildew of barley in selections from Tunisian barley landraces. Ph.D. thesis, Montana State University, Bozeman, USA, p 1-144.

Czembor J.H. 2005. Powdery mildew [*Blumeria graminis* (DC.) E. O. Speer f. sp. *hordei*] resistance in landraces of barley (*Hordeum vulgare* L.) – habilitation monography. *Monographies and Dissertations of IHAR* no. 2005(24), p 1-164.

## **P45. Powdery mildew resistance - progress and results of spring barley breeding in the Hordeum Ltd., plant breeding station**

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The annual area sown to spring barley during 2000 - 2008 years was cca 194,000 hectares in the Slovak Republic. The most frequent and main disease on spring barley is powdery mildew caused by *Blumeria graminis* DC.f.sp.hordei Marchal. The aim of this work is illustrate the progress and the results in spring barley breeding for resistance to powdery mildew in the HORDEUM Ltd., Plant Breeding Station in Sládkovičovo where barley breeding has a long tradition. Eleven new varieties were listed in the List of Registered Varieties during above mentioned period. Varieties Sladar, Donaris, Levan, Slaven, Ezer and Ludan carrying the resistance Mlo which is effective against all known pathotypes. Variety Nitran is heterogeneous, composed of more components, one is Mlo. Varieties Poprad, Pribina, Nadir, Cyril carrying resistances which can not provide protection against barley powdery mildew. In the poster are presented five breeding lines in State Variety Trial 2009. The majority of them have high level of resistance based on mlo gene, one breeding line has the same resistance as variety Pribina . Significant differences were identified in fifty breeding lines resistances tested in pre-official trials in last three years. Resistance which can provide protection was found in thirty-nine breeding lines (78%). Comparing with the previous years it is considerable increase. According to the pedigree and evaluation of pathogen isolates reaction types one line probably carrying resistance derivated from *Hordeum vulgare* ssp. *spontaneum*. Unknown resistances were identified in two lines, two lines were heterogeneous and six lines possess non-effective resistances.

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## **P46. Effectiveness of oat resistance sources to powdery mildew and crown rust in Tunisia's conditions**

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Oat crown rust (*Puccinia coronata* fsp *avenae*) is present in Tunisia on cultivated, spontaneous and on volunteer oats. Aecial stage of oat crown rust was found on the *Rhamnus lycioides*, a forest plant native in the mountainous northern part of the country. This ubiquitous pathogen habitually constitutes a threat to cultivated oats. Powdery mildew (*Erysiphe graminis* fsp *avenae*) is the second fungus disease on oats causing important damages particularly before flowering. Crown rust and powdery mildew were studied during two years on 15 oat lines with different predisposition to powdery mildew and on 16 *Pc*-gene oat lines with variable effectiveness to crown rust. On susceptible lines, powdery mildew infected the leaves early in the season (January) and tended to invade the basal part of the foliage during the cold period before decreasing its severity to become rare at the beginning of April when warm weather prevails. Meanwhile, crown rust epidemics occurred on susceptible plants in March to generalize attacks in April then remained active performing polycyclic infections even on the upper leaves until maturity at the end of May. The rainy year (> 500 mm) was favourable to crown rust; the relative dry year ( $\approx$  300 mm) was more suitable for the development of powdery mildew. Different oat lines proved their resistance to powdery mildew such as Mostyn, Cc 4761, Melys, OM 1387, OM 1621, APR 122, APR 166. The lines Manod and Cc 4146 belonging respectively to the OMR-1 and OMR-2 oat mildew resistance groups were relatively susceptible with a severity of 20 and 10 % respectively. The above-mentioned lines having resistance to powdery mildew were, at the same time, highly attacked by crown rust whose severity ranged between 50 and 90 %. For the *Pc*-gene lines, the *Pc*54 showed a slight attack with powdery mildew around 5 % but expressed a high crown rust severity of 45 %. Furthermore, other lines possessed intermediary reaction towards these two pathogens with a powdery mildew severity nil to 6 % and a crown rust severity between 5 and 20 %, i.e. Maelor, Maldwyn, Roxton, SG K 95708, *Pc*56. Results showed that many sources of resistance are still effective against crown rust and/or powdery mildew, however cumulating the two types of resistance in one cultivar should improve oat productivity.

## **P47. Pathogenicity of powdery mildew (*Blumeria graminis* (DC.)Speer) on triticale (x *Triticosecale* Wittm.) in Poland**

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*Blumeria graminis*, which causes powdery mildew is one of the most severe disease in cereals. In short time powdery mildew became an important widespread disease of triticale in Central Europe. From early 80's when the first Polish triticale varieties were registered, the cultivation area increased very quickly to more than one million hectares. For many years in comparison with parental species, triticale was generally known as free from diseases. However, at first the appearance of the septoria then leaf rust and fusarium were recorded. In years 2002-2005 on the basis of the observations in different regions of Poland a rapid increase of powdery mildew on triticale were noted on many wide grown cultivars. One of the best Polish variety 'Lamberto' after several years of extensive cultivation in Poland and Germany became very sensitive to *B. graminis* population. Nowadays fungus *B. graminis* may cause yield losses in triticale production up to 25%. The plant-pathogen interaction is controlled by specific resistance genes in the plant and matching fungal avirulence genes according to the gene-for-gene hypothesis. Identification of race-specific resistance and pathogenicity of powdery mildew on triticale is of a major importance in triticale breeding for efficient pathogen control strategy.

The objectives of this study were to define pathogenicity of powdery mildew populations collected from different regions of Poland. The isolates were obtained from diseased leaves of susceptible variety 'Lamberto' collected from 19 various parts of the country. From infested leaves from each place, around 40 single spore isolates were received. During the experiment isolates were stored on leaves of susceptible variety placed in 0,5% agar. Each single spore isolate was tested on differential set of 28 varieties. After 8–10 days of incubation, the disease reaction types of seedlings were scored. Scoring was done according to a 0–4 scale adopted from Mains and Dietz (1930). Plants with infection types 0–2 were classified as resistant, while plants that scored 3 and 4 were classified as susceptible.

The research was supported by the grant of the Polish Ministry of Education (grant No. HCU3148).

## **P48. Virulence of the powdery mildew (*Blumeria graminis*) population on Triticale in Belgium (Flanders)**

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In the past, triticale was known as a very healthy crop regarding to leaf diseases. However, as its area grows, triticale is increasingly exposed to various pathogens, and the opportunities for these pathogens to produce physiological variants with severe virulence on major cultivars are enhanced accordingly. During the last growing seasons more varieties become heavily infected and show high susceptibility to powdery mildew in Belgium. Severe outbreaks of the disease in epidemic proportions was also observed in Poland and Germany. The situation in triticale may become more serious than in wheat because fewer disease resistance mechanisms are known today. This is why resistance breeding will be the most important issue in the coming years. The obligate, biotrophic fungus *Blumeria graminis* (DC) Speer (Syn. *Erysiphe graminis* DC), is the causal agent of powdery mildew in cereals and grasses (subfamily *Pooideae*). Strict host specialization was described for *B. graminis*, with infection limited to a single host genus. Based on the host specialization, eight *formae specialis* (ff.spp.) were distinguished within *B. graminis*. Triticale was believed to carry resistance to pathogens superior to that of the parental species, but susceptibility of triticale to *Blumeria graminis* ff.spp. *tritici* and *secalis* has been recorded. A profound study started to find out more about the sudden resistance breakthrough of triticale to powdery mildew to become a better insight of the host-pathogen interaction. The variation in reaction of different genotypes of triticale, wheat and rye to isolates obtained from natural populations of *Blumeria graminis* on triticale, wheat and rye was determined by inoculation of leaf segments.

## **P49. Molecular characterization of *Blumeria graminis* f. sp. *hordei* using AFLP markers**

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Turkey is the sixth country among top ten barley producers (FAO-Stat). Among many diseases, powdery mildew decreases the yield of barley nearly 40 % in temperate regions worldwide (BluGen). The disease also is a treat to the region. The screening of powdery mildew virulence is not periodically conducted toward testing newly emerging races in the country. Therefore, our study might be the first one on detecting local pathogen variation, their genetic similarities and distances to the isolates with known virulances. The samples of *Blumeria graminis* f. sp. *hordei* (*Bgh*) were collected from Cukurova region, near the Mediterranean Sea. The genetic characterizations of were carried out using AFLP fingerprinting after the purified isolates were obtained. The thirty eight of them in comparison to nine universal isolates with known virulence genes (differentiating races) were analyzed in this study. The AFLP profiles showed unique bands only present in some of the isolates, these fragments may be utilized as “isolate specific markers” once they are further characterized. Using 16 AFLP primer sets, isolates from Cukurova were clustered as three major groups, distantly related to the differentiating races. We believe that these analyses can be valuable to assess newly emerging isolates in future studies, also for genetic relationship and evolutionary analysis.

## P50. Induction of resistance to leaf rust in wheat by barley powdery mildew

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Induction of resistance by primary inoculation with barley powdery mildew *Blumeria graminis* f. sp. *hordei* (*Bgh*) has been published in wheat against a second infection by wheat powdery mildew or stem rust (Érsek 1973, Barna et al. 1996) and in barley by an incompatible *Bgh* race against a compatible *Bgh* race (Ouchi et al. 1976). In this work we report on the induction of resistance to leaf rust in wheat, a non-host by barley powdery mildew pathogen.

Seven wheat cultivars or breeding lines (MvC 1535-02, Mv 336-02, Mv 12-02, Mv22-2001, Mv Emma, Mv Mambo és Mv Mezőföld) and a barley cultivar (Ingrid), as well as 2 races (12 and 61) of wheat leaf rust and a race mixture of barley powdery mildew were used in the experiments. Primary inoculation with *Bgh* induced resistance in all wheat genotypes at first and third leaf stages against both leaf rust races. The pretreatment with *Bgh* did not change the reaction type, but the number of pustules were reduced by 10 to 90 % depending on the genotype, leaf stage and rust race.

Light and UV microscopy revealed that germination of conidia was partly inhibited and hypersensitive reaction of epidermal (and some mesophyll) cells and papillae formation could be detected on *Bgh* infected wheat leaves. Interestingly enough the above non-host incompatible reactions depended largely on wheat genotype. At sites of penetration attempts accumulation of H<sub>2</sub>O<sub>2</sub> was shown by DAB staining in accordance with the finding of Trujillo et al. (2004). In addition, ascorbate peroxidase (APX), catalase (CAT) and glutathione S-transferase (GST) enzyme activities were augmented in wheat leaves already one day after *Bgh* infection, similarly to the barley-*Bgh* hypersensitive incompatible interaction (Harrach et al. 2008).

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## P51. High-resolution mapping of the wheat *Lr46* pleiotropic rust resistance locus

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Rust diseases are the most important diseases of wheat globally, and genetic resistance is the most effective method for controlling all three rusts. Numerous resistance genes have been characterized and reported. For typical resistance genes, the mechanism of resistance and the basis of race specificity are due to gene-for-gene interactions between host resistance (*R*) gene products and pathogen avirulence (*Avr*) gene products. Durable rust resistance based on *R* genes remains elusive in wheat, but some resistance genes are thought to be durable because they are not dependent on the recognition of a single *Avr* gene product. The pleiotropic *Lr46* gene confers durable, race non-specific resistance to leaf rust and stripe rust but little is known about its mechanism of action. Using a population of 3931 lines with a genetic resolution of approximately 0.01 cM, fine-scale mapping of the *Lr46* locus was carried out for both the development of molecular markers and map-based cloning of the gene. Existing markers were used to probe wheat hexaploid and tetraploid bacterial artificial chromosome (BAC) libraries and, following low-pass sequencing of selected BACs, contigs were assembled that were the source of many additional markers. Because no recombination was detected between BAC-derived markers, synteny was explored with the model grass genome, *Brachypodium distachyon*. Markers spanning the *Lr46* locus were colinear with a 90 kbp physical region from *Brachypodium* supercontig 2, which was used to identify wheat ESTs and develop new markers. Subsequently, we delimited *Lr46* to a 20 kbp physical region of *Brachypodium distachyon*. The newly developed markers were evaluated on diverse wheat germplasm to test their efficacy for marker-assisted selection for the *Lr46* locus.

## P52. QTL analysis of powdery mildew resistance in an introgressive line of bread wheat

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Tetraploid wheats of the timopheevii group are considered a useful reservoir of disease resistance genes. The introgressive line 8/1, a derivative of the susceptible spring wheat cultivar 'Tähti' and *Triticum militinae* (2n=28 A<sup>1</sup>A<sup>1</sup>GG), is characterized by extremely high resistance to *Blumeria graminis* (DC) E. O. Speer f.sp. *tritici* (*Bgt*) both at the seedling and adult plant stages of plant growth. Screening the genome of the line 8/1 with 180 microsatellite markers, *T.militinae* translocations on chromosomes 1A, 1B, 2A, 4A, 5B, 5A and 7A were detected. QTL analysis of a F<sub>2</sub> mapping population indicated at least three significant for resistance loci including the powdery mildew resistance locus *QPm.tut-4A* which was mapped on distal end of long arm of wheat chromosome 4A in region flanked by markers *wmc232* and *gwm160*. The region explained up to 50% of the quantitative trait variance.

In order to detect recombinations in the QTL regions, the initial F<sub>2</sub> population was enlarged and a population of recombinant DH lines (335 genotypes) was developed from the F<sub>2</sub>-F<sub>4</sub> plants.

DH lines were screened for seedling resistance to 3 different isolates of *Bgt*, each tested by two single-conidia-derived subisolates. Adult plant resistance was tested in the field under natural infection. As a result, we have shown that powdery mildew resistance in the introgressive line 8/1 is a quantitative trait in all stages of plant development. The seedling resistance response to the analysed *Bgt* isolates showed no race-specificity. Both at the seedling and in adult stage of plant growth, the main effect on the resistance response is explained by the *T. militinae* allele at the *Xgwm160* locus.

### **P53. Effectiveness of resistance specific genes wheat lines to population of powdery mildew (*Blumeria graminis* f.sp. *tritici*)**

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For identification of reactions of *B. graminis* f.sp. *tritici* under field conditions the range of sixteen winter and spring lines of wheat with specific genes resistance was used. Reactions of single lines in six years (2003-2008) of experiments were different and were influenced by weather progress. The highest disease severity was in 2003 on the line Sonora (*Pm3c*), in 2004 on Khapli (*Pm4a*), in 2005 on Michigan Amber (*Pm3f*), in 2006 on Khapli (*Pm4a*), in 2007 on Chul (*Pm3b*) and in 2008 on Axminster (*Pm1*). The lowest disease severity was in 2003 (zero value) on Asosan (*Pm3a*), Rektor (*Pm5*) and Amigo (*Pm17*), in 2004 on Ralle (*Pm3d*) it was zero, in 2005 on Amigo (*Pm17*) it was zero, in 2006 on Amigo (*Pm17*), in 2007 on Amigo (*Pm17*) and in 2008 on Amigo (*Pm17*). Effectiveness of gene resistance *Pm4a* (Khapli) and *Pm2a* (Ulka) to population of powdery mildew was low in monitored years. On the contrary, genes *Pm17* (Amigo), *Pm3d* (Ralle), *Pm2+Mld* (Maris Dove), *Pm6* (NK-747), *Pm5* (Rektor) and *Pm4b* (Ronos) affected high protection to local population of powdery mildew. The genes of resistance *Pm2,6* (Maris Huntsman), *Pm2*, 4b, 8 (Apollo), *Pm3a* (Asosan), *Pm3b* (Chul), *Pm1* (Axminster), *Pm3c* (Sonora) and *Pm8* (Disponent) expressed medium resistance to powdery mildew. Experiments indicated that most genes of resistance in differential assortment of wheat provided middle or high resistance to local powdery mildew population.

## **P54. VIGS of an F-box protein decreases powdery mildew resistance in barley**

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In our previous studies, we found a new Zeitrlope (ZTL) type F-box protein which is expressed at a higher level upon avirulent pathogen infection (Bozkurt et al., 2007). F-box proteins mark the proteins to be degraded through 26S proteasome system by ubiquitination. Since the information on the role of ubiquitin mediated proteolysis in disease responses is advancing rapidly, we sought to understand the way which F-box functions in resistance response as part of ubiquitin-proteasome pathway. Interestingly, in response to silencing of this F-box gene *via* BSMV mediated virus induced gene silencing (VIGS) method, barley plants lost resistance in incompatible pathogen inoculations. The Pallas-01 line having *Mla1 R*-gene showed hyphae formations when inoculated with avirulent powdery mildew race, Bgh1013, after 4-fold silencing. This observation suggests that F-box protein plays a key role as a positive regulator in disease resistance by ubiquitinating a target, yet to be characterized.

## P55. Identification of donors and breeding material resistant to wheat stripe rust

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Yellow, or stripe, rust of wheat (*Puccinia striiformis* f. sp. *tritici*) is one of the most widespread and dangerous diseases of wheat. The region of Central Asia is one of the most important wheat areas in the world. In this area, wheat stripe rust over the past few years is the major factor that adversely affects wheat yield and quality and causes considerable economic damage; yield losses reached 30-50%. Effectiveness of the most known Yr-genes is break down due to changes of pathogen race composition. It is therefore necessary to choose new sources of resistance to yellow rust.

It is known that the success of breeding depending on the knowledge of pathogen race composition and genotype of breeding material. The international and European sets of wheat differentials, Yr-gene isogenic lines of Avocet Susceptible (AVS) and commercial cultivars were used to study race composition of *Puccinia striiformis* f. sp. *tritici* and to postulate genes for resistance to yellow rust in some winter wheat cultivars in Kazakhstan. Urediniospores of stripe rust, collected from cultivars grown in farms and experimental stations were used as inoculum for infection. As a result 20 pathotypes of the stripe rust pathogen were identified in Kazakhstan. The most virulent pathotypes were 7E159, 15E159, 47E143, 111E158, 7E223, 7E157, 7E21, 4E144, and 6E145 that overcome resistance controlled by Yr9 or Yr10 genes out of 16 Yr genes studied. The use of these pathotypes for evaluating wheat germplasm for resistance could help to improve breeding for yellow rust resistance. The valuable sources of yellow rust resistance are L-19, L-572, L-796, Almaly, Taza, Zhalyn (Kazakhstan), Aichurec (Kyrgyzstan), Ulugbek 600 (Uzbekistan), and Ekinchi (Azerbaijan).

The study of winter wheat germplasm from different nurseries allowed to evaluate the value of the lines for genetic and breeding programs directed on improvement of wheat stripe rust resistance. It was found that cultivars Taza, Krasnovodopadskaya 25, and Ulugbek 600 have all-stage resistance. The most effective resistant sources against stripe rust in this region are the genotypes possessing genes Yr2+, Yr4+, Yr5, Yr10, and Yr15. Among the USA differentials, cultivars that were resistant to stripe rust included Paha, Druchamp, Riebesel 47/51, Lee, Tres, Express, Clement, Heines VII and Hybrid 46. Virulences to resistance genes Yr1, Yr2, Yr6, Yr7, Yr8, and Yr9 have been detected. The number of genes and character of gene interaction conferring resistance to yellow rust of the most important wheat samples were determined. Based on genetic studies of the resistant stocks, we will be able to develop cultivars possessing effective gene or combination of genes regarding to known virulent races of the stripe rust pathogen.

Using complex of morphological and molecular markers, associated to Yr18/Lr34 gene, 49 carriers of wheat yellow and leaf rust resistance genes were identified. The use of microsatellite marker Xgwm130 and morphological marker leaf tip necrosis (Ltn) are the reliable approach to identification of carriers for effective slow rusting Lr34 gene. Based on genetics of donors it will be possible to develop the cultivars possessing effective gene or combination of genes regarding to known virulent race of pathogen.

**P56. Introduction of two resistance genes against powdery mildew (*Blumeria graminis* f. sp. tritici) and leaf rust (*Puccinia recondita* f. sp. tritici) to winter wheat (*Triticum aestivum*)**

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Powdery mildew is a fungal disease caused by the *Blumeria graminis* and leaf rust is also a fungal disease, affecting wheat caused by the *Puccinia recondita*. They are the most prevalent of all of the wheat diseases in many regions of the world including Poland. These two diseases had caused serious epidemics in Poland. The aim of presented research was to use genetic markers in process of introduction of effective genes of resistance to modern polish and german winter wheat cultivars. A powdery mildew resistance gene *Pm21* derived from *Haynaldia villosa* (syn. *Dasyphyrum villosum*) and leaf rust resistance gene *Lr41* derived from *Aegilops tauschii*, were transferred into the very best polish and german varieties (Nadobna and Lexus). A 6VS/6AL translocation line Yangmai 5 was used as the resistance source of *Pm21* for powdery mildew and the line WGRC10 was used as the resistance source of *Lr41* for leaf rust. In foreground selection several molecular markers for *Pm21* and *Lr41* were resorted. For the detection of gene *Pm21* in breeding materials with powdery mildew resistance were used three molecular markers: SCAR<sub>1250</sub>, SCAR<sub>1400</sub> and NAU/xibao15. To detect the *Lr41* gene were developed three SSR markers, *Xgdm35*, *Xbarc124*, *Xgwm210*. After the first backcrosses background selection was conducted using the 5 AFLP primers combinations (P38/T43, P41/T41, P39/T41, P35/T43, P36/746). After the last crosses selected lines were inoculated in the greenhouse at the three-leaf stage with a natural pathogen population of *B. graminis* and *P. recondita*.

## **P57. Leaf rust (*Puccinia triticina*) resistance genes in IWWIP winter-facultative wheat (*triticum aestivum* L.) genotypes**

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Leaf (brown) rust induced by *Puccinia triticina* is a common wheat disease causing serious yield losses around the world. Breeding for genetic resistance is the most economic way of avoiding yield losses. Therefore, wheat breeding programs in many countries apply resistance breeding in their programs for resistance, especially a long lasting one, against leaf rust. The most preferred long-lasting resistance type is slow rusting on these days. Slow rusting acquires both prediction of *Lr* genes of genotypes in the greenhouse and an efficient screening of leaf rust under field epidemy. Here, we studied *Lr* genes of winter-facultative wheat genotypes improved by the International Winter Improvement Program (IWWIP) besides 40 Thatcher leaf rust isolines. Greenhouse and field studies were in CIMMYT (Mexico) headquarters in 2005. The genotypes were inoculated with twelve common leaf rust races in the greenhouse. The *Lr* genes identified in the genotypes and number of varieties with the genes, either in combination or alone were the following: *Lr1* (in 8 genotypes), *Lr3a* (7), *Lr10* (17), *Lr13* (12), *Lr14a* (7), *Lr10* (1), *Lr16* (7), *Lr17* (8), *Lr23* (12), *Lr24* (6), and *Lr26* (3), *Lr27*(2), *Lr31* (2).

All genotypes were also inoculated with leaf rust spores in the field and then screened using modified Cobb scale every succeeding week. Based on the screenings, the Area Under The Disease Progress Curve (AUDPC) was later calculated separately for each genotype. The variation for AUDPC and a clear indication of slow rusting in winter wheat genotypes adequately existed.

## **P58. Selection for resistance sources of wheat to the most harmful diseases for creation durable resistant cultivars**

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*Puccinia triticina*, *Blumeria graminis*, *Stagonospora nodorum* and *Septoria tritici* are the most harmful diseases of wheat in Russia. For creation cultivars with durable resistance the initial material should be characterized by a genetic variety of resistance, constraining evolution of pathogen. For evaluation and selection of wheat sources and donors of resistance to a complex of the most harmful diseases all diversification of pathogen cultures from All Russian Research Institute of Phytopathology collection were used. More than 1500 samples of spring and winter wheat from USD-ARS, N. I. Vavilov All Russian Research Institute of Plant Industry and “Arsenal” collections were tested for resistance to leaf rust, *Septoria* and powdery mildew. Evaluations for resistance to diseases were carried out in the infection nurseries of All Russian Research Institute of Phytopathology (ARRIP), Agriculture Research Institute of Non Chernozem Zone (ARINCZ) and Lukyanenko Research Institute of Agriculture (KNIISH) differed agro-ecology zones. The genotypes possessed different leaf rust resistance types were selected as a result of evaluations for resistance of spring durum and common wheat. The race-specific and adult plant resistance genes to leaf rust and powdery mildew were identified by the phytopathologic testing and the STS and SSR-markers. Wheat cultivars possessed high level of partial resistance to leaf rust, *Septoria* and compatible resistance to leaf rust and powdery mildew are of interest for breeding. The greatest quantity of resistant and weak susceptible cultivars to *Septoria* was allocated among samples of winter soft wheat from the West European and East European ecology-geographical groups, spring soft wheat from the South American group, spring durum wheat from the East European group. Resistant wheat cultivars to leaf rust, *Septoria* and powdery mildew from NSGC with complex of economically valuable features for improvement of wheat cultivars in Central region of Russia were selected.

## **P59. Evaluation of species from genus *Aegilops* for resistance to leaf rust and septoriosis**

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One hundred and eighty-two samples of 20 species closely related to genus *Aegilops* were tested for resistance to leaf rust and septoriosis local populations in greenhouse experiments (seedling stage) and in field nurseries on an artificial infection background. As a result of the testing *Aegilops* species in ontogenesis there were revealed 88 samples possessed high race-specific resistance to leaf rust. Results of researches showed that the diploid forms ( $2n=14$ ) were very resistant to leaf rust, 7 species from 11 weren't infected pathogen in all phases of plant growth (*Ae. speltoides*, *Ae. aucheri*, *Ae. longissima*, *Ae. searsii*, *Ae. umbellulata*, *Ae. commosa*, *Ae. heldreichii*). Species of *Ae. bicornis*, *Ae. scharanensis*, *Ae. tauschii*, *Ae. unirstata* were differed by susceptibility to pathogen. There weren't found completely leaf rust resistant samples among tetraploid group ( $2n=28$ ). However there were found forms of *Ae. ovata*, *Ae. columnaris*, *Ae. triuncialis*, *Ae. variabilis* resistant to pathogen in all phases of plant vegetation. The hexaploid group ( $2n=42$ ) was represented by strong leaf rust susceptible species of *Ae. vavilovi*, *Ae. trivialis*, *Ae. juvenalis*. The evaluation for resistance to septoriosis has revealed samples differed levels of susceptibility. Prevailing quantity of samples were susceptibility to disease. Nine samples were septoriosis resistant (disease intensity 10-15%). There were 3 diploid samples (1 sample of *Aegilops bicornis* and 2 samples of *Aegilops longissima*) and 6 tetraploid samples of *Aegilops columnaris*. Two samples of *Ae. longissima* and 5 samples of *Ae. columnaris* possessed the group resistance to leaf rust and septoriosis were of interest as sources for improvement of wheat.

## **P60. Breeding for resistance to rust diseases of wheat in Kyrgyzstan**

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Bread wheat in Kyrgyzstan is an important strategic crop as it is major food the population. It occupies more than half the total arable area of agricultural crops cultivated in the country. At present, virtually all farms do not have the possibility of growing intensive varieties with a high level of productivity and high quality of grain. The main factors, limiting wheat yield, quality grain, are problems with connection rust diseases.

Annual crop losses caused by rust diseases (*P.striiformis*, *P.recondita*) in Kyrgyzstan are 10-30%. In 2001 an epiphytotic of yellow rust was recorded in Kyrgyzstan with crop losses estimated at between 40-60% (M.Dzhunusova, A.Yahyaoui, A.Morgounov, 2002).

Although wheat research in Kyrgyzstan started in 1936, breeding wheat for biotic stresses such as rust diseases was initiated only in recent years.

Since 2002 wheat breeding program in Kyrgyzstan for resistance to yellow rust started with close collaboration with CIMMYT and ICARDA. In this period about 1000 samples of wheat were introduced and screened at Chui region under natural epidemic conditions. A few hundred resistance lines were selected and went through breeding program for adaptation to ecological conditions, and yield trials were conducted for selection of the most varieties (Azibrosh, Zubkov, Zagadka, Almira, Djamin, Hans, Petr). In present time advanced lines starting from PYT were sent to CIMMYT, ICARDA for tests to stem rust resistance.

Monitoring of the yellow rust population started in 1999 based on CWAYRTN distributed by ICARDA. The results of evolution the reaction to yellow rust of differentials of CWAYRTN in Chui region of Kyrgyzstan indicated, that most effective against *Puccinia striiformis* in this region are the sources of genes Yr2+, Yr4+, Yr5, Yr10, Yr15, demonstrated R-MR infection types.

In 2006 we are screening nurseries CWANA-6<sup>th</sup> RWKLDN 2006-2007. In this years were drought, because we are can not selected to resistance rust diseases. We are selected best germplasm for yield breeding and resistance to abiotic stress (AO41, Emu'S'/TEVEE'S'3/SD8036, Entry#27, #75.#76, #88, #91, #94).

In 2007-2008 we are screening nurseries 1<sup>st</sup> Ug99IRSTN2007-2008. These years were drought, because we are can not selected to resistance rust diseases. We are selected best germplasm for yield breeding program and resistance to abiotic stress (Cook, Pavon76, Vernstein, CnSSrTmp, Bt/Wld)

In 2008 we are starting monitoring nurseries CWANA-1<sup>st</sup> Stem Rust Resistance Spring Bread Wheat Yield Trial 2008-2009. Monitoring this nurseries showed that 17 varieties were resistance to yellow rust diseases, demonstrated R-MR infection types, 5 varieties were susceptible to yellow rust (Amir1, Amir2, Zafir-3, Borej-2, Zain-2). There are varieties demonstrated MS-S infection types. We are selected for future breeding program 4 best lines of wheat for productivity, resistance to rust diseases (Jawahir-14, Durra-1, Durra-4, Durra-5).

## **P61. Combining ability for plant height, spike length and thousand kernel weight in crosses of powdery mildew resistant wheat lines**

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Powdery mildew (*Erysiphe graminis*) is widely distributed in wheat production regions which have cooler and moist spring season and it influences the grain yield and end use quality negatively. Because of these adverse effects, together with high yielding it is necessary to develop powdery mildew resistant bread wheat genotypes. F<sub>2</sub> generations of 15 crosses by design II mating scheme (North Carolina) between five powdery mildew resistant lines obtained from CIMMYT (International Maize and Wheat Improvement Center) and three commercial varieties (Atilla-12, Basribey-95 and Golia), and these eight parents were grown in a three replicated experiment with artificial inoculation in 2006-2007 cropping season in Bornova-İzmir location. The estimates of general combining ability effects were obtained for plant height, spike length and 1000 kernel weight and inheritance mode of powdery mildew was investigated in F<sub>2</sub> generation. The contrasts of parents vs. crosses indicating average heterosis effect in F<sub>2</sub> generation were found to be significant for the spike length and the 1000 kernel weight but not for the plant height. Similarly, it was determined that general combining ability effects, being significant level for spike length and 1000 kernel weight which differences among F<sub>2</sub> populations were significant, accounted for 96.4 % and 83.6 % of variability among crosses, respectively. Portion of general combination ability effect in significant level (GCA) for plant height which differences among crosses were not significant, in total variability was obtained as lower (%44.9) than those of other two traits. Specific combining ability effects (SCA) for all traits were not significant. Among the parent genotypes, lines 48 and 72 also cultivar Atilla-12 for spike length, line 27 and Atilla-12 for 1000 kernel weight showed significant GCA effects. Besides, GCA effects of parent genotypes were observed to be not significant for plant height. Considering Khi square tests, it was understood that powdery mildew resistance in the crosses derived from Atilla-12 and Basribey with line 48, Golia and Basribey with line 27, and Golia with line 35 was governed by single gene in F<sub>2</sub> generation. It was concluded that combinations of Atilla-12 x 48 and Basribey x 27 as the promising F<sub>2</sub> populations which the genotypes with long spike, high kernel weight and acceptable plant height could be developed in addition to resistance to powdery mildew may be used in future wheat breeding programs regarding the mean values of F<sub>2</sub> generations for measured traits.

## **P62. Introduction of powdery mildew (*Blumeria graminis* (DC.)Speer) resistant gene Pm21 into winter triticale (x *Triticosecale* Wittm.)**

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Since 1984 when the first Polish winter triticale variety named 'Grado' was registered, the cultivation area of triticale spike upward very quickly to more than one million hectares. Dozens of new varieties were registered at that time, nonetheless triticale in comparison with parental species was known as generally free from diseases. On the basis of long time experience conducted in Germany (1988-2001) scientists recorded that triticale is the most "healthy" cereal. However, in 2005 on the basis of the field observations, powdery mildew became serious triticale threat in Poland. One of the best polish variety 'Lamberto' after several years of extensive cultivation in Poland and Germany became very sensitive to *Blumeria graminis* population. It has been proven that fungus *B. graminis* may cause yield losses in triticale production up to 25%. Nowadays, resistance to diseases became one of the most important benchmark of variety mileage for intensive, integrated and organic triticale production. The development and utilization of triticale cultivars with resistance genes is an efficient, economic and environmentally safe strategy to control powdery mildew.

The aim of these studies is introduction an effective resistant gene of powdery mildew into winter triticale variety. Therefore 'Moderato' widely grown fodder, traditional winter triticale variety, well-adapted to the Polish environmental conditions was presumed. The gene *Pm21*, located on short arm of chromosome 6V is one of the most effective resistant genes against *B. graminis* races in Europe as well as in Asia, conveys high resistance to all known races of *B. graminis*. *Pm21*, originally introduced into wheat line 'Yangmai5' from *Haynaldia villosa* through chromosome translocation of 6VS/6AL. It is noteworthy that the 6VS/6AL translocation also carries a resistant gene (*Yr26*) to *Puccinia striiformis*.

The population F<sub>1</sub> derived from the cross of 'Moderato'x'Yangmai5' was developed. Afterwards, this population was backcrossed with 'Moderato'; population F<sub>1</sub> and then the next generation were obtained. At each following generation fertility has been estimated. Marker-assisted selection of *Pm21* was conducted by using a co-dominant marker NAU/xibao15 linked with resistant gene. Only lines carrying *Pm21* resistant gene and full fertility were selected for further research.

### **P63. Improvement of drought tolerant winter and facultative wheat promising lines resistant to yellow rust and stem rusts (Ug99)**

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Yellow rust (*Puccinia striiformis* f.sp. *tritici*) and stem rust (*P. graminis*) diseases of wheat are significant threat to wheat production, even in drought stressed regions of Iran. Although research efforts have been effectively reduced the crop damage, but rust populations has been evolved due to changes in the environment and wheat production system in the region. Pathogenic variation is the underlying cause of the elusive rust resistance. So, new yellow rust race (*166E134A+*, *Yr27+*) widespread and breakdown of resistance and virulence took place for most wheat cultivars grown in Iran in 2006-07. Observation of stem rust epidemic and emerging new race (*Ug99*) in Africa and its spread to the neighboring countries such as Yemen (2006) and Iran (2007) is also a big threat for wheat production. Breeding for qualitative type of resistance is a widely adapted method. The objective of this paper is to introduce 9 drought tolerant, stem and yellow rusts resistant and high yielding bread wheat promising lines. Two sets of wheat materials including forty four F3 Segregating Populations (SP) and 100 drought tolerant high yielding lines (AYT) received from ICARDA and 20 national newly improved lines were evaluated in Early Generation tests and Advanced Yield Trial, respectively, at Seed and Plant Improvement Institute (SPII), Karaj-Iran during 2003-04 cropping season in two sets of experiments. In first set of experiment, forty-four F3 SP were planted under mist conditions and Selected Bulk method was used to handle the F3 generation. Selected plants from SP were planted under terminal drought stress (TDS) conditions for assessing their drought tolerance until F6 generation. Selected pure lines were then tested for grain yield in Regional Preliminary Yield Trial at Karaj (temperate zone) and Ardabil stations (cold zone) in F7 generation in 2007-08. The selected lines were then sent to Kenya for evaluation of their reaction to new stem rust race (*Ug99*). Data received from Kenya showed resistance to *Ug99* in four selected lines with the pedigrees of 130L1.11//F35.70/MO73/4/YMH/TOB//MCD/3/LIRA/5/TIRCHMIR1/LCO//SABALAN; SKAUZ/4/TJB916.46/CB306//2\*MHB/3/BUC/5/TIRCHMIR1/LCO//SABALAN;474S101/BOLERO/3/TIRCHMIR1/LCO//SABALAN and Sabalan/KINACI97. The selected lines showed the acceptable level of resistance to yellow rust race (*166E134A+*) of yellow rust, too. Determination of yield stability was carried out, using non-parametric ranking statistic method, and results showed that these four lines with good resistance to *Ug99* and *166E134A+*, *Yr27+* races were also yield superior to the check cultivars. In second set of experiments, one hundred advanced lines were planted in alpha lattice design (AYT) under TDS conditions and necessary agronomic and morphological traits and their reactions to rusts and grain yield were recorded. Selected lines were evaluated for rust resistance in several hot spots over 3 years and evaluated for grain yield stability and adaptability in four agricultural research stations for two years. Considering both the recorded stem rust data from Kenya, and yellow rust data from hotspot sites in Iran, and based on grain yield, five superior drought tolerant lines were selected from this trial, too. The lines with pedigrees of Bkt/Zhong; OK82282//BOW/NKT/3/SARDARI-HD75; SARDARI-HD93/6/SN64//SKE/2\*ANE/3/SX/4/) were resistant to *166E134A+*, *Yr27* yellow rust race, but were susceptible to *Ug99*. Although the line with pedigree of AGRI/BJY//VEE/3/PRINIA was moderately-resistant to *Ug99*, it showed high susceptibility (90 S) to new yellow rust race. The new line (SARDARI-HD83//LINFEN875072/KAUZ) was resistant to both stem rust (*Ug99*) and yellow rust (*166E134A+*, *Yr27+*) with high yield under both irrigation and TDS conditions.

## P64. Improvement of leaf rust resistance of spring bread wheat in the North Kazakhstan

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The North Kazakhstan is one of the largest rainfed cereal crop producers ranking the 6<sup>th</sup> in the world in terms of the cropping area. Virtually all kinds of cereal crops are grown in Kazakhstan with the wheat crop dominating and occupying 12.5 million ha. The widest-spread and most important disease for the North Kazakhstan is leaf rust. For the last decade, Leaf Rust outbreaks were reported in various regions in 5-7 years out of 10 with the wheat yield reduced by 25-40% and highly deteriorated grain quality. Apparently, the higher occurrence of rust, *Septoria* and *Helminthosporium* spots is associated with a global climate warming resulting in locally higher rainfall during the vegetation and higher temperatures in winter. Accumulation and distribution of the leaf diseases is also facilitated by a wider extension of zero tillage as one of its components is retention of crop residues on the soil surface. But up to now the priority wheat breeding objectives have been yield potential, drought resistance and grain quality. Disease resistance breeding was not paid a proper attention and as the result the current cultivars are susceptible to a number of important diseases, and rusts, in particular. In the recent years, the rust resistance breeding of wheat was conducted partly in the framework of the Kazakhstan-Siberian Spring Wheat Improvement Network (KASIB) facilitated by CIMMYT in 2000 and currently embracing 15 research institutions in Kazakhstan and Russia with a potential impact on the area of 20 million ha. CIMMYT has contributed new rust resistance donors and sources to national breeding programs (Thatcher isogenic lines LR, ISRTN, SRRSN). The shuttle breeding program between Mexico and Kazakhstan-Russia resulted in a promising breeding material possessing high resistance to rusts. The results of the KASIB research show that the Kazakhstan leaf rust population is strongly virulent for commercial varieties and many *Lr* isogenic lines. Highly effective resistance genes in the region are *Lr9*, *Lr24*, *Lr25* and *Lr36*. Genes *Lr17*, *Lr20*, *Lr28*, *Lr29*, *Lr34* considered as resistant previously, proved to be susceptible to some leaf rust isolates. A number of genes (*Lr12*, *Lr29*, *Lr30*) possess slow rusting effect with low rust severity. Highly effective stem rust resistance genes to local population are *Sr24*, *Sr33*, *Sr35* and *Sr36*. Due to Ug 99 (TTKS) outbreak, over 1000 genotypes were assessed for resistance to this race in Kenya. The assessment results demonstrate that the majority of Kazakhstan and Russian varieties from the West Siberia, Altay and Ural regions are susceptible to Ug 99. However, highly resistant varieties such as Stepnaya 62, Omskaya 37 and Omskaya 38 were identified under strong infection pressure in Kenya. These varieties were submitted to the State Yield Trial and are considered as promising for a broad release in the West Siberia and North Kazakhstan. Therefore, a stronger emphasis on the theoretic research on resistance genetics, involvement of new genetic resources and close collaboration between CIMMYT and national breeding programs of Kazakhstan and Russia allow to believe that issues of leaf disease resistance enhancement be successfully resolved for commercial varieties.

## **P65. Research on inheritance of yellow rust resistance in İzgi-2001 wheat cultivar**

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Wheat (*Triticum* spp.) is one of the most important and strategic crop in the world. Yellow rust, caused by the fungal pathogen *Puccinia striiformis* f. sp. *tritici* is one of the most damaging diseases affecting yield and quality in wheat in almost throughout in Turkey as well as on large lands of all over the world. The disease is controlled through resistant genotypes and fungicides. Among all the control measures of yellow rust, genetic resistance is the only economic and practical control measure, causing no additional cost to the farmer. Use of genetic resistance and development of resistant cultivar are very important to control of the disease. On the other hand, genetic resistance is environmentally safe control measure. The most wide resistance mechanism of yellow rust is specific (generally under mono or oligo-genic control) resistance and at present, more than 30 resistance genes have been described.

Izgi 2001 is resistant to yellow rust and was released for Central Anatolia region. The objective of the study was to determine the inheritance of resistance in İzgi 2001 bread wheat cultivars. In this study, resistance of adult and seedling stage of F<sub>2</sub> plants which are obtained from İzgi 2001 X ES 14 crossed was determined to the yellow rust under disease conditions which artificially were produced using appropriate inoculation methods in the field and greenhouse. The experiment was conducted at the Research Station of the Central Research Institute for Field Crops in Yenimahalle and Haymana locations, Ankara, Turkey. For adult plant stage test, the F<sub>2</sub> plants and the parents were sown by hand in 2 m-long rows with 33 cm space on the rows in October 2006. The F<sub>2</sub> plants for seedling stage test, the parents and the susceptible checks were sown in small pots and they were kept in the greenhouse until scored in March 2007. As a susceptible check, Little Club was used for both seedling and adult plant reactions test. Yellow rust isolate (effective on Yr 6, Yr 7, Yr 8, Yr 9, Yr 2+, Yr SD, Yr A) was used for inoculation both seedling stage and adult plant stage. As a result, Chi-Square test has confirmed this study and resistance controlled by a major gene in this resistant is cultivar.

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**Key Words:** Wheat (*Triticum* spp), Yellow rust (*Puccinia striiformis* f.sp. *tritici*), Inheritance

## **P66. Model genome interrogator: a PLEXdb module that leverages sequenced genomes for motif discovery via meta-promoter extraction and analysis**

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The language that provides instruction to the transcriptional machinery of cells remains poorly understood. Characterization of motif function (word meaning) and relative position (word order, usage, grammar) helps further our understanding of this cis-regulatory language. The two commonly used approaches for identifying putative regulatory motifs capitalize on 1) co-expression during development and/or across treatments, and 2) motif retention in independent lineages as evolution occurs. Model Genome Interrogator (MGI) facilitates a powerful fusion of these approaches. For single or batch queries, MGI uses the sequenced genomes of rice and/or Arabidopsis to identify the putative orthologues (metagenes) corresponding to probesets from any of the 16 microarray platforms supported by PLEXdb ([www.plexdb.org](http://www.plexdb.org)). For each metagene identified, MGI allows researchers to view annotations and gene models, and to extract sequence data from promoters, exons, introns and UTRs. The following use case scenario illustrates several of many MGI utilities. For a set of cereal genes found to be co-expressed across several conditions via a microarray experiment, cereal probe identifiers are used in an MGI query to find orthologous genes in rice. The promoters of these metagenes can then be retrieved by the researcher using the MGI web interface, and subsequently subjected to cis-regulatory element searches using standard methods. When a motif is shared among the metapromoters beyond what could occur by chance, coordinate regulation of these genes in both cereal and rice can be inferred. If such an hypothesis can be borne out via further testing, it suggests that the regulatory mechanism is sufficiently important to have been conserved through 50 million years of evolution in both independent lineages. Thus, MGI facilitates identification of conserved non-coding sequences (CNS) that are predicted to regulate whole sets of genes, creating a synergism of co-expression and CNS approaches for documenting the language of promoters.

## **P67. PLEXdb: Plant and pathogen expression database and tools for comparative and functional genomic analysis**

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PLEXdb is a plant expression database that supports all Affymetrix microarray designs for plants and plant pathogens. PLEXdb provides microarray annotation and visualizations of raw and normalized experiment data across treatments in the form of boxplots, heatmaps, dendrograms, expression graphs, scatterplots and quality control plots. Experiment data deposited in PLEXdb is hand-checked for MIAME/Plant compliance and completeness then processed by normalizing the raw data using RMA and MAS5.0, producing tables and graphics from the resulting data. To facilitate the use of PLEXdb at any stage of data analysis, experiments may be kept private, shared worldwide with collaborators and reviewers, or released directly to the public. A suite of analysis and visualization tools permits in-depth exploration including searching for homologous genes on different microarray platforms and translating microarray probe sets onto model genomes. New features include: submission of experiment data to GEO on researchers' request; GO, EC, and KEGG annotation of probe sets; the Microarray Platform Translator; a gene variance tool which searches experiments by the amount of variation in the expression of a specific gene; and tools for creating, analyzing, and annotating gene lists. PLEXdb uses standardized vocabularies such as the gene and plant ontologies to improve searching and cross-experiment comparisons. PLEXdb also has interactive links to plant and pathogen databases such as PlantGDB, Gramene, GrainGenes, HarvEST, and UniProt. PLEXdb currently houses over 1,000 GeneChip hybridizations from 21 rust and mildew experiments from barley, wheat, Arabidopsis, soybean, and grape.

## **P68. Omics approaches to understand the nature of virulence in *Puccinia striiformis* f.sp. tritici**

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New genomic and transcriptomic methods greatly facilitate the study of the biology and evolution of fungal plant pathogens. The obligate biotrophic and asexually reproducing rust fungus *Puccinia striiformis* f.sp. *tritici* (*Pst*) forms haustoria during plant infection and delivers proteins and other effector molecules into the plant cell. Rust effectors take a key function in the rust-host interaction: on the one hand they are target of the plant resistance protein system and on the other hand they mediate virulence as subjects of diversifying selection. The virulence of *Pst* also depends on the presence and rapid evolution of such effectors. *Pst* causes yellow rust on wheat, one of the most devastating diseases worldwide, and poses an imminent danger to previously resistant wheat varieties in unaffected geographic regions due to aerial long-distance dispersal and adaptation to a warmer climate. We are applying the next-generation Solexa/Illumina sequencing technology to both DNA and RNA of *Pst*. The assembled genomic and transcriptomic information is likely to facilitate the analysis of *Puccinia striiformis* effectors and to provide new insights into the molecular basis of virulence/avirulence as well as pathogen evolution.

## **P69. Identification of proteins differentially expressed upon BTH treatment of *Triticum aestivum* (wheat) by proteomics approach**

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The systemic acquired resistance (SAR) mechanism is a process of biological and chemical activation upon infection with disease agents function as a part of the innate immunity response system of plants. Externally applied fungicide, benzo (1, 2, 3)-thiadiazole-7-carbothioic acid (BTH) is known to induce the SAR response in plants. The molecular level studies identifying BTH induced or suppressed genes are limited to this date; few genes responding to BTH are determined, none of which were identifies the differentially expressed proteins by proteomics approach. The focus of this preliminary study is to identify the gene products controlled by BTH in wheat. The 2D-PAGE analysis was carried out for BTH treated *Triticum aestivum* cv. 'Gerek-79' and mock treatments. Following 2D-PAGE image analysis, the selected differentially expressed protein spots were identified by nano-LC-ESI-MS/MS. Among the protein spots studied, five increased upon BTH treatment, another set of five spots were absent in the control sample, thus they were apparent only in the gel of the BTH treated sample, whereas three protein spots disappeared in BTH treated gel. Up-regulation of some proteins are such as oxygen evolving enhancer protein (OEE2) and cold-responsive late embriogenesis abundant (LEA)/RAB-related COR protein and down-regulated ones are such as Rubisco LSU and fructose 1,6 biphosphate aldolase shown for the first time.

## P70. Identification of Resistance to Yellow Rust in Wheat Germplasm in Iran

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Wheat grain yield is reduced by limiting factors such as wheat diseases. Rusts with different physiological races, virulence and the ability of development of new races are the most important wheat diseases. Thirty seven genes for resistance to yellow rust have been identified and used in different wheat breeding programs. However, many of these resistance genes were effective for a short period, but by incidence of new races of yellow rust they are not effective any longer. Therefore, development of new resistance cultivars demands continuous efforts. Response to yellow rust in 90 cultivars and advance breeding lines including commercial cultivars, crossing block germplasm, and introductions from CIMMYT, Mexico were studied using four yellow rust isolates collected from Karaj, Zarghan, Ardabil and Mashhad. Using observed reactions and recorded values for each differential variety (Johnson *et al.*, 1972), 134E134A<sup>+</sup>, 166E134A<sup>+</sup>, 6E2A<sup>+</sup> and 6E22A<sup>+</sup> races were determined for isolates from Karaj, Zarghan, Ardabil and Mashhad, respectively. The race 6E22A<sup>+</sup> from Mashhad was virulent on Yr8 gene in addition to Yr2, Yr6, Yr7, Yr9, YrA, Yr22 and Yr23 genes. This characteristic differentiates this race from many other races identified in Iran. In order to evaluate the responses, the genotypes were inoculated at 12 stage in Zadoks scale (Zadoks *et al.*, 19784) using talk powder in 1:4 ratio. Latent period and infection type were recorded. Genotype × race interaction was significant which implies specific resistance. Non-additive test for Replication × genotype interaction was not significant for all four races. In cluster analysis two traits with four races, considering different reactions of different genotypes to four different races, the genotypes were classified in two main groups. Group 1 included 24 genotypes which were resistant/moderate resistant to all four races. Group 2 comprised of 66 genotypes which were at least susceptible to one race.

**Keywords:** Bread wheat, Yellow rust, Physiological races, Latent period, Infection type

## P71. Diallel Analysis of Yellow Rust Resistance Components in Wheat Genotypes

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Yellow rust caused by *Puccinia striiformis* westend f. sp. *tritici* is one of the most important fungal diseases in Iran and many other countries. Knowledge of the inheritance of traits of interest is essential for determination of appropriate breeding approach and evaluation of genetic gains in different generations. In order to determine the heritability and gene action for yellow rust resistance components, six cultivars and advance breeding lines including; Pishtaz, MV17, Moghan3, Seri/Kauz, Kauz//Kauz/PVZ as well a susceptible cv. Bolani were crossed using half diallel 7 × 7 design. Evaluation of F1s and parents were carried out using a Randomized Complete Blocks Design with three replications. Plants were inoculated with 134E134A<sup>+</sup> race using talk powder. The first resistance component, latent period, was determined by the number of days from inoculation to the appearance of the first pustules and the second component, infection type, was recorded using MacNeal *et al.*, 1971 method. Pustules density and size were measured on infected leaves kept in lacto-phenol solution using microscope. Analysis of variance for combining ability using Griffing method showed that general combining ability (GCA) and specific combining ability (SCA) for four traits of interest were significant which implies the additive and non-additive effects in genetic control of these traits. Among parents cv. MV17 with high GCA for all four traits was identified as suitable parent to be used in breeding programs for enhancement of resistance to yellow rust. Bolani had the highest GCA next to MV17 but for susceptibility. Crosses of MV17 with Bolani, Pishtaz, Kauz//Kauz/PVZ had higher SCA (-) for infection type, number of pustules and pustule size, and the highest SCA (+) for latent period in enhancement of resistance which implies non-additive (dominance and epistasis) gene effects in these crosses. Graphic analysis of Vr and Wr indicated that resistance components; latent period, infection type, pustule size, and number of pustules were recessive in some genotypes but dominant in some others. Broad and narrow sense heritabilities for all four traits were relatively high. Therefore, selection for enhancement of resistance to this race using these resistance components is useful.

**Keywords:** Wheat, Diallel crossing, Heritability, Yellow rust, Resistance Component

## **P72. Inheritance and Gene Action for Resistance to Stripe Rust in Bread Wheat**

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In order to determine the genetic parameters, number of genes and gene action, for resistance genes to yellow rust, F1, F2, F3, BC1, BC2, BC1S1, BC2S1 generations derived from a cross made between MV17 (resistant) and Bolani (susceptible) were grown and evaluated using Randomized Complete Blocks Design with three replications in Field Research Station, Seed and Plant Improvement Institute, Karaj, Iran. These generations were inoculated in different stages by mixture of pathogen, 134E134A<sup>+</sup> race, and talk powder. Following the diseases establishment; infection severity and infection type on flag leaves of single plants, in three times with 7 days intervals, were recorded. Using these records, coefficient of infections (CI) for each interval were calculated, and then final coefficient of infections (FCI) and areas under diseases progress curve (AUDPC) and relative areas under diseases progress curve (rAUDPC) were estimated and used in statistical analyses for estimation of genetic components, means and variances. Generation mean analysis using joint scaling test revealed that additive, dominance and epistasis effects, particularly, additive × dominance and additive × additive interactions, significantly affected reduction in FCI, AUDPC and rAUDPC. Finally, considering all three traits, five parameters model m, [d], [h], [i], [j] was determined as suitable model. Dominance degree for all traits was complete dominance but negative which indicates that dominance is inclined towards parents with lower AUDPC, FCI and rAUDPC. Broad sense heritability for different resistance components was high, but narrow heritability was intermediate.

Keywords: Bread wheat, Yellow rust, Heritability, Generation mean analysis

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