Proceedings of the 2012 National Fusarium Head Blight Forum

December 4-6, 2012
Wyndham Orlando Resort
Orlando, Florida USA

Proceedings compiled and edited by: S. Canty, A. Clark, A. Anderson-Scully and D. Van Sanford

Cover designed by: Kris Versdahl of Kris Versdahl & Associates
Red Lake Falls, MN

Photo Source: High Plains Journal, Emma Misener - All Aboard Wheat Harvest
(http://www.allaboardharvest.com/)

U.S. Wheat & Barley Scab Inititiative

©Copyright 2012 by individual authors.

All rights reserved. No part of this publication may be reproduced without prior permission from the applicable author(s).

Printed in the United States by ASAP Printing, Inc., Lansing, MI 48911

Copies of this publication can be viewed at http://www.scabusa.org.

REFERENCING ITEMS IN THE FORUM PROCEEDINGS

When referencing abstracts or papers included in these proceedings, we recommend using the following format:

Last Name and Initial(s) of Author, [followed by last names and initials of other authors, if any]. Year of Publication. Title of paper. In: [Editor(s)] (Eds.), *Title of Proceedings* (Page Numbers.); Place of Publication: Publisher.

Sample Reference:

S. Ali, M. Eldakak, P. Gautam, K. Glover, J.S. Rohila, J. Gonzalez and W. Berzonsky. 2012. “Gibberella zeae Chemotypes Diversity on Moderately FHB Resistant Wheat Genotypes in South Dakota.” In: S. Canty, A. Clark, A. Anderson-Scully and D. Van Sanford (Eds.), *Proceedings of the 2012 National Fusarium Head Blight Forum (* pp. 115-117). East Lansing, MI/Lexington, KY: U.S. Wheat & Barley Scab Inititiative.

Proceedings of the 2012 National Fusarium Head Blight Forum

Wyndham Orlando Resort

Orlando, FL USA

December 4-6, 2012

We would like to acknowledge our sponsors and partners for their generous contributions to the 2012 National Fusarium Head Blight Forum.

American Bakers Association

American Malting Barley Association

Anheuser-Busch Companies

BASF
The Chemical Company

Bayer CropScience

General Mills

American Bakers Association
American Malting Barley Association
Anheuser-Busch Companies
BASF
Bayer CropScience
General Mills

Minnesota Wheat Research
& Promotion Council

MillerCoors
Minnesota Wheat Research and Promotion Council
Monsanto
North American Millers' Association
North Dakota Wheat Commission
Sierra Nevada Brewing Company
U.S. Department of Agriculture-Agricultural Research Service (USDA-ARS)

FORUM ORGANIZING COMMITTEE

Co-Chairs: Mike Davis, American Malting Barley Association, Milwaukee, WI,
Jane DeMarchi, National Association of Wheat Growers, Washington, DC

Members: Gary Bergstrom, Cornell University, Ithaca, NY
Fred Kolb, University of Illinois, Urbana, IL
Gene Milus, University of Arkansas, Fayetteville, AR
Jim Pestka University of Wisconsin, Madison, WI
Paul Schwarz, North Dakota State University, Fargo, ND
Nilgun Tumer, Rutgers University, New Brunswick, NJ
Jochum Wiersma, University of Minnesota, Crookston, MN
Jin-Rong Xu, Purdue University, West Lafayette, IN
USWBSI Co-Chairs: Art Brandli, Warroad, MN; and
Dave Van Sanford, University of Kentucky, Lexington, KY

ORGANIZED BY:

U.S. Wheat & Barley
Scab Initiative

Table of Contents

SESSION 1: FHB MANAGEMENT

SESSION 2: FOOD SAFETY AND TOXICOLOGY

SESSION 3: VARIETY DEVELOPMENT AND HOST PLANT RESISTANCE

SESSION 4: PATHOGEN BIOLOGY AND GENETICS

SESSION 5: GENE DISCOVERY AND ENGINEERING RESISTANCE

OTHER PAPER

SESSION 1:

FHB MANAGEMENT

Co-Chairpersons: Gary Bergstrom and
Gene Milus

EFFECT OF PLANT RESIDUE TREATMENT ON THE EPIDEMIOLOGY OF FUSARIUM HEAD BLIGHT

N. Aitkhozina*

Plant Pathology Laboratory, Institute of Microbiology and Virology,
Ministry of Education and Science, Almaty, Kazakhstan
*Corresponding Author: PH: (727) 292921852, Email: nag_aitkhozhina@yahoo.com

ABSTRACT

Kazakhstan is the largest producer of wheat in the central part of Asia. Each year different fungal pathogens cause yield losses from 10-40% in the country. FHB has affected all wheat and barley classes. The disease is caused by the set of *Fusarium* and *Microdohium* species. Integrated disease control in fields includes resistant varieties, cultural practices, and fungicide application. Within the fungicide reduction policy, it's important to develop alternative means to control wheat diseases. In the case of soil borne diseases, the crop residues contribute to the disease severity. Thus, to control FHB epidemics, it's important to suppress the disease by means of reducing pathogens surviving on plant residues. Several *Fusarium* species, esp. *Fusarium graminearum*, *F.culmorum* were isolated from surface soil, anthesis and plants at dough, and from plant residues after wheat harvest in 2010. The inhibition effect of crushed horseradish (*Armoraceae lapathifolia*) tuber tissue volatiles and active compounds on FHB in two small farms in 2010-2012 years was evaluated. Ground horseradish tuber tissue (100 g/10 plant residues) was spread onto wheat residues in October 2010-2011. Every year the occurrence of FHB during the vegetation season was monitored. Plots repeatedly treated with horseradish every year in October had significantly reduced FHB up to 50%. Commercial fungicides or tuber tissue of different plants (carrot, potato) failed to reduce FHB and *Fusarium* inoculum in plant residues and soil. Populations of *Streptomyces* spp., *Bacillus* spp., *Pseudomonas* spp. in soil after the plant tuber tissue amendment gradually expanded (7-fold in 2012) in comparison with those ones in nontreated plots. This approach has the potential to reduce FHB to acceptable level.

DETERMINING 'RAIN FAST' TIME AND RESIDUAL LIFE OF PROSARO ON WHEAT SPIKES

Kelsey F. Andersen[1], Leslie A. Morris[2], Richard C. Derksen[2], Larry V. Madden[1] and Pierce A. Paul[1*]

[1]Department of Plant Pathology, The Ohio State University, OARDC, Wooster, OH 44691; and [2]USDA-ARS, OARDC, Wooster, OH 44691
[*]Corresponding Author: PH: (330) 263-3842, Email: paul.661@osu.edu

ABSTRACT

Fusarium head blight (FHB) is a disease of wheat and other small grain crops caused by the fungal pathogen *Fusarium graminearum* (teleomorph, *Gibberella zeae).* Chemical control has become an important component of FHB management practices and, when applied correctly, can reduce FHB and as well as its associated toxin, Deoxynivalenol (DON). Rainfall events surrounding anthesis are most conducive to FHB development and DON accumulation and thus it is under these conditions that fungicide application is most warranted. It is unclear, however, how rainfall directly following application of certain fungicide chemistries affects deposition, coverage, absorption and efficacy. The aim of the current study was to determine the 'Rain Fast' time for Prosaro® (tebuconazole + prothioconazole). A field trail was conducted for the first time during the 2011/12 growing season in Wooster, OH. A moderately susceptible cultivar (cv. Hopewell) was used in a randomized complete block design with five simulated rainfall treatments applied at different times after a single application of Prosaro® (6.5 fl. Oz/A + 0.125% Induce v/v) at anthesis; no rainfall (1) rainfall 60 minutes after application (2), rainfall 105 minutes after application (3), rainfall 150 minutes after application (4), and rainfall 195 minutes after application (5). Rainfall treatments were applied using an artificial rain simulator, at an intensity of approximately 39 mm/h, for 6 minutes. Plots were spray inoculated with *F. graminearum* spore suspensions approximately 12 hours after fungicide application. Fungicide residue on wheat spikes over time was quantified by collecting a sample of spikes from each plot every four days after fungicide treatment application and analyzing for Tebuconazole residue using GC-MS. FHB intensity was rated approximately 3 weeks after anthesis and DON was quantified post-harvest. In general, rainfall closer to fungicide application (60, 105 and 150 minutes after) reduced fungicide efficacy, resulting in higher mean FHB intensity and DON than later rainfall (195 minutes) or no rainfall (0). Rapid decrease in Tebuconazole residue was seen after 7 days in all treatments, and the rate of decrease appeared to be similar among rainfall timings. This study will be repeated in the 2012/13 growing season using a similar protocol.

EFFECTS OF LOCAL CORN DEBRIS MANAGEMENT ON FHB AND DON LEVELS IN FOURTEEN U.S. WHEAT ENVIRONMENTS IN 2011 AND 2012

G.C. Bergstrom[1*], J.A. Cummings[1], K.D. Waxman[1], C.A. Bradley[2], A.L. Hazelrigg[3], D.E. Hershman[4], M. Nagelkirk[5], L.E. Sweets[6] and S.N. Wegulo[7]

[1]Dept. of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, NY 14853; [2]Crop Sciences Dept., University of Illinois, Urbana, IL 61801; [3]Plant and Soil Science Dept., University of Vermont, Burlington, VT 05405; [4]Dept. of Plant Pathology, University of Kentucky, Princeton, KY 42445; [5]Michigan State University Extension, Sanilac County, Sandusky, MI 48471; [6]University of Missouri, Columbia, MO 65211; and [7]Plant Pathology Dept., University of Nebraska, Lincoln, NE 68583
[*]Corresponding Author: PH: 607-255-7849, E-mail: gcb3@cornell.edu

ABSTRACT

Reduction or elimination of within-field sources of inoculum of *Fusarium graminearum* is the basis for cultural control measures such as crop rotation sequences in which cereals follow non-cereal crops. In USWBSI-supported microplot experiments conducted in twenty-one winter wheat fields over five states in 2009 and 2010, DON level differed significantly between corn debris and no debris microplots in only one location, strongly suggesting that regional atmospheric inoculum is the strongest contributor to infection even when corn debris is present in a wheat field. Small area sources of debris, however, may result in an underestimation of the contribution of spores from a larger field of corn debris to FHB and DON. The goal of the current USWBSI research project is to provide realistic estimates of 'DON reduction' that can be expected from cultural controls that reduce within-field inoculum sources. We utilized moldboard plowing of corn debris as a proxy for planting after a non-cereal crop to compare directly with wheat planted no-till into corn debris in commercial-scale wheat fields planted following grain corn harvest in Illinois, Kentucky, Michigan, Missouri, Nebraska, New York, and Vermont. Following corn harvest, replicated wide (60 ft) strips were moldboard plowed or left non-plowed prior to sowing wheat over the entire field with a no-till drill. Wheat in each strip was monitored for FHB and sampled for laboratory quantification of head infection by *F. graminearum* and contamination of grain by DON. Results were collected over two years, 2011 and 2012, from winter wheat in six states (IL, KY, MI, MO, NE, and NY) and spring wheat in one state (VT).

In 2011, FHB symptoms at soft dough stage were low to moderate at every location except Missouri. Yet, at crop maturity, a high percentage of wheat heads was found to be infected by *F. graminearum* in all locations except Nebraska and Vermont. Measurable DON was found in grain from every environment and the levels were lowest in Vermont and highest in Kentucky and Nebraska. It is interesting that the Nebraska site showed the lowest disease index and lowest incidence of head infection, but the highest average toxin level. Moldboard plowing resulted in a significant decrease in FHB index in four environments (IL, MO, NY, MI), though the magnitude of the difference was large only in Missouri. In Nebraska, FHB index was significantly higher in the moldboard-plowed treatment in which the wheat crop matured earlier than in the no-till corn debris treatment. Moldboard plowing was associated with a small but significant decrease in recovery of *F. graminearum* from mature heads in three environments (IL, MI, NY). There was no significant effect of plowing on DON level in five environments (IL, KY,

MO, NY, VT) and there were small but significant decreases in toxin in moldboard-plowed compared to no-till strips in two environments (MI and NE). An additional treatment of minimum tillage (chisel plow) was added in the Michigan experiment; DON levels in the minimum-till plots were intermediate between moldboard and no-till but not significantly different from no-till.

In 2012, a generally warm and dry cropping season across the experimental region, FHB symptoms at soft dough stage were not observed in four locations (KY, MI, NY, VT) and were observed at low levels at three locations (IL, MO, NE); plowing had no significant effect on FHB index in any location. At crop maturity, a moderate percentage of wheat heads (i.e., greater than 10%) was found to be infected by *F. graminearum* only in Missouri and Vermont; in both environments there was a significantly greater incidence of heads infected in no-till than in moldboard-plowed strips. DON was not detected in Nebraska, and was detected at low levels in all other states. Moldboard plowing resulted in a significant decrease in already low DON levels in New York and Vermont. A similar level of reduction in DON level was observed in wheat from moldboard-plowed strips in Michigan, but DON was assayed in small samples that were pooled from the replicate strips, so no statistical comparison could be made.

There is a strong trend in two years of data suggesting that inoculum from area atmospheric sources exerts a far greater effect than inoculum from in-field corn residue on the level of DON contamination. A third year of experimentation in three additional wheat environments in 2013, in Illinois, Nebraska and New York, will provide increased evidence of the magnitude of the effect of corn residue management on DON reduction.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported in part by the U.S. Department of Agriculture under agreement No. 59-0206-9-056. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

2012 TRIAL OF THE PERFORMANCE OF SELECTED BIOLOGICAL CONTROL AGENTS FOR THE SUPPRESSION OF FUSARIUM HEAD BLIGHT IN SOUTH DAKOTA AND NORTH DAKOTA

B.H. Bleakley[1,2*], K.R. Ruden[1], N. Srinivasa Murthy[2], A. Arens[3] and S. Halley[3]

[1]Plant Science Department, South Dakota State University, Brookings, SD; [2]Biology/Microbiology Department, South Dakota State University, Brookings, SD; and [3]North Dakota State University, NDSU Langdon Research Extension Center, Langdon, ND

[*]Corresponding Author: PH: (605) 688-5498, Email: bruce.bleakley@sdstate.edu

ABSTRACT

Fusarium Head Blight (FHB) or Wheat Scab, caused by *Fusarium graminearium (Gibberella zeae*) is an economically important disease of wheat and barley. Yield losses could be controlled or reduced through the use of fungicides alone or in combination with biological control agents (BCAs). Field plot trials were conducted in Brookings, South Dakota and Langdon, North Dakota to analyze the efficacy of *Bacillus* strains in biological control of FHB. Spray applications of *Bacillus* BCAs alone or in combination with Prosaro® (fungicide) and/ or Induce NIS (non-ionic surfactants) and/ or colloidal chitin, and/or plant oil were done on Durum and Briggs spring wheat heads at Feekes 10.51. In the Brookings spring wheat trial, the combination of *Bacillus* 1BA, plant oil and Prosaro® reduced the FHB incidence to 5.5%, which was less than the FHB incidence observed for Prosaro alone (6.5%) or the untreated control (16.5%). Treatments of *Bacillus* 1BA with plant oil and Prosaro reduced the disease index to 0.76%, while the treatment *Bacillus* 1D3 + plant oil + colloidal chitin + Prosaro reduced it to 0.74%. Treatment of Prosaro alone reduced the FHB disease index to 0.93%, while for the untreated control it was observed to be 2.74%. Treatment differences were observed for Disease DON (deoxynivalenol), Disease FDK and Disease Protein as well. In the Langdon durum wheat trial, treatment differences were observed for FHB incidence, severity, index, yield and test weight. These trials demonstrated that *Bacillus* strains 1BA or 1D3 in combination with Prosaro and/or colloidal chitin and/or plant oil can reduce FHB incidence and FHB disease index in wheat, more than a single application of Prosaro.

INFLUENCE OF MANAGEMENT PRACTICES ON *FUSARIUM* MYCOTOXINS IN WHEAT STRAW

C.A. Bradley[1*], K.A. Ames[1], Y. Dong[2], E.A. Brucker[1] and F.L. Kolb[1]

[1]University of Illinois, Urbana, IL; and [2]University of Minnesota, St. Paul, MN
[*]Corresponding Author: PH: (217) 244-7415, Email: carlbrad@illinois.edu

ABSTRACT

The effect of foliar fungicides and resistant cultivars have been evaluated for their effects on mycotoxins in grain associated with Fusarium head blight (FHB) of wheat; however, little is known about how these FHB management practices affect mycotoxins in wheat straw. High mycotoxins levels in wheat straw could be a serious problem for livestock producers who use wheat straw for bedding in their facilities. This could be most detrimental to non-ruminant animals such as swine sows, which can eat 2 to 4 kg of wheat straw bedding per day. Research trials were conducted in 2011 and 2012 in Illinois to determine mycotoxin levels present in wheat straw (stems only) and if typical FHB management practices had an effect on mycotoxin levels. To determine the mycotoxin levels, stem samples were collected immediately after harvest, were ground into small particles, and then sent to the University of Minnesota mycotoxin testing laboratory.

Fungicide trials were conducted at four locations in Illinois (Brownstown, Dixon Springs, Monmouth, and Urbana) to determine the effects of Headline® (pyraclostrobin; BASF Corp.), Caramba® (metconazole; BASF Corp.), Prosaro® (prothioconazole + tebuconazole; Bayer CropSciences), and Folicur® (tebuconazole; Bayer CropSciences) on mycotoxins in wheat straw. All locations were planted into corn stubble and were mist-irrigated. Headline was applied at Feekes growth stage (FGS) 9, while all other fungicides were applied at FGS 10.5.1. Ranges of DON, 3ADON, 15ADON, NIV, and ZEA in wheat stems at these locations were 0.6-104.6 ppm, 0.01-5.7 ppm, 0.1-17.8 ppm, 0-1.6 ppm, and 0-1.3 ppm, respectively in 2011, and 1.1-13.3, 0-0.5, 0.2-3.2, 0-0.3, and 0-0.2 ppm, respectively in 2012. In 2011, when averaged over all locations, none of the fungicides decreased mycotoxin levels compared to the non-treated control, but Headline fungicide significantly ($P \leq 0.10$) increased 3ADON and 15ADON compared to the non-treated control. In 2012, when averaged over all locations, none of the fungicides significantly decreased or increased mycotoxins compared to the non-treated control.

Integrated management trials designed to evaluate cultivar (susceptible vs. moderately-resistant) × fungicide (Prosaro vs. non-treated) effects were conducted at Dixon Springs, Urbana, and Monmouth, IL. Two trials (mist-irrigated and non-irrigated) were conducted at Urbana each year. Ranges of DON, 3ADON, 15ADON, NIV, and ZEA in these trials were 0.1-33.5, 0-1.39, 0-10.1, 0-0.5, and 0-1.5 ppm, respectively in 2011, and 0.2-10.0, 0-0.6, 0-2.2, 0-1.1, and 0 ppm, respectively in 2012. When averaged over all trials in 2011, foliar fungicides did not affect mycotoxin levels, but the susceptible cultivar (Pioneer 25R47) had significantly greater DON levels compared to the moderately resistant cultivar (BW5228). When averaged over all trials in 2012, no significant differences were observed among any of the treatments for mycotoxins levels in the wheat stems.

A mist-irrigated cultivar evaluation trial was conducted at Urbana in 2011 and 2012. Ranges of DON, 3ADON, 15ADON, NIV, and ZEA at these locations were 8.9-54.1, 0.6-3.1, 4.9-16.6, 0, and 0-0.5 ppm, respectively in 2011, and 0.3-6.0, 0-0.5, 0.2-2.1, 0-0.7, and 0 ppm, respectively in 2012. In 2011,

significant differences in levels of DON, 3ADON, and 15ADON were observed among the cultivars, and a significant, positive Spearman correlation ($P = 0.0001$; $R = 0.50$) was detected between DON levels in straw and DON levels in grain. In 2012, no significant differences in stem mycotoxin levels were observed among cultivars, but a significant, positive Spearman correlation ($P = 0.0132$; $R = 0.36$) was detected between DON levels in straw and DON levels in grain. The significant correlations detected between DON in grain and DON in straw indicates that a cultivar's FHB resistance level may play a role in the level of DON observed in the straw.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture under agreement number 59-0206-9-076. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

EFFICACY OF CLO-1 BIOFUNGICIDE ON SUPPRESSING PERITHECIAL PRODUCTION OF *GIBBERELLA ZEAE* ON CROP RESIDUES

Yuanhong Chen[1], Allen G. Xue[1*], Harvey D. Voldeng[1], George Fedak[1], Tobias Längle[2], Jinxiu Zhang[2], Gary E. Harman[3] and Gordon Genge[4]

[1]Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada (AAFC), Ottawa, ON, Canada K1A 0C6; [2]Pest Management Centre, AAFC, Ottawa, ON, Canada K1A 0C6; [3]Department of Plant Pathology, Cornell University, Geneva, NY, USA 14456-0462; and [4]ICUS Canada, St. John's, NL, Canada A1C 2R2

*Corresponding Author: PH: 613-759-1513, Email: allen.xue@agr.gc.ca

ABSTRACT

Fusarium head blight (FHB), caused by *Gibberella zeae*, is a destructive disease of wheat. Previous studies demonstrated that *Clonostachys rosea* strain ACM941 is a *G. zeae* antagonist by inhibiting mycelial growth and reducing FHB severity. The objective of this research was to evaluate the efficacy of CLO-1, a formulated product of ACM941 for reducing perithecial production on various crop residues in comparison with registered fungicide Folicur (tebuconazole) under field conditions. When applied on *G. zeae* inoculated corn, soybean and wheat residues in spring each year of 2009 and 2010, CLO-1 significantly inhibited the perithecial production on all the crop residue types, reducing daily perithecial production (DPP) by 89.4% on corn residue, 92.2% on soybean residue and 88.6% on wheat residue, compared with the untreated control. When applied on naturally infected wheat residues in the fall each year of 2009 and 2010, CLO-1 significantly reduced DPP in the following growing season by 72.3% on peduncle, 51.0% on spikelet, and 57.2% on stem. These effects were better but not significantly different from those achieved by Folicur fungicide used as positive control in the same experiments. Results of this study suggest that CLO-1 is a promising biofungicide against *G. zeae* and may be used as a control measure in an integrated FHB management program to reduce the initial inoculum, and thus reduce FHB severity and increase yield and grain quality.

KNOWN KNOWNS AND KNOWN UNKNOWNS: ASSESSING ADOPTION OF SCAB MANAGEMENT TOOLS

C. Cowger*

USDA-ARS, Department of Plant Pathology, NCSU, Raleigh, NC
*Corresponding Author: PH: (919) 513-7388, Email: Christina.Cowger@ars.usda.gov

ABSTRACT

Over the past two decades, researchers have developed and disseminated recommendations for management of Fusarium head blight of wheat in the U.S. Of several recommendations, the three key ones are: increase acreage of moderately resistant varieties; monitor scab risk leading up to heading and flowering; and, when indicated, make a timely application of an effective fungicide. Use of these techniques has been demonstrated to significantly reduce kernel abortion, kernel damage, and mycotoxin contamination.

From anecdotal evidence, it appears that wheat grower adoption of these techniques is uneven. This suggests the need for: 1) a more systematic assessment of the extent of adoption in scab-prone areas, and 2) a deeper understanding of factors that hinder or promote adoption.

In an initial inquiry, two available sets of data on adoption were evaluated.

The first dataset was of subscriptions to the USWBSI-supported system of email and/or text message alerts. These alerts are received free of charge when advisory updates are posted to the scab risk forecasting web site. Data on subscribers consisted of state, occupation, and type of alert received. As expected, the five largest categories of subscribers were farmers, fungicide industry, grain purchasers, consultants, and extension personnel. States with the largest number of subscriptions tended to have a high percentage of farmers as subscribers. Subscriptions per 1,000 wheat farms ranged from one to 22, and varied substantially among states.

The second dataset was of 2011 acreage by variety. Those data were assessed together with ratings of commonly planted wheat varieties for 21 states that are covered by USWBSI scab risk forecasts, have significant wheat acreage, and are subject to scab epidemics. Among the results:

- In five states that produce hard wheat and durum wheat, estimates were made for an area covering over 22 million acres of wheat planted annually by about 57,000 wheat farming operations (National Agricultural Statistics Service, or NASS). Annual variety surveys by state and market class allowed estimation of percentages of acreage planted to specific wheat varieties. FHB resistance ratings for those varieties were used to classify the varieties as moderately resistant (MR), moderately susceptible (MS), or susceptible (S). The MR percentage estimates ranged from 0 to 62 and indicated where major progress has been made in breeding and growing scab-resistant varieties.

- By contrast, out of 16 scab-vulnerable states that primarily grow soft wheat, only one had published a recent survey of wheat variety acreage that allowed estimation of percentages of MR, MS, and S cultivars. In this 16-state soft wheat area, about 6.36 million acres of wheat were planted annually by about 61,300 wheat farms, according to NASS. In addition, rough estimates of market shares

of top varieties were obtained for some soft wheat states from private consultants, seed producers, and seed certification agencies.

The data suggest that major progress has been made in adoption of recommended scab techniques in some areas. It also appears there is a need for new systems to regularly obtain estimates of variety acreage in the primarily soft-wheat states. Such information would allow a stronger focus on adoption of variety resistance, and would permit monitoring of progress to occur. The data also suggest that lessons could be learned from states that have been especially successful in raising awareness and use of scab risk forecasts among growers and their advisors.

EVALUATION OF INTEGRATED METHODS FOR MANAGING FHB AND DON IN WINTER WHEAT IN NEW YORK IN 2012

J.A. Cummings, K.D. Waxman and G.C. Bergstrom*

Dept. of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, NY 14853
*Corresponding Author: PH: 607-255-7849, Email: gcb3@cornell.edu

OBJECTIVE

To evaluate the individual and interactive effects of moderately resistant cultivars and the foliar fungicide Prosaro® on wheat yield and the integrated management of Fusarium head blight (FHB) and deoxynivalenol (DON) under two environments in New York.

INTRODUCTION

In response to the USWBSI goal to validate integrated management strategies for FHB and DON, the Disease Management RAC of USWBSI initiated a multi-state, multi-year, coordinated field study. In New York during 2012, we observed the disease and yield impact of cultivar susceptibility, inoculation with *Fusarium graminearum*, and treatment with Prosaro fungicide in two different experimental environments.

MATERIALS AND METHODS

Both experiments were performed at the Musgrave Research Farm in Aurora, NY following cultural practices recommended for soft red winter wheat in the region. The four cultivars included were 'Pioneer 25R34' (classified as moderately susceptible to FHB), 'Pioneer 25R46' (classified as moderately resistant to FHB), 'Otsego' (classified initially as moderately resistant to FHB), and 'Truman' (established as moderately resistant to FHB). The two experimental environments, both planted on September 26, 2011, were characterized by the planting of winter wheat no-till into 1) soybean residue and 2) corn residue in immediately adjacent parcels of land. Each experimental design was a split-split plot with four wheat cultivars as whole plots, inoculation treatment as subplot, and fungicide treatment as sub-subplot, in four replicated blocks. Main plots were planted with a 10 ft wide commercial grain drill and were 20 ft long. Spray treatments applied at Feekes GS10.5.1 on 5/23/12 were 1) non-sprayed, non-inoculated 2) Prosaro 6.5 fl oz/A & Induce 0.125%, non-inoculated 3) non-sprayed and inoculated with *F. graminearum*; and 4) Prosaro 6.5 fl oz/A & Induce 0.125% and inoculated with *F. graminearum*. Treatments 3 and 4 were inoculated with a conidial suspension of *F. graminearum* (40,000 conidia/ml) on the same day as the Prosaro application after the fungicide had dried and in early evening to provide a better environment for infection. Prosaro and *F. graminearum* applications were applied with a tractor-mounted sprayer with paired Twinjet nozzles mounted at an angle (30° from horizontal) forward and backward and calibrated to deliver at 20 gallons per A. FHB and foliar diseases were assessed at soft dough stages. Grain was harvested from a 4 ft wide x 20 ft long area in each subplot using a Hege plot combine on July 3, 2012. Grain moistures, plot yields, and test weights were recorded with the latter two adjusted for moisture. Means were calculated and subjected to Analysis of Variance. Fisher's protected LSD was calculated at $P = 0.05$. Analysis of DON content in grain was conducted in the USWBSI-supported mycotoxin laboratory of Dr. Dong.

RESULTS AND DISCUSSION

Both experimental environments were located in the same field that in the previous year was split, growing corn in one half and soybean in the other. Flowering occurred simultaneously in both environments during a relatively dry period, considered low risk for FHB infection. FHB

incidence at soft dough stage was well below 1% in all plots so was recorded as zero.

The impact of *F. graminearum* inoculation was determined by comparing the non-inoculated and inoculated treatments (combining non-sprayed and Prosaro treatments) in both experiments (environments). Inoculation did not significantly affect yield, FHB index, or DON in either experiment. Cultivars did not respond differentially to inoculation in either environment.

Significant differences in yield were detected among cultivars in each environment. Following soybean, only Otsego had significantly higher yield than that of all other cultivars. Following corn, Otsego had the highest yield, which was significantly higher than Pioneer 25R34 and Pioneer 25R46; and Pioneer 25R34 had the lowest yield, which was significantly lower than Otsego and Truman. Yield for each cultivar was significantly higher following soybean than following corn. This may be attributable to increased nitrogen following soybean, but this was not measured. However, within each environment, there was no significant difference in yield within each cultivar, regardless of spray treatment or inoculation, except for the non-inoculated, non-sprayed P25R34. For some unknown reason this treatment resulted in a yield significantly lower than all other treatments for this variety. In the virtual absence of disease pressure, cultivars differed significantly in yield with Otsego consistently yielding highest.

When results of all the cultivars were combined, the overall impact of the Prosaro applications was not significant, regardless of environment, for yield or FHB index. No significant difference was detected for DON contamination among any of the spray treatments in either environment. The only significant difference in DON contamination of grain was for the cultivar Pioneer 25R34, which although still far below the threshold of agronomic importance, i.e., 2 ppm, was significantly higher than that of the other varieties following corn. In this experiment, with little to no fungal disease pressure, the Prosaro application caused no significant improvement of yield or reduction in FHB index or DON contamination of grain.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported in part by the U.S. Department of Agriculture under agreement No. 59-0206-9-056. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

Table 1. Main effect of treatment on grain yield, Fusarium head blight index, and deoxynivalenol contamination at Aurora, NY.

	Adjusted grain yield (bu/A)		
Treatment:	After corn	After soybean	Average
Non-sprayed, non-inoculated	90	123	107
Prosaro, non-inoculated	89	124	107
Non-sprayed, inoculated	90	129	110
Prosaro, inoculated	92	127	110
LSD (*P*=0.05)	NS	NS	
	Fusarium head blight index (%)		
Treatment:	After corn	After soybean	Average
Non-sprayed, non-inoculated	0	0	0
Prosaro, non-inoculated	0	0	0
Non-sprayed, inoculated	0	0	0
Prosaro, inoculated	0	0	0
LSD (*P*=0.05)	NS	NS	
	Contamination of grain by DON (ppm)		
Treatment:	After corn	After soybean	Average
Non-sprayed, non-inoculated	0.05	0.02	0.04
Prosaro, non-inoculated	0.06	0.03	0.05
Non-sprayed, inoculated	0.04	0.04	0.04
Prosaro, inoculated	0.05	0.03	0.04
LSD (*P*=0.05)	NS	NS	

Table 2. Main effect of cultivar on grain yield, Fusarium head blight index, and deoxynivalenol contamination at Aurora, NY.

	Adjusted grain yield (bu/A)		
Cultivar:	After corn	After soybean	Average
Otsego	101 a	137 a	119
Pioneer 25R34	78 c	124 b	101
Pioneer 25R46	87 bc	125 b	106
Truman	96 ab	118 b	107
LSD (*P*=0.05)	11.5	10.9	
	Fusarium head blight index (%)		
Cultivar:	After corn	After soybean	Average
Otsego	0	0	0
Pioneer 25R34	0	0	0
Pioneer 25R46	0	0	0
Truman	0	0	0
LSD (*P*=0.05)	NS	NS	
	Contamination of grain by DON (ppm)		
Cultivar:	After corn	After soybean	Average
Otsego	0.02 b	0.03	0.03
Pioneer 25R34	0.12 a	0.04	0.08
Pioneer 25R46	0.04 b	0.03	0.04
Truman	0.05 b	0.02	0.04
LSD (*P*=0.05)	0.040	NS	

EVALUATING THE EFFECT OF FUNGICIDE CHEMISTRY AND APPLICATION TIMING ON FHB, DON AND *FUSARIUM GRAMINEARUM* BIOMASS IN SOFT RED WINTER WHEAT

Daisy D'Angelo, Katelyn Willyerd, Antonio Cabrera, Laurence Madden and Pierce Paul*

Department of Plant Pathology, The Ohio State University, OARDC, Wooster, OH 44691
*Corresponding Author: PH: (330) 263-3842, Email: paul.661@osu.edu

ABSTRACT

Quinone Outside Inhibitors (QoI) and Demethylation Inhibitors (DMI) are highly effective fungicides against foliar diseases, and as such, are important components of wheat disease management programs. However, while the DMIs are highly recommended for Fusarium head blight (FHB) and deoxynivalenol (DON) control, the QoIs are not. This is largely because some members of the latter group of fungicides have been shown to increase DON in wheat grain, especially when applied close to anthesis. However, it is unclear whether DON response to QoI fungicides is consistent across active ingredients (AIs); whether the response in influence by application timing and weather conditions; and whether it is associated with an increase in fungal colonization of the grain. A field study was conducted in Wooster, OH, to evaluate the effects of two QoI fungicide AIs (Azoxystrobin and Pyraclostrobin), applied at different growth stages, FHB, DON and *F. graminearum* biomass (FBM) in wheat. DMI fungicide treatments were also included as references for comparison with the QoI treatments. The experimental design was a randomized complete block with 9 treatments plus an untreated check. The treatments consisted of Headline® (6 fl. oz./A), Quadris® (9 fl. oz./A), and Prosaro® (6.5 fl. oz./A) applied at Feekes growth stages (GS) 8 (flag leaf emergence), GS 10 (boot), and GS 10.5.1 (flowering). A nonionic surfactant was added to each treatment at a rate of 0.125% v/v. All plots were spray-inoculated with a spore suspension of F. graminearum approximately 24 h after the anthesis treatments were applied. FHB intensity was rated approximately three weeks after anthesis, and a sample of grain from each plot was visually rated for *Fusarium* damaged kernels (FDK) and analyzed for DON and fungal biomass (*Fusarium* infection) using gas chromatography-mass spectrometry and quantitative real-time PCR laboratory techniques. The highest levels of DON and fungal biomass (FBM) were observed in the QoI treatments (AI x timing combination), with two of those treatments (Quadris at Feekes GS 8 and GS 10) being significantly different from the untreated check for DON contamination and one (Quadris at GS 10) significantly different from the check for fungal biomass. However, the levels of FHB index and FDK in the Quadris GS 8 and Quadris GS 10 treatments were not significantly different from the untreated check, suggesting that relative to the check, these two treatments resulted in disproportionately higher levels of DON contamination and fungal colonization of the grain than expected based on visual symptoms. This was confirmed by the fact that these same two treatments had significantly lower index:DON, FDK:DON, and index:FBM ratios than the untreated check.

BOOSTED REGRESSION TREES IDENTIFY PRE- AND POST-ANTHESIS WEATHER VARIABLES FOR PREDICTING FUSARIUM HEAD BLIGHT EPIDEMICS

E. De Wolf[1*], D. Shah[1], P. Paul[2], K. Willyerd[2] and L. Madden[2]

[1]Department of Plant Pathology, Kansas State University, Manhattan KS; and
[2]Department of Plant Pathology, The Ohio State University, Wooster OH
[*]Corresponding Author: PH: 785 537-7671, Email: dewolf1@ksu.edu

ABSTRACT

The project's goal was to identify weather variables useful for predicting major Fusarium head blight (FHB) epidemics in the United States. The dataset consisted of 527 unique observations of major (severity $\geq 10\%$) and non-major (severity $< 10\%$) epidemics, linked to 380 weather-based predictors summarizing temperature (t), relative humidity (rh) or rainfall (r) in fixed-length windows pre- and post-anthesis; as well as binary indicators for the level of genetic resistance, the presence of crop residue as a local inoculum source, and the type of wheat (winter vs. spring wheat). Boosted Regression Trees (BRTs) were fit to a subset consisting of 70% of the data, and model test error was estimated on the remaining 30% of the data. The BRT models have deepened the understanding of how variables are associated with FHB epidemics, and have suggested novel representations of weather that may be more associated with FHB epidemics than predictors now used. BRT models confirmed that rh is an important predictor of major FHB epidemics, but also indicated a greater importance of t and r than previously suggested by earlier modeling approaches. The BRT models also identified variable interactions that were difficult to detect in previous analyses. Results of this analysis will be integrated into existing models for FHB epidemics to improve predictive accuracy.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-7-072 and 59-0206-9-071. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture

FUSARIUM HEAD BLIGHT MANAGEMENT: PROGRESS AND POSSIBLE KNOWLEDGE GAPS

E. De Wolf *

Department of Plant Pathology, Kansas State University, Manhattan KS
*Corresponding Author: PH: 785 537-7671, Email: dewolf1@ksu.edu

ABSTRACT

The cooperative effort to improve the management of Fusarium head blight (FHB) within the United States has made several important advances since the beginning of the US Wheat and Barley Scab Initiative (USWBSI). Early advances included an improved understanding of fungicide efficacy for various active ingredients and application technologies that could maximize the value of these products. As varieties with genetic resistance became more widely available, the group began investigating the potential benefits of combining genetic resistance with fungicides for improved FHB control. Advances in disease forecasting models for FHB complimented the integrated management research and help growers evaluate the risk of severe epidemics and the need for fungicide applications. Despite these advances, the management of FHB remains a challenge for many growers because of incomplete information about the FHB reaction of common varieties, and factors that compromise the efficacy of fungicide applications. To address these challenges, we must address these knowledge gaps with a renewed commitment to FHB management research and communication efforts.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-7-072. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

UNIFORM FUNGICIDE TRIAL RESULTS FOR MANAGEMENT OF FHB AND DON, 2012

M. McMullen[1*], A. Friskop[1], J. Jordahl[1], S. Meyer[1], G. Bergstrom[2], C.A. Bradley[3], R. Dill-Macky[4], M. Smith[5], J. Wiersma[5], S. Halley[6], A. Arens[6], G. Milus[7], K. Ruden[8] and B. Schatz[9]

[1]North Dakota State University, Fargo, ND; [2]Cornell University, Ithaca, NY; [3]University of Illinois, Urbana, IL; [4]University of Minnesota, St. Paul, MN; [5]University of Minnesota, Crookston, MN; [6]Langdon Research Extension Center, Langdon, ND; [7]University of Arkansas, Fayetteville, AR; [8]South Dakota State University, Brookings, SD; and [9]Carrington Research Extension Center, Carrington, ND
[*]Corresponding Author: PH: (701) 231-7627, Email: marcia.mcmullen@ndsu.edu

ABSTRACT

Uniform fungicide trials were conducted on multiple classes of wheat (durum, hard red spring, hard red winter, and soft red winter) across six states in 2012. Trials were established at multiple locations in Illinois, North Dakota and South Dakota, and one location in Arkansas, Minnesota and New York. All sites had either added inoculum in the form of *Fusarium graminearum* infested grain spawn, infested residue, or the fungus was spray inoculated at flowering. Several sites used mist or overhead irrigation to promote disease development. Four test sites (Fargo, ND; Ithaca, NY; Fayetteville, AR; and Groton, SD) were too hot and too dry for adequate development of FHB to separate treatments. Twelve site/variety combinations did have adequate FHB for determining efficacy of treatments. Thirteen treatments were evaluated across six of these location/variety sites, while 10 treatments were evaluated across 12 location/variety sites. Treatments included triazole fungicides - Caramba® (metconazole), Prosaro® (prothioconazole + tebuconazole); and Folicur® (tebuconazole) applied alone at Feekes 10.5, 10.5.1 or 5 days after Feekes 10.5.1; and a strobilurin fungicide - Headline (pyraclostrobin) applied alone at Feekes 9.0, or combinations of Headline at Feekes 9 followed by Caramba, Prosaro, or Folicur at Feekes 10.5.1. Sites that had only 10 treatments generally did not include the Caramba or Prosaro applied at Feekes 10.5 or the Headline treatment at Feekes 9. Preliminary examination of the data indicates:

- FHB Index (% Field Severity): Triazole fungicide treatments generally reduced FHB index from that of the untreated. For sites with all 13 treatments, the highest FHB index among fungicide treatments generally was with Headline applied alone, at Feekes 9. For sites that did not have this treatment, the highest FHB index value most frequently observed among fungicide treatments was Headline at Feekes 9 followed by Folicur at Feekes 10.5.1, and lowest values generally with a triazole fungicide applied at Feekes 10.5.1.

- DON (ppm): For sites with all 13 treatments, Headline applied once at Feekes 9 generally had the highest DON level apart from the untreated check. Triazole fungicides applied at Feekes 10.5.1 or five days after Feekes 10.5.1 generally resulted in lowest DON levels across sites.

- Yields ranged from 30+ bu in some hard red spring wheat sites to 80 to 90+ bushels in some Illinois winter wheat sites, and yield responses to treatment varied among sites. When yields were converted to percent of untreated for each trial, average yield responses to fungicides generally ranged from 5 to 35%, but greater responses were observed in MN.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-064 and is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

UNIFORM TESTS OF BIOLOGICAL CONTROL AGENTS FOR MANAGEMENT OF FHB AND DON, 2012

M. McMullen[1*], G. Bergstrom[2], J. Cummings[2], C. Jochum[3], G. Yuen[3], B.H. Bleakley[4], N.K.S. Murthy[4] and K. Ruden[4]

[1]North Dakota State University, Fargo, ND; [2]Cornell University, Ithaca, NY; [3]University of Nebraska, Lincoln, NE; and [4]South Dakota State University, Brookings, SD
[*]Corresponding Author: PH: 701-231-7627, Email: marcia.mcmullen@ndsu.edu

ABSTRACT

A uniform set of eight biological or biological plus fungicide treatments were compared to an untreated check for evaluation of control of Fusarium Head blight (FHB) and DON (deoxynivalenol) in wheat. Taegro (bacterium *Bacillus subtilis* var. *amyloliquefacians* strain FZB24 containing 5.0 x 1010 cfu/g, Novozymes Biologicals Inc.) was the test biological agent, applied either alone at five to seven days after Feekes growth stage 10.5.1, or following application of a triazole fungicide at Feekes growth stage 10.5.1, and with or without canola oil as an adjuvant. Treatments were applied to soft red winter wheat (Aurora, NY), hard red winter wheat (Mead and Havelock, NE), and hard red spring wheat (Volga, SD). Disease levels were low at the four sites in 2012, because of the occurrence of high temperatures and drought, although each site had either natural inoculum, infested grain spawn, or sprayed inoculum to increase disease potential. FHB Index (% field severity) values were determined at three of the four sites, and were non-significant among treatments at the two NE sites, and generally not significantly different than the untreated check at the SD site. *Fusarium* damaged kernels (FDK) values were not different among treatments. DON levels were below detectable levels at the two NE sites, and less than one ppm for all treatments at the NY and SD sites. Results in NY and SD indicated that DON levels generally were significantly reduced with the triazole fungicide treatments or with Taegro if applied in combination with a triazole fungicide. Yield impacts were non-significant at three locations. At the SD site, yield was lowest in the untreated check, and significantly improved with several fungicide or Taegro plus fungicide treatments.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture under Agreement No. 59-0206-9-064 and is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

BARLEY MONOCULTURE SYSTEM: EFFECT OF TILLAGE PRACTICES ON DEOXYNIVALENOL (DON) CONTENT

D. Pageau[1*], J. Lajeunesse[1], J. Lafond[1] and B. Blackwell[2]

[1]Agriculture and Agri-Food Canada, Research Farm, Normandin, Quebec, Canada; and [2]Agriculture and Agri-Food Canada, Eastern Cereal and Oilseed Research Centre, Ottawa, Ontario, Canada
[*]Corresponding Author: PH: (418) 274-2941, Email: denis.pageau@agr.gc.ca

ABSTRACT

Fusarium head blight (FHB) associated with the presence of the fungus *Fusarium graminearum* is probably one of the most feared diseases in barley (*Hordeum vulgare*) production in Eastern Canada. In addition to reducing grain yields, the fungus produces a toxin (deoxynivalenol or DON) which can affect the health of livestock. The objective of this study was to determine the impact of different tillage systems on DON content in barley. The tillage systems were: T1: Conventional (fall moldboard plowing and spring harrowing – 2 passes with a cultivator), T2: Chisel (fall) and spring harrowing (2 passes with a cultivator), T3: Chisel (fall) and spring harrowing (1 pass with cultivator), T4: No tillage (fall) and spring harrowing (1 pass with a rotative harrow), T5: No tillage (fall) and spring harrowing (1 pass with a cultivator) and T6: No-till (no tillage the previous fall and no harrowing in spring). Those tillage systems were initiated in 1990 and since then, barley was grown without rotation. The DON content of grain has been determined since 2003. In general, the highest DON contents were obtained with direct seeding (T6) while the lowest DON contents were obtained with plowing (T1) or chiseling (T2 and T3). Because the fungus that causes FHB survives on residue left on the soil, and according to the results of this trial, tillage practices that bury cereal residue could be used to reduce deoxynivalenol (DON) content in barley.

2012 UNIFORM FUNGICIDE PERFORMANCE TRIALS FOR THE SUPPRESSION OF FUSARIUM HEAD BLIGHT IN SOUTH DAKOTA

K.R. Ruden*, G.S. Redenius, K.D. Glover and J.L. Kleinjan

Plant Science Department, South Dakota State University, Brookings, SD 57007
*Corresponding Author: PH: (605) 688-6246; Email: kay.ruden@sdstate.edu

ABSTRACT

Fusarium head blight (FHB – scab) has been a serious concern for wheat and barley producers in South Dakota for over twenty years. The objective of this study was to continue to evaluate the effects of various fungicides and fungicide combinations along with different application timings for the suppression of Fusarium head blight and other wheat diseases. Two hard red spring wheat cultivars, 'Brick' and 'Oklee', were planted at three South Dakota locations (Groton, South Shore/Watertown and Volga). 'Wesley' winter wheat study sites were also established at South Shore/Watertown and Volga. Studies at Groton and South Shore were conducted under ambient conditions. The Volga site was under ambient conditions until anthesis, after which mist irrigation was applied. Trial treatments from the Uniform Fungicide Trial treatments list for the suppression of FHB included an untreated check and the following fungicides: Applied at Feekes growth stage 10.51: Caramba® (14 fl oz/A), Prosaro® (6.5 fl oz/A) and Onset® (4 fl oz/A); Applied at Feekes growth stage 9: Headline SC® (6 fl oz/A) followed by Caramba (14 fl oz/A) at Feekes growth stage 10.51; Applied at Feekes growth stage 9: Headline SC (6 fl oz/A) followed by Prosaro (6.5 fl oz/A) at Feekes growth stage 10.51; Applied at Feekes growth stage 9: Headline SC (6 fl oz/A) followed by Onset (4 fl oz/A) at Feekes growth stage 10.51; Applied at Feekes growth stage 10.51: Onset (4 fl oz/A) plus Caramba (10 fl oz/A) together; Applied at 3-7 days after Feekes growth stage 10.51: Caramba (14 fl oz/A) and Prosaro (6.5 fl oz/A). All treatments except the Headline SC treatments included Induce, a non-ionic surfactant, applied at 0.125% v/v. Spring wheat trials were planted in a factorial randomized complete block design with six replications. Winter wheat locations were planted in a randomized complete block design with four replications. Spring wheat plots at the Volga location were inoculated by spreading *Fusarium graminearum* (isolate Fg4) inoculated corn (*Zea mays*) grain throughout the field and providing overhead mist irrigation applied from 5:00 pm until 10:00 pm each day for two weeks following anthesis. Other sites had natural inoculum from corn stalk residue and natural moisture conditions. Twenty-one days following treatment, plots were evaluated for leaf diseases, FHB incidence, FHB head severity, and FHB field severity. Samples were collected for Fusarium damaged kernels (FDK), deoxynivalenol (DON), grain yield and test weight. No significant treatments were found at the Groton location because ambient conditions were too hot and dry for FHB to develop. At the South Shore/Watertown location for spring wheat, the resistant variety, 'Brick', did not have any FHB or DON present for any of the treatments whereas in the susceptible variety "Oklee", visual scab ratings were not found but one treatment was positive for DON, namely the Onset + Caramba treatment sprayed at Feekes 10.51. At the Volga location for spring wheat, no significant treatments for FHB Incidence, Severity and Field Index for the resistant variety "Brick" were found, but all treatments were significant in reducing FDK levels. On the susceptible variety "Oklee", three treatments showed significance for FHB Incidence: Caramba applied at 3-7 days after Feekes 10.51, Headline applied at Feekes 9 + Prosaro applied at Feekes 10.51 and Prosaro applied at 3-7 days after Feekes 10.51. No treatments caused a significant reduction in FDK or DON on Oklee at Volga.

Overall, the hot and dry weather was not conducive for FHB development this year in the state of South Dakota. Even at the Volga site where artificial inoculation and misting were used, no significant amount of DON was produced.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-050. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

CHARACTERIZATION OF DIFFERENTIAL SPIKE COLONIZATION AND DEOXYNIVALENOL ACCUMULATION IN SUSCEPTIBLE AND RESISTANT WHEAT CULTIVARS

Jorge David Salgado, Larry V. Madden and Pierce A. Paul*

Department of Plant Pathology, The Ohio State University, OARDC, Wooster, OH 44691
*Corresponding Author: PH: (330) 263-3842, Email: paul.661@osu.edu

ABSTRACT

Fusarium head blight (FHB) development and deoxynivalenol (DON) contamination of wheat are difficult to control. The two major means of combating FHB are the application of fungicides and the use of resistant cultivars in an effort to reduce visual symptoms (spike necrosis) and associated grain yield and quality losses. However these approaches do not always provide adequate reduction of DON levels. Although different types of resistance to FHB are reported and commonly used in FHB research, these have not been completely characterized. There is little research-based information on how different types of resistance are interrelated and how weather conditions affect their manifestation. Type II resistance (fungus spread) does not always parallel Type III resistance (mycotoxin accumulation). Under certain weather conditions, cultivars with high levels of Type II resistance may have DON accumulation comparable to that of cultivars with lower levels of Type II resistance. The goal of this study was to better characterize resistance to FHB in wheat through a better understanding of infection, colonization, and DON accumulation in symptomatic and asymptomatic spikes of resistant and susceptible cultivars. Two soft red winter wheat cultivars, one moderately resistant (Truman) and the other susceptible (Cooper) to FHB (based on visual symptoms), were grown under greenhouse conditions. The experimental design was a randomized complete block with a split-split-plot arrangement of temperature (whole plot), cultivar (sub-plot), and moisture durations (sub-sub plot). Individual plants reaching anthesis (Feekes GS 10.5.1) were point-inoculated in the central floret of the central spikelet with a highly aggressive isolate of *F. graminearum*. After inoculation, separate groups of plants were incubated in growth chambers at one of three temperatures (15, 20 and 25°C), at high relative humidity (>90%). A subset of spikes was sampled at three-day intervals from each growth chamber, for a total of 21 days. For each spike, spikelets, rachises and grain were sampled at regular intervals above and below the point of inoculation and assayed for fungal growth and DON. Spread within the spike was quantified by measuring the distance of mycelial growth and necrosis along the rachis from the point of inoculation. At 15°C, fungal growth inside the rachis, both above or below the point of inoculation, was observed in the susceptible cultivar after 6 to 9 days of high moisture and after 9 to 12 days in the resistant cultivar. When the cultivars were compared at higher temperatures (20 and 25°C), fungal growth in the rachis was observed after 3 days in the susceptible cultivar and after 6 days in the resistant cultivar. The mean number of bleached spikelets above or below the point of inoculation ranged from 0 to 2 at 15°C and from 0 to 7 at 20 and 25°C, depending on the moisture duration. At 15°C, it took less time (15 to 18 days) for two spikelets above and/or below the inoculated spikelet to become bleached on the susceptible than the resistant cultivar (21 days). When compared at 20°C, two bleached spikelets were observed at 6 days after inoculation (dai) on the susceptible cultivar compared to 9 dai on the resistant cultivar. However, when compared at 25°C, no visual differences in the number of bleached spikelets were observed between Cooper and Truman, and both cultivars had two bleached spikelet above or below the point of inoculation at 12 dai. Averaged across all moisture durations, measuring from the point of inoculation, mean necrotic damage of the rachis at the low temperature (15°C) ranged from

0 to 10.71 mm for Cooper and from 0 to 3.39 mm for Truman; at the mild temperature (20°C), mean necrotic damage ranged from 0 to 14.94 mm and 0 to 33.13 mm for Cooper and Truman, respectively; and at the high temperature (25°C) from 0 to 34.56 mm and 0 to 23.67 mm for Cooper and Truman, respectively. DON results were not available at the time this report was being prepared.

INTEGRATED MANAGEMENT STRATEGIES FOR FUSARIUM HEAD BLIGHT OF SOFT RED WINTER WHEAT IN MISSOURI: SUMMARIZATION OF SIX YEARS OF TRIAL DATA

Laura E. Sweets*

Division of Plant Sciences, University of Missouri, Columbia, MO
*Corresponding Author: PH: (573) 884-7307, Email: SweetsL@missouri.edu

OBJECTIVE

To evaluate the importance of crop sequence, variety selection and fungicide application as components of an integrated management program for Fusarium head blight (FHB) of soft red winter wheat in Missouri.

INTRODUCTION

The severity of FHB or scab epidemics in the United States has caused enormous yield and quality losses in both wheat and barley over the last decade or more. The development of this disease is dependent on the genetics of the host, favorable environmental conditions, the prevalence of the causal fungus and the survival and spread of the causal fungus. Control of this disease has been difficult because of the complex nature of the host-pathogen interaction. Management of FHB and the associated mycotoxin DON have not been achieved by any single control measure. An integrated approach is critical to attaining the best possible management of FHB and DON in any given environment.

As a result of a workshop sponsored by the Chemical, Biological and Cultural Control Research Area of the U.S. Wheat & Barley Scan Initiative in 2006, a protocol for a multi-state project focusing on integrated management strategies for FHB was developed. The research portion of the project has been multi-state trials evaluating crop sequence, variety selection and fungicide application as an integrated management program for FHB.

The University of Missouri has participated in the multi-state integrated management project for the past six growing seasons. Results from the six years are summarized in this poster abstract.

MATERIALS AND METHODS

During the fall of 2006 two adjacent fields at the University of Missouri Bradford Research and Extension Center just east of Columbia, MO, were identified for this study. The fields had been in a corn/soybean rotation for at least five years prior to the initiation of the study and were separated by a small drainage ditch. The wheat trials were planted into standing corn residue or soybean residue on the same day. The remainder of each field was planted into the normal rotation crop of corn or soybean. In subsequent years, the wheat trials were shifted to other areas of the same fields with the remainder of the fields planted to the normal rotational crop.

Five soft red winter wheat varieties with similar heading times and varying reactions to FHB were selected for the trial. The five varieties included the public varieties Bess (tolerant) and Roane (moderately tolerant) which are widely grown in Missouri, the Agri-Pro variety Elkhart (susceptible) and the Pioneer varieties 25R47 (moderately susceptible) and 25R54 (moderately tolerant).

In the fall of 2006 the trials were planted no-tillage into either soybean residue or standing corn residue on the same day. Individual plots were 7 rows (~7.5" row spacings) by 30' in length. Each trial was set up as a split plot trial with fungicide application as the main plot and variety

as the sub-plot. There were six replicates in each trial. Sub-plots were separated by buffer plots. The foliar fungicide Prosaro® (6.5 fl oz/A) was applied at Feekes growth stage 10.5.1. A non-ionic surfactant was added to the fungicide at the rate of 0.125% v/v, and application was made using a CO2 pressurized backpack sprayer with TwinJet XR8002 nozzles mounted at an angle (30 and 60 degrees) forward and backward.

Plots were evaluated for incidence and severity of FHB, yield was taken, grain samples were submitted to North Dakota State University for DON analysis and grain samples were rated for percent of Fusarium damaged kernels (FDK). Analysis of variance was used to determine the effects of variety, fungicide and their interactions on yield, DON levels and FHB index (average of 100 wheat heads per plot) and percent FDK for each residue type.

The trial was repeated following the same protocol during the next five growing seasons.

RESULTS

Weather conditions during the 2007 growing season were not conducive for the development of FHB at the Columbia, MO location. Conditions as the wheat crop was flowering were too dry for infection to occur and for disease to develop. However, the following five seasons have been more conducive for the development of FHB. In 2008, 2009 and 2010 weather conditions were unusually wet and cool as the wheat crop flowered and after flowering so both scab and DON levels were high. The 2011 and 2012 seasons were wet during flowering so scab developed but were hot and dry during grain fill so FHB index and DON levels were lower than in the previous three years.

The results from the six years of this trial demonstrate the importance of crop sequence, variety selection and fungicide application in reducing FHB and DON levels in soft red winter wheat grown in Missouri. Planting wheat after soybean rather than corn showed a reduction in both FHB and DON even in years which were not particularly favorable for the development of FHB. Crop sequence and variety selection appear to be valuable preventative measures for reducing FHB and DON levels. The application of the fungicide Prosaro at FGS 10.5.1 tended to reduce FHB levels and increase yields for most of the varieties on both crop sequences with effects being more pronounced on susceptible and moderately tolerant varieties. The data from the six years of this trial indicate that an integrated management approach employing crop sequence and variety selection as pre-plant preventative management measures and fungicide application during the growing season if weather conditions at flowering warrant application may be beneficial in reducing FHB, reducing DON levels and increasing yield for soft red winter wheat grown in Missouri.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based on work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-078. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions or recommendations expressed in this publication are those of the author and do not necessarily reflect the view of the U.S. Department of Agriculture.

UNIFORM FHB INTEGRATED MANAGEMENT TRIALS: A SUMMARY FROM 2012

K. Willyerd[1], K. Ames[2], G. Bergstrom[3], C. Bradley[2], J. Cummings[3], P. Gross[4], L. Madden[1], J. Ransom[4], K. Ruden[5], J.D. Salgado[1], L. Sweets[6], K. Wise[7] and P. Paul[1*]

[1]The Ohio State University/OARDC, Wooster, OH 44691; [2]University of Illinois, Urbana, IL 61801; [3]Cornell University, Ithaca, NY 14853; [4]North Dakota State University, Fargo, ND 58102; [5]South Dakota State University, Brookings, SD 57007; [6]University of Missouri, Columbia, MO 65211; and [7]Purdue University, West Lafayette, IN 47907
[*]Corresponding Author: (PH) 330-263-3842; Email: paul.661@osu.edu

OBJECTIVE

To evaluate the integrated effects of fungicide and genetic resistance on FHB and DON in small grain crops in different environments.

INTRODUCTION

From 2009 to 2011, coordinated, uniform trials were conducted in multiple states to evaluate the effects of grain class, crop rotation, cultivar resistance, and fungicide application on management of FHB and DON. Results from over 40 wheat trials demonstrated using fungicide or moderate resistance alone resulted in approximately 53 and 54% control in FHB index, respectively; and 39 and 51% control in DON, respectively (Willeyard et al). Combining moderate resistance (in hexaploid wheat) and fungicide resulted in 76 and 71% control of index and DON; furthermore, the efficacy of this integrated approach was stable across different environments and cropping systems. However, more research is required to evaluate these effects under higher disease pressure and in other small grain classes such as barley and durum wheat. In a given year of this coordinated effort, 20 to 54% of trials were eliminated from analysis due to little to no FHB and/or DON in the susceptible, untreated check, as low disease in the reference makes calculating the effects of treatment impossible. This report summarizes results from trials conducted during the 2012 season, including trials with the factor of artificial inoculum, which was hypothesized to increase the amount of "useable" data (that is, trials with significant disease in the check).

MATERIALS AND METHODS

Trials were established in fields following a host or non-host crop of *F. graminearum*. At least three commercial small grain cultivars, classified as susceptible (S), moderately susceptible (MS) or moderately resistant (MR), were planted in three to six replicate blocks in each trial. The standard experimental design was a randomized complete block, with a split-split-plot arrangement of cultivar (whole-plot), inoculation (sub-plot) and fungicide treatment (sub-sub-plot; UT, untreated and TR, treated). Some trials used fungicide as whole-plot and cultivar as sub-sub-plot; while others did not include inoculation as a factor. Fungicide (Prosaro®, 6.5 fl. oz/A + NIS) was applied at anthesis, using CO_2 powered sprayers, equipped with Twinjet XR8002 or paired XR8001 nozzles, mounted at a 30 or 60^o angle, forward or backward. For trials with artificial inoculations, either *F. graminearum*-colonized corn kernels were spread on the soil surface of plots prior to anthesis or plots were spray-inoculated with a spore suspension of the fungus approximately 24 hours following fungicide treatments. FHB index (plot severity) was assessed during the dough stages of grain development. Milled grain samples were sent to a USWBSI-supported laboratory for toxin analysis. Proc GLIMMIX of SAS was used to evaluate the effects of fungicide, cultivar, (and inoculation, when appropriate) and their interactions on index

and DON (assuming a significance level $\alpha = 0.05$). Percent control was calculated to compare the effect of cultivar resistance and fungicide treatment combinations (S_TR, MS_UT, MS_TR, MR_UT and MR_TR) on index and DON to the susceptible, untreated check (S_UT).

RESULTS AND DISCUSSION

At the time of this summary, data were collected from 22 trials, conducted in 7 states (IL, IN, MO, ND, NY, OH and SD) (Table 1). These included 9 soft red winter wheat (SRWW), 4 hard red winter wheat (HRWW), 4 six-row barley, 3 hard red spring wheat (HRSW) and 2 two-row barley trials. FHB index and DON accumulation varied among locations and grain classes (Table 1). Overall, mean FHB was 0% in many locations (environments 1, 2, 3, 7, 8, 9, 10 and 11), and as a result, DON accumulation was also low or samples were simply not sent for toxin analysis (Table 1). Only environments with > 5% mean index (4, 5, 6, 12, 13, 21 and 22) and/or >1 ppm mean DON (4, 5 and 6) in the susceptible, untreated check were included in this analysis (Table 1). Means for cultivar resistance class x fungicide treatment combinations and percent control of index and DON, relative to the untreated susceptible check (S_UT), are found in Table 2.

Illinois. Six soft red winter wheat cultivars were planted into host residues in two trials that included artificial inoculation as a factor. Despite the presence of FHB in both trials, DON levels were well below 1 ppm perhaps due to rapid grain maturation and relatively early harvest in 2012. **Monmouth (ENV 21).** Index, DON and yield observations ranged from 0 to 100%; 0 to 0.2 ppm and 75.7 to 106.7 bu/A, respectively. Neither cultivar, fungicide treatment (Table 3) nor inoculation (data not shown) had significant effects on index. In this environment the MS_TR combination resulted in the greatest control in index (97%), followed by MS_UT and MR_TR (both approximately 70%, Table 2). **Urbana (ENV 22).** Index, DON and yield observations ranged from 0 to 18%; 0 to 0.4 ppm and 52.8 to 94.4 bu/A, respectively. Both cultivar and fungicide had significant effects on index and DON (Table 3). Inoculation did not have a significant effect on either response (data not shown). All management combinations resulted in <3% FHB index and reduced index by over 60% compared to the S_UT check (Table 2)

Missouri. Five soft red winter wheat cultivars were planted into host and non-host crop residues near Columbia, MO. These two trials relied on ambient inoculum. Overall, mean FHB index, DON and yield were greater in the non-host (soybean) environment compared to that of the trial planted following corn, despite the same planting and treatment dates. **Host residues (ENV 4).** Index, DON and yield observations ranged from 6.7 to 32%; 0 to 4 ppm and 45.7 to 90.0 bu/A, respectively. Both cultivar and fungicide treatment had significant effects on index and DON (Table 3). Generally, index and DON decreased with improved resistance and fungicide treatment application, however, S_TR showed improved DON control compared to MS_UT (Table 2). The MR_TR combination resulted in the greatest control in index and DON (57 and 89%, respectively) compared to the S_UT check. **Non-host residues (ENV 5).** Index, DON and yield observations ranged from 7.3 to 39.1%; 0 to 12.5 ppm and 52.2 to 95.5 bu/A, respectively. The cultivar x fungicide interaction had significant effects on DON only (Table 3). Similar to the host residue environment, S_TR showed improved DON control compared to MS_UT and MR_TR had the greatest DON control (89%, Table 2). Both cultivar and fungicide treatment had significant effects on index (Table 3). Mean index decreased with improved resistance level and treatment. MR_TR resulted in the greatest control of index (66%, Table 2).

North Dakota **(ENV 6).** Six hard red winter wheat cultivars were planted into non-host residue near Carrington, ND. This trial was inoculated with colonized corn spawn followed by supplemental misting. Index and DON observations ranged from 0.05 to 62% and 2.1 to 14.9 ppm, respectively. Yield data were not available for this environment.

Cultivar and fungicide factors each had significant effects on index and DON (Table 3). Within each resistance class, fungicide treatment increased control of both index and DON, with MR_TR resulting in approximately 95 and 55% control of index and DON, respectively (Table 2).

South Dakota. Three trials, with three small grain cultivars each, were planted near Volga, SD. **HRSW (ENV 12).** This trial was established in non-host residue and was not artificially inoculated. Index, DON and yield observations ranged from 0 to 21.9%; 0 to 0.8 ppm and 34.9 to 55 bu/A, respectively. Only cultivar had significant effects on index (Table 3) and only the MR cultivar had significantly less index than that of S or MS cultivars (data not shown). The MR_TR combination had the greatest control of index (59%) compared to the S_UT. **6-Row Barley (ENV 13).** This trial was also established in non-host residue and was not artificially inoculated. An MS cultivar was not included in this trial. Index, DON and yield observations ranged from 8.6 to 17.9%; 0 to 1.2 ppm and 32 to 69.2 bu/A, respectively. Neither cultivar nor treatment had significant effects on index in this trial (Table 3). In general, the MR cultivar only provided between 4-5% control of index compared to the S_UT (Table 2). **HRWW (ENV 14).** This trial was established in host residue and artificial inoculation with colonized corn spawn was included as a third factor. Index, DON and yield observations ranged from 0 to 50%; 0 to 1.1 ppm and 36.2 to 61.6 bu/A, respectively. While the S_UT had index <5% and DON <1 ppm, the overall trial means exceeded these values, which is why this trial was included in the analysis. Neither cultivar nor treatment had significant effects on index in this trial (Table 3). Inoculation also did not have a significant effect on index ($F = 0.16$, $P = 0.69$; data not shown). Interestingly, the S cultivar had the lowest levels of mean index and DON out of all resistance classes. This may be explained, in part, by differences in flowering date among the cultivars. Reportedly, rain events occurred during flowering of the MS and MR cultivars, but the S cultivar may have "escaped", reaching peak flowering during a dry period.

CONCLUSIONS

In most trials, the use of an MR cultivar reduced both index and DON, relative to the untreated, susceptible check. The effect of fungicide was slightly more variable across trials, potentially due to interactions between fungicide efficacy and environmental conditions. In general, fungicide application increased percent control of index and DON within each resistance category. However, there were some exceptions to this, observed within the MS cultivar this year. Most frequently, the combination of MR_TR resulted in the greatest level of control across trials. The degree of this control was dependent on each environment's unique cultivars and cropping system. Several trials incorporated artificial inoculations this year in an effort to provide more useable data (>5% index and >1 ppm DON in the S_UT); however, unless supplemental misting accompanied inoculations (as done in corn spawn-inoculated trials), the dry conditions prevalent in many small grain growing regions this year likely thwarted potential infections and disease development.

AKNOWLEDGEMENTAND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-071. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

LITERATURE CITED

Willyerd, K.T., C. Li, L.V. Madden, C.A. Bradley, G.C. Bergstrom, L.E. Sweets, M. McMullen, J.K. Ransom, A. Grybauskas, L. Osborne, S.N. Wegulo, D.E. Hershman, K. Wise, W.W. Bockus, D. Groth, R. Dill-Macky, E. Milus, P.D. Esker, K.D. Waxman, E.A. Adee, S E. Ebelhar, B.G. Young, and P.A. Paul. 2012. Efficacy and Stability of Integrating Fungicide and Cultivar Resistance to Manage Fusarium Head Blight and Deoxynivalenol in Wheat. Plant Dis. 96:957-967.

Table 1. Study descriptions, trial-wide mean index, DON and yield (across all treatments and reps) and mean index and DON for the susceptible, untreated (S_UT) check from 22 coordinated integrated management trials (ENV, environments) in 2012.

					Trial-wide means			S_UT Check	
ENV	LOCATION	CLASS	PREVIOUS CROP	INOC?	Index %	DON ppm	Yield bu/A	Index %	DON ppm
1	IN	SRWW	host	Y	0.00	.	62.40	0.00	.
2	Aurora, NY	SRWW	host	Y	0.00	0.06	93.08	0.00	0.06
3	Aurora, NY	SRWW	non-host	Y	0.00	0.02	125.83	0.00	0.04
4	Bradford, MO	SRWW	host	N	13.62	1.25	66.98	24.27	3.07
5	Bradford, MO	SRWW	non-host	N	16.71	2.85	75.05	32.40	9.45
6	Carrington, ND	HRWW	non-host	Y	8.61	7.17	.	23.85	10.00
7	Forman, ND	HRWW	host	N	0.00	0.00	75.72	0.00	0.00
8	Forman, ND	HRSW	non-host	N	0.00	0.00	63.89	0.00	0.00
9	Prosper, ND	HRWW	host	N	0.00	0.00	79.06	0.00	0.00
10	Finley, ND	HRSW	non-host	N	0.00	0.00	71.71	0.00	0.00
11	Finley, ND	6rowBARLEY	non-host	N	0.00	0.05	67.56	0.00	0.05
12	Volga, SD	HRSW	non-host	N	11.48	0.12	46.72	17.75	0.12
13	Volga, SD	6rowBARLEY	non-host	N	12.87	0.51	44.95	12.96	0.51
14	Volga, SD	HRWW	host	Y	6.40	0.09	50.64	3.31	0.09
15	Wooster, OH	SRWW	non-host	Y	2.34	0.11	93.45	4.24	0.21
16	Fargo, ND	2rowBARLEY	host	N	0.389	.	41.87	0.54	.
17	Fargo, ND	2rowBARLEY	non-host	N	0.184	.	17.95	0.18	.
18	Fargo, ND	6rowBARLEY	non-host	N	0.229	.	20.58	0.39	.
19	Fargo, ND	6rowBARLEY	host	N	0.159	.	49.32	0.24	.
20	Dixon Springs, IL	SRWW	host	Y	4.64	0.04	51.41	4.06	0.15
21	Monmouth, IL	SRWW	host	Y	17	0.02	88.46	39.11	0.08
22	Urbana, IL	SRWW	host	Y	11.96	0.06	74.81	34.48	0.18

Table 2. Mean FHB index and DON and percent control for each management combination, relative to the untreated, susceptible check (from trials (ENV) with >5% index and/or >1 ppm DON in the check).

		Resistance x Treatment Combination*						% Control Compared to S_UT				
Response	**ENV**	**S_UT**	**S_TR**	**MS_UT**	**MS_TR**	**MR_UT**	**MR_TR**	**S_TR**	**MS_UT**	**MS_TR**	**MR_UT**	**MR_TR**
Index (%)	4	24.27	19.47	14.32	12.65	11.33	10.49	19.79	41.01	47.88	53.33	56.76
	5	32.40	26.78	19.52	16.70	12.80	11.08	17.34	39.76	48.46	60.49	65.79
	6	23.85	5.54	9.14	2.73	9.40	1.01	76.78	61.68	88.57	60.59	95.75
	12	17.75	11.13	11.86	12.48	8.40	7.28	37.28	33.16	29.68	52.68	59.00
	13	12.96	13.29	.	.	12.35	12.40	-2.51	.	.	4.69	4.35
	14	3.31	3.50	5.83	11.25	10.42	4.08	-5.74	-76.23	-239.88	-214.70	-23.36
	21	39.11	27.19	11.25	1.25	19.35	11.95	30.48	71.23	96.80	50.53	69.44
	22	7.19	2.13	1.31	1.13	2.61	0.00	70.45	81.75	84.35	63.71	100.00
DON (ppm)	4	3.07	1.70	2.60	1.62	0.82	0.34	44.63	15.31	47.34	73.22	88.78
	5	9.45	4.43	5.10	3.27	1.32	0.49	53.09	46.03	65.43	86.07	94.78
	6	10.00	10.05	7.59	5.73	5.09	4.55	-0.50	24.13	42.75	49.13	54.50

*Resistance x treatment combinations included: susceptible, untreated check (S_UT); susceptible, treated (S_TR); moderately susceptible, untreated (MS_UT); moderately susceptible, treated (MS_TR); moderately resistant, untreated (MR_UT); moderately resistant, treated (MR_TR).

Table 3. Effects of cultivar, fungicide and their interactions on FHB index and DON for those coordinated management trials with greater than 5% index and/or 1 ppm DON in the check.

		Cultivar Resistance		Fungicide Treatment		Cultivar x Treatment Interaction	
Response	**ENV**	***F Statistic***	***P-value***	***F Statistic***	***P-value***	***F Statistic***	***P-value***
Index (%)	4	77.31	<0.01	6.90	0.01	2.54	0.09
	5	52.52	<0.01	4.91	0.03	0.64	0.53
	6	4.37	0.02	14.33	<0.01	1.60	0.21
	12	6.50	0.02	3.62	0.08	3.06	0.08
	13	0.87	0.36	0.05	0.82	0.03	0.86
	14	0.63	0.54	0.00	0.95	0.76	0.48
	21	3.99	0.10	3.08	0.08	0.08	0.93
	22	6.25	<0.01	9.07	<0.01	1.96	0.15
DON (ppm)	4	61.41	<0.01	31.37	<0.01	3.01	0.06
	5	60.24	<0.01	25.58	<0.01	6.42	<0.01
	6	19.22	<0.01	0.49	0.53	0.66	0.52

SESSION 2:

FOOD SAFETY AND TOXICOLOGY

Co-Chairpersons: Jim Pestka and
Paul Schwarz

IMPACT OF BRAN PROPERTIES ON *FUSARIUM* MYCOTOXIN LEVELS IN WINTER WHEAT (*TRITICUM AESTIVUM* L.) KERNELS

H.D.P. Damecharla[1], W.A. Berzonsky[1*], N.R. Reese[2] and P.G. Krishnan[3]

[1]Seed Technology Laboratory, Department of Plant Science, South Dakota State University; [2]Department of Biology and Microbiology, South Dakota State University; and [3]Crop Quality Lab, Department of Health and Nutritional Sciences, South Dakota State University
[*]Corresponding Author: PH: 605-688-5334, Email: william.berzonsky@sdstate.edu

ABSTRACT

Wheat bran is an important source of dietary fiber, and according to consumers, they prefer the appearance and taste of whole-wheat products made from white wheat compared to those made from red wheat. However, since the predominant FHB mycotoxin, deoxynivalenol (DON) accumulates primarily in the bran layer of kernels, the bran may be a significant source of DON, especially in white-wheat products made using white wheat. Moreover, differences in bran between white and red wheat kernels may impact the DON in such products, presenting a potential food contamination risk. Near-isogenic lines (NILs) of red and white winter wheat were developed for use in this study to examine potential bran differences and identify the impact of any genetic differences on the accumulation of DON in bran. In two greenhouse trials we artificially infected parents, NILs, and control varieties with *Fusarium graminearum* (teleomorph *Gibberella zeae*) to induce FHB, and samples were collected from both infected and uninfected plants. To compare accumulation of DON in the bran layer, the samples were pearl-milled to produce bran and non-bran fractions. DON accumulation in bran fractions was significantly higher than in non-bran fractions. Though the differences were not statistically significant, generally, DON accumulation was higher in the bran fractions of red kernel genotypes compared to accumulation in the bran fractions of white kernel genotypes. To determine the nature of these differences, we are also analyzing possible morphological and chemical bran differences between the white and red NILs. Using the NILs, we will measure bran layer thickness from kernel cross-sections and determine the impact of bran extractions on fungal growth in vitro. Data from these analyses will be compared and presented.

A BARLEY UDP-GLUCOSYLTRANSFERASE FORMING A NOVEL ZEARALENONE-GLUCOSIDE

M.P. Kovalsky Paris[1], W. Schweiger[1], S. Shin[2], G. Muehlbauer[2], C. Hametner[3], R. Krska[4], F. Berthiller[4] and G. Adam[1*]

[1]University of Natural Resources and Life Sciences, Vienna (BOKU), Department of Applied Genetics and Cell Biology, A-3430 Tulln, Austria; [2]University of Minnesota, Department of Agronomy and Plant Genetics, St Paul, MN 55108, USA; [3]Vienna University of Technology, Institute of Applied Synthetic Chemistry, A-1060 Vienna, Austria; and [4]BOKU, Department IFA-Tulln and Christian Doppler Laboratory for Mycotoxin Metabolism, A-3430 Tulln, Austria
[*]Corresponding Author: PH: (0043-1) 47654-6380, Email: gerhard.adam@boku.ac.at

ABSTRACT

During work aiming to identify deoxynivalenol (DON) inactivating UDP-glucosyltransferases, several barley genes were DON inducible, but did not confer DON resistance upon heterologous expression in yeast [1]. We found that one of the candidate genes, designated *HvZOG2*, encodes an enzyme capable of glycosylating the second prominent *Fusarium* mycotoxin, zearalenone. Besides the previously described [2] product zearalenone-4-O-glucoside (Z4G) also a second compound consistent with the mass of a glucoside was formed. The two products, Z4G and, ZON-2-O-glucoside were produced in about 1:1 ratio. According to a new nomenclature and numbering system for zearalenone derivatives [3] the novel product should be named and abbreviated ZEN-16-glucoside. Yeast expressing *HvZOG2* was grown in a fermenter and treated with zearalenone. The zearalenone-glucosides released into the medium were purified by solid phase extraction. The ZEN-16-Glc was eluted with 60% methanol and concentrated before further purification by preparative HPLC (Waters Sun Fire C18 column, 40-80% methanol gradient). The structure of the purified compound was confirmed by NMR. The occurrence of ZEN-16-Glc in crop plants is currently under investigation. We will also determine, whether ZEN-16-Glc, like the previously described ZON-4-Glc/ZEN-14-Glc, is easily hydrolyzed back to the parental Fusarium mycotoxin and should therefore be considered as an additional masked mycotoxin [4].

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based on work funded by the Austrian Science Fund (SFB F37), the Christian Doppler Society, the Minnesota small grains initiative and the USDA. This is a cooperative project with the US Wheat and Barley Scab Initiative. Any opinions, findings and conclusions or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the US Department of Agriculture.

REFERENCES

Schweiger et al. (2010) Validation of a candidate deoxynivalenol-inactivating UDP-glucosyltransferase from barley by heterologous expression in yeast. Mol Plant Microbe Interact 23: 977-86.

Poppenberger et al. (2006) Heterologous expression of *Arabidopsis* UDP-glucosyltransferases in Saccharomyces cerevisiae for production of zearalenone-4-O-glucoside. Appl Environ Microbiol 72: 4404-10.

Metzler (2010) Proposal for a uniform designation of zearalenone and its metabolites. Mycotoxin Res 27: 1–3.

Berthiller et al. (2012) Masked mycotoxins: A review. Mol Nutr Food Res [in press].

DON – PAST, PRESENT AND FUTURE

David Miller*

Carleton University, Department of Chemistry, Ottawa, ON K1S 5B6, Canada
*Corresponding Author: PH: 613-520-2600 ext 1053, Email: david_miller@carleton.ca

ABSTRACT

Deoxynivalenol (DON) was discovered by the Japanese from grain that caused 'red mold disease'. However, Fusarium head blight was long recognized in Europe as a substantial problem in both the old and new world. Spanish cultivars of wheat were imported to the new world (by Columbus) followed by English and French cultivars by fisherman and emigrants in the 16th century. What we know as FHB was reported in the 17th century in New England and Quebec & Acadia (New Brunswick). Quebec priests wrote that the grain was in some way toxic. Ergot epidemics were by then known to be caused by a fungus. Modern awareness that that FHB resulted in grain that was toxic began in the 1920s during a series of bad epidemics. American wheat exported to Germany was reported to cause emesis in swine by German researchers in 1929. Reporting in a 1926 Phytopathology, Russian workers discussed scabby wheat producing "inebriant bread" associated with the now recognizable symptoms of acute DON toxicosis. Large FHB screening programs began in the US during that time. Many of these older cultivars have reasonable tolerance to FHB but have poor yield by today's standards.

When FHB devastated farmers around the Great Lakes in the 1980s and later in the mid-1990s, much research was again stimulated. Programs to understand agronomic factors and the nature of the resistance have produced thousands of papers in the past 30 years. None-the-less, after a century of research, cultivars that have adequate resistance to FHB and other diseases and have appropriate quality characteristics remain elusive. We remain in a period where it is necessary to manage the problem to ensure that there is an adequate supply of soft and hard wheat in the affected areas. The strategies to do this involve resistance screening programs, forecasting systems to ensure appropriate use of fungicides, and regulations.

As witnessed by the differing views on appropriate regulation of aflatoxin and ochratoxin A, over-regulation affects all partners in the chain from farm to fork. A determination of reasonable certain of no harm of a particular toxin requires a sophisticated understanding of the mode of action of the compound. Increased understanding allows the uncertainties to be better defined. In 2004, an ILSI-EU workshop on DON identified a number of priorities for action. One of these was that the mechanism of action of the mechanism of food refusal in non-human primates. Although these data have so far not indicated that the PMTDI is insecure, the widespread occurrence of the DON-glycoside in European cultivars forces a re-examination of the question.

QUALITY ASSURANCE ISSUES IN DON TESTING

Paul Schwarz*

Department of Plant Sciences, North Dakota State University, Fargo, ND
*Corresponding Author: PH: 701-799-4419, Email: paul.schwarz@ndsu.edu

ABSTRACT

Mycotoxin testing is an essential component to the USWBSI mission of developing effective control measures that minimize the threat of Fusarium head blight (scab), including the reduction of mycotoxins, to the producers, processors, and consumers of wheat and barley. The four diagnostic labs that are supported through USWBSI funding analyze in excess of 50,000 samples of wheat and barley each year, and support all research areas of the Initiative. As such, an understanding of factors that impact accuracy and precession of deoxynivalenol (DON) data, as well as laboratory throughput, are very important. The intent of this presentation is to review the basics of DON testing methodology, sources of error, and then the limitations on throughput. Discussion of check samples, limits of detection (LOD), limits of quantitation (LOQ), and DON recovery will address the interpretation of data, in terms of quality assurance. Sampling issues, which were previously discussed as the major source of error, will be reiterated.

SESSION 3:

VARIETY DEVELOPMENT AND HOST PLANT RESISTANCE

Co-Chairpersons: Fred Kolb and
Jochum Wiersma

OVERVIEW OF BREEDING FOR FHB RESISTANCE IN WHEAT – WHERE WE'VE COME FROM AND WHERE WE ARE

James A. Anderson*

University of Minnesota, Department of Agronomy & Plant Genetics, St. Paul, MN
*Corresponding Author: PH: (612) 625-9763, Email: ander319@umn.edu

ABSTRACT

The cultivation of wheat varieties resistant to Fusarium head blight (FHB) is one of the most important components to diminish losses due to this disease. Although there is no known immunity to this disease in wheat germplasm, considerable improvements in genetic resistance have been achieved in the past 20 years by concentrated breeding efforts that have relied primarily upon repeated field and greenhouse-based screening. Dedicated breeding efforts for resistance to FHB in the Upper Midwest spring wheat region date to the late 1980's when 'Sumai 3' was first introduced as a source of resistance. Since consecutive epidemics beginning in 1993, breeding for FHB has been a top priority for programs in the region. At the time of those epidemics, most cultivars available were susceptible, with only 'Pioneer 2375' rated as having an intermediate (MR-MS) level of resistance. 'Alsen', released by NDSU in 2000, was the first moderately resistant cultivar to be widely grown in the region. Today, half of the cultivars available to growers in the spring wheat region are classified as moderately resistant or better for FHB reaction and were grown on 43% of the region's spring wheat acreage in 2011. Despite these genetic gains and improved fungicides, even the most resistant materials available today can incur damage when environmental conditions are conducive for an epidemic. DNA markers have been identified for many QTL using biparental mapping populations and a few are being routinely used in marker-assisted selection (MAS). FHB resistance is a quantitative trait, and the best QTLs are able to reduce damage by 20-30%, but most QTL have far smaller effects. The *Fhb1* QTL was present in cultivars grown on 40% of the region's wheat acreage in 2011. We routinely screen with DNA markers all of our pre-yield trial F_5 lines, about 1,000 total, as well as BC_1 and TC_1 plants segregating for *Fhb1* and the 5AS QTL. Enriched populations undergo phenotypic selection for FHB resistance, and other yield, disease resistance, and end-use quality testing necessary to produce FHB resistant germplasm and variety candidates. Substantial efforts in phenotypic assessments for FHB resistance will still be necessary, even with an increase in MAS for this trait, because there are likely to be numerous genes with minor effects, which need to be combined with the major QTLs in order to obtain the desired level of resistance. We are initiating genomic selection with a primary goal of identifying and discarding susceptible lines prior to entry into yield trials, thus eliminating a major bottleneck in our breeding program.

ACKNOWLEDGEMENTS AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement Nos. 59-0790-4-091, 59-0206-9-052, and 59-0206-9-066. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author and do not necessarily reflect the view of the U.S. Department of Agriculture.

FINE MAPPING OF WHEAT *FHB1* USING SEQUENOM MASSARRAY SNP GENOTYPING PLATFORM

A.N. Bernardo[1], S. Chao[2] and G-H. Bai[3*]

[1]Dept. of Plant Pathology, Kansas State University, Manhattan, KS; [2]USDA-ARS, Cereal Crops Research Unit, Fargo, ND; and [3]USDA-ARS, Hard Winter Wheat Genetics Research Unit, Manhattan, KS
[*]Corresponding Author: PH: (785) 532-1124, Email: guihua.bai@ars.usda.gov

ABSTRACT

Reduction in grain yield and quality due to Fusarium head blight (FHB) is a serious problem in bread wheat (*Triticum aestivum* L.), and the *Fhb1* QTL in chromosome 3BS provides a major source of FHB resistance. *Fhb1* has been linked to restriction fragment length polymorphisms, simple sequence repeats (SSR), amplified fragment length polymorphisms and sequence tagged site (STS) markers. In an effort to fine map the *Fhb1* QTL region using high-throughput genotyping platform, we looked at single nucleotide polymorphisms (SNPs) to saturate the region with these biallelic markers. SNPs are the most abundant form of polymorphism and are ideal for fine mapping. Fifty-five SNPs were identified between two near-isogenic lines (NILs) that were genotyped using the Illumina Infinium 9K SNP array. The NILs were derived from continuous backcross of Clark (recurrent FHB-susceptible parent) to Ning7840 (*Fhb1* donor). Two sets of multiplex SNPs representing the 55 SNPs were analyzed in 91 Ning7840/Clark BC_7F_5 lines that showed recombination in the QTL region using the Sequenom MassArray. A total of 45 SNPs showed segregation in the population and were analyzed together with known SSR, STS and SNP markers that were closely linked to *Fhb1*. Six new SNP markers were mapped between *Xgwm533* and *Xgwm493*, SSR markers flanking the *Fhb1* in 3BS. These SNPs should be useful for high resolution mapping of *Fhb1* and high-throughput marker-assisted selection in combination with other markers using Sequenom MassArray.

ACKNOWLEDGEMENTS AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

GRAIN YIELD PERFORMANCE OF WESLEY BACKCROSS *FHB1* HRWW GERMPLASM

W.A. Berzonsky[1*], A. Bakhsh[2], P.S. Baenziger[3] and G. Bai[4]

[1]Dept. of Plant Science, South Dakota State Univ., Brookings, SD; [2]Dept. of Plant Breeding & Genetics, College of Agric. Dera Ghazi Khan, Pakistan; [3]Dept. of Agronomy & Horticulture, Univ. of Nebraska, Lincoln, NE; and [4]Hard Winter Wheat Genetic Research Unit, USDA-ARS, Manhattan, KS
[*]Corresponding Author: PH: (605) 688-5334, Email: William.berzonsky@sdstate.edu

ABSTRACT

Transferring *Fhb1*, a widely utilized and effective QTL for resistance to Fusarium head blight (FHB) from spring to winter wheat (*Triticum aestivum* L.) is important for producing more resistant winter wheat varieties. However, to release resistant winter wheat varieties with the broadest appeal to growers, the transfer and expression of resistance to FHB should also be accomplished without a significant reduction in grain yield performance. Our objective was to assess the impact of *Fhb1* on grain yield performance when the QTL was transferred to Wesley, an adapted and widely grown variety in the Northern Hard Winter Wheat (HWW) region of the US. Compared with Wesley and two other regionally grown HWW varieties, Lyman and Overland, we evaluated the grain yield performance of 20 Wesley backcross (BC) breeding lines, each of which was verified as being homozygous for *Fhb1*. The evaluated lines were produced by initially hybridizing spring wheat breeding line ND2928 as the parental source of *Fhb1* to Wesley, followed by two backcrosses to Wesley. Lines homozygous for *Fhb1* were identified and selected from among BC progeny using molecular markers STS256, Gwm533, Gwm493, and Umn10, which were applied at the various cross and BC generations. Utilizing replicated field trials; yearly grain yield evaluations were conducted from 2010 to 2012 at Mead and Lincoln, NE and at Brookings and Dakota Lakes, SD. When averaged over all years and locations, the highest yielding BC line performed at 94% of the grain yield of Wesley and at 83% of the grain yield of Overland, the highest yielding variety in the trials. Only at the Brookings location did at least one BC line exhibit a higher grain yield than Wesley in each year of the trial, and compared with 2012, approximately twice as many BC lines exhibited higher grain yield than Wesley in 2010 and 2011. Lines evaluated in 2010, 2011, and 2012 were at the BC_2F_5, BC_2F_6, and BC_2F_7 generation, respectively. In addition to the impact of seasonal and location effects on the expression of grain yield, segregation for background genes may have impacted the grain yield performance of the BC lines over years. Marker selection was applied only during the derivation of the BC lines to identify lines homozygous for *Fhb1,* and no marker background selection for the recurrent parent or within-line selection was practiced for grain yield thereafter. Consequently, background segregation for genes influencing grain yield may have been responsible for some of the differences observed between BC lines over years. Irrespective of grain yield differences due to environment, these results emphasize the importance of combining selection for grain yield performance with selection for the presence of *Fhb1* when transferring the QTL from spring wheat breeding lines to winter wheat varieties. To transfer *Fhb1* from spring wheat lines and develop high-yielding winter wheat germplasm lines, this tandem selection approach is likely to be even more important when relatively few backcrosses are made to the winter wheat parent.

ADOPTION OF WHEAT CULTIVAR EVEREST SIGNIFICANTLY LOWERED THE KANSAS STATEWIDE FUSARIUM HEAD BLIGHT PHENOTYPE

W.W. Bockus[1*], M.A. Davis[1], E.D. De Wolf[1] and A.K. Fritz[2]

[1]Department of Plant Pathology, and [2]Department of Agronomy, Kansas State University, Manhattan, KS
[*]Corresponding Author: PH: (785) 532-1378, Email: Bockus@ksu.edu

ABSTRACT

Fusarium head blight (FHB) is a serious disease of small grains such as wheat. Significant losses can occur due to the blighting of many heads in the field. One of the best ways to manage FHB is by planting cultivars with some level of resistance. As a result of funding from the U.S. Wheat and Barley Scab Initiative, significant effort has been placed on developing cultivars adapted to Kansas with improved levels of FHB resistance. Because of this effort, the moderately-resistant cultivar Everest was released in 2009 and has gained popularity such that it now is the second most planted cultivar in Kansas and the most popular cultivar in the eastern third of the state where FHB traditionally occurs. The goal of this project was to quantify the impact that the adoption of Everest has had on the average statewide FHB phenotype and the average phenotype for the eastern third of the state. From 2003 through 2012, data were obtained from the USDA National Agricultural Statistics Service, Kansas Field Office relative to the percentage acreage planted to various wheat cultivars in Kansas. In addition, each cultivar's reaction to FHB was obtained from De Wolf et al. (2012. Wheat Variety Disease and Insect Ratings 2012. Kansas Coop. Extension Service publication MF-991. 4 pp). The above data were used to calculate a score for an average cultivar for the entire state and the eastern third of the state. The decimal equivalent of the percentage acreage planted to a cultivar in a given year was multiplied by its FHB score (1-9 scale where 1=resistant and 9=susceptible). Acreage planted to "blends" (average of 11.3% of the acres), unknown cultivars, or to cultivars where the FHB phenotype was not known was ignored. Nevertheless, the average acreage used each year was 80.5% statewide and 78.9% for the eastern third. Calculated values for all planted cultivars in a given year were added together and divided by the decimal of the total acreage for all known cultivars to obtain an average FHB phenotype. Prior to 2011, the average statewide phenotype was above 7 indicating high susceptibility. Beginning in 2009, and becoming more pronounced in 2012, there has been a noticeable decline in the statewide FHB phenotype. This has been especially evident in the eastern third of the state where the average phenotype has gone from 7.74 in 2008 to 5.73 in 2012. Although other cultivars have been recently released with improved levels of resistance (e.g. Art and Hitch), Everest has contributed the most to the observed recent improvement in FHB phenotypes. It now occupies over 23% of the acres in the eastern third of the state where FHB is especially important. Gower adoption of Everest has significantly lowered the vulnerability of the Kansas wheat crop to FHB.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under agreement No. 59-0206-1-110. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

GENOTYPE BY SEQUENCING AND MARKER ASSISTED SELECTION: BREAKING THE BOTTLENECK

Robert Brueggeman[1*], Tim Friesen[2] and Jared LeBoldus[1]

[1]Department of Plant Pathology, North Dakota State University, Fargo, ND; and [2]USDA-ARS, Cereal Crops Research Unit, Northern Crop Science Laboratory, Fargo, ND 58105
[*]Corresponding Author: PH: 701-231-7078, Email: Robert.Brueggeman@ndsu.edu

ABSTRACT

Genotyping is the major resource bottleneck when developing populations to characterize disease resistance in barley or utilizing genomic selection. Until recently the Illumina Bead Express was the cheapest technology available for barley genotyping, however the cost is still prohibitive. To identify novel FHB resistance QTL in barley and the markers associated with these loci, it is essential to develop bi-parental populations and have the technology and resources available for robust genotyping and phenotyping. The Ion Torrent next generation sequencing platform presents the opportunity to overcome the resource bottleneck by generating up to ten million 400 base sequences (40 Gb of sequence) per a single run. To evaluate the potential of this technology we developed 50 barcoded adaptors with optimized restriction enzymes to reduce the complexity of the barley genome (~5,000 Mb) down to 18,243 unique sequence sites. Similarly we were able to reduce the complexity of the fungal pathogen, *Pyrenophora teres* f. *teres* (~40 Mb genome) to 14,143 unique sequence sites. Utilizing the barcoded genomic libraries, we sequenced 40 individuals from a bi-parental population of *P. teres* f. *teres* in a single Ion Torrent run. Using this data we identified >1,000 single nucleotide polymorphisms (SNPs) representing unique loci. We expect that by reducing the barley genome complexity down to 18,243 loci, we will be capable of running genotype by sequencing (GBS) on barley populations for ~$16/ line and generate ~1,000 SNP markers. The ability to genotype barley populations will allow us to characterize novel resistance sources and rapidly develop markers that can be utilized in marker-assisted selection strategies, including genomic selection. The GBS method is also being utilized to characterize virulence genes in fungal pathogens through analysis of bi-parental populations and association mapping in natural populations where crossing is not an option.

COMPARISON OF RESISTANCE OF MODERN WHEAT CUTIVARS TO THE DON PRODUCER *FUSARIUM GRAMINEARUM* AND THE T2/HT2 PRODUCER *FUSARIUM SPOROTRICHIOIDES*

H. Buerstmayr[1*], M. Lemmens[1], L. Duchalais[2], D. Hourcade[3], L. Guerreiro[3], V. Laurent[4] and O. Robert[4]

[1]BOKU-University of Natural Resources and Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, Konrad Lorenz Str. 20, A-3430 Tulln, Austria; [2]RAGT, 60 rue Léon Beauchamp, BP45, 59930 La Chapelle d'Armentières, France; [3]ARVALIS – Institut du végétal, 3 rue Joseph et Marie Hackin, 75116 Paris; [4]Bioplante – Florimond Desprez, BP41, 59242 Cappelle en Pévèle
[*]Corresponding Author: PH: 43-2272-66280-201, Email: hermann.buerstmayr@boku.ac.at

ABSTRACT

While resistance of wheat to deoxynivalenol (DON) producing Fusarium species (*e.g. F. graminearum, F. culmorum*) has been well studied, the resistance response to T2/HT2 producers (e.g. *F. sporotrichioides*) is much less investigated. Likewise, while the role of DON in the pathogenesis process has been confirmed, a potential role of T2/HT2 as aggressiveness factors is currently unknown. We therefore performed artificial inoculation trials at three locations (two in France, one in Austria) during two seasons with 48 (2011) or 96 (2012, trials in progress) wheat lines or cultivars using *F. graminearum, F. sporotrichioides* or a mix of both species. The wheat lines comprised mainly current cultivars from France and Austria and several experimental lines. We scored Fusarium head blight (FHB) visual symptoms and other morphological traits such as plant height and anther extrusion.

Based on first year results we found that 1) there was a large genetic variation in FHB resistance among current cultivars, ranging from moderately resistant to highly susceptible. 2) Experimental lines, which were selected for high FHB resistance have been confirmed. 3) Resistance to both investigated Fusarium species was highly correlated ($r>0.9$), indicating a common mechanism of resistance against DON and T2/HT2 producers. 4) The extent of anther extrusion was negatively correlated with FHB severity ($r=-0.76$). Anther extrusion may thus be a useful trait for indirect selection. Trials to re-evaluate these findings are underway.

ACKNOWLEDGEMENT

We acknowledge funding of this work by FSOV (Fonds de Soutien à l'obtention Végétale, France).

ADVANCED BACK-CROSS QTL MAPPING OF RESISTANCE TO FUSARIUM HEAD BLIGHT DERIVED FROM *TRITICUM MACHA* (GEORGIAN SPELT WHEAT)

M. Buerstmayr*, M. Lemmens, B. Steiner and H. Buerstmayr

BOKU-University of Natural Resources and Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, Konrad Lorenz Str. 20, A-3430 Tulln, Austria
*Corresponding Author: PH: 43-2272-66280-201, E-mail: maria.buerstmayr@boku.ac.at

ABSTRACT

While many reports on genetic analysis of *Fusarium* resistance in bread wheat have been published, only limited information is available on Fusarium head blight (FHB) resistance derived from wheat relatives. We report about genetic analysis of FHB resistance derived from *Triticum macha* (Georgian spelt wheat). In order to introduce valuable alleles from the landrace *T. macha* into a modern genetic background we used an advanced back-cross QTL mapping scheme (Tanksley and Nelson 1996). A back-cross-two derived recombinant inbred line population of over 300 BC_2F_3 lines was developed from a cross of *T. macha* with the Austrian winter wheat cultivar Furore. The population was evaluated for Fusarium resistance in six field experiments. The population was genetically fingerprinted with > 600 markers. The obtained linkage map covered 37 linkage groups with 563 markers. Five novel FHB resistance QTL, all descending from *T. macha*, were found on four chromosomes (2A, 2B, 5A, 5B). The largest effect QTL overlapped with the *Q-locus* (spelt type) on chromosome 5A and appears therefore an interesting QTL especially for spelt wheat improvement. For details see Buerstmayr et al. (2011).

ACKNOWLEDGEMENTS

We acknowledge funding by the FWF (Austrian Science Fund), project number: 17310-B05. We acknowledge Clare Nelson (Kansas State University, USA) for helping us with CarthaGène.

REFERENCES

Buerstmayr M, Lemmens M, Steiner B, Buerstmayr H (2011) Advanced backcross QTL mapping of resistance to Fusarium head blight and plant morphological traits in a *Triticum macha* × *T. aestivum* population. Theor Appl Genet 123:293-306

Tanksley SD, Nelson JC (1996) Advanced backcross QTL analysis: a method for the simultaneous discovery and transfer of valuable QTLs from unadapted germplasm into elite breeding lines. Theor Appl Genet 92:191-203

ASSOCIATION OF THE SEMI-DWARFING ALLELES *RHT-B1A/B* OR *RHT-D1A/B* WITH FUSARIUM HEAD BLIGHT RESPONSE IN A WINTER WHEAT DOUBLED HAPLOID POPULATION AND NEAR ISOGENIC LINES

M. Buerstmayr and H. Buerstmayr*

BOKU-University of Natural Resources and Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, Konrad Lorenz Str. 20, A-3430 Tulln, Austria
*Corresponding Author: PH: 43-2272-66280-201, E-mail: hermann.buerstmayr@boku.ac.at

ABSTRACT

Recent publications have shown that the widely used dwarfing genes *Rht-B1* (syn. *Rht1*) and *Rht-D1* (syn. *Rht2*) are associated with Fusarium head blight (FHB) resistance. The semi-dwarf allele *Rht-D1b* and to a lesser extent *Rht-B1b* appear to increase FHB susceptibility in wheat (Miedaner and Voss 2008, Holzapfel et al. 2008, Srinivasachary et al. 2009). In order to further evaluate the effects of these alleles we 1) developed and tested back-cross derived sister lines differing in their *Rht* alleles in a highly FHB resistant recipient line and 2) evaluated one doubled haploid population segregating at both loci.

On average across seven NIL-pairs for *Rht-B1* we found that lines with the semi-dwarf allele *Rht-B1b* showed about 90% increased FHB severity compared to their sister lines, which had the tall allele *Rht-B1a*. The difference was even more pronounced for *Rht-D1*, where on average across six NIL-pairs lines with the semi-dwarf allele *Rht-D1b* had about 160% higher FHB severity compared to lines with the *Rht-D1a* allele. Similarly in the DH population *Rht-D1b* lines were significantly higher diseased than *Rht-B1b* lines. Our data are in agreement with previous findings that semi-dwarfing alleles reduce FHB resistance and that *Rht-B1b* is less damaging than *Rht-D1b*. However, the negative effect of the semi-dwarf alleles can be balanced by selecting lines with other known or unknown FHB resistance QTL in their genome. Therefore, selection of semi-dwarf cultivars with good FHB resistance is quite difficult but feasible, and *Rht-D1b* should be avoided if high FHB resistance is desired.

ACKNOWLEDGEMENTS

We thank Jutta Foerster (Saaten Union Research Laboratory, Hovedissen, DE) for the *Rht* allele analysis within the 'Short-Wheat' project (ERA-Net EUROTRANS-BIO-1) and EUROTRANSBIO for financial support.

REFERENCES

Miedaner T, Voss HH (2008) Effect of dwarfing *Rht* genes on Fusarium head blight resistance in two sets of near-isogenic lines of wheat and check cultivars. Crop Sci 48:2115-2122.

Holzapfel J, Voss HH, Miedaner T, Korzun V, Häberle J, Schweizer G, Mohler V, Zimmermann G, Hartl L (2008) Inheritance of resistance to Fusarium head blight in three European winter wheat populations. Theor Appl Genet 117:1119-1128.

Srinivasachary, Gosman N, Steed A, Hollins TW, Bayles R, Jennings P, Nicholson P (2009) Semi-dwarfing *Rht-B1* and *Rht-D1* loci of wheat differ significantly in their influence on resistance to Fusarium head blight. Theor Appl Genet 118:695-702.

MOLECULAR MAPPING OF RESISTANCE TO FUSARIUM HEAD BLIGHT IN TETRAPLOID WHEAT

Maria Buerstmayr[1], Abdallah Alimari[1,2], Karin Huber[1,3], Marc Lemmens[1], Barbara Steiner[1] and Hermann Buerstmayr[1*]

[1]BOKU-University of Natural Resources and Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, Konrad Lorenz Str. 20, A-3430 Tulln, Austria; [2]Current Address: Department of Plant Biotechnology, National Agricultural Research Centre (NARC), Palestine; and [3]Current Address: Agrarisches Informationszentrum, Schauflergasse 6, A-1014 Vienna
[*]Corresponding Author: PH: 43-2272-66280-201, Email: hermann.buerstmayr@boku.ac.at

ABSTRACT

While many reports on genetic analysis of *Fusarium* resistance in bread wheat have been published, only limited information is available on Fusarium head blight (FHB) resistance derived from tetraploid wheats. In this contribution we report about genetic analysis of FHB resistance derived from two tetraploid *Triticum* sources: 1) *Triticum dicoccum* (cultivated emmer) and 2) *Triticum dicoccoides* (wild emmer). Back-cross-one derived recombinant inbred line populations were developed from crosses of the resistance donors with adapted Austrian durum wheat cultivars. The populations were evaluated for *Fusarium* resistance in well replicated experiments with artificial inoculation. The *T. dicoccum* derived populations were tested in field trials using spray inoculations and the *T. dicoccoides* derived mapping population was greenhouse tested using single-floret inoculations. The same lines were genetically analysed using SSR and AFLP markers. Map construction based on the back-cross derived RIL populations was done with *CarthaGène* (De Givry et al. 2005) and QTL mapping in *Qgene* (Nelson 1997).

In *T. dicoccum* the most consistent QTL effect mapped to chromosome 4B, and overlapped with the *Rht-B1a* allele. Further QTL mapped to 3B, 6A, 6B and 7B.

QTL for type 2 FHB resistance were detected in wild emmer (*T. dicoccoides*) mapping to chromosomes 3A and 6B. Wild and cultivated emmer wheat are promising sources for improving FHB resistance in durum wheat.

ACKNOWLEDGEMENTS

We acknowledge funding of this work by FWF (Austrian Science Fund), project number: 17310-B05; Abdallah Alimari was supported by a North-South Dialogue grant from the Austrian Academic Exchange Service (OeAD). We sincerely thank Clare Nelson (Kansas State Univ.) for his support in mapping with CarthaGène. We acknowledge Jeannie Gilbert (AG Canada, Winnipeg) for supplying the *T. dicoccum* line and Tzion Fahima and Tami Krugman (Univ. Haifa) for supplying the *T. dicoccoides* line. We also acknowledge Peter Jack and Christopher James (RAGT, Cambridge, UK) for SSR genotyping of the *T. dicoccum* populations.

REFERENCES

De Givry S, Bouchez M, Chabrier P, Milan D, Schiex T (2005) Multipopulation integrated genetic and radiated hybrid mapping. Bioinformatics 21:1703-1704.

Nelson JC (1997) QGENE: Software for marker-based genomic analysis and breeding. Mol Breeding 3:239–245.

META-ANALYSIS OF FUSARIUM HEAD BLIGHT RESISTANCE QTL IN CHINESE WHEAT LANDRACES

Jin Cai[1], Guihua Bai[1*] and Xiaofei Wang[2]

[1]USDA-ARS, Small Grain Genotyping Laboratory, Manhattan KS; and
[2]Department of Agronomy, Kansas State University, Manhattan KS
[*]Corresponding Author: PH: (785) 532-1124, Email: gbai@ksu.edu

ABSTRACT

Fusarium head blight (FHB) is one of the most devastating diseases in wheat. FHB not only causes significant losses in grain yield and quality, but also produces mycotoxins such as deoxynivalenol that are toxic to humans and animals. Growing resistant cultivars is one of the most effective strategies to minimize the disease damage. FHB resistance has been reported from many sources, but Chinese sources, especially Chinese landraces, such as Sumai3 and Wangshuibai, show the best resistance. Among them, Quantitative trait loci (QTL) for FHB resistance in Sumai3 have been well characterized in multiple studies, however, QTL in many other Chinese landraces are poorly characterized. Meta-analysis, a statistic method to combine QTL mapping results across independent studies, has been widely applied in human genetics research. In this study, five populations were developed from different Chinese wheat landraces (Haiyanzhong (HYZ), Wangshuibai (WSB), Baishanyuehuang (BSYH), Huangfangzhu (HFZ) and Huangcandou (HCD)). QTL have been identified in each population, however, only a selected set of markers were mapped in each population and some QTL may not be mapped due to poor coverage of markers in that population. Adding new markers to all possible QTL regions of all the five populations may recover the missing QTL. In total, after adding new markers, 12 QTL were remapped and 3 additional QTL mapped on 6 chromosomes (3A, 3BS, 3DL,5AS, 6BS, 7DL) in this study. Four QTL were identified on the consensus maps with one QTL each on chromosomes 3D and 3A, and two on chromosome 3BS. The QTL 95% confidence intervals were shortened by using a new clustering approach based on a Gaussion mixture model in MetaQTL V1.0. Thus, meta-analysis using the new maps will facilitate validation of QTL and identification of closely linked markers for marker-assisted selection.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

CHARACTERIZATION OF WHEAT MUTANTS WITH REDUCED FUSARIUM HEAD BLIGHT SYMPTOMS

Anthony Clark* and David Van Sanford

University of Kentucky, Dept. of Plant and Soil Sciences, Lexington, KY 40546
*Corresponding Author: PH: (859) 218-0809, E-mail: anthony.clark@uky.edu

ABSTRACT

M2-derived lines with reduced Fusarium head blight symptoms have been obtained from two gamma-irradiated susceptible soft red winter wheat lines, KY93C-1238-17-1 and KY96C-0786-3-2. In 2011 and 2012 these were tested in 2 rep RCB designs in inoculated irrigated nurseries in Lexington, KY. In 2011 29 KY93C-1238-17-1 and 23 KY96C-0786-3-2 mutants were tested. In 2012 the number of mutants was reduced to 14 KY93C-1238-17-1 and 7 KY96C-0786-3-2 lines. In 2011 disease ratings (0-9), %FDK, heading date and height were collected. Additionally, in 2012 DON, incidence, severity and index were recorded. Between years there were significant overall differences ($P<0.05$) for heading date (20 days earlier in 2012), rating (1.3 in 2012 versus 1.8 in 2011) and FDK (7.6% in 2012 versus 15.1% in 2011). Nonetheless, the reduced ratings and %FDK that were seen for the mutant lines in 2011 were repeated in 2012. In 2012 we observed significant differences ($P<0.05$) among KY93-1238-17-1-derived lines for: disease rating (parental mean 2.0, mutant mean 0.6 (range 0-2.5)); severity (parental mean 31%, mutant mean 18.2% (range 8.0-35.2%) and %FDK (parental mean 10.6%, mutant mean 5.75% (range 2.9-11%)). Mean DON for KY93C-1238-17-1 was 12.6 ppm, the mutant average was 8.5 ppm (range 5.45-12.7ppm). Mean DON for Truman from neighboring tests was 8.9 ppm. Among KY96C-0786-3-2 mutants only index was significantly different at $P<0.05$ (parental mean 25.7%, mutant mean 17.9% (range 8.8-28.9%)). Reduced means for KY96C-0786-3-2 mutants were seen: DON (parental mean 22.2 ppm, mutant mean 16.7 ppm (range 12.7-20.1 ppm); FDK (parental mean 14.4%, mutant mean 9.89% (range 5.76-13.67%); disease rating (parental mean 3.5, mutant mean 2.14 (range 1-3)) and severity (parental mean 36.8%, mutant mean 31.2% (range 19.7-40.9%)).

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-054. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

MAPPING RESISTANCE TO FUSARIUM HEAD BLIGHT IN A DOUBLED HAPLOID WHEAT POPULATION FROM THE CROSS MD01W233-06-1/SS8641

Benjamin Conway[1], J. Paul Murphy[2], Gina Brown-Guedira[3], Yanhong Dong[4], Shiaoman Chao[5], Carl Griffey[6] and Jose Costa[1*]

[1]PSLA Dept., University of Maryland College Park, MD 20742-4452; [2]Dept. of Crop Science, North Carolina State University, Raleigh, NC, 27695-7620; [3]USDA-ARS, Plant Science Research Unit, Raleigh, NC 27695; [4]Plant Pathology Dept., University of Minnesota, St. Paul, MN 55108, [5]USDA-ARS, BRL, Fargo, ND; and [6]Dept. of Crop and Soil Environmental Sciences, Virginia Tech, Blacksburg, Virginia 24061
[*]Corresponding Author: PH: 301-405-1317, Email: costaj@umd.edu

ABSTRACT

Fusarium head blight (*Fusarium graminearum*) is one of the urgent fungal pathogen threats to global agriculture. It is capable of causing significant yield losses, and concomitantly lowers test weight and causes the accumulation of the mycotoxin deoxynivalenol (DON), lowering farmer incomes and potentially rendering grain unfit for human and animal consumption. Fungicides can be applied to control these fungal diseases, but there are few effective fungicides to control scab in epidemic conditions, and applying fungicides can be difficult to time correctly and can have unintended consequences for environmental and human health. Breeding for disease resistance reduces the need for fungicide application, controlling disease without the harmful side effects. The soft red winter wheat line MD01W233-06-1 is moderately resistant to FHB, without previously described sources of resistance. It was crossed with the FHB susceptible line SS8641. From this cross a doubled haploid (DH) mapping population of 124 lines was created via the maize wide cross method and evaluated in 2011 and 2012 across four field locations for FHB resistance. Phenotypic data for mapping was collected by visual estimation for scab incidence (INC) and scab severity (SEV) in the field, and seed samples were analyzed post harvest for percent *Fusarium* damaged kernel (FDK) and assayed to determine DON content. The DHs were screened with one morphological marker (red coleoptile) and 29 SSR markers and assayed on a recently developed Illumina Infinium assay with 9000 SNPs. Linkage analysis was performed with genetic markers that were polymorphic and did not show segregation distortion. The map consisted of a morphological marker, 24 polymorphic SSRs, and 1786 polymorphic SNPs. QTL IciMapping v. 3.1 was used to construct the linkage map and perform QTL analysis. Markers with previously mapped locations were anchored to chromosomes and linkage analysis produced 25 linkage groups. QTL analysis revealed 31 QTLs with LOD scores ranging from 8.0 to 3.0 on 8 chromosomes. Most QTLs were linked to markers on chromosomes 3B, 2D, and 7B. Chromosome 3B showed consistent QTLs for INC, SEV, DON, and FDK that appeared in multiple location/years. Chromosome 2D had consistent QTLs for INC, DON, and FDK across multiple location/years. Chromosome 7B had consistent QTLs for SEV and FDK across multiple location/years.

GENERALIZED LINEAR MODELS FOR GENETIC PREDICTION OF SCAB RESISTANCE FROM REGIONAL DISEASE NURSERIES

J.T. Eckard[1], J.L. Gonzalez-Hernandez[1*], K.D. Glover[1], W.A. Berzonsky[1], J.A. Anderson[2] and M. Mergoum[3]

[1]South Dakota State University, Department of Plant Science, Brookings, SD; [2]University of Minnesota, Dept. of Agronomy and Plant Genetics, Minneapolis, MN; and [3]North Dakota State University, Dept. of Plant Sciences, Fargo, ND
[*]Corresponding Author: PH: (605) 688-6907, Email: jose.gonzalez@sdstate.edu

ABSTRACT

Each year, joint efforts from the collaborators of the USWBSI generate upwards of 100,000 phenotypic data points for scab severity on promising wheat lines through the Uniform Regional Scab Nurseries, in addition to related traits such as FDK and DON concentrations. These data are quite complex, with observed responses of often being categorical or bound (proportions), and conditional variance structures being correlated and heteroscadastic. Regardless, statistical analysis of these data has primarily been limited to ANOVA (fixed effect) models to compare entry means within and among sites, which are designed for the analysis of continuous and normally distributed data with homogeneous variance. Although the ANOVA F-test is fairly robust, inference regarding the genetic differences among entries using LSD methods can be misleading when applied to categorical and bound data. Furthermore, any imbalance in the data results in different levels of precision regarding the genetic values among entries, and can greatly bias the estimates of genetic merit for the entries.

Statistical models that have less restrictive assumptions than the ANOVA method are available to model complex data. These models do not require strict distributional assumptions and can more efficiently combine the data among locations and years into a single analysis to improve the results generated from the Uniform Regional Scab Nurseries. For this project, we are evaluating and comparing different statistical methods for estimating the genetic value of entries in multi-site scab nurseries, including linear fixed-effects models (ANOVA), linear mixed-effects models (BLUP analysis), and generalized linear mixed-effects models. Simulations are being conducted to compare the accuracy and precision of the estimates for genetic merit derived from these models. The utility of the models will then be validated by application to several years of uniform regional nursery data and comparison with the existing analysis results. If more sophisticated statistical methods are capable of even marginally improving the estimation of genetic value for scab resistance from regional disease nurseries, then they provide essentially a zero cost means for improving selection for scab resistance.

MAPPING AND PYRAMIDING SCAB RESISTANCE QTL IN EARLY GENERATION SPRING AND WINTER WHEAT BREEDING POPULATIONS USING A FAMILY-BASED MAPPING APPROACH

J.T. Eckard, J.L. Gonzalez-Hernandez*, K.D. Glover and W.A. Berzonsky

South Dakota State University, Department of Plant Science, Brookings, SD
*Corresponding Author: PH: (605) 688-6907, Email: jose.gonzalez@sdstate.edu

ABSTRACT

Resistance to Fusarium head blight in wheat is a complex trait, for which numerous QTL have been identified. While the use of *Fhb1* for development of host resistance is commonplace in wheat breeding programs, the vast genetic variation for resistance represented by smaller effect QTL has not been routinely exploited. This infrequent transition from QTL discovery to marker-assisted selection stems from disparity between mapping and breeding populations. Family-based linkage analysis developed for human pedigrees has been used successfully to identify *Fhb1* in early generation breeding populations (Rosyara et al. 2009. Theor. Appl. Genet. 118: 1617-1631), suggesting utility for integrating QTL mapping and marker-assisted selection.

This project extends the family-based mapping approach to identify and pyramid multiple scab resistance QTL in breeding populations with an *Fhb1* background. HRSW breeding populations consisting of 44 double-cross families have been developed by crossing novel sources of resistance with 16 experimental lines carrying *Fhb1* from the SDSU spring wheat breeding program. Winter wheat breeding populations consisting of 28 double-cross families have also been developed by crossing resistant varieties with 'Wesley'-Fhb1 backcross lines. Double-cross F_1 individuals were screened for resistance in the greenhouse by single head spray inoculations with *Fusarium graminearum* isolate Fg4. The heritability of individual plant severity in the greenhouse was between 0.33 and 0.39. Spring wheat F_2 lines were planted and rated for scab severity in inoculated disease nurseries by SDSU, NDSU and UMN to progeny test the F1 individuals. Winter wheat F_2 lines have been planted in disease nurseries by SDSU, NDSU, and KSU for evaluation in 2013. F_3 seed from top performing lines have been advanced to winter nurseries for further evaluation and increase.

Founder lines and double-cross F_1 individuals are being genotyped using SSR markers to provide a genome-wide scan for resistance QTL. Chromosomes 2B, 3B, 7B and 4D have already been genotyped to target previously reported QTL from the parent lines. Existing software packages MERLIN (Abecasis et al. 2002. Nat. Genet. 30(1): 97-101) and MENDEL (Lange et al. 2001. Amer J Hum Genetics 69: 504) are being used to conduct family-based linkage and association tests. Markers associated with identified resistance QTL will be used to assist selection and pyramiding of those QTL in F_4 lines for the purpose of validating the QTL and generating resistant germplasm for use in wheat breeding programs.

HISTORICAL COMPARISON OF THE NORTH AMERICAN BARLEY SCAB EVALUATION NURSERY (NABSEN)

P.L. Gross[1], R.D. Horsley[2], K.P Smith[3], J. Menert[4], W.G. Legge[5], J.R.Tucker[5] and R. Brueggeman[1*]

[1]Dept. of Plant Pathology, and [2]Dept. of Plant Sciences, North Dakota State University, Fargo, ND, USA; [3]Dept. of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN, USA; [4]Busch Ag. Resources Inc., Fort Collins, CO, USA; and [5]Agric. and Agri-Food Canada Research Centre, Brandon, Manitoba, CANADA

[*]Corresponding Author: PH: (701) 231-7078, Email: Robert.Brueggeman@ndsu.edu

ABSTRACT

The North American Barley Scab Evaluation Nursery (NABSEN) was established to screen elite two-rowed and six-rowed barley germplasm for resistance to Fusarium head blight (FHB) in regional uniform nurseries in North America and China. Participants included breeding programs from North Dakota State University (NDSU), University of Minnesota (UM), Busch Ag. Resources Inc. (BARI), and Agriculture & Agri-Food Canada Research Centre (AAFC). Each year (2002-2012) seven misted inoculated sites were established in the upper Midwest of the United States and one in Canada. From 2007-2012 a site was also established in China. NABSEN participants use different criteria to select elite material for testing; NDSU submits both two-rowed and six-rowed lines, BARI and UM programs submit primarily six-rowed entries, and the Canadian breeding programs submit mostly two- rowed lines. Here we report on the resistance to FHB of entries submitted from the NDSU, UM, BARI and AAFC breeding programs. Comparisons were made with Conlon (two-rowed resistant check) and Chevron (six-rowed resistant check) for deoxynivalenol (DON) content and FHB severity. These two resistant checks have been used consistently throughout all eleven years. AAFC and NDSU had at least one two-rowed entry equal to Conlon for DON accumulation in 63.6% and 54.5% of the years, respectively. AAFC and NDSU also had at least one entry equal to Conlon for FHB severity in 90.9 % and 81.8 % of the years, respectively. AAFC, NDSU, BARI and UM, had at least one six-rowed entry equal to Chevron for DON accumulation in 18.2%, 36.7%, 18.2% and 36.7% of the years, respectively. The NDSU breeding program has had one entry equal to Chevron for FHB severity in one of the eleven years. Over the past eleven years six North American varieties that are commercially available have been included in NABSEN testing. They include Tradition, and Quest six-rowed lines and Pinnacle, CDC Mindon, Norman and Taylor two-rowed barley lines. Several of these lines have improved resistance and the collective increased resistance of materials in the NABSEN through the years and release of varieties with FHB resistance are indicators of the progress in FHB resistance and the essential role that the NABSEN has played.

GETTING FUSARIUM HEAD BLIGHT (FHB) RESISTANCE WHERE IT COUNTS, IN EXISTING COMMERCIAL CULTIVARS

Steve Haber[1*], Jeannie Gilbert[1], Stephen Fox[1] and Steve Robinson[2]

[1]AAFC, Cereal Research Centre, 195 Dafoe Rd., Winnipeg, MB, R3T2M9, Canada; and [2]AAFC, Saskatoon Research Centre, 107 Science Place, Saskatoon, SK, S7N0X2, Canada
[*]Corresponding Author: PH: (204) 983-1467, Email: steve.haber@agr.gc.ca

ABSTRACT

For the deployment of host genetic resistance to Fusarium head blight (FHB) to confer meaningful benefits it must preserve, or possibly enhance, the combinations of useful traits expressed by elite germplasm. In wheat, multiple genes must interact in complex ways to express FHB resistance that is sufficient to protect against losses to yield and quality. This poses a severe challenge to efforts to achieve this level of resistance while retaining all the desirable attributes of a commercial cultivar. Indeed, no contemporary cultivar has derived a high level of FHB resistance by introgression of genes from known resistance sources. An alternative approach opened with the finding that FHB-resistant 'Sumai 3' and its susceptible near-isogenic lines do not differ in the general plant defence genes that are induced in response to inoculation by *Fusarium graminearum*. With this indication that improved FHB resistance might be gained by changing the control of expression of existing genes rather than introgressing new ones, we sought to evolve heritable traits *de novo* in the descendants of germplasm subjected to systemic stresses, an approach that has already yielded new sources of resistance in wheat to wheat streak mosaic virus (WSMV) and rust infections. We chose as our starting material the contemporary Canadian hard red spring wheat cultivar 'Waskada' (intermediate FHB reaction), and subjected succeeding generations to systemic stresses including virus infection, heat and cold. In each cycle, we selected and advanced plants that differed visibly from their progenitors in a range of traits including FHB resistance, which we evaluated for the first time in the 2010 FHB nursery. The three sublines which exhibited better resistance than relevant checks became the founders of three families of sublines that were advanced in cycles of further selection in the field over the next two years before entering them in the 2012 field nursery. In this trial the original 'Waskada' progenitor had an FHB index score of 22.5, the best family of sublines had a mean score of 4.2 while the most resistant, but non-elite, checks scored 0.5-1.0. A preliminary agronomic trial, conducted in the absence of disease pressure, produced seed used to determine that the families of sublines evolved from 'Waskada' possessed quality traits similar to the cultivar progenitor.

ASSOCIATION MAPPING OF FUSARIUM HEAD BLIGHT RESISTANCE IN U.S. HARD WINTER WHEAT

Feng Jin[1], Dadong Zhang[1], Chengsong Zhu[1], William Bockus[2], P. Stephen Baenziger[3], Shiaoman Chao[4] and Guihua Bai[1,5*]

[1]Dept. of Agronomy and [2]Dept.of Plant Pathology, Kansas State University, Manhattan, KS, 66506; [3]Dept. of Agronomy and Horticulture, University of Nebraska, Lincoln, NE, 68583; [4]USDA-ARS, Fargo, ND; and [5]USDA-ARS-HWWGRU, Manhattan, KS, 66506
[*]Corresponding Author: PH: 785-532-1124, Email: guihua.bai@ars.usda.gov

ABSTRACT

Fusarium head blight (FHB) is one of the most destructive diseases of wheat worldwide. FHB epidemics can cause severe losses in both grain yield and quality of wheat. One of the most effective approaches to reduce the disease losses is to grow FHB resistant cultivars. To characterize resistance quantitative trait loci (QTLs) in U.S. winter wheat, association mapping was conducted using 135 hard winter wheat, 66 soft winter wheat accessions and Sumai 3 as resistant control. All accessions were evaluated for FHB severity (type II) using single-point inoculation in greenhouse. Total 282 SSR, 21 other types of markers and 9000 genome-wide SNP markers were genotyped. Population structure analysis divided the population into four subgroups, including two hard winter wheat groups and two soft winter wheat groups, and Q model was the best for association mapping. In the greenhouse experiments, 4 SSR and 15 SNP markers were associated with type II resistance mainly located in chromosome of 1A, 2A, 2B, 3B, 3D, 4A, 6A, 6B, etc. Several QTL and significant markers associated with type II resistance identified in this study should be useful for improvement of FHB resistance in hard winter wheat.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

EVALUATION OF FHB RESISTANCE AND AGRONOMIC PERFORMANCE IN SOFT RED WINTER WHEAT ACROSS POPULATIONS

Jerry Johnson*, Dan Bland, Yuanfeng Hao and Zhenbang Chen

Crop and Soil Sciences, Griffin Campus, The University of Georgia, Griffin, Georgia 30223
*Corresponding Author: PH: 770-468-5101, Email: jjohnso@uga.edu

ABSTRACT

Development of resistant wheat cultivars is the most efficient approach to control FHB. Local broadly adaptive cultivars have been crossed with *Fhb1* derived lines, Truman, and Jamestown to introduce FHB resistant QTL into local adaptive genetic backgrounds. Eight elite lines (GA051173W-S11, GA051173-S18, GA051173-S25, GA 051207-S19, GA 051207-S21, GAMD08-27-E9-S13 GAMD08-27-E9-S14, and GAMD08-27-E9-S15) with resistance from either Truman, IN981359C1, or Ning 7840, were evaluated in the field in 2012 for FHB resistance and agronomic performances. The closely linked markers were also used to detect the resistant QTL for FHB and other critical diseases. GAMD08-27-E9-S13, GAMD08-27-E9-S14, and GAMD08-27-E9-S15 maintained the *Fhb1* and 5A QTL from the resistant donor Ning 7840. Elite lines, GA051173-S11 and GA051173-S18, selected from the cross of Truman and AGS 2010, showed a high level of FHB resistance which was similar to the resistant controls, Bess and Jamestown. These five elite lines also included important resistant genes for Hessian fly (*H13*) and leaf rust (*Lr37/Yr17/Sr38*). GA 051207-S19 and GA 051207-S21 were the highest yielding lines with moderate resistance for FHB index and ISK index when compared to the check "AGS 2035". In addition, an elite line GA041052-11E51 evaluated in the GAWN also showed high level of FHB resistance and had very high grain yield. Several other lines with Jamestown as source of resistance will be further evaluated for FHB and grain yield.

GENETIC ANALYSIS OF FUSARIUM HEAD BLIGHT RESISTANCE IN TUNISIAN-DERIVED DURUM WHEAT POPULATIONS

Shahryar F. Kianian*, Seyed M. Pirseyedi, Farhad Ghavami, Ajay Kumar and Elias M. Elias

Department of Plant Sciences, North Dakota State University, Fargo, ND
*Corresponding Author: PH (701) 231-7574, Email: s.kianian@ndsu.edu

ABSTRACT

Host plant resistance is recognized as the most effective means of controlling FHB infection. Resistant FHB varieties in hexaploid wheat have been released; however, the progress toward the same goal in durum wheat (*Triticum turgidum* ssp. *durum* Desf.) wheat has been limited. Sources of resistance in durum wheat are few and transferring the resistance genes from hexaploid wheat have met with limited success. The new Tunisian resistant durum sources found recently show promising amount of resistance comparable to the hexaploid sources. We have used these sources in several studies to identify and mark genomic regions of interest for introgression into cultivated durum varieties.

In one study we used 171 BC_1F_6 and 169 BC_1F_7 lines derived from crossing of four Tunisian tetraploid sources of resistance (Tun7, Tun18, Tun34, Tun36) with durum cultivars 'Ben', 'Maier', 'Lebsock' and 'Mountrail' for association studies. The Tun18 and Tun7 FHB resistances were found to be comparable to the best hexaploid wheat sources. A new significant QTL for FHB resistance was identified on the long arm of chromosome 5B (*Qfhs.ndsu-5BL*) with both association and classical QTL mapping analysis. Linkage disequilibrium (LD) blocks extending up to 40 cM were evident in these populations. The linear mixed model considering the structure (Q or P) and the kinship matrix estimated by REML (K_T) identified as the best for association studies in a mixture of wheat populations from a breeding program. The results of association mapping analysis also demonstrated a region on the short arm of chromosome 3B as potentially linked to FHB resistance. This region is in proximity of major FHB resistance gene "*Fhb1*" reported in hexaploid wheat. A possibility of having susceptibility or suppressor of resistance gene(s) on durum wheat chromosome 2A was further confirmed in this material explaining the problem in developing resistant genotypes without counter selection against this region.

In another study, two additional Tunisian- derived advanced backcross populations, Tun 108 × Lebsock/ Lebsock and Tun 108 × Ben/Ben, were screened for FHB resistance in both greenhouse and field. A total of 171 BC_1F_7 lines derived from Tun 108 × Ben/Ben were phenotyped for their reaction to FHB in two greenhouse and two field experiments. Analysis of variance showed significant effect on FHB infection rate for the genotypes and also environments, as well as G×E interactions. Broad sense heritability for FHB infection rate was calculated to be around 40.4%±0.09. The correlation between the two greenhouse seasons and also the two field scab nurseries were positive and significant while there was correlation between only one of the greenhouse data and the field data. Transgressive segregation for FHB severity was observed and approximately 5% of the lines performed better than the resistant parents in the field and 25-30% while evaluated in the greenhouse. A total of 329 markers were mapped to 239 unique loci with coverage of 1887.6 cM, and an average of 7.89 cM between any two marker loci. QTL analysis for FHB resistance revealed six different QTL on 5 different chromosomes (1A,1B, 2B, 3B, 5A, 5B, 7A and 7B). The QTL on 5A and 7A were both effective in the field and greenhouse and

together explained ~ 9% of total phenotypic variation and 22.5% of genetic variation in Tun108×Ben/Ben population. The QTL regions on chromosomes 2B and 3B both were associated with resistance to severity, incidence, FDK and DON.

We used the same procedure for screening a population of 174 BC1 F7 individuals derived from cross between Tun 108 and Lebsock/Lebsock in 2 greenhouse and 2 field trial during 2010 to 2011. Additionally, FDK, DON, 3ADON, 15ADON and Nivalenol were measured on samples collected from 2011 field experiment. Analysis of variance indicated a significant effect on FHB infection rate for the genotypes and also environments, as well as G×E interactions. Broad sense heritability for FHB infection rate was estimated to be around 33.4%±0.09. The correlation between the two greenhouse seasons and also the two field scab nurseries were positive and significant while there was no correlation between the greenhouse data and the field data. Remarkable correlations between disease incidence and FDK ($r = 0.53$, $p < 0.0001$) was assessed, while there was no significant correlation between severity and FDK. There was no correlation between disease incidence and DON, severity and DON. Transgressive segregation for FHB severity was observed within this population. Nearly 5% of the lines with highest resistance in the field, lowest FDK and DON accumulation were selected for incorporation into the breeding program. The QTL analysis is underway and the results will be reported.

The most FHB resistant Tunisian derived backcross lines and associated markers are being employed to incorporate valuable regions into advanced durum breeding lines for cultivar improvement.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-063. This is a cooperative project with the U.S. Wheat and Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

METABOLIC PROFILING REVEALS NOVEL INSIGHTS INTO THE BIOTRANSFORMATION OF DON IN WHEAT

B. Kluger[1], C. Bueschl[1], R. Krska[1], M. Lemmens[2], G. Adam[3*] and R. Schuhmacher[1]

[1]Center for Analytical Chemistry, Department for Agrobiotechnology (IFA-Tulln); [2]Institute for Biotechnology in Plant Production, Department for Agrobiotechnology (IFA-Tulln), Konrad-Lorenz-Str. 20; and [3]Department of Applied Genetics and Cell Biology, Konrad-Lorenz-Str. 24, 3430 Tulln, University of Natural Resources and Life Sciences (BOKU), Vienna, Austria
[*]Corresponding Author: PH: 0043-1-47654-6380, Email: gerhard.adam@boku.ac.at

ABSTRACT

In a recent study we have developed a novel approach for the untargeted screening of metabolites of xenobiotics in plants. The approach was successfully applied to wheat plants after treatment with a mixture of $^{13}C_{15}$– and non-labeled deoxynivalenol (DON) and resulted in the assignment of a total of nine different DON conjugates. Besides the well-known DON-3-*O*-glucoside, for the first time the occurrence *in planta* of DON-glutathione (GSH), DON-S-cysteinyl-glycine, DON-S-cysteine have been reported together with five other DON conjugates [1].

In the present work we have further characterized the molecular structures of the remaining five DON conjugates by liquid chromatography - tandem mass spectrometry (LC-MS/MS). Additionally, DON- and *Fusarium graminearum* treated near isogenic wheat lines, which differed in two major resistance quantitative trait loci (QTLs) against Fusarium head blight (FHB) *Qfhs.ndsu-3BS* and *Qfhs.ifa-5A*, were monitored for all nine DON conjugates.

For the two parent lines Remus (susceptible) and CM-82036 (resistant) and each NIL, wheat ears were challenged at anthesis in two inoculation variants with *F. graminearum*, and DON and harvested at 0, 14, 48 and 96h after inoculation. For each time point and inoculation variant 5 plants (1 ear per plant) were treated. After the respective inoculation period, treated ears were harvested, immediately frozen in liquid nitrogen and stored at -80°C until analysis. Frozen plant samples were homogenized in a ball mill with liquid nitrogen, extracted and analyzed by LC-high resolution-MS.

This contribution will present the putative molecular structures of the identified (novel) DON conjugates and will compare the formation of these DON biotransformation products over a time period of up to 96 hours after inoculation with either DON or *F. graminearum* strain PH-1. The results will be evaluated and discussed in view of the different combinations of resistance QTLs present in the tested near isogenic wheat lines.

ACKNOWLEDGEMENT

This work was funded by the Austrian Science Fund FWF (SFB F37).

REFERENCE

[1] Kluger B, Bueschl C, Lemmens M, Berthiller F, Häubl G, Jaunecker G, Adam G, Krska R, Schuhmacher R (2012) Stable i sotopic labelling-assisted untargeted metabolic profiling reveals novel conjugates of the mycotoxin deoxynivalenol in wheat. Analytical and Bioanalytical Chemistry:1-6. doi:10.1007/s00216-012-6483-8

COMPARISON OF VISUAL AND DIGITAL IMAGE ANALYSIS METHODS FOR ESTIMATION OF *FUSARIUM* DAMAGED KERNELS IN WHEAT

P.V. Maloney[1], S. Petersen[1], R.A. Navarro[1], D. Marshall[2], A.L. McKendry[3], J.M. Costa[4] and J.P. Murphy[1*]

[1]North Carolina State University, Raleigh, NC; [2]USDA-ARS, Raleigh, NC; [3]University of Missouri, Columbia, MO; and [4]University of Maryland, College Park, MD
[*]Corresponding Author: PH: (919) 513-0000, Email: paul_murphy@ncsu.edu

ABSTRACT

Fusarium head blight (FHB), or head scab, causes a reduction in grain yield and quality, as well as, the formation of shriveled, dull-grey seeds called "tombstones" or *Fusarium* damaged kernels (FDK). FDK is commonly quantified on a percentage basis by visually separating damaged kernels from the healthy kernels following harvest, a process that is both time consuming and labor intensive. The objective of this study was to evaluate an alternative method for quantifying FDK through the use of digital imagery and the digital image analysis program ImageJ. The 'NC-Neuse' x 'AGS 2000' recombinant inbred population of 172 lines and the NC-Neuse x 'Bess' double haploid population of 112 lines were used in this study. NC-Neuse and Bess were moderately resistant and AGS 2000 was susceptible to FHB. Both populations were evaluated under moderate to heavy FHB pressure in a total of five environments in North Carolina, Maryland and Missouri with two to three replications per environment. Wheat heads from each plot were harvested, dried, threshed, and cleaned by hand. Digital image analysis estimates were obtained by applying a hue, balance, saturation filter in ImageJ to images captured using a standard digital SLR camera. The filter was set to exclude the less color saturated (grey) kernels. ImageJ would then output the proportionate area of damaged kernels.

Significant genetic variation was observed using both visual and digital image analysis methods to estimated FDK. These methods' correlation values ranged from 0.72 to 0.80 over all environments. A lower correlation value of 0.54 was observed in Columbia, MO because of cracked and broken kernels in the samples. Digital image analysis was three times faster than the visual method, and was able to estimate FDK on a larger per plot sample whereas labor and time constraints limited the sample size for the visual method. Digital image analysis was consistent over different samples and appears well suited as an alternative form of FDK detection in unbroken grains samples.

PRE-BREEDING THROUGH RECURRENT MASS SELECTION

Francois Marais*, David Cookman, Bradley Bisek and Tyler Larson

Department of Plant Sciences, North Dakota State University, Fargo, ND 58108
*Corresponding Author: PH: 701-231-8155, Email: Gideon.marais@ndsu.edu

ABSTRACT

Recurrent mass selection (RMS) is an ideal breeding strategy for the enrichment of a breeding population with respect to genes that govern complex, polygenic traits. In North Dakota, primary targets for hard red winter wheat breeding include increased winter-hardiness, high yield potential, good processing quality and effective resistance against major diseases such as Fusarium head blight, leaf and stem rust, tan spot and the septoria complex. When RMS is applied to pre-breeding, generation intervals can be shortened considerably through single seed descent. Accelerated single seed descent (SSD) inbreeding can be achieved by utilizing small soil volume, extended daylight hours, elevated temperature during growth, premature harvesting and seed drying. During SSD inbreeding, the integration of seedling screening for rust resistance or low temperature tolerance, marker aided selection for key genes and field selection for FHB resistance amid artificial epidemics can help to raise the target gene frequencies and eventually facilitate gene pyramiding. A highly diverse pre-breeding base population has been derived by making an *Ms3*- (dominant male sterility) assisted complex cross with 110 diverse lines and varieties contained within five populations. These included genotypes with native and exotic resistance and adaptation genes, primarily from spring wheat or less cold-hardy winter wheat. This base population will be subjected to recurrent selection during which F_1 female plants (1-year cycle) will be cross-pollinated with F_4-derived F_5 lines that had been field selected once during a 3-year breeding cycle.

IS THE FUSARIUM HEAD BLIGHT RESISTANCE IN TRUMAN SOFT RED WINTER WHEAT NOVEL?

Anne L. McKendry*

Division of Plant Sciences, University of Missouri, Columbia 65211
*Corresponding Author: PH: (573) 882-7708, Email: mckendrya@missouri.edu

ABSTRACT

At the University of Missouri, we have been addressing losses associated with Fusarium head blight (FHB) mainly caused by *Fusarium graminearum* Schwabe [telomorph: *Gibberella zeae* Schw. (Petch)] through the identification and use of 'native' sources of resistance identified in soft red winter wheat germplasm of U.S. origin. 'Truman', developed and released by the University of Missouri has excellent, broad-based FHB resistance, coupling low incidence and severity, good kernel quality retention, and low DON with good adaptation and agronomic performance throughout the northern soft red winter wheat region. It serves as the resistant check in the Northern Uniform Scab Nursery and the Northern Preliminary Scab Nursery. Truman carries none of the known markers for FHB resistance used by the Wheat Genotyping Laboratory at Raleigh NC therefore, it may provide different FHB resistance genes that could complement other widely-used sources of resistance. Anecdotal evidence from the Missouri breeding program suggests that the source of resistance in Truman is partially dominant, and highly penetrant. Results of a diallel study of 20 winter wheat varieties confirmed that across a wide range of resistant and susceptible genotypes, Truman had very good general combining ability. A small but significant specific combining ability component was also detected which appeared to be associated with dominance gene action rather than epistasis. With funding from the USWBSI, a QTL study was undertaken on an $F_{6:8}$ recombinant inbred line mapping population developed at the Univ. of Missouri from the cross Truman x MO 94-317. Greenhouse type II phenotypic data were collected at the Univ. of Missouri while phenotypic data for field-based traits (incidence, severity, and *Fusarium* damaged kernels and DON) were collected at Missouri, Purdue, and/or the University of Kentucky. DON analyses were done at the University of Minnesota. Genetic linkage maps were constructed from 160 SSR markers, and 530 Dart markers. QTL analyses of greenhouse type II resistance combined over years identified four QTL on chromosomes 1BSc, 2BL, 2DS, and 3BSc that accounted for 10.9, 16.1, 19.9 and 7.3% of the variability respectively. QTL on 1BSc, 2BL and 3BSc do not appear to be novel. The QTL on 2DS, linked with the DArT marker wPt666223, however, may be unique to Truman. The same 2DS QTL was also detected for incidence, severity, FHBI (incidence x severity), and DON accounting for 22.9, 23.0 and 25.3, and 20.7% of the phenotypic variance, respectively as well as for incidence based on Purdue data and DON based on KY data where it accounted for 24.3 and 20.1% of the phenotypic data, respectively. Although significant for FDK the 2DS QTL had a smaller effect, accounting for 7.5% of the phenotypic variance. A second QTL on 2ASc that was also common across incidence, *FhbI*, FDK and DON accounted for 6.7, 8.2, 8.5, and 10% of the variation respectively, and may also be novel in Truman although a QTL near this region has been identified in Wangshuibai for both DON and FDK. Finally, a QTL on 3DS associated with incidence in Truman, accounting for 10% of the variance also appears to be novel. Other QTL identified in this population appear to have been reported in European, Asian or U.S. germplasm. Among the unique QTL in Truman, the region on 2D has the largest effect and providing significant reductions in DON, incidence, and disease spread within the head. Once validated, it may provide gene(s) complementary to other widely used sources that should prove valuable for marker-assisted-selection.

MAPPING OF FUSARIUM HEAD BLIGHT RESISTANCE IN WILD BARLEY ACCESSION PI 466423

J. Menke[1], K. Beaubien[2], T. Szinyei[1], Y. Dong[1], S. Chao[3], P. Olivera[1], B. Alsop[1], S. Dahl[1], K. Smith[2] and B. Steffenson[1*]

[1]Department of Plant Pathology and [2]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108; and [3]USDA-ARS Cereal Crops Research Unit, Fargo, ND 58105
[*]Corresponding Author: PH: (612) 625-4735, Email: bsteffen@umn.edu

OBJECTIVE

To map loci conferring Fusarium head blight resistance in wild barley.

INTRODUCTION

The devastating Fusarium head blight (FHB) epidemics occurring over the past two decades contributed to the decline of the malting barley industry in the Red River Valley region of the USA and Canada. The best strategy for reducing disease and mycotoxin levels is an integrated approach of cultural practices, fungicide application, and host resistance. To identify resistance to FHB, over 23,000 *Hordeum* accessions were screened in field nurseries in the Midwest and China (*1, 2*). These studies showed that resistance to FHB was extremely rare as less than 2% of accessions carried useful levels of resistance for breeding. Moreover, resistance was only partial as no immune or even highly resistant accessions were identified.

Several molecular mapping studies involving cultivated barley accessions and landraces revealed that FHB resistance is controlled, in most cases, by a number of loci with relatively small effects that are scattered across the barley genome (*3, 4, 5, 6, 7*). Additionally, some agronomic (e.g. heading date and plant height) and spike architecture (e.g. row type and kernel density) traits were found to map at coincident chromosomal positions as the identified FHB resistance loci (*8*). These data raise important questions as to whether these associations are due to closely linked genes or possibly pleiotropy.

Wild barley is a very rich source of disease resistance genes, but has not been fully exploited for FHB resistance. From the extensive evaluations of *Hordeum* germplasm, we identified 27 wild barley accessions with partial resistance to FHB (*1*). One of the most resistant accessions was PI466423, which was originally collected near the village of Mehola west of the Jordan River in Israel. Our overall goal is to reduce the losses caused by FHB, especially quality discounts due to the accumulation of DON. This can be best achieved by identifying and incorporating into barley cultivars genes that confer a high level of resistance to FHB and the accumulation of mycotoxins. To increase the diversity of FHB resistance alleles in cultivated barley, we developed an advanced backcross population involving PI466423 and cultivar Rasmusson. Our specific objective was to map loci conferring FHB resistance in wild barley accession PI466423.

MATERIALS AND METHODS

Plant materials. A recombinant chromosome substitution line (RCSL) population was developed for the advanced backcross strategy of Tanksley and Nelson (*9*) using the crossing scheme of Matus et al. (*10*). The final population consisted of 258 BC_2F_4 lines

FHB phenotyping. The RCSL population was assessed for FHB severity (in percent) at St. Paul and Crookston, Minnesota in 2010, 2011 and 2012 and in Hangzhou, China in 2011. DON accumulation (in parts per million) also was assayed in all environments, except the 2012 Minnesota nurseries (pending) and 2011 China

nursery. Nurseries at Crookston and Hangzhou were inoculated using the "grain spawn" method where ascospores are the primary inoculum source (*8*). At St. Paul, a calibrated load of *Fusarium graminearum* conidia was applied to plants at the heading stage using the "spray inoculation" method. FHB and DON assessments were made according to the methods of Ma et al. (*5*) and Fuentes et al. (*11*), respectively.

Extraction of genomic DNA. Sections (5 cm) of leaf material were harvested from 2-week-old BC_2F_4 seedlings grown in a greenhouse. Tissue was flash frozen in liquid nitrogen, stored at -80˚C, and lyophilized to dryness. Leaf tissue was ground to powder using a Qiagen TissueLyser II and 3 mm tungsten carbide beads. Genomic DNA was isolated from ground tissue using a Qiagen BioSprint 96 workstation and the BioSprint 96 DNA Plant Kit. Quality and quantity of isolated genomic DNA was assessed via agarose gel electrophoresis and spectrophotometry, respectively.

Single nucleotide polymorphism (SNP) genotyping. Genotyping of parents and progeny was conducted at the USDA-ARS Cereal Crops Research Unit in Fargo, North Dakota. Samples of genomic DNA were genotyped using the Illumina Infinium assay. The Illumina genotyping array is capable of interrogating 7,863 SNPs within the barley genome simultaneously. Genotype calls for all SNP markers were visually inspected and validated using the GenomeStudio Genotyping Module. This analysis yielded 6,710 informative markers, 2,252 of which were polymorphic between the parental lines.

Genetic map construction. Genetic maps of the seven barley chromosomes were constructed using Join Map 4.0. All 2,252 polymorphic SNPs were considered for map construction, though only a single marker representing markers with greater than 98% redundancy was included in the final map. Linkage groups were determined by recombination frequency and only marker pairings with LOD > 3.0 were included in the final map. Kosambi's mapping function was used for map construction.

Marker analysis and QTL mapping. Only six-rowed RCSLs were used in the QTL analysis to eliminate the possible confounding factor of row type. Chromosome bin positions were approximated based on Munoz-Amatriain et al. (*12*). Single marker analysis to detect marker associations was conducted using QTL Cartographer 2.5_011 (*13*). Data were analyzed as a trait mean for each environment with a total of four to seven environments per trait (environments per trait are listed in Table 1). Marker-trait QTL were defined if the association was detected in two or more environments at or above LOD 3.0. QTL were defined as major if they were detected in greater than 75% of all trait environments. Allele effects at the QTL location were calculated in reference to the Rasmusson allele. Correlations for trait means were conducted using Excel.

RESULTS AND DISCUSSION

FHB and DON levels varied markedly across locations and also years (see summary data in Table 1). QTL analysis revealed a major effect QTL for FHB severity and DON concentration on chromosome 2H, bin 4 (Fig. 1). This QTL was observed in six of the seven environments, and the percentage of phenotypic variance explained ranged from 40% in 2010 to 7% in 2012 for FHB and from 25% in 2011 to 18% in 2010 for DON. Analysis of heading date and plant height revealed QTL coincident with the one for FHB and DON in the bin 4 region of chromosome 2H (Fig. 1). It is not known whether closely linked loci control these different traits or if pleiotropy may be involved. If the former case, the resistance allele may be unique and therefore useful for enhancing FHB resistance of breeding lines carrying other resistance alleles. The question of linkage vs. pleiotropy will be resolved with additional crosses as was done previously with FHB resistance loci and heading date loci by Nduulu et al. (14). In previous studies on mapping FHB resistance, QTL for heading date

(*5*,*15*), height (*15*), and DON accumulation (*3*) were identified on chromosome 2H bin 4.

The allelic effect (α) for the chromosome 2 bin 4 FHB QTL was inconsistent with four environments (Crookston 2010, 2011, and 2012 and St. Paul 2010) showing a negative α for Rasmusson, and two environments (St. Paul 2011 and 2012) showing a positive α for Rasmusson. Phenotyping barley germplasm for FHB resistance and DON accumulation is fraught with many sources of variation (*8*). Some of the variation we observed across environments may be due to the different inoculation methods employed (grain spawn spread on ground with ascospore inoculum source vs. spray inoculation directly on heading plants with conidial inoculum source), timing of disease assessments (St. Paul is taken at set intervals after inoculation whereas Crookston is not) and perhaps local climatic factors (Crookston is cooler than St. Paul and hence plants have a longer plant maturation period). Nevertheless, the same QTL was detected across different environments.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-1-119. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

REFERENCES

1. S. K. Dahl et al., Evaluation of *Hordeum* accessions for resistance to Fusarium head blight. *Proc. 2009 National Fusarium Head Blight Forum.* Orlando, FL (2009).

2. Y. Huang et al., Haplotype diversity and population structure in cultivated and wild barley evaluated for Fusarium head blight responses. *Theor. Appl. Genet.* (2012). (in press)

3. R. C. de la Pena et al., Quantitative trait loci associated with resistance to Fusarium head blight and kernel discoloration in barley. *Theor. Appl. Genet.* **99**, 561-569 (1999).

4. H. Zhu et al., Does function follow form? Principal QTLs for Fusarium head blight (FHB) resistance are coincident with QTLs for inflorescence traits and plant height in a doubled-haploid population of barley. *Theor. Appl. Genet.* **99**, 1221-1232 (1999).

5. Z. Ma, B. J. Steffenson, L. K. Prom, and N. L. V. Lapitan. Mapping of quantitative trait loci for Fusarium head blight resistance in barley. *Phytopathology*. **90**, 1079-1088 (2000).

6. A. Mesfin et al., Quantitative trait loci for Fusarium head blight resistance in barley detected in a two-rowed by six-rowed population. *Crop Sci.* **43**, 307-318 (2003).

7. R. D. Horsley et. al., Identification of QTLs associated with Fusarium head blight resistance in barley accession CIho 4196. *Crop Sci*. **46**, 145-156 (2006).

8. B. J. Steffenson, in: *Fusarium Head Blight of Wheat and Barley.* K. J. Leonard, W.R. Bushnell, Eds. (APS, St. Paul, 2003), pp. 241-295.

9. S. D. Tanksley, J. C. Nelson, Advanced backcross QTL analysis: a method for the simultaneous discovery and transfer of valuable QTLs from unadapted germplasm into elite breeding lines. Theor. Appl. Genet. **92**, 191-203 (1996).

10. Matus et al., Development and characterization of recombinant chromosome substitution lines (RCSLs) using *Hordeum vulgare* subsp. *spontaneum* as a source of donor alleles in a *Hordeum vulgare* subsp. *vulgare* background. Genome **46**, 1010–1023 (2003).

11. R. G. Fuentes et. al., Resource Allocation and Cultivar Stability in Breeding for Fusarium Head Blight Resistance in Spring Wheat. *Crop Sci*. **45**, 1965-1972 (2005).

12. M. Muñoz-Amatriaín et. al., An improved consensus linkage map of barley based on flow-sorted chromosomes and single nucleotide polymorphism markers. *Plant Genome*. **4**, 238–249 (2011).

13. S. Wang , C. J. Basten, Z-B. Zeng, Windows QTL Cartographer 2.5. Department of Statistics, North Carolina State University, Raleigh, NC. (2012). (http://statgen.ncsu.edu/qtlcart/WQTLCart.htm)

14. L. M Nduulu, A. Mesfin, G. J. Muehlbauer, K. P. Smith, Analysis of the chromosome 2(2H) region of barley associated with the correlated traits Fusarium head blight resistance and heading date. *Theor. Appl. Genet.* **115**, 561-570 (2007).

15. P. M. Hayes et. al., Quantative trait locus effects and environmental interaction in a sample of North American Barley germplasm. *Theor. Appl. Genet.* **87**, 392-401 (1993).

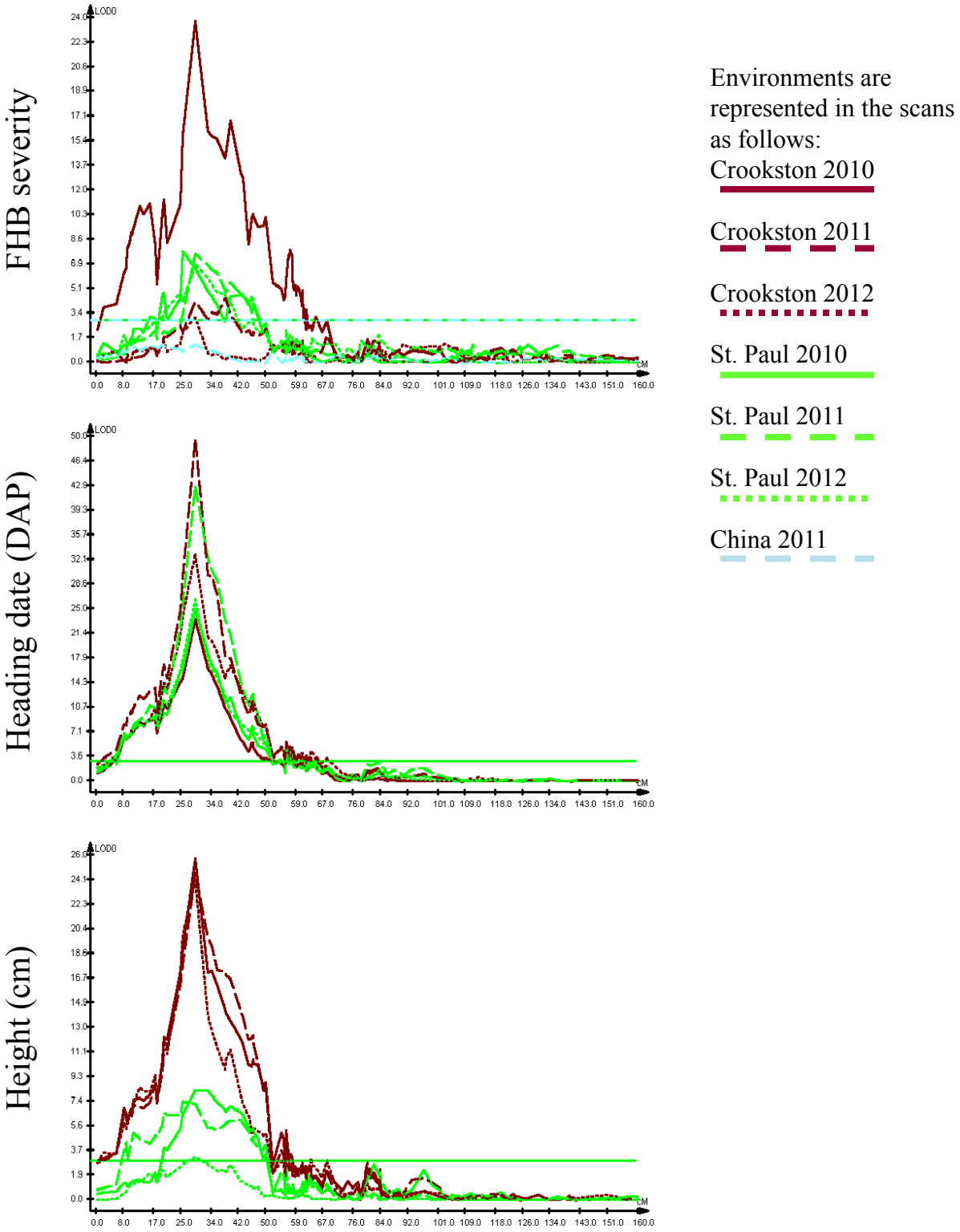

Figure 1. LOD scans for major QTL at chromosome 2 bin 4 for FHB severity, heading date and height in the PI466423/Rasmusson population. Allelic effect and R2 values for these peaks are presented in Table 1.

Table 1. Phenotypic and QTL summary for the PI466423/Rasmusson population for FHB severity, heading date, height, DON accumulation and spike angle.

Trait[§]	Environment[#]	PI466423	Rasmusson	Mean[†]	Min[†]	Max[†]	S.E.[†]	QTL Peak[£]	R^2	Alpha[‡]	QTL Peak[£]	R^2	Alpha[‡]
FHB	CR 2010	nd*	14	21	3	90	11.37	2 -- 4	0.409	-10.651			
	CR 2011	13	39	32	11	69	10.21	2 -- 4	0.090	-4.500			
	CR 2012	8	9	12	2	35	4.61	2 -- 4	0.070	-1.748			
	SP 2010	nd	9	13	1	37	6.78	2 -- 4	0.159	-3.787			
	SP2011	nd	12	11	1	34	7.30	2 -- 4	0.156	4.271			
	SP 2012	nd	nd	17	2	58	9.67	2 -- 4	0.141	5.402			
	CH 2011	1	1	1	0	4	0.42						
HD	CR 2010	58	52	51	47	56	1.86	2 -- 4	0.404	1.740			
	CR 2011	57	50	50	42	57	3.13	2 -- 4	0.661	3.739			
	CR 2012	63	59	59	50	69	4.49	2 -- 4	0.521	4.728			
	SP 2010	nd	58	55	41	69	3.70	2 -- 4	0.424	3.523			
	SP 2011	73	65	64	52	71	3.57	2 -- 4	0.608	4.094			
	SP 2012	nd	nd	57	45	75	6.04	2 -- 4	0.437	6.028			
HT	CR 2010	72	76	83	59	110	8.18	2 -- 4	0.429	7.890	3 -- 12	0.076	-3.219
	CR 2011	81	63	70	48	92	8.13	2 -- 4	0.416	7.721	3 -- 12	0.067	-3.360
	CR 2012	80	80	79	39	105	9.48	2 -- 4	0.420	8.922	3 -- 12	0.092	-4.337
	SP 2010	nd	84	83	58	114	9.29	2 -- 4	0.168	5.574	3 -- 12	0.069	-3.503
	SP 2011	nd	60	62	35	82	6.96	2 -- 4	0.155	3.819	3 -- 12	0.076	-3.042
	SP 2012	nd	66	67	39	86	7.77	2 -- 4	0.068	3.034	3 -- 12	0.067	-2.843
DON	CR 2010	3.00	23.17	15.07	1.00	56.90	8.19	2 -- 4	0.184	5.018			
	CR 2011	28.50	36.45	34.06	3.70	83.00	15.05	2 -- 4	0.255	10.621			
	SP 2010	nd	19.37	20.83	3.40	54.40	8.62	2 -- 4	0.159	4.877			
	SP 2011	nd	12.97	8.49	0.29	29.30	6.12	2 -- 4	0.162	3.631			
SA	CR 2010	1	1	1.1	1	3	0.35				5 -- 10	0.081	-0.157
	CR 2012	1.2	1	1.2	1	2.5	0.35				5 -- 10	0.044	-0.110
	SP 2010	nd	1	1.1	1	3	0.36	2 -- 6	0.141	-0.251			
	SP 2012	nd	1	1.1	1	3	0.28	2 -- 6	0.137	-0.165	5 -- 10	0.068	-0.125

[§]Trait abbreviations are FHB (FHB Severity), HD (heading date in days after planting), HT (height in cm), DON (deoxynivalenol accumulation), SA (spike angle on 1—3 scale where 1 is upright and 3 is nodding).

[#]Environments are abbreviated: CR denotes Crookston, MN, SP denotes St. Paul, MN, CH denotes Hangzhou, China.

*nd denotes no data available for parent at this environment

[†] Mean, minimum, maximum and standard error are presented for the PI466423/Rasmusson population

[£]QTL Peaks are listed by chromosome and bin position. Numbers listed are: (chromosome) – (bin)

[‡]Allelic effect of the Rasmusson allele at the QTL peak

'PROSPER': A NEW HARD RED SPRING WHEAT CULTIVAR COMBINING HIGH YIELD AND GOOD RESISTANCE TO FUSARIUM HEAD BLIGHT, LEAF DISEASES, AND QUALITY ATTRIBUTES

Mohamed Mergoum[1*], Richard C. Frohberg[1], Senay Simsek[1], Tika B. Adhikari[2], Jack W. Rasmussen[2], Shaobin Zhong[2], Maricelis Acevedo[2], Mohammed S. Alamri[3], Pawan K. Singh[4], Timothy L. Friesen[5] and James A. Anderson[6]

[1]Dept. of Plant Sciences and [2]Dept. of Plant Pathology, North Dakota State University, Fargo, ND, 58108; [3]Nutrition and Food Sciences Dep., King Saud Univ., P.O. Box 2460, Riyadh 11451, Saudi Arabia; [4]CIMMYT, Km. 45, Carretera México-Veracruz, El Batán, Texcoco CP 56130, Edo. de México, Mexico; [5]USDA-ARS, Fargo, ND, 58108; and [6]Dept. of Agronomy and Plant Genetics, Univ. of Minnesota, 411 Borlaug Hall, 1991 Buford Cir., St. Paul, MN 55108
[*]Corresponding Author: PH: (701) 231-8478, Email: Mohamed.mergoum@ndsu.edu

OBJECTIVES

The main objective of this research project is develop new improved hard red spring wheat (HRSW) cultivar with resistance to Fusarium Head Blight (FHB) disease and superior grain yield and bread-making quality.

INTRODUCTION

For decades, scab or FHB, caused mainly by *Fusarium graminearum Schwabe* [telomorph *Gibberella zeae* (Schwein.)], has been a serious threat to wheat production throughout the world (Schroeder and Christenson, 1963; Bai and Shaner, 1994; McMullen et al., 1997; Stack, 2003). In North America and particularly in the US spring wheat region (North Dakota and neighboring states), FHB has been a major disease for HRSW produced since 1993 (Bai and Shaner, 1994; McMullen et al., 1997). Some economic reports (Nganje et al., 2004) estimate combined direct and secondary economic losses caused by FHB for all crops were at $7.7 billion. ND and MN account for about 68% ($5.2 billion) of the total dollar losses. Direct losses from 1993 through 2001 for wheat only were estimated to $2.492 billion (Nganje et al., 2004). The use of genetically resistant cultivars is believed to be the most efficient and economical method of controlling this FHB in wheat. This has been demonstrated in ND since 2002 when 'Alsen' (Frohberg et al., 2006), a moderate FHB resistance cultivar derived from the Chinese source 'Sumai 3' (PI 481542), was released by NDSU (with the support of the scab initiative funds). Sumai3, a spring wheat from China, is arguably the most used source of resistance to FHB in the world. 'Alsen' was the leading cultivar in the spring region between 2002 and 2006. Up to 2.4 million acres (37.4% of ND wheat acreages) was grown to 'Alsen' (N.D. Agricultural Statistics Service, USDA. 2006). A similar scenario was repeated with 'Glenn' (Mergoum et al., 2006a), a 2005 NDSU release which dominated the HRSW wheat region from 2007 to 2011 (N.D. Agricultural Statistics Service, USDA. 2012). The rapid increase in acreage planted to 'Alsen', Glenn, and other HRSW cultivars such 'Barlow' (Mergoum et al., 2011), indicates the desire of ND wheat growers to produce such HRSW cultivars with FHB resistance.

MATERIAL AND METHODS

The HRSW cultivar Prosper a sister line of 'Faller' (Mergoum et al., 2008) was developed using a modified bulk breeding procedure. It was selected from the "ND2857/ND2814" cross made at NDSU in the fall of 1997. ND2857 (ND2709/ ND688) is a hard red spring experimental line

that has good resistance to FHB originating from ND2709 line derived from a cross involving Sumai3. Both ND2709 and ND688 are HRSW experimental lines developed by the NDSU breeding program. ND2814 ('KITT' (PI 518818)/'AMIDON' (PI 527682)//'GRANDIN' (PI 531005) /'STOA S' (PI 520297)) is a HRSW line developed by NDSU HRSW breeding program. Kitt is HRSW cultivar released in1975 by the Minnesota Agricultural Experiment Station and the USDA-ARS while Amidon, Grandin and Stoa are HRSW cultivars released by NDAES in 1988, 1989, 1984, respectively. Prosper was selected from a bulk of one purified $F_{5:6}$ row-plot planted in 2001 at Christchurch, NZ. Prosper was tested in the breeding yield trials in many locations in ND from 2001 to 2004. Subsequently, Prosper was tested as ND 808 in the North Dakota Variety Trials (NDVT) from 2005 to 2010 and in the HRSW Uniform Regional Nursery (URN) in 2009 and 2010. The URN is conducted in various location in the US and Canadian spring wheat region. The first seed increase of Prosper was grown in Prosper, ND in the summer of 2009. Prosper was tested for its reaction to FHB and different races of tan spot, leaf and stem rusts, SNB, and STB under greenhouse and field conditions during the period of 2004 to 2010. The SNB, STB and tan spot are the major components of the leaf spotting disease complex of wheat in North America. A complex of these diseases occurs in nature. Hence managing leaf spots is difficult; however, resistant cultivars are the most effective and economical means of controlling leaf spot.

RESULTS

Prosper (ND 808) was released because it combines very high yield (Tables 1 and 2), resistance to FHB and leaf diseases (Table 3), and good end-use quality (Table 4). Prosper was named after the small town of Prosper, which is located in Cass County, ND, where the NDSU HRSW breeding program conducts its main breeding activities.

Grain yield and other agronomic traits were based on as many as 63 location-years of testing in the NDVT (Table 1). Based on data from these trials, grain yield of Prosper (4415 kg ha^{-1}) was similar to that of Barlow (4232 kg ha^{-1}), Faller (4409 kg ha^{-1}), 'Howard' (Mergoum et al., 2006b) (4214 kg ha^{-1}), and 'Steele-ND' (Mergoum et al., 2005b) (4110 kg ha^{-1}). However, Prosper yielded significantly ($p < 0.05$) more than several previously released NDSU cultivars, including 'Alsen' (3799 kg ha^{-1}), Glenn (4003 kg ha^{-1}), 'Dapps' (Mergoum et al., 2005a) (3852 kg ha^{-1}), 'Parshall' (PI 613587) (3633 kg ha^{-1}), and 'Reeder' (PI 613586) (3867 kg ha^{-1}) (Table 1). Yield data shows that Prosper is more adapted to eastern North Dakota (5389 kg ha^{-1}) compared with its yield (3897 kg ha^{-1}) in western North Dakota (Table 1). In the URN trials conducted in 2009 and 2010 (28 location-years), Prosper had a yield of 4773 kg ha^{-1}, which was significantly ($P < 0.05$) higher than all other cheeks, including 'Keene' (PI 598224) (3985 kg ha^{-1}), 'Verde' (PI 592561) (4287 kg ha^{-1}), and 'Chris' (CItr 13751) (3104 kg ha^{-1}) (Table 2). The performance of Prosper in the URN as compared to other HRSW cultivars for other agronomic traits is reported in Table 2.

Artificial inoculation with FHB disease was used to cause intense disease pressure. The average severity (Stack and Frohberg, 2000) recorded for Prosper in the field scab nursery (23%) was significantly lower than that for the very susceptible check '2398' (70%) (Table 3). In the same trials, the average FHB severity of the best FHB resistant grown checks 'Alsen' and Glenn were 22 and 25%, respectively while ND2710 (PI 633976), the most resistant elite line was 13%. Under artificial greenhouse and artificial inoculation conditions (data not shown), the average FHB severity of Prosper was 33%, which was similar to the scores of 'Alsen' (28%) and Glenn (30%); and significantly lower than the 87% and 71% registered for the susceptible checks 2398 and Reeder. Field testing and screening tests of seedlings and adult-plants conducted under greenhouse conditions from 2004 to 2010 showed that Prosper is resistant to the pathotypes of the predominant race of leaf rust in the region. Gene postulation shows that Prosper possesses Lr21, which confers resistance

to the major races of leaf rust in the spring region. Recently, however, a new race that has overcome Lr21 was observed in Minnesota and North Dakota (Table 3). Prosper is also resistant to the stem rust races TPMK, TMLK, RTQQ, QFCQ, and QTHJ under field and greenhouse conditions (Table 3). Prosper was screened for tan spot, STB, and SNB under greenhouse conditions. Prosper had an average scores of 2.2, 2.2, and 3.6 for tan spot races 2, 3, and 5, respectively, while 'Alsen' scored 3.5, 1.9, and 2.0 for the same races (Table 3). In the same screening tests, the reactions to the races 2, 3, and 5 of the check 'Salamouni' (PI 182673) were 1.4, 1.4, and 1.3 and of the check 'Glenlea' (CItr 17272) were 4.3, 2.0, and 1.9. Salamouni is considered among the best sources of resistance to tan spot, whereas Glenlea is usually used as the susceptible check to race 2. The scores for the reaction of Prosper to STB and SNB were 2.2 and 2.4, while 'Alsen' had scores of 2.7 and 4.4; Salamouni, 1.7 and 1.7; and Glenlea, 2.4 and 3.7 (Table 3).

The critical quality parameter including Falling number, Flour extraction, dough and baking parameters for Prosper and checks included in the HRSW-VT grown in North Dakota from 2004 to 2010 are reported in Table 4. The falling number of Prosper (414 s) was not significantly different than that of the most commonly grown HRSW cultivars, including Howard (422 s), Glenn (394 s), Steele-ND (432 s), 'Alsen' (414 s), and Reeder (430 s) (Table 4). Similarly, the flour extraction value (Table 4) of Prosper (709 g/kg) was similar to that of Howard (696 g/kg) and Steele-ND (692 g/kg); however, it was significantly superior to that of Glenn (680 g/kg), 'Alsen' (688 g/kg), and Reeder (684 g/kg). Mixing peak time of Prosper was 8.2 min, not significantly different from the checks, except Glenn (10 min) and 'Alsen' (10.1 min). The mixing tolerance score (14.5 min) was significantly shorter than for Glenn (20.4 min) but comparable with the scores of rest of the checks (Table 4). Bread loaf volume produced from Prosper (1000 mL) was comparable with that of all of the checks, except for Glenn (1056 mL) (Table 4). Similarly, the water absorption of Prosper (64.4%) was not significantly different from that of the checks, except for Steele-ND (66.4%) (Table 4).

AKNOWLEDGEMENTS AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-066. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

The authors thank the NDSU Research and Extension Center for all logistics to conduct the yield trials.

REFERENCES

Anderson, J.A., R.W. Stack, S. Liu, B.L. Waldron, A.D. Fjeld, C. Coyne, B. Moreno-Sevilla, J.M. Fetch, Q.J. Song, P.B. Cregan, and R.C. Frohberg. 2001. DNA markers for Fusarium head blight resistance QTL in two wheat populations. Theor. Appl. Genet. 102:1164-1168.

Bai, G.H., and G. Shaner. 1994. Scab of wheat: Prospects for control. Plant Disease. 78: 760-766.

Frohberg, R.C., R.W. Stack, and M. Mergoum. 2006. Registration of 'Alsen' wheat. Crop Sci. 46:2311–2312. doi:10.2135/cropsci2005.12.0501

McMullen, M. P., R. Jones, and D. Gallenberg. 1997. Scab of wheat and barley: A re-emerging disease of devastating impact. Plant Disease 81:1340-1348.

Mergoum, M., R.C. Frohberg, J.D. Miller, T. Olson, and J.B. Rasmussen. 2005a. Registration of 'Dapps' wheat. Crop Sci. 45:420–421. doi:10.2135/cropsci2005.0420

Mergoum, M., R.C. Frohberg, J.D. Miller, and R.W. Stack. 2005b. Registration of 'Steele-ND' wheat. Crop Sci. 45:1163–1164. doi:10.2135/cropsci2004.308CV

Mergoum, M., R.C. Frohberg, T. Olson, T.L. Friesen, J.B. Rasmussen, and R.W. Stack. 2006a. Registration of 'Glenn' wheat. Crop Sci. 46:473–474. doi:10.2135/cropsci2005.0287

Mergoum, M., R.C. Frohberg, T. Olson, T.L. Friesen, J.B. Rasmussen, and R.W. Stack. 2006b. Registration of 'Howard' wheat. Crop Sci. 46:2702–2703. doi:10.2135/cropsci2006.03.0185

Mergoum, M., R.C. Frohberg, T. Olson, T.L. Friesen, J.B. Rasmussen, and R.W. Stack. 2008. Registration of 'Faller' spring wheat. J. Plant Reg. 2:224–229. doi:10.3198/jpr2008.03.0166crc

Mergoum, M., R. C. Frohberg, T. L. Friesen, J. B. Rasmussen, G. Harland, and S. Simsek. 2011. 'Barlow': A High Quality and Yielding Hard Red Spring Wheat Cultivar Adapted to the North-Central Plains of the USA. Journal of plant Registration 5: 62-67.

N.D. Agricultural Statistics Service, USDA. 2006. North Dakota wheat varieties.

N.D. Agricultural Statistics Service, USDA. 2012. North Dakota wheat varieties.

Nganje W. E., S. Kaitibie, W. W. Wilson, F. L. Leistritz, and D. A. Bangsund. 2004. Economic Impacts of Fusarium Head Blight in Wheat and Barley: 1993-2001. Agribusiness and Applied Economics Report No 538.

Schroeder, H. W., and J. J. Christensen. 1963. Factors affecting resistance of wheat to scab caused by *Gibberella zeae*. Phytopathology 53: 831-838.

Stack, R. W. 2003. History of Fusarium head blight with emphasis on North America. P. 1-34. In K. J. Leonard and W. R. Bushnell (ed.) Fusarium head blight of wheat and barley. APS Press, St Paul, MN.

Stack, R.W., and R.C. Frohberg. 2000. Inheritance of resistance to Fusarium head blight in spring wheat F-1 hybrids. In: Proceedings of the International Symposium on Wheat Improvement for Scab, Nanjing, China. 5–10 May 2000. p. 94–97.

Table 1. Summary of agronomic data for Prosper and check cultivars tested in the North Dakota Hard Red Spring Wheat Variety Trials, 2005 to 2010.

	Grain yield								
Cultivar	North Dakota	Eastern North Dakota	Western North Dakota	Grain protein	1000-kernel weight	Grain volume weight	Heading date	Height	Straw strength
		kg ha^{-1}		%	g	kg m^{-3}	d after June 1st	cm	0–9†
Prosper	4415	5389	3897	14.2	33.5	763	61.6	83	1.5
Barlow	4232	4938	3857	14.8	31.6	782	58.4	85	1.5
Faller	4409	5310	3930	14.1	32.9	758	61.3	83	1.6
Glenn	4003	4630	3671	15.0	31.5	802	57.9	87	1.3
Howard	4214	5026	3783	14.5	31.7	778	59.7	85	2.0
Steele-ND	4110	4804	3742	14.8	32.2	777	59.5	85	2.2
Dapps	3852	4472	3340	16.1	29.7	756	58.2	91	1.9
Alsen	3799	4325	3577	15.2	30.5	774	59.8	81	1.3
Parshall	3633	4443	3143	15.2	27.3	774	59.4	90	1.1
Reeder	3867	4485	3671	14.9	30.9	768	60.9	81	0.8
LSD (0.05)	209	157	260	1.1	3.1	22	1.7	3.3	.9
No. of environments	63	22	41	61	29	63	98	63	35

† 0 = completely erect; 9 = completely flat at harvest.

Table 2. Summary of agronomic data for Prosper and check cultivars tested in the Hard Red Spring Wheat Uniform Regio Nursery, 2009 and 2010.

Cultivar	Grain yield	Grain volume weight	Grain protein	Heading	Height	Straw strength
	kg ha^{-1}	kg m^{-3}	g kg^{-1}	d after June 1st	cm	0–9†
Prosper	4773	770	14.3	30.5	87	1.0
Verde	4287	765	14.0	28.2	83	0.7
2375	4178	771	14.0	26.9	85	2.2
Keene	3985	776	14.3	27.5	98	2.1
Chris	3104	753	15.0	29.5	102	5.1
Marquis	3048	758	14.4	30.3	104	3.9
LSD (0.05)	432	11	0.8	1.1	3.3	1.0
No. of environments	28	28	28	28	28	28

†0 = completely erect; 9 = completely flat at harvest.

Table 3. Disease reactions of Prosper and check cultivars tested in the North Dakota Hard Red Spring Wheat Variety Trials between 2004 and 2010.

		Leaf rust		Stem rust		Tan spot				
Cultivar	FHB† Severity	Green house‡	Field	Green house§	Field	Race 2	Race 3	Race 5	Septoria blotch	Stagonospora blotch
	%						1–5¶			
Prosper	23	R/MR#	R/S	R-MR	R–MR	2.2	2.2	3.6	2.2	2.4
Alsen	22	R	MR/MS	R-MR	tR	3.5	1.9	2.0	2.7	4.4
Glenn	25	R	R	R-MR	R	–	–	–	–	–
Traverse	–	R	MR/MS	R	R	3.6	3.1	3.2	2.9	2.6
Knudson	–	–	R	R	R	1.6	1.6	1.8	2.2	1.6
Reeder	–	R	S	MR/R	5R	2.5	1.5	3.9	2.9	2.2
Steele-ND	–	R	R	R	R	2.1	2.0	4.0	2.7	4.0
2398	70	R	R	R	R	–	–	–	–	–
2710	13	R	R	R	R	–	–	–	–	–
Baart	–	–	S	–	50S	–		–	–	–
Glenlea	–	–	–	–	–	4.3	2.0	1.9	2.4	3.7
Salamouni	–	–	–	–	–	1.4	1.4	1.3	1.7	1.7
No. of environments	11	9	5	4	9	6	6	6	4	4

† FHB, Fusarium head blight ;% severity scored on 10 spikes (Stack and Frohberg (2000).
‡ Greenhouse reactions for leaf rust races MCDL and THBJ.
§ Greenhouse reactions for *P. graminis* f. sp. *tritici* races TPMK, TMLK, RTQQ, QFCQ, and QTHJ.
¶ 1 = resistant ; 5 = susceptible ; (Lamari and Bernier, 1989).
#R, resistant; MR, moderately resistant; MS, moderately susceptible; S, susceptible; TR, trace/resistant; 5R, resistant with 5% disease severity; 50MS, moderately susceptible with 50% disease severity.

Table 4. Quality parameters for Prosper and check cultivars tested in the North Dakota Hard Red Spring Wheat Variety Trials, 2005 to 2010.

Cultivar	Falling number	Flour extraction	Peak time	Mixing tolerance	Loaf volume	Water absorption
	s	g kg^{-1}		min	mL	%
Prosper	414†	709	8.2	14.5	1000	64.4
Howard	422	696	8.2	15.0	1006	66.1
Glenn	394	680	10.0	20.4	1056	65.3
Steele-ND	432	692	8.1	14.1	1015	66.4
Alsen	414	688	10.1	17.0	1018	65.3
Reeder	430	684	6.9	12.1	979	64.3
LSD (0.05)	28	19	1.7	3.7	22	1.3
No. environments	35	35	35	35	35	35

FIELD EVALUATION OF EXOTIC FUSARIUM HEAD BLIGHT RESISTANCE QTL IN SOFT RED WINTER WHEAT

Daniela Miller[1], Gina Brown-Guedira[2] and Jose Costa[1*]

[1]University of Maryland, PSLA Dept. 2102 Plant Sciences Bldg., College Park;
and [2]USDA-ARS, Plant Science Research Unit, Raleigh, NC 27695
*Corresponding Author: PH: (301) 405-1317, Email: costaj@umd.edu

ABSTRACT

Fusarium Head Blight (FHB), caused by the fungus *Fusarium graminearum*, is a devastating disease of wheat and other cereals that colonizes the grain directly. It results in severe yield losses and accumulation of the deoxynivalenol (DON) toxin, which may leave the grain unfit for human consumption. Breeding for disease resistance is the most efficient and sustainable method of controlling FHB. Ning7840, a Chinese spring wheat cultivar, has high resistance to FHB, derived from at least three resistance QTL located on chromosomes 3B, 2D, and 5A respectively. Two of these exotic resistance QTL from Ning7840 (on 3B and 2D) were introduced into the locally-adapted soft red winter wheat cultivar McCormick by backcrossing with a final cross with the highly susceptible cultivar SS8641. F5-derived progenies were developed by self-pollination. During each selfing generation, progeny were selected based on the presence of resistance alleles on SSR markers Umn10, cfd79, and Xgm533 (on 3B) and gwm539 and cfd233 (on 2D), and for a high background of McCormick and SS8641 alleles using markers spread throughout the genome. The current population is comprised of F5-derived F6 recombinant lines with a near isogenic background and differing presence of resistance alleles flanking both QTL. The present study sought to see if there were any significant differences in phenotypic expression between lines with different combinations of QTL-flanking alleles. Disease resistance was evaluated through an FHB-inoculated and misted field study conducted in Salisbury (MD). Scab incidence (Inc), scab severity (Sev), scab index, Fusarium damaged kernels (FDK), 1000-kernel weight and deoxynivalenol (DON) were analyzed using Proc GLM in SAS. It was found that lines with resistance alleles flanking both sides of the 2D QTL and one side of the 3BS showed lower FDK than other recombinant lines as well as Ning7840 itself. This research is the first stage of a larger study which seeks to fine-map the 3B and 2D QTL by correlating phenotypic resistance to the presence/absence of a high density of molecular markers flanking the QTL.

TRENDS IN FHB RESISTANCE AMONG WHEAT CULTIVARS IN ARKANSAS AND ENTRIES IN THE USSRWWSN: 2008-2012

Gene Milus*, David Moon and Peter Rohman

Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701
*Corresponding Author: PH: 479-575-2676, Email: gmilus@uark.edu

ABSTRACT

Developing cultivars with resistance to FHB and evaluating existing cultivars for FHB resistance have been important objectives of wheat breeding programs funded by the Scab Initiative. Advanced breeding lines that are adapted to the southern soft red winter wheat region are evaluated in the Uniform Southern Soft Red Winter Wheat Scab Nursery (USSRWWSN) that is coordinated through the Scab Initiative. Cultivars grown on at least 5% of the acreage in states with public wheat breeding programs funded by the Scab Initiative are evaluated for FHB resistance so that the FHB reactions can be made available to growers, extension personnel, and consultants. The objective of this report is to compare the trends in FHB resistance and DON level among entries in the USSRWWSN and cultivars grown in Arkansas.

Entries in the USSRWWSN and 20 commonly-grown and promising replacement cultivars (picked by Jason Kelley, University of Arkansas Extension Wheat Agronomist) were evaluated annually in inoculated, misted FHB nurseries at two locations since 2008. The experimental design at each location was a non-randomized complete block with three replications. Coker 9835 and Bess were included in each nursery as susceptible and resistant checks, respectively. Individual plots were two 1-m-long rows. The percentage of florets blighted was estimated visually at soft dough stage as a measure of FHB severity. Both rows of all plots were harvested at maturity and threshed using low air flow to retain scabby grain. Grain samples were lightly cleaned, evaluated for the percentage of *Fusarium* damaged kernels (FDK) by comparing samples to a set of known standards, and sent to the mycotoxin lab at the University of Minnesota for DON analysis. The distribution of mean values of FHB severity, FDK and DON for USSRWWSN entries and Arkansas cultivars in each environment (year-location) were plotted using box plots to allow visual comparisons among all of the data.

For FHB severity, FDK and DON, the distributions of Arkansas cultivars and USSRWWSN entries were similar in each environment except that the distributions for USSRWWSN entries were more skewed toward greater susceptibility than the distributions for Arkansas cultivars. Arkansas cultivars almost always had lower values than Coker 9835, whereas the USSRWWSN always had entries with values greater than Coker 9835. Some entries had DON values more than double the value for Coker 9835. Only a few entries and cultivars had values that were lower than the values for Bess. The results of this study indicate there has been a consistent trend over the past 5 years for Arkansas cultivars to be slightly more resistant than entries in the USSRWWSN.

THE 2011-12 SOUTHERN UNIFORM SOFT RED WINTER WHEAT SCAB NURSERY

J.P. Murphy* and R.A. Navarro

Department of Crop Science, Box 7629, North Carolina State University, Raleigh, NC 27695
*Corresponding Author: PH: (919) 513-0000, Email: paul_murphy@ncsu.edu

ABSTRACT

The Southern Uniform Winter Wheat Scab Nursery provides breeders in the public and private sectors with an opportunity for multi-environment, independent evaluations of FHB resistance in advanced generation breeding lines compared with the current most resistant check varieties Ernie, Bess and Jamestown. Valuable data are provided on resistance to other important fungal and viral diseases, milling and baking quality and agronomic characteristics. In addition, genotypic analyses identify alleles present at numerous important loci. The nursery is the primary method to facilitate the sharing of the best resistant materials throughout the breeding community.

The 2011-12 nursery comprised 47 advanced generation breeding lines and four check cultivars, 'Ernie', 'Bess', 'Jamestown' (partially resistant) and 'Coker 9835' (susceptible). Eight U.S. public programs (Univ. of Arkansas, Univ. of Georgia, Univ. of Illinois, Louisiana State University, Univ. of Maryland, N.C. State Univ., VA Tech., and USDA-ARS), and one private company, Agripro-Coker, submitted entries. The nursery was distributed to 11 U.S., one Romanian, and one Hungarian cooperator for field, and/or greenhouse evaluations. In addition, three USDA-ARS laboratories conducted evaluations for Hessian fly resistance, milling and baking quality and marker genotypes based on established diagnostic markers.

Copies of the full report will be available at the 2012 National Fusarium Head Blight Forum and subsequently on line at the USWBSI web site: http://www.scabusa.org/.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-083. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

	Cultivar/ Designation	FHB Incidence	RANK	FHB Severity	RANK	FHB Index	RANK	FDK	RANK	ISK	RANK	DON	RANK
1	ERNIE	30	5	13	10	4	5	7	9	16	3	5	25
2	COKER 9835	76	50	49	49	35	49	40	49	49	49	15	49
3	BESS	21	1	12	5	2	1	4	2	11	1	3	5
4	JAMESTOWN	44	19	20	24	7	17	7	9	26	19	4	13
5	NC09-21916	34	9	24	29	5	8	6	5	23	9	3	5
6	VA08W-613	45	21	13	9	4	5	8	12	23	9	3	5
7	M08-8036#	45	22	23	27	7	17	4	2	18	5	3	5
8	IL02-18228	25	2	18	21	2	1	2	1	12	2	1	1
9	ARS07-1214	82	51	51	50	45	50	40	49	54	50	26	50
10	ARS09-173	45	26	33	43	12	35	15	34	32	36	9	42
11	ARS09-367	75	49	51	51	45	50	44	51	58	51	26	50
12	ARS09-446	57	44	38	46	22	45	27	46	48	48	11	47
13	ARS09-513	55	42	39	48	18	41	31	47	47	47	8	40
14	ARS09-595	56	43	38	45	18	41	25	45	40	43	6	32
15	ARS09-643	47	31	35	44	14	39	18	42	39	42	10	45
16	ARS09-724	46	29	27	33	11	33	8	12	29	28	4	13
17	GA 051173W-S11	39	13	28	35	9	25	13	31	27	21	9	42
18	GA 051207-S19	49	38	28	36	12	35	16	35	33	37	5	25
19	GA 051207-S21	46	28	29	38	10	28	8	12	29	28	5	25
20	GA 051173-S25	55	41	38	47	22	45	23	44	42	44	9	42
21	GA 051173-S18	36	11	22	26	5	8	11	25	25	17	4	13
22	GAMD08-27-E9-S13	47	33	25	31	8	21	12	28	30	31	4	13
23	GAMD08-27-E9-S14	47	32	13	8	6	13	8	12	23	9	4	13
24	GAMD08-27-E9-S15	45	25	11	4	8	21	11	25	28	25	4	13
25	LA'04039C-14-8	42	17	15	15	8	21	6	5	29	28	3	5
26	LA'04039C-10-6	42	15	17	18	6	13	9	17	25	17	4	13
27	LA05102C-1-2	32	7	16	16	7	17	6	5	27	21	1	1
28	LA05102C-8-8	33	8	9	3	4	5	5	4	24	14	2	3
29	LA05079D-55	57	45	25	30	25	48	17	40	45	46	7	35
30	LA05079F-P01	53	39	27	32	18	41	16	35	37	40	5	25
31	LA05079F-P03	49	37	19	22	10	28	16	35	30	31	8	40
32	LA05145D-12	58	46	30	40	16	40	16	35	35	39	7	35
33	LA05145D-24	63	48	31	42	23	47	31	48	42	44	11	47
34	MD03W61-11-2(11PW#108)	45	23	21	25	13	37	16	35	38	41	7	35
35	MD03W61-11-3(11PW#109)	46	27	17	19	10	28	9	17	28	25	5	25
36	UX0066-4-79(11PW#183)	41	14	14	12	6	13	10	22	20	6	4	13
37	MD08-22-1-6-4(11PW#189)	28	3	8	1	2	1	9	17	20	6	2	3
38	MD08-26-H2-23(11CVM-3)	35	10	12	7	5	8	7	9	22	8	3	5
39	MH07-7483	60	47	23	28	19	44	18	42	33	37	10	45
40	M09-9826#	42	16	14	14	6	13	17	40	30	31	4	13
41	NC08-23323	45	24	31	41	10	28	9	17	27	21	7	35
42	NC08-23324	54	40	28	37	9	25	12	28	30	31	7	35
43	NC09-22422 (Fhb1)	38	12	14	11	5	8	12	28	27	21	3	5
44	NC09-20986 (Fhb1)	28	4	8	2	3	4	8	12	17	4	3	5
45	NC8355-4 (Fhb1)	31	6	12	6	5	8	13	31	23	9	4	13
46	NC8452-2	47	34	14	13	7	17	10	22	23	9	6	32
47	VA09W-52	48	36	20	23	10	28	14	33	31	35	5	25
48	VA09W-73	46	30	28	34	13	37	9	17	28	25	6	32
49	VA09W-75	44	18	16	17	8	21	6	5	26	19	4	13
50	VA10W-42	47	35	17	20	9	25	10	22	24	14	5	25
51	VA10W-140	44	20	30	39	11	33	11	25	24	14	4	13
	Mean			24		12		14		30		6	
	LSD (0.05)			28		16		16		15		7	
	CV%			60.0		71.4		56.8		25.1		53.9	

	Cultivar/ Designation	Heading Date	RANK	Plant Height	RANK	Hessian Fly Biotype L	Stagon. nodorum % F'VILLE AR	Stripe Rust % N'PORT AR
1	ERNIE	108	9	30	13	0-15	7	68
2	COKER 9835	110	19	31	15	0-13	15	78
3	BESS	113	38	35	43	0-14	30	17
4	JAMESTOWN	106	3	30	8	0-16	15	0
5	NC09-21916	113	38	34	40	0-3	2	12
6	VA08W-613	108	9	32	18	0-18	2	37
7	M08-8036#	111	26	33	28	0-20	2	37
8	IL02-18228	116	43	37	49	0-16	15	7
9	ARS07-1214	116	43	33	30	0-16	15	43
10	ARS09-173	106	3	30	5	21-0	15	1
11	ARS09-367	113	38	30	14	0-17	30	48
12	ARS09-446	108	9	33	32	0-14	30	25
13	ARS09-513	110	19	32	21	0-14	15	68
14	ARS09-595	107	6	32	22	0-17	7	57
15	ARS09-643	106	3	21	1	0-15	15	0
16	ARS09-724	110	19	36	47	0-14	7	1
17	GA 051173W-S11	112	33	40	51	15-3	15	2
18	GA 051207-S19	111	26	34	38	14-0	15	15
19	GA 051207-S21	110	19	35	44	17-0	15	12
20	GA 051173-S25	112	33	34	37	13-2	15	0
21	GA 051173-S18	105	1	30	9	22-0	7	1
22	GAMD08-27-E9-S13	121	49	35	46	0-13	2	20
23	GAMD08-27-E9-S14	121	49	34	35	0-18	2	1
24	GAMD08-27-E9-S15	121	49	33	25	0-16	7	15
25	LA'04039C-14-8	107	6	30	6	0-16	2	57
26	LA'04039C-10-6	110	19	35	45	0-17	2	25
27	LA05102C-1-2	105	1	34	33	0-15	7	1
28	LA05102C-8-8	108	9	35	42	0-16	30	1
29	LA05079D-55	109	17	31	17	0-18	15	0
30	LA05079F-P01	108	9	32	23	0-16	15	0
31	LA05079F-P03	110	19	28	2	0-16	15	0
32	LA05145D-12	107	6	30	7	0-17	15	2
33	LA05145D-24	116	43	39	50	0-17	7	70
34	MD03W61-11-2(11PW#108)	111	26	30	10	0-15	7	0
35	MD03W61-11-3(11PW#109)	112	33	33	26	0-17	2	0
36	UX0066-4-79(11PW#183)	111	26	32	19	0-18	15	57
37	MD08-22-1-6-4(11PW#189)	118	48	34	39	0-19	2	80
38	MD08-26-H2-23(11CVM-3)	111	26	35	41	0-17	15	12
39	MH07-7483	116	43	36	48	0-16	30	0
40	M09-9826#	108	9	29	4	0-20	7	78
41	NC08-23323	111	26	32	20	0-13	7	89
42	NC08-23324	112	33	33	31	0-20	7	94
43	NC09-22422 (Fhb1)	111	26	30	11	20-0	7	20
44	NC09-20986 (Fhb1)	110	19	31	16	16-0	7	10
45	NC8355-4 (Fhb1)	108	9	29	3	0-20	7	6
46	NC8452-2	116	43	33	27	0-16	7	11
47	VA09W-52	109	17	30	12	0-23	30	7
48	VA09W-73	113	38	33	29	0-14	7	0
49	VA09W-75	108	9	34	34	0-17	2	3
50	VA10W-42	112	33	34	36	0-18	2	0
51	VA10W-140	113	38	33	24	0-18	15	71
	Mean	112		32			11	25
	LSD (0.05)	6		7			.	.
	CV%	2.5		10.5			.	.

	DESIGNATION	Rht-B1b	Rht-D1b	Rht8	Ppd-D1a	Vrn-A1	Lr34/Yr18	Lr37/Yr17	Sr36	Sr24/Lr24	Sr2	Lr9	Qyr.uga-2AS	Fhb1	Fhb 5A ERNIE
1	ERNIE	het	no	no	het	het	no	no	yes	no	no	no	no	no	yes
2	COKER 9835	no	yes	no	yes	vrn-A1b	no	no	yes	no	no	yes	no	no	no
3	BESS	yes	no	no	no	vrn-A1b	no	no	no	no	no	no	no	no	no
4	JAMESTOWN	no	yes	no	yes	vrn-A1a	no	no	no	no	no	no	no	no	no
5	NC09-21916	no	yes	no	yes	vrn-A1b	no	no	no	no	no	no	no	no	no
6	VA08W-613	no	yes	no	yes	vrn-A1a	no	no	no	no	no	yes	no	no	no
7	M08-8036#	no	no	no	yes	vrn-A1b	no	no	yes	no	no	yes	no	no	no
8	IL02-18228	yes	het	no	no	vrn-A1b	no	no	no	no	no	no	no	no	no
9	ARS07-1214	yes	no	no	no	vrn-A1b	no	no	no	yes	no	no	no	no	no
10	ARS09-173	yes	no	no	yes	vrn-A1b	no	yes	no	no	no	no	no	no	no
11	ARS09-367	yes	no	no	no	vrn-A1b	no	no	no	yes	no	no	no	no	no
12	ARS09-446	no	no	no	no	vrn-A1b	no	no	yes	no	no	no	no	no	no
13	ARS09-513	no	no	het	yes	vrn-A1b	no	no	het	no	no	no	no	no	no
14	ARS09-595	no	no	no	yes	vrn-A1a	no	no	yes	no	no	no	no	no	no
15	ARS09-643	no	yes	no	yes	vrn-A1b	no	het	no	no	no	no	no	no	no
16	ARS09-724	yes	no	no	yes	vrn-A1b	no	yes	no	no	no	no	no	no	no
17	GA 051173W-S11	no	yes	no	het	vrn-A1b	no	yes	no	no	no	no	no	no	no
18	GA 051207-S19	no	yes	no	yes	vrn-A1b	no	yes	no	no	no	no	no	no	no
19	GA 051207-S21	no	yes	no	yes	vrn-A1b	no	yes		no	no	no	no	no	no
20	GA 051173-S25	yes	no	no	yes	vrn-A1b	no	yes	no	no	no	no	no	no	no
21	GA 051173-S18	no	yes	no	yes	vrn-A1b	no	yes	no	no	no	no	no	no	no
22	GAMD08-27-E9-S13	no	yes	no	no	vrn-A1b	no	het	no	no	no	no	no	yes	no
23	GAMD08-27-E9-S14	no	yes	no	no	vrn-A1b	no	yes	no	no	no	no	no	yes	no
24	GAMD08-27-E9-S15	no	yes	no	no	vrn-A1b	no	het	no	no	no	no	no	yes	no
25	LA'04039C-14-8	no	no	no	het	vrn-A1b	no	no	yes	no	no	yes	no	no	no
26	LA'04039C-10-6	no	no	no	het	vrn-A1b	no	no	yes	yes	no	no	yes	no	no
27	LA05102C-1-2	no	no	no	yes	het	no	no	yes	no	no	no	yes	no	no
28	LA05102C-8-8	no	no	no	yes	vrn-A1b	no	no	yes	no	no	no	no	no	no
29	LA05079D-55	yes	no	no	no	vrn-A1b	no	no	no	no	no	no	no	no	no
30	LA05079F-P01	yes	no	no	no	vrn-A1b	no	no	no	no	no	yes	no	no	no
31	LA05079F-P03	yes	no	no	het	vrn-A1b	no	no	yes	no	no	yes	no	no	no
32	LA05145D-12	no	yes	no	yes	vrn-A1a	no	no	yes	no	no	no	no	no	no
33	LA05145D-24	no	yes	no	yes	vrn-A1a	no	no	no	no	no	yes	no	no	no
34	MD03W61-11-2(11PW	no	yes		yes	vrn-A1b	no	no	no	no	no	no	no	no	
35	MD03W61-11-3(11PW	no	yes	no	yes	vrn-A1b	no	no	yes	no	no	no	no	no	no
36	UX0066-4-79(11PW#1	no	yes	no	yes	vrn-A1b	no	no	yes	no	no	no	yes		het
37	MD08-22-1-6-4(11PW#	no	yes	no	no	vrn-A1b	no	no	no	yes	no	no	no	yes	no
38	MD08-26-H2-23(11CVI	no	yes	no	yes	vrn-A1b	no	yes	no	yes	no	no	no	yes	no
39	MH07-7483	yes	no	no	yes	vrn-A1b	no	no	no	no	no	no	no	no	no
40	M09-9826#	yes	no	no	het	vrn-A1b	no	no	yes	no	no	yes	no	no	no
41	NC08-23323	no	yes	no	yes	vrn-A1b	no	no	no	no	no	yes	no	no	no
42	NC08-23324	no	yes	no	het	vrn-A1b	no	no	no	no	no	yes	no	no	no
43	NC09-22422 (Fhb1)	het	no	no	het	vrn-A1b	no	yes	no	no	no	no	no	yes	no
44	NC09-20986 (Fhb1)	yes	no	no	het	vrn-A1b	no	yes	no	no	no	no	no	yes	no
45	NC8355-4 (Fhb1)	no	yes	no	yes	vrn-A1a	no	no	yes	no	no	no	no	yes	no
46	NC8452-2	no	yes	no	yes	vrn-A1b	no	no	het	yes	no	yes	het	no	het
47	VA09W-52	no	yes	no	yes	vrn-A1a	no	no	no	no	no	no	no	no	no
48	VA09W-73	no	het	no	no	vrn-A1b	no	no	no	no	no	yes	no	no	no
49	VA09W-75	no	yes	no	het	vrn-A1b	no	no	no	no	no	yes	no	no	no
50	VA10W-42	yes	no	no	yes	vrn-A1b	no	no	no	no	no	no	no	no	no
51	VA10W-140	no	yes	no	yes	vrn-A1b	no	no	no	no	no	yes	no	no	no

	DESIGNATION	Fhb Ernie 3Bc	Fhb 5A Ning7840	Fhb 2DL-Wuhan1/W14	1RS:1AL	1RS:1BL	H13	H9	Bdv2/3	Sbm1	Bx7 over-expressing	Glu-A1	Glu-D1	TaSus2-2B
1	ERNIE		no	no	no	no	no	no	no	no	no	Ax1 or null	het	yes
2	COKER 9835	yes	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	yes
3	BESS	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	2+12	no
4	JAMESTOWN	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	no
5	NC09-21916	no	no	no	no	no	no	no	no	yes	no	Ax2*	5+10	no
6	VA08W-613	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	no
7	M08-8036#	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	yes
8	IL02-18228	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	5+10	no
9	ARS07-1214	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	2+12	no
10	ARS09-173	no	no	no	no	no	het	no	no	yes	no	Ax2*	2+12	no
11	ARS09-367	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	2+12	no
12	ARS09-446	no	no	no	no	no	no	no	no	no	no	Ax2*	5+10	yes
13	ARS09-513	no	no	no	no	no	no	no	no	no	no	Ax1 or null	2+12	het
14	ARS09-595	no	no	no	no	no	no	no	no	yes	no	Ax2*	5+10	yes
15	ARS09-643	no	no	no	yes	no	no	no	no	yes	het	Ax2*	2+12	no
16	ARS09-724	no	no	no	no	no	no	no	no	no	no	Ax1 or null	2+12	no
17	GA 051173W-S11	no	no	no	no	no	yes	no	no	yes	no	het	2+12	no
18	GA 051207-S19	no	no	no	no	no	no	yes	no	yes	no	Ax2*	5+10	no
19	GA 051207-S21	no	no	no	het	no	no	yes	no	yes	no	Ax2*	5+10	no
20	GA 051173-S25	no	no	no	no	no	yes	no	no	yes	no	Ax1 or null	2+12	no
21	GA 051173-S18	no	no	no	no	no	yes	no	no	yes	no	Ax1 or null	2+12	no
22	GAMD08-27-E9-S13	no	yes	no	yes	no	no	no	no	yes	no	Ax2*	2+12	no
23	GAMD08-27-E9-S14	no	yes	no	yes	no	no	no	no	yes	no	Ax2*	2+12	no
24	GAMD08-27-E9-S15	no	yes	no	yes	no	no	no	no	yes	no	Ax2*	2+12	no
25	LA'04039C-14-8	no	no	no	no	yes	no	no	no	no	no	Ax2*	5+10	yes
26	LA'04039C-10-6	no	no	no	no	yes	no	no	no	no	no	Ax2*	5+10	yes
27	LA05102C-1-2	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	yes
28	LA05102C-8-8	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	yes
29	LA05079D-55	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	2+12	no
30	LA05079F-P01	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	2+12	no
31	LA05079F-P03	no	no	no	no	no	no	no	no	het	yes	Ax2*	5+10	yes
32	LA05145D-12	no		no	no	yes	no	no	no	yes	het	Ax2*	5+10	yes
33	LA05145D-24	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	no
34	MD03W61-11-2(11PW#108)	no		no	no	no	no	no		yes	no	Ax2*	2+12	het
35	MD03W61-11-3(11PW#109)		no	no	no	no	no	no	no	yes	no	Ax2*	2+12	yes
36	UX0066-4-79(11PW#183)	no	no	no	no	no	no	no	no	yes	no	Ax2*	2+12	yes
37	MD08-22-1-6-4(11PW#189)	no	yes	no	yes	no	no	no	no	yes	no	Ax2*	2+12	no
38	MD08-26-H2-23(11CVM-3)	no	het	no	no	no	no	no	no	yes	no	Ax2*	2+12	no
39	MH07-7483	no	no	no	no	yes	no	no	no	yes	no	Ax1 or null	het	no
40	M09-9826#	no	no	no	no	no	no	no	no	yes	no	Ax2*	het	yes
41	NC08-23323	het?	no	no	het	no	no	no	no	yes	no	Ax1 or null	2+12	no
42	NC08-23324	no	no	no	yes	no	no	no	no	yes	no	Ax1 or null	2+12	no
43	NC09-22422 (Fhb1)	no	no	no	no	no	yes	no	no	yes	no	Ax2*	nknow	no
44	NC09-20986 (Fhb1)	no	no	no	no	no	het	no	no	yes	no	Ax2*	het	no
45	NC8355-4 (Fhb1)	no	no	no	no	no	no	no	no	yes	no	Ax1 or null	5+10	yes
46	NC8452-2	no	no	no	het	no	no	no	no	yes	no	Ax2*	2+12	het
47	VA09W-52	no	no	no	het	no	no	no	no	yes	no	Ax1 or null	2+12	no
48	VA09W-73	no	no	no	no	no	no	no	no	no	no	Ax2*	2+12	no
49	VA09W-75	no	no	no	yes	no	no	no	no	no	no	Ax2*	2+12	no
50	VA10W-42	no	no	no		no	no	no	no	yes	no	Ax2*	2+12	no
51	VA10W-140	no	no	no	no	no	no	no	no	no	no	Ax2*	5+10	no

BREEDING FOR FUSARIUM HEAD BLIGHT (FHB), RUST AND BARLEY YELLOW DWARF (BYD) RESISTANCE IN PURDUE SOFT WINTER WHEAT LINES 05247 AND 02444

Herb W. Ohm*

Department of Agronomy, Purdue University, 915 W. State Street, West Lafayette, IN 47907
*Corresponding Author: PH: (765) 494-8072, Email: hohm@purdue.edu

ABSTRACT

Fusarium head blight (FHB) of wheat, caused mainly by *Fusarium graminearum* Schwabe (telemorph *Gibberella zeae*), results in serious yield loss and vomitoxin (DON) contamination in wheat grain. In addition to FHB, other diseases such as rust and barley yellow dwarf (BYD) all cause reduced yield and lowered grain quality. Stacking various disease resistance genes in wheat cultivars is an important strategy to reduce production losses. The wheat breeding program at Purdue University aims at developing soft winter wheat lines with FHB resistance and other various disease resistances. Both 05247 and 02444 have high yield potential, producing 16317 kg ha^{-1} and 14536 kg ha^{-1}, respectively (compared with 13505 kg ha^{-1} for Branson) averaged two years (2011 and 2012) at Lafayette, IN. Both lines also have various disease resistances, such as FHB, BYD, HF and rust. As indicated by the FHB inoculation data from year 2010 and 2011, line 05247 and 02444 have good FHB type II resistance, with an average of 2.75 and 2.50 spikelets for disease spread, respectively. In year 2012, due to a dry and early season, no FHB disease had developed and a disease score of 0.5 for Type II resistance was seen for almost all of the lines. However, the environment was optimum for rust development. With disease readings of one for both stripe rust and leaf rust, line 05247 shows excellent rust resistance.

HIGH SPEED SORTING OF *FUSARIUM* DAMAGED WHEAT KERNELS

Tom Pearson[1*], Sven Halvorson[2] and Anthony Clark[3]

[1]USDA-ARS-CGAHR, Manhattan, KS; [2]University of Oregon, Department of Mathematics, Eugene, OR; and [3]University of Kentucky, Department of Plant and Soil Sciences, Lexington, KY
[*]Corresponding Author: PH: 785-776-2729, Email: thomas.pearson@ars.usda.gov

OBJECTIVES

To demonstrate feasibility of removing *Fusarium*-damaged wheat kernels from breeder samples using a low-cost, high-speed, sorting machine capable of measuring several visible and near infrared (NIR) spectral bands.

INTRODUCTION

Use of visible and NIR light for phenotypic sorting of single wheat kernels has been shown to be a useful tool to improve several quality traits (Dowell et al., 2009). One such application is to use NIR to distinguish single wheat kernels with high levels of deoxynivalenol (DON) from kernels having low DON levels using chemometric models implemented on the automated Perten single kernel NIR system (SKNIR). Additionally, the SKNIR has proven to be a useful tool to objectively separate kernels having low, intermediate, and high levels of *Fusarium* damaged kernels (FDK) (Peiris et al., 2010). This is important for breeding programs as it has been shown that resistance to Fusarium head blight (FHB) and accumulation of DON can be inherited (Buerstmayr et al., 1999, Miedaner et al., 2003). However, instrumentation for automatically and rapidly segregating single wheat kernels on the basis of NIR characteristics is expensive and no longer readily available. To fill this void, a simple light emitting diode (LED) based sorting machine was developed to rapidly measure light reflection within a limited number of visible and NIR spectral bands and sort kernels at a rate of about 20 kernels/s or 2.3 Kg/hr (Pearson et al., 2012). Compared with the SKNIR, the LED system is potentially much lower cost, ~$3000 vs ~$100,000, and the throughput is much higher, 20 kernels/s vs 0.5 kernels/s. The only significant drawback is that the LED sorter could have lower accuracy in some applications due to the limited number of spectral bands used.

MATERIALS AND METHODS

IIn the LED based sorting instrument developed by Pearson et al. (2012), visible and NIR reflectance data was acquired by rapidly blinking six different LEDs one at a time. For this study, the number of LEDs were increased to nine with peak emission wavelengths at: 1070nm, 1050nm, 970nm, 940nm, 880nm, 850nm, 624nm (red), 527nm (green), and 470nm (blue). Selection of LEDs was limited to those with a peak emission wavelength less than 1100nm due to the silicon based sensor used. An image of the LED circuit board illuminating a wheat kernel exiting the end of a chute is shown in Figure 1. Reflected light from the kernel is projected by the lens onto a photo-diode. The photo-diode signal is amplified and input to a micro-controller on the same circuit board which also controls the LEDs, performs all computations, classifies kernels based on reflected light from several of the LEDs, and outputs a digital signal to activate an air valve to divert kernels based on the classification.

Ten different samples of hard red spring (HRS) wheat and soft red winter (SRW) wheat were used to test the sorter. The varieties of HRS used were: Altona, Clearwater, Crystal, Cypress River, Grandin, Fairfax, Hollsus, Kane, Plum and Westroc. The incidence of FDK averaged 21.2% and ranged from 12.4% to 30.5%. Sorter performance can be affected by kernel size variation; the 1000 kernel weights of undamaged kernels averaged 32.7g and

ranged between 27.7g to 42.2g so these samples encompass a fairly broad range of sizes. The ten SRW samples were comprised of breeding population samples supplied by the University of Kentucky. The incidence of FDK averaged 11.8%, ranging from 6.3% to 19.6%. The 1000 kernel weights of undamaged SRW kernels averaged 37.7g, ranging between 32.6g to 39.3g.

While separate calibrations for the LED sorter were developed and used for SRW and HRS, the calibration procedure was the same. Two hundred undamaged kernels and 200 kernels classified as having an intermediate level of FDK were separated by the SKNIR instrument using the calibration developed by Peiris et al. (2010) and these kernels were used for calibrating the LED instrument. KY07C-1056 breeding population SRW and Clearwater variety HRS were used for the calibration samples. These calibration samples were then fed through the LED instrument and the responses from the nine LEDs for all kernels were saved to a personal computer. Letting each of the nine LED responses be represented by $\lambda 1 ... \lambda 9$, every possible unique combination of $(\lambda x - \lambda y)/\lambda z$ were computed except for cases where $\lambda x = \lambda y$. These difference ratios have the benefit of cancelling out background noise, fluctuations in illumination levels, and help characterize where differences in spectra occur. Stepwise discriminant analysis (Hintze, 2001) was then used to select a small subset of these ratios to be used in sorting after they were programmed back into the micro-controller for sorting.

The nine HRS and SRW wheat samples not used in the calibration were sorted by the LED sorter into three groups: (a) undamaged, (b) intermediate, and (3) FDK. While this sorter is only capable of performing two way classifications, the three groups were created by passing all kernels through the sorter twice and grouping the samples based on the number of times they were accepted or rejected. Samples that were accepted twice were called "undamaged", samples rejected once and accepted once comprised the "intermediate" group, and samples rejected twice were classified as "FDK"..

To determine the sorting accuracy of the LED sorter, the SKNIR was used to sort each of the three LED sorted groups. The SKNIR instrument sorted 1,000 kernels of each LED sort stream into three categories per the Peiris et al. (2010) calibration: (a) undamaged – Bin 1; (b) Intermediate – Bin 2, and (c) FDK – Bins 3 and 4. After completion, the kernel distribution was recorded and compared with the LED sorted classification.

RESULTS AND DISCUSSION

Table 1 displays the average agreement between the LED and SKNIR sorters for classifying HRS samples into the three FDK categories. Overall, after sorting HRS in the LED sorter twice, FDK concentration was reduced from an unsorted average of 21.2% to 4.8% in the undamaged group, a 77% reduction, while the false positive reject rate for undamaged kernels was 12%. The false positive error rate was computed by combining the weight percent of the LED sorted samples (Table 1) with the percentage of undamaged kernels in the intermediate and FDK categories. Of the HRS kernels that the LED sorter classified as undamaged, the SKNIR found that 4.8% were FDK (range: 2.5 to 6.5%). However, visual inspection of these indicated that less than 1/10 of these kernels show visual symptoms of severe FDK such the "tombstone" appearance. The LED sorter is able to distinguish many kernels with minor symptoms of FDK as 61% of the kernels classified as intermediate by the LED sorter were also classified as intermediate or FDK by the SKNIR. Undamaged kernels that were lighter in color, possibly due to weathering, tended to be classified as FDK or intermediate by the LED sorter but were classified as undamaged by the SKNIR. Therefore, results may vary depending on other environmental effects during the growing season.

As shown in Table 2, the LED sorter removed a greater percentage of FDK from SRW while maintaining similar false positive rates. Two passes through the LED sorter resulted in an 87% reduction of FDK compared the original unsorted

FDK concentration (11.8% unsorted to 1.5% sorted) with a false positive rate of 14.7%. Only 1.5% of the kernels classified as undamaged by the LED sorter were considered FDK by the SKNIR and none of these had the "tombstone" appearance. The fact that nearly half of the kernels classified as intermediate by the LED sorter were classified as intermediate or FDK by the SKNIR indicates that, as with HRS, the LED sorter was able to separate many kernels with minor symptoms of FDK from undamaged kernels.

The spectral band difference ratios selected during the calibration procedure for HRS were: (blue-green)/1070nm, (green-850nm)/970nm, and (red-850nm)/1070nm, (1050nm-1070nm)/1070nm; and for SRW: (blue-red)/red, (850nm-970nmnm)/970nm, and (blue-1070nm)/red. An additional study is currently underway to build and test a similar LED sorter circuit board that uses an InGaAs sensor so that LEDs in the range of 900 to 1700nm can be used. Delwiche and Harland (2004) showed that FDK could be detected with high accuracy using a few spectral bands in this region. Nevertheless, the results indicate that the sorter in its current form is able to remove FDK from breeder samples, making it a viable tool for use in developing Fusarium resistant wheat varieties. The LED sorter has also proven effective for separating red and white wheat as well as sorting kernels on the basis of protein content (Pearson et al., 2012), attesting to the versatility of this instrument.

ACKNOWLEDGEMENT AND DISCLAIMER

We thank Elizabeth Maghirang, Agricultural Engineer, USDA-ARS-CGAHR, for assistance with this work.

Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture. USDA is an equal opportunity provider and employer.

REFERENCES

Buerstmayr, H., Lemmens, M., Fedak, G., and Ruckenbauer, P. 1999. Back-cross reciprocal monosomic analysis of Fusarium head blight resistance in wheat (*Triticum aestivum* L.). Theor. Appl. Genet. 98:76-85.

Delwiche, S.R. and G.A. Hareland. 2004. Detection of scab-damaged hard red spring wheat kernels by near-infrared reflectance. *Cereal Chemistry*. 81(5):643-649.

Dowell, F.E., Maghirang, E.B., and Baenziger, P.S. 2009. Automated single-kernel sorting to select for quality traits in wheat breeding lines. *Cereal Chemistry*. 86(5):527-533.

Hintze,J. 2001. NCSS and PASS. Number Cruncher Statistical Systems. Kaysville, UT, USA. www.ncss.com

Miedaner, T., Schneider, B., and Geiger, H. H. 2003. Deoxynivalenol (DON) content and Fusarium head blight resistance in segregating populations of winter rye and winter wheat. Crop Sci. 43:519-526.

Pearson, T.C Maghirang, E.B., and Dowell, F.E. 2012. A multispectral sorting device for wheat kernels. *American Journal of Agricultural Science and Technology*, submitted September 2012. available at: https://www.sugarsync.com/pf/D6985352_8431431_6909673

Peiris, K.H.S., Pumphrey, M.O., Dong, Y., Maghirang, E.B., Berzonsky, W., and Dowell, F.E. 2010. Near-Infrared Spectroscopic method for identification of Fusarium Head Blight Damage and Prediction of Deoxynivalenol in Single Wheat Kernels. *Cereal Chemistry*. 87(6):511-517.

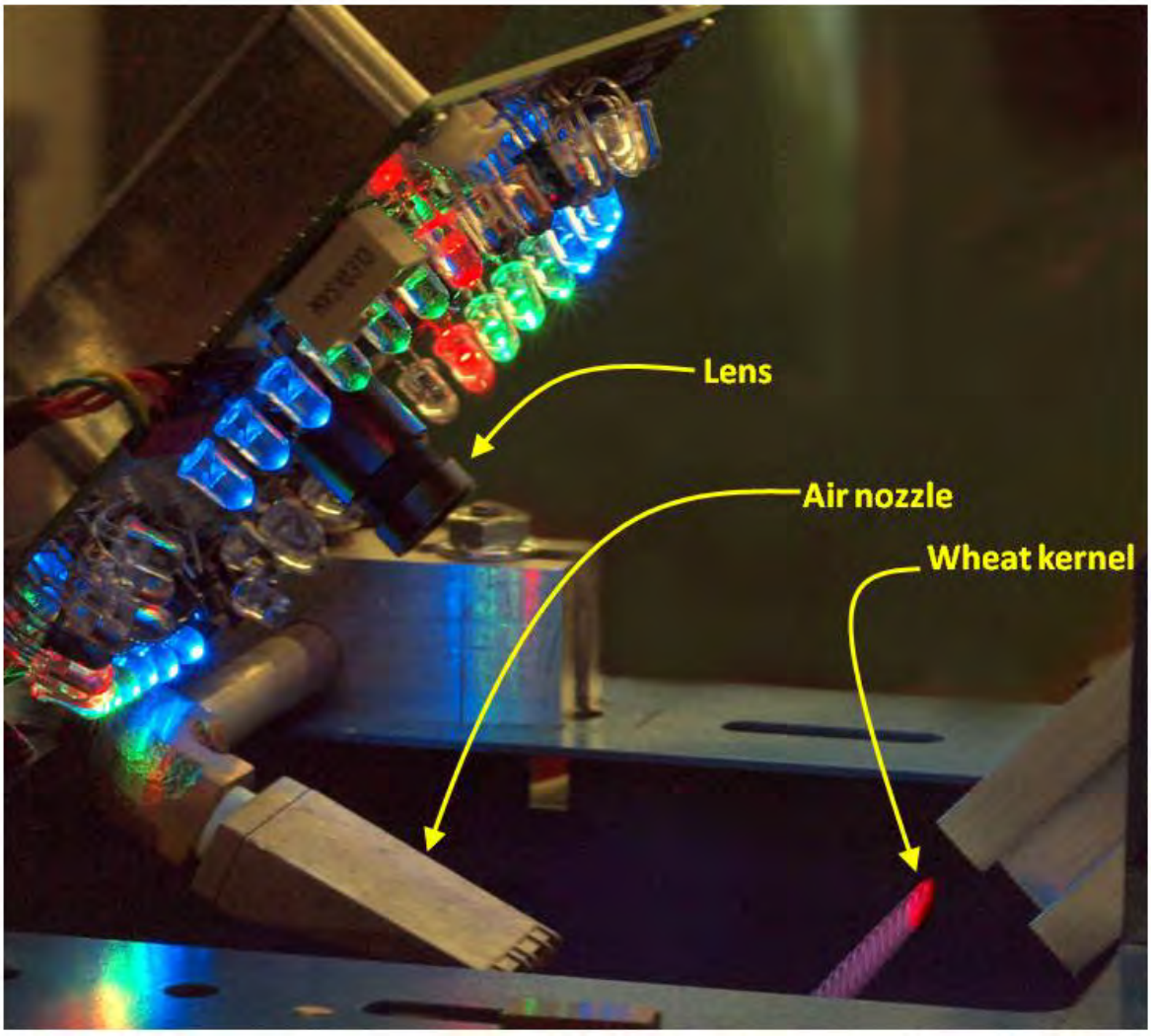

Figure 1. LEDs illuminating a wheat kernel as it falls off of the chute. The air nozzle can be activated to re-direct the falling kernel if the light reflected from the kernel indicates it is FDK. Note that even though several LEDs appear to be on at the same time, they actually blink much more rapidly than the camera exposure time; so at any given time, only one type of LED is energized. The pattern in the streak ahead of the kernel represents the distance the kernel travels in the time for all nine LEDs to blink.

Table 1. Average agreement (± std. dev.) of HRS between LED and SKNIR instruments.

LED Sorter classification	% Number of kernels sorted into groups using Perten SKNIR sorter			weight % of original sample
	undamaged	Intermediate	FDK	
undamaged	83.2 ±5.4	12.0 ±4.8	4.8 ±1.7	54.8 ±7.3
Intermediate	38.7 ±19.5	38.9 ±18.9	22.4 ±7.8	23.5 ±4.6
FDK	14.4 ±10.5	19.4 ±10.4	66.1 ±20.2	21.6 ±5.1

Table 2. Average agreement (± std. dev.) of SRW between LED and SKNIR instruments.

LED Sorter classification	% Number of kernels sorted into groups using Perten SKNIR sorter			weight % of original sample
	undamaged	Intermediate	FDK	
undamaged	94.3 ±2.0	4.2±1.7	1.5 ±0.6	66.3 ±8.7
Intermediate	56.5 ±13.0	26.4 ±7.7	17.2 ±6.5	21.1 ±5.0
FDK	21.7 ±11.3	23.6 ±5.3	54.7 ±9.6	12.6 ±3.9

QTL ASSOCIATED WITH FUSARIUM HEAD BLIGHT RESISTANCE IN THE NC-NEUSE X AGS 2000 RECOMBINANT INBRED POPULATION

S. Petersen[1], P.V. Maloney[1], R.A. Navarro[1], J.H. Lyerly[1], C. Cowger[2], G. Brown-Guedira[2], D. Marshall[2], J.M. Costa[3], C.A. Griffey[4] and J.P. Murphy[1*]

[1]Department of Crop Science, Box 7629, North Carolina State University, Raleigh, NC 27695; [2]USDA-ARS, Plant Science Research Unit, Raleigh, NC 27695; [3]Department of Plant Science and Landscape Architecture, University of Maryland, College Park, MD 20742; and [4]Department of Crop and Soil Environmental Sciences, Virginia Tech, Blacksburg, VA 24061
[*]Corresponding Author: PH: (919) 513-0000, Email: paul_murphy@ncsu.edu

ABSTRACT

Breeding for resistance to Fusarium Head Blight is of major importance, as the disease can have serious negative impacts on wheat production in warm and humid regions of the world, including the state of North Carolina. Fusarium Head Blight can cause significant grain yield reduction, but also severely affect the grain quality due to accumulation of mycotoxins produced by the pathogen. The importance of finding native sources of resistance in U.S. soft red winter wheat lines has been emphasized in recent years. 'NC-Neuse' is a North Carolina soft red winter wheat cultivar, released in 2003, which is moderately FHB resistant.

A population of 179 random F_5-derived recombinant inbred lines derived from a cross between 'NC-Neuse' and the FHB susceptible line 'AGS 2000' was evaluated for FHB resistance at one field location (3 reps) in the 2010-11 field season, and at two field locations (2 reps/loc) in the 2011-12 season. The FHB related traits evaluated included disease incidence (INC), severity (SEV), *Fusarium* damaged kernels (FDK), and accumulation of the mycotoxin deoxynivalenol (DON). A linkage map developed prior to this QTL mapping study with a total of 1536 polymorphic SSR, DArT and SNP markers across 31 linkage groups was utilized for mapping of QTL (with additional 345 polymorphic unlinked markers). QTL analysis for individual environments and across environments was conducted using Multiple Interval Mapping (MIM) with WinQTLCart v. 2.5. The critical LOD value to declare QTL significance was 3.2, based on 1000 permutations.

QTL associated with one or more FHB resistance traits were identified on chromosomes 1A, 1B, 2AL, 4A, 5A, 5B, 6AL, and 7B. Their LOD score values ranged from 3.2 to 4.38 with R^2 values of 5.5% to 13.3%.

Incidence data from Salisbury, MD 2012 were used for analysis. One QTL associated with decreased disease incidence (type I resistance) was identified on chromosome 1B explaining 7.4% of the phenotypic variation, and one QTL associated with increased incidence (and later heading date) was identified on chromosome 5B explaining 10.3% of the phenotypic variation.

Severity data from Kinston, NC 2011 and Salisbury, MD 2012 were analyzed. Two QTL associated with decreased severity (type II resistance) were identified on chromosomes 4A and 6AL that explained 13.3 and 11.1% of the phenotypic variation, respectively.

Fusarium damaged kernel data from Kinston, NC 2011 and 2012, as well as Salisbury, MD 2012 were analyzed. A total of three QTL associated with decreased proportion of FDK was found. Two QTL were found on chromosome 1A explaining 8.6 and 9.4% of the phenotypic variation, as well as one QTL on chromosome 7B explaining 12.4% of the phenotypic variation.

Deoxynivalenol data from Kinston, NC 2011 and 2012 were used for analysis. Four QTL associated with decreased DON content were found on chromosomes 2AL, 4A, 5A and 6AL explaining between 5.5 and 10.1% of the phenotypic variation.

The evaluations are continuing in the 2012-13 season where we will test the consistency of these tentative QTL.

SOFT WINTER WHEAT RESPONSES TO *FHB1* AND *QFHS.NAU-2DL* QTL FOR FUSARIUM HEAD BLIGHT RESISTANCE IN F_2 DERIVED POPULATIONS

Daniela Sarti[1], Anthony Clark[1], Gina Brown-Guedira[2], Yanhong Dong[3] and David Van Sanford[1*]

[1]University of Kentucky, Plant & Soil Science Dept., Lexington, KY 40546;
[2]USDA-ARS, North Carolina State University, Raleigh, NC 27695; and
[3]University of Minnesota, Plant Pathology Dept., St Paul, MN 55108
[*]Corresponding Author: PH: 859-338-2409, Email: dvs@uky.edu

ABSTRACT

Fusarium Head Blight (FHB), caused by *Fusarium graminearum* Schwabe [teleomorph: *Giberella zeae* Schein. (Petch)] is recognized as one of the most destructive diseases of wheat (*Triticum aestivum* L. and *T. durum* L.) and barley (*Hordeum vulgare* L.). Breeding for FHB resistance is one of the most efficient approaches to reducing this damage. Disease resistance must be accompanied by selection for desirable agronomic traits. Donor parents with two FHB resistance quantitative trait loci (QTL) *Fhb1* (chromosome 3BS) and *QFhs.nau-2DL* (chromosome 2DL) were crossed to four adapted SRW wheat lines to generate backcross and forward-cross progeny. F_2 individuals were genotyped and assigned to 4 different groups according to presence/ absence of resistance alleles at both QTL. The effectiveness of these QTL in reducing FHB in F_2 derived lines was assessed in a misted, inoculated scab nursery for 2 years. Backcross-derived progeny from four genetic backgrounds were planted in replicated plots and in the scab nursery at Lexington, KY in 2011 and 2012. Traits measured included rating (1-9), severity, incidence, FHB index (severity * incidence), FDK (*Fusarium* damaged kernels) and DON (deoxynivalenol). FDK and DON were predicted with Near Infrared Reflectance (NIR) and compared with actual values. One of our objectives was to explore the utility of F_2 populations as indicators of expression levels of QTL prior to extensive backcrossing. The *Fhb1* + 2DL combination showed higher resistance and lower FDK than other QTL classes in most of the populations. FDK was reduced by resistance alleles at one or both QTLs in all four populations. Rating values were significantly ($P \leq 0.05$) reduced by the presence of resistance alleles. In some cases where the average QTL effect was not significant, there was significant ($P \leq 0.05$) variation among $F_{2:4}$ lines within QTL class for FDK, Rating and FHB index. Significant QTL effects on FDK were also detected using NIR. Correlations between FDKNIR and actual FDK ranged from 0.53 to 0.77 across the four populations. Correlations between DONNIR and FDK ranged from 0.55 to 0.74 among populations. BC_1F_3 lines revealed that one backcross had restored yield potential, in that there were lines with yields not significantly different from commercial checks. In population 7, almost 58% of the lines showed competitive yield that did not significantly differ ($P \leq 0.05$) from the commercial checks. Preliminary results indicated that BC1 populations may be a useful source of breeding lines. F_2 populations should be used for genotyping, ensuring QTL are effective before extensive backcrossing.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-054. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

QTL MAPPING TO INVESTIGATE POSSIBLE INHIBITION OF *FHB1*

Brian Seda[1], Ruth Dill-Macky[2], Shiaoman Chao[3] and James Anderson[1*]

[1]University of Minnesota, Department of Agronomy & Plant Genetics, St. Paul, MN;
[2]University of Minnesota, Department of Plant Pathology, St. Paul, MN;
and [3]USDA-ARS, Biosciences Research Lab, Fargo, ND
[*]Corresponding Author: PH: 612-625-9763, Email: ander319@umn.edu

ABSTRACT

Fhb1 is the major Fusarium head blight resistance gene in wheat, and its lineage traces back to introgression from the Chinese cultivar Sumai 3. This gene is most strongly associated with Type II resistance (resistance to fungal spread within the wheat head), but also contributes to other forms of resistance. While investigating *Fhb1* candidate genes as part of our ongoing efforts to clone this gene, we discovered that the recipient genotype, 'Bobwhite' failed to consistently express the effect of *Fhb1*. In addition to this particular gene, there have been other QTL mapping studies published in which a source of resistance is contributed by the susceptible parent in the cross. The failure of *Fhb1* to express in Bobwhite, supported by these other findings, seems to indicate either the interaction of a gene inhibitor with *Fhb1*, or the presence of multiple segregating QTL regions in the resistant parent that confer the segregating response. In order to investigate these hypotheses, we generated a recombinant inbred mapping population of 129 $F_{5:7}$ lines from the cross between '260-2' and 'Bobwhite' where all lines were homozygous positive for the *Fhb1* resistance allele. Bobwhite is negative for the *Fhb1* resistance allele, while 260-2 is a near isogenic line containing *Fhb1* from a Sumai3/Stoa//MN97448 background. Two seasons of Type II resistance screening were undertaken in the greenhouse. Inoculations were conducted using the single-floret (point) inoculation method, and the phenotype assessed as the percent spread of symptoms from the inoculated spikelet. Replicated field trials were conducted in St. Paul, MN in 2011 and 2012 and Crookston, MN in 2012 using both the mapping population and a population of 114 $F_{5:7}$ RILs from the same cross that were homozygous negative for *Fhb1*. Symptomatic spikelet counts (20 heads/plot)were taken at approximately 21 dai to estimate FHB severity (FHBS). Thirty head weights, the percentage of visually scabby kernels (VSK) and the deoxynivalenol content of grain (DON) were assessed on the harvested grain. Phenotypes for all traits analyzed indicate multiple gene segregation. The population was genotyped using the Illumina 9K SNP Infinium assay. The traits examined have now been mapped and 11 QTL regions contributing to FHB resistance have thus far been identified.

DEVELOPMENT OF INTERNATIONAL FUSARIUM HEAD BLIGHT SCREENING NURSERIES OF WHEAT AT CIMMYT, MEXICO

Pawan K. Singh*, Xinyao He and Etienne Duveiller

International Maize and Wheat Improvement Center (CIMMYT),
Apdo. Postal 6-641, 06600 Mexico DF, Mexico
*Corresponding Author: PH: (612) 605 5205 ext 2217, Email: pk.singh@cgiar.org

ABSTRACT

Fusarium head blight (FHB) or head scab, is a fungal disease of small grain cereals that has become a major threat to wheat production globally. *Fusarium graminearum*, the predominant species causing FHB worldwide, is known to produce two important mycotoxins, deoxynivalenol (DON) and zearalenone, which can contaminate the diseased grain. FHB disease causes yield loss, low test weights, low seed germination and contamination of grain with mycotoxins which makes it unfit for human and animal consumption. Changes in agricultural practices including more maize-wheat rotation and adaptation of conservation agriculture practices and global warming have contributed to enhanced incidence and severity of FHB globally. Fungicide application, biological control and adoption of specific agronomic practices have shown limited success in FHB control, increase cost of production, pollution concerns and pose practical constrains in their large scale adaptation. The most effective, economical and environmentally friendly means of managing FHB involves incorporation of genetic resistance into commercial cultivars. FHB research began at the International Maize and Wheat Improvement Center (CIMMYT), Mexico in the early 1980's, and since then, large-scale FHB screening has been conducted to identify and incorporate new resistance genes into elite CIMMYT germplasm. To develop FHB resistant germplasm, large scale FHB screening work has been performed on the promising breeding lines from different CIMMYT breeding programs, genbank accessions and other resources, and crosses have been made between parents with complimentary disease resistance and agronomic traits. The screening field at El Batan, Mexico covers 2 hectares with a yearly screening capacity of up to 10,000 plots, and a programmable misting system is equipped to provide uniform humidity conditions favourable to FHB development. Spray inoculation is being used in the field to assess the combined effect of Type I and Type II resistance, and DON contamination is assayed in the laboratory for promising lines. Furthermore, a haplotyping system has also been established to diagnose well known FHB QTL. Promising lines with good FHB resistance based on multiple years of screening are regularly compiled as a Fusarium Head Blight Screening Nursery (FHBSN) and distributed worldwide. In 2012, the 20 entries comprising the 14th FHBSN were tested at El Batan, Mexico and distributed to 94 institutes in 26 countries. The FHBSN comprise agronomically superior breeding lines possessing Sumai3 and/or non-Sumai3 resistance.

EVALUATING GENOMIC SELECTION FOR DON IN A COLLABORATIVE BARLEY BREEDING EFFORT

Kevin P. Smith[1*], Vikas Vikram[1], Aaron Lorenz[2], Jean-Luc Jannink[3], Shiaoman Chao[4] and Richard Horsley[5]

[1]University of Minnesota, Dept. of Agronomy and Plant Genetics, St. Paul, MN 55108; [2]University of Nebraska, Dept. of Agronomy and Horticulture, Lincoln, NE 68583; [3]USDA-ARS, Robert W. Holley Center for Agriculture and Health, Ithaca, NY 14853; [4]USDA-ARS, Northern Crop Science Lab, Fargo, ND 58105; and [5]North Dakota State University, Dept. of Plant Sciences, Fargo, ND 58108
[*]Corresponding Author: PH: 612-624-1211, Email: smith376@umn.edu

ABSTRACT

Accelerating the development of new varieties with low deoxynivalenol (DON) is a critical component of the USWBSI mission to reduce the risk of Fusarium head blight (FHB). To meet this goal, we have established a project that couples 1) a collaborative breeding effort between three Midwest barley breeding programs to exploit elite FHB germplasm and 2) genomic selection (GS) breeding methods to reduce the breeding cycle time and increase the rate of gain from selection. Using a large marker and trait database assembled through the Barley Coordinated Agricultural Project (CAP), we constructed several training populations with elite breeding lines from the Midwest breeding programs and trained a ridge regression model to calculate genomic estimated breeding values (GEBVs) for DON. Model accuracy was calculated as the correlation between GEBV and the observed phenotype divided by the square root of the heritability. Models trained with breeding lines from a single breeding program were better predictors of lines from that same program. Pooling lines from several breeding programs produced a single model that gave accurate predictions for all lines from all breeding programs, but did not increase accuracy over the other models. The results of this validation study prompted us to implement GS and then evaluate model accuracy on actual breeding progenies. A collaborative breeding population was established by crossing among elite breeding lines with enhanced FHB resistance from the University of Minnesota (M), North Dakota State University (N) and Busch Agriculture (B) breeding programs to generate 1440 F3 progeny. These lines were genotyped with a custom 384 SNP assay and predictions were made using models trained with the Barley CAP marker and trait data. Three breeding progeny populations (MxM, NxN, MxN) of 100 randomly selected lines were evaluated in field trials for DON. As predicted by the results of the prior validation study, models trained with M breeding lines were more accurate for MxM progenies (r=0.58) then for NxN progenies (r=0.07). Likewise the N model was more accurate for NxN (r=0.48) then for MxM progenies (r=0.26). The M+N training population produced accuracies of 0.56 for MxM, 0.40 for NxN, and 0.35 for MxN progenies. This suggests that pooling lines from the two breeding programs will be important for predicting progenies that result from crosses between the programs.

ACKNOWLEDGMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-072. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

PHENOTYPIC ANALYSIS OF A SOFT WHEAT POPULATION THAT WILL BE USED FOR ASSOCIATION ANALYSIS AND GENOMIC SELECTION

C.H. Sneller[1*], A. Cabrera[1], P. Paul[1], D. Van Sanford[2], A. Clark[2], A. Mckendry[3], F. Kolb[4], H. Ohm[5], R. Freed[6] and M.E. Sorrells[7]

[1]The Ohio State University, Wooster OH; [2]University of Kentucky, Lexington KY; [3]University of Missouri, Columbia, MO; [4]University of Illinois Urbana-Champaign, Urbana IL; [5]Purdue University, West Lafayette, IN; [6]Michigan State University, East Lansing MI; and [7]Cornell University, Ithaca NY
[*]Corresponding Author: PH: (330) 263-3944, Email: sneller.5@osu,edu

ABSTRACT

Soft winter wheat (SWW) from the Eastern US has considerable resistance to FHB. Our objectives are to use a large SWW population to i) elucidate the genetics of resistance using association analyses and ii) to develop genomic selection models. This poster presents analysis of the 2011 and 2012 phenotypic data from the population. The population is comprised of 49 checks and 700 RILs. The population will be phenotyped by seven co-PIs in 2011, 2012, and 2013. Each co-PI phenotyped 149 entries comprised of the 49 checks and 100 RILs from their program: a RIL was only tested at one location each year. The traits are incidence, severity, index, *Fusarium* damaged kernels (FDK), and DON. The mean and standard deviation of the 49 checks from an environment were used to standardize data from all entries from that environment.

Over all trait/entry combinations, 31% of the combinations had lower trait values than Truman (R check), 76% were less than Freedom (MR check) and only 5% were greater than Pioneer 2545 (S check). The 700 RILs come from 76 crosses with 41 crosses having ≥ six entries. Over the 222 trait/cross combinations involving these 41 crosses, 73% showed evidence of segregation while in 27% of the combinations all RILs from the cross were at least moderately resistant for the trait: none of these crosses produced only susceptible RILs for any trait.

Resistance to toxin accumulation (RTA) was assessed by regressing DON on estimates of grain infection (FDK or fungal biomass assessed by qPCR) within in each environment using 2011 data. Entries with negative residuals have less DON than predicted based in grain infection and may have high RTA. Several entries had negative residuals in all environments indicating they consistently resist toxin accumulation despite grain infection. Other entries repeatedly had positive residuals indicating they have low RTA.

In conclusion, data standardization eliminated environment effects and minimized entry by environment effects. The population shows good segregation for all traits both overall and within most individual crosses and should be well suited for future association analyses and genomic selection. There is evidence for genetic variation for RTA.

REPORT ON THE 2011-2012 NORTHERN UNIFORM WINTER WHEAT SCAB NURSERIES (NUWWSN AND PNUWWSN)

C. Sneller[1*], P. Paul[2], L. Herald[1] and B. Sugerman[1]

[1]Dept. of Hort. and Crop Science, and [2]Dept. of Plant Pathology,
The Ohio State University, Wooster, Ohio 44691
[*]Corresponding Author: PH: (330) 263-3944, E-mail: sneller.5@osu.edu

OBJECTIVES

This is a summary of the report on the 2011-2012 Northern Uniform Winter Wheat Scab Nursery (NUWWSN) and the Preliminary Northern Uniform Winter Wheat Scab Nursery (PNUWWSN). A full report will be available on the USWBSI web site after the 2012 forum. The objective of these tests is to screen winter wheat genotypes adapted to the northern portion of the eastern US for scab resistance.

MATERIAL AND METHODS

The traits assessed and locations that reported data are listed in Table 1. The 56 (+ 4 checks) entries in the NUWWSN came from 14 programs while the 46 (+ 4 checks) entries in the PNUWWSN entries came from nine programs (Table 2).

RESULTS

Many entries in the PNUWWSN and the NUWWSN showed very good resistance to FHB. In the NUWWSN 22% were not significantly different from the most resistant entry for all seven FHB traits. Only 9% of the entries were susceptible for four or more traits. Over 31% of the entries had DON levels < 4 ppm from these inoculated and listed nurseries where the average DON was 6.5 ppm. In the PNUWWSN, over 43% were not significantly different from the most resistant entry for all seven FHB traits, though 26% of the entries were susceptible for four or more traits. Over 47% of the entries had DON levels < 4 ppm from these inoculated and listed nurseries where the average DON was 5.3 ppm.

Table 1. Traits assessed in the 2011-12 PNUWWSN and NUWWSN tests.

Code	Trait	Description	PNUWWSN Locations*	NUWWSN Locations*
INC	Disease incidence	% of heads with at least one infected spikelets	MI,VA,KY,MO	MI,MD,VA,NY,KY,NE,MO
SEV	Disease severity from field tests	% of infected spikelets in an infected head.	MI,VA,KY,MO	MI,MD,VA,NY,KY,NE,MO
IND	Disease index	IND = (SEVxINC)/100	MI,VA,KY,MO,OH	MI,MD,VA,NY,KY,NE,MO,OH
FDK	Fusarium damaged kernels	Percentage of grain ishowing sypmotoms of Fusarium infection	KY,MO	MD,OH,NY,KY,NE,MO
ISK	Composite of head and kernel traits	ISK Index = 0.3 (Severity) +0 .3 (Incidence)+ 0.4 FDK)	KY,MO	MD,OH,NY,KY,NE,MO
DON	DON (vomitoxin)	PPM of vomitoxin in grain	VA,KY	OH,VA,KY
GH	Greenhouse severity	Same as SEV except from greenhouse	IL,MO	IL,MO
HD	Heading date	Julian date when 50% of heads have emerged	MI,OH,VA,KY	MI,MD,OH,VA,NY,KY

Table 2. Entries in the 2011-12 PNUWWSN (PNU) and NUWWSN (NU)

NU	ERNIE	
NU	FREEDOM	
NU	TRUMAN	
NU	PIONEER2545	
NU	NY103-208-7263	Cayuga/Caled.
NU	NY94052-3329	Pioneer2737w/Harus
NU	NY05072&75-1	Superior*6/Pinb-a
NU	KWS007	IL87-2834-1 / 960314
NU	KWS001	TOTEM / M98-2152
NU	KWS002	TWO44-094 / HONEY
NU	KWS003	95-3245/ Ernie
NU	LCS19214	T814/L900819
NU	LCS19209	Goldfield//IL84-3010/T812
NU	LCS19231	VA99W-200/Patton
NU	LCS19103	IL84-3010/T812
NU	LCS19104	Auburn/T812
NU	E9021R	Pioneer 2552/D8006
NU	OH05-200-74	OH629/HOPEWELL
NU	OH06-159-6	P.92145E8-7-7-1-9-1 / OH728
NU	F0065	Pioneer 25R37/D6234
NU	OH08-133-25	HONEY / COKER 9663
NU	OH06-180-57	KY90C-042-37-1/OH687
NU	OH07-166-41	OH708 / OH684
NU	OH07-263-3	OH748 / BRAVO
NU	04606RA1-1-7-1-6	Truman/INW0316
NU	P0537A1-3-12	IN0411/2754//IN0412/98134
NU	P0566A1-3-1-67	INW0412/992060
NU	P05222A1-1-2-7	99840/INW0304//INW0304/INW0316
NU	MH07-7483	M95-2994-1/P 25R57
NU	MH07-7474	M97-1048/ELKHART
NU	M08-8036#	COKER 9511/BRANSON
NU	M08-8214	COOPER/PIO2552
NU	DH1-46	Superior x D8006W
NU	DH1-62	Superior x D8006W
NU	DH2-4	25R47 x ADV Dyno
NU	DH2-45	25R47 x ADV Dyno
NU	DH5-56	25R56 X Emmit
NU	IL06-13721	IL00-8530 / IL97-3632
NU	IL06-23571	IL96-6472/ Pioneer 25W33 // 94-1653
NU	IL07-4415	P96169RE2-3-6-4 / IL01-34159
NU	IL07-19334	IL01-36115 / IL79-008T-B-B
NU	KY04C-2004-1-1-3	Roane/Allegiance
NU	KY03C-1224-10-12-3	25R18/VA87W-375ws//KY96C-0767-1
NU	KY03C-1195-10-8-5	KY92-0010-17//25R18/KY92C0017-17
NU	KY04C-2031-29-7-3	Truman/VA97W-375ws
NU	MD08-22-1-6-2	Ning7840/McCormick*3
NU	MD08-22-32	Ning7840/McCormick*3
NU	MO090932	980829/Ernie
NU	MO081320	980525//981020/AP Patton
NU	MO090478	980429/Ernie
NU	MO091068	Ernie/Colorben 4
NU	NE10514	NE99533-3/NE99464
NU	NE10449	NI03418/Camelot
NU	NW03666	N94S097KS/NE93459
NU	NW10401	SHARK/F4105W2.1//NI02425
NU	NE10418	OK99212/Overland
NU	VA09W-52	GF921221E16 / McCormick"S" // VA99W-200
NU	VA09W-73	SS 520 (VA96W-158) / VA99W-188 // TRIBUTE
NU	VA10W-21	Z00-5018 / VA01W-158
NU	VA09W-75	SS 520 (VA96W-158) / VA99W-188 // TRIBUTE
PNU	ERNIE	
PNU	FREEDOM	
PNU	TRUMAN	
PNU	PIONEER2545	
PNU	KWS006	PUR 89118 / RINGO
PNU	KWS004	RAVEN / ATLAS
PNU	KWS005	HARVARD / DINGO
PNU	F0051R	Gold./CJ9306//Caled/CJ9403/3/Caled/4/Caled
PNU	F0014	Pioneer 2552 / E0029
PNU	F0038	D8006 / CJ9306 // Caled. /3/ Caled. /4/ Caled.
PNU	F0036R	D6234/W14/E0038-1/3/E0038-1
PNU	OH08-172-42	DOUGLAS / JEKYL
PNU	7x831-1-I-03Ser (2)	Malabar*4/Karl
PNU	OH08-172-42	DOUGLAS / JEKYL
PNU	OH08-180-48	DOUGLAS / MCCORMICK
PNU	OH08-269-58	P.92226E2-5-3 / OH708
PNU	OH07-254-11	OH728 / VA97W-361WS
PNU	05247A1-7-7-3-1	99840*2/03726//99794
PNU	P0566A1-3-1-65	INW0412/992060
PNU	P05247A1-7-3-27	99840*2/03726//99794
PNU	P05247A1-7-3-120	99840*2/03726//99794
PNU	P0762A1-2-8	981129/99793//INW0301/92145/3/981477/981312//INW0316
PNU	P07287RA1-14	INW0304/INW0316//97462/3/Truman
PNU	M09-9811#	TRUMAN/CK9511
PNU	M09-9826#	CK9511/M03-3002
PNU	IL07-21847	IL99-2536/ IL97-3632// IL00-8061
PNU	IL08-8844	IL00-8109 / IL02-24251
PNU	IL08-22206	IL00-8530 / VA01-476 // IL79-002DH
PNU	IL08-33373	IL79-005T-B-B / IL00-8530
PNU	IL07-20728	McCormick/IL97-1828// IL00-8061
PNU	IL07-20743	McCormick/IL97-1828// IL00-8061
PNU	KY04C-2031-29-6-1	Truman/VA97W-375ws
PNU	KY03C-2022-16-18-1	KY93C-0876-66/25R18
PNU	KY04C-2006-45-5-1	Roane/KY93C-1238-17-1
PNU	KY04C-2150-66-16-5	25R18/KY93C-1238-17-1
PNU	KY04C-2150-64-16-1	25R18/KY93C-1238-17-1
PNU	KY04C-2150-64-17-1	25R18/KY93C-1238-17-1
PNU	MO100295	981020/010895
PNU	MO101235	001164/IL 96-6472
PNU	MO100532	000925//980525/433-1-2
PNU	MO100314	010708/AP Patton
PNU	MO100410	980829/IL 96-346
PNU	MO101259	002409/980525
PNU	MO081765	L910097/MO 92-599
PNU	VA08W-613	FREEDOM / NEUSE"S" // VA98W-688
PNU	VA09W-608	P97397B1-4-5 / McCORMICK // COKER 9511
PNU	VA10W-663	P97397B1-4-5 / McCORMICK // COKER 9511
PNU	VA10W-28	SS-MPV57 (VA97W-24) / M99*3098
PNU	VA09W-654	VA98W-749 / IL96-3073 // P9793A1-5
PNU	VA10W-617	VA98W-749 / IL96-3073 // COKER 9474

Table 3. Most resistant entries from the 2011-12 NUWWSN (top of table) and PNUWWSN (bottom of table).

NAME	INC	SEV	IND	FDK	ISK	DON	GHSEV	HD	#l	#h
MD08-22-1-6-2	20.4 l	7.4 l	1.9 l	5.7 l	10.0 l	1.2 l	13.9 l	136	7	0
MD08-22-32	21.5 l	8.1 l	2.0 l	9.7 l	11.4 l	1.4 l	10.0 l	134	7	0
IL07-4415	22.2 l	5.1 l	2.1 l	9.6 l	12.8 l	1.6 l	11.7 l	130 l	7	0
IL07-19334	25.2 l	6.5 l	2.5 l	5.3 l	11.5 l	3.0 l	8.3 l	134	7	0
MO091068	19.3 l	10.0 l	2.9 l	11.0 l	13.9 l	2.2 l	8.2 l	133	7	0
M08-8214	28.2 l	7.2 l	3.0 l	6.5 l	13.8 l	3.1 l	24.3 l	131 l	7	0
IL06-13721	21.3 l	9.5 l	3.5 l	11.1 l	15.9 l	2.3 l	19.0 l	129 l	7	0
LCS19103	23.1 l	9.5 l	3.8 l	11.2 l	16.3 l	2.5 l	17.9 l	131 l	7	0
IL06-23571	33.0 l	11.3 l	5.2 l	7.0 l	16.6 l	2.6 l	11.8 l	129 l	7	0
M08-8036#	35.9 l	13.1 l	5.3 l	9.1 l	18.7 l	2.6 l	21.0 l	130 l	7	0
LCS19214	34.2 l	11.1 l	5.7 l	7.7 l	18.5 l	2.9 l	38.0 hl	131 l	7	1
MO090478	24.9 l	13.0 l	6.3 l	13.8 l	18.2 l	3.5 l	19.0 l	129 l	7	0
MO090932	21.7 l	8.4 l	3.1 l	7.5 l	10.4 l	6.4	6.0 l	139	6	0
TRUMAN	21.2 l	6.0 l	3.8 l	8.5 l	12.8 l	6.3	8.5 l	139	6	0
NW10401	25.9 l	14.0 l	5.0 l	10.5 l	15.9 l	3.9 l	63.0 h	131 l	6	1
LCS19209	33.9 l	11.0 l	6.1 l	8.8 l	19.3	2.2 l	24.8 l	131 l	6	0
MO081320	24.6 l	7.2 l	6.3 l	10.3 l	17.8 l	3.6 l	34.3 hl	131 l	6	0
LCS19104	27.1 l	16.7	6.6 l	12.7 l	18.0 l	3.5 l	15.8 l	132	6	0
OH07-166-41	28.3 l	15.2 l	6.7 l	7.5 l	18.0 l	3.9 l	57.9 h	134	6	1
LCS19231	34.6 l	18.5	7.5 l	10.1 l	19.0 l	4.2 l	33.8 hl	133	6	1
KY03C-1224-10-12-3	29.1 l	15.4 l	7.9 l	10.5 l	17.3 l	7.7	8.5 l	134	6	0
NE10514	28.8 l	12.5 l	5.6 l	17.8	18.6 l	9.9	23.3 l	138	5	0
ERNIE	26.5 l	9.6 l	6.4 l	14.2 l	19.7	6.7	17.8 l	129 l	5	0
VA09W-52	36.5	13.1 l	8.2 l	8.3 l	20.1	2.7 l	24.6 l	130 l	5	0
P0762A1-2-8	23.7 l	4.0 l	2.2 l	8.3 l	20.0 l	3.2 l	11.4 l	135 l	7	0
MO101259	30.0 l	7.9 l	2.6 l	11.1 hl	17.2 l	5.2 l	16.8 l	140 h	7	1
KY04C-2150-66-16-5	33.7 l	6.4 l	3.1 l	7.6 l	22.2 l	2.9 l	17.1 l	139 h	7	0
MO100295	29.0 l	12.3 l	3.9 l	4.5 l	18.6 l	3.7 l	30.6 hl	136	7	1
MO100314	33.9 l	8.8 l	4.0 l	5.9 l	18.6 l	2.3 l	6.9 l	138	7	0
MO081765	32.3 l	9.7 l	4.2 l	7.7 l	22.8 l	1.7 l	9.8 l	136	7	0
MO100532	32.6 l	11.5 l	4.5 l	3.7 l	20.1 l	2.2 l	24.8 l	134 l	7	0
VA10W-617	26.4 l	11.2 l	5.0 l	3.7 l	19.0 l	2.5 l	22.8 l	134 l	7	0
KY04C-2150-64-16-1	42.4 l	10.6 l	5.2 l	8.7 l	24.8 l	3.6 l	13.4 l	138	7	0
IL07-20743	34.4 l	9.1 l	5.3 l	7.5 l	22.8 l	4.5 l	26.4 l	137	7	0
MO101235	32.4 l	13.7 l	5.5 l	5.1 l	19.2 l	4.8 l	14.5 l	136	7	0
IL07-21847	32.5 l	9.7 l	5.8 l	13.5 hl	22.6 l	2.3 l	21.5 l	134 l	7	1
P05247A1-7-3-27	39.8 l	8.4 l	5.9 l	17.4 hl	28.1 l	5.2 l	35.4 hl	134 l	7	2
VA09W-608	31.7 l	12.4 l	5.9 l	9.6 l	23.0 l	1.6 l	10.3 l	134 l	7	0
IL08-33373	39.4 l	6.6 l	6.1 l	15.2 hl	24.8 l	2.7 l	10.6 l	140 h	7	1
TRUMAN	42.4 l	15.8 l	6.2 l	4.8 l	23.7 l	6.5 l	7.9 l	142 h	7	0
IL07-20728	40.5 l	14.8 l	6.3 l	3.4 l	20.4 l	2.0 l	30.0 hl	137	7	1
IL08-22206	35.6 l	8.1 l	6.6 l	4.8 l	24.3 l	1.8 l	23.8 l	134 l	7	0
MO100410	33.7 l	7.8 l	6.9 l	7.4 l	21.3 l	5.3 l	6.9 l	142 h	7	0
IL08-8844	35.3 l	17.5 l	7.1 l	9.0 l	22.2 l	3.3 l	19.0 l	134 l	7	0
VA09W-654	28.6 l	14.0 l	7.7 l	4.7 l	18.3 l	2.1 l	33.0 hl	139	7	1
VA08W-613	33.6 l	9.0 l	8.0 l	18.2 hl	30.0 l	4.7 l	13.3 l	132 l	7	1
P07287RA1-14	29.9 l	11.9 l	5.3 l	17.4 hl	27.4 l	3.5 l	47.0 h	133 l	6	2
M09-9811#	36.8 l	20.8 h	6.2 l	8.1 l	23.4 l	3.1 l	21.4 l	135	6	1
KY04C-2150-64-17-1	48.7 h	9.2 l	6.8 l	5.3 l	28.5 l	4.8 l	15.5 l	139	6	1
ERNIE	36.3 l	7.6 l	8.8 l	11.7 hl	27.7 l	5.3 l	16.9 l	133 l	6	0

l,h indicate a mean that is not significantly different than the lowest (l) or highest (h) mean in that column

Table 4. Summary of results of the 2011-12 NUWWSN.

NAME	INC		SEV		IND		FDK		ISK		DON		GHSEV		HD		HGT		#l	#h
ERNIE	26.5	l	9.6	l	6.4	l	14.2	l	19.7		6.7		17.8	l	129	l	34		5	0
FREEDOM	46.0	h	20.5		14.2		13.3	l	27.4		3.9	l	7.6	l	134		38		3	1
TRUMAN	21.2	l	6.0	l	3.8	l	8.5	l	12.8	l	6.3		8.5	l	139		41		6	0
PIONEER2545	54.4	h	32.6	h	28.1	h	23.2	h	40.7	h	11.2		58.7	h	135		38		0	6
NY103-208-7263	43.1	h	24.9	h	13.6		16.9		26.4		10.7		14.8	l	143	h	43		1	2
NY94052-3329	49.2	h	25.5	h	16.9		21.5	h	31.3		17.1	h	21.3	l	142	h	39		1	4
NY05072&75-1	39.2	h	22.7	h	12.1		20.7	h	25.3		6.2		25.8	l	143	h	47	h	1	3
KWS007	45.4	h	25.1	h	14.8		16.4		29.4		9.0		44.5	h	136		38		0	3
KWS001	39.1	h	19.7		11.9		18.5		28.0		10.7		33.8	hl	135		34		1	2
KWS002	42.1	h	19.3		9.1	l	15.7		24.9		7.4		9.9	l	136		39		2	1
KWS003	41.0	h	17.4		8.1	l	13.2	l	22.5		4.1	l	23.6	l	136		34		4	1
LCS19214	34.2	l	11.1	l	5.7	l	7.7	l	18.5	l	2.9	l	38.0	hl	131	l	35		7	1
LCS19209	33.9	l	11.0	l	6.1	l	8.8	l	19.3		2.2	l	24.8	l	131	l	35		6	0
LCS19231	34.6	l	18.5		7.5	l	10.1	l	19.0	l	4.2	l	33.8	hl	133		43		6	1
LCS19103	23.1	l	9.5	l	3.8	l	11.2	l	16.3	l	2.5	l	17.9	l	131	l	40		7	0
LCS19104	27.1	l	16.7		6.6	l	12.7	l	18.0	l	3.5	l	15.8	l	132		40		6	0
E9021R	42.7	h	27.7	h	13.7		10.0	l	25.7		7.1		45.3	h	133		39		1	3
OH05-200-74	31.9	l	7.3	l	5.7	l	15.9		20.2		6.3		15.4	l	136		38		4	0
OH06-159-6	29.7	l	10.4	l	5.2	l	18.6		20.8		7.6		62.3	h	130	l	35		3	1
F0065	46.3	h	33.6	h	20.5		14.9	l	33.6	h	10.1		55.7	h	134		37		1	4
OH08-133-25	43.0	h	26.1	h	15.1		21.7	h	31.9	h	5.4	l	51.4	h	132		37		1	5
OH06-180-57	38.4	h	21.6		15.0		12.3	l	27.0		8.0		56.9	h	134		37		1	2
OH07-166-41	28.3	l	15.2	l	6.7	l	7.5	l	18.0	l	3.9	l	57.9	h	134		38		6	1
OH07-263-3	40.9	h	17.7		11.6		8.7	l	22.5		4.9	l	47.1	h	131	l	38		2	2
P04606RA1-1-7-1-6	45.9	h	12.1	l	10.0		16.5		26.3		15.3	h	5.3	l	136		41		2	2
P0537A1-3-12	34.5	l	17.4		8.4	l	15.6	l	23.3		7.2		21.2	l	132		35		4	0
P0566A1-3-1-67	38.0	h	11.5	l	7.1	l	20.3	h	25.1		4.2	l	8.6	l	130	l	33		4	2
P05222A1-1-2-7	39.2	h	11.3	l	7.5	l	18.2		24.5		8.6		30.1	hl	130	l	29	l	3	2
MH07-7483	40.4	h	18.1		9.9		20.6	h	27.3		9.9		27.1	l	134		36		1	2
MH07-7474	34.3	l	19.7		8.4	l	14.0	l	21.5		8.0		35.3	hl	134		36		4	1
M08-8036#	35.9	l	13.1	l	5.3	l	9.1	l	18.7	l	2.6	l	21.0	l	130	l	35		7	0
M08-8214	28.2	l	7.2	l	3.0	l	6.5	l	13.8	l	3.1	l	24.3	l	131	l	40		7	0
DH1-46	33.6	l	15.4	l	10.4		23.4	h	25.4		10.3		53.8	h	141		42		2	2
DH1-62	43.7	h	26.1	h	13.6		25.6	h	29.4		13.0	h	13.0	l	143	h	42		1	4
DH2-4	34.5	l	18.8		7.9	l	29.0	h	24.0		12.2		7.7	l	141	h	38		3	1
DH2-45	44.7	h	22.2		13.7		22.7	h	29.8		11.0		37.3	hl	134		36		1	3
DH5-56	37.8	h	26.0	h	12.9		19.9	h	30.0		7.2		34.2	hl	136		38		1	4
IL06-13721	21.3	l	9.5	l	3.5	l	11.1	l	15.9	l	2.3	l	19.0	l	129	l	35		7	0
IL06-23571	33.0	l	11.3	l	5.2	l	7.0	l	16.6	l	2.6	l	11.8	l	129	l	37		7	0
IL07-4415	22.2	l	5.1	l	2.1	l	9.6	l	12.8	l	1.6	l	11.7	l	130	l	35		7	0
IL07-19334	25.2	l	6.5	l	2.5	l	5.3	l	11.5	l	3.0	l	8.3	l	134		38		7	0
KY04C-2004-1-1-3	37.3		18.6		8.4	l	20.0	h	24.5		8.3		9.8	l	135		37		2	1
KY03C-1224-10-12-3	29.1	l	15.4	l	7.9	l	10.5	l	17.3	l	7.7		8.5	l	134		32	l	6	0
KY03C-1195-10-8-5	50.2	h	12.7	l	10.6		15.2	l	26.3		8.5		9.8	l	136		35		3	1
KY04C-2031-29-7-3	36.8		20.9		10.9		9.7	l	22.5		6.6		39.4	h	133		33		1	1
MD08-22-1-6-2	20.4	l	7.4	l	1.9	l	5.7	l	10.0	l	1.2	l	13.9	l	136		34		7	0
MD08-22-32	21.5	l	8.1	l	2.0	l	9.7	l	11.4	l	1.4	l	10.0	l	134		35		7	0
MO090932	21.7	l	8.4	l	3.1	l	7.5	l	10.4	l	6.4		6.0	l	139		41		6	0
MO081320	24.6	l	7.2	l	6.3	l	10.3	l	17.8	l	3.6	l	34.3	hl	131	l	37		6	0
MO090478	24.9	l	13.0	l	6.3	l	13.8	l	18.2	l	3.5	l	19.0	l	129	l	33		7	0
MO091068	19.3	l	10.0	l	2.9	l	11.0	l	13.9	l	2.2	l	8.2	l	133		35		7	0
NE10514	28.8	l	12.5	l	5.6	l	17.8		18.6	l	9.9		23.3	l	138		38		5	0
NE10449	32.2	l	16.3		7.8	l	26.2	h	25.9		8.3		33.3	hl	138		41		3	2
NW03666	38.0	h	15.1	l	8.0	l	18.3		22.9		10.6		27.0	l	136		40		3	1
NW10401	25.9	l	14.0	l	5.0	l	10.5	l	15.9	l	3.9	l	63.0	h	131	l	38		6	1
NE10418	36.4		17.7		7.0	l	17.1		22.7		5.3	l	26.2	l	130	l	40		3	0
VA09W-52	36.5		13.1	l	8.2	l	8.3	l	20.1		2.7	l	24.6	l	130	l	34		5	0
VA09W-73	41.3	h	22.5	h	10.2		16.7		24.6		3.8	l	25.0	l	131	l	35		2	2
VA10W-21	34.3	l	17.1		8.3	l	12.5	l	21.2		5.3	l	42.3	h	131	l	34		4	1
VA09W-75	42.8	h	22.8	h	10.7		17.8		25.4		5.6	l	32.1	hl	133		36		2	3
AVERAGE	34.9		16.2		8.7		14.6		22.1		6.5		26.9		134		37			
MINUMUM	19.3		5.1		1.9		5.3		10.0		1.2		5.3		129		29			
MAXIMUM	54.4		33.6		28.1		29.0		40.7		17.1		63.0		143		47			
LSD(0.05)	16.9		11.1		7.3		10.3		9.3		4.5		34.1		3		3			
# LOCATIONS	7		7		8		6		6		3		2		6		2			

l,h indicate a mean that is not significantly different than the lowest (l) or highest (h) mean in that column

Table 5. Summary of results of the 2011-12 NUWWSN.

NAME	INC		SEV		IND		FDK		ISK		DON		GH		HG		HGT	#l	#h
ERNIE	36.3	l	7.6	l	8.8	l	11.7	hl	27.7	l	5.3	l	16.9	l	133	l	35	6	0
FREEDOM	53.7	h	25.6	h	20.0	h	23.8	h	38.6	h	6.2	l	13.2	l	139		38	2	5
TRUMAN	42.4	l	15.8	l	6.2	l	4.8	l	23.7	l	6.5	l	7.9	l	142	h	41	7	0
PIONEER2545	67.7	h	25.0	h	26.6	h	25.9	h	45.3	h	12.8	h	22.6	l	138		39	1	6
KWS006	39.2	l	8.0	l	12.1		26.3	h	28.2	l	8.8		9.6	l	136		38	4	1
KWS004	36.6	l	25.9	h	11.2	l	26.0	h	32.0	hl	4.0	l	60.6	h	136		41	4	4
KWS005	48.4		24.6	h	10.6	l	28.4	h	39.1	h	4.9	l	30.1	hl	140	h	40	3	4
F0051R	57.5	h	20.0	h	11.1	l	12.5	hl	37.5	h	6.9	l	14.6	l	140	h	38	4	4
F0014	61.0	h	33.2	h	28.0	h	25.3	h	45.3	h	15.3	h	22.9	l	137		35	1	6
F0038	56.7	h	29.1	h	20.2	h	15.3	hl	36.3	h	13.1	h	30.3	hl	139		38	1	6
F0036R	52.6	h	28.2	h	17.4		16.3	hl	36.4	h	10.5	h	22.9	l	137		35	1	4
OH08-172-42	51.1	h	16.6	l	12.3		16.2	hl	31.9	hl	7.2		15.3	l	137		39	4	3
7x831-1-l-03Ser (2)	45.5		15.2	l	9.0	l	14.6	hl	31.7	hl	8.1		40.3	h	141	h	47	4	2
OH08-172-42	52.5	h	20.6	h	12.7		20.2	hl	34.2	h	7.8		36.4	hl	137		41	2	5
OH08-180-48	52.5	h	14.1	l	13.8		19.9	hl	39.6	h	11.2	h	37.8	hl	138		35	3	5
OH08-269-58	46.0		17.9	l	15.6		14.5	hl	28.9	l	7.5		43.3	h	137		41	3	2
OH07-254-11	50.3	h	28.6	h	12.9		24.4	h	36.3	h	12.8	h	26.5	l	135		37	1	5
P05247A1-7-7-3-1	54.8	h	18.0	l	11.3	l	10.9	hl	34.4	h	5.4	l	34.6	hl	140	h	38	5	3
P0566A1-3-1-65	48.9	h	18.2	l	9.6	l	5.3	l	32.8	h	5.5	l	13.6	l	133	l	36	5	2
P05247A1-7-3-27	39.8	l	8.4	l	5.9	l	17.4	hl	28.1	l	5.2	l	35.4	hl	134	l	34	7	2
P05247A1-7-3-120	48.6	h	16.0	l	10.2	l	16.2	hl	34.3	h	5.8	l	46.7	h	134	l	34	4	4
P0762A1-2-8	23.7	l	4.0	l	2.2	l	8.3	l	20.0	l	3.2	l	11.4	l	135	l	34	7	0
P07287RA1-14	29.9	l	11.9	l	5.3	l	17.4	hl	27.4	l	3.5	l	47.0	h	133	l	34	6	2
M09-9811#	36.8	l	20.8	h	6.2	l	8.1	l	23.4	l	3.1	l	21.4	l	135		40	6	1
M09-9826#	46.1		9.7	l	8.6	l	25.5	h	34.9	h	4.8	l	16.0	l	134	l	34	4	2
IL07-21847	32.5	l	9.7	l	5.8	l	13.5	hl	22.6	l	2.3	l	21.5	l	134	l	38	7	1
IL08-8844	35.3	l	17.5	l	7.1	l	9.0	l	22.2	l	3.3	l	19.0	l	134	l	39	7	0
IL08-22206	35.6	l	8.1	l	6.6	l	4.8	l	24.3	l	1.8	l	23.8	l	134	l	37	7	0
IL08-33373	39.4	l	6.6	l	6.1	l	15.2	hl	24.8	l	2.7	l	10.6	l	140	h	40	7	1
IL07-20728	40.5	l	14.8	l	6.3	l	3.4	l	20.4	l	2.0	l	30.0	hl	137		38	7	1
IL07-20743	34.4	l	9.1	l	5.3	l	7.5	l	22.8	l	4.5	l	26.4	l	137		39	7	0
KY04C-2031-29-6-1	58.9	h	27.8	h	10.9	l	22.1	h	37.5	h	5.4	l	25.8	l	140	h	41	3	4
KY03C-2022-16-18-1	61.2	h	25.6	h	10.7	l	13.2	hl	36.8	h	3.7	l	18.4	l	140	h	36	4	4
KY04C-2006-45-5-1	55.4	h	21.0	h	9.7	l	9.5	l	32.8	h	4.0	l	11.0	l	138		40	4	3
KY04C-2150-66-16-5	33.7	l	6.4	l	3.1	l	7.6	l	22.2	l	2.9	l	17.1	l	139	h	35	7	0
KY04C-2150-64-16-1	42.4	l	10.6	l	5.2	l	8.7	l	24.8	l	3.6	l	13.4	l	138		38	7	0
KY04C-2150-64-17-1	48.7	h	9.2	l	6.8	l	5.3	l	28.5	l	4.8	l	15.5	l	139		36	6	1
MO100295	29.0	l	12.3	l	3.9	l	4.5	l	18.6	l	3.7	l	30.6	hl	136		43	7	1
MO101235	32.4	l	13.7	l	5.5	l	5.1	l	19.2	l	4.8	l	14.5	l	136		40	7	0
MO100532	32.6	l	11.5	l	4.5	l	3.7	l	20.1	l	2.2	l	24.8	l	134	l	41	7	0
MO100314	33.9	l	8.8	l	4.0	l	5.9	l	18.6	l	2.3	l	6.9	l	138		50	7	0
MO100410	33.7	l	7.8	l	6.9	l	7.4	l	21.3	l	5.3	l	6.9	l	142	h	51	7	0
MO101259	30.0	l	7.9	l	2.6	l	11.1	hl	17.2	l	5.2	l	16.8	l	140	h	42	7	1
MO081765	32.3	l	9.7	l	4.2	l	7.7	l	22.8	l	1.7	l	9.8	l	136		37	7	0
VA08W-613	33.6	l	9.0	l	8.0	l	18.2	hl	30.0	l	4.7	l	13.3	l	132	l	35	7	1
VA09W-608	31.7	l	12.4	l	5.9	l	9.6	l	23.0	l	1.6	l	10.3	l	134	l	35	7	0
VA10W-663	45.7		28.0	h	13.8		4.0	l	32.1	h	1.8	l	18.2	l	132	l	27	3	2
VA10W-28	44.5		26.9	h	11.2	l	3.4	l	30.2	l	2.0	l	22.0	l	133	l	34	5	1
VA09W-654	28.6	l	14.0	l	7.7	l	4.7	l	18.3	l	2.1	l	33.0	hl	139		43	7	1
VA10W-617	26.4	l	11.2	l	5.0	l	3.7	l	19.0	l	2.5	l	22.8	l	134	l	37	7	0
AVERAGE	42.6		16.1		9.5		12.9		28.7		5.3		22.8		137		38		
MINUMUM	23.7		4.0		2.2		3.4		17.2		1.6		6.9		132		27		
MAXIMUM	67.7		33.2		28.0		28.4		45.3		15.3		60.6		142		51		
LSD(0.05)	19.2		14.7		9.3		17.8		14.8		5.3		32.6		3		.		
# LOCATIONS	4		4		5		2		2		2		2		4		1		

l,h indicate a mean that is not significantly different than the lowest (l) or highest (h) mean in that column

SSR MARKER HAPLOTYPES CONFIRM FIVE NOVEL FHB RESISTANCE SOURCES FROM THE CIMMYT SCAB RESISTANCE SCREENING NURSERY

S.L. Sydenham*, C. de Villiers and J.A.N. Asiwe

ARC-Small Grain Institute, Private Bag X 29, Bethlehem, 9700, South Africa
*Corresponding Author: PH: 27 58 3073408, Email: sydenhams@arc.agric.za

ABSTRACT

Wheat production in South Africa under irrigation is periodically under threat from fungal diseases. Fusarium head blight (FHB) caused predominantly by *Fusarium graminearium*, has become the most prevalent disease on wheat in different irrigation production areas. Under severe disease pressure, in combination with the planting of highly susceptible cultivars, maize-wheat crop rotation and no-tilling practices, yield losses of up to 40% are possible. A secondary concern is the FHB infected grain contaminated with mycotoxins, such as Deoxynivalenol (DON), which are harmful to humans and animals after consumption. Currently in South Africa there is very little enforcement of mycotoxin monitoring regulations within contaminated wheat grain. Additionally, there is no officially registered fungicide to control Fusarium head blight in South Africa to date. The most environment friendly and efficient FHB control method is host plant resistance. There are different forms of FHB resistance documented that make up the FHB disease complex: Type I- resistance to initial infection, Type II- resistance to the spread of disease symptoms within the spike and Type III- resistance to the accumulation of mycotoxins in infected grain. To date there are no moderately tolerant or moderate-highly FHB resistant wheat cultivars available in South Africa for commercial use. Seven sources of resistance were used for targeted SSR haplotype comparison namely, Sumai 3 (*3B-Fhb1, 5A, 6B-Fhb2 & Fhb7AC*), Frontana (*3A*), Asozaria III (QTL?), Baisanyuehuang (*3BSc* & *Fhb1*), Huangcandou (*Fhb1, 3Bsc & 2D*), Huangfangzhu (*7A* & *3B*), Haiyanzhong (*7D*) and Wangshuibai (*Fhb4 & Fhb5*). For a number of years the well documented FHB resistant QTL (*3B-Fhb1, 5A, 6B-Fhb2 & Fhb7AC*) in Sumai 3 have been used throughout the world primarily transferring the *Fhb1* gene into different wheat varieties. However, it is important to further diversify FHB resistance gene pool available and continually search for new novel, FHB resistant sources to prevent total dependence on the existing well characterised sources which are predominantly of Asian origin. In this study, five resistant lines identified during a two year phenotypic screening process were further genotyped and characterised with the use of 22 Simple sequence repeat (SSR) markers associated with a number of different FHB QTL/genes from the seven known FHB resistant sources listed. The lines originated from the Wheat germplasm nurseries (Scab Resistant Screening Nursery - SRSN) imported from CIMMYT, Mexico and were tested with the South African FHB complex isolates. Pedigree analysis of selected lines showed no related kinship to any of the seven known resistance sources. A number of the SSR markers showed clear allelic differences between the five CIMMYT lines and the seven known resistance sources tested. These findings indicate the true novelty of the resistance in these five sources from the CIMMYT SRSN. Mapping populations of these sources will need to be developed to attempt to map and identify the different QTL/genes present and regulating FHB resistance expression. This will be an important step in improving and further diversifying FHB resistant levels available in South African wheat, as we look into the future.

FHB SYMPTOMS AND DON ACCUMULATION IN WINTER DURUM CULTIVARS AND DOUBLE HAPLOID LINES

L. Tamburic-Ilincic*, M. Camerini, P. Pandey, J. Brinkman and A. Schaafsma

University of Guelph, Ridgetown Campus, Ridgetown, ON, N0P2C0, Canada
*Corresponding Author: PH: (519) 674-1500 x 63557, Email: ltamburi@uoguelph.ca

ABSTRACT

Fusarium graminearum (FG) Schwabe [teleomorph *Gibberella zeae* Schwein. Petch] is the predominant *Fusarium* species pathogenic to cereals in North America. The most important mycotoxin produced by FG is deoxynivalenol (DON). In general, durum (*Triticum turgidum* subsp *durum* L) is more susceptible to FHB than common wheat (*Triticum aestivum* L.). The most practical way to control FHB is through the development of resistant cultivars. OAC Amber is the only winter durum registered in Canada. The objective of this study was to investigate the level of resistance to FHB in several winter durum cultivars and double haploid (DH) lines. Fifteen winter durum cultivars from Italy and 73 DH lines (cross OAC Amber x NSD 11/00) were screened for FHB resistance. The plots were spray inoculated with a mixture of *F. graminearum* isolates at 50% anthesis. FHB symptoms were recorded as a product of incidence and severity. Deoxynivalenol was quantified in harvested grain by ELISA (EZ-Quant® www.diagnostix.ca). Mean FHB index and DON level across durum cultivars from Italy were 55.7 % and 9.5 ppm, respectively, with the lowest FHB index (23.8%) and DON level (2.6 ppm) detected in the cv Arna Coris. FHB index and DON level in OAC Amber were 44.9% and 11.2 ppm, respectively, while mean DON across the DH population was 30.1 ppm, ranging from 10.5 ppm to 68.6 ppm. A number of winter durum genotypes with lower DON levels than OAC Amber have been identified. If found to be phenotypically stable in coming years, they would be considered as FHB donor parents for future crosses. In general, higher DON levels were detected in winter durum compared with common wheat lines grown in the same FHB nursery in Ridgetown, Ontario, Canada in 2012. These results indicate that breeding for FHB resistance and lower DON accumulation in winter durum grain is a realistic and promising goal.

COORDINATED EVALUATION AND UTILIZATION OF MARKER ASSISTED SELECTION FOR FHB RESISTANCE

David Van Sanford[1*], Fred Kolb[2], Anne McKendry[3], Herb Ohm[4], Clay Sneller[5], Mark Sorrells[6], Gina Brown-Guedira[7], Janet Lewis[8,9], Russ Freed[8] and Lee Siler[8]

[1]University of Kentucky, Dept. of Plant and Soil Sciences, Lexington, KY; [2]University of Illinois, Dept. of Crop Sciences, Urbana, IL; [3]University of Missouri, Dept. of Plant Sciences, Columbia, MO; [4]Purdue University, Department of Agronomy, West Lafayette, IN; [5]Ohio State University/OARDC, Dept. of Hort. and Crop Science, Wooster, OH; [6]USDA-ARS, Plant Science Research Unit, Raleigh, NC; [7]Michigan State University, Dept. of Plant, Soil and Microbial Sciences, East Lansing, MI; and [8]Current Address: Bayer CropScience, Global Wheat Breeding, 202 Keim Hall, Lincoln, NE
[*]Corresponding Author: PH: 859-338-2409, Email: dvs@uky.edu

ABSTRACT

The objectives of this project were to 1) evaluate the effectiveness of use of FHB-resistance QTL in the NWW breeding programs through marker assisted selection (MAS); 2) quantify the effects of these QTL in reducing FHB and DON, and 3) measure their impact on other important traits such as yield and milling and baking quality. Soft winter wheat breeding programs in NY, MI, OH, IN, IL, MO and KY each identified ~10 lines from their programs for inclusion in the study. All lines had shown FHB resistance in individual program FHB nurseries. Based on pedigree, it was assumed that in some lines the resistance derived from exotic QTL such as *Fhb1* and in other cases, resistance was considered to be native. The lines were genotyped at the USDA-ARS Eastern Regional Small Grain Genotyping Lab in Raleigh, NC. Two resistant check cultivars were included in the study: Truman and Pioneer Brand 25R32. Yield trials were grown in 2011 and 2012 in MI and KY; FHB nurseries were grown both years in OH, IL, IN, and MO. Due to extremely dry conditions in 2012, FHB data was recorded at only two locations: MO and OH. Samples from the MI yield trial were submitted both years to the USDA-ARS Soft Wheat Quality Lab in Wooster, OH for milling and baking quality evaluation.

For incidence, severity, index, FDK (2011-2012), and DON (2011) the mean score for the QTL based resistance class was numerically lower than that of the native class but the difference was rarely significant at $P < 0.05$. QTL based resistance had no measurable effect on yield or test weight; the difference between classes for these traits was non-significant and the top yielding lines were from both classes of resistance. Milling and baking quality of this group of lines was not great, based on 2011 samples from MI. In general, the two classes of resistance differed very little for quality traits. Numerically, QTL - derived resistance was associated with higher flour yield, softer kernels and stronger gluten than was native resistance.

Other soft winter wheat studies have reported successful use of resistance QTL without a negative effect on milling and baking quality. The picture from this study should be clarified when the 2012 quality data become available.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-054. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

MAPPING OF FHB RESISTANCE IN SRW WHEAT CULTIVAR JAMESTOWN

E. Wright[1], C. Griffey[1*], S. Malla[1], D. Van Sanford[2], S. Harrison[3], J.P. Murphy[4], J. Costa[5], G. Milus[6], J. Johnson[7], A. McKendry[8], D. Schmale III[9], A. Clark[2] and N. McMaster[9]

[1]Dept. of Crop and Soil Environmental Sciences, Virginia Tech, Blacksburg, VA 24061; [2]Dept. of Plant Sciences, University of Kentucky, Lexington, KY 40546; [3]Dept. of Plant, Environmental and Soil Sciences, Louisiana State University, Baton Rouge, LA 70803; [4]Dept. of Crop Science, North Carolina State University, Raleigh, NC 27695; [5]Dept. of Plant Science and Landscape Architecture, University of Maryland, College Park, MD 20742; [6]Dept. of Plant Pathology, University of Arkansas, Fayetteville, AR 72701; [7]Dept. of Crop and Soil Science, University of Georgia, Griffin, GA 30223; [8]Department of Plant Sciences, University of Missouri, Columbia, MO 65201; and [9]Dept. of Plant Pathology, Physiology, and Weed Science, Virginia Tech, Blacksburg, VA 24061
[*]Corresponding Author: PH: 540-231-9789, Email: CGriffey@vt.edu

ABSTRACT

A major pathogen of wheat (*Triticum aestivum* L.), Fusarium Head Blight (FHB), is caused by the pathogen *Fusarium graminearum* Schwabe. Infection of wheat with FHB results in yield loss, reduced seed quality, and accumulation of mycotoxins such as deoxynivalenol (DON) and nivalenol (NIV). An important strategy for FHB control is pyramiding multiple resistance genes to provide a broad range of resistance, Type I (resistance to initial infection) and Type II (resistance to spread in wheat spike). The objective of this study is to identify quantitative trait loci (QTL) for FHB resistance in the native soft red winter (SRW) wheat cultivar Jamestown. A total of 77 Jamestown/LA97113UC-124 (JT/LA) $F_{4:6}$ recombinant inbred lines (RILs) lines and 186 Pioneer 25R47/Jamestown (P47/JT) $F_{5:7}$ RILs were evaluated for FHB incidence, severity, index, and concentrations of DON and NIV in three environments (JT/LA:Arkansas, Georgia, Louisiana, and Virginia ; P47/JT: Maryland, North Carolina, and Virginia). Both public and proprietary single nucleotide polymorphism (SNP) markers were used to genotype all of the JT/LA RILs and 42 of the P47/JT RILs at Monsanto Company. A 9k SNP platform was used and about 2,000 polymorphic markers were identified in the JT/LA population and about 250 markers in the P47/JT population. The linkage map was constructed using Map Manager QTX, based on the consensus map provided by the Monsanto Company. Windows Cartographer (WinQTLCart version 2.5) was used to identify possible QTLs. For the Jamestown/LA97113UC-124 population evaluated for FHB in 2011, 108 QTLs were detected on all wheat chromosomes except for 4D, 5D, 6B, 6D, and 7D. There were eight, six, four, and nine QTLs associated with FHB incidence, severity, and DON and NIV content, respectively. Among the 108 QTLs, 13 were consistent in that a given QTL controlled multiple FHB traits and 4 of these QTL were observed across environments. These consistent QTL were located on chromosomes 1A, 1B, 1D, 2A, 3B, 4A, 5A, 5B, 6B, 6D, 7A, and 7B. In 2012, nine QTLs for FHB were detected on chromosomes 1A, 2B, 2D, 3B, 6A, 7A, and 7B in the JT/LA population. For the Pioneer 25R47/Jamestown population, six and four QTLs were identified in 2011 and 2012, respectively, and the QTLs were located on chromosomes 1B, 2B, 3A, and 6A in 2011 and 2A, 3A, 3B, and 6A in 2012. Among the ten QTLs identified, two were found to be consistent across multiple environments and multiple traits. The consistent QTLs were located on chromosome 3A and 6A. In these populations, the RIL genotypes are being further characterized using SSR markers. More informative linkage map and QTL results will be reported with the addition of SSR markers in these populations.

QTL MAPPING IN A DOUBLED HAPLOID POPULATION OF WHEAT TO EXPLORE THE RELATIONSHIP BETWEEN PLANT MORPHOLOGICAL TRAITS AND FUSARIUM HEAD BLIGHT RESISTANCE

Yaopeng Zhou[1], Gina Brown-Guedira[2] and Jose M. Costa[1*]

[1]Department of Plant Science and Landscape Architecture, University of Maryland, College Park, MD 20742-4452; and [2]USDA-ARS, 4114 Williams Hall, Plant Science Research Unit, Raleigh, NC 27695-7620
[*]Corresponding Authors: PH: 301-405-1317, Email: costaj@umd.edu

ABSTRACT

Fusarium head blight (FHB) is a serious wheat disease in major wheat growing regions all over the world. Many reports have been published on genetic analysis of FHB resistance, which is a quantitative trait governed by polygenes. Plant height, spike length, leaf length and leaf width are crucial morphological traits that have been reported to contribute to plant architecture and indirectly affect host FHB resistance and grain yield. In this study, we are interested in the plant morphological QTLs that are coincident with or closely linked to FHB resistance loci and will conduct QTL mapping based on a wheat doubled haploid (DH) population of 124 lines derived from the cross between MD01W233-06-1 and SS8641. MD01W233-06-1, a soft winter wheat germplasm released in 2010, shows resistance to powdery mildew, leaf rust and FHB. SS8641 is a commercial cultivar with broad spectrum rust and mildew resistance but is susceptible to FHB. The DH lines were tested for grain yield, plant height, spike length, flag leaf length and flag leaf width in 2012. The DH population showed a very wide range of variation for these morphological traits in addition to scab resistance. A genetic linkage map with ~1800 SSR and SNP markers was constructed. Data from 2012 was analyzed using Inclusive Composite Interval Mapping (ICIM) methods, which detected 16 potential QTLs explaining 8.2 to 55.8% of the corresponding phenotypic variance with LOD scores greater than 3.0. Three QTLs were detected for flag leaf length and flag leaf width, respectively, with LOD scores ranging from 3.0 to 4.2, 4.5 to 5.6 and corresponding R^2 ranging from 14.1 to 55.8%, 12.0 to 15.9%. One QTL for spike length with LOD=3.4 and R^2=12.1%, six QTLs for plant height with LOD scores ranging from 3.7 to 8.3 and R^2 ranging from 8.2 to 22.7% and three QTLs for grain yield with LOD scores ranging from 3.1 to 3.5 and R^2 ranging from 9.9 to 21.7% were also identified. Detected QTLs for morphological traits will be examined with FHB resistance loci (study in progress) to provide insight into their relationship. Furthermore, the DH population will be evaluated in 2013 at three field locations: Upper Marlboro (MD), Queenstown (MD), and Clarksville (MD) as well as at a research greenhouse at the University of Maryland.

FUSARIUM HEAD BLIGHT REACTIONS OF LANGDON DURUM D-GENOME DISOMIC SUBSTITUTION LINES

X. Zhu[1], S. Zhong[2], S.S. Xu[3] and X. Cai[1*]

Departments of [1]Plant Sciences and [2]Plant Pathology, North Dakota State University, Fargo, ND 58108; and [3]USDA-ARS, Northern Crop Science Laboratory, Fargo, ND 58102
[*]Corresponding Author: PH: (701) 231-7404, Email: xiwen.cai@ndsu.edu

ABSTRACT

An effective source of resistance to Fusarium Head Blight (FHB) like Sumai 3 has not been found in durum wheat. Limited progress has been made in the improvement of FHB resistance in durum by utilizing resistance sources derived from hexaploid wheat and tetraploid relatives of durum. We have observed that FHB resistance QTL exhibited less effectiveness of resistance in durum than hexaploid wheat. It has been anticipated that D-genome chromosomes of hexaploid wheat might play a role in the expression of FHB resistance genes in wheat. Here we report the FHB reaction of a complete set of Langdon (LDN) durum D-genome disomic substitution lines (DSLs), where one pair of A- or B-genome chromosomes of LDN were replaced by one pair of homoeologous D-genome chromosomes from common wheat 'Chinese Spring (CS)'. The 14 DSLs were evaluated for Type II resistance with point inoculation method under three greenhouse environments in two seasons (Fall 2011 and Spring 2012). Homogeneity test indicated there was no significant difference among three greenhouse environments ($p<0.05$). So combined statistical analyses were performed with the DSLs. LDN exhibited a mean FHB severity of 57.47%. The mean FHB severity for each of the DSLs over the three environments ranged from 27.75% for the DSL LDN5D(5A) to 84.42% for LDN6D(6A). FHB severity of LDN5D(5A) was significantly lower than LDN and other DSLs. In addition, LDN5D(5A) had long and slim spikes due to the absence of *Q* gene on chromosome 5A. This modified spike structure might constrain the infection and/or spreading processes of the fungal pathogen and then reduce FHB disease in LDN5D(5A). Additional studies are under way to further determine the effect of this 5D(5A) chromosome substitution on FHB resistance. On the other hand, two DSLs, including LDN6D(6A) and LDN2D(2B), exhibited significantly higher severity than LDN. These results suggest that chromosome 6A and 2B might contain genes enhancing FHB resistance in LDN or chromosome 6D and 2D contain the genes for FHB susceptibility and/or suppression of FHB resistance in LDN background. A better understanding of the effects of these chromosome substitutions on FHB resistance/susceptibility will facilitate the development of durum varieties and germplasm with improved resistance to FHB.

SESSION 4:

PATHOGEN BIOLOGY AND GENETICS

Chairperson: Jin-Rong Xu

GIBBERRELLA ZEAE CHEMOTYPE DIVERSITY ON MODERATELY FHB RESISTANT WHEAT GENOTYPES IN SOUTH DAKOTA

S. Ali*, M. Eldakak, P. Gautam, K. Glover,
J.S. Rohila, J. Gonzalez and W. Berzonsky

Plant Science Department, South Dakota State University, Brookings, SD 57007
*Corresponding Author: PH: (605) 688-6996, Email: Shaukat.ali@sdstate.edu

INTRODUCTION

Gibberrella zeae (anamorph: *Fusarium graminearum*) causes Fusarium head blight (FHB) or scab on small grains worldwide, especially barley and wheat in the USA. The disease impacts wheat production by affecting both yield (poor grain filling and reduced seed test weight) and quality (contaminating grains with mycotoxins). Fungal isolates can be classified primarily into two groups based on the production of 8-Ketotrichothecenes deoxynivalenol (DON) and nivalenol (NIV). In the USA, The population that produces DON is predominant. deoxynivalenol producing isolates can be further grouped as one of two chemotypes, 3-acetyl-Deoxynivalenol (3ADON) or 15- acetyl-Deoxynivalenol (15ADON) based on their trichothecene profile. These mycotoxins are injurious to both human and animal health (Desjardins 2006). The FDA has set 1ppm the maximum contamination limit in the food products fit for human consumption (Schmale and Bergstrom 2003)

The pathogen virulence and aggressiveness pattern can be affected due to the continuous selection pressure of host resistance, excessive use of a single fungicide or fungicides with similar chemistry and adverse environmental conditions. A population shift in *G. zeae* (15ADON to 3ADON) has been observed in the USA, especially in the northern Great Plains (Burlakoti et al. 2008; Gale, 2007), and in Canada (Ward et al. 2008). Several independent studies also indicated that the 3ADON population is more aggressive in scab development and DON production compared to the 15ADON population (Puri and Zhang, 2010; Ward et al. 2008; Ali et al. 2009). The reasons behind the population shift are still not fully understood but host resistance, excessive fungicides application, and change in weather conditions are thought to be the most likely suspects.

OBJECTIVES

1. Analyze *F. graminearum* isolates for their chemotypes recovered from hard red spring wheat genotypes in South Dakota

2. Determine if the host resistance favors 3ADON population over 15ADON population

MATERIALS AND METHODS

FHB samples collection and recovery of Fusarium graminearum isolates - Twenty-four FHB diseased head samples of 10 advanced breeding lines with moderate FHB resistance and two FHB susceptible cultivars 'Oxen' and 'Briggs' were collected from spring wheat breeding nurseries grown at SDSU experimental research Stations near Volga and Watertown in 2012 (Table 1). Five of 10 breeding lines had different genetic backgrounds but all had *Fhb1* gene as source of FHB resistance (Table 1). Ten diseased heads of each genotype were collected from each location. To recover fungal isolates, scabby grains (tombstones) were recovered from each sample. Five tombstones from each sample were randomly selected and plated on half strength potato dextrose agar (PDA) medium in 15 x 100 mm plastic petri plates. Five tombstones of each sample were plated on one plate. The fungal colonies grown out of the plated grains were transferred individually onto new ½ PDA plates.

The identity of fungal isolates was determined based on their colony and spore morphology described in (Nelson et al. 1983). In total 93 *F. graminearum* isolates were recovered from all 24 collected samples. Ten isolates from each genotype (five/location) were recovered and stored in 15% glycerol at -80C in the freezer.

DNA Extraction and PCR-based Chemotyping - To recover DNA from the isolates, all 93 *F. graminearum* isolates were grown individually for 2 days on cellophane membrane placed on ½ PDA plates. Mycelia of each isolate were harvested by scraping the surface of cellophane membrane with a sterile spatula. DNA of all 93 isolates was extracted from the harvested mycelia using the protocol described in Liu et al., (2000). Trichothecene chemotype was determined for all the isolates using the trichothecene specific primers (3CON, 3NA, 3D15A, and 3D3A) (Starkey et al., 2007, Ward et al., 2002). PCR amplification was performed in a C-1000 thermal cycler (BioRad, USA) using amplification steps of 94°C for 2 min, followed by 32 cycles of 94°C for 30s, 52°C for 30s and 72°C for 1 min with final extension of 72°C for 5 min. The PCR amplified products were run on 1.5% (wt/vol) agrose gels and scored with reference to 100 bp DNA ladder (New England Biolabs, USA). The PCR amplification produced bands of 610 and 243bp corresponding to the 15ADON and 3ADON chemotypes, respectively (Fig 1)

RESULTS AND DISCUSSION

Ninety-three of the 120 plated scabby grains produced *F. graminearum.* Twenty of the plated grains were infected with other *Fusarium* species, (i.e., *F. sporotrichioides*, F. a*venaceum* and F. *equiseti*). Recovery of *F. graminearum* from most plated samples seems to indicate that it is still the primary pathogen associated with FHB development in the state. The majority (94%) of isolates were grouped as 15ADON; whereas, 6% of the isolates were grouped as 3ADON (Fig. 1). 3ADON isolates were recovered from both FHB susceptible (n=2) and resistant (n=3) wheat genotypes. The 2012 growing season was generally an FHB disease free year as it was hot and dry; however, occurrence of overnight dew periods provided some opportunity for FHB development on some heads in most of the plots in the breeding nurseries and in the commercial spring wheat field plots visited. The results of this study indicate that FHB moderately resistant cultivars may not have any potential role at least in favoring the recent fungal population shift from 15ADON to 3ADON in South Dakota. Also, the 15ADON population is still the predominant population in the state. However, the presence of a 3ADON population in the state and their higher aggressiveness than the 15ADON population in FHB development and DON production (Puri and Zhong 2010; Ali et al. 2009) warrant the use of 3ADON isolates for screening breeding material for FHB resistance to obtain durable resistant cultivars. More *F. graminearum* isolates, recovered from spring wheat and winter wheat FHB samples collected from breeding nurseries and commercial fields over multiple years, are under investigation to obtain a more complete picture of the fungal population chemotypes present in the state.

ACKNOWLEDGEMENT

The authors thank Richard Geppert and Manali Shirke for their help in samples collection and in the lab for DNA extraction, respectively. This research work was funded by the South Dakota Wheat Commission.

REFERENCES

Ali et al. 2009. Proceedings of the 2009 National Fusarium Head Blight Forum held at Orlando, Florida, USA from 7-9 December. Pp 19-21.

Burlakoti et al. 2008. Phytopathology 98:969-97

Desjardins, A. E. 2006. *Fusarium* mycotoxins chemistry, genetics and biology. The American Phytopathological Society press, St. Paul, Minnesota, USA

Gale et al. 2007. Phytopathology 97:1434-1439.

Liu et al. 2000. Journal of Clinical Microbiology 38:471

Nelson et al. 1983. The Pennsylvania State University Press. Pp226.

Puri, K, and S. Zhong 2010. Phytopathology 100:1007-1014

Schmale III, D. G. and G. C. Bergstrom 2003. The Plant Health Instructor. DOI:10.1094/PHI-I-203-0612-01

Starkey et al. 2007. Fungal Genet. Biol. 44: 1191-1204.

Ward et al. 2008. Fungal Genetics and Biology 45:473-484.

Ward et al. 2002. Natl. Acad. Sci. USA 99:9278-9283.

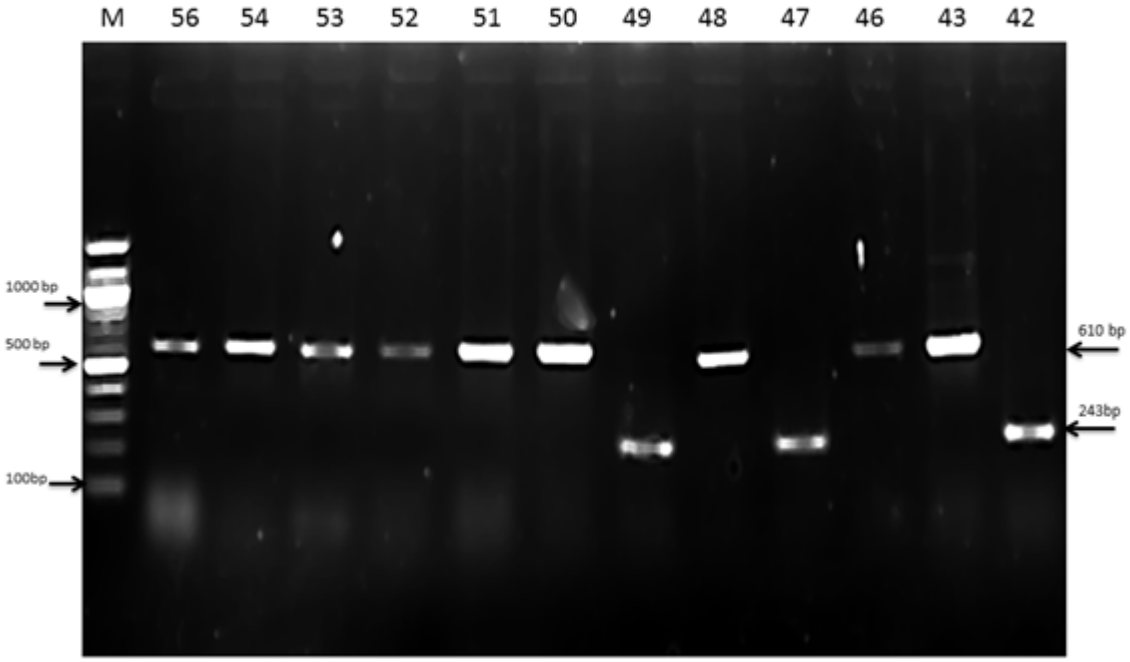

Fig1. Determination of chemotypes of *F. graminearum* isolates using the multiple PCR amplification method (Ward et al. 2008). The bands at 610bp and 243bp amplified from the *F. graminearum* isolates (42 to 56) correspond to the 15ADON and 3ADON chemotypes, respectively. M represents the 100bp DNA marker.

Table1. South Dakota hard red spring wheat genotypes and their pedigree from which *F. graminearum* isolates were recovered and analyzed for their chemotypes in 2012

Wheat genotype	Pedigree	Number of *F. graminearum* isolates tested
SD 4415	SD3934/SD4101/SD3934	3
SD 4417	SD3934/SD3948//Oxen	10
SD 4419*	SD3934/Granger//Briggs	10
SD 4420*	SD3934/Granger//Briggs	4
SD 4421	SD3934/Granger//Briggs	10
SD 4422	SD3934/Granger//Briggs	8
SD 4424	SD3934/Granger//Briggs	4
SD 4426	SD3934/Granger//Briggs	9
SD 4428	SD3934/SD3948//SD3944	10
SD 4429	SD3934/SD4102//Briggs	5
Briggs	BW114/BERGEN//SD3097	10
Oxen*	YW352/SBZ004A	10

*=3ADON isolates were recovered

THE RELATIVE EXPRESSION OF *TRI5* GENE DURING WHEAT-*FUSARIUM GRAMINEARUM* COLONIZATION

C.C. Amarasinghe and W.G.D. Fernando*

Department of Plant Science, University of Manitoba, Winnipeg, R3T2N2, Canada
*Corresponding Author: PH: (204) 474-6072, Email: Dilantha.Fernando@ad.umanitoba.ca

ABSTRACT

TRI5 is the key gene in the deoxynivalenol (DON) biosynthesis pathway. It encodes for the enzyme trichodiene synthase that catalyses the first step in DON biosynthesis. The objective of this study is to examine the level of expression of *TRI5* in resistant and susceptible wheat cultivars after inoculating with *Fusarium graminearum* 3-ADON and 15-ADON isolates. In this study the two wheat cultivars Glenn (rated moderately resistant to FHB) and Roblin (rated highly susceptible to FHB) were grown in the greenhouse and were inoculated with ON-06-39, Q-06-32 (3-ADON) and Q-06-10, ON-06-05 (15-ADON) isolates. The level of expression of *TRI5* gene was evaluated at 0, 6, 12, 24, 48, 72 hrs and 7 days after inoculation. The relative expression of *TRI5* gene was analysed in comparison with the fungal GAPDH house-keeping gene. The expression of *TRI5* gene was initiated at 72 hrs after inoculation and no significant expression was observed before this time period. The level of expression was higher in 7 dai than 72 hai. In this study relative expression of *TRI5* was higher in the cv. Roblin than in cv. Glenn in most of the treatments. It is expected to have a higher expression of *TRI5* gene in highly susceptible cv. Roblin as it showed the highest spread of the pathogen. Also at 7 days, 3-ADON isolates showed higher level of expression than 15-ADON isolates except for one treatment. In several treatments, *TRI5* gene expression differed among the two isolates within the same chemotype group confirming the isolate variation during plant-pathogen interaction. Findings from this study would help in understanding the cross-talk between Wheat-*F. graminearum* during colonization. Further work is being carried out to examine the expression of other genes in DON biosynthetic pathway.

PROFILING TRICHOTHECENE GENOTYPES OF *FUSARIUM GRAMINEARUM* ISOLATED FROM CORN, WHEAT AND POTATOES IN EASTERN CANADA

R.R. Burlakoti[1*], L. Tamburic-Illincic[2], V. Limay-Rios[2], R.D. Peters[3] and P. Burlakoti[2]

[1]Weather INnovations Incorporated, Chatham, Ontario, Canada; [2]University of Guelph, Ridgetown Campus, Ridgetown, Ontario, Canada; and [3]Agriculture and Agri-Food Canada, Charlottetown, Prince Edward Island, Canada
[*]Corresponding Author: PH: 519-352-5334 ext 225, Email: rburlakoti@weatherinnovations.com

ABSTRACT

Fusarium head blight (FHB) and Gibberella ear rot (GER) are economically important diseases of wheat and corn in Eastern Canada and cause millions of dollars in losses. *Fusarium graminearum* is the principal causal agent of both diseases. In addition to huge yield losses, the fungus is responsible for quality degradation and mycotoxin contamination in grain. The fungus is also reported as a storage rot pathogen of potatoes in the United States and Canada. Several studies were previously carried out on population structure and mycotoxin diversity of *F. graminearum* infecting Canadian wheat. However, there are limited studies on the diversity of *F. graminearum* populations collected from corn and potatoes in Canada. The purpose of the present study was to understand the influence of host, cultivars, geography, and weather variables on the diversity of trichothecene profiles of *F. graminearum* isolates in Ontario and eastern Canada. *F. graminearum* isolates were recovered from grain samples of popularly grown wheat cultivars and corn hybrids from diverse geographic locations of south western and central Ontario in 2010 and 2011. As well, *Fusarium graminearum* was isolated from potato samples collected from Manitoba, Quebec, New Brunswick, and Prince Edward Island (PEI) during national potato surveys. A total of 298 single spore isolates of *F. graminearum* were recovered: 54 isolates from wheat, 227 isolates from corn and 17 isolates from potatoes. All isolates were characterized to the species level. Out of 298 *F. graminearum* isolates, 176 isolates (117 from corn, 49 from wheat and 10 from potatoes) were genotyped using *TRI3*- and *TRI-12* based molecular markers. All the tested *F. graminearum* isolates were DON genotypes. Only 3% of isolates from corn and 2% of isolates from wheat were 3-ADON genotypes, and rest of isolates were 15-ADON genotypes. Interestingly, all the *F. graminearum* isolates from potatoes were 3-ADON genotypes. Molecular genotyping of more isolates is in progress. The ability of representative isolates to produce DON, 3-ADON and 15-ADON will be assessed with chemical analysis by growing the fungal isolate in rice culture. This study will provide base-line data on 3-ADON and 15-ADON profiles of *F. graminearum* isolates of corn and potatoes. The outcome will provide information to screen wheat, corn, and potato germplasms against FHB, GER, and Fusarium rot, respectively.

COMPARATIVE GENOMICS REVEALS NEW INSIGHTS INTO THE EVOLUTION OF *FUSARIUM* PATHOGENESIS IN WHEAT

Donald M. Gardiner, Kemal Kazan* and John M. Manners

CSIRO Plant Industry, Inc., Queensland Bioscience Precinct, St Lucia, Brisbane, 4067, Queensland, Australia
*Corresponding Author: PH: 61-7-3214-2678, Email: kemal.kazan@csiro.au

ABSTRACT

Fusarium head blight and crown rot are globally important fungal diseases of cereal crops such as wheat and barley. In Australia, these diseases are caused by two related *Fusarium* pathogens: *Fusarium graminearum* and *F. pseudograminearum*. Our earlier work on the inducers of trichothecene (DON) biosynthesis in *F. graminearum* has led to the identification of a variety of amine compounds that induce the fungal *TRI5* gene involved in DON biosynthesis in *F. graminearum* (Gardiner et al., 2009). We also found the activation of wheat genes encoding polyamine biosynthetic enzymes in infected heads prior to detectable DON accumulation (Gardiner et al., 2010), suggesting that the pathogen exploits this host stress response as a signal for production of trichothecene mycotoxins.

More recently, we have undertaken a comparative genomics approach to help understand how *Fusarium* pathogens cause disease on cereals. For this, we have sequenced the genome *F. pseudograminearum* using next generation sequencing technologies. As expected, the *F. pseudograminearum* genome was highly similar to the previously sequenced genome of *F. graminearum*. Interestingly, however, comparison of the predicted proteins encoded by the *F. pseudograminearum* genome to those from a range of other cereal and non-cereal pathogens revealed genes that have orthologues only in certain cereal pathogens. The fungal deletion mutants for two of these genes showed reduced virulence on wheat, indicating the importance of these genes for fungal virulence (Gardiner et al., 2012). Therefore, the comparative genomics approach appears to be useful for not only studying the evolution of *Fusarium* pathogenesis but also for better understanding of infection strategies used by these pathogens. This latter knowledge may lead to the development of new crop protection strategies.

REFERENCES

Gardiner DM, Kazan K, Manners JM. (2009) Nutrient profiling reveals potent inducers of trichothecene biosynthesis in *Fusarium graminearum*. Fungal Genet. Biol. 46:604-613.

Gardiner DM, Kazan K, Praud S, Torney FJ, Rusu A, Manners JM. (2010) Early activation of wheat polyamine biosynthesis during Fusarium head blight implicates putrescine as an inducer of trichothecene mycotoxin production. BMC Plant Biol. 10:289.

Gardiner DM, McDonald MC, Covarelli L, Solomon PS, Rusu AG, Marshall M, Kazan K, Chakraborty S, McDonald BA, Manners JM. (2012). Comparative pathogenomics reveals horizontally acquired novel virulence genes in fungi infecting cereal hosts. PLoS Pathog. 8:e1002952.

RNA-SEQ REVEALED GENE EXPRESSION DIFFERENCES BETWEEN 3ADON AND 15ADON POPULATIONS OF *FUSARIUM GRAMINEARUM IN VITRO* AND *IN PLANTA*

K.D. Krishna[1], C. Yan[2] and S. Zhong[1*]

[1]Department of Plant Pathology; and [2]Department of Computer Science, North Dakota State University, Fargo, ND
[*]Corresponding Author: PH: (701) 231-7427, Email: shaobin.zhong@ndsu.edu

ABSTRACT

Fusarium graminearum is the major causal agent of Fusarium head blight (FHB), a devastating disease of wheat and barley in North America and worldwide. The fungus produces several trichothecenes [Deoxynivalenol (DON) and its acetylated derivatives, 3-acetyldeoxynivalenol (3ADON) and 15-acetyldeoxynivalenol (15ADON) as well as nivalenol (NIV)], which are harmful to humans and animals. Recent studies showed that the 3ADON-producing isolates dramatically increased in the fungal population and were more aggressive and accumulated more DON in wheat grains than the prevalent 15ADON-producing isolates in North America. However, the genetic and molecular basis for the differences between the two populations is still unclear. In this study, we compared transcriptome of the 3ADON and 15ADON populations *in vitro* and *in planta* using the RNA-seq technology. The *in vitro* RNA samples were isolated from bulked mycelia of 10 3ADON-type isolates or 10 15ADON-type isolates grown on mung bean agar plates for five days. The *in planta* RNA samples were extracted from spikelets of the susceptible spring wheat cultivar 'Briggs', which were harvested at 48, 96 and 144 hours after inoculation (HAI) with equally mixed spore suspension (10^5 spores/ml) of 10 isolates from each population. Two biological samples (replicates) were taken from each treatment or time point, and thus two (1×2) *in vitro* and six (3×2) *in planta* RNA samples were sequenced for each of the fungal populations using the Illumina HiSeq 2000 platform. Total numbers of reads generated from each sample (replicate) ranged from 26.4 to 49.5 million for the 3ADON population and 27.8 to 39.1 million for the 15ADON population. Over 80% of the sequence reads from each of the *in vitro* RNA samples (replicates) mapped to the reference genome sequence of *F. graminearum* (PH-1). However, the percentages of sequence reads mapped to the fungal genome ranged from 5.3 to 13.3% for the *in planta* RNA samples from inoculation with 3ADON isolates and 6.5-8.2% for the RNA samples from inoculation with 15ADON isolates. Pearson's Correlation Coefficient (PCC) between two biological replicates within each treatment was significantly high, ranging from 0.903 to 0.997 ($p<0.0001$). Comparative analyses of *in planta* versus *in vitro* gene expression profiles revealed 2,159, 1,981 and 2,095 genes up-regulated in 3ADON isolates, and 2,415, 2,059, 1,777 genes up-regulated in 15ADON isolates during infection at 48, 96 and 144 HAI, respectively. Of these genes, 633, 526 and 668 were up-regulated at the three time points, respectively, only in the 3ADON population. Among the 65 genes up-regulated at all the three time points, 29 were found to be in the category of unclassified proteins while 25 had functions related to protein synthesis (ribosome biogenesis and translation), amino acid metabolism (aspartate, threonine, tryptophan, and pyruvate family), DNA processing and degradation, polyketides metabolism, and peptide, antigen and GTP binding, and cation transport (H+, Na+, K+, Ca2+, NH4+ etc.). Gene expression profile comparison between the 3ADON and 15ADON population grown *in-vitro* identified a total of 479 genes up-regulated and 801 genes down-regulated in the 3ADON population. Of the 479 up-regulated genes in the 3ADON population, 21.2% and 8.95% of them are involved in functions for C-compound and carbohydrate metabolism, and for polysaccharide, respectively, although a majority

of the genes (58.3%) encode for unclassified proteins. Pair-wide comparisons between the two fungal populations *in planta* revealed 484, 451 and 310 differently expressed genes, respectively, at the three time points. The 3ADON isolates had 186, 87, and 63 genes up-regulated, respectively, at the three time points *in planta*, compared to the 15ADON isolates. Although a majority of these genes (69.3%) are in the category of unclassified proteins, genes involved in C-compound and carbohydrate metabolism, non-vesicular cellular import, cellular transport, allantoin and allantoate transport, C-compound and carbohydrate transport, amino acid metabolism, C-compound and carbohydrate metabolism, secondary metabolism, and detoxification were identified among the others. Our RNA-seq analyses provide a foundation for further understanding of the molecular mechanisms contributing to the higher aggressiveness and DON production of the recently emerged 3ADON population.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-8-067. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

EFFECT OF CLIMATE ON THE DISTRIBUTION OF *FUSARIUM* SPECIES CAUSING CROWN ROT OF WHEAT IN THE PACIFIC NORTHWEST OF THE U.S.

Grant J. Poole[1], Richard W. Sailey[2], Carl Walker[3], David Huggins[4], Richard Rupp[5], John Abatzoglou[6], Kimberly Garland-Campbell[7*] and Timothy C. Paulitz[8]

[1]South Australia Research and Development Institute, Urrbrae, SA, Australia; [2]Columbia Basin Agricultural Research Center, Oregon State University, Pendleton, OR, USA; [3]Pioneer Hibred Intl., Johnston, IA, USA; [4]USDA-ARS Land Management and Water Conservation Research Unit, Pullman, WA, USA; [5]Dept. of Crop and Soil Sciences, Washington State University, Pullman, WA, USA; [6]Dept. of Geography, Univ. of Idaho, Moscow, ID, USA; [7]USDA-ARS Wheat Genetics, Quality, Physiology and Disease Research Unit, Pullman, WA, USA; and [8]USDA-ARS Root Disease and Biological Control Unit., Pullman, WA, USA
[*]Corresponding Author: PH: 509-335-0582, Email: kgcamp@wsu.edu

ABSTRACT

Fusarium crown rot, caused by multiple *Fusarium* spp. including *F. culmorum* and *F. pseudograminearum*, is one of the most widespread root and crown diseases of wheat in the Pacific Northwest (PNW) of the U.S. Fusarium crown rot (FCR) occurrence and distribution has been associated with temperature and precipitation. Our objectives were to characterize crown rot disease severity and distribution throughout the PNW by conducting a survey of 210 fields covering the diverse dryland wheat-producing areas of Washington and Oregon. We used a factor analysis statistical approach to determine the effects of climate and geography on species distribution and disease severity. Climatic variables of mean annual temperature (MAT), mean temperature in the coldest month (MTCM), mean temperature in the warmest month (MTWM), mean annual precipitation (MAP), elevation, soil type and cropping intensity were highly intercorrelated and used in a factor analysis. The factor analysis detected two latent factors that could be used as predictor variables in linear mixed models with repeated measures of FCR disease scores and in generalized linear mixed models for the presence/absence of *Fusarium* spp. Isolates of *Fusarium* spp. were obtained from 99% of 105 fields sampled in 2008 and 97% of 105 fields sampled in 2009. Results of the factor analysis showed that the distribution of *F. pseudograminearum* occurred in a greater frequency in areas on the PNW at lower elevations with lower moisture and higher temperatures, whereas *F. culmorum* occurred in greater frequency from areas at higher elevations with moderate to high moisture and cooler temperatures. The factor analysis approach allowed us to quantify the effects of several environmental and climate variables on disease and species occurence for *Fusarium* spp. in the PNW.

THE 3-ADON AND 15-ADON GENOTYPES OF *FUSARIUM GRAMINEARUM* IN NEW YORK ARE NOT DISCRIMINATED BY PATHOGENIC OR SEXUAL REPRODUCTIVE FITNESS

P. Spolti[1,2], E.M. Del Ponte[1] and G.C. Bergstrom[2*]

[1]Universidade Federal do Rio Grande do Sul, Porto Alegre RS, Brazil; and [2]Cornell University, Department of Plant Pathology and Plant-Microbe Biology, Ithaca NY
[*]Corresponding Author: PH: (607) 255-7849, Email: gcb3@cornell.edu

ABSTRACT

Some recent studies in the northern U.S.A. and Canada have suggested that isolates of *Fusarium graminearum* sensu stricto with the 3-ADON trichothecene chemotype have greater pathogenic fitness on wheat than isolates of the 15-ADON chemotype, enough to displace 15-ADON chemotypes within local populations of the fungus. We tested the hypotheses that contemporary 3-ADON isolates are more fit pathogenically and reproductively than contemporary 15-ADON isolates in New York. A geographically diverse collection of *F. graminearum* sensu stricto was made from spikes of commercially produced wheat in New York in 2011, and the isolates were categorized for B-trichothecene genotype (corresponding to presumptive chemotypes) based on PCR amplification of the *TRI3* and *TRI12* genes. Trichothecene genotypes were recovered in the proportion of 15% 3-ADON isolates and 85% 15-ADON isolates, with no NIV isolates found. Twenty-five isolates each of 3-ADON and 15-ADON genotypes were selected arbitrarily for the fitness study. For the pathogenicity assay, 10 spikes of the susceptible wheat cultivar Norm were inoculated into the central spikelet for each isolate, and Fusarium head blight (FHB) severity (proportion of symptomatic spikelets) was recorded 10 days later. For the sexual reproduction assay, corn stalk segments (3cm x 2.5cm) were prepared, autoclaved and inoculated with each isolate, and incubated in moist chambers. Twenty-one days after inoculation the segments were evaluated for the percent segment area covered with perithecia into four classes (0 = no perithecia, 1 = 1-10%, 2 = 11-30%; 3 = >30%). Following enumeration of perithecia, stalk segments were incubated an additional 48 h and the number of discharged ascospores was estimated based on the number of colonies counted on water-agar plates that were positioned 1.5 cm above two segments. Permutation tests were used to test the effect of isolates and orthogonal contrast to test the association of trichothecene genotype with the three variables of FHB severity, perithecia formed, and ascospores discharged. There was strong evidence (P <0.0001) of the effect of isolate for the three variables analyzed but no evidence of difference between the two genotype groups (P>005). Thus the hypotheses that isolates of the 3-ADON genotype are more fit pathogenically and reproductively than those of the15-ADON genotype were rejected. The mean FHB severity was 41% (range 8-74%) and 40% (range 7-92%) for the 3-ADON and 15-ADON isolates, respectively. The number of isolates in classes 0, 1, 2 and 3 for the production of perithecia among isolates of 3-ADON was 1, 11, 9, 4, respectively, and similar to that observed among isolates of 15-ADON: 1, 10, 9, 5. The number of ascospores per plate ranged from 0 to 100 for the 3-ADON genotypes and from 0 to 97 for the 15-ADON genotypes with a mean of 38 and 34 ascospores per plate, respectively. These results suggest that, within the population of *F. graminearum* infecting wheat in New York, isolates with a 3-ADON genotype do not possess any obvious pathogenic or sexual reproductive advantage over 15-ADON isolates.

TOTAL DEOXYNIVALENOL (DON), 15-ADON AND 3-ADON DETECTED BY GC-MS IN WHEAT GRAIN AFTER INOCULATION WITH TWO *FUSARIUM GRAMINEARUM* CHEMOTYPES AND FUNGICIDE APPLICATION

Lily Tamburic-Ilincic[1*], Chami Amarasinghe[2], Anita Brule-Babeli[2], Dilantha Fernando[2] and Jeannie Gilbert[3]

[1]University of Guelph, Ridgetown Campus, Ridgetown, ON, N0P2C0, Canada; [2]Department of Plant Science, University of Manitoba, Winnipeg, MB, R3T2M9, Canada; and [3]Agriculture and Agri-Food Canada, Cereal Research Centre, Winnipeg, MB, R3T2M9, Canada
[*]Corresponding Author: PH: 519-674-1500 x 63557, Email: ltamburi@ridgetownc.uoguelph.ca

ABSTRACT

Fusarium graminearum (Schwabe) is the principal cause of Fusarium head blight (FHB) in North America, one of the most serious diseases of wheat. Deoxynivalenol (DON) is the most important mycotoxin produced by *F. graminearum* (FG), but 15-acetyl DON (15-ADON) and 3-acetyl DON (3-ADON) analogs are also produced at low levels. The fungicides FOLICUR® (tebuconazole), PROLINE® (prothioconazole), PROSARO® (tebuconazole + prothioconazole) and CARAMBA® (metaconazole) are commonly used to control FHB in Canada. The objective of this study was to investigate the effect of the fungicides on DON, 15-ADON and 3-ADON levels after inoculation with 15-ADON and 3-ADON *F. graminearum* isolates in inoculated, misted wheat plots. Moderately resistant (MR) cultivars 'Alsen' and 'Glenn' were planted in Ridgetown, ON and Winnipeg, MB, respectively. The highly susceptible (HS) cultivar 'Roblin' was also planted at both locations. The cultivars were sprayed with the fungicides at 50% anthesis and inoculated individually with six *F. graminearum* isolates (both chemotypes) two days later. The harvested grain was analyzed for total DON, 15-ADON and 3-ADON using gas chromatography-mass spectrometry (GC-MS).

The highest level of total DON was recorded in 'Roblin' in MB without fungicide application and after inoculation with 3-ADON FG isolates (69.4 ppm in 2009 and 29.9 ppm in 2010). In addition, 1.4 ppm of 3-ADON (2% of total DON) and 0.5 ppm of 3-ADON (1.8% of total DON) were detected in the same samples in 2009 and 2010, respectively. In 2009, in addition to total DON, both analogs (15-ADON and 3-ADON) were detected in 'Roblin' in MB after inoculation with 15-ADON isolates of FG, while only 3-ADON was detected after inoculation with 3-ADON FG isolates. Neither analog was detected in wheat grain from Ontario because of low levels of total DON. Our results indicate that all four fungicides controlled FHB in spring wheat regardless of the *F. graminearum* chemotype inoculated. A higher levels of DON were detected in the HS cultivar compared to the MR cultivars, suggesting that planting wheat with increased level of resistance to FHB combined with fungicide application is the best strategy to lower DON levels in harvested grain.

THE IMPACT OF THE *FUSARIUM GRAMINEARUM* GENOME SEQUENCE ON THE QUEST FOR CONTROL OF HEAD BLIGHT

Frances Trail*

Department of Plant Biology, Plant Biology Lab, 612 Wilson Rd,
Room 342, Michgian State University, East Lansing, MI 48824
*Corresponding Author: PH: 517-432-2939, Email: trail@msu.edu

ABSTRACT

In the last ten years, genome sequences have become available for hundreds of filamentous fungi, including mycotoxigenic *Fusarium* species *Fusarium graminearum*. The availability of genome sequences has stimulated and facilitated research on this important pathogen worldwide. The information has enhanced our ability to dissect the molecular basis of pathogenicity, explore the biosynthesis and regulation of mycotoxins, understand population structure, and elucidate elements of the life cycle. We have, in particular, used this information to understand the life cycle, including development and dispersal of ascospores, overwinter survival in crop residues and biosynthesis of mycotoxins during plant infection and colonization. The results of our research and that of others continue to reveal information on this important agricultural species that can be harnessed to reduce the impact of this mycotoxigenic fungus on grain crops.

SESSION 5:

GENE DISCOVERY AND ENGINEERING RESISTANCE

Chairperson: Nilgun Tumer

LIPID TRANSFER PROTEIN-MEDIATED RESISTANCE TO A TRICHOTHECENE MYCOTOXIN – NOVEL PLAYERS IN FHB RESISTANCE

Anwar Bin Umer[1], John McLaughlin[1], Susan McCormick[2] and Nilgun Tumer[1*]

[1]Department of Plant Biology and Pathology, S.E.B.S, Rutgers University, New Brunswick, NJ; and [2]Bacterial Foodborne Pathogens and Mycology Unit, USDA-ARS-NCAUR, Peoria, IL
[*]Corresponding Author: PH: 848-932-6359, Email: tumer@aesop.rutgers.edu

ABSTRACT

Lipid transfer proteins are a class of basic cysteine rich proteins characterized by an eight cysteine motif backbone with intrinsic antimicrobial activities against bacterial and fungal pathogens. Previously, we identified two type IV nonspecific lipid transfer protein (nsLTP) genes (LTP4.4 and LTP4.5) from screening 250,000 activation-tagged *A.thaliana* seeds. Overexpression of both genes enhanced resistance to trichothecin. We set up yeast as a model system to investigate the mechanism by which the LTP4.4 and LT4.5 mediate resistance to trichothecenes. LTP4.4 and LTP4.5 expression conferred resistance to 2µM and 3µM Tcin in yeast. LTP4.4 provided a greater level of resistance than LTP4.5. In contrast, expression of a different nsLTP (LTP 1.1) did not provide resistance to trichothecin. Moreover, expression of LTP4.4 and LTP4.5 did not provide any resistance to other translation inhibitors, such as cycloheximide, anisomycin or chloramphenicol. These results suggest that resistance to trichothecenes is not a general response, but a feature unique to LTP4.4 and LTP4.5.

Cell fractionation assays showed that while LTP4.4 remained largely in the cytosol, LTP4.5 was primarily associated with the membrane fractions, suggesting a difference in localization of the two nsLTPs. To explore the mechanism of nsLTP-mediated resistance to Tcin, we investigated the effects of Tcin on cytosolic and mitochondrial translation, two known targets of trichothecenes. Cytosolic translation was inhibited significantly (>65%), but mitochondrial translation was inhibited only minimally (<23%) by Tcin in cells overexpressing LTP4.4 and LTP4.5 relative to cells transformed with the vector alone. Reactive oxygen species (ROS) generation, an early time point event during trichothecene toxicity, was also alleviated in yeast overexpressing LTP4.4 and LTP4.5 with less than 2% of the cells generating any significant ROS at 2µM and 3µM Tcin. Taken together, these results suggest a likely role for mitochondria in nsLTP-mediated resistance to trichothecene toxicity.

2012 NORTH DAKOTA TRANSGENIC BARLEY FHB NURSERY REPORT

Lynn S. Dahleen[1*], Robert Brueggeman[2], Tilahun Abebe[3] and Ron Skadsen[4]

[1]USDA-ARS, Fargo, ND; [2]Dept. of Plant Pathology, North Dakota State University, Fargo, ND; [3]Dept. of Biology, University of Northern Iowa, Cedar Falls, IA; and [4]USDA-ARS, Madison, WI
[*]Corresponding Author: PH: 701-239-1384, Email: lynn.dahleen@ars.usda.gov

ABSTRACT

The 2012 North Dakota transgenic field trials consisted of 23 barley lines, tested in three misted and three non-misted replicates. Plots were sown on May 9, 2012 in hill plots with 10 seed per hill spaced at 30 cm, and all plots were inoculated using the grain spawn method at heading. Lines included Conlon, the resistant checks Quest and CI4196, twelve primary transgenic lines derived from Conlon, and four transgenic-null pairs derived from crosses between primary Golden Promise transgenics and Conlon. FHB severity was evaluated approximately three weeks after anthesis, by counting the total and infected number of seed on ten randomly selected spikes per row. DON concentrations in the barley samples were determined by gas chromatography with electron capture detection using the method of Tacke and Casper. FHB and DON data were analyzed by SAS (SAS Institute, Cary, NC) with means adjusted for the nearest checks. Average FHB severity was 16% over all six replicates, 9% in the non-irrigated plots and 23% in the irrigated plots. Average DON contamination was 5.1ppm over all plots with 1.9ppm in the non-irrigated plots and 8.5ppm in the irrigated plots. Standard deviations for DON among the replicates were extremely high, reducing the power of comparisons. These lines will be analyzed again in the 2013 transgenic FHB nursery.

ACKNOWLEDGEMENT

This material is based upon work supported by the U.S. Department of Agriculture. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative.

PROTEOMIC DISSECTION OF NEAR ISOGENIC LINES FOR THE DISCOVERY OF SCAB RESPONSIVE GENES IN WHEAT

Moustafa Eldakak[1], Ansuman Roy[1], Yongbin Zhuang[2], Karl Glover[2], Shaukat Ali[2], Yang Yen[1] and Jai S. Rohila[1,2*]

[1]Department of Biology & Microbiology, and [2]Department of Plant Science, South Dakota State University, Brookings, SD 57007
[*]Corresponding Author: PH: (605) 688-4453, Email: jai.rohila@sdstate.edu

ABSTRACT

Fusarium head blight (FHB) or scab, is a disease of economic importance affecting small grain crops every year causing loss of billions of dollars in grain quality and quantity. In wheat, FHB affects the developing heads directly and has been regarded as a severe threat to U.S. and global food security. The molecular mechanisms that underscore the complex disease etiology leading to the suppression of innate resistance in a susceptible line or keep on maintaining the resistance levels in a resistant line of wheat, caused by scab are not well understood. Given the complexity of breeding for FHB resistance/susceptibility, a very first step is to get the fundamental and comprehensive knowledge for FHB responsive wheat functional proteins after *Fusarium* infection. This knowledge should accelerate the wheat breeding efforts to develop FHB resistant wheat cultivars. In the present investigation, we have evaluated the effect of scab infection on wheat heads of a resistant near isogenic line (NIL) and a susceptible NIL at cellular levels. The young heads of the two NILs, resistant (260-2) and susceptible (260-4), were challenged with *Fusarium* and the infected heads were subjected to 2D-DIGE analysis for the identification of *Fusarium* responsive proteins in wheat. A total of 80 protein spots were recorded displaying significantly differential expression on polyacrylamide gel. These protein spots were cut, trypsin digested and the protein was identified through MALDI-TOF mass spectrometry. Further, using the Gene Ontology (GO), functional pathways of these altered proteins discovered several up- and down- regulated biological processes and plant cellular components and organelles of wheat. Analysis of the various pathways affected in wheat plants by the *Fusarium* infection is done. This study shows that, there are significant functional differences in the regulation of host proteins that could be a cause and/or result of a successful or a defeated scab infection. In this study, combined use of a proteomic platform and GO analysis facilitated a better understanding of host-pathogen interactions at cellular levels.

A NOVEL SOURCE OF FUSARIUM HEAD BLIGHT RESISTANCE DERIVED FROM *ELYMUS TSUKUSHIENSIS*

B. Friebe[1*], J. Cainong[1], P. Chen[2], W.W. Bockus[3], and B.S. Gill[1]

[1]Wheat Genetic Resources Center and Department of Plant Pathology, Kansas State University; [2]The National Key Laboratory of Crop Genetics and Germplasm Enhancement, Nanjing Agricultural University, Nanjing Jiangsu, PR China; and [3]Department of Plant Pathology, Kansas State University
[*]Corresponding Author: PH: (785) 532-2364, Email: friebe@ksu.edu

ABSTRACT

Elymus tsukushiensis Honda (2n=6x=42, $S^{ts}S^{ts}H^{ts}H^{ts}Y^{ts}Y^{ts}$, syn. *Roegneria kamoji* C. Koch) is a perennial cross-pollinating hexaploid species native to China, Korea, and Japan. *E. tsukushiensis* is a distantly related wild relative of bread wheat and a source for resistance to Fusarium head blight (FHB). Previously, we reported the production and characterization of wheat-*E. tsukusiensis* chromosome addition lines and showed that the disomic addition having a group-1 *E. tsukushiensis* chromosome, $1E^{ts}$#1, or a wheat- *E. tsukushiensis* translocation chromosome TW·$1E^{ts}$#1S added to the wheat genome, conferred resistance to FHB.

We used *ph1b*-induced homoeologous recombination to produce wheat- *E. tsukushiensis* recombinants. The screening of 488 progenies of plants homozygous for *ph1b* and heterozygous for TW·$1E^{ts}$#1S identified one distal TWL·WS-$1E^{ts}$#1S and one interstitial TiWL·WS-$1E^{ts}$#1S-WS recombinant. Stocks homozygous for both recombinant chromosomes were recovered, conferred type-2 resistance to FHB after point inoculation in a greenhouse test, and may be used in cultivar improvement.

ETHYLENE-SIGNALING IS ESSENTIAL FOR BASAL RESISTANCE TO FUSARIUM HEAD BLIGHT IN WHEAT

Megan E. Gillespie[1], Amanda S. Brandt[1] and Steven R. Scofield[1,2*]

[1]Department of Agronomy, Purdue University, 915 West State Street, West Lafayette, IN, 47907-2054; and [2]USDA-ARS, Crop Production and Pest Control Unit, 915 West State Street, West Lafayette, IN, 47907-2054
[*]Corresponding Author: PH: 765-494-3674, Email: scofield@purdue.edu

ABSTRACT

The role of ethylene (ET)-signaling in the mechanism of resistance to Fusarium head blight (FHB) has been controversial. Expression profiling analyses have identified induction of ET-signaling within the first 6-hours after challenge by *Fusarium graminearum* as a significant event correlated with resistance. However, another study employing an *Arabidopsis* model system and experiments in wheat concluded that ET-signaling was essential for FHB susceptibility. To further address this controversy we employed a virus-induced gene silencing (VIGS) system to silence genes required for ET-biosynthesis and signaling in an FHB resistant wheat genotype. These studies indicated that silencing genes encoding either S-adenosyl-methionine synthetase or an ET-responsive transcription factor, TaERF7-1, caused FHB resistant plants to become susceptible. These results were confirmed by methods independent of VIGS. Inhibition of ET-receptors by treatment with 1-methylcyclopropene caused FHB resistant plants to become susceptible and susceptible genotypes to develop significantly increased disease. Conversely, treatment of FHB susceptible plants with the chemical precursor of ET, 1-aminocyclopropane-1-carboxylic acid (ACC), at a concentration sufficient to induce the expression of TaERF7-1, resulted in increased resistance to FHB. The fact that these studies indicate manipulation of ET signaling affects FHB interactions of both resistant and susceptible genotypes indicates that ET signaling is controlling a component of basal defense. Taken together, these results strongly support ET-signaling as having an essential role in activating basal defense against FHB in wheat.

IDENTIFYING FHB RESISTANCE GENES IN WHEAT USING A NEXT-GENERATION SEQUENCING APPROACH

Anna Hofstad[1], Haiyan Jia[1], Benjamin P. Millett[1], Eduard Akhunov[2] and Gary J. Muehlbauer[1*]

[1]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108; and [2]Department of Plant Pathology, Kansas State University, Manhattan, KS 66506
[*]Corresponding Author: PH: 612-624-2755, Email: muehl003@umn.edu

ABSTRACT

The *Fhb1* QTL on wheat chromosome 3BS confers type II resistance to Fusarium Head Blight (FHB). To gain a better genetic understanding of the *Fhb1* QTL locus, a near-isogenic line (NIL) pair carrying the resistant and susceptible alleles for *Fhb1* was subjected to SNP genotyping and gene expression analysis. We genotyped the NIL pair with 9,000 SNPs and found the lines to be 98% identical. Of the SNPs that could be mapped, 132 SNPs map to the 3BS region and 40% of the total SNP differences were found on chromosome 3BS. We established three experiments to examine gene expression in the NIL pair and to identify candidates for the *Fhb1* gene. We used next-generation sequencing of RNA from the following experiments to obtain the gene expression data: (1) point inoculation of spikelets with *Fusarium graminearum* and sampling the inoculated spikelets at 96 hours after inoculation; (2) point inoculation of deoxynivalenol (DON) or sterile water and sampling the inoculated spikelets at 12 hours after inoculation; and (3) point inoculation of *F. graminearum* and sampling of the rachis at 96 hours after inoculation. For all experiments at least 100 million sequencing reads were obtained for each genotype. For all analyses, differential expression is defined with at least a 2-fold change in expression and a q-value less than 0.05. For experiment #1, we identified 5,973 sequences that showed differential expression between the two genotypes. For experiment #2, we identified 2,210 sequences that showed differential expression between the two genotypes when DON inoculated and 866 sequences that showed differential expression when water inoculated. For experiment #3, we identified 4,771 sequences that showed differential expression between the two genotypes. We also mapped the sequencing reads from experiment #1 and experiment #3 to the *F. graminearum* transcripts to identify genes that are being expressed by the fungus during infection. For experiment #1, we identified 137 transcripts that showed differential expression between the two genotypes. For experiment #3, we identified 419 transcripts that showed differential expression between the two genotypes. Results of the differentially expressed genes will be presented.

IDENTIFICATION AND CHARACTERIZATION OF BARLEY GENES THAT PROVIDE RESISTANCE TO TRICHOTHECENES

Yadong Huang[1], Sanghyun Shin[1], Benjamin P. Millett[1], Xin Li[1], Gerhard Adam[2], Susan McCormick[3], Kevin P. Smith[1], Brian J. Steffenson[4] and Gary J. Muehlbauer[1*]

[1]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108, USA; [2]Center of Applied Genetics, BOKU-University of Natural Resources and Applied Life Sciences, Muthgasse 18, A-1190 Vienna, Austria; [3]Mycotoxin Research Unit, USDA-ARS-NCAUR, Peoria, IL 61604, USA; and [4]Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108, USA
[*]Corresponding Author: PH: (612) 624-2755; E-mail: muehl003@umn.edu

ABSTRACT

Developing transgenic barley and wheat cultivars with increased trichothecene resistance is an important strategy to cope with Fusarium head blight (FHB). We have previously conducted several RNA microarray experiments using barley spikes inoculated with *Fusarium graminearum* or deoxynivalenol (DON). A number of up-regulated transcripts were identified and selected for further characterization. These genes are potentially involved in the metabolism or transport of trichothecenes, which include UDP-glucosyltransferases, glutathione-S-transferases, cytochrome P450s, ABC transporters and an epoxide hydrolase. We used a rapid *in planta* assay to test their efficacy against trichothecenes by overexpressing them in *Arabidopsis thaliana* and growing transgenic plants on media containing trichothecenes. The HvUGT13248 gene has been shown to confer resistance to DON in yeast, Arabidopsis and wheat. By re-sequencing and association analysis, a single nucleotide polymorphism (SNP) which causes a nonsynonymous mutation in the conserved substrate binding domain of HvUGT13248 was identified to be associated with FHB susceptibility. Transgenic Arabidopsis plants overexpressing this HvUGT13248 allele, as well as other aforementioned genes, are being tested on DON media and the results will be reported.

TRANSPOSON MUTAGENESIS FOR IDENTIFICATION OF STRESS RESPONSIVE GENES IN CEREALS

Ravneet Kaur, Surinder Singh and Jaswinder Singh*

Plant Science Department, McGill University, Ste Anne De Bellevue, QC, H9X3V9, Canada
*Corresponding Author: PH: (514) 398-7906, Email: Jaswinder.Singh@mcgill.ca

ABSTRACT

Wheat and Barley are two major food grain crops around the globe, which feed a large population of the world. Fusarium head blight (FHB) is an epidemic disease of wheat and barley causing heavy economic losses to farmers due to yield decreases. Mycotoxin produced by *Fusarium* makes these useless for flour and malt products. Varieties resistant to FHB are a matter of high priority in many areas where they are grown, but the complex nature of resistance makes this a highly challenging task. In barley, two major quantitative trait loci's (QTL) have been identified viz. QTL1 and QTL2 on chromosome 6H and 2H respectively which have a large effect on kernel discoloration. The resistant allele of QTL2 decreases the occurrence of head blight by nearly 50% in varieties in which it is present thus proving its importance. Efforts have been made to clone important QTL for better understanding of the mechanisms involved for FHB tolerance. Maize *Ac/Ds* system is one of the important tools that can be utilized for dissecting and saturating QTL through saturation mutagenesis. Previous and ongoing mapping studies in our lab indicate an added advantage of *Ds* transpositions, in gene rich linked positions; making this technique appropriate to dissect FHB QTL. Currently, our main focus is to saturate QTL2 region using maize *Ds* elements eventually facilitating identification and characterization of genes associated with FHB resistance. Plants with single *Ds* insertions (TNPs), mapping near QTL of interest are important vehicles for gene identification through re-activation and transposition of *Ds*. Recently we have reported that the frequency of Ds reactivation is higher using *in-vitro* transformation methods as compared to conventional breeding. Thus, *Ds* elements from TNP 41 (mapped near QTL2) will be re-activated by transforming the immature embryos with construct containing *Ac*TPase. The purpose is to identify phenotypes, morphology of which may be associated with FHB tolerance.

GENETIC AND GENOMIC APPROACHES FOR MANAGING *FUSARIUM* PATHOGENS CAUSING HEAD BLIGHT AND CROWN ROT IN WHEAT

Kemal Kazan*, Tim Fitzgerald, Chunji Liu, Mick Ayliffe and John Manners

CSIRO Plant Industry, Queensland Bioscience Precinct, St Lucia, Brisbane, 4067, Queensland, Australia
*Corresponding Author: PH: 61-7-3214-2678, Email: kemal.kazan@csiro.au

ABSTRACT

Fusarium head blight and crown rot are globally important fungal diseases of cereal crops such as wheat and barley. In Australia, these diseases are caused by two related *Fusarium* pathogens: *Fusarium graminearum* and *F. pseudograminearum*. We are employing genetic and genomic approaches to minimize crop losses by these pathogens through an improved understanding of pathogen biology and host plant resistance. In this talk, an overview of the work being conducted in CSIRO Plant Industry, Australia will be presented. Firstly, to better understand the weaponry used by the pathogen, we are comparatively analyzing the genomes of cereal infecting *Fusarium* spp. This topic will be covered in detail in *Pathogen Biology and Genetics* session of the Forum. Secondly, we are employing forward and reverse genetic approaches in wheat and Brachypodium to identify host factors that promote disease resistance or susceptibility. In particular, we are exploring the inactivation of susceptibility genes as a strategy to achieve novel resistance. Initially, wheat lines lacking selected putative susceptibility genes were identified within a mutant population and are currently being tested for disease resistance. Thirdly, quantitative trait loci (QTL) that confer *Fusarium* resistance in wheat are being identified and incorporated into elite germplasm. Efforts are also underway to clone and study the mode of action of these QTL. Finally, we have developed a high-throughput wheat transformation technique that allows testing the efficacy of large number of transgenes rapidly. The complementary approaches being explored should assist with development of wheat germplasm that is highly resistant to *Fusarium* spp.

TESTING TRANSGENIC SPRING WHEAT AND BARLEY LINES FOR REACTION TO FUSARIUM HEAD BLIGHT: 2012 FIELD NURSERY REPORT

Koeritz, E.J.[1], Elakkad, A.M.[1], Dahleen, L.S.[2], Skadsen, R.[6], Abebe, T.[7], Shah, J.[3], Nalam, V.J.[3], Klossner, G.[3], Tumer, N.[4], Di, R.[4], Muehlbauer, G.J.[5], Li, X.[5], Shin, S.[5] and Dill-Macky, R.[1*]

[1]Department of Plant Pathology, University of Minnesota, St. Paul, MN; [2]USDA-ARS, RRVARC, Fargo, ND; [3]Department of Biological Sciences, University of North Texas, Denton, TX; [4]Department of Plant Biology and Plant Pathology, Rutgers University, New Brunswick, NJ; [5]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN; [6]USDA-ARS, Madison, WI; and [7]Department of Biology, University of Northern Iowa, Cedar Falls, IA

[*]Corresponding Author: PH: (612) 625-2227, Email: ruthdm@umn.edu

ABSTRACT

The 2012 field screening nursery consisted of 42 wheat and 24 barley entries evaluated in side by side experiments. Entries within each species experiment were arranged in a randomized complete block design with four replications in a field located at UMore Park, Rosemount MN. Trial entries, and untransformed controls, were submitted by the University of North Texas (5+1 wheat), Rutgers University (7+1 wheat), the University of Minnesota (19+5 wheat), and the USDA (20+1 barley). Lines with known reactions to Fusarium head blight (FHB) were also included as checks. The wheat checks used were the moderately resistant Alsen, the susceptible cultivars Wheaton and Roblin, and a non-inoculated Wheaton check. The barley checks were the moderately resistant Quest and the susceptible cultivars, Robust and Stander. Individual plots were 2.4 m long single rows. The trial was planted on May 22, 2012. All plots, except a non-inoculated Wheaton check, were inoculated twice. The first inoculation, 6 July 2012, was applied at anthesis for wheat and at head emergence for barley. The second inoculation was applied three days after the initial inoculation (dai) for each plot. The inoculum was a composite of 30 *F. graminearum* isolates at a concentration of 100,000 macroconidia ml-1 with Tween 20 (polysorbate) added at 2.5 ml.L-1 as a wetting agent. The inoculum was applied using a CO2-powered backpack sprayer fitted with a SS8003 TeeJet spray nozzle with an output of 10ml.sec-1 at a working pressure of 275 kPa. Mist-irrigation was applied from before the first inoculation on July 5 through July 25 to facilitate FHB development. FHB incidence and severity were assessed visually 19 dai for wheat and 14 d.a.i. for barley on 20 arbitrarily selected heads per plot. FHB incidence was determined by the percentage of spikes with visually symptomatic spikelets of the 20 heads observed. FHB severity was determined as the percentage symptomatic spikelets of the total of all spikelets observed. Plots were harvested at maturity on August 13 (barley) and August 22 (wheat). Fifty (barley) and 50 (wheat) heads where harvested from each plot, threshed and the seed cleaned manually. The wheat sub-samples were used to determine the percentage of visually scabby kernels (VSK) and then all samples (wheat and barley) were ground and submitted for deoxynivalenol (DON) analysis. The data indicated that resistance was expressed in some of the transformed lines.

ACKNOWLEDGEMENTS AND DISCLAIMER

We would like to acknowledge Beheshteh Zargaran, Alencar Xavier, Rebecca Schneider, Jared Schuster and Vadym Matyash for their assistance in completion of the work reported. We would also like to acknowledge Dr. Yanhong Dong for conducting the mycotoxin analysis.

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-9-069. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

TRANSGENIC WHEAT CARRYING A BARLEY UDP-GLUCOSYLTRANSFERASE EXHIBIT HIGH LEVELS OF FUSARIUM HEAD BLIGHT RESISTANCE

Xin Li[1], Sanghyun Shin[1,7], Ruth Dill-Macky[3], Franz Berthiller[4], Thomas Clemente[5], Susan McCormick[6] and Gary Muehlbauer[1,2*]

[1]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108, USA; [2]Department of Plant Biology, University of Minnesota, St. Paul, MN 55108, USA; [3]Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108, USA; [4]Department of Agrobiotechnology (IFA-Tulln), University of Natural Resources and Life Sciences, Vienna, 3430 Tulln, Austria; [5]Department of Agronomy and Horticulture, University of Nebraska, Lincoln, NE 68588, USA; [6]USDA-ARS, Bacterial Foodborne Pathogen and Mycology Research Unit, Peoria, IL 61604, USA; and [7]Present address: National Institute of Crop Science (NICS), RDA, Suwon 441-857, Korea
[*]Corresponding Author: PH: (612) 624-2755, Email: muehl003@umn.edu

ABSTRACT

Fusarium head blight (FHB) is a devastating disease of wheat and barley, mainly caused by *Fusarium graminearum*, leading to huge economic losses worldwide. During infection, the fungal pathogen produces trichothecene mycotoxins, such as deoxynivalenol (DON), that increase fungal virulence. Moreover, grain products contaminated with trichothecenes threatens the health of humans and animals that consume them. Previous work had identified a barley UDP-glucosyltransferase (HvUGT13248) gene that exhibited resistance to DON via the conversion to DON-3-O-glucoside (D3G) in transgenic yeast and *Arabidopsis*. We developed transgenic wheat lines constitutively overexpressing the HvUGT13248 gene in the background of cultivar Bobwhite and CB037. We performed point-inoculation tests in the greenhouse for three seasons (2011 spring, 2011 fall and 2012 spring) and found that transgenic wheat exhibited significantly higher type II resistance compared with the untransformed parental lines. Moreover, in field tests HvUGT13248-overexpressing wheat lines also showed significantly less disease severity compared to the untransformed controls. To assess the mechanism of resistance, we inoculated plants with DON and examined the concentration of DON and D3G from 1-21 days after inoculation. HvUGT13248-overexpressing wheat plants converted DON to D3G more rapidly to a higher extent than Bobwhite, indicating that the barley HvUGT13248 gene provides FHB type II resistance in transgenic wheat by converting DON to D3G.

ACTIVATION TAGGING IN *ARABIDOPSIS* IDENTIFIES TWO NOVEL NON-SPECIFIC LIPID TRANSFER PROTEINS WHICH PROVIDE ENHANCED RESISTANCE TO A TRICHOTHECENE MYCOTOXIN

John E. McLaughlin[1], Mohamed Anwar Bin-Umer[1], Thomas Widiez[1], Emily Salmon-Denikos[2], Debaleena Basu[1], Susan McCormick[3], Brian Gregory[2] and Nilgun E. Tumer[1*]

[1]Department of Plant Biology and Pathology, School of Environmental and Biological Sciences, Rutgers University, New Brunswick, NJ; [2]Department of Biology, University of Pennsylvania, Philadelphia, PA; and [3]Bacterial Foodborne Pathogens and Mycology Unit, USDA-ARS-NCAUR, Peoria, IL
[*]Corresponding Author: PH: 848-932-6359, E-mail: tumer@aesop.rutgers.edu

ABSTRACT

Trichothecene mycotoxins are potent virulence factors produced by *Fusarium graminearum* during wheat and barley infection leading to the development and spread of Fusarium Head Blight (FHB). They inhibit cytosolic and mitochondrial protein synthesis in addition to having other complex cytotoxic effects. A genetic screen in *Arabidopsis* was undertaken to identify genes that provide resistance to trichothecenes. Approximately 250,000 activation tagged M2 generation *Arabidopsis* seeds were screened for resistance to trichothecin (Tcin), a type B trichothecene, and several lines from this population were identified that showed resistance. These plants were able to form roots on 4 µM Tcin, a concentration which severely restricts root initiation and elongation of the Col-0 wild type following germination. Characterization of one of these resistant lines using qRT-PCR identified an activation genotype, *Arabidopsis thaliana resistant root formation1* (*AtTRRF1)*. In *AtTRRF1*, two closely linked novel non-specific lipid transfer protein (nsLTP) genes, *LtpIV.4* and *LtpIV.5*, were found to be overexpressed compared to the wild-type control. Both proteins are classified as type IV nsLTPs, a largely uncharacterized class of nsLTPs with limited structural and functional information. Overexpression of both *LtpIV.4* and *LtpIV.5* independently in *Arabidopsis* confirmed resistance to trichothecin based on differences in the ability to form roots when grown on solid media containing 4 µM Tcin. Overexpression of *LtpIV.4* in *Arabidopsis* induced a high level (72.7% ± 10.5) of tolerance, as measured by the percentage seedlings that develop roots when grown on media containing 4 µM Tcin, as compared with control plants (5.6 ± 2.7). Overexpression of *LtpIV.5* induced a moderate level (58.6% ± 11.0) of tolerance. Expression of *LtpIV.4:GFP* and *LtpIV.5:GFP* was examined by transient expression in tobacco leaves and in the transgenic *Arabidopsis* lines by confocal microscopy. The GFP tagged *LtpIV.4* and *LtpIV.5* both localized near the cell wall, suggesting that they are likely targeted to the apoplast. In addition, coexpression analysis using an ER-mcherry marker indicates that *LtpIV.4* may also localize to the ER. This study identified two novel nsLTPs which function to provide enhanced resistance to a trichothecene mycotoxin in plants.

DEVELOPING FUSARIUM HEAD BLIGHT RESISTANT WHEAT

Muehlbauer, G.J.[1,2*], S. Shin[2], X. Li[2], J. Boddu[2], S. Heinen[2], J.A. Torres-Acosta[3], M.P.K. Paris[3], W. Schweiger[4], T. Clemente[5], R. Dill-Macky[6], S. McCormick[7], M. Lemmens[8], F. Berthiller[8] and G. Adam[3]

[1]Department of Plant Biology, University of Minnesota, St. Paul, MN 55108; [2]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108; [3]Department of Applied Genetics and Cell Biology, University of Natural Resources and Life Sciences, 3430 Tulln, Austria; [4]Department for Agrobiotechnology - IFA-Tulln, Institute for Biotechnology in Plant Production, University of Natural Resources, 3430 Tulln, Austria; [5]Department of Agronomy and Horticulture, University of Nebraska, Lincoln, NE 68588; [6]Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108; [7]USDA-ARS, Bacterial Foodborne Pathogens and Mycology Research Unit, Peoria, Illinois 61604, USA; and [8]Department for Agrobiotechnology - IFA-Tulln, Center for Analytical Chemistry (Christian Doppler Laboratory for Mycotoxin Metabolism), University of Natural Resources, 3430 Tulln, Austria

[*]Corresponding Author: PH: 612-624-2755, Email: muehl003@umn.edu

ABSTRACT

Fusarium head blight (FHB), caused by *Fusarium graminearum*, is a major disease problem in wheat and barley around the world. During infection, *F. graminearum* produces a host of trichothecene mycotoxins, such as deoxynivalenol (DON), that act as virulence factors and cause a reduction in grain quality. Therefore, developing approaches to detoxify trichothecenes will serve the dual function of increasing disease resistance and improving grain quality. Numerous gene expression experiments were conducted to identify genes that are differentially expressed in barley and wheat after *F. graminearum* inoculation or treatment with DON. Previous work in *Arabidopsis thaliana* has shown that a UDP-glucosyltransferase can detoxify trichothecenes. We identified a set of barley UDP-glucosyltransferases that exhibited homology to the *Arabidopsis* gene. Examining these genes resulted in the identification of the barley UGT (*HvUGT13248*) gene that provided resistance to DON in yeast and *Arabidopsis*. Resistance to DON was shown to be via conjugation of DON with UDP glucose to form DON-3-O-glucoside (D3G). Transgenic wheat overexpressing *HvUGT13248* were developed in the FHB susceptible Bobwhite and CB037 backgrounds. These transgenics exhibited a statistically significant increase in type II resistance compared to the nontransgenic controls. In several of the lines the level of type II resistance was equivalent to that observed in the Sumai3 genotype. Field screening of the transgenic wheat showed a statistically significant decrease in disease severity compared to non transgenic controls. As in yeast and *Arabidopsis*, resistance in the transgenic wheat appears to be via conversion of DON to D3G. Our results show that developing wheat with the increased capacity to detoxify DON results in increased FHB resistance.

EXPRESSION-QTL MAPPING IN WHEAT TO IDENTIFY REGULATORY HOTSPOTS INVOLVED IN THE RESISTANCE TO *FUSARIUM GRAMINEARUM*

M. Samad-Zamini, W. Schweiger*, E. Sam, G. Siegwart, B. Steiner, M. Lemmens and H. Buerstmayr

BOKU-University of Natural Resources and Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, Konrad Lorenz Str. 20, A-3430 Tulln, Austria
*Corresponding Author: PH: 43-2272-66280-205, Email: wolfgang.schweiger@boku.ac.at

ABSTRACT

Expression QTL studies provide a technique to identify genetic markers (transcript derived marker, TDM), which can be used to generate a genetic map and subsequently identify eQTL based on differences in transcript abundance. This approach allows validating reported phenotypical QTL and simultaneously identifying candidate genes that are encoded or governed by these QTL.

In wheat more than 200 QTL have been reported to contribute to resistance against Fusarium head blight. Most are minor contributors, although (prominently *Fhb1*, located on 3BS and *Qfhs.ifa-5A* on 5AS) explain up to 25 % of the observed resistance. To date, none of these QTL has been cloned and characterized. To characterize reported and newly identified QTL on the transcriptome level, we performed an eQTL study using a population of 200 doubled haploid (DH) lines segregating for *F. graminearum* resistance. This population derives from the resistant line CM82036 (derived from Sumai 3) and the susceptible European spring wheat cultivar Remus. Six central spikelets were inoculated with a Fusarium spore suspension at anthesis and samples were taken at two time points after inoculation. RNA was hybridized onto a custom-build microarray (Agilent 8x60k), comprising 44.000 wheat genes, several hundred wheat candidate genes, that have been reported responsive to *Fusarium* in literature and the entire transcriptome of *Fusarium graminearum* (ca. 14.000 genes). In total, we hybridized about 500 microarrays. We detected 2240 transcripts that show differential abundance between the parental lines. These transcripts and differential transcripts that are not differentially accumulating between the parental lines (due to transgressive segregation) are used as markers. At the time of abstract submission data analysis and QTL analysis as well as the identification of cis- and trans-regulated eQTL corresponding to each time point are still ongoing. These results, the identified regulative hotspots and related genes are presented.

FUNCTIONAL GENOMICS OF UDP-GLUCOSYLTRANSFERASES: HETEROLOGOUS EXPRESSION IN YEAST TO TEST FOR DEOXYNIVALENOL DETOXIFICATION CAPABILITY OF CANDIDATE GENES

W. Schweiger[1*], M.P. Kovalsky Paris [2], G. Wiesenberger [2], F. Berthiller [3], M. Lemmens[1], S. Shin[4], J.A. Torres Acosta[2], G.J. Muehlbauer[4] and G. Adam[3]

[1]Institute for Biotechnology in Plant Production, [2]Department of Applied Genetics and Cell Biology, [3]Center for Analytical Chemistry, Department of Agrobiotechnology, BOKU-University of Natural Resources and Life Sciences Vienna, IFA Tulln, 3430 Tulln, Austria; and [4]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108, USA

[*]Corresponding Author: PH: 43-2272-66280-205, Email: wolfgang.schweiger@boku.ac.at

ABSTRACT

The plant pathogenic fungus *Fusarium graminearum* produces the trichothecene toxin deoxynivalenol (DON). This protein biosynthesis inhibitor has been shown to be a virulence factor in wheat required for spread of infection. Resistance to the spreading of *Fusarium* is mediated by the ability of the plant to glucosylate DON into the non-toxic conjugate DON-3-O-glucoside by UDP-glucosyltransferases (UGT). The first gene encoding an enzyme with this activity from a monocotyledonous plant was recently described (HvUGT13248, Schweiger *et al.* 2010). Since the genomes of both wheat and barley have not yet been fully sequenced, we investigated the homologs of HvUGT13248 in the sequenced genome of the monocot model species *Brachypodium distachyon* (Bd) and the genomes of rice and sorghum. *Brachypodium* is closely related to wheat and barley and should therefore provide an insight into the architecture of wheat and barley UGTs. The gene family of Bd UGTs consists of 177 predicted genes. We characterized the cluster of six Bd UGTs with the highest amino acid sequence similarity to HvUGT13248 by expression in yeast. Only two of the candidate Bd UGTs were able to glucosylate DON to DON-3-O-glucoside. Also from the cluster of rice genes only one gene is capable of detoxifying DON. This is also the case for the single copy gene from *Sorghum*. Seemingly, the UGT genes undergo rapid evolution, and due to different copy numbers in gene clusters it is difficult to identify true orthologues.

Overexpression of a DON glucosylating UGT should increase DON resistance in plants. Yet, UGTs have also been shown to be capable of altering the activity of plant hormones, such as brassinosteroids, by glucosylation, which in turn leads to dwarfing. We tested HvUGT13248 and the two DON detoxifying Bd UGTs for their ability to glucosylate the brassinosteroid castasterone in a yeast assay. No castasterone-glucoside formation was detectable in yeast expressing the three candidate UGT genes. Overexpression of the barley HvUGT13248 gene in *Arabidopsis thaliana* led to increased DON resistance of seedlings without unwanted side effects such as dwarfing (Shin *et al*., 2012).

REFERENCES

Schweiger, W., *et al* (2010) Validation of a candidate deoxynivalenol-inactivating UDP-glucosyltransferase from barley by heterologous expression in yeast. Mol. Plant-Microbe Interact. 23, 962-76.

Shin, S., *et al* (2012) Transgenic *Arabidopsis thaliana* expressing a barley UDP-glucosyltransferase exhibit resistance to the mycotoxin deoxynivalenol. J. Exp. Bot. 63:4731-40.

TRANSCRIPTOMIC CHARACTERIZATION OF THE *FUSARIUM* RESISTANCE QTL *FHB1* AND *QFHS-IFA.5A*

W. Schweiger[1*], B. Steiner[1], C. Ametz[1], G. Siegwart[1], G. Wiesenberger[4], F. Berthiller[2], M. Lemmens[1], H. Jia[3], G. Adam[4], G.J. Muehlbauer[3], D.P. Kreil[5] and H. Buerstmayr[1]

[1]BOKU - University of Natural Resources and Life Sciences, Institute for Biotechnology in Plant Production and [2]Christian Doppler Laboratory for Mycotoxin Metabolism, IFA-Tulln, 3430 Tulln, Austria; [3]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108 U.S.A.; [4]Department of Applied Genetics and Cell Biology, University of Natural Resources and Life Sciences, 3430 Tulln, Austria; [5]Chair of Bioinformatics, Department of Biotechnology, University of Natural Resources and Life Sciences, 1190 Vienna, Austria and School of Life Sciences, University of Warwick, Coventry CV4 7AL, U.K.
[*]Corresponding Author: PH: 43-2272-66280-205, Email: wolfgang.schweiger@boku.ac.at

ABSTRACT

Fusarium head blight caused by *Fusarium graminearum* is a devastating disease of wheat. We have developed near-isogenic lines (NILs) differing in the two major *F. graminearum* resistance quantitative trait loci (QTL), *Qfhs.ndsu-3BS* (also known *Fhb1*) and *Qfhs.ifa-5A*, which are located on the short arm of chromosome 3B and on 5A, respectively. These NILs show different levels of resistance and were used to identify transcripts that are significantly changed in a QTL-specific manner in reaction to the pathogen and between mock-inoculated samples. After inoculation with *F. graminearum* spores, 16 and 352 transcripts showed a significantly different response for the *Fhb1* and *Qfhs.ifa-5A* NIL pairs, respectively. Notably, we identified a lipid-transfer protein, corresponding to Ta.1282.4.S1_at that is 50-fold more abundant in plants carrying the *Qfhs.ifa-5A* resistant allele. In addition to the *Qfhs.ifa-5A*-associated candidate gene, we identified a UDP-glycosyltransferase, designated TaUGT12887, exhibiting a difference in response to the pathogen in lines harboring both QTL compared to lines carrying only the Qfhs.ifa-5A resistance allele, suggesting Fhb1 dependence of this transcript. Yet, this dependence was observed only in the NIL with higher basal resistance. The complete cDNA of TaUGT12887 was reconstituted from available wheat genomic sequences and expressed in a toxin sensitive strain of *Saccharomyces cerevisiae*. DON resistance, albeit weaker compared to the previously characterized barley HvUGT13248 was conferred. We will discuss possible interpretations of this result.

TARGETING DEFENSE REGULATORY GENES AND HOST SUSCEPTIBILITY FACTORS FOR ENHANCING FUSARIUM HEAD BLIGHT RESISTANCE IN WHEAT

Jyoti Shah[1*], Vamsi Nalam[1], Guy Klossner[1], Syeda T. Alam[1], Sujon Sarowar[1], Hyeonju Lee[2], Dehlia McAfee[2] and Harold Trick[2]

[1]Department of Biological Sciences, University of North Texas, Denton, TX 76203; and
[2]Department of Plant Pathology, Kansas State University, Manhattan, KS 66506
[*]Corresponding Author: PH: (940) 565-3535, Email: shah@unt.edu

ABSTRACT

Fusarium graminearum is one of the major causative agents of Fusarium head blight (FHB), a destructive disease of small grains. Genetic studies in *Arabidopsis thaliana* have provided insights into plant defense mechanisms that control severity of disease caused by *F. graminearum* (Makandar et al., 2006, 2010). Comparable mechanisms are also involved in controlling FHB severity in wheat (Makandar et al. 2006, 2012). In addition, host mechanisms that predispose plant tissue to fungal infection have also been identified. Several *Arabidopsis* genes that regulate defenses targeting *F. graminearum* have been constitutively expressed in wheat to enhance host plant resistance against FHB. Many of these have shown promise in greenhouse studies. In addition, wheat homologues of lipoxygenase genes that contribute to disease susceptibility have been targeted for silencing in wheat. Efforts are also underway to target genes involved in non-host resistance to promote FHB resistance in wheat.

ACKNOWLEDGEMENTS AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-8-060. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

REFERENCES

Makandar, R., Essig, J. S., Schapaugh, M. A., Trick, H. N. and Shah, J. (2006). Genetically engineered resistance to Fusarium head blight in wheat by expression of Arabidopsis *NPR1*. Mol. Plant-Microbe Interact. 19:123-129.

Makandar, R., Nalam, V., Chaturvedi, R., Jeannotte, R., Sparks, A.A., and Shah, J. (2010). Involvement of salicylate and jasmonate signaling pathways in *Arabidopsis* interaction with *Fusarium graminearum*. Mol. Plant-Microbe Interact. 23:861-870.

Makandar, R., Nalam, V., Lee, H., Trick, H.N., Dong, Y., and Shah, J. (2012). Salicylic acid regulates basal resistance to Fusarium head blight in wheat. Mol. Plant-Microbe Interact. 25, 431-439.

TRANSCRIPTOME SEQUENCING OF *FUSARIUM* CHALLENGED WHEAT NEAR ISOGENIC LINES: A COMPARISON OF METHODS

G. Siegwart, C. Ametz, M. Zamini, B. Steiner, W. Schweiger* and H. Buerstmayr

BOKU-University of Natural Resources and Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, A-3430 Tulln, Austria
*Corresponding Author: PH: 43-2272-66280-205, Email: wolfgang.schweiger@boku.ac.at

ABSTRACT

In an effort to identify differentially expressed transcripts in response to *Fusarium graminearum* in a quantitative trait loci (QTL) dependent manner, we performed transcriptome sequencing on NILs segregating for two prominent resistance QTL (*Qfhs.ndsu-3BS* and *Qfhs-ifa-5A*).

In order to identify the method best suitable for the entire set of samples, we compared two different sequencing approaches - full length RNA-seq and MACE (Massive Analysis of cDNA Ends), generating only one read/cDNA from the 3' end - on samples of the parental lines (susceptible cv. Remus and QTL-donor CM-82036). RNA from *F. graminearum* or mock inoculated floret tissue (50 hours after inoculation, hai) of both genotypes was provided to the sequencing facilities of 'GATC' (Konstanz, Germany) for RNA-seq and 'GenXPro' (Frankfurt/Main, Germany) for MACE. Both used one lane of an Illumina HighSeq2000 flow cell (yielding potentially 180 million reads). GATC used 8x multiplexing (22 M reads/sample) whereas GenXPro used 10x multiplexing, (18 M reads/ sample). Here higher multiplexing was used, because of the higher specificity of MACE, which should compensate for lower read numbers.

The generated number of reads for the respective methods differed significantly: For RNA-seq about 25 M reads/sample were produced, in contrast to 7 M reads/sample for MACE. After trimming, we were able to map 84% of the MACE reads and 72% of the RNA-seq reads to the publically available wheat flcDNA- (16k) and unigene-collections (65k). A pairwise comparison between *F. graminearum*/mock inoculated samples in both genotypes revealed about 19.500 and 9.500 differentially expressed genes between *F. graminearum* and mock-inoculated samples using RNA-Seq and MACE, respectively, with an overlap of 7.600 genes. Taken together, in our experiment the high amount of reads generated by RNA-seq outweighs the higher specificity and relatively higher number of mappable reads provided by MACE when given a limited budget. RNA-seq identifies most of the differentially expressed transcripts in response to *F. graminearum* and additionally, the high median number of reads per target allows for detection of polymorphisms and reconstruction of gene models. Consequently, the remaining samples were sequenced using conventional RNA-seq. Quality assessments of the sequenced samples attest good quality and confirms the choice of RNA-Seq over MACE.

Data analysis including test for differential expression in response to *F. graminearum* between NILs differing in either or both QTL is currently ongoing and will be presented as a poster.

STRUCTURAL AND FUNCTIONAL ANALYSIS OF A STRESS RESPONSIVE GENE FROM BARLEY

Kashmir Singh, Ravneet Kaur and Jaswinder Singh*

Plant Science Department, McGill University, Ste Anne De Bellevue, QC, H9X3V9, Canada
*Corresponding Author: PH: (514) 398-7906, Email: Jaswinder.Singh@mcgill.ca

ABSTRACT

Biotic and abiotic stresses pose major threats to crop species, causing heavy yield losses worldwide. Barley being an excellent model grain crop used as food for humans and animal feed stock; it is beneficial to study stress tolerance mechanism in this plant. Insertional mutagenesis has been widely employed to characterize stress-responsive genes in plants such as *Arabidopsis* and rice. We are currently employing this tool in barley to identify important traits that can be used to enhance plant's performance. We have identified a Wall–associated receptor-like kinase 1 (WAK1) by analyzing flanking sequence of *Ds* transposon insertion site in barley cv Golden Promise. Wall–associated receptor-like kinases (WAKs) are candidates for directly linking the extracellular matrix with intracellular compartments and are involved in developmental processes and studies have shown that this gene is involved in defense response against pathogen attack. For example, Induction of *WAK1* expression by salicylic acid (SA) is required by arabidopsis plants to survive infection by the bacterial pathogen, *Pseudomonas syringae*. SA is a signaling molecule that accumulates in plants in response to pathogen attack and is required for the establishment of systemic acquired resistance (SAR). Rice *WAK1* is induced by infection of an incompatible race P131 of *Magnaporthe oryza* which provides evidence that WAK take part in plant fungal disease resistance. For characterization of *WAK1* gene from barley, three BAC clones corresponding to WAK fragment were sequenced and full length *WAK1* gene was identified. The gene has 3 exons and two short introns with a coding region of 2178 bp encoding a protein of 725 amino acids. Regulatory region was also identified and analyzed in -1000 bp sequence upstream to start codon. Using CDD and SMART, various conserved domains such as GUB WAK Bind, EGF_CA and PKc including other regions like signal peptides, active sites and transmembrane domains were identified. The gene organization of *WAK1* was compared with that of wheat and arabidopsis, which was found to be similar, thus, suggesting that *WAK1* gene remained unchanged during the evolution. Nonetheless, WAK1 shared very low similarity with protein sequences available from barley cultivar Haruna Nijo, rice, wheat, *Arabidopsis*, maize and was found to be 50%, 46%, 21%, 25% and 19% respectively. This divergence may have helped the plants to adapt themselves according the surrounding environment, as WAKs are main proteins helping in exchange between the cytoplasm and outer environment through the cell wall. Semi-quantitative RT-PCR based expression analysis indicates its expression is specific to roots.

IDENTIFICATION OF CANDIDATE GENES OF MAJOR FHB-RESISTANT QTL IN WHEAT CULTIVAR SUMAI 3

Yongbin Zhuang, Aravind Gala and Yang Yen*

Department of Biology and Microbiology, South Dakota State University, Brookings, SD 57007
*Corresponding Author: PH: (605) 688-4567, Email: yen.yang@sdstate.edu

ABSTRACT

Major FHB resistant QTL have been identified in Sumai 3. To identify the genic components of these QTL, 406 FHB-related wheat ESTs revealed by microarray assay were investigated in two sets of material: two bulked resistant/susceptible pools constructed from 10 $F_{2:8}$Sumai3 / Y1193 RILs per pool and two near-isogenic lines (NILs) differed only in the *Fhb1* containing region on chromosome arm 3BS. The plants grown in greenhouse were investigated for FHB severity by calculating the rate of FHB-diseased kernels and for expression of the FHB-related genes by qPCR assay. Genes showed significant differential expression (≥2 fold) either between the two bulks or the two NILs in both years were subjected to eQTL mapping for their association with FHB-resistance QTL. The identified candidate genes were then physically mapped to their carrier chromosomes with the Chinese Spring nulli-tetra deficiency set. One gene, designated *WFhb1_c1* (Wheat*Fhb1* candidate gene 1), was both functionally associated with and physically located within *Fhb1*, and was found to be weakly similar (*E* = 5e+0) to a gene encoding pectin methyl esterase inhibitor. Two other genes, designated as *WFI_6BL1* and *WFI_6BL2* (Wheat-*Fusarium* interaction gene 6BL1/6BL2), were functionally associated with *Fhb_6BL*, but physically mapped on chromosomes 7D and 5A, respectively. *WFI_6BL1* was annotated as a 13-lipoxygenase gene and *WFI_6BL2* might encode a PR-4 like protein. Study of the dynamic expression of the three genes in the early stage of FHB pathogenesis suggested that: 1) *Fhb1* seems to contribute to FHB resistance by reducing susceptibility in the first 60 hours; 2) *Fhb_6BL* made its contribution to FHB resistance via the JA-mediated pathways; and 3) wheat seemed to activate its defense mechanism in the biotrophic phase of FHB pathogenesis.

OTHER PAPERS

OVERVIEW OF THE USWBSI WEB SITE

D. Hane[1*], S. Canty[2], D. Van Sanford[3] and O. Anderson[4]

[1]USDA-ARS-WRRC, 800 Buchanan Street, Albany, CA; [2]USWBSI-NFO, Michigan State University, East Lansing, MI; [3]University of Kentucky, Dept. of Plant and Soil Sciences, Lexington, KY; and [4]USDA-ARS-WRRC, 800 Buchanan Street, Albany, CA
[*]Corresponding Author: PH: (510) 559-6194, Email: davidhane@gmail.com

ABSTRACT

The US Wheat and Barley Scab Initiative (USWBSI) maintains a web site (http://www.scabusa.org) that offers information and services in support of the USWBSI's mission. The web site employs many technologies to facilitate rapid communication of information to the community. Users can browse the site anonymously as guests with limited access to applications and information. They may also register and have expanded access to information as well as the ability to contribute data using the site's various applications. Several technologies employed by the USWBSI site will also push information to users who have subscribed to the appropriate services. Some of these technologies include the FHB Alert system, various mailing lists, and RSS feeds. This poster provides an overview of the USWBSI web site and some of the features and applications.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement Nos. 59-0206-9-057 and 59-0206-1-082. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

GLEANINGS FROM THE 4TH INTERNATIONAL SYMPOSIUM ON FUSARIUM HEAD BLIGHT, NANJING 2012

Gene Milus*

Department of Plant Pathology, University of Arkansas, Fayetteville, AR 72701
*Corresponding Author: PH: 479-575-2676, Email: gmilus@uark.edu

ABSTRACT

Relatively few members of the USWBSI community participated in the Symposium. For those who could not participate, I am providing a somewhat biased view of what I found interesting and informative. Sumai-3 is still considered the most resistant cultivar, but combining a level of resistance similar to Sumai-3 with high yield, etc. has not been accomplished yet for winter wheat. Dozens of exotic resistant lines were reported, but no resistant cultivars have been developed from adapted x exotic crosses. Dozens of QTL for resistance were reported. Except for *Fhb-1*, these make small contributions to the overall resistance, and use of MAS has not led to the release of any resistant cultivar. The most successful strategy for developing resistant cultivars has been to make crosses among adapted moderately susceptible to susceptible cultivars, select first for high yield and then select for resistance in inoculated, misted nurseries. Additive effects and transgressive segregation for resistance were reported frequently, and these were supported by molecular results showing hundreds of genes being involved in resistance. There were several reports on the degree of anther extrusion among wheat lines affecting the level of type I resistance. Closed flowering or high anther extrusion were associated with low levels of initial infection. The Sha-3/Catbird line used as a parent in some of my resistant ARGE lines has high anther extrusion.

There were several reports on the *Fusarium* species and chemotypes causing head blight around the world. Most of these reports showed that the pathogen population has been changing over the past several years. Expanded maize production was the major factor that has been associated with the increase in *F. graminearum*. *F. asiaticum* and 3A-DON chemotypes were more common in warm areas, whereas *F. graminearum* and 15A-DON chemotypes were more common in cool areas. NIV chemotypes of both *F. asiaticum* and *F. graminearum* also appear to be on the increase. There was concern, but no definitive evidence, that the pathogen has been evolving toward greater aggressiveness. In China, the wheat area affected by severe head blight has shifted from the Yangtze River region northward in response to increased maize production, and there was a widespread, severe head blight epidemic in 2012.

Growing moderately resistant cultivars and applying a fungicide at flowering are universally recognized as the two most important management practices for reducing losses and mycotoxins. Application technology that increases the amount of fungicide applied to heads increases efficacy. The Chinese have been using carbendazim since the early 1970s. Resistance to this fungicide was first found in 1992 and is now common. The Jiangsu Pesticide Institute developed 2-cyano-3-amino-3-phenylancryic acetate that is now being used for FHB.

COMPARATIVE USE OF BOTANICAL OIL EXTRACTS IN PEST MANAGEMENT

Olotuah*, O.F.

Department of Plant Science and Biotechnology, Adekunle Ajasin University,
P.M.B.001, Akungba – Akoko, Ondo State, Nigeria
*Corresponding Author: E-mail: lanrose2002@yahoo.com

OBJECTIVES

1. To emphasize the adoption of use of plant extracts in pest control.

2. To consider the potential of use of oil extracts in the control of Fusarium head blight

INTRODUCTION

Cowpea, (*Vigna unguiculata*) (L). Walp is the one of the most ancient crops known to man. It is an annual legume and is commonly referred to as Southern pea, black eye pea, etc. The largest production is in Africa, with Nigeria and Niger predominating. The cowpea is predominantly a hot weather crop. It is more tolerant to drought, water logging, infertile soils, and acid stress than common bean. Cowpea can be grown successfully under conditions that are totally unsuitable for the common bean. They are much less tolerant to cold soils than common bean. Cowpea is considered nutritious with a protein content of about 24.8%, fat content of 1.9%, fibre content of 6.3%, carbohydrate content of 63.6% & water content of 8-9%.

The cowpea bruchid, *Callosobruchus maculatus* (F.) (Chrysomelidae: Bruchin), is a worldwide pest of stored cowpea grain (*Vigna unguiculata* (L.) Walp. Several methods had been adopted in its control. Some were effective while there were draw backs observed in some, especially in the use of synthetic chemicals.

Research in recent years has been turning more towards selective bio-rational pesticides, generally perceived to be safer than the synthetics (Anarson *et al*, 1989) and extensive works on the use of plant extracts in pest control have also been documented. Consequently, plant derived oils and powders have recently been evaluated and shown to be effective against a number of insect pests (Butler and Henneberry, 1990). This research work was based on the laboratory evaluation of use of three botanical oils in the control of insect pest of cowpea.

MATERIALS AND METHODS

This experiment was carried out at the Department of Plant Science and Biotechnology, Adekunle Ajasin University, Akungba-Akoko. The insect pest, C*allosobruchus maculatus* used was obtained from a culture of infected cowpea seeds maintained at ambient condition in the Department of Plant Science and Biotechnology Laboratory. Fresh plant leaves of *Hyptis suaveolens, Eucalyptus globulus* and *Cymbopogon citratus* was collected at maturity from different locations at Akungba Akoko,Ondo State, Nigeria during the rainy season. The leaves were detached from the stalk, washed and sun-dried for about 3 weeks before being pulverized using an electric blender.

The soxhlet apparatus (extractor) was used for the extraction of oils of *Eucalyptus* globulus, *Hyptis suaveolens* and *Cymbopogon citratus* and the different oils stored in McCartney bottle which was kept in a refrigerator at 5°C before use.

10ml of *Hyptis suaveolens* oil, *Eucalyptus globulus oil* and *Cymbopogon citratus* oil were prepared separately for the experiment while 40 seeds of Cowpea (*Vigna unguiculata*) were put in each Petri-dish, with three replicate and one

control. The control was a petri dish of cowpea seeds without treatment application. Twenty (20) pairs consisting of 20 males and 20 females of *C. maculatus* were put in each Petri-dish and the botanical oils were applied at each setup. This experiment was monitored for 3 weeks. At the end of the experiment, data collection was based on mortality rate at each concentration levels and data was subjected to analysis of variance and means compared using Tukey's Honestly Significant Test at 5% level of probability.

RESULTS AND DISCUSSION

Tables 1 to 3 show the cumulative mortality rate of insect pests with the application of the three botanicals for three weeks.

In tables 1to 3 it could be observed that the three botanicals are significantly different, $P > 0.05$, compared to one another, which makes *Hyptis suaveolens* the most effective. The least performance observed in *Eucalyptus globulus* was not an indication of weak potentials as it has equally been reported in some research works as being effective. Consequently, this variance in potentials may likely be dependent on the concentration levels adopted in the research. Observations made in this experiment corroborate the view of several researchers on the adoption and use of plant oils in pest control. Consequently, the high infestation of cowpea at almost every stage of its growth and concomitant damage necessitate a proactive and promising approach to its control. Although the use of synthetics had been adopted but since its use were faced with several challenges, the use of botanicals had been gaining attention in recent times. The use of essential oils had equally been adopted and proven effective. Raja *et al*., (2001) reported that pulse stored in gunny bags and treated with aqueous extracts from leaves of *Melia azadirachta, Hyptis suaveolens* and tuber of *Cyperus rotundus,* were effectively protected without any infestation for up to 6 months. Kim *et al,* 2003 reported the insecticidal activities of aromatic plant extracts and essential oils against *Sitophilus oryzae* and *Callosobruchus chineensis.* Also, Keita *et al*, (2001) reported that seeds treated with botanical extract oils did not lose their viability and they also established that powder made from essential oil of different basils provided complete protection against *C. maculatus* and did not show any significant effect on the seed germination rate. In a similar experiment on pest control focused on the adoptive use of extract, Tapondjou *et al*., (2002) showed that the dry ground leaf of *Chenopodium ambrosioides* inhibited F_1 progeny production and adult emergence of the *Callosobruchus chinensis* and *C. maculatus*, while Olotuah (2010) reported that the use of Cashew Nut shell liquid is effective in the control of Okra field insect pests, *Podagrica uniforma* and *P. sjostedti*.

Essential oils are used in perfumery, aromatherapy, cosmetics, incense, medicine, household cleaning products and for flavoring food and drink. They are responsible for the aroma and flavor associated with herbs, spices and perfumes. They are also called volatile oils because they easily diffuse into the air where they are then detectable by our olfactory senses. Essential oils are usually terpenoids another large class of secondary chemicals. Their presence in certain plant parts probably reflects their functions. It has also been reported that some organic pest control product such as Orange Guard use a citrus fruit peel base, such as from lemons and oranges. Citrus oils kill many flying and crawling insects on contact by destroying the waxy coating of the insect's respiratory system. Other organic pesticides use natural extracts to repel rather than kill pests. Some products use garlic or hot peppers and essential oils of herbs such as cloves to repel insects and other pest.

In addition to terpenoids, phenolic compounds are responsible for the aroma and flavour of some spices. For example eugenol is a phenolic compound found in both cinnamon (*Cinnamonum spp*) and cloves from *Syzygium aromaticum*. The functions of essential oils were once considered waste products. However, the biosynthetic pathways that yield essential oils are specialized and imply an expenditure of energy by the plant of their production. Another debated role for essential

oils is to inhibit competing plants or allelopathy. For example, essential oils in sheds leaves can leach into the soil where they may inhibits the germination or growth of other plants competing for the same resources (e.g. light, minerals nutrient, water). As with many other secondary compounds, they are now believed to also defer herbivory and prevent infections by pathogens i.e. bacteria and fungi. Nature had provided plant extracts that make very effective pesticides and insect repellants, which seems to be cheaper, safer and more easily produced than the synthetic insecticides.

On the basis, the use of botanical oil extracts should be seen as an effective protective measure in pest control and adoption of use extended to other crops of economic value such as the control of the Fusarium head blight.

REFERENCES

1. Arnason, J.T., B.J.R. Philogen and P. Morand. 1989. Insecticides of Plant Origin. ACS, Symposium Series No. 387, American Chemical Society, Washington, D.C. USA, pp: 213.
2. Butler, G.D., Jr and Henneberry, T.J. 1990. Pest Control on Vegetables and Cotton with Household Cooking Oils and Liquid Detergents. Southwest. *Entomol. 15, 123-131.*
3. Keita, S.M., C.Vincent, J.Schmit, P, Arnason and J. T. Belange,2001. Efficacy of Essential oil and *Ocimum basilicum L.* And *O. gratissimum* L. applied as an insecticidal fumigant and powder to control *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae). *Journal of Stored Products Research.37 (4):339-349.*
4. Kim, S.I., J.Y. Roh, D.H. Kim, H.S. Lee and Y.J Ahn, 2003. Insecticidal activities of Aromatic plant extracts and essential oils against *Sitophilus oryzae and Callosobruchus chineensis. Journal of Stored products Research 39(3): 293-303.*
5. Olotuah, O.F. and Ofuya, T.I. (2010)a: Comparative Protection of Cowpea, *Vigna unguiculata*, (L.) Walpers against Field Insect Pests using Cashew Nut Shell Liquid and Cypermethrin (Cymbush). *P. J. Nut.9 (2): 158-161.*
6. Raja, N., A. Babu, S. Dorn and S. Ignacimuthu, 2001. Potential of Plants for protecting Stored pulses from *Callosobruchus maculatus* (Coleoptera: Bruchidae) infestation. *ESN Occasional Publication,31: 227-235.*
7. Taponjou, L.A., C. Adler, H.Bouda and D.A Fontan,2002. Efficacy of Powder and Essential Oil from *Chenopodium ambrostodes* leaves as post-harvest grain Protestants against six-stored product beetles, *Journal of Stored Product Research. 38(4) 395-402.*

Table1. Cumulative mortality rate of insect pests determined at different times at the first week of the experiment.

	2 MINUTES	4 MINUTES	6 MINUTES
Hyptis suaveolens	40.0±0.0a (100%)	0±0.0c (0%)	0±0.0b (0%)
Cymbopogon citratus	37.5±0.5b (93.75%)	2.5±0.5b (6.25 %)	0±0.0b (0%)
Eucalyptus globulus	27.0±1.0c (67.5%)	7.5±1.5a (18.75%)	5.5±0.5a (13.75%)
Control	0±0.0d (0%)	0±0.0c (0%)	0±0.0b (0%)

Means in each column bearing the same letter are not significantly different at the 5 % level of probability using Tukey's Test

Table2. Cumulative mortality rate of insect pests determined at different times at the second week of the experiment.

	2 MINUTES	4 MINUTES	6 MINUTES
Hyptis suaveolens	40.0±0.0a (100%)	0±0.0c (0%)	0±0.0b (0%)
Cymbopogon citratus	35.5±0.5b (88.75%)	4.5±0.5b (11.25%)	0±0.0b (0%)
Eucalyptus globulus	26.0±0.5c (65%)	8.5±0.5a (21.25%)	5.5±0.5a (13.75%)
Control	0.0±0.0d (0%)	0.0±0.0c (0%)	0.0±0.0b (0%)

Means in each column bearing the same letter are not significantly different at the 5 % level of probability using Tukey's Test

Table3. Cumulative mortality rate of insect pests determined at different times at the third week of the experiment.

	2 MINUTES	4 MINUTES	6 MINUTES
Hyptis suaveolens	40.0±0.0a (100%)	0±0.0c (0%)	0±0.0b (0%)
Cymbopogon citratus	38.0±0.5b (95%)	2.0±0.0b (5%)	0±0.0b (0%)
Eucalyptus globulus	28.0±0.5c (70%)	8.0±0.5a (20%)	4.0±0.5a (10%)
Control	0±0.0d (0%)	0±0.0c (0%)	0±0.0b (0%)

Means in each column bearing the same letter are not significantly different at the 5 % level of probability using Tukey's Test

INDEX OF AUTHORS

NOTES

NOTES

NOTES

NOTES

NOTES

NOTES

U.S. Wheat & Barley

Scab Initiative