

Proceedings of the 2014 National Fusarium Head Blight Forum



December 7-9, 2014
Hyatt Regency St. Louis at the Arch
St. Louis, Missouri

**Proceedings of the
2014 National Fusarium
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Proceedings compiled and edited by: S. Canty, A. Clark, N. Turcott and D. Van Sanford

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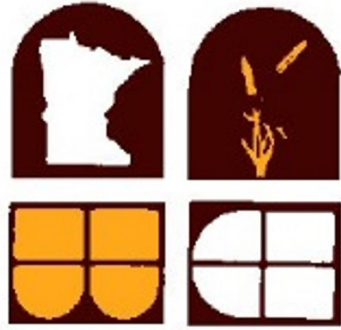
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FHB MANAGEMENT

TRIAZOLE SENSITIVITY IN POPULATIONS OF *FUSARIUM GRAMINEARUM*: PRELIMINARY FINDINGS, NEEDED RESEARCH, AND IMPLICATIONS FOR MANAGEMENT

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ABSTRACT

As part of a wider survey effort to assess genetic and phenotypic diversity among contemporary isolates of *Fusarium graminearum* in New York, we screened 50 isolates for sensitivity to two triazole fungicides, tebuconazole and metconazole. Our objective was to establish a baseline of sensitivity against which future and more extensive surveys could be referenced. One of the 50 isolates was found to be highly resistant to tebuconazole based on a laboratory determination of EC₅₀ (effective concentration leading to a 50% reduction of mycelial growth) at 8.09 mg/l. This was not just a laboratory phenomenon; suppression of FHB and DON was significantly reduced when a commercial rate of tebuconazole was applied to wheat plants inoculated with the resistant isolate as compared to plants inoculated with a sensitive isolate. Following treatment with tebuconazole, more individuals of the resistant isolate were recovered from wheat plants inoculated with an equal mixture of the resistant and sensitive isolate; in the absence of tebuconazole application, equal numbers of the resistant and sensitive isolates were recovered from co-inoculated plants. The tebuconazole-resistant isolate was an outlier among the 50 isolates though a wide range of sensitivity, EC₅₀ of 0.28 to 2.5 mg tebuconazole per l, was found among the other 49 isolates. None of the 50 isolates was resistant to metconazole and the range of EC₅₀ was narrower, from 0.05 to 0.86 mg/l. Putting these findings into some perspective, there has been no documented failure of control of *Fusarium* head blight with tebuconazole or any other triazole fungicides in North America; a partial reduction in control due to fungicide resistance build-up would be very hard to discern. It is not uncommon to find low frequencies of fungicide resistance in native fungal populations even before exposure to a particular fungicide. Natural variation in fungicide resistance should be expected in this fungus that is well known for its high degree of genetic variability. We suggest that more isolates with resistance at various levels will be found as larger surveys are conducted. We will share our perspectives on needed future research and on what implications that triazole resistance may have on the management of *Fusarium* head blight.

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EFFECT OF GLYPHOSATE ON FUSARIUM HEAD BLIGHT IN WHEAT AND BARLEY UNDER DIFFERENT SOIL TILLAGES IN EASTERN CANADA

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ABSTRACT

Fusarium head blight (FHB) is an important disease of wheat and barley, particularly in the wet conditions of eastern Canada. The principal pathogen associated with FHB, *Fusarium graminearum*, produces deoxynivalenol (DON), a mycotoxin that makes the grain unfit for food or feed. Surveys conducted in eastern Saskatchewan in 2005 and 2007 revealed that glyphosate application in the previous 18 months within minimum-till system was significantly associated with higher FHB levels in wheat and barley. This study aimed to determine the effect of glyphosate on FHB development in wheat and barley and on *F. graminearum* inoculum production under different soil tillages in eastern Canada. The experiment was performed during two years (2007-2008) at two different sites in Quebec, Canada (Saint-Augustin-de-Desmaures and Saint-Mathieu-de-Beloeil). Six trials were set in both sites, combining two cereal species, wheat and barley, and three soil tillages: moldboard plough, spring tillage (minimum-till) and direct drilling. For each trial, glyphosate or other herbicides chosen according to weed species were applied as main plot treatments on Roundup Ready™ soybean the year preceding cereal crops. The next year, three wheat and three barley cultivars with a distinct FHB resistance level were sown in the main herbicide plots, constituting the subplots. In each main plot, two Petri plates containing a *Fusarium*-selective medium were placed facing the ground in order to capture spores coming from the soybean residues. FHB index, *Fusarium*-damaged kernels (FDK), deoxynivalenol (DON) content and *F. graminearum* inoculum production were measured. Glyphosate had no significant effect on FHB index, FDK or DON content, whatever the trial and the site. *F. graminearum* inoculum production was enhanced by glyphosate in only one trial out of twelve. The relationship between *F. graminearum* inoculum from soybean residues and DON content was weak. Therefore, it seems that glyphosate used on soybean the year preceding wheat or barley crop has no or low impact on FHB development and *F. graminearum* inoculum production under Quebec cropping conditions, whatever the tillage practices used.

ACCUMULATION OF *FUSARIUM GRAMINEARUM* MYCOTOXINS
IN WHEAT STRAW AT VARIOUS INTERVALS AFTER
ANTHESIS FOR WHEAT CULTIVARS RANGING IN
SUSCEPTIBILITY TO FUSARIUM HEAD BIGHT

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ABSTRACT

Mycotoxins are known to be present in grains from plants affected by Fusarium head blight (FHB), but little is known about their presence in wheat straw. Wheat straw is commonly used as bedding material for livestock. Non-ruminants, such as swine, are especially sensitive to mycotoxins and may eat up to 4 kg of wheat straw bedding per day. When straw from fields affected by FHB is used as bedding, livestock are at risk of exposure to FHB-associated mycotoxins.

A field trial was conducted in Urbana, Illinois in 2013 and 2014 to test for the accumulation of mycotoxins in different parts of straw tissue. The trial was mist-irrigated, and *Fusarium graminearum*-infested corn kernels were spread throughout the trial to serve as an inoculum source. Twelve soft red winter wheat cultivars ranging in susceptibility to FHB were planted. Whole plants were sampled from each plot in 15 cm linear row sections at four different times during the growing season: 7 days after anthesis (daa), 14 daa, 21 daa, and 28 daa. These plants were split equally into lower and upper sections of the plant with all head and root tissue removed. The samples were then dried under forced air and ground into smaller particles. After harvest, stubble (consisting of only stem tissue) also was collected, dried, and ground. All grain and straw samples were sent to the University of Minnesota for mycotoxin analysis.

Five mycotoxins were tested for in this study, which were deoxynivalenol (DON), 3-acetyl-deoxynivalenol (3ADON), 15-acetyl-deoxynivalenol (15ADON), nivalenol (NIV), and zearalenone (ZEA). Due to low levels of NIV and ZEA, only DON, 3ADON, and 15ADON are reported.

DON concentrations in the top portion of the stems ranged from 0 to 3.8 ppm at 14 daa, 0 to 28.2 ppm at 21 daa, and 0 to 39.6 ppm at 28 daa. DON concentrations in the lower portion of the stem ranged from 0 to 1.3 ppm at 14 daa, 0 to 4.5 ppm at 21 daa, and 0 to 21.8 ppm at 28 daa. Post-harvest DON concentrations in the straw tissue ranged from 0.7 to 31.9 ppm, and concentrations of DON in harvested grain ranged from 0.2 to 14.2 ppm.

3ADON concentrations in the top portion of the stems ranged from 0 to 0.4 ppm at 14 daa, 0 to 0.7 ppm at 21 daa, and 0 to 1.2 ppm at 28 daa. 3ADON concentrations in the lower portion of the stem ranged from 0 ppm at 14 daa, 0 to 0.2 ppm at 21 daa, and 0 to 0.8 ppm at 28 daa. Post-harvest 3ADON concentrations in the straw tissue ranged from 0 to 3.8 ppm, and concentrations of 3ADON in harvested grain ranged from 0 to 0.1 ppm.

15ADON concentrations in the top portion of the stems ranged from 0 to 0.9 ppm at 14 daa, 0 to 2.1 ppm at 21 daa, and 0 to 8.0 ppm at 28 daa. 15ADON concentrations in the lower portion of the stem

ranged from 0 to 0.4 ppm at 14 daa, 0 to 0.8 ppm at 21 daa, and 0 to 1.7 ppm at 28 daa. Post-harvest 15ADON concentrations in the straw tissue ranged from 0 to 16.3 ppm, and concentrations of 15ADON in harvested grain ranged from 0 to 0.3 ppm.

At the 14 and 21 daa sampling timings, differences in DON concentration between upper and lower stem tissue did not differ within each cultivar. At 28 daa, significantly ($P \leq 0.05$) greater levels of DON were observed in the upper stem tissue compared to the lower stem tissue for ‘Pioneer 25R47’, ‘Kaskaska’, and ‘Sisson’, but no differences in DON concentration between upper and lower stem tissue were observed for any other cultivar.

Spearman’s correlation analysis was conducted to determine relationships between mycotoxin concentrations in grain and stems collected post-harvest. This analysis revealed that positive, significant correlations were present for DON in grain and stems ($P = 0.0001$; $R = 0.80$), for 3ADON in grain and stems ($P = 0.0007$; $R = 0.34$), and for 15ADON in grain and stems ($P = 0.0001$; $R = 0.70$). These results indicate that cultivars with resistance to mycotoxin accumulation in the grain may also have a low risk of mycotoxin accumulating in the straw tissue.

ACKNOWLEDGEMENT AND DISCLAIMER

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PRELIMINARY ANALYSIS OF NATIONAL SURVEY OF WHEAT & BARLEY PRODUCERS ON SCAB MANAGEMENT

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ABSTRACT

In late 2012, the USWBSI commissioned the National Agricultural Statistics Service (NASS) of the USDA to survey wheat and barley producers in 17 states that have experienced epidemics of Fusarium head blight in small grains. The purpose of the survey was to gather information from grain producers that would help us better assist them in managing the damaging disease.

A four-page questionnaire was mailed to more than 16,000 producers in March 2014. The survey asked questions about which practices are used to manage scab. Such practices include planting moderately resistant varieties, staggering planting dates, using scab risk forecasts, and applying fungicides. The survey probed both the degree of adoption of management techniques, and also barriers to adoption. It also asked how producers obtain information about scab management.

Survey responses were collected both in writing and over the phone. State-by-state response rates ranged from 43% to 68%. Within each state, counties with similar production practices were grouped together in one to nine districts. Responses are being analyzed at the district, state, and national levels.

EVALUATION OF INTEGRATED METHODS FOR MANAGING FHB AND DON IN WINTER WHEAT IN NEW YORK IN 2014

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OBJECTIVE

To evaluate the individual and interactive effects of moderately resistant cultivars and application timings of the fungicide Prostaro® on wheat yield and the integrated management of Fusarium head blight (FHB) and deoxynivalenol (DON) 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 2014, we observed the disease and yield impact of cultivar susceptibility, inoculation with *Fusarium graminearum*, and treatment with Prostaro fungicide at two timings.

MATERIALS AND METHODS

The trial was conducted at the Musgrave Research Farm in Aurora, NY in a Lima silt loam soil planted with four soft red winter wheat varieties, 'Pioneer Brand 25R40' (susceptible to Fusarium head blight (FHB)), 'Emmit' (moderately susceptible to FHB), 'Otsego' (moderately susceptible to FHB), and 'Pioneer Brand 25R46' (moderately resistant to FHB), following soybean harvest on 25 Sep 2013. The experiment was set up as a completely randomized block design with a split-plot arrangement, with cultivar as the main plot and the treatments as subplots, randomized in six replicated blocks. Main plots were sown with wheat at 118.8 lb/A with a 10 ft wide commercial grain drill. Subplots were 20 x 10 ft including 15 rows with 7-in. row spacing. The plots were

fertilized at planting (200 lb/A of 10-20-20) and topdressed on 10 Apr (170 lb/A of a 50/50 mix of ammonium sulfate and urea, providing ca. 57 lb/A of nitrogen) and again on 21 Apr (30 lb/A of urea, providing an additional 13.8 lb/A of nitrogen). The first Prostaro application was at anthesis (Feekes growth stage, FGS 10.51) on 2 Jun including the surfactant Induce at 0.125% V/V, and inoculated with a conidial suspension of *F. graminearum* (40,000 conidia/ml) after the fungicide had dried to augment the development of FHB. The second Prostaro application occurred seven days after anthesis on 9 Jun including the surfactant Induce at 0.125% V/V, and inoculated with a conidial suspension of *F. graminearum* (40,000 conidia/ml) after the fungicide had dried. Fungicide and *F. graminearum* treatments were applied with a tractor-mounted sprayer with paired TJ-60 8003vs nozzles mounted at an angle (30° from horizontal) forward and backward, 20-in. apart, pressurized at 30 psi, and calibrated to deliver 20 gal/A. Incidence and severity (percent of symptomatic spikelets on symptomatic heads) of FHB in each plot were rated on 23 Jun and used to calculate FHB Index, where FHB index = (FHB severity * FHB incidence)/100. Foliar diseases were rated on 23 Jun as percent severity on flag leaves (average rating for whole plot). Grain was harvested from a 20 x 4 ft area in each subplot using an Almaco plot combine on 25 Jul. Grain moistures, plot yields, and test weights were recorded. Yields and test weights were adjusted to bu/A at 13.5% moisture. *Fusarium* damaged kernels (FDK) were evaluated post-harvest as a percentage of kernels visibly affected by FHB out of a 100 kernel subsample from each plot. Analysis of deoxynivalenol (DON) content in grain was conducted in the US Wheat and Barley Scab Initiative-supported mycotoxin

analysis laboratory at the University of Minnesota, St. Paul, MN. Treatment means were calculated, subjected to analysis of variance, and separated by Fisher's protected LSD test ($P = 0.05$).

RESULTS AND DISCUSSION

The incidence of FHB over all plots ranged from 0.7 to 16%. The impact of supplemental inoculation with *F. graminearum* was determined by comparing the non-inoculated and inoculum only treatment. Overall, inoculation resulted in significantly reduced yield and increased FHB and DON as compared with the non-inoculated plots. FHB and DON development in 2014 were attributed primarily to supplemental rather than background inoculum.

Significant cultivar responses to inoculation were observed for yield, FHB and DON for the susceptible variety Pioneer 25R40, but only for FHB and DON, for the moderately susceptible varieties Emmit and Otsego and for the moderately resistant variety Pioneer 25R46. These data support the current qualitative designations of varieties as susceptible (Pioneer 25R40), moderately susceptible (Emmit and Otsego), and moderately resistant (Pioneer 25R46).

Under moderately low disease pressure, significant differences were detected in yield among the varieties with both Pioneer varieties yielding highest and Otsego yielding lowest, regardless of treatment. Pioneer 25R40 had significantly higher FHB index and DON than all the other varieties, regardless of treatment, and was the only variety to have an overall DON level above the 2.0 ppm threshold observed by grain buyers. Despite its high yield potential, planting of the susceptible variety carries an increased risk of docked or rejected grain even under moderate disease pressure. Prosaro fungicide application at either FGS 10.51 or 7 days later reduced DON risk in the susceptible variety by more than 50%. With excellent choice of high yielding varieties in the moderately susceptible and moderately resistant categories, we counsel New

York growers to no longer plant susceptible soft red winter wheats.

Environmental conditions encountered in our plots in 2014 were more favorable for *Fusarium* infection at 7 days after FGS 10.51 than they were at 10.51 and, therefore, the spores applied later served as the more important inoculum for infection. When results of all the cultivars were combined, the overall impact of each of the two Prosaro application timings was to significantly decrease FHB incidence, index, DON, and foliar diseases, as compared with the inoculum only treatment. The Prosaro application at 7 days after the initiation of flowering resulted in the lowest FHB and DON, i.e., the fungicide applied later did the best job of suppressing FHB and DON resulting from fungal spores that arrived at the later timing. FHB and DON applied at 7 days after FGS 10.51 were significantly lower than for the Prosaro application at FGS 10.51, and did not differ from the non-inoculated, no-fungicide control treatment. But it is also worth noting that sufficient fungicide remained on spikes from the FGS 10.51 application to give significant suppression of FHB and DON resulting from fungal spores deposited on plants at 7 days after 10.51. It is unlikely that we would have seen any advantage of the late fungicide application over the earlier if spores had only been applied at the early timing. This underscores the necessity to apply supplemental inoculum corresponding to all timings that fungicides are applied in an unbiased experiment to assess comparative efficacy of fungicide timings.

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

Treatment	FHB Incidence (%)	FHB Index	FDK (%)	DON (ppm)	Yield (bu/A)
No-fungicide, non-inoculated control.....	1.9 c	0.2 c	1.5 b	0.33 c	110.9 a
No-fungicide, inoculated FGS 10.51, and again 7 days later.....	11.4 a	2.6 a	10.3 a	2.19 a	104.7 b
Prosaro SC (6.5 fl oz) at FGS 10.51, inoculated FGS 10.51, and again 7 days later.....	7.7 b	1.3 b	4.2 b	0.91 b	110.1 ab
Prosaro SC (6.5 fl oz) at 7 days after FGS 10.51, inoculated FGS 10.51, and again 7 days later.....	3.8 c	0.4 c	2.8 b	0.47 bc	110.1 ab
LSD ($P=0.05$).....	2.31	0.70	3.66	0.59	5.99

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

Cultivar	FHB Incidence (%)	FHB Index	FDK (%)	DON (ppm)	Yield (bu/A)
Pioneer 25R40.....	8.3 a	1.6 a	9.9 a	2.01 a	115.2 a
Otsego.....	8.2 a	1.7 a	4.8 b	0.79 b	97.4 c
Emmit.....	6.8 a	1.0 a	2.7 b	0.78 b	104.6 b
Pioneer 25R46.....	1.5 b	0.1 b	1.2 b	0.33 b	118.6 a
LSD ($P=0.05$).....	2.72	0.83	3.68	0.63	3.53

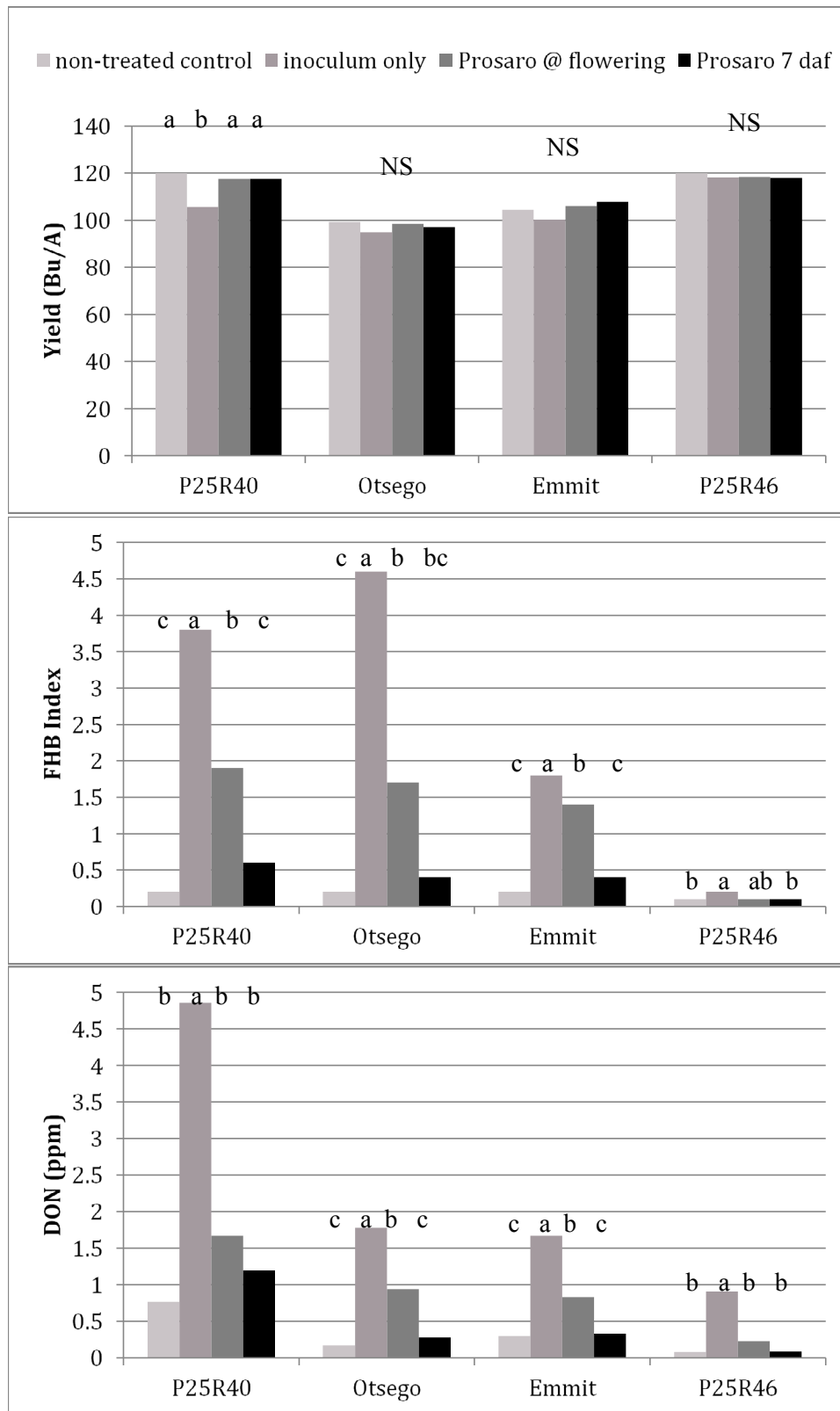


Figure 1. Effect of Prosaro fungicide application and *F. graminearum* inoculation on yield, FHB index and DON contamination of four winter wheat cultivars in Aurora, NY.

CORRELATION AND CAUSALITY ANALYSES OF
METEOROLOGICAL VARIABLES TO EXAMINE
PREDICTORS OF *FUSARIUM GRAMINEARUM*
ASCOSPORE RELEASE

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ABSTRACT

Fusarium graminearum is spread via macroconidia (asexual spores) and ascospores (sexual spores). Ascospores have been shown to travel long distances (>100 m) from sources of inoculum. We are investigating the meteorological variables and conditions associated with ascospore release to improve our understanding of temporal variation and spore emission rates. We aim to produce a useful model that includes source and at-risk wheat fields, spore emission rates, and built-in functionality capable of analyzing for favorable conditions for ascospore release. Our goal is to produce a model that provides stakeholders with an accurate representation of potential pathways of disease spread that may assist in making better informed management decisions, such as the application of fungicides.

Field trials were conducted in 2011 and 2012 at Virginia Tech's Kentland Farm, and statistical analyses were performed to examine potential relationships between spore release and a variety of meteorological variables. Our objective was to determine the correlation and causality relationships between environmental variables and spore release based on data obtained from a series of field experiments conducted over two growing seasons. In each of these growing seasons, a wheat field was artificially inoculated with *F. graminearum*, an active sampler was used to capture atmospheric spores, and meteorological conditions including air temperature, relative humidity, rainfall, and solar radiation were gathered from a nearby weather station. The spore concentration data obtained was assumed to be representative of release events within the inoculated field. Statistical analyses revealed significant relationships between ascospore release and time, solar radiation, wind speed, and relative humidity. Causality analyses were performed to determine if any of the environmental variables appeared to be causal agents for ascospore release events. The results indicated that solar radiation and relative humidity were the most important external driving factors in this system.

Based on the information gained from our correlation and causality analyses, we have designed a series of controlled laboratory experiments to assess ascospore release as a function of environmental variables. Our objective is to define the relationship between spore release and temperature, relative humidity, and light under controlled conditions. We have configured an embedded dual chamber using a growth chamber to maintain temperature and light and an acrylic chamber containing perithecia and a saturated salt solution to maintain relative humidity. We are testing combinations of temperature (15°C and 25°C), relative humidity (75%, 85%, and 95%), and light (light or complete darkness) and obtaining

temporal particle (ascospore) counts using an aerodynamic particle sizer. We aim to incorporate the knowledge gained about the environmental variables associated with spore release into a model to more accurately represent the atmospheric transport of ascospores from their source to their final destination. The results will further inform farmers and growers on the timing of potential ascospore release events, allowing them to make timely field management decisions.

EXAMINATION OF COMMERCIAL GRAIN SAMPLES TO ASCERTAIN HOW DEOXYNIVALENOL CONTAMINATION EXCEEDED ANTICIPATED LEVELS IN SOME 2014 WHEAT CROPS FROM WESTERN KENTUCKY

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ABSTRACT

Fusarium head blight (FHB or scab) re-emerged in the USA over two decades ago, first appearing in the Upper Midwest states of Minnesota and North Dakota in 1993. Since that time, FHB epidemics and associated mycotoxin contamination, especially by deoxynivalenol (DON), have been reported in all classes of wheat and in all production regions except the Pacific Northwest. The principal strategy to manage FHB has been the development of wheat varieties with moderate resistance and varieties with improved resistance to FHB are now deployed in all FHB-prone production regions in the USA. Effective fungicides are also available following a national effort to test the efficacy of fungicides to control FHB. In addition to the multi-state uniform fungicide testing program, effective technologies for the application of fungicides to the wheat head have been developed and deployed. Thus, growers do now have viable options for chemical control. A national forecasting system, The *Fusarium* Risk Assessment Tool, was developed, and has been deployed in many states for over a decade, to aid growers in deciding if conditions are favorable for FHB development and thus if fungicide applications are warranted. Best management practices recommend that growers combine the best available genetic resistance with a fungicide when conditions are favorable for FHB development. In some instances however, it seems that despite using best management practices, grain is still occasionally contaminated with *Fusarium*-mycotoxins. In 2014, a number of soft red winter wheat crops in western Kentucky were rejected because of DON contamination. This appeared to have occurred when the forecasting model had suggested that the risk of FHB development was moderate or low and/or where fungicides had been used and were anticipated to be effective. To better understand the level of mycotoxin contamination of crops in this region, samples from 21 commercial fields, representative of crops in this region, were collected and examined. These samples were evaluated for visual damage to the grain and subsequently tested for the presence of *Fusarium*-mycotoxins. The percent of visually scabby kernels (VSK), determined by visually matching the 21 grain samples to check samples, ranged from one to thirty percent. The percent of symptomatic kernels, in the samples by weight, ranged from three to thirty three percent. In addition to visual inspections of the grain, 100 seeds from each sample (50 symptomatic and 50 non-symptomatic seeds) were plated on a semi-selective growth media to assess the level of *F. graminearum* infestation. In one sample *F. graminearum* was isolated from 86% of the seeds identified as symptomatic and from 70% of the seeds that appeared to be sound. By contrast in another grain sample, *F. graminearum* was only isolated from 18% of the seeds identified as symptomatic and from 6% of the seeds that appeared sound. The deoxynivalenol content of the 21 samples ranged from 0.13 ppm to 16.4 ppm. Our examination of the grain samples confirm that *Fusarium* infection

was generally sufficient to cause some visual damage to grain and that contamination by *Fusarium* mycotoxins appeared closely correlated to visual symptoms. It appears that the crops examined had flowered in mid-May and at least some of the crops had a fungicide, generally Prosaro or Caramba, applied at flowering, with applications reported to have been applied between May 5 and May 20. In the earlier part of this period (May 5-11) the FHB risk assessment tool for this part of Kentucky indicated a low risk of FHB though medium risk was evident in parts of western Kentucky from May 12-20 and an area of high risk was evident within that medium risk area from May 13-18. The prediction of FHB in this region was likely hindered by uneven crop development that followed unusually cool conditions in winter and spring and we speculate that a lag time in the response of the model to the changing in environmental conditions may have resulted in the model underestimating risk. The winter wheat model is considered more prone to this type of inaccuracy than the spring wheat model used in the *Fusarium* Risk Assessment Tool. Challenging weather conditions may also have hindered the timely application of fungicides to some crops. Unfortunately there was insufficient information available to determine the risk predicted for the individual crops for which grain samples were provided. It would appear however that visual inspection of the grain for *Fusarium* damage at the buying point may have allowed for the segregation of the most heavily contaminated grain from other crops that were less damaged.

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THE EFFECTIVENESS OF AN INTEGRATED STRATEGY TO MANAGE FUSARIUM HEAD BLIGHT IN BARLEY PRODUCTION USING A META-ANALYSIS APPROACH

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ABSTRACT

An integrated management strategy combining host resistance, a timely fungicide application and crop rotation is recommended to help reduce the risk of Fusarium head blight (FHB) in barley production. As part of a research collaboration within the United States Wheat and Barley Scab Initiative, 19 barley field trials were conducted from 2008 to 2014 across North Dakota, Minnesota and South Dakota. The objective of these trials was to determine the efficacy of an integrated FHB management strategy in reducing scab index and deoxynivalenol (DON) levels. The experimental design used was a randomized complete block with either a split-split plot or split plot arrangement, with the treatment factors being previous crop residue, variety, and fungicide application. Three to eight two-row and/or six-row barley varieties, varying in FHB resistance, were included at each location. Fungicide applications using prothioconazole + tebuconazole (Prosaro 421C, Bayer CropScience, Research Triangle Park, NC) were made at 50% heading or 4 to 5 days after 50% heading, and a non-treated check was included at each trial. Previous crop residue was classified as either a host or non-host for *Fusarium graminearum*. As a way to summarize the findings, the data from the trials will be subjected to a multivariate meta-analysis. Data sets will be organized into six host resistance-fungicide combinations; susceptible treated, susceptible non-treated, moderately susceptible treated, moderately susceptible non-treated, moderately resistant treated, and moderately resistant non-treated. The results of the meta-analysis will be used to assess the stability and relative effectiveness of implementing an integrated management strategy for FHB in barley production.

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THE USE OF INTEGRATED MANAGEMENT STRATEGIES TO LOWER FHB DON IN BARLEY

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ABSTRACT

Fusarium Head Blight (FHB) has reduced the quality of barley grown in the Midwest for the last two decades due to discolored kernels, and more importantly the presence of the toxin, deoxynivalenol (DON). Six-rowed and two-rowed barley cultivars with different levels of DON resistance and timing of fungicide applications showed lower DON levels. The first year of these Integrated Pest Management studies were performed at two locations, Fargo and Hope, ND. The Fargo trial investigated the effects of two different application times of a single fungicide application in an artificially inoculated FHB nursery and the experiment at Hope, ND only had one fungicide treatment and was under natural infection. Disease incidence, severity and DON were evaluated along with test weight and yield at both locations. Both treatments at the Fargo location showed significantly lower FHB incidence, severity and DON accumulation when Prosaro® fungicide was applied at Feekes 10.4-10.5 (50%-75% head emergence) and five days later compared to the untreated checks. The study at Hope, ND also showed significantly lower DON accumulation when a single treatment of Prosaro was applied at the Feekes 10.5 stage (0.26 ppm) compared to the untreated control (0.47 ppm). There were significant differences among cultivars at both locations for test weight and yield compared to the untreated checks.

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This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No.59-0206-9-064. 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.

META-ANALYSIS OF 19 YEARS OF FUNGICIDE TRIALS FOR THE CONTROL OF FUSARIUM HEAD BLIGHT OF WHEAT

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ABSTRACT

Twenty years ago many were skeptical that Fusarium head blight (FHB) could be managed with fungicides (McMullen et al. 2012). Early work by Marcia McMullen and colleagues from 1995-1997 showed that a single application of a triazole (DMI) fungicide had the potential to reduce FHB index. This led to the establishment of the Uniform Fungicide Trials (UFT) by the USWBSI, with the first field studies being conducted in 1998. Initially, the studies focused on the use of propiconazole (Tilt) or tebuconazole (Folicur) for reducing index and DON. In the 2000s, other DMI-active ingredients such as prothioconazole, metconazole, and mixtures of actives such as tebuconazole + prothioconazole were added to the collection of fungicides being tested. Some of these were initially tested as experimental products before being registered and given trade names such as Proline®, Caramba® and Prosaro®, respectively. Other treatments were considered in a small number of trials, but were not of sufficient number for analytical purposes. Preliminary analyses were based on qualitative or ad hoc syntheses of the conducted trials. Paul, Madden, and colleagues then performed quantitative research syntheses of the trials using univariate and multivariate meta-analyses to estimate the expected treatment effects for FHB index, DON, yield, and test weight (Madden & Paul 2011, Paul et al. 2007, 2008, 2010). These meta-analyses were based on trials conducted up through 2005 for index and DON, and through 2007 for yield and test weight. Overall, Proline, Caramba, and Prosaro applied at anthesis performed much better than the other tested fungicides, and there were only minor differences in efficacy among these three.

Nevertheless, mean percent control (percent reduction relative to the untreated control) was typically only 50% for index and 40% for DON for the best treatments (averaged across environments and wheat market classes). Therefore, the UFTs have been continued to: determine the stability of efficacy and economics of these fungicides under a wide range of environments; explore alternative fungicide treatments that may result in higher percent control, especially for DON; and allow greater flexibility in terms of timing of applications. New treatments included: other mixtures of triazole fungicides applied at anthesis (typically as tank mixes); different timings of the best triazoles (before, at, or after anthesis); strobilurin fungicides (especially pyraclostrobin [Headline]) applied at different times; or combinations of Headline early and a triazole at anthesis.

The full data set analyzed consisted of 309 trials, from 1995 through 2013; 27 separate treatments were included as having been tested in a sufficient number of trials for the meta-analysis. Trials were conducted in up to 12 states per year. A multivariate meta-analysis showed large variability in percent control for the different treatments, and none of the new treatments provided significantly better control of index and DON than the original three best treatments, i.e., prothioconazole (Proline), metconazole (Caramba), and tebuconazole + prothioconazole (Prosaro) applied at anthesis. Percent control for these three treatments remained generally stable over time, although treatment efficacy for FHB index declined somewhat for spring wheat relative to winter wheat. For index and DON, a tank mix of tebuconazole

+ metconazole applied at anthesis, and metconazole applied 5 days after anthesis, resulted in percent control about the same as the original best treatments, the latter suggesting that there is some flexibility in applying the single DMI fungicide. A strobilurin application at a single time led to moderate or low percent control of index relative to the original best treatments; however, as hypothesized, a strobilurin application led to significantly *higher* DON in the grain relative to the untreated control. For instance, applying pyraclostrobin at heading produced an average 22% decrease in index and an 18% increase in DON relative to no treatment at all. Applying pyraclostrobin at boot (useful for controlling foliar diseases) and tebuconazole + prothioconazole at anthesis achieved a percent control of index similar to the triazole-only application at anthesis; however, the percent control of DON was considerably lower than for the triazole alone. That is, an anthesis application of a triazole could not counteract the negative effects of an earlier application of a strobilurin.

In conclusion, the best triazole fungicides applied at anthesis, or shortly thereafter, either alone or as a mixture, provide significant levels of control of index and DON. There is no evidence that substantially higher levels of control can be achieved with a single fungicide application without coupling this with other integrated control tactics. Additional analysis is needed to characterize the impact of all the fungicide treatments, especially for yield and test weight.

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SCREENING FOR FHB SUSCEPTIBILITY IN BARLEY AND WHEAT CULTIVARS IN THE WESTERN US

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ABSTRACT

Ten years ago, the incidence of FHB in the irrigated west was regarded as a minor and relatively rare occurrence. With the substantial increase in corn acreage directly due to the increase in the dairy industry, and with changes in irrigation practices, FHB has become a regularly occurring problem with economically significant impacts on small grain producers. The objective of this study was to determine host resistance levels in wheat and barley varieties released for the arid irrigated production areas of the PNW that have been selected without screening for FHB disease reaction. Varieties and advanced breeding lines from public and private breeding programs in the PNW and Intermountain West included in extension variety trials were tested for FHB susceptibility. Small plots were planted April 8, 2014 with two replicates per variety. Inoculum was developed from local isolates of *Fusarium graminearum*. The field nursery was sprayed twice (heading and flowering) with conidial suspension at 5×10^4 spores per ml (Chen et al., 2006). Plots were rated for disease incidence and severity three weeks after inoculation. Significant differences in spring wheat varieties for FHB index were recorded, but no significant differences in yield were found. The hard white spring wheat 'Klasic' was very susceptible, but varieties showing significantly lower levels of infection included the soft white spring wheat varieties Alpowa, Babe, UI Stone, UI Pettit, and advanced line WA 8162. However, there was very little infection that occurred in the barley trials.

OBJECTIVE

The objective of this study was to determine host resistance levels in wheat and barley varieties released for the arid irrigated production areas of the PNW.

INTRODUCTION

Ten years ago, the incidence of FHB in the irrigated west was regarded as a minor and relatively rare occurrence. With the substantial increase in corn acreage directly due to the increase in the dairy industry, and with substantial changes in irrigation practices, FHB has become a regularly occurring problem with economically significant impacts on small grain producers. Unacceptable levels of DON toxin have been found consistently in irrigated wheat and barley in areas of the PNW and intermountain West in the past five years. Corn debris, where high levels of *Fusarium graminearum* reside, takes up to three or four years to degrade in arid west environments. Changes in crop rotation have shifted the predominant species of *Fusarium* to *F. graminearum*, which produce airborne ascospores that can disperse many miles in the wind. Disease management approaches have changed depending on level of susceptibility of the varieties being grown. Control strategies must incorporate varieties that are less susceptible to FHB.

MATERIAL AND METHODS

Varieties and advanced breeding lines from public and private breeding programs in the PNW and Intermountain West were tested for degree of

susceptibility. An irrigated FHB disease nursery was established at the University of Idaho's Aberdeen Research and Extension Center. The spring wheat and spring barley nurseries were planted separately. Eight-foot plots consisting of two rows were planted April 8, 2014 in a randomized complete block with two replicates per variety. Inoculum was developed from local isolates of *Fusarium graminearum*. The field nursery was sprayed twice (at heading and flowering) based on heading and flowering dates with conidial suspension at 5×10^4 spores per ml (Chen et al., 2006). A CO₂ backpack sprayer with 8003 VS nozzle tips calibrated at 40 psi was used to apply inoculum at a rate of 1 sec/ft. Plots were irrigated two hours daily to maintain irrigation requirements and humid conditions. Thirty heads per plot were assessed for disease incidence and severity three weeks after inoculation and the percentage of FHB-colonized seeds and DON will be tested after harvest. A FHB index was calculated as (% severity x % incidence)/100. Plots were harvested 8 September with a small plot combine. Yield was determined with the HarvestMaster® system on the combine. Data were analyzed using GLIMMIX in SAS. Fisher's protected LSD was used for mean comparisons.

RESULTS AND DISCUSSION

Disease developed quicker in the durum wheat, then in the other spring wheat varieties. Disease did not develop in the barley trials. The spring was cooler than average with very cool nights, likely contributing to low disease pressure. Recommendations for 2015 will be to postpone

planting for 2 to 4 weeks, in order to increase the likelihood of warmer weather occurring during anthesis and inoculation to facilitate infection and disease development. There were significant differences in varieties for the FHB index ($\alpha = 0.01$), which varied from 1.6 in the advanced line WA 8162, the most resistant variety, to 57.2 in Klasic, a very susceptible hard white spring wheat (Table 1). The three lines showing the highest level of resistance included the soft white spring wheat UI Stone, UI Pettit and WA 8162. UI Stone was selected prior to release based on FHB resistance and carries two known molecular markers; *UMN10* associated with resistance gene *Fhb1* and *Xbarc117*, a QTL on chromosome 5AS. WA 8162 is an advanced soft white line from Washington State University's spring wheat breeding program.

There was a high degree of variability between reps for FHB index and yield. Yield varied from 61 bu/A to 143 bu/A, but there were no significant differences at $\alpha = 0.05$. FDK and DON levels will be measured and reported at a later date.

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Table 1. Yield and FHB index ratings of spring wheat varieties, 2014 trial, Aberdeen R&E Center, Aberdeen, ID.

Variety	Class ^z	Yield bu/A		FHB Index	
Bullseye	hrs	143.4	a	15.5	e-l
SY Basalt	hrs	118.7	ab	20.7	c-j
WB 6121	sws	116.5	abc	23.1	c-h
Alturas	sws	115.4	a-d	31.1	bc
LCS Star	hws	111.4	a-e	16.2	e-l
Dayn	hws	111.1	a-f	19.0	c-j
WB 6430	sws	108.9	a-g	16.0	e-l
Kelse	hrs	108.2	a-g	26.7	cde
UI Pettit	sws	106.7	b-g	3.6	m
Alpowa	sws	106.7	b-g	12.9	f-m
SY-40292	hrs	103.1	b-g	19.1	c-j
Penawawa	sws	101.6	b-g	24.9	c-g
WA 8162	sws	101.6	b-g	1.6	m
11SB0096	sws	100.9	b-g	10.3	i-m
IDO851	sws	100.2	b-g	14.8	e-l
Babe	sws	99.8	b-g	12.4	g-m
IDO852	sws	98.4	b-h	17.9	d-j
LL3419	hrs	94.7	b-i	39.9	b
WA 8166	hrs	94.7	b-i	15.6	e-l
LL 3361	hrs	94.0	b-i	14.9	e-l
Cabernet	hrs	94.0	b-i	28.9	b-e
BZ908-41	hrs	92.6	b-i	10.1	j-m
LL 3378	hrs	91.1	b-i	9.2	j-m
SY-10136	hrs	89.7	b-i	11.5	h-m
LCS Atomo	hws	89.3	b-i	26.5	cde
Alzada	durum	87.5	b-i	23.3	c-h
Snow Crest	hws	86.8	b-i	31.0	bc
IDO862E	hrs	86.0	b-i	22.1	c-j
WB 9229	hrs	84.2	b-i	23.3	c-h
IDO862T	hrs	83.1	b-i	23.0	c-j
WA 8189	sws	82.8	b-i	16.7	e-k
Utopia	durum	82.4	b-i	30.2	b-e
Jefferson	hrs	82.0	c-i	21.9	c-j
IDO1202S	hrs	80.6	c-i	9.0	j-m
WB 9668	hrs	80.6	c-i	23.0	c-i
UI Stone	sws	79.9	d-i	3.9	klm
Westbred 936	hrs	79.1	d-i	25.5	c-f
Klasic	hws	75.5	e-i	57.2	a
Buck Pronto	hrs	74.8	f-i	22.6	c-j
UI Winchester	hrs	73.7	ghi	19.0	c-j
UI Platinum	hws	63.2	hi	30.6	bcd
WB-Paloma	hws	61.3	i	22.6	c-j
	Num DF	41			
	F Value	1.54			
	Pr > F	0.0894			

^zClass definitions of wheat varieties: hrs = hard red spring, hws = hard white spring, sws = soft white spring.

RAINFASTNESS OF CARAMBA® AND PROTECTION AGAINST FUSARIUM HEAD BLIGHT AND DEOXYNIVALENOL IN SOFT RED WINTER WHEAT

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ABSTRACT

Fusarium head blight (FHB), caused by the fungal pathogen *Fusarium graminearum*, is a common and devastating disease of wheat and other small grain crops in the United States and other parts of the world. FHB damages and contaminates grain with harmful mycotoxins such as deoxynivalenol (DON). Fungicide application is critical to controlling FHB, conferring 48-52% reduction in disease and 42-45% reduction in DON in the absence of cultivar resistance. Research has shown that the efficacy of fungicides may be reduced if rainfall occurs during or shortly after application. The goal of this study was to quantify the protection that the commonly-recommended Caramba® fungicide confers if it rains within the first few hours after application. Seven-row plots (5 x 10 ft) of FHB susceptible soft red winter wheat cultivar Pioneer 25R45 were planted at a seeding rate of 1.6×10^6 seeds/acre and a row spacing of 7.5 in. The experimental design was a randomized complete block, with seven experimental units (plots) per block. Caramba was applied at anthesis at a rate of 14 fl oz/A to all but one plot in each block. Simulated rain was applied at 79 to 112 mm h⁻¹ (for a total volume of 4.1 mm across the plot) to five randomly selected plots in each block. The rainfall applications were made at 0, 60, 105, 150, or 195 min after Caramba application. The untreated plot (Check 1) and one fungicide-treated plot (Check 2) within each block were not subjected to simulated rain. All plots were spray-inoculated with a spore suspension of *F. graminearum* approximately 36 hours after fungicide application, and FHB index (IND) and incidence (INC) as well as *Fusarium* damaged kernels (FDK) and DON were quantified for each plot. Mean IND, INC, FDK, and DON in Check 1 were 16%, 41%, 23%, and 10 ppm, compared to 8%, 25%, 14%, 6 ppm in Check 2. All fungicide-treated plots had significantly lower mean INC than Check 1, regardless of whether or when rainfall treatments were applied. Plots subjected to rainfall at least 105 minutes after Caramba application had significantly lower mean IND and FDK than Check 1 (untreated). Mean IND, INC, DON, and FDK were not significantly different among plots exposed to rain between 105 and 195 min after Caramba application and Check 2 (fungicide treated, without rain). Mean IND and FDK in plots that received rain at 0 and 60 min after Caramba application were not significantly different from Check 1. Percent control of IND and DON relative to Check 1 ranged from 45 to 50% for plots exposed to rain at least 105 min after Caramba application, 25 to 40% for those that received rain at 0 or 60 min, and 46 to 52% for treated plots that were not subjected to rain (Check 2). Determining a minimum length of time between application of Caramba and subsequent rainfall that results in significant disease and toxin control (the rainfast time) can advise application recommendations that account for weather. The results of this study suggest that growers should apply Caramba at least 105 minutes before it rains.

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INFLUENCE OF PRE-ANTHESIS RAINFALL PATTERNS AND INOCULUM SOURCES ON FUSARIUM HEAD BLIGHT AND DEOXYNIVALENOL IN WHEAT

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OBJECTIVES

To investigate the effects of intermittent moisture during the 8-day pre-anthesis window and of inoculum sources on Fusarium head blight (FHB) and deoxynivalenol (DON) in wheat.

effect of frequency and timing of rainfall within the 7- to 10-day immediately prior to anthesis is still poorly understood. Thus, research is needed to better characterize how the number of days and distribution of pre-anthesis moisture affect FHB development and DON contamination.

INTRODUCTION

The effect of variable pre-anthesis high-moisture or rainfall patterns on FHB and DON constitutes a major knowledge gap in the epidemiology of FHB. This has led to uncertainty in the assessment of the risk of this disease and toxin. Producers and researchers alike have questioned the “low-risk” prediction of the FHB risk tool in some seasons when pre-anthesis rainfall is intermittent (spotty). Empirical observations and results from controlled-environment studies show that infection cycle events critical for FHB development may occur under (or even require) conditions of intermittent moisture. For instance, ascospore release was associated with cyclic wet and dry periods under both laboratory and field conditions (4,7). Rossi et al. (6) found that the number of *F. graminearum* macroconidia sampled in wheat fields often spiked during the day following a rain event, rather than on the day of the rain event.

FHB development and DON accumulation are strongly influenced by environmental conditions before and during anthesis and early grain-fill. It is well known that pre-anthesis temperature and moisture are critical for both processes. However, very few studies have investigated the effects of moisture or rainfall patterns during the pre-anthesis window on FHB and DON (1,2). In particular, the

MATERIALS AND METHODS

Field plots were planted on 10 October 2013 at the Ohio Agricultural Research and Development Center in Wooster, OH. Plots of Hopewell, an awnless, susceptible soft red winter wheat cultivar, were planted into a field previously cultivated with oats, and managed according to standard agronomic practices for Ohio. Each experimental unit (plot) consisted of seven 6-m-length rows, spaced 19 cm apart, and planted at a seeding rate of 4×10^6 seeds ha^{-1} . The experimental design was a randomized complete block, with a split-plot arrangement of pre-anthesis simulated rainfall patterns (five levels) as whole-plot and inoculum sources (three levels) as sub-plot. There were 2 replicate blocks. Beginning 8 days prior to anthesis and ending at 50% anthesis, the rainfall patterns (treatments) were: 1) rainfall every day; 2) rainfall on the first and last two days, separated by a four-day period without rainfall; 3) no rainfall on the first and last two days, separated by four days with rainfall; 4) rainfall every other day, and 5) check (no supplemental rainfall/irrigation; ambient rainfall). Irrigation risers were mounted in each whole plot, with separate timers programmed to run 4 minutes every 12 minutes between 5:00 and 9:00 h and 17:00 and 22:00 h on the scheduled day of rainfall. On average, 16 mm of “rain” was delivered by the irrigation system on each designated “rainy day”.

The inoculum sources were corn spawn (colonized corn kernels) and naturally-infected corn residue, spread between the rows at jointing (Feekes GS 6), and without in-field inoculum (check).

Twenty spikes were harvested daily from each plot and assayed for spores of *F. graminearum* as previously described (5). FHB index (field- or plot-level disease severity, defined as mean proportion of diseased spikelets per spike) was evaluated at soft dough (Feekes GS 11.2) on 20 spikes at 5 arbitrarily selected locations within each sub-plot. A sample of grain from each plot was used to estimate percent *Fusarium* damaged kernels (FDK, the percentage of small, shriveled, whitish-pink kernels) with the aid of a diagrammatic rating scale (3), and then sent to the U.S. Wheat and Barley Scab Initiative-funded laboratory at the University of Minnesota for DON quantification.

RESULTS AND DISCUSSION

We observed that mean FHB index, FDK, and DON were numerically higher for plots that received simulated rain compared to the check, and for plots that received rain, the means were higher for those with corn spawn than those with naturally-infected corn residue or without in-field inoculum (Fig. 1). Pre-anthesis rainfall patterns tended to influence FHB development and DON contamination in plots with corn spawn, but appeared to have little discernable effect on disease and toxin in plots with corn residue or without inoculum. This was likely because plots with corn spawn had more spores on the infection court (Fig. 2A and B). In plots with corn spawn, the every-day rainfall treatment (Rain_1) resulted in numerically higher mean FHB index than treatments with intermittent rainfall (Rain_2, Rain_3 and Rain_4) (Fig. 1). Interestingly, however, although mean FHB index was highest for Rain_1, plots that received rainfall on the first and last two days (Rain_2) or every other day (Rain_4) during the 8-day pre-anthesis window had higher or comparable mean FDK and DON. Rain_1 and Rain_2 also resulted in more spores reaching the spikes than Rain_3 and

Rain_4 during the week before anthesis (Fig. 2C and D). Disease and toxin responses were lowest and most variable for Rain_3. These results are probably a reflection of the effects of dry-wet moisture cycles on spore production and release, and stimulation of mycotoxin production. This is consistent with findings from previous field and greenhouse experiments (1,2), suggesting that the pattern and distribution of rainfall may affect FHB and grain quality.

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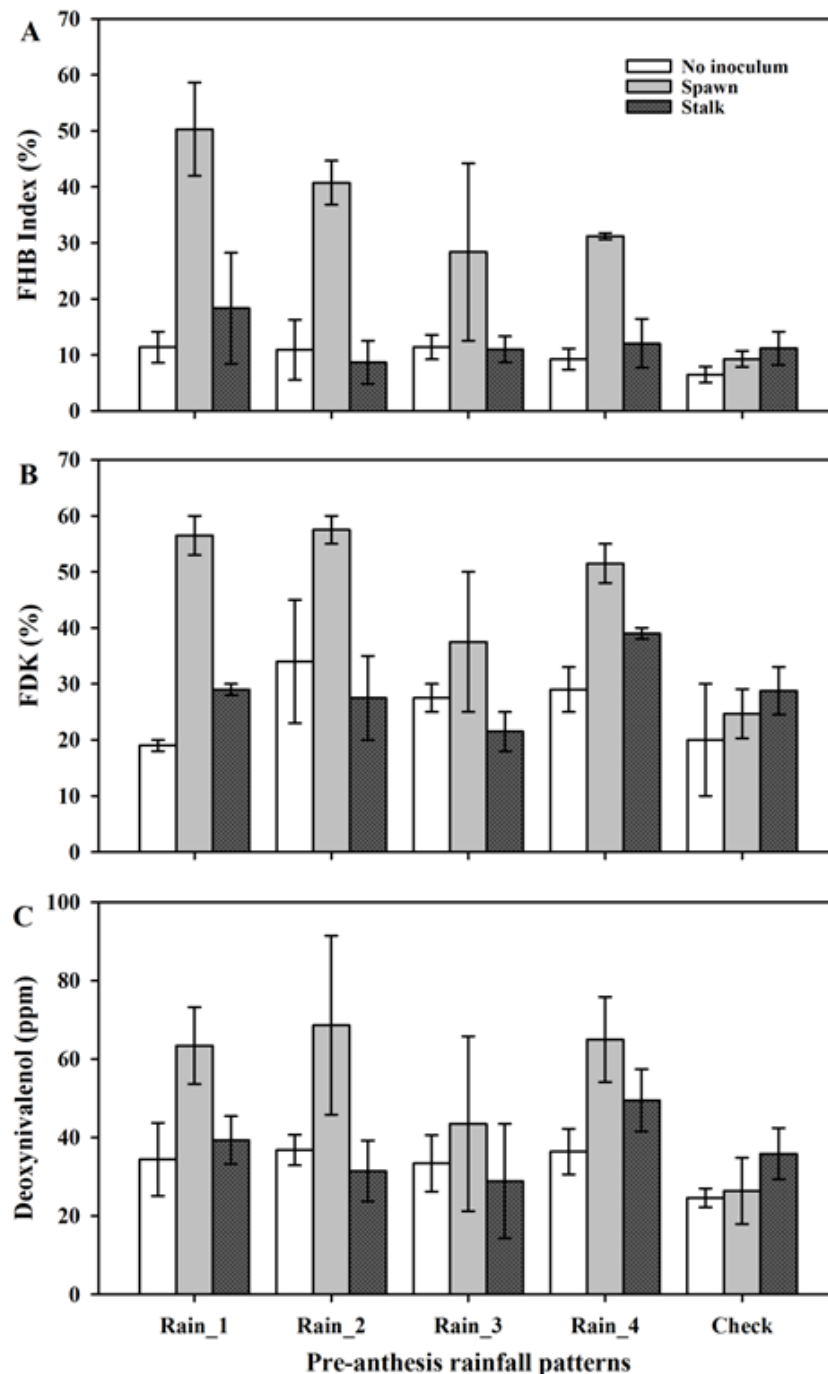


Figure 1. Mean Fusarium head blight (FHB) index (A), *Fusarium* damaged kernels (FDK) (B), and deoxynivalenol (C) content of harvested grain (ppm) under different pre-anthesis rainfall patterns and inoculum sources. Rain_1 = rain every day for 8 days, Rain_2 = rain only on the first and last two days of the window, Rain_3 = rain only on the middle four days of the window, Rain_4 = rain every other day, Check = ambient rainfall. No inoculum = without in-field inoculum, Spawn = *F. graminearum* colonized corn kernels, Stalk = naturally-infected corn crop residue. Each bar represents the treatment arithmetic mean from two replicate plots, and error bars are standard error of the mean.

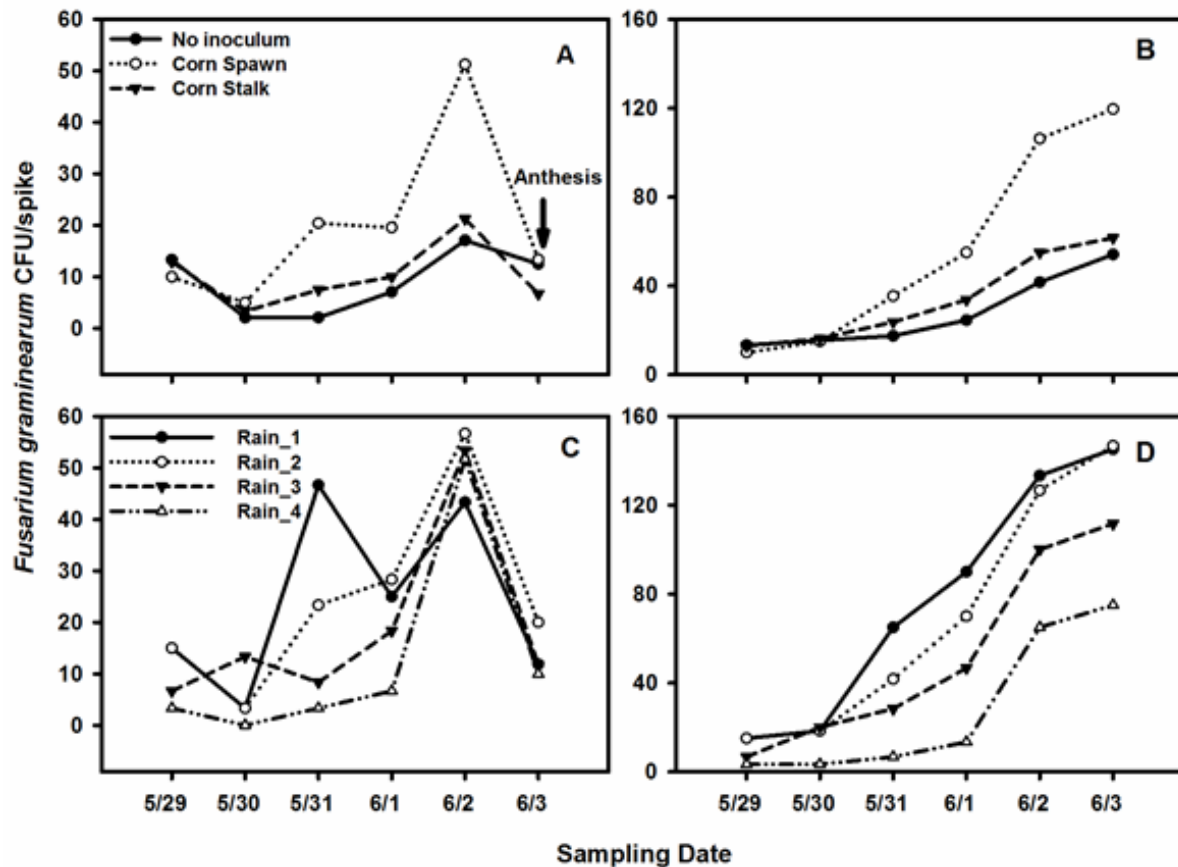


Figure 2. Mean (A and C) and cumulative number of (B and D) CFU/spike for different inoculum sources, averaged across rainfall patterns (A and B), and for different rainfall patterns applied to plots with corn spawn inoculum (C and D). Rain_1 = rain on every day for 8 days immediately prior to anthesis, Rain_2 = rain only on the first and last two days of the 8-day window, Rain_3 = rain only on the middle four days of the window, Rain_4 = rain on every other day. No inoculum = no in-field inoculum, and Corn Spawn and Corn Stalk = *F. graminearum* colonized corn kernels and naturally-infected corn residue, respectively, spread between the rows of the plot. Note: an outlier (1,236 CFU/spike on June 2 for Rain_1 plus corn spawn in block 2) was removed.

2013 AND 2014 FIELD PLOT TRIALS FOR BIOLOGICAL CONTROL OF FUSARIUM HEAD BLIGHT IN SOUTH DAKOTA USING *BACILLUS AMYLOLIQUEFACIENS* STRAINS

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ABSTRACT

Fusarium Head Blight (FHB) or Wheat Scab, caused by *Fusarium graminearum* is an economically important disease of wheat and barley. Yield losses can 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 to analyze the efficacy of *Bacillus amyloliquefaciens* strains 1BA and 1D3 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 were done on Briggs spring wheat heads at Feekes 10.51. In the 2013 Field Plot trials, multiple treatments exhibited statistically significant reduced levels of DON in comparison to the untreated control. *Bacillus* strain 1BA amended with colloidal chitin, Prosaro and plant oil reduced DON levels significantly (P=0.10), in comparison to treatment with Prosaro alone. Only the DON results from 2013 plots are presented in this abstract, as the other results were presented for the 2013 FHB Forum.

For 2014, no statistically significant treatment differences were observed for FHB incidence, severity, index and yield. The combination of *Bacillus* 1BA, plant oil, colloidal chitin and Prosaro reduced the FHB incidence to 35.5%, which was less than the FHB incidence observed for Prosaro alone (42.5%) or for the untreated control (48.5%). The treatment combination of *Bacillus* strains 1BA, plant oil, and Induce NIS reduced the FHB severity to 51.52%, which was less than the FHB severity observed for Prosaro alone (51.81%) or the untreated control (67.64%). The treatment of *Bacillus* strain 1BA and 1D3 with plant oil, colloidal chitin and Prosaro reduced the disease index to 21.24%, while the treatment of Prosaro alone reduced the disease index to 22.16%. The FHB index of untreated control was 31.57%; further, the treatment of *Bacillus* strain 1D3 with plant oil, colloidal chitin and Prosaro increased the yield to 56.68 bu/acre, while the treatment of Prosaro alone increased the yield to 54.8 bu/acre. The yield for the untreated control was 49.43 bu/acre. Several treatments with the BCAs showed significant differences (P=0.10) for grain test weight in comparison to the untreated control. The Disease Protein, FDK and DON data are not yet available as of November 2014.

These trials demonstrated that *Bacillus* strains 1BA or 1D3 in combination with Prosaro and/or colloidal chitin with plant oil can reduce FHB in wheat, more than a single application of Prosaro.

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INTEGRATING CULTIVAR RESISTANCE AND FUNGICIDE APPLICATION TO MANAGE FUSARIUM HEAD BLIGHT OF WHEAT

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ABSTRACT

Fusarium head blight (FHB) is a devastating disease of wheat in the southern cone of South America. FHB represents one of the main constraints for wheat production in Uruguay, where moderate to severe outbreaks have occurred in one of every four years over the past two decades. In order to optimize disease control measures, cultivar resistance and fungicides were investigated and their interaction was evaluated. Commercial cultivars and advanced lines were characterized under intermediate to high disease pressure in nurseries and field trials during 2011 to 2013. Few commonly grown cultivars had high levels of resistance and comprised 7, 10, and 15% of the area planted to wheat in 2011, 2012, and 2013, respectively. Metconazole alone or in combination with epoxiconazole were the most effective fungicides in controlling FHB, by reducing FHB index (FHBI), *Fusarium* damaged kernels (FDK) and deoxynivalenol (DON) content and increasing grain yield. Although some mixtures of triazoles + strobilurins and triazoles + carboxamides + strobilurins reduced FHBI and FDK, they increased DON content. Fungicide efficacy in reducing FHB and DON and in increasing grain yield was greater in a moderately resistant cultivar (INIA-Genesis 2375) than in a susceptible one (INIA Don Alberto). These results suggest that it may be possible to manage FHB by cultivar resistance and timely fungicide applications with recommended triazoles.

CONTROL OF FHB WITH RESISTANT GENOTYPES AND FUNGICIDES: 2014 RESULTS

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ABSTRACT

A series of variety by fungicide trials were conducted in 2014 in the three classes of wheat that are grown in North Dakota: hard red spring, hard red winter and durum. Experiments consisted of a factorial combination of variety and fungicide at Feekes 10.51 stage. The number of varieties varied by class and location and included those varieties that had been released and were likely to be grown in the state. Treatments were replicated three times. The fungicide used was a commercial combination of tebuconazole + prothioconazole (Prosaro™, Bayer CropScience) at a rate of 6.5 fl oz per acre with NIS. Winter wheat was planted no-till after spring wheat. Durum and spring wheat followed various other crops depending on the location and were planted after tillage at some locations and no-till at others. All locations were subject to natural FHB infestation and rainfall. Yield, disease severity and DON levels were recorded and data were analyzed using standard statistical techniques. Fairly high levels of FHB occurred especially in the winter wheat and the durum at Minot. In winter wheat, fungicide reduced DON levels from 5.9 ppm to 3.5 ppm at Prosper and from 12.1 to 9.1 ppm at Forman. Within the treatments that received no fungicide, varieties had DON levels of 13.8 to 1.2 ppm and 32.5 to 2.8 ppm at Prosper and Forman, respectively. The cultivar Emerson which was developed in Canada showed excellent resistance to FHB. With fungicide treatment, yields increased by 10 bu/a at Prosper and 18 bu/a at Forman. In spring wheat, there was little FHB development at Forman. DON levels were reduced from 0.4 to 0.1 ppm and yield increased from 49.0 to 51.9 bu/a with the fungicide treatment. At Hope, where there was slightly more FHB pressure, fungicide reduced DON from 1.0 to 0.1 ppm averaged across varieties. Within the no fungicide treatment, varieties ranged from 0.0 to 3.2 ppm DON. Yield increased by 5 bu/a on average with fungicide treatment. In the durum experiment at Minot, DON levels were reduced from 16 to 12 ppm with the fungicide treatment, and varied from 23 to 10 ppm between varieties. These data show the importance of varietal resistance relative to fungicide in the control of FHB. Fungicide can play an important role in reducing DON levels to an acceptable level when resistant cultivars are used or when disease pressure is not excessive.

ACKNOWLEDGEMENT AND DISCLAIMER

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EVALUATION OF HRWW AND HRSW CULTIVARS FOR FHB MANAGEMENT IN SOUTH DAKOTA

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ABSTRACT

Fusarium head blight (FHB – scab) remains a serious concern for wheat and barley producers in South Dakota. One of the sustainable and affordable means of FHB management is through host resistance. The objective of this study was to evaluate the different commercial hard red winter and spring wheat cultivars for FHB management in South Dakota. Fourteen winter wheat cultivars and nineteen spring wheat cultivars were evaluated. The winter wheat cultivars were planted at two locations, Volga and South Shore; whereas the spring wheat cultivars were planted only at Volga. Experimental design used was complete randomized block with four replications. The spring wheat cultivars were under ambient conditions until anthesis, after which mist irrigation was applied. Winter wheat was left under ambient conditions at both locations. Twenty-one days following anthesis, plots were evaluated for leaf diseases, FHB incidence, FHB head severity, FHB field severity, and FHB disease index (FHB incidence x severity). At harvest, grain yield and test weight were determined. Grain samples were collected for assessing *Fusarium* damaged kernels (FDK) and deoxynivalenol (DON).

The Volga location generally had high FHB pressure (highest FHB index of 38.6) while the South Shore location had low FHB pressure (highest FHB index of 11.5) At the Volga Research Farm location, the winter wheat cultivars that had the lowest FHB disease index were Arapahoe (4.51%), Lyman (6.54%) Ideal (7.31%), and Matlock (9.65%) whereas at South Shore, the cultivars that had the lowest FHB Disease Index were Matlock (0.91%), Redfield (1.05%), Arapahoe (1.06%), Everest (1.51%), Expedition (2.29%), and Ideal (3.20%), . The winter wheat cultivars that had the highest yield were Matlock and Arapahoe at Volga and Matlock and Redfield at South Shore. For the spring wheat cultivars that were tested at the Volga Research Farm, the cultivars that had the lowest FHB Disease Index were LCS Iguacu (1.09%), LCS Albany (1.99%), and WB9507 (3.15%). Forefront (3.25%), Sabin (3.72%), SY Ingmar (3.93%), and SY Soren (4.24%) The highest yielding spring wheat cultivars at Volga were Forefront and Prevail.

These results indicate FHB moderate resistance for both winter and spring commercial wheat cultivars in South Dakota for the management of FHB.

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BEST FHB MANAGEMENT PRACTICES: A 2014 MULTI-STATE PROJECT UPDATE

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OBJECTIVE

Evaluate the integrated effects of fungicide and genetic resistance on FHB and DON in all major grain classes, with emphasis on different application timings and new genotypes to develop more robust “*best-management practices*” for FHB and DON.

INTRODUCTION

Over the last 15 years, considerable progress has been made to develop management strategies to minimize FHB-associated grain yield and quality losses in wheat and barley. Several new resistant cultivars have been developed, efficacious fungicides registered, accurate disease forecasting models deployed to help guide fungicide applications, and the value of integrating multiple in-field and grain harvesting strategies to manage this disease-toxin complex has been demonstrated (Salgado et al., 2014; Willyerd et al., 2012; McMullen et al., 2012). For instance, results from several years of coordinated integrated management trials showed that relative to the untreated susceptible check, the combination of moderately resistant cultivar and Prosaro application at anthesis resulted in more than 70% control of both FHB index and DON (Willyerd et al., 2012). However, weather conditions, fungicide

and spray associated costs, cultivar yield potential and other factors often prevent the adoption of current management recommendations. For instance, wet, soggy field conditions may make it impossible for ground applications of fungicides at the recommended anthesis growth stage. Moreover, even if such applications are made, research shows the rainfall during or shortly after treatment may reduce fungicide efficacy (Andersen et al., 2014). Other limitations to adequate timing of fungicide applications include uneven crop development and variable anthesis window within a field, and the inability to correctly determine the anthesis growth stage. These limitations have led to questions being asked about the efficacy of applying fungicides before or after anthesis.

MATERIALS AND METHODS

Field experiments were established in 12 US wheat-growing states (AR, DE, IL, IN, MD, MI, MN, ND, NE, NY, OH and SD) to investigate the effects of cultivar resistance and fungicide application timing on FHB and DON. Plots were established following host or non-host crops of *F. graminearum*, according to standard agronomic practices for each location. At least three commercial wheat cultivars, classified as susceptible (S), moderately susceptible (MS), or moderately resistant (MR), were planted in most

trials. However, some trials only included one or two of these resistance categories. Plots were planted in four to six replicate blocks. The standard experimental design was a randomized complete block, with a split-split-plot arrangement of cultivar as whole-plot and fungicide (Prosaro, 6.5 fl. oz/A + NIS) application timing as sub-plot (untreated or treated at anthesis [A] or 2 to 7 days post-anthesis [A+2 ... A+7, respectively]). All plots were artificially inoculated with either *F. graminearum*-colonized corn kernels spread on the soil surface or spray-inoculated with a spore suspension of the fungus approximately 24-36 hours following the anthesis fungicide treatment. FHB index (plot severity) was assessed during the soft dough stages of grain development. Milled grain samples were sent to a USWBSI-supported laboratory for toxin analysis. For the purpose of this report, percent control of FHB index and DON was estimated for each cultivar x fungicide application timing combination relative to the untreated susceptible check, and the best management practice, based on percent control, was highlighted for each trial/environment.

RESULTS AND DISCUSSION

For this report, data from 14 trials, representing seven soft red winter wheat, two hard red winter wheat, four hard red spring wheat, and one soft white winter wheat classes were summarized (Table 1). Means for each cultivar resistance class x fungicide application timing combination are shown in Table 1. Mean FHB index in the untreated susceptible check ranged from 0 to 49%, and mean DON from 0.5 to 15.6 ppm. In some locations, FHB did not develop due to unfavorable weather conditions. In addition, DON data were not available for some trials at the time of this report, therefore trials with missing data or nominal disease and toxin levels (< 4% index and < 2 ppm DON, Table 1) were not used to estimate percent control. Percent control of FHB index and DON, relative to the untreated susceptible check is shown in Figs. 1 and 2 for trials with the highest levels of mean index and DON in the check (and where possible, representative of each market

class). The best management combinations, based on the highest percent control of index, for trials/environment with index > 4% are presented in Fig. 3.

Fungicide alone reduced FHB index and DON in each resistance category and wheat market class, however, the combination of cultivar resistance and fungicide application was most effective at reducing FHB and DON in most trials (Table 1 and Figs. 1-3, in 8 out of 11 trials reporting FHB index > 4%). In some cases (ENV = 3, 8 and 9) fungicide-treated MS cultivars had the highest percent control of both FHB and DON, and post-anthesis treatments in ENV 3, 8, 9, 10, and 13 were as effective as or more effective than anthesis treatments (Figs. 1-3). Based on these results, there is evidence suggesting that applying fungicides post-anthesis may be as efficacious against FHB and DON as treatments applied at anthesis in all wheat classes and environments.

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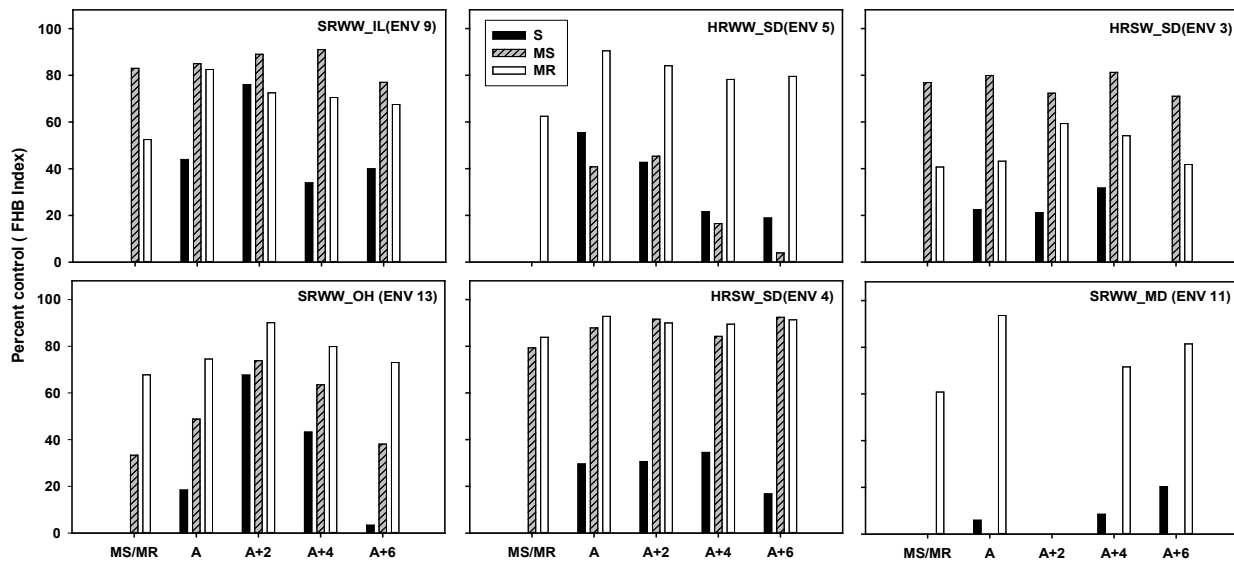


Figure 1. Percent control of FHB index relative to the untreated susceptible check (Table 1) for different FHB management combinations in three different wheat classes (HRWW, HRSW, and SRWW). Cultivar resistance (susceptible, S; moderately susceptible, MS; and moderately resistant, MR). MS/MR, represents the effect of cultivar resistance alone (untreated MR or MS cultivar). Prosaro (6.5 fl oz/A) was applied either at anthesis (A), or 2, 4, or 6 days post-anthesis (A+2, A+4 or A+6, respectively).

Table 1. Mean FHB index and DON for different cultivar x fungicide timing management combinations from 14 coordinated integrated management trials (ENV, environments) representing different wheat classes (TYPE = HRWW, HRSW, SRWW and SWWW). Results are organized by cultivar FHB resistance reaction (susceptible, S; moderately susceptible, MS; and moderately resistant, MR) and fungicide treatment (untreated [No] or treated [TR] at anthesis [Yes] or 2, 4, 5, 6 or 7 days post-anthesis.

Response	ENV	LOCATION	TYPE	Susceptible (S)						Moderately Susceptible (MS)						Moderately Resistant (MR)								
				Anthesis TR		Post anthesis TR (days)		Anthesis TR		Post anthesis TR (days)		Anthesis TR		Post anthesis TR (days)		Anthesis TR		Post anthesis TR (days)						
				No	Yes	2	3	4	5	6	7	No	Yes	2	4	5	6	7	No	Yes	2	4	5	6
FHB Index ^a (%)	1	Langdon, ND	HRSW	10.1	4.6	2.1	1.7	0.6	1.6	...
	2	Fargo, ND	HRSW	0.0	0.1	0.0	0.0	0.0	0.5	...
	3	Volga, SD	HRSW	48.8	37.8	38.4	...	33.3	...	50.6	11.3	9.8	13.5	9.2	...	14.1	...	28.9	27.7	19.8	22.4	...	28.4	...
	4	South Shore, SD	HRSW	18.8	13.2	13.0	...	12.3	...	15.6	3.9	2.3	1.6	3.0	...	1.4	...	3.0	1.4	1.9	2.0	...	1.6	...
	5	Volga, SD	HRWW	19.0	8.5	10.9	...	14.9	...	15.4	30.6	11.3	10.4	15.9	...	18.3	...	7.2	1.8	3.0	4.2	...	3.9	...
	6	Mead, NE	HRWW	3.1	3.1	2.4	...	2.4	...	2.7	1.5	1.6	1.7	1.8	...	1.8	...
	7	Georgetown, DE	SRWW	7.0	4.9	4.1	...	3.3	0.4	0.1	0.7	...
	8	Dixon Springs, IL	SRWW	7.3	8.5	3.8	...	0.7	...	1.1	1.6	1.1	0.6	0.0	...	0.7	...	1.9	3.0	1.9	1.7	...	0.8	...
	9	Urbana, IL	SRWW	12.5	7.0	3.0	...	8.3	...	7.5	2.1	1.9	1.4	1.1	...	2.9	...	5.9	2.2	3.4	3.7	...	4.1	...
	10	West Lafayette, IN	SRWW	4.4	2.3	1.8	...	2.8	...	1.7	0.5	0.4	0.4	0.5	...	0.2	...	0.4	0.2	0.2	0.2	...	0.5	...
	11	Wye, MD	SRWW	13.2	12.4	12.0	...	10.5	5.2	0.8	3.7	...
	12	Aurora, NY	SRWW	4.2	1.8	0.5	...	1.0	0.8
	13	Wooster, OH	SRWW	12.6	10.3	4.1	...	7.2	...	12.2	8.4	6.5	3.3	4.6	...	7.8	...	4.1	3.2	1.3	2.5	...	3.4	...
	14	Deckerville, MI	SWWW	0.6	0.3	0.1	...	0.2
DON ^b (ppm)	6	Mead, NE	HRWW	0.6	1.2	0.8	...	1.1	...	0.7	0.6	0.6	1.3	0.9	...	0.9	...
	7	Georgetown, DE	SRWW	2.0	1.2	1.2	...	1.2	0.3	0.3	0.2	...
	8	Dixon Springs, IL	SRWW	4.0	7.9	3.0	...	2.4	...	3.0	0.8	0.9	0.8	0.3	...	1.0	...	1.2	3.3	1.3	1.1	...	1.7	...
	9	Urbana, IL	SRWW	7.6	5.7	5.1	...	4.4	...	4.5	1.2	1.9	1.8	2.1	...	1.9	...	2.8	2.6	2.3	2.7	...	2.1	...
	10	West Lafayette, IN	SRWW	7.1	7.3	4.5	...	4.1	...	4.1	2.4	2.3	2.1	2.0	...	1.8	...	3.9	2.6	3.0	2.5	...	2.1	...
	11	Wye, MD	SRWW	1.9	1.6	1.1	...	0.9	0.6	0.5	0.4	...
	12	Aurora, NY	SRWW	3.2	1.3	0.8	...	1.3	0.5
	13	Wooster, OH	SRWW	15.6	9.0	7.3	...	9.1	...	9.2	6.1	5.3	4.6	4.4	...	5.0	...	4.2	3.5	2.1	2.5	...	2.4	...
	14	Deckerville, MI	SWWW	0.5	0.5	0.1	...	0.1

^aFHB index = mean proportion of disease spikelets per spike.

^bDON = deoxynivalenol content of harvested grain in ppm.

^cFungicide application = Prostaro applied at 6.5 fl. oz./A + NIS at or after anthesis.

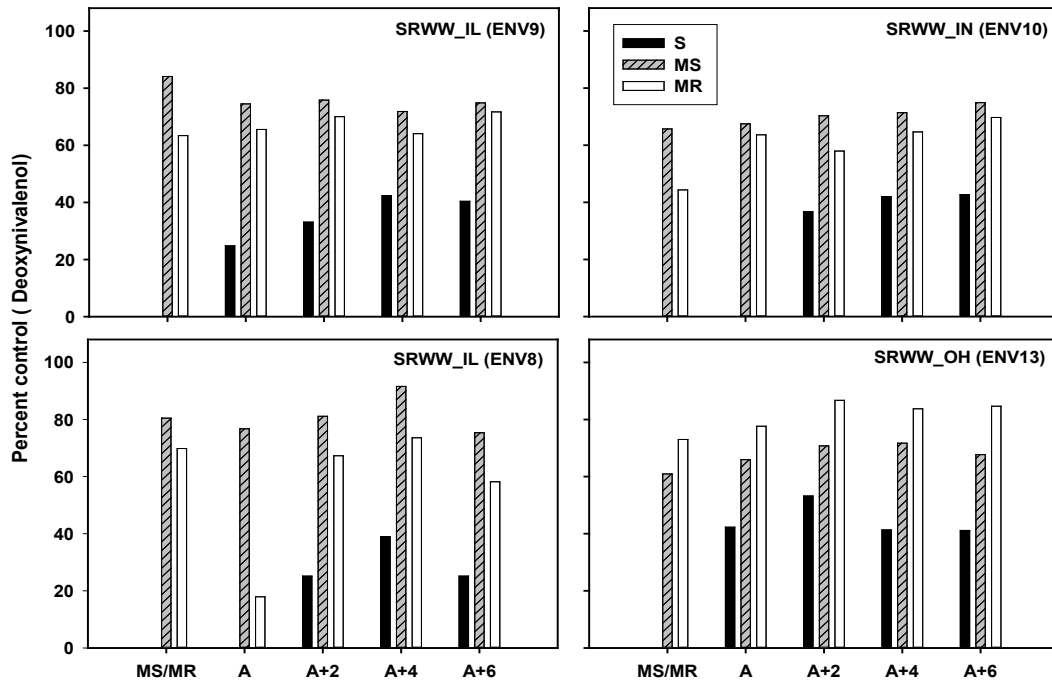


Figure 2. Percent control of DON relative to the untreated susceptible check (Table1) for different FHB management combinations in trials with mean DON check > 2 ppm. Cultivar resistance (susceptible, S; moderately susceptible, MS; and moderately resistant, MR). MS/MR, represents the effect of cultivar resistance alone (untreated MR or MS cultivar). Prostaro (6.5 fl oz/A) was applied either at anthesis (A), or 2, 4, or 6 days post-anthesis (A+2, A+4 or A+6, respectively).

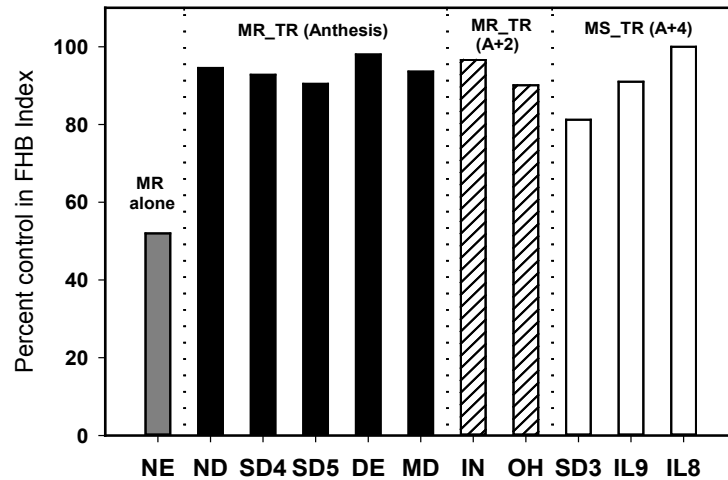


Figure 3. Percent control of FHB index relative to the untreated susceptible check for the best management combinations in environments that reported FHB index levels in the susceptible untreated check above 4% (Table 1). Environments are grouped based on the cultivar x fungicide application timing combination with the highest percent control. Cultivar FHB resistance reaction (susceptible, S; moderately susceptible, MS; and moderately resistant, MR). Plots were treated (TR) with Prosaro (6.5 fl oz/A) either at anthesis (A), or 2, 4, or 6 days post-anthesis (A+2, A+4 or A+6, respectively). MR alone = the effect of moderate resistance in the absence of fungicide.

INTEGRATING CULTIVAR RESISTANCE AND FUNGICIDE APPLICATION RATE AND TIMING FOR FHB MANAGEMENT IN OHIO

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OBJECTIVES

The objective of this study was to develop more robust strategies for FHB management in soft red winter wheat in Ohio, by reevaluating the efficacy of post-anthesis fungicide treatments as influenced by cultivar resistance, fungicide chemistry, and application rate.

INTRODUCTION

Best-management practices for *Fusarium* head blight (FHB) and deoxynivalenol (DON) in wheat include the use of moderately resistant cultivars, crop rotation, tillage, and fungicide application at anthesis. However, weather and other farm-related factors may prevent the adoption of these strategies or reduce their efficiency. For instance, rainfall during anthesis may reduce the efficacy of fungicides or even prevent them from being applied at the recommended time. Previous research has shown that Prosaro applications made after anthesis may be just as effective as or sometime more effective than applications made at anthesis (D'Angelo et al 2014). However, it is unclear whether the efficacy of post-anthesis applications will be influenced by active ingredient, application rate, or resistance of the cultivar being treated.

MATERIALS AND METHODS

Two field experiments were established in Wooster, Ohio during the 2014 season, using a split-plot arrangement as the experimental design. In the first study, Prosaro (6.5 fl. oz./A) was applied to four cultivars with different levels of resistance to FHB

(Hopewell, susceptible; Bromfield, moderately susceptible; and Truman and Malabar, both moderately resistant), and in the second, Prosaro and Caramba were applied to Hopewell at low and high rates (6.5 and 8.2 fl. oz./A for Prosaro and 13.5 and 17 fl. oz./A, for Caramba). In both studies, treatments were either applied at 50% anthesis or between 2 and 7 days after anthesis. All plots were spray-inoculated at anthesis with a spore suspension of *F. graminearum*, and FHB intensity and *Fusarium* damaged kernel (FDK) were rated, grain yield estimated, and grain samples tested for DON. FHB intensity and DON data were arcsine-square root- and log-transformed, respectively, and analyzed using a linear mixed modeling approach.

RESULTS AND DISCUSSION

Mean FHB index (IND) ranged from 1.5 to 21% and 0.7 to 16% in untreated and fungicide-treated plots, respectively. The corresponding ranges for mean DON were 3.3 to 16.7 and 0.9 to 15 ppm. The effects of cultivar and fungicide x rate combination on IND, FDK, DON, and grain yield did not depend on application time (the interactions were not significant, $P > 0.05$) (**Table 1 and 2**). Differences in mean IND and DON among cultivars and fungicide treatments ($P < 0.05$) were statistically significant (**Table 3 and 4**). Averaged across application time, Truman and Malabar had significantly lower mean IND, FDK, and DON than Hopewell and Bromfield (**Table 3 and Fig.1**), and Prosaro at the high rate had significantly lower mean IND, and numerically, but not always statistically, lower mean FDK and DON than the other tested fungicide x rate combinations (**Table**

4 and Fig.2). Fungicide-treated plots generally had significantly lower mean IND, FDK, DON, and higher mean grain yield than the untreated check. Averaged across cultivars (experiment 1) or fungicide x rate combinations (experiment 2), treatments applied between two and five days post-anthesis had significantly lower mean IND and DON ($P < 0.05$) than those applied at anthesis, and comparable or significantly lower mean FDK (Table 3, Table 4, Fig. 1 and Fig. 2). The effects of treatments made more than five days after anthesis varied between the two experiments (Table 3 and Table 4). Cultivar and fungicide x rate combination did not have a significant effect on grain yield (Table 1 and Table 2); however, for all application times, treated plots had significantly higher mean yield (between 307 and 644 kg/ha) than the untreated check (Table 3 and Table 4). In general, mean yields were comparable among anthesis and post-anthesis treatments. The only exception was for the treatment applied two days after anthesis in experiment 1, which had significantly higher mean yield than the anthesis treatment.

Our results showed that using a moderately resistant cultivar reduced mean IND by 59 to 76% and DON by 72 to 74% when compared to untreated-susceptible check, and fungicide alone

reduced IND by 3 to 84% and DON by 41 to 68%, relative to the check. However, combinations of moderate resistance and fungicide, particularly treatments made between at 2 and 4 days after anthesis, were the most efficacious, with mean percent control relative to the susceptible-untreated ranging from 68 to 96% for IND and 81 to 88% for DON, and percent yield increase ranging from 8 to 12%.

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Table 1. Summary statistics from linear mixed model analyses of the effect of cultivar and fungicide timing on arcsine-transformed FHB incidence (INC), index (IND), *Fusarium* damaged kernels (FDK) and log-transformed deoxynivalenol (DON) grain contamination and grain yield (YLD) in soft red winter wheat in Ohio.

Factor	IND	FDK	DON	YLD
Cultivar	<0.001	<0.001	<0.001	0.086
TIME	<0.001	<0.001	<0.001	<0.001
Cultivar*TIME	0.397	0.050	0.499	0.219

Cultivar = Hopewell, susceptible; Bromfield, moderately susceptible; and Truman and Malabar, both moderately resistant. Time = Time of Prosaro application - anthesis or 2, 4 or 6 days after anthesis

Table 2. Summary statistics from linear mixed model analyses of the effect of fungicide and application timing on arcsine-transformed FHB incidence (INC), index (IND), *Fusarium* damaged kernels (FDK) and log-transformed deoxynivalenol (DON) grain contamination and grain yield (YLD) in soft red winter wheat in Ohio.

Factor	IND	FDK	DON	YLD
Fungicide	0.001	0.080	0.019	0.314
TIME	<0.001	0.001	<0.001	0.220
Fungicide*TIME	0.054	0.922	0.250	0.807

Fungicide = Prostaro and Caramba applied at low and high rates (6.5 and 8.2 fl. oz./A for Prostaro and 13.5 and 17 fl. oz./A, for Caramba).

Time: Time of fungicide application - anthesis or 2, 5 or 7 days after anthesis

Table 3. Probability values for pairwise differences of least square means from linear mixed model analyses of the effect of cultivar and fungicide timing on arcsine-transformed FHB incidence (INC), index (IND), *Fusarium* damaged kernels (FDK) and log-transformed deoxynivalenol (DON) grain contamination and grain yield (YLD) in soft red winter wheat in Ohio.

Contrast	IND	FDK	DON	YLD
Check vs A	0.013	<0.001	0.002	0.001
Check vs A+2	<0.001	<0.001	<0.001	<0.001
Check vs A+4	<0.001	<0.001	<0.001	0.001
Check vs A+6	0.319	0.001	<0.001	0.001
A vs A2	<0.001	0.087	0.001	0.001
A vs A4	0.002	0.844	0.026	0.927
A vs A6	0.120	0.449	0.063	0.977
Hopewell vs Bromfield	0.010	<0.001	0.001	0.362
Hopewell vs Malabar	<0.001	<0.001	<0.001	0.016
Hopewell vs Truman	<0.001	<0.001	<0.001	0.195
Truman vs Bromfield	<0.001	<0.001	0.002	0.670
Truman vs Malabar	0.003	0.016	0.569	0.153
Bromfield vs Malabar	0.015	0.001	0.005	0.077

Check = untreated and A = fungicide application at anthesis or 2 (A+2), 4 (A+4) or 6 (A+6) days after anthesis. Hopewell, susceptible; Bromfield, moderately susceptible; and Truman and Malabar, both moderately resistant.

Table 4. Probability values for pairwise differences of least square means from linear mixed model analyses of the effect of fungicide and application timing on arcsine-transformed FHB incidence (INC), index (IND), *Fusarium* damaged kernels (FDK) and log-transformed deoxynivalenol (DON) grain contamination and grain yield (YLD) in soft red winter wheat in Ohio

Contrast	IND	FDK	DON	YLD
Check vs A	<0.001	<0.001	<0.001	<0.001
Check vs A+2	<0.001	<0.001	<0.001	<0.001
Check vs A+5	<0.001	<0.001	<0.001	<0.001
Check vs A+7	<0.001	<0.001	<0.001	<0.001
A vs A2	<0.001	0.047	0.003	0.379
A vs A5	<0.001	<0.001	<0.001	0.164
A vs A7	0.001	0.063	0.002	0.595
Prosaro low vs Prosaro high	0.049	0.550	0.019	0.128
Prosaro low vs Caramba low	0.003	0.048	0.297	0.730
Prosaro low vs Caramba high	0.449	0.487	0.436	0.172
Prosaro high vs Caramba low	<0.001	0.017	0.003	0.219
Prosaro high vs Caramba high	0.014	0.211	0.072	0.851
Caramba low vs Caramba high	0.009	0.153	0.087	0.288

Check = untreated and A = fungicide application at anthesis or 2 (A+2), 4 (A+5) or 6 (A+7) days after anthesis. Prosaro and Caramba applied at low and high rates (6.5 and 8.2 fl. oz./A for Prosaro and 13.5 and 17 fl. oz./A, for Caramba).

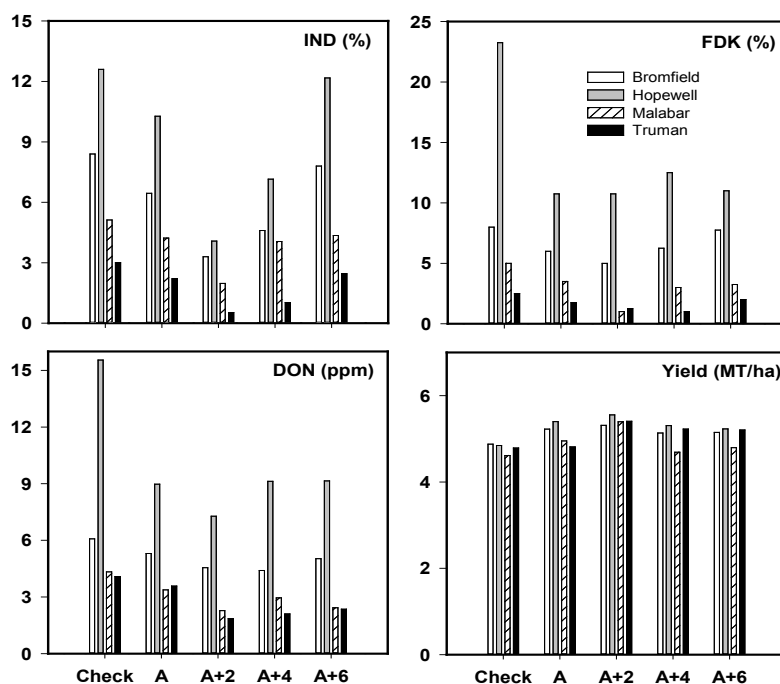


Fig. 1. Mean FHB index (IND), FDK, DON and grain yield from untreated (Check) and Prosaro-treated (6.5 fl oz/A) plots of soft red winter wheat cultivars Bromfield (moderately resistant), Hopewell (susceptible), and Malabar and Truman (moderately resistant). Treatments were made at anthesis (A), or 2, 4, or 6 days post-anthesis (A+2, A+4 or A+6, respectively).

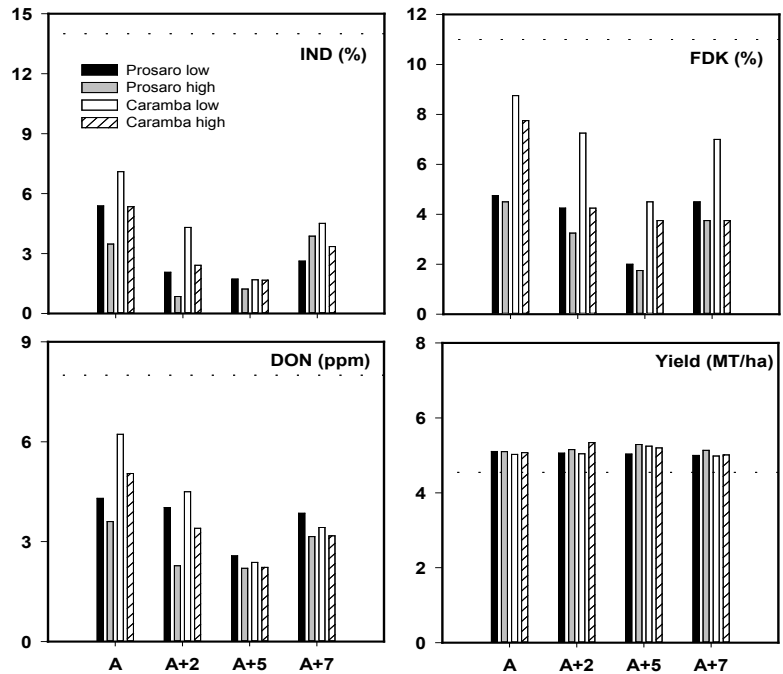


Fig. 2. Mean FHB index (IND), FDK, DON and grain yield from plots of susceptible cultivar Hopewell treated with four fungicide-rate combinations: Prosaro at low and high rates (6.5 and 8.2 fl. oz./A) and Caramba at high and low rates (13.5 and 17 fl. oz./A, respectively). Treatments were applied at anthesis (A) or 2, 5, or 7 days post-anthesis (A+2, A+5 and A+7, respectively). Dashed line indicates untreated check values.

WEATHER TIME SERIES CURVES IN RELATION TO FUSARIUM HEAD BLIGHT EPIDEMICS

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ABSTRACT

Our long term goal is to develop and deliver predictive models for Fusarium head blight (FHB) epidemics in the U.S. The current FHB observational data matrix was updated to include data from 2010 onwards sent by collaborators participating in U.S. Wheat & Barley Scab Initiative (USWBSI) FHB coordinated integrated management projects. The new data expanded the data matrix from 527 to 865 observations, a 64% increase. Sixteen states are now represented, with 74% of the observations coming from winter wheat and the remainder from spring wheat. FHB epidemics, defined as FHB index $\geq 10\%$ (this threshold having been determined by observing the most susceptible varieties), had occurred in 236 of the observations. No FHB (i.e. FHB index = 0) was recorded in 184 of the remaining 629 observations. Latitude and longitude coordinates associated with each location-year were used to identify the closest reporting weather station with air temperature, dew point and pressure data. Weather data, from September 01 of the year preceding anthesis to 30 days post-anthesis, were downloaded via Mathematica scripts, and summarized to hourly data after data integrity checks and cleaning. Missing values were imputed by interpolation or by an algorithm designed specifically for multivariate time series. Relative humidity and vapor pressure deficit were calculated from temperature and dew point. The hourly weather data were summarized to daily values. Mean curves for epidemics and non-epidemics were then plotted for the period beginning 120 days before anthesis and ending 20 days post-anthesis. During this period, mean daily temperature and dew point increased approximately linearly, with some apparent separation between the temperature curves a few days on either side of anthesis. Pressure showed a decreasing trend during this period, with multiple crossing-overs between the epidemic and non-epidemic curves. With relative humidity, there was a clear and consistent separation between the mean epidemic and non-epidemic curves, beginning around 35 days pre-anthesis and continuing into the post-anthesis period, with the epidemic relative humidity curve being above the non-epidemic curve. Similarly, for vapor pressure deficit, the mean epidemic curve was consistently below the non-epidemic curve from about 17 days pre-anthesis through 20 days post-anthesis. These exploratory time series analyses suggest that the signal capturing the difference between FHB epidemics and non-epidemics is strongest in moisture-related variables, beginning about 3 to 4 weeks pre-anthesis and extending as far as 3 weeks into the post-anthesis period.

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UNIFORM FUNGICIDE TRIAL RESULTS FOR MANAGEMENT OF FHB AND DON, 2014

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ABSTRACT

Data from prior USWBSI-funded uniform fungicide trials have shown that DMI fungicides Caramba® 90 SL, Prosaro® 421 SC were two of the most effective products against FHB and DON. However, for the grower these products can be relatively expensive. Tebuconazole fungicide is now off-patent, and several “generic” formulations are available at relatively low costs (some reports of less than \$3 per acre). In years where the risk of scab is predicted as low, and growers have fields planted to moderately FHB resistant varieties, many are making the economic decision to use generic tebuconazole products. The aim of this project was to compare generic formulations of tebuconazole products to evaluate any differences in efficacy and to examine bio control formulations for the suppression of FHB. Trials were conducted at multiple locations across six states (Arkansas, Illinois, North Dakota, South Dakota, Minnesota and New York) in 2014. All sites were inoculated with *Fusarium graminearum* infested corn spawn, infested residue, or spray inoculation with spores at flowering. In several locations, mist irrigation was used to promote disease development. Eleven common treatments were evaluated across locations; Prosaro, Caramba, Monsoon, Muscle, Onset, Orius, Tebustar, Toledo, Aproach, Aproach Prima and the biological control Taegro in combination with Prosaro. Additional rates of Caramba and/or Prosaro were tested. All treatments were applied at Feekes 10.5.1 (early anthesis). Preliminary analysis of the data revealed that all treatments helped reduce the incidence of FHB in comparison with plots that received no fungicide application. The fungicides Caramba and Prosaro appeared to provide the best control of FHB, yield increase and reduction in DON for those locations which reported DON levels. Not all generic tebuconazole products (Monsoon, Muscle, Onset, Orius, Tebustar and Toledo) performed equally across all locations.

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**FOOD SAFETY
AND
TOXICOLOGY**

TOWARDS UNDERSTANDING TRICHOHECENE-MEDIATED
DISRUPTION OF RIBOSOMAL FUNCTION
AND HENCE ITS TOXICITY

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ABSTRACT

Trichothecene mycotoxins are macrocyclic fungal metabolites known to inhibit protein synthesis in eukaryotic ribosomes. These toxins contaminate kernels of wheat and barley as a result of infection with *Fusarium* species. The major trichothecenes associated with *Fusarium*-infected kernels belong to the Type B class of trichothecenes and include deoxynivalenol (DON) and its acetylated derivatives, 3-acetyl DON and 15-acetyl DON. However, Type A class of trichothecenes, including T-2 toxin, have also been found in *Fusarium*-infected kernels. Different degrees of toxicity have been observed among the different trichothecenes, and these differences are specific to the class of organism in question. For instance, DON is known to be more phytotoxic than T-2 toxin, whereas T-2 toxin is more harmful in mammalian systems than DON. While it is known that these toxins inhibit protein synthesis by disrupting peptidyl transferase activity, the exact mechanism of this inhibition is poorly understood. Furthermore, it is not known how differences in trichothecene structure can affect different levels of toxicity. Our long-term goals are to better understand the toxicity mechanisms of these compounds, and as an initial step towards this end, we have employed a series of solid-state and solution-state nuclear magnetic resonance (NMR) spectroscopy experiments to study the three-dimensional structures and hydrogen-bonding patterns of both Type A and B trichothecenes. In our study, the epoxide ring (essential for toxicity) seems to constrain the configuration of the other side of the molecule (specifically the tetrahydropyranyl pocket). This tetrahydropyranyl pocket appears to be conserved in ribosomal binding, as was observed in the recently published structure of the yeast ribosome co-crystallized with different trichothecenes (Loubresse et al. 2014). We propose that the epoxide ring is not directly involved in trichothecene toxicity, but rather is essential in stabilizing the three dimensional structure required for establishing the configuration necessary for inhibition of protein synthesis.

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CHARACTERIZATION OF A DEOXYNIVALENOL-INACTIVATING UDP-GLUCOSYLTRANSFERASE AND ITS UTILIZATION FOR ENZYMATIC PRODUCTION OF DON- AND NIV-GLUCOSIDE

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ABSTRACT

Trichothecene mycotoxins such as the *Fusarium* metabolites deoxynivalenol (DON) and nivalenol (NIV) can be inactivated *in planta* by formation of glucose conjugates. Since such metabolites escape routine detection and the parental toxin can be reactivated by hydrolysis, they are termed “masked mycotoxins”. To date, there is insufficient information concerning the toxicological relevance of glucosylated mycotoxins. It has been shown that DON-3-O- β -D-glucoside (D3G) can be efficiently cleaved by intestinal bacteria, and is indeed cleaved in the intestinal tract of rats and pigs. While currently the concentrations of mycotoxin glucosides are rather low in infected cereals ongoing attempts to increase *Fusarium* resistance by wheat breeding may increase the contribution of glucosides to the total mycotoxin load. We identified a plant glucosyltransferase 1 family member which could be successfully expressed in *Escherichia coli* and efficiently glucosylates DON and NIV. This enzyme was purified by affinity chromatography and its biochemical properties were characterized. Substrate inhibition occurs at higher DON concentrations, and the enzyme is strongly inhibited by the second reaction product UDP. The enzyme is able to glucosylate both DON and NIV regioselectively at the position C3-OH, as confirmed by NMR of the purified glucosides. Using UDP-glucose as co-substrate, the complete conversion of larger amounts of DON (50 mg range) is possible within short reaction times (< 4 hours) *in vitro*. This enzyme is therefore a suitable biocatalyst to efficiently produce relevant amounts of DON and NIV-glucosides for use as analytic standards and for toxicological risk assessment, e.g. in animal feeding trials.

THE ZEARALENONE DETOXIFICATION PATHWAY
OF *TRICHOSPORON MYCOTOXINIVORANS*:
ELUCIDATION OF THE FIRST STEP

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ABSTRACT

Zearalenone (ZEN), a potent estrogenic mycotoxin produced by several *Fusarium* species, can be inactivated by the basidiomycete yeast *Trichosporon mycotoxinivorans*. ZEN is converted to a non-estrogenic metabolite [1], termed ZOM-1. We proposed a two-step mechanism involving a Baeyer-Villiger reaction leading to the formation of a hypothetical intermediate with a newly formed lactone bond, followed by lactone opening by an esterase. At this time the predicted intermediate could not be detected by LC-MS/MS in the cultures of wild-type *Trichosporon mycotoxinivorans* cells [1].

The gene encoding the first enzymatic step of the proposed pathway could be identified by a genetic approach. *Trichosporon mycotoxinivorans* was mutagenized by irradiation in a TRIGA Mark II nuclear reactor, followed by screening for mutants unable to degrade ZEN, which were detected using a *Saccharomyces cerevisiae* estrogen receptor bioassay. The determination of the genome sequences of the wild-type and mutant *Trichosporon mycotoxinivorans* strains and bioinformatical analysis revealed a large deletion containing a Baeyer-Villiger type monooxygenase candidate gene. Expression of a codon-optimized cDNA of this candidate gene in baker's yeast led to production of a metabolite of the mass of the expected hypothetical intermediate in ZEN treated transformants. After SPE pre-clean up and enrichment the metabolite was isolated by prep-HPLC and characterized by NMR. The estrogenicity of the new metabolite was tested using a yeast bioassay (based on induction of a *lacZ* reporter protein mediated by an expressed human estrogen receptor alpha fusion-gene). Nearly 100-fold higher concentrations of iZOM than ZEN are needed for activation of the reporter construct, demonstrating that the first step alone already leads to detoxification.

REFERENCE

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NEW TRICKS OF AN OLD ENEMY: ISOLATES OF
FUSARIUM GRAMINEARUM PRODUCE A TYPE
A TRICHOHECENE MYCOTOXIN

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ABSTRACT

The ubiquitous filamentous fungus *Fusarium graminearum* causes the important disease Fusarium head blight on various species of cereals, leading to contamination of grains with mycotoxins. In a survey of *F. graminearum* (*sensu stricto*) on wheat in North America several novel strains were isolated, which produced none of the known trichothecene mycotoxins despite causing normal disease symptoms. In rice cultures a new trichothecene mycotoxin (named NX-2) was characterized by liquid-chromatography-tandem-mass spectrometry. NMR measurements identified NX-2 as 3 α -acetoxy-7 α ,15-dihydroxy-12,13-epoxytrichothec-9-ene. Compared to the well-known 3-acetyl-deoxynivalenol it lacks the keto group at C-8 and hence is a type A trichothecene. Wheat ears inoculated with the isolated strains revealed a ten-fold higher contamination with its deacetylated form, named NX-3, (up to 540 mg kg⁻¹) compared to NX-2. The toxicities of the novel mycotoxins were evaluated utilizing two *in vitro* translation assays and the alga *Chlamydomonas reinhardtii*. NX-3 inhibits protein biosynthesis to almost the same extent as the prominent mycotoxin deoxynivalenol, while NX-2 is far less toxic, similar to 3-acetyl-deoxynivalenol. Genetic analysis revealed a different *TRII* allele in the N-isolates which was verified to be responsible for the difference in hydroxylation at C-8.

The occurrence of isolates producing the new toxin raises the question whether such strains have a selective advantage, and in the worst case can counteract progress made by plant breeders in the last decade. We will discuss the hypothesis that production of a toxin with an acetylated C3-OH may be a response of the fungus to circumvent inactivation by glycosylation, while lacking the keto-group may prevent glutathione-mediated detoxification. Population genetic studies to determine whether the frequency of NX-producers is changing seem highly warranted.

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**GENE DISCOVERY
AND
ENGINEERING
RESISTANCE**

A RAPID ASSAY FOR SYNTHETIC siRNA ACTIVITY AGAINST *TRI5*

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ABSTRACT

Host Induced Gene Silencing (HIGS) has been demonstrated in multiple plant species as an effective and novel type of resistance to pathogens. This system functions via RNA interference (RNAi), which is initiated by double stranded RNA (dsRNA). RNAi can be initiated by Dicer-mediated production of siRNA 21mers derived from the dsRNA, or siRNA can be synthesized. Certain species of siRNA that are homologous to target pathogen gene sequences can effectively suppress gene expression. Hundreds of possible 21mers can be derived from a long dsRNA, or single siRNAs can be designed based on conserved characteristics. Here we directly test synthesized 21mer siRNA silencing capability against GFP induction driven by the *TRI5* promoter in *Fusarium graminearum* strain *TRI5prom::GFP*. GFP fluorescence can be used to determine function of the trichothene-producing pathway via induction of *TRI5*. This strain also constitutively expresses RFP. The best induction of *TRI5* occurred with TBI media containing putricine. GFP fluorescence peaked at 44 h post-inoculation with 30,626 +/- 6165 RFU then dropped to a moderate level of expression at 54:00 h (8,422 +/- 1,491 RFU). Peak OD600 and RFP—both measure of fungal growth—were observed at 60:00 and 74:00, respectively. This system is the basis for our method of measuring down-regulation of *TRI5* via direct exposure to siRNAs designed against *TRI6* and *TRI10*, both transcription factors shown to induce *TRI5*.

ACKNOWLEDGEMENT AND DISCLAIMER

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TESTING TRANSGENIC SPRING WHEAT AND BARLEY LINES
FOR REACTION TO FUSARIUM HEAD BLIGHT:
2014 FIELD NURSERY REPORT

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ABSTRACT

The 2014 field screening nursery consisted of 49 wheat and 11 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 Minnesota (39 wheat lines + Bobwhite*, CB037* and Rollag*), Rutgers University (9 wheat lines + Bobwhite*) and the USDA (7 barley lines + Conlon and ND20448*). Lines with known reactions to Fusarium head blight (FHB) were also included as checks. The wheat checks included were the moderately resistant cultivars Alsen, RB07, Rollag and Sumai 3 and the susceptible cultivar Wheaton. The barley checks were the moderately resistant cultivar Quest and the susceptible cultivar Robust. Individual plots were 2.43 m long single rows. The trial was planted on June 6, 2014. All plots were inoculated twice. The first inoculation was applied at anthesis for wheat (July 16-July 29) and at head emergence (July 21-July 25) for barley. The second inoculation was applied three days after the initial inoculation (d.a.i.) for each plot. The inoculum was a composite of 39 *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 10 ml.sec⁻¹ at a working pressure of 275 kPa. Mist-irrigation was applied from the first inoculation on July 16 through August 14 to facilitate FHB development. FHB incidence and severity were assessed visually 22-27 d.a.i. for wheat and 20-23 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 hand harvested at maturity on September 9 (UMN wheat & barley) and September 16 (Rutgers wheat). Approximately sixty heads were harvested from each plot, threshed and the seed cleaned manually. The wheat grain was 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. In 2014, the disease severities were generally higher than the 2013 nursery. Mean FHB severities for the untransformed wheat checks, Bobwhite, CB037 and Rollag were 63, 34 and 22%, respectively. Mean FHB severities for the standard wheat checks, Alsen, Sumai 3 and Wheaton were 26, 18 and 83%, respectively. For barley, the untransformed check variety Conlon had a mean FHB severity of 22%. The barley standard checks, Quest and Stander had mean FHB severities of 17 and 34%, respectively. For the wheat entries in Bobwhite, CB037 and Rollag backgrounds, the

FHB severity data indicated that resistance was significantly expressed ($P < 0.05$) in some transformed lines compared to the untransformed check. Similarly the FHB severities of several barley entries appeared to be statistically better than the corresponding untransformed check. The harvested grain is currently being analyzed for DON, though the data are not yet available they will be included in the poster presented at the forum.

We would like to acknowledge Beheshteh Zargaran her assistance in preparing inoculum. We would also like to acknowledge Dr. Yanhong Dong for conducting the mycotoxin analysis.

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CHROMOSOME ENGINEERING AND NEXT GENERATION
SEQUENCING ASSISTED TRANSFER AND DEPLOYMENT OF
ALIEN GRASS SPECIES RESISTANCE TO FHB IN WHEAT

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ABSTRACT

The successful transfer of effective resistance to FHB from the perennial grass *Elymus tsukushiensis* Honda (2n=6x=42, S^{ts}S^{ts}H^{ts}H^{ts}Y^{ts}Y^{ts}, syn. *Roegneria kamoji* C. Koch) into wheat is the culmination of a research effort that began with the funding of a project by the McKnight Foundation between Nanjing Agricultural University (NAU) and Kansas State University (KSU) in 1995. In the 1990s, NAU scientists reported on the production of wheat x *E. tsukushiensis* hybrids and the disomic addition (DA) lines 1E^{ts}#1 and its derivative TWL·1E^{ts}#1S with effective resistance to FHB in greenhouse and field nurseries in China. The TWL·1E^{ts}#1S chromosome consists of the short arm of chromosome 1E^{ts}#1 joined to an unknown chromosome arm of wheat at the centromere. We crossed DATWL·1E^{ts}#1S with the *ph1ph1* (a mutant of *Ph1* gene that allows pairing between wheat and alien chromosomes) stock of wheat and identified plant progenies that were homozygous *ph1ph1* and carried one copy of TWL·1E^{ts}#1S. In these plants, we expected the 1E^{ts}#1S arm of TWL·1E^{ts}#1S to pair randomly with one of the short arms of the group-1, wheat chromosomes 1A, 1B or 1D. To monitor recombination, we designed primers from 50 ESTs mapped to group-1S arms of wheat. One proximal (BF202643/*Hae*III) and one distal (BE591682/*Hae*III) EST-STS polymorphic markers were identified. We screened 488 progenies using the EST-STS marker and identified one proximal (#74) and one distal (#107) recombinant. To further characterize the recombinant chromosomes, we designed 20 SNPs using 1AS, comparative sequence analysis, and KASParTM marker assays. Recombinant #74 carried the unidentified wheat arm from TWL·1E^{ts}#1S and proved to be agronomically undesirable. The distal recombinant #107 involved chromosome 1A of wheat, where the distal tip of 1AS was substituted by a homoeologous segment from 1E^{ts}#1S of TWL·1E^{ts}#1S and was designated as T1AL·1AS-1E^{ts}#1S. Plants where T1AL·1AS-1E^{ts}#1S substituted for chromosome 1A of wheat were fully fertile. Recombinant #107, together with susceptible (Overlay) and moderately susceptible (Everest, Chinese Spring and Karl 92) controls, was screened in greenhouse tests for FHB resistance using a single-point inoculation method. The FHB index ratings of recombinant #107 carrier progenies ranged from 4.2 to 13.3%, compared to 31.7–42.5% for noncarrier progenies from the same cross, similar to the susceptible controls. The gene symbol *Fhb6* has been assigned to designate this source of resistance. We developed two KASPar SNP markers to monitor the introgression of *Fhb6* into adapted wheats. The material has been released as KS14WGRC61 (W-ELTSU T1AL·1AS-1E^{ts}#1S (TA5655//CS *ph1B* MUT (TA3809)*2//FULLER*2 F₃) and distributed to breeders. Further prebreeding and evaluation of *Fhb6* wheat lines in field nurseries is underway. (This research was supported by a grant from the USBWSI.)

RNA-SEQ CHARACTERIZATION OF TWO BARLEY
FUSARIUM HEAD BLIGHT RESISTANT QTL

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ABSTRACT

Fusarium species cause Fusarium head blight (FHB) disease in wheat and barley. Resistance to the disease is controlled by quantitative trait loci (QTL). Previous QTL mapping studies in barley have identified two QTLs located on chromosome 2H bin8 and 6H bin7, respectively. To gain an understanding of the molecular mechanisms of FHB resistance, near isogenic line (NIL) pairs with contrasting QTL alleles for the 2H bin8 and 6H bin7 were developed and examined using RNA-sequencing. The transcriptomic changes of both NIL pairs were examined at 48 and 96 hours after *Fusarium* or mock inoculation. The host response to infection differed dramatically from 48 hours after inoculation (hai) to 96 hai. Comparative analysis of defense responses of the 2hb8 NIL pair revealed that the resistant (R) NIL exhibited broad and constitutive defense responses when compared with the susceptible (S) NIL. Cellulose synthases, UDP glycosyltransferases, cytochrome P450 enzymes, pectinesterase inhibitors, cytokinin signaling components, peptidases and lipid transfer proteins are enriched in the 2hb8 R NIL defense responses. A pair of cysteine rich receptor-like kinases were identified as promising candidate genes for the 2hb8 QTL. The 6hb7 R NIL displayed a more rapid induction of a set of defense genes than the S NIL at 48 hai and the transcript expression difference between the R and S NIL diminished at 96 hai, indicating that the R allele at the 6hb7 QTL responds more rapidly to infection. Overlap of differentially accumulated genes was identified between the two NIL pairs at 48 hai, suggesting that certain resistance mechanisms are co-regulated by the two QTL, including the DON-inactivating *HvUGT13248* gene. Long noncoding RNAs (lncRNAs) have emerged as key regulators of transcription. A total of 10,338 lncRNAs were identified from our barley spike samples, among which 486 were FHB responsive. Examples of co-induction of lncRNAs and their neighboring transcripts were identified. The current transcriptomic analysis of two barley FHB QTL NIL pairs revealed the dynamics of host response to *Fusarium* infection and identified genes and lncRNAs that are associated with FHB resistance.

FUSARIUM CONTROL BY HOST-INDUCED GENE SILENCING

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ABSTRACT

Fusarium Head Blight (FHB) and Seedling Blight (FSB), which is caused by mycotoxin-producing fungi of the genus *Fusarium*, is an economically important crop disease. We assessed the potential of host-induced gene silencing (HIGS) targeting the three fungal *cytochrome P450 lanosterol C-14 α -demethylase* (*CYP51*) genes, which are essential for ergosterol biosynthesis, to restrict fungal infection. *In vitro* feeding of *CYP3RNA*, a 791 nucleotides (nt) dsRNA complementary to all three paralogs *CYP51A*, *CYP51B*, and *CYP51C*, resulted in growth inhibition (half maximum growth inhibition [IC₅₀] = 0.9 nM) as well as altered fungal morphology, similar to that observed after treatment with the azole fungicide tebuconazole, for which *CYP51* is a target. This inhibition of fungal growth correlated with *in fungus* production of siRNAs corresponding to the targeted *CYP51* sequences. Expression of the same dsRNA in *Arabidopsis* and barley rendered susceptible plants highly resistant to fungal infection. Microscopic analysis revealed that mycelium formation on *CYP3RNA*-expressing leaves was restricted to the inoculation sites, and that inoculated barley caryopses were virtually free of fungal hyphae. This inhibition of fungal growth correlated with *in planta* production of siRNAs corresponding to the targeted *CYP51* sequences, as well as highly efficient silencing of the fungal *CYP51* genes. The high efficiency of fungal inhibition suggests that HIGS targeting of the *CYP51* genes is an alternative to chemical treatments for the control of devastating fungal diseases such as FHB and FSB.

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

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ABSTRACT

Fusarium head blight (FHB) is an old yet unsolved problem of cereal crops, mainly caused by the fungal pathogen *Fusarium graminearum*. During infection, trichothecenes produced by *Fusarium* increase fungal virulence and decrease grain quality. Previous work identified a barley UDP-glucosyltransferase gene (*HvUGT13248*) that detoxifies deoxynivalenol (DON) by the conversion to DON-3-O-glucoside (D3G) in transgenic yeast and *Arabidopsis*. Here we report successful development of transgenic wheat and barley overexpressing *HvUGT13248* gene. The transgenic wheat show high levels of FHB type II resistance in the greenhouse point inoculation tests. The FHB severity of the transgenic lines were reduced by up to 91% compared to untransformed lines. We also tested these transgenic wheat in inoculated (spray inoculated with macroconidia) and mist-irrigated field experiments in three consecutive years, and they also show high levels of FHB resistance. Moreover, transgenic wheat carrying *HvUGT13248* converted DON to D3G more rapidly than untransformed plants, and there was also reduced DON accumulation in the grains of the transgenic wheat harvested from the field tests. To screen wheat and barley resistance to trichothecenes, we developed a fast and convenient method by monitoring root growth of seedlings on trichothecene-containing growth media. We used this root assay to show that transgenic barley overexpressing *HvUGT13248* exhibit resistance to DON. We also introduced the *HvUGT13248* transgene into the elite wheat cultivar Rollag, and the backcross-derived lines exhibited high levels of FHB resistance in the greenhouse and field tests, however, the FHB severity levels were only slightly reduced from Rollag. This result suggests that the FHB resistance mechanisms provided by the *Fhb1* QTL and the *HvUGT13248* transgene may overlap.

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TWO NOVEL NON-SPECIFIC LIPID TRANSFER PROTEINS
PROVIDE ENHANCED RESISTANCE TO A TRICHOHECENE
MYCOTOXIN BY REDUCING OXIDATIVE STRESS

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ABSTRACT

Fusarium head blight (FHB) caused by *Fusarium graminearum* is one of the most important cereal diseases worldwide. Trichothecene mycotoxins, which are produced during infection accumulate in the grain, posing a significant health threat to humans and animals. To identify genes that improve resistance to trichothecenes, we screened an activation tagged *Arabidopsis* population against trichothecin (Tcin), a type B trichothecene in the same class as deoxynivalenol (DON). One of the resistant lines identified contained an activation tag upstream of two nonspecific lipid transfer protein (nsLTP) genes, *AtLTP4.4* and *AtLTP4.5*. Expression of both nsLTP genes were induced in the mutant over 10-fold relative to wild type. Overexpression of either nsLTP gene conferred resistance to Tcin in *Arabidopsis* and in *Saccharomyces cerevisiae*. In both systems *AtLTP4.4* provided greater resistance than *AtLTP4.5* relative to wild type and vector transformed lines. In *Arabidopsis* and yeast, Tcin treatment increased reactive oxygen species (ROS) accumulation and overexpression of *AtLTP4.4* attenuated ROS levels relative to the controls. Exogenous addition of GSH and other antioxidants enhanced resistance to Tcin while the addition of buthionine sulfoximine (BSO), an inhibitor of GSH synthesis, increased sensitivity to the toxin, indicating that oxidative stress contributes to trichothecene sensitivity. To further evaluate ROS induction in plants by trichothecenes, confocal microscopy was performed using *Arabidopsis* and tobacco leaves infused with trichothecenes and ROS was detected by staining with 2',7'-dichlorofluorescein diacetate (H2DCF-DA), which is converted to the highly fluorescent dichlorofluorescein (DCF) when oxidized by ROS. DON and Tcin treatments revealed DCF stain that colocalized with chloroplasts, the cell wall region, and possibly the apoplast. Increasing the dosages of Tcin and DON intensified the DCF staining of chloroplasts. Overnight treatments with a high dose of DON (240 µM) released chlorophyll into the cytoplasm as observed after treatment with a low concentration of paraquat. These results demonstrate that trichothecenes target chloroplasts and induce ROS and that overexpression of a specific *Arabidopsis* nsLTP protects against trichothecene-induced oxidative stress possibly by increasing the antioxidant defense.

DEVELOPING TRANSGENIC WHEAT AND BARLEY THAT EXHIBIT
RESISTANCE TO *FUSARIUM GRAMINEARUM* VIA GLUCOSIDE
CONJUGATION OF TRICHOHECENE MYCOTOXINS

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ABSTRACT

Fusarium graminearum infection of wheat and barley results in production of trichothecene mycotoxins including deoxynivalenol (DON) and nivalenol (NIV). These mycotoxins result in increased fungal virulence and reduce grain quality. Numerous transcriptomic studies have been conducted by our lab on the wheat/barley – *F. graminearum* interaction. These studies have identified a set of genes that may provide resistance to *F. graminearum* infection via conjugation, degradation or transport of trichothecenes. In addition, these studies also provide an understanding of *F. graminearum* genes that are expressed during infection. For example, the *F. graminearum* transcriptome responds differently to wheat carrying either resistant or susceptible alleles for *Fhb1*. From these studies we identified a barley UDP-glucosyltransferase (*UGT13248*) that exhibited DON resistance in yeast. Transgenic wheat expressing *UGT13248* exhibited a high level of type II resistance in the greenhouse and resistance in the field that approaches the level of resistance conferred by Sumai 3. The mechanism of resistance conferred by *UGT13248* is via conjugation of DON to DON-3-O-glucoside. Backcross families carrying *Fhb1* (type II resistance) derived from Rollag and the *UGT13248* transgene were screened in the greenhouse and field. The level of resistance in plants carrying *Fhb1* alone and those carrying *Fhb1* and the *UGT13248* transgene were similar with a few *Fhb1/UGT13248* containing lines exhibiting a slight reduction in severity compared to those carrying *Fhb1* alone. The lack of reduction in disease severity may be due to either (1) *Fhb1* and the *UGT13248* acting in the same manner or (2) the level of resistance conferred by *Fhb1* is so high that it is difficult to obtain increased resistance. It is noteworthy that transgenic wheat carrying *UGT13248* also exhibits type II resistance to 3-ADON- and NIV-producing strains of *F. graminearum*, indicating that *UGT13248* acts on a wide range of trichothecene mycotoxins. Interestingly, overexpression of *UGT13248* in barley resulted in resistance to DON in root assays. Overall, our results demonstrate that *UGT13248* is an effective gene for conferring resistance to *F. graminearum* infection.

EQUAL GENOMIC AND PHENOTYPIC SELECTION GAIN FOR FHB RESISTANCE AND DON ACCUMULATION IN BARLEY

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ABSTRACT

Genomic selection (GS) is a marker based selection method that promises to improve and accelerate the breeding process in plants and animals. Numerous studies have investigated the gain per unit time; however few have compared the gain from GS and phenotypic selection (PS) using empirical data. In this study, we used Fusarium head blight (FHB) severity and deoxynivalenol (DON) data from five consecutive years of selection (2006 – 2010) to compare the gain between GS and PS. In each year, about ninety six barley breeding lines were phenotypically evaluated in FHB and DON trials and that data was used to conduct PS. A set 168 parental lines, that were genotyped with 1,536 SNP markers and phenotyped for FHB and DON, were used as a training population to predict the performance of the breeding lines in each of the five years using RR-BLUP. We selected best (top 10%) breeding lines in each year using PS and GS. These lines were re-evaluated together in 4 trials in Minnesota and North Dakota to compare the gain from selection using the two selection schemes. In general, the gain from GS and PS were similar across the 5 years indicating that GS could maintain similar gains per cycle of selection, but at a reduced cost and shorter cycle time.

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ENGINEERING RESISTANCE AGAINST *FUSARIUM GRAMINEARUM*

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ABSTRACT

Fusarium graminearum is the principal causative agent of Fusarium head blight (FHB), a destructive disease of wheat and barley. Under laboratory conditions, *F. graminearum* can also cause disease on leaves and flowers of *Arabidopsis thaliana*, a model plant for molecular-genetic studies. Our results have shown that pathogen-associated molecular patterns (PAMPs), can stimulate PAMP-triggered immunity (PTI), which can enhance resistance against *F. graminearum* in *Arabidopsis*. Furthermore, application of a bacterial PAMP was capable of enhancing FHB resistance in wheat, thus suggesting that the PTI mechanism can potentially be targeted for enhancing resistance against *F. graminearum* in wheat. We have engineered one of these defense elicitors for expression in *Arabidopsis* and find that these plants exhibit elevated resistance to leaf and floral inoculation with *F. graminearum*. Transgenic wheat plants expressing the defense elicitor have been generated and are being evaluated for resistance to FHB. In addition, we have engineered wheat to constitutively express a transcription factor involved in the activation of PTI. Results obtained with these plants will be presented, in addition to other strategies for enhancing FHB resistance that are underway in our lab.

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**PATHOGEN
BIOLOGY
AND
GENETICS**

PRESENCE OF *FUSARIUM GRAMINEARUM* IN AIR ASSOCIATED
WITH SORGHUM FIELDS

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ABSTRACT

Sorghum can be included in crop rotations with wheat. However, there are no known reports on the effects of sorghum grown in rotation with wheat on the epidemiology of head scab caused by *Fusarium graminearum*. Conidia in air samples within two sorghum fields were collected by passive spore trapping for two years at four plant stages (vegetative, anthesis, grain development or maturity) during the sorghum growing season. Spores were collected and germinated on a general fungal medium (potato dextrose agar; PDA) and a medium semi-selective for *Fusarium* spp. (pentachloronitrobenzene-containing agar; PCNB). Colonies cm⁻² hr⁻¹ on PCNB ranged from 31.0 to 85.7 percent of colonies cm⁻² hr⁻¹ on PDA, depending on environment and growth stage. A subsample of *Fusarium* isolates from PCNB traps were identified molecularly by comparing sequences from a portion of the translation elongation factor (*TEF*) 1 α gene with those in the FUSARIUM-ID database (<http://isolate.fusariumdb.org/index.php>). Surprisingly, 26.8% were *F. graminearum*, the most numerous *Fusarium* species or genotype. Phylogenetic analyses of these isolates, as well as *F. graminearum* from sorghum leaf tissue and grain, using *TEF*, the rRNA internal transcribed spacer region and a portion of the histone-3 gene (*H3*), revealed that these isolates were highly similar to one another and to previously characterized *F. graminearum* isolates. Further research to determine whether isolates associated with sorghum production produce tricothecenes or zearalenone and are pathogenic to wheat, will need to be conducted to determine whether *F. graminearum* associated with sorghum production can affect head scab levels in wheat.

RESISTANCE MECHANISMS AND MANAGEMENT OF *GIBBERELLA ZEA* TO BENZIMIDAZOLE FUNGICIDE CARBENDAZIM AND A NOVEL FUNGICIDE PHENAMACRIL (JS399-19)

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ABSTRACT

In China, *Gibberella zea* is the dominant pathogen causing wheat head blight. Carbendazim (MBC) has been widely used to control this disease since the 1970s. However, the resistance to MBC is currently serious in *G. zea*. Though the MBC-resistance was controlled by one major gene and is involved in mitotic division, no mutation in the target β -tubulin was found, which was different from other filamentous fungi. To identify the MBC-resistance mechanism of *G. zea*, other tubulin genes were analyzed. Alterations at amino-acid codon 17 or 167 or 198 or 200 in β 2-tubulin were found to correspond to the different phenotypes of MBC-sensitivities. Deletion/complementation of the β 2-tubulin gene as well as mononucleotide displacement and affinity of MBC binding tubulins validated the point mutations conferring resistance of *G. zea* to MBC. It is interested to find that MBC-resistance mutation leads to increased expression of deoxynivalenol (DON) biosynthesis genes. Compared to the wild-type MBC-sensitive strain, the resistant strain produced twice as much DON in infected grains. Novel chemical, phenamacril (development code no. JS399-19) 2-cyano-3-amino-3-phenylacrylic acetate is recommended as a *Fusarium* specific fungicide to control MBC-resistance *Fusarium* head blight.

Phenamacril is a novel cyanoacrylate fungicide discovered and patented by the Jiangsu Branch of the National Pesticide Research & Development South Center of China. The fungicide exhibited specific activity against fungal plant pathogens of the genus *Fusarium* with which it strongly interferes with mycelial growth and it has an excellent control effect on *Fusarium* head blight. We have monitored phenamacril-resistance population in the field for 3 years and do not find any resistant mutants. In the lab, phenamacril-resistant mutants were obtained. Through the genome sequence of the sensitive strain and resistant mutants, and homologous double exchange between the gene locus of the sensitive strain and the resistant mutant, we found that the point mutations in the gene myosin-5 (at codon 216, 217, 418, 420, or 786) confer resistance to phenamacril.

FUSARIUM GRAMINEARUM INTERACTION WITH
THE EPIDERMIS OF THE BARLEY PALEA
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ABSTRACT

The first step in disease initiation is the penetration by the pathogen of the host surface. Preventing entry of host targets by *Fusarium graminearum* is an intriguing goal that requires better understanding of the mechanisms of *F. graminearum* ingress of host tissues. We are investigating the role of the various epidermal cell types in the interaction between host and pathogen. The epidermis of the barley palea features several cell types including multiple phytolith (trichomes and cells of similar origin) morphotypes and stomata. We have used a histological approach to demonstrate that the fungus preferentially interacts with particular cell types on the palea of excised barley florets. In particular, one phytolith morphotype common on two-row barley appears to be a common point of contact between the fungus and the host. Elucidation of the initial targets of infection will aid future strategies for disease control.

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CO-PRODUCTION OF 3ADON AND 15ADON BY CULTURES OF
FUSARIUM GRAMINEARUM 15ADON STRAINS, BUT NOT 3ADON
STRAINS, IS DUE TO DIFFERENCES IN ACETYLTRANSFERASE
ACTIVITY AND SUBSTRATE SPECIFICITY

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ABSTRACT

Fusarium graminearum strains can be assigned to chemotypes, e.g. 3ADON or 15ADON, on the basis of PCR analysis using polymorphisms in the trichothecene biosynthetic genes *TRI3* and *TRI12*. Trichothecene production in liquid culture is consistent with the chemotype predicted with PCR analyses, i.e., acetylated DON (3ADON or 15ADON) is produced but no DON is detected. In contrast, grain infected with 3ADON or 15ADON strains are predominantly contaminated with DON with small amounts of 3ADON or 15ADON, or sometimes both, detected. A mixture of acetylated deoxynivalenols is also sometimes found in rice cultures. Although *F. graminearum* 70E1, a 3ADON strain, produced DON and 3ADON, *F. graminearum* GZ3639, a 15ADON strain, produced a mixture of DON, 15ADON, 3ADON and 3,15-diADON. We have previously shown that differences in the *TRI8* esterase gene determine the 3ADON or 15ADON chemotype. Disruption of *Tri8* in either 3ADON or 15ADON strains results in the accumulation of 3,15-diADON, the common precursor of 3ADON and 15ADON. Yeast expressing *TRI8* from a 3ADON strain removed the C-15 acetyl group from 3,15-diADON while yeast expressing *TRI8* from a 15ADON strain removed the C-3 acetyl group from 3,15-diADON. In order to determine if differences in trichothecene acetyltransferase genes might contribute to the production of both acetylated forms in the cultures of one chemotype, *TRI3* and *TRI101* from 3ADON and 15ADON chemotypes were expressed in yeast. In trichothecene biosynthesis, *Tri101* acetylates at C-3, converting isotrichodermin into isotrichodermin, and *Tri3* acetylates at C-15, converting 15-decalonectrin into calonectrin. Feeding experiments with yeast expressing *Tri101* from a 3ADON or a 15ADON strain indicated that *Tri101* can convert DON into 3ADON. Feeding experiments with yeast expressing *Tri3* indicated that *Tri3* from 3ADON or 15ADON strains were functional, i.e. able to convert 15-decalonectrin into calonectrin. *Tri3* from a 15ADON strain was also able to convert DON into 15ADON but *Tri3* from a 3ADON strain did not convert DON into 15ADON. These differences in *Tri3* activity and substrate specificity can account for both 3ADON and 15ADON being produced in a 15ADON strain but not a 3ADON strain.

THE NIVALENOL-PRODUCING *FUSARIUM GRAMINEARUM*
GENOTYPE IN SCABBY NORTH CAROLINA WHEAT SPIKES

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ABSTRACT

Fusarium head blight (or scab), which in the U.S. is caused primarily by *F. graminearum*, leads to drastic decreases in yield and test weight of small grains. In addition, *Fusarium* mycotoxins in grain heads can render the crop unsuitable for human or animal consumption. In livestock, scabby grain can lead to feed refusal and/or poor weight gain. Although this fungus produces various mycotoxins, the most important ones in small grains are deoxynivalenol (DON) and nivalenol (NIV). Both can cause severe toxicoses in humans and livestock; compared to DON, NIV has greater mammalian toxicity. While DON is the dominant wheat scab toxin in most of the U.S., a high proportion of *Fusarium* isolates from southern Louisiana wheat had been found by other researchers to be NIV producers. Although a 2006 wheat survey detected about 10% NIV producers in each of two NC counties, the distribution of NIV strains across the state was unknown. DON contamination is often measured in North Carolina grain crops, but NIV is not.

In this study, we sampled commercial wheat heads symptomatic for scab from 60 fields in 24 NC counties in the 2013-14 growing season. From each infected head, a single *Fusarium* strain was isolated and, using polymerase chain reaction (PCR), categorized as a 3-ADON, 15-ADON, or NIV genotype. Partial results showed that, of the 776 isolates that amplified successfully, 96% were 15-ADON. NIV types were 3% of the total sample, and up to 8% of strains from a single field; they did not seem to be concentrated in any county or region of the state. As in the 2006 survey, the 3-ADON genotype was found in several counties at a very low frequency (<1% of the total sample), raising the question of whether it will increase in frequency in North Carolina as it has in the northern U.S. and Canada, or alternatively if selective forces are keeping it rare. In practical terms, by assessing the distribution of NIV-producing *Fusarium* strains in North Carolina wheat fields, we will determine whether and where NIV may warrant monitoring in severe scab years.

GENOTYPING BY SEQUENCING FOR FOOTPRINTS
OF SELECTION IN *FUSARIUM GRAMINEARUM*
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ABSTRACT

Previous population genetic studies of *Fusarium graminearum* have found evidence for frequent outcrossing as well as genetic clustering into populations associated with genotypes at the trichothecene gene cluster. Notably, several studies have reported the increase in frequency of isolates with the 3ADON chemotype from Fusarium head blight (FHB) infected wheat, though the specific cause of this shift is still uncertain. We argue that new population genomic studies will provide an important complement to experimental studies that investigate functional differences between populations. The sequencing of the *F. graminearum* genome has revealed how the genome is functionally organized and which regions are most dense with polymorphisms. However, there is an urgent need to use sequence-based markers on a genome-wide scale to describe patterns of variation along chromosomes and among different geographical regions. This information can lead to the identification of the genetic basis of functional differences between populations that can affect pathogen management and strategies for developing host plant resistance.

Here, we provide preliminary results from our FY14 USWBSI project that uses genotyping by sequencing (GBS) markers for the population genomic analysis of isolates from multiple regions in the Americas. Though our sample is still expanding, we have revisited population structure with nearly 300 isolates and asked how it relates to isolate collection location and trichothecene genotype. Our GBS markers let us investigate how patterns of genetic differentiation between populations vary across the genome, identifying candidates for local adaptation. To determine whether genome-wide association studies are feasible, we have characterized the decay of linkage disequilibrium with distance along chromosomes and how this varies by genome location. Finally, we are scanning the genome to determine whether footprints of selection support the hypothesis that natural selection acting directly on genetic changes at the FHB-related trichothecene loci has caused the 3ADON population shift. By investigating the cause of population shifts and their relationship to mycotoxin chemotypes, we may identify novel genes critical for fungal fitness against which we can develop strategies for toxin reduction and FHB control.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-1-113. 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.

SPREAD AND COLONIZATION OF *FUSARIUM GRAMINEARUM*
DURING INFECTION IN A RESISTANT WHEAT
CULTIVAR CARRYING *FHBI* RESISTANCE

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ABSTRACT

The spread and colonization of *F. graminearum* in rachis of a resistant wheat cultivar carrying *FHBI* resistance was examined by fluorescence microscopy using a fungal strain constitutively expressing GFP. To preserve GFP fluorescence, a simple cryohistological process was employed. At 6 dpi in the upper portion of the rachis segment, immediately adjacent to the inoculation point, fungal hyphae were visible predominantly in the parenchyma and, to a lesser extent, in the vascular bundles. In the parenchyma, hyphae were observed inside the cells and in the apoplast. Fungal hyphae that appear in apoplast tend to surround cells that appeared to be healthy, with intact chloroplasts. In parenchymatous cells that had intracellular hyphae, chloroplasts were damaged. Histochemical investigation identified massive depositions of condensed catechol-type tannins, visible starting at 3 dpi, and the presence of pectin associated with the fungal hyphae in the upper portion of the rachis segment. Accumulation of phenolic compounds and pectin were detected within and along the vascular bundles, within the parenchyma cells, and in the apoplast. Our findings suggest that phenolic compounds represent a part of the response of the plant to fungal infection. Phenolic compounds and pectin may restrict fungal spread in the apoplast thus playing a role in controlling the spread of the infection. Investigation of the genes involved in this plant resistance response is in progress.

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MODIFICATION OF THE MYCOTOXIN DEOXYNIVALENOL WITH ENZYMES AND MICROORGANISMS

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ABSTRACT

New strategies are needed to mitigate deoxynivalenol (DON) contamination in wheat and barley. This research is aimed at discovering novel enzymes and microorganisms to degrade DON to improve food safety and decrease economic loss producers face. We have first developed a tool to screen for potential candidate DON enzymes by engineering several yeast strains to be sensitive to DON. One of the yeast strains is sensitive to DON at 100 ppm, but not sensitive to de-epoxy DON (the detoxified product) at the same concentration. Second, enzyme candidates to transform DON were selected using the BRENDA database, an enzyme repository, based on their functionality; promising epoxide hydrolases and cycloisomerases have been identified. Third, we bioprospected for DON detoxifying microorganisms from the environment and cultured the samples in the presence of 100 ppm DON. Three mixed cultures and one pure culture consistently detoxify DON in laboratory experiments; the organisms responsible for DON detoxification are in the process of being characterized. Organisms and genes that demonstrate DON detoxification will be tested in contaminated wheat and barley samples in future studies. This research will offer new strategies for detoxifying DON in wheat and barley.

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**VARIETY
DEVELOPMENT
AND
HOST PLANT
RESISTANCE**

GENOMIC SELECTION FOR FUSARIUM HEAD BLIGHT
RESISTANCE IN A SOFT RED WINTER WHEAT (*TRITICUM
AESTIVUM* L.) BREEDING PROGRAM

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ABSTRACT

Genomic Selection (GS) is breeding strategy aiming at selecting superior individuals based on their genomic estimated breeding values (GEBVs). The strategy is particularly promising for quantitative traits and requires dense, genome-wide marker data. Fusarium head blight (FHB), primarily caused by *Fusarium graminearum* in the US, has been shown to be quantitatively inherited. The aim of this work is to assess the effect of the training population size, level of genotypic missing data, imputation methods, and statistical models on GS accuracy. A total of 273 lines from the University of Illinois soft red winter wheat program, and from breeding programs around the Midwest and Eastern US were included in this study. Genotypic data were obtained using the Genotyping-by-Sequencing (GBS) protocol. Libraries were constructed using three two-enzyme combinations, where a rarer cutter (*PstI*) and three common cutters (*MspI*, *HinPI*, and *BfaI*) were combined. A different set of barcodes were used for each enzyme combination. Sequence data were obtained from 96-plex Illumina HiSeq2000 runs, and then analyzed with the UNEAK pipeline. Two data sets were obtained according to their maximum proportion of missing data per marker: 20%, and 50%. After applying the Fisher's exact test, the number of SNPs called for each data set was 5K and 16K. Four imputation methods were tested: mean imputation (MNI), singular value decomposition (SDVI), random forest regression (RFI), and expectation maximization (EMI). We also tested different sizes of the training population (96, 144, 192, and 218), as well as different proportions of the training population in relation to the validation population (0.5, 0.6, 0.7, 0.8, and 0.9). The phenotypic data were collected in a field nursery in Urbana, IL, in 2011, 2013, and 2014. Best unbiased linear predictors (BLUPs) were calculated for FHB severity, incidence, deoxynivalenol (DON) concentration, *Fusarium*-damaged kernels, the ISK and FHB indexes. Accuracy significantly increased for all traits when the largest training population (218) was used. Using eighty percent of individuals as the training set resulted in the best combination of mean accuracy and variance. No statistically significant differences were detected for accuracy when different genotypic data sets (5K and 16K SNPs) were compared. The imputation methods performed equally well, with a numerical advantage for EMI. Also, MNI and EMI were the least computationally intensive. For all traits except incidence, rr-BLUP outperformed LASSO and ELASTIC-NET. The highest five-fold cross-validated accuracies were recorded for incidence, ranging from 0.68 to 0.81, depending on the GS model. The lowest values were obtained for severity, ranging from 0.45 to 0.48. Other traits showed intermediate values. In conclusion, this study shows that measurements associated with FHB resistance can be predicted with GS models with moderate accuracy, even without a reference genome.

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USING MARKER-ASSISTED SELECTION TO IMPROVE HARD WINTER WHEAT FHB RESISTANCE

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ABSTRACT

Epidemics of wheat Fusarium head blight (FHB), incited by *Fusarium graminearum*, are more frequent and severe in hard winter wheat (HWW, *Triticum aestivum* L.) growing region due to reduced tillage and the continued expansion of corn (*Zea mays* L.) into the region. Thus, improving FHB resistance becomes a major breeding objective in most HWW breeding programs. Although quantitative trait loci (QTL) for FHB resistance have been reported from different sources including some from HWW in the Great Plains, some resistant sources from China show the best resistance. To identify and validate QTL from Chinese landraces, we construct a consensus map of five mapping populations with Chinese landraces as resistant parents using genotyping-by-sequencing (GBS) generated single nucleotide polymorphism (SNP) markers. The consensus map was used for QTL meta-analysis to identify SNPs tightly linked to overlapping QTL across populations. Among QTL identified, *Fhb1* is the QTL with the largest effect across the populations. By screening recombinants in *Fhb1* region using a large segregation population derived from Ning7840/Clark through marker-assisted backcross, a small fragment co-segregating with *Fhb1* were identified. Markers from the region were developed for marker-assisted selection. Because *Fhb1* is not present in HWW cultivars in the Great Plains, we developed marker-assisted backcross project to transfer *Fhb1* to US adapted HWW backgrounds. The lines with *Fhb1* in different US winter wheat backgrounds showing a high level of type II FHB resistance were selected. To date, *Fhb1* has been transferred to 17 adapted HWW cultivars. Some of the *Fhb1* lines have been used as resistant parents in different breeding programs, and others are in double haploid production and seed increasing stage and will be distributed to breeding programs for further yield testing.

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PHENOTYPIC ANALYSIS OF FHB RESISTANCE IN A SOFT
WHEAT POPULATION FOR GENOME-WIDE ANALYSES

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ABSTRACT

Breeding for Fusarium Head Blight (FHB) resistance is vital to controlling this disease. In this study, a set of 649 soft winter wheat lines, termed the FHBGS population, were genotyped by GBS and phenotyped over three years in multiple uniform scab nurseries adapted to the Corn Belt (OH, NY, KY, MI, MO and IL). Traits assessed included incidence (INC), severity (SEV), index (IND, *Fusarium*-damaged kernel (FDK), INC+SEV+FDK (ISK), and concentration of deoxynivalenol (DON). Within each location the data was standardized for the mean and standard deviation of a set of 49 checks. Our results showed high heritability (0.88 to 0.94) for all traits. Best linear unbiased predictors (BLUPs) obtained from each trait were highly correlated to one another. Principal component (PC) analysis among all traits revealed high percentage of variance explained by the first PC (~81%). The most superior 5% of individuals performed better than the resistant check (Truman) for all traits except DON. Cluster analysis of marker data among all individuals showed clear differentiation of lines from New York from all others. The remaining lines were placed in two less differentiated clusters: one of these groups had a high proportion of parentage from Truman. On average the NY cluster was the most susceptible and the Truman cluster the most resistant. The results from this analysis will constitute the basis for subsequent association analysis and genomic selection study.

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PRELIMINARY ANALYSIS OF GENOMIC
SELECTION FOR FHB RESISTANCE

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ABSTRACT

Resistance to FHB in soft winter wheat appears to be polygenic and phenotypic selection is slow and relatively inefficient. Genomic selection may be a useful alternative selection tool. A population of 649 soft winter wheat lines, termed the FHBGS population, was phenotyped for multiple FHB traits over multiple environments and genotyped with 4,643 GBS markers. FHB index (IND) was also evaluated in a population of 273 lines, termed the TCAP population, and genotyped with 3919 markers from the 90K SNP chip. The prediction accuracy of GS was assessed using cross-validation approach with ridge regression best linear unbiased prediction (RR-BLUP) model. In the FHBGS population GS accuracy across all individuals ranged from 0.43 to 0.55 (for SEV and FDK, respectively), while accuracy for IND in the TCAP population was 0.62. In both populations, genetic relatedness affected the accuracy of prediction. Higher accuracies were observed when individuals in training and predicting sets belonged to the same cluster: GS models developed in one cluster generally did not predict the observed phenotypes of the other clusters. The findings are directly applicable for breeders to implement GS schemes to improve FHB resistance in the Northern U.S. soft winter wheat breeding programs.

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

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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 human and animal. FHB resistance has been reported from many sources, especially Chinese landraces, such as Sumai3, Wangshuibai. Among them, quantitative trait loci (QTLs) for FHB resistance in Sumai3 have been well characterized in multiple studies, however, QTLs 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). QTLs have been identified in each population using low-density maps constructed with a set of simple sequence repeats (SSR) and sequence tagged site (STS) markers. However, low density maps may not be able to cover all QTL regions in those populations and some QTLs may be missed. Genotyping-by-sequencing (GBS) is a novel approach that provides a rapid and robust tool for discovery of high-density SNPs for QTL mapping. In the current study, we analyzed the five populations with GBS SNP, developed high-density maps for the five populations and constructed a consensus map for meta-analysis of the QTLs. Using the new maps, 21 QTLs were remapped on 9 chromosomes (1AS, 3A, 5AS, 7AL, 3BS, 6BS, 2D, 3DL, 7DL) after adding GBS-SNP to original SSR maps, among which 10 QTLs are new QTL identified on 7 chromosomes (6A, 2B, 4B, 1D, 4D, 5D, 6D). QTLs were then projected onto the consensus maps by referring the original QTL confidence intervals (CIs) and QTL contributions (R^2). The FHB-resistance QTLs with the 95% CIs were shortened by using a clustering approach based on Gaussian mixture model in MetaQTL V1.0. Consistent QTLs among two or more populations were identified and tightly linked markers for these QTLs were identified. Thus, meta-analysis using GBS-SNP maps facilitate the validation of QTLs and identification of closely linked markers for marker-assisted selection.

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COMPARING OPTICAL SORTING, AIR SEPARATION
AND DIGITAL IMAGE ANALYSIS ESTIMATIONS
OF WHEAT *FUSARIUM* DAMAGED KERNELS

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ABSTRACT

One aspect of Fusarium head blight (FHB) of wheat (*Triticum aestivum* L.), caused by *Fusarium graminearum* Schwabe [teleomorph *Gibberella zeae* (Schwein.:Fr.) Petch], is grain mummification. The proportion of the characteristic resultant “tombstones”, pale, shriveled grains, is quantified as percent *Fusarium*-damaged kernels (FDK). FDK is one of the most valuable measurements in the development of solutions to FHB. For instance, breeding programs can make thousands of FDK measurements each year. Because of the labor required, FDK is typically estimated visually rather than by hand separation and counting, the gold standard. We compare alternative methods for their accuracy and efficiency. Two methods are based on separating the *Fusarium*-damaged kernels, by optical sorting or air separation. FDK is then expressed as the proportion of the mass of the sample. We compare the results from these methods with a third method where estimates were obtained using an ImageJ program on digital images of grain samples.

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MAPPING OF QUANTITATIVE TRAIT LOCI FOR FHB
AND DON RESISTANCE IN A DOUBLED HAPLOID
POPULATION OF EVEREST X ART

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ABSTRACT

The current project is aimed at mapping and identifying quantitative trait loci for resistance in wheat to spread of Fusarium head blight (FHB) symptoms (type II resistance) and spread and accumulation of deoxynivalenol toxin (DON) (type III resistance) throughout the spike. The mapping population consists of 148 doubled haploid lines from an Everest x Art cross. The disease study was performed using a point inoculation technique in a greenhouse with a 14 hour 30°C day and 10 hour 18.9° C night. Inocula of *F. graminearum* consisted of a conidial suspension using field isolate GZ 3639 native to Kansas. The conidial suspension was adjusted to 100 µL⁻¹ for inoculations. Pots were arranged in a completely randomized design on four greenhouse benches. Twenty to forty heads were randomly selected from each line and inoculated after the heads emerged from the leaf sheath. Selected spikes were injected with 10 µL⁻¹ of conidial suspension, via a micropipette; into a central spikelet (approximately the tenth fully developed spikelet from the base). Spikes were rated for percent infection after 14 days. Kernels were harvested separately from each spikelet in order to determine how DON moves throughout the spike. DON spread and accumulation were measured using a single kernel near-infrared spectroscopy (SKINR) instrument. FHB resistance was significantly correlated with DON resistance ($r = 0.82$, $p < 0.001$). Genotyping-by-sequencing (GBS) was used to discover and genotype SNP markers. TASSEL software was then used to call and filter markers resulting in 7,311 SNPs that were bi-allelic between the parents. A linkage map was created with MSTMap® software using a LOD threshold of 8 and a no mapping distance threshold of 15 cM. The final linkage map consisted of 2,211 markers covering 31 linkage groups (LG) with an average of 71 markers per LG, average LG length of 93.6 cM, and average marker spacing of 1.3 cM. Standard interval mapping and composite interval mapping were performed separately for FHB and DON via the QTL package in RStudio v0.98.1080®. Four QTL for type II resistance were found on LGs 1, 2, 10, and 17. The QTL on LGs 1 and 2 were associated with alleles from Everest and explained 13.4% and 10.0% of the additive phenotypic variance, respectively. The QTL on LG 10 was associated with alleles from Art and explained 21.3% additive phenotypic variation. There were two LOD peaks on LG 17 at 52 cM using SIM (LOD = 3.12, $p = 0.0501$) and 79 cM using CIM (LOD = 3.36, $p = 0.032$). The more significant peak obtained from CIM was used as the location of the QTL and explained 11% of the additive phenotypic variation. The narrow-sense heritability estimate for type II resistance was slightly low at 0.13. There were no significant QTL observed for type III resistance to spread of DON toxin. This experiment is being repeated and the results will be useful in determining if the sources of type II and type III resistance in the cultivars Art and Everest evolved independent of each other. This will allow for the potential to combine QTL for the release of germplasm with high resistance.

‘PARSHALL’: AN INDIGENOUS AND NOVEL FHB RESISTANCE SOURCE FOR FUSARIUM HEAD BLIGHT WITH HIGH QUALITY AND ADAPTED HARD RED SPRING WHEAT CULTIVAR

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ABSTRACT

Fusarium head blight (FHB) is a devastating disease affecting wheat growing regions worldwide. Economically, FHB epidemics have resulted in losses of hundreds of millions in the US alone since the 1990s. Developing wheat cultivars with FHB resistance that meet the producers and processors needs is critical. ‘Parshall’ is a NDSU released cultivar with good FHB resistance. However, the genetics underlying Parshall’s FHB resistance have yet to be characterized. A RIL population was generated from the cross Parshall × ‘Reeder’ (PR) and tested in several locations across three states (ND, MN, and SD). Five FHB traits {severity, incidence, disease index, level of deoxynivalenol (DON), and *Fusarium* damaged kernels (FDK)} and one agronomic trait (heading date) were evaluated in field and greenhouse experiments over three years (2010-2012). The PR population was genotyped using DArT and SNP markers. A genetic map consisting of 504 markers was used for composite interval mapping to identify corresponding FHB QTL traits. In total, 81 (genome A=41; B=38 and D=2) QTL were identified on 15 different chromosomes, across locations and years. In total five QTL for resistance type I, 17 for type II, 13 for type III, 11 for type IV, 12 for FHB-NDX and 23 for heading dates (HD) were identified. Among these, 3, 8, 2, 3, 5, and 13 were identified as stable QTL for resistances type I, II, III, and IV; and NDX and HD, respectively. Similarly, the number of major QTL detected for resistance type I, II, III, IV, NDX, and HD were 3, 13, 9, 9, 8, and 14, respectively. Most importantly, major and stable QTL were identified on 2A2 and 4B regions and explained respectively, 16-50% and 7-40% of the FHB traits phenotypic variation. . Some of the FHB resistance regions identified in this study were previously reported to be associated with loci for salinity tolerance, defense response genes, high yield, and quality traits. Since the pedigree of Parshall does not include Sumai3 background; we conclude that Parshall is a new source of FHB resistance with specific adaptation to the Northern American Central Plains region. As such, Parshall may be especially useful in wheat improvement and marker-based wheat breeding.

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GENETIC MAPPING OF QUANTITATIVE TRAIT LOCI FOR
FUSARIUM HEAD BLIGHT RESISTANCE IN SPRING
BARLEY 'KUTAHYA' AND WILD BARLEY 'W-365'

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ABSTRACT

The deployment of resistant cultivars is one of the best methods for controlling Fusarium head blight (FHB) in barley and reducing the impact of mycotoxin contamination in harvested grain. Here, we report on the identification of quantitative trait loci (QTL) for FHB resistance in two advanced backcross populations. The resistant donor parents are Kutahya, a two-rowed Dutch cultivar, and W-365, a wild barley from Iraq. The recurrent parent of both populations, Quest, is a six-rowed Minnesota barley with a moderate level of FHB resistance. From 2012-2014, the Kutahya/Quest population was screened for FHB resistance and accumulation of deoxynivalenol (DON) in 6-7 environments and the W-365/Quest population in 4-5 environments. Agronomic traits affecting disease development (i.e. plant height, heading date, row type, spike density, and spike angle) were also measured in multiple environments. The populations were genotyped using the Illumina Infinium Assay. Single nucleotide polymorphism (SNP) markers were used to construct the Kutahya/Quest (2,983 markers in total) and W-365/Quest (2,162 markers) maps. QTLs for FHB resistance were identified on every chromosome in the Kutahya/Quest population, explaining from 3.25-7.58% of the phenotypic variation for FHB severity. Of the 6 QTLs identified, those on chromosomes 1H, 2H, and 5H were consistently detected in most environments. In the W-365/Quest population, QTLs were identified on chromosomes 1H, 2H, 3H, 4H, and 5H and explained from 4.60-23.76% of the phenotypic variation for FHB severity. Previously described QTL contributed by Quest were confirmed on chromosomes 1H, 2H, 3H, 4H, 5H, and 6H. Resistance QTLs contributed by the Kutahya and W-365 identified on chromosome 2H, 3H, and 2H, 4H, respectively. In addition to identifying these resistance QTLs, putative transgressive segregants were identified.

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USING ASSOCIATION ANALYSIS AND GENOMIC SELECTION
TO IMPROVE FUSARIUM HEAD BLIGHT RESISTANCE
IN SOFT RED WINTER WHEAT

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ABSTRACT

With new marker platforms continuing to become available for crop species and the cost of genotyping reducing, breeders are now able to use this technology for crop improvement. Association analysis aims to identify marker trait relationships and is based on linkage disequilibrium between the quantitative trait loci (QTL) and markers: these associations can be used in marker-assisted selection (MAS). However, most quantitative traits are controlled by many small effect QTL making MAS ineffective. An alternative approach is to use genomic selection (GS) to estimate all marker effects simultaneously and determine the breeding value of the individuals. The first objective of this research was to identify, locate, and determine the magnitude of QTL effects for Fusarium head blight (FHB) resistance. The second objective was to determine the accuracy of GS models for predicting FHB resistance. We used a population of 470 elite breeding lines genotyped using genotyping-by-sequencing. Lines were phenotyped for FHB index for two years. The heritability of FHB Index was 0.59. We identified four significant QTL with R^2 values ranging from 2.6 to 2.8% and allele effects ranging from 1.07 to 1.76%. No QTLs were found in common between FHB and heading date (HD) or height. The relative efficiencies of GS models for FHB prediction ranged from 0.22 to 0.5. Based on these results we believe these technologies will be useful for improving FHB resistance in this soft red winter wheat population.

FUSARIUM HEAD BLIGHT RESISTANCE IN
SOFT RED WINTER WHEAT
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ABSTRACT

Development of resistant wheat cultivars is the most efficient approach to control Fusarium Head Blight. Local broadly adaptive cultivars have been crossed with *Fhb1* derived lines, Truman, Neuse, Jamestown, Oakes, and derived lines with *Fhb1* to introduce FHB resistant QTL into adaptive genetic backgrounds. Elite lines with resistance from Truman, Neuse, GAMD08-27-E9 and Jamestown, were evaluated in the field during 2014 for FHB resistance and agronomic performances. Several elite lines have been identified with good FHB resistant derived from Jamestown. GA051477-13ES4 from the cross of AGS 2020 / Jamestown had similar ratings as Jamestown for incidence, index and ISK. Lines with moderate levels of FHB resistance from either Jamestown or Neuse were identified with high yield potential. In addition, GA04151-10E29 was evaluated in the 2012 Uniform Southern Wheat Nursery and also showed moderate level of FHB resistance with high grain yield was released in 2014. Several other lines with Jamestown, Truman, Oakes, and IN 97397 as source of resistance were identified with moderate level for FHB index and ISK and high grain yield when compared to the checks “SS 8641” and “AGS 2035”. These lines will be further evaluated for FHB and grain yield. Double haploid lines, NC 10014-38 (NC 06-198-96/NC 08-140) and NC 10435-11 (NC 05-21937 / Oakes // Jamestown) showed a high level of FHB resistance and high yield performance.

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IDENTIFICATION OF FHB RESISTANCE QTL IN NATIVE SRW WHEAT CULTIVAR TRIBUTE

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ABSTRACT

Deployment of native sources of resistance to Fusarium head blight (FHB), caused by *Fusarium graminearum*, has been a priority in wheat (*Triticum aestivum*) breeding programs because of local adaptation and minimum yield drag. Pyramiding different sources of resistance would be an effective approach to enhance FHB resistance. The objectives of the study were to identify the FHB resistance QTL in the native soft red winter (SRW) wheat cultivar Tribute and develop diagnostic markers for use in marker-assisted breeding. A total of 115 double haploid (DH) lines, developed at NCSU, were evaluated for FHB incidence and FHB severity by cooperators in AR, KY, MD, NC, and VA during 2013 and 2014 (except MD). Grain samples from each location were visually assessed for *Fusarium* Damaged Kernels (FDK) and analyzed for deoxynivalenol (DON) toxin content. The population was also evaluated for type II resistance to disease spread in the greenhouse at Virginia Tech. A set of SSR markers were used to genotype the mapping population. Genotype-by-location interaction was significant for the population. Composite interval mapping identified seven putative QTL on chromosomes 1A, 1B, 2A, 2D, 3BS, 5A, and 7D for FHB incidence, FHB severity, FDK, and DON content. Putative QTL for FHB resistance were detected on 1A, 1B, 2A, 3BS, and 5A, whereas putative QTL for FHB susceptibility were detected on 7D. The putative QTL on 2D was associated with both resistance and susceptibility to FHB. The putative QTL for FHB on 2A and 2D were linked to loci governing heading date and flowering date across locations, whereas the putative QTL for FHB on 1B was linked to plant height but only in MD. The variation explained by putative QTL on 1A, 1B, 2A, 2D, 3BS, 5A, and 7D was 14% to 17% (Additive = -3.8 to -8.6), 8% to 20.6% (Additive = -1.1), 8% to 26% (Additive = -0.1 to -9.0), 40% to 42% (Additive = -8.4 and 9.0), 11% to 17% (Additive = -4.0 to -8.9), 12.5% to 15.7% (Additive = -5.3 to -8.6), and 11% to 15.7% (Additive = 5.2 to 10.4), respectively. The population is being genotyped for 90K SNP and diagnostic markers for the putative QTL on 1A, 3BS, and 5A will be identified for utilization in marker-assisted breeding.

**A BREEDING TOOL FOR ESTIMATING GENETIC VARIANCE
AND CORRELATED RESPONSE IN BI-PARENTAL CROSSES:
TARGETING HIGH-YIELD AND LOW-DEOXYNIVALENOL (DON)
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ABSTRACT

Breeding programs of self-pollinated crops usually focus on crosses between the most elite germplasm. While elite \times elite crosses ensure a desired population mean, they do not guarantee any level of genetic variation (V_G). Unlike population mean, which is simply predicted by the mid-parent value, prediction of genetic variance relies on the knowledge of population parameters that are not readily known. The purpose of this research was to use genome-wide markers to predict V_G of DON and yield of simulated bi-parental RIL populations resulting from pairwise crosses among parent candidates using a procedure that combines genomic simulation and genome-wide prediction. In our example, we demonstrate the utility of the procedure to evaluate the 435 possible crosses resulting from a half-diallel of 30 parents through simulation to identify a manageable number of crosses that could then be potentially advanced to field-based trials. We show that the procedure can be used to screen among high \times high crosses for yield and among low \times low crosses for DON based on the expected progeny variances across crosses. Since yield and DON are unfavorably correlated, we also demonstrate how equally yielding crosses may differ in their correlated response for DON. Finally, we propose a genome-wide equation to quantify the “coefficient of gene distribution” theoretically outlined in classical quantitative genetics texts as a measure of genetic distance in the context of a given trait. This computational resource will be provided in a package in the R environment and should be useful to breeders who are designing crosses to develop improved FHB resistant varieties.

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THE 2013-14 SOUTHERN UNIFORM SOFT RED
WINTER WHEAT SCAB NURSERY

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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 2013-14 nursery comprised 58 advanced generation breeding lines and four check cultivars, 'Ernie', 'Bess', 'Jamestown' (partially resistant) and 'Coker 9835' (susceptible). Seven U.S. public programs (Univ. of Arkansas, Univ. of Georgia, Louisiana State University, Univ. of Maryland, N.C. State Univ., VA Tech., and USDA-ARS), and three private companies (Agripro-Coker, KWS, and Limagrain) 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.

The mean level of FHB resistance in the nursery was high. Between 83 and 97 percent of entries had significantly better means than the susceptible check for Severity, Index and ISK. DON data are still being reported. Sources of resistance included Chinese, South and North American germplasms.

Copies of the full report will be available at the 2014 National Fusarium Head Blight Forum and subsequently on line at the USWBSI web site: <http://www.scabusa.org/>.

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Table 1. Means across locations and genotypic content of regions associated with FHB resistance.

	Cultivar/ Designation	FHB Incidence		FHB Severity		FHB Index		FDK		ISK		DON	
		RANK		RANK		RANK		RANK		RANK		RANK	
1	ERNIE	54	15	23	10	14	10	22	14	26	16		
2	COKER9835	88	62	62	62	56	62	51	61	63	62		
3	BESS	48	7	23	10	13	8	18	6	21	6		
4	JAMESTOWN	52	12	22	8	14	10	18	6	22	7		
5	M10-1615	51	9	21	6	12	4	22	14	26	16		
6	AR00179-2-2	60	27	26	16	19	21	16	4	29	24		
7	AR00334-5-2	66	44	27	18	21	27	22	14	31	29		
8	AR01136-3-2	62	33	32	29	24	37	29	30	31	29		
9	AR04001-3	54	15	29	23	16	17	25	21	24	12		
10	AR04084-1-3	59	26	34	34	20	25	27	24	29	24		
11	ARGE07-1347-6-7-9	43	2	20	2	12	4	19	9	18	2		
12	ARGE07-1354-2-6-1	55	19	27	18	19	21	21	13	24	12		
13	ARGE07-1355-16-6-6	54	15	28	22	19	21	18	6	23	8		
14	ARS09-228	63	37	33	32	23	34	32	39	31	29		
15	ARS10-028	74	57	29	23	24	37	30	34	36	43		
16	ARS10-038	68	48	59	61	41	60	55	62	57	61		
17	ARS10-043	81	61	40	48	34	57	39	51	49	58		
18	ARS10-172	78	59	52	59	44	61	43	56	51	59		
19	ARS10-389	40	1	20	2	9	2	20	11	15	1		
20	B09-0002	46	6	20	2	13	8	14	2	19	4		
21	B09*900256	65	42	43	53	30	53	46	58	45	56		
22	B08-91993	58	24	36	39	24	37	32	39	33	35		
23	GA04494-13ES1	61	29	49	57	31	54	48	59	41	52		
24	GA051477-13ES2	61	29	40	48	26	44	30	34	35	40		
25	GA051477-13ES4	45	5	32	31	15	15	28	27	24	12		
26	GA051207-13ES11	64	39	41	52	27	47	32	39	33	35		
27	GA061050-13ES18	62	33	40	48	23	34	37	49	34	38		
28	GA06586-13ES21	76	58	51	58	36	58	48	59	51	59		
29	GA06390-13ES24	62	33	38	43	27	47	43	56	34	38		
30	GA061050-13ES17	61	29	39	46	26	44	32	39	35	40		
31	KWS 013	65	42	39	46	28	49	34	46	38	46		
32	KWS 026	55	19	26	16	14	10	15	3	27	20		
33	KWS 027	68	48	36	39	26	44	41	55	38	46		
34	LA06149C-P7	71	53	34	34	24	37	29	30	38	46		
35	LA08201C-57	54	15	34	34	18	20	23	19	29	24		
36	LANC8170-41-1	64	39	43	53	24	37	32	39	38	46		
37	LA07085CW-P4	56	22	30	25	20	25	29	30	27	20		
38	LANC8170-41-2	55	19	31	27	16	17	23	19	26	16		
39	LCS 08577-4	73	55	52	59	36	58	39	51	48	57		
40	LCS 229	68	48	44	56	31	54	33	45	39	50		
41	M09-9547	60	27	34	34	21	27	32	39	33	35		
42	M11-1027#	64	39	31	27	21	27	28	27	35	40		
43	M11-2298	51	9	24	12	12	4	22	14	23	8		
44	MD08-22-22-13-4	56	22	25	14	19	21	13	1	23	8		
45	MD26-H2-23-13-1	66	44	25	14	22	30	20	11	29	24		
46	MD09W272-8-4-13-3	58	24	24	12	15	15	26	23	28	23		
47	MDC07026-F2-19-13-4	68	48	30	25	22	30	30	34	31	29		
48	NC11-21401	52	12	22	8	12	4	25	21	23	8		
49	NC11-22289	43	2	19	1	8	1	19	9	20	5		
50	NC11-22291	44	4	20	2	11	3	16	4	18	2		
51	NC8170-45-17	50	8	21	6	14	10	22	14	24	12		
52	NC8170-86-2	53	14	27	18	17	19	28	27	27	20		
53	NC9305-7	51	9	27	18	14	10	27	24	26	16		
54	NC09-21916	61	29	36	39	24	37	27	24	30	28		
55	VA10W-96	63	37	36	39	24	37	31	37	32	33		
56	VA11W-108†	73	55	40	48	32	56	38	50	44	54		
57	VA11W-230	62	33	32	29	22	30	31	37	32	33		
58	VA11W-278	66	44	43	53	28	49	40	53	39	50		
59	VA12W-102	69	52	38	43	29	52	34	46	41	52		
60	VA12W-150	66	44	38	43	23	34	29	30	37	45		
61	VA12FHB-37	78	59	34	34	28	49	40	53	44	54		
62	VA12FHB-85	71	53	33	32	22	30	36	48	36	43		
	Mean	61		33		22		29		32			
	LSD (0.05)	24		20		19		21		15			
	CV%	20.1		31.4		43.4		37.2		23.7			

Table 1. Continued

Cultivar/ Designation	Heading Date		Plant Height		Flour Yield %		Softness Equivalent %		Hessian Fly Biotype L	Fib1	Fib Massey 3BL	Fib 5A	Fib 2DL- Wuhan1/W14
	RANK		RANK		RANK		RANK						
1 ERNIE	131	7	33	12	66	46	55	36	0-14	-	Het	Ernie	-
2 COKER9835	135	51	31	3	65	59	63	1	0-15	-	-	no data	-
3 BESS	134	39	38	57	67	31	63	1	0-15	-	-	-	-
4 JAMESTOWN	130	2	34	25	68	18	60	13	0-20	-	-	-	-
5 M10-1615	131	7	35	38	69	10	57	26	0-16	-	-	-	-
6 AR00179-2-2	133	26	39	59	67	31	61	7	0-15	-	-	-	-
7 AR00334-5-2	133	26	39	58	67	31	58	21	0-20	-	Het	-	-
8 AR01136-3-2	134	39	37	51	67	31	58	21	0-16	-	-	-	-
9 AR04001-3	132	15	40	62	69	10	58	21	0-14	-	-	-	-
10 AR04084-1-3	135	51	36	46	70	6	59	17	0-11	-	-	-	-
11 ARGE07-1347-6-7-9	133	26	37	53	66	46	55	36	0-11	Het	Het	-	Het
12 ARGE07-1354-2-6-1	135	51	37	48	66	46	61	7	0-12	-	-	-	-
13 ARGE07-1355-16-6-6	133	26	40	61	67	31	59	17	0-11	-	-	-	-
14 ARS09-228	134	39	35	30	72	4	35	62	0-13	-	-	-	-
15 ARS10-028	134	39	34	18	73	2	44	58	0-12	-	-	-	-
16 ARS10-038	135	51	36	44	72	4	43	59	0-15	-	-	Ernie	-
17 ARS10-043	136	58	33	13	73	2	39	61	0-9	-	-	Ernie	-
18 ARS10-172	135	51	32	7	70	6	51	51	0-12	-	-	-	-
19 ARS10-389	130	2	34	23	74	1	41	60	0-13	-	-	-	-
20 B09-0002	132	15	36	45	68	18	53	45	0-10	-	-	-	-
21 B09*900256	133	26	34	20	65	59	60	13	0-14	-	-	Ernie	-
22 B08-91993	131	7	38	56	66	46	60	13	0-15	-	Yes	-	-
23 GA04494-13ES1	131	7	31	4	69	10	52	48	0-16	-	-	Ernie	-
24 GA051477-13ES2	131	7	36	41	68	18	62	6	0-14	-	-	-	-
25 GA051477-13ES4	131	7	35	31	67	31	63	1	0-14	-	-	-	-
26 GA051207-13ES11	132	15	36	39	70	6	58	21	0-14	-	-	-	-
27 GA061050-13ES18	132	15	32	6	66	46	58	21	0-15	-	-	-	-
28 GA06586-13ES21	132	15	36	42	69	10	50	54	0-7	-	-	-	-
29 GA06390-13ES24	132	15	31	2	68	18	55	36	0-12	-	-	-	-
30 GA061050-13ES17	132	15	35	26	66	46	59	17	0-15	-	-	-	-
31 KWS 013	129	1	35	36	67	31	57	26	0-16	-	-	Ernie	-
32 KWS 026	132	15	36	40	68	18	57	26	0-15	-	Het	-	-
33 KWS 027	137	61	38	54	70	6	53	45	0-18	-	-	-	-
34 LA06149C-P7	135	51	37	50	66	46	61	7	0-19	-	-	-	-
35 LA08201C-57	134	39	39	60	68	18	56	33	0-17	-	-	-	-
36 LANC8170-41-1	134	39	35	27	67	31	49	56	0-13	Yes	-	-	-
37 LA07085CW-P4	131	7	34	19	68	18	60	13	0-15	-	-	-	-
38 LANC8170-41-2	133	26	32	8	68	18	51	51	0-13	Yes	-	-	-
39 LCS 08577-4	133	26	35	33	69	10	63	1	0-15	-	-	-	-
40 LCS 229	132	15	35	35	67	31	63	1	0-15	-	Yes	-	-
41 M09-9547	134	39	37	47	68	18	54	43	0-15	-	-	-	-
42 M11-1027#	134	39	34	24	66	46	61	7	0-14	-	Het	-	-
43 M11-2298	132	15	37	52	67	31	55	36	0-12	-	Het	-	-
44 MD08-22-22-13-4	134	39	33	9	66	46	54	43	0-14	Yes	-	Ning	Yes
45 MD26-H2-23-13-1	136	58	34	17	67	31	51	51	0-13	Yes	-	Ning	Yes
46 MD09W272-8-4-13-3	133	26	33	14	66	46	56	33	0-14	Yes	-	-	-
47 MDC07026-F2-19-13-4	133	26	34	16	69	10	55	36	0-19	Yes	-	-	-
48 NC11-21401	133	26	32	5	65	59	56	33	0-15	Yes	-	-	Yes
49 NC11-22289	130	2	34	21	66	46	50	54	0-16	-	-	-	-
50 NC11-22291	130	2	35	29	67	31	52	48	0-13	-	-	-	-
51 NC8170-45-17	134	39	36	43	67	31	52	48	0-14	Yes	-	Ning	-
52 NC8170-86-2	133	26	35	32	65	59	55	36	0-17	Yes	-	-	-
53 NC9305-7	134	39	38	55	66	46	55	36	0-12	-	-	-	-
54 NC09-21916	133	26	35	34	68	18	61	7	0-17	-	-	-	-
55 VA10W-96	130	2	35	37	68	18	53	45	0-15	-	-	-	-
56 VA11W-108†	133	26	35	28	67	31	61	7	0-12	-	-	-	-
57 VA11W-230	132	15	33	10	68	18	57	26	0-12	-	-	-	-
58 VA11W-278	131	7	33	15	67	31	57	26	0-14	-	Yes	-	-
59 VA12W-102	135	51	31	1	69	10	59	17	0-15	-	-	-	-
60 VA12W-150	134	39	34	22	66	46	57	26	0-10	-	-	-	-
61 VA12FHB-37	137	61	33	11	68	18	57	26	0-15	-	-	-	-
62 VA12FHB-85	136	58	37	49	69	10	49	56	0-15	-	Het	-	-
Mean	133		35		68		55						
LSD (0.05)	4		3		.		.						
CV%	1.4		5.0		.		.						

QTL CONFERRING TYPE II RESISTANCE TO FUSARIUM
HEAD BLIGHT IN ADAPTED WHEAT CULTIVAR
'UI STONE' AND ITS EFFECT ON YIELD

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ABSTRACT

Fusarium head blight (FHB) is an emerging wheat disease in Southeastern Idaho. Most cultivars grown in this region are susceptible to FHB. Several exotic sources of resistance are available to fill this hole, however, lack of adaptation and often complication arises due to "linkage drag" limit their direct utilization. Therefore, identification and development of adapted FHB resistant cultivars is one of major objectives in the University of Idaho wheat breeding program. The objectives of this study were to map QTL associate with type II FHB resistance in the adapted soft white spring (SWS) wheat cultivar 'UI Stone' and evaluate side effect of identified QTL on yield. A total of 151($F_{4,6}$) recombinant inbred lines (RILs) derived from the cross between resistant cultivar 'UI Stone' and a moderately susceptible cultivar 'Alturas' were evaluated for type II FHB resistance by measuring disease severity expressed as a percentage of infected spikelets (PIS) in four greenhouse experiments over three years. The RILs were genotyped with 154 markers (77 SSR and 77 SNP). A Linkage map was constructed using MapMaker 3.0b and QTL analysis was performed using WinQTL Cartographer Ver. 2.5. Two major QTL for type II FHB resistance, *QFhbuis.ab-2B* and *QFhbuis.ab-3B*, were identified by both single marker analysis and composite interval mapping (CIM) methods and the two QTL together explained 23.6 to 24.8% of phenotypic variation. These QTL had no significant effect on yield based on the regression analyses. This study also identified 4 lines with better FHB resistance and higher grain yield than UI Stone. These four lines could be used as germplasm and/or released as new resistant cultivars after further evaluation. More phenotypic evaluation for FHB severity by cooperators in 2015 and 90K SNP marker data will be utilized to validate the effect of these two QTL for potential application in marker-assisted breeding.

FT-NIR OPTICAL CHARACTERISTICS OF SOUND AND *FUSARIUM* DAMAGED WHEAT AT TWO MOISTURE CONTENT LEVELS

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ABSTRACT

In prior research, we developed near-infrared spectrometric methods to detect *Fusarium*-damaged kernels (FDK) of wheat and their deoxynivalenol (DON) levels using the Single Kernel Near-Infrared (SKNIR) system which is operated in the 950-1650 nm spectral range. Those studies showed that DON has NIR absorption bands with peaks around 1408, 1904 and 1919 nm. These DON absorption bands may be interfered by the broad water absorption bands around 1450 and 1940 nm. Water and DON bands may be shifted to lower or higher wavelengths depending on the moisture content and *Fusarium* damage levels of the grain matrix and this may affect the performance of calibrations for detection and quantification of DON levels in FDK, especially when kernels with different moisture content levels are evaluated. Therefore, it is useful to study the NIR absorption patterns of sound and FDK at different moisture content levels to identify specific NIR absorption bands associated with moisture and *Fusarium* damage in kernels independent from each other. That information may be useful to improve NIR calibrations and also may help improve FDK sorting by the new rapid LED-based grain sorters. To study the FDK and moisture absorption bands, sound and FDK of cultivar 'Art' harvested from a FHB screening nursery were selected. Half of the kernels (~5-6 g) were partly dried at 80C for one hour (dry) and the rest were at equilibrium moisture level (wet) at room temperature. The kernels were scanned with two repacks in glass vials (14mm x 45mm) using the PerkinElmer Spectrum 400 FT-NIR spectrometer in the 1000-2500 nm spectral range. The spectrometer conditions used were data interval = 2nm; resolution = 16nm, number of scans = 25 with 0.2 cm/s scan mirror speed. The second derivatives of the spectra were constructed with 25 data points for slope calculation using PerkinElmer Spectrum software. Spectral subtractions were performed between wet and dry kernels of sound and FDK groups, and between sound and FDK of dry and wet kernel groups to identify peak NIR absorptions due to water and *Fusarium* damage, respectively. *Fusarium* damage related peak absorptions were observed at 1013 nm, 1198nm, 1274 nm, 1355-1362 nm, 1418-1428 nm, 1585nm, 1698nm, 1744nm, 1782nm, 1826nm, 2276nm, 2328 nm and 2371 nm regions. Moisture content related peak absorptions were detected at 1162 nm, 1337 nm, 1405-1408nm and 2002 nm regions. These absorption peaks which are isolated and independent from the influence of absorptions due either to moisture or *Fusarium* damage may be useful for improving performance of NIR calibrations for scab sorting and DON estimation and for modifications in LED-based high speed sorting instruments by incorporating LEDs for those wavelengths.

FUSARIUM HEAD BLIGHT RESISTANCE QTL IN THE NC-NEUSE /
AGS2000 RECOMBINANT INBRED LINE POPULATION

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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. The North Carolina cultivar NC-Neuse is a moderately FHB resistant soft red winter wheat, released in 2003.

A population of 170 random F₅-derived recombinant inbred lines derived from a cross between ‘NC-Neuse’ and the FHB susceptible line ‘AGS 2000’ was evaluated for FHB resistance over several years and locations. Suitable data for at least some FHB traits were collected from a total of seven environments (2-3 reps/env). These included Kinston, NC in 2011, 2012, 2013, and 2014; Salisbury, MD in 2012; and Lake Wheeler, NC in 2013 and 2014. The FHB related traits evaluated were disease incidence (INC), severity (SEV), *Fusarium* damaged kernels (FDK), and accumulation of the mycotoxin deoxynivalenol (DON).

Least squares means (lsmeans) were calculated from the phenotypic data within and across environments. In environments where heading date (HD) was significant, this was used as covariate in the data analysis and calculations of lsmeans.

A linkage map containing a total of 1839 polymorphic SSR, DArT and SNP markers across 27 linkage groups was developed and utilized for mapping of QTL in this population. QTL analysis was conducted using Composite Interval Mapping (CIM) and then Multiple Interval Mapping (MIM) with WinQTLCart 2.5. The critical LOD value to declare QTL significance was 3.0, based on 1000 permutations.

We identified QTL associated with FHB resistance (from several environments and/or several resistance traits) on chromosomes 1A, 1B, 2A, 4A, 5B, and 6A. Their LOD scores ranged from 3.0 to 5.4 with effects between 5.5-11.5%. At all QTL except the one on chromosome 5B, the NC-Neuse allele contributed resistance. The QTL on 5B (co-localized with the *Vrn-B1* locus) showed up only in 2012 environments, probably due to an usually mild winter. QTL for HD and plant height were mapped to chromosomes 2B, 4A, 5B, 6A, and 7D. The resistance QTL on chromosomes 4A and 6A did not co-localize with QTL controlling HD and/or plant height.

In the coming months, markers associated with the identified resistance QTL will be run on a broader set of wheat lines to test their usefulness and to test frequencies of resistance alleles.

ACKNOWLEDGEMENT AND DISCLAIMER

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VALIDATION OF FUSARIUM HEAD BLIGHT
RESISTANCE QTL USING THE NC-NEUSE / BESS
DOUBLED HAPLOID POPULATION

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ABSTRACT

Fusarium Head Blight (FHB) is one of the most damaging diseases of wheat. It lowers the grain yield and quality, and contaminates grain with the mycotoxin deoxynivalenol (DON). Genetic resistance is a critical control measure and breeding objective. Many studies have focused on the genetic basis of FHB resistance in Asian wheat sources, while resistance in native sources has not been characterized as well yet. In addition, the need for validation of mapped QTL remains an important research objective in order to use markers more efficiently in marker-assisted selection (MAS).

The two cultivars 'NC-Neuse' and 'Bess' display moderate resistance to FHB. NC-Neuse was developed and released from North Carolina State University. Bess is one of the most FHB resistant lines in the Southeast, and is a full-sib of the cultivar 'Truman'. Bess and Truman were both developed and released from the University of Missouri, and have similar resistance levels.

Quantitative trait loci (QTL) associated with FHB resistance in NC-Neuse were very recently identified and mapped to chromosomes 1A, 1B, 2A, 4A, 5B, and 6A (Abstract presented at this Forum also).

The wheat breeding group at University of Missouri recently identified and mapped QTL associated with resistance in Truman (*in press*). The QTL were mapped to chromosomes 1B, 2A, 2B, 2D, 3B, 4B, 6B, and 7B.

The objective of this study was to map QTL for FHB resistance in the NC-Neuse / Bess DH population, and use these results to validate FHB resistance QTL found in previous studies including NC-Neuse and Truman.

A population of 100 doubled haploid (DH) lines derived from a cross between NC-Neuse and Bess was evaluated for FHB resistance over several years and locations. Suitable data for at least some FHB traits was collected from a total of seven environments (2-3 reps/env). These included Kinston, NC in 2012, 2013, and 2014; Columbia, MO in 2012 and 2013; and Lake Wheeler, NC in 2013 and 2014. The FHB related traits evaluated were disease incidence (INC), severity (SEV), *Fusarium* damaged kernels (FDK), and accumulation of DON.

Least squares means (lsmeans) were calculated from the phenotypic data within and across environments. In environments where heading date (HD) was significant, it was used as covariate in the data analysis and calculations of lsmeans.

A linkage map containing a total of 4013 polymorphic SSR and SNP markers across 51 linkage groups was developed and utilized for mapping of QTL associated with FHB resistance in this population. QTL analysis using lsmeans from the phenotypic data (across and within individual environments) was conducted using Composite Interval Mapping (CIM) and then Multiple Interval Mapping (MIM) with WinQTLCart 2.5. The critical LOD value to declare QTL significance was 3.0, based on 1000 permutations.

Preliminary results showed QTL associated with one or more FHB resistance traits on chromosomes 1A, 1B, 2A, 2B, 3A, 3B, 4A, 4B, 5A, and 6A. At the QTL on chromosomes 1A, 2A, 3A, 4A, 4B, and 6A the NC-Neuse allele conferred resistance. At the QTL on chromosomes 1B, 2B, 3B, and 5A the Bess allele conferred resistance. Their LOD scores ranged from 3.13 to 10.33 with effects between 4.4-23.6%.

An update on pertinent results and map comparisons will be presented at the Forum.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0206-0-083. 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.

CHANGES IN THE WHEAT PRIMARY METABOLISM DURING DEFENSE AGAINST *FUSARIUM GRAMINEARUM*

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ABSTRACT

Wheat is commonly infected by *Fusarium graminearum*, causing Fusarium head blight, which leads to severe losses in grain yield and quality. A decisive factor in the mounted defense response is how plants counteract the effects of the *F. graminearum* toxin deoxynivalenol, which elicits oxidative stress and inhibits protein biosynthesis. In response plants generate secondary metabolites, inactivate the toxin by metabolization and try to compensate for the reduced levels of protein biosynthesis. All these incur at significant cost and require restructuring of the primary metabolism to meet the elevated needs in energy, carbon and nitrogen equivalents. We have investigated changes to the primary metabolism in response to the pathogen in transcriptomic and metabolomic datasets and additionally to the toxin in metabolomic datasets. These data have been generated by RNAseq and GC-MS from wheat near-isogenic lines segregating for the resistance QTL *Fhb1* and *Qfhs.ifa-5A* in a series of time points after inoculation with the fungus or deoxynivalenol. We observed increased levels in the respiration including the pentose phosphate pathway, which produces also erythrose-4-phosphate, required as a precursor in the shikimate pathway, which ultimately leads to the production of defense-associated phenylpropanoids. The detrimental effects on translation by the toxin are met by the increased synthesis of amino acids and tRNA ligases. Significant differences for *Fhb1* were observed in metabolite levels after DON treatment and to a lesser extent for lines lacking *Qfhs.ifa-5A* after *Fusarium graminearum* treatment.

PYRAMIDING *FHB1* WITH USEFUL RUST RESISTANCE GENES
IN A WINTER-HARDY WHEAT GENETIC BACKGROUND

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ABSTRACT

A winter wheat breeding program was initiated at North Dakota State University in 2011. The first objective of the new program is to develop a productive breeding population with adequate variation for cold-hardiness, yield, disease resistance and processing quality.

This study is part of a larger pre-breeding effort to develop new parental materials carrying useful genes for disease resistance and adaptation. Norstar is an old Canadian variety with exceptional cold-hardiness, yet is lacking in disease resistance and is too tall under North Dakota growing conditions. In view of the difficulty to pyramid cold-hardiness (low heritability) with disease resistance through regular crosses, it was decided to upgrade Norstar for its future use as a breeding parent. Marker-assisted backcrosses were therefore employed to transfer and pyramid combinations of resistance genes into the Norstar background.

The targeted genes included a Fusarium head blight resistance gene (*Fhb1*), two leaf rust resistance genes (*Lr34*, *Lr53*) and four stem rust resistance genes (*Sr2*, *Sr26*, *Sr39*, *Sr50*). An attempt was also made to co-transfer the reduced height gene, *Rht-B1b*, with the disease resistance genes. Following the third backcross to Norstar, the various near-isogenic progenies were inter-mated to derive progeny having combinations of *Fhb1* and *Rht-B1b* plus targeted leaf and/or stem rust resistance genes. Five different near-isogenic lines (each carrying *Fhb1* and *Rht-B1b*) that differ for the leaf and stem rust resistance genes they possess, were recovered following selfing of the intercrossed F₁ progenies.

With respect to their utility as cross parents, the set of NILs: (i) will be used in direct crosses with other breeding parents; (ii) will first be inter-crossed to derive more complex F₁ parents that are homozygous *Fhb1*, *RhtB1b*, but simultaneously heterozygous for two or more rust resistance genes.

DEVELOP SNP MARKERS FOR *FHB1* THROUGH FINE
MAPPING USING WHEAT 90K SNP ARRAYS
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ABSTRACT

Wheat Fusarium head blight (FHB) is one of the most destructive diseases limiting wheat production worldwide. *Fhb1*, located on chromosome 3BS, is the most stable quantitative trait locus (QTL) with the largest effect on wheat resistance to spread of diseases in a spike (type II). *Fhb1* has been widely used in breeding to improve FHB resistance in wheat, but the gene underlying *Fhb1* has not been cloned. This study used single nucleotide polymorphism (SNP) from wheat 90K SNP arrays to saturate the *Fhb1* region. A high-density linkage map was constructed by adding 25 SNP markers to the region using a recombinant inbred population and a backcross-derived near isogenic line (NIL) population derived from Ning7840 x Clark BC₂F₂. *Fhb1* was delimited to a 0.88 cM interval between SNP3026 and SNP241 containing 11 SNPs. The SNP markers in the *Fhb1* region were converted to competitive allele-specific PCR (KASP) markers for further fine mapping. SNP79259 and SNP77323 co-segregated with *Fhb1* in the 376 NILs that have recombination between *Xgwm533* and *Xgwm493*. The physical distance of the two SNP markers is about 300 kb on the reference sequences of chromosome 3B contig 0954 of Chinese Spring. The two KASP SNPs can be used for marker-assisted pyramiding of *Fhb1* with other resistance QTLs. This result demonstrated that wheat 90K SNP array is a useful tool for increasing the marker density in the candidate gene region to facilitate fine mapping in wheat.

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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.

VALIDATION OF FUSARIUM HEAD BLIGHT RESISTANCE
QTLs IN WHEAT USING DOUBLE HAPLOIDS
DERIVED FROM FOUR-WAY CROSSES

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ABSTRACT

Fusarium head blight (FHB), caused primarily by *Fusarium graminearum*, is one of the most devastating plant diseases in the world. Specifically in wheat, FHB has become responsible for significant economic and health concerns worldwide due to mycotoxin accumulation in infected grain, as well as yield and quality losses. To date, sources of resistance conferring complete resistance to FHB have not been identified in wheat. Thus, extensive research efforts worldwide has focused on development and use of resistant cereal cultivars for the control of FHB. QTLs for FHB resistance have been mapped to almost all wheat chromosomes when different mapping populations were investigated. In our research, we are using double haploid (DH) wheat lines derived from selected four-way crosses combining several sources of resistance to validate putative QTLs (Xmc758, Gwm33, xbacr176, Xgm120, Xwmc317, Xwmc332, Xwmc522 and Xwmc296) that could minimize the threat of FHB including the reduction of mycotoxins, to the producers, processors, and consumers of wheat. We use molecular techniques to validate DH lines and their corresponding parents. In this study, we report on our work on the DH derived lines screen for FHB in three northern plains location. Our finding will assist ongoing efforts aimed to develop resistance wheat varieties, minimize the impact of the disease, and provide resources that can possibly assist in the advancement of wheat germplasm research.

**CHANGES IN FUSARIUM HEAD BLIGHT AND GRAIN YIELD
TRAITS OVER THREE CYCLES OF GENOMIC SELECTION
IN A BARLEY BREEDING POPULATION**

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ABSTRACT

To date, the potential of genomic selection (GS) has been documented largely by theoretical and simulation-based research. While not entirely absent, empirical evidence of genomic selection's efficacy, principally gain from selection, is lacking in the literature. The barley program at the University of Minnesota has implemented GS and has so far completed five cycles of advanced cycle breeding. The first three breeding cycles, each comprised of 50 selected individuals accompanied by 50 randomly selected individuals, along with the "cycle 0" parents have been grown as a single experiment in multiple Minnesota environments and phenotyped for multiple traits, including Fusarium head blight, grain deoxynivalenol concentration, and grain yield. This study will assess gain from selection over the first three breeding cycles and provide part of the empirical evidence for the effectiveness of GS in plant breeding.

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GENETIC AND MOLECULAR ANALYSIS
OF NOVEL FHB RESISTANCE

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ABSTRACT

Fusarium head blight (FHB) caused by *Fusarium graminearum* (*Fg*) is one of the most prevalent diseases of wheat (*Triticum aestivum* L.) and other small grain cereals. It has proven difficult to move existing sources of FHB resistance into adapted Canadian varieties due to poor agronomics and low yield. Recently, a novel source of FHB resistance, FL62R1, which has high FHB resistance and high agronomics and yield, was created by germplasm developers at Agriculture and Agri-Food Canada. In this study, we will use genetic and molecular approaches to characterize this new FHB resistance and with the goal of eventually introgressing desirable alleles into Canadian elite wheat varieties. The spread of *Fg* in heads of FL62R1 was considerably reduced compared to susceptible varieties, and the high level of type II resistance observed was similar to the well-known FHB resistant variety, Sumai 3. Fungal progression was monitored by using a GFP-tagged *Fg* strain. Microscopic data showed that *Fg* was effectively blocked in the rachis and did not spread to uninoculated spikelets of FL62R1. Double haploid mapping populations of FL62R1 crossed with two Canadian elite wheat varieties have been generated for genetic analysis.

IDENTIFICATION OF NEW QTL FOR NATIVE
RESISTANCE TO FHB IN SRW WHEAT

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ABSTRACT

Fusarium Head Blight (FHB), caused by *Fusarium graminearum* Schwabe, is a major disease of wheat (*Triticum aestivum* L.). Frequently FHB results in severe yield loss, reduced seed quality, and accumulation of mycotoxins such as deoxynivalenol (DON) and nivalenol (NIV). Control of FHB can be achieved via pyramiding multiple genes conferring resistance to initial infection, disease spread, kernel damage, and toxin accumulation and in turn provide for broad and effective cumulative resistance. The objective of this study was to identify quantitative trait loci (QTL) associated with FHB in the native soft red winter (SRW) wheat cultivar Jamestown. A total of 186 F_{5,7} recombinant inbred lines (RILs) derived from a cross of Pioneer 25R47 / Jamestown (P47/JT) were evaluated for FHB incidence, FHB severity, FHB index, *Fusarium* damaged kernels (FDK) and DON concentration for two years in three environments (MD, NC, and VA). Both public and proprietary single nucleotide polymorphism (SNP) markers were used at Monsanto Company to initially genotype 42 of the P47/JT RILs having contrasting phenotypes for FHB. Subsequently, a set of 142 RILs were genotyped with public 90K SNP. Bulk segregant analysis was used to select microsatellite markers (SSRs) associated with FHB. Linkage maps were constructed using JoinMap. Windows Cartographer (WinQTLCart version 2.5) was used to identify possible QTLs. Six consistent QTL identified in P47/JT and located on chromosomes 1B, 3B, 5A, 5B, and 6A were associated with FHB incidence, FHB severity, FDK and DON content. The putative QTL on 1B and 6A were associated with resistance to FHB, whereas the putative QTL on 3B, 5A, and 5B were associated with susceptibility to FHB. The variation explained by putative FHB resistance QTL on 1B and 6A was 7% to 19.5% (Additive = -0.3 to -7.4) and 7.2% to 14.7% (Additive = -0.8 to -6.0). The most diagnostic marker for the QTL on 1B was *WMC500*; flanked by *GWM18* and *GWM273* (12.2 cM interval). The QTL on 6A was flanked by *Barc146* and *D_GBUVHFX01CSU22_382* (10.3 cM interval). These QTL are being validated in FG95195 / Jamestown and Jamestown / LA97113UC-124 mapping populations. Diagnostic markers for FHB resistance QTL in Jamestown would facilitate marker-assisted breeding.

IDENTIFICATION AND MAPPING OF QUANTITATIVE TRAIT
LOCI FOR FUSARIUM HEAD BLIGHT RESISTANCE
IN EMMER AND DURUM WHEAT

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ABSTRACT

Fusarium head blight (FHB), caused by *Fusarium graminearum*, presently threatens durum wheat (*Triticum turgidum* subsp. *durum*) production in many durum-growing regions. It is critical to identify useful sources of FHB resistance for durum wheat. A domesticated emmer wheat (*T. turgidum* subsp. *dicoccum*) accession, PI 41025, was previously shown to be moderately resistant to FHB. This study was undertaken to identify quantitative trait loci (QTL) associated with FHB resistance in PI 41025. A population of 200 recombinant inbred lines developed from a cross between the durum variety 'Ben' and PI 41025 was evaluated for reaction to *F. graminearum* in one field and three greenhouse environments. The disease severity data and a single nucleotide polymorphism marker-based linkage map from this population were used for QTL analysis. The results showed that a QTL on chromosome 2A derived from Ben and two QTL on 3A and 5A derived from PI 41025 were associated with FHB resistance. The 2A and 3A QTL were detected only in the greenhouse experiments and they each explained 8% of the phenotypic variation. The QTL on 5A, which mapped very close to the domestication gene *Q*, explained 11% and 35% of phenotypic variation in greenhouse and field evaluations, respectively. The identification of the 2A QTL from Ben confirmed the presence of FHB resistance in North Dakota durum cultivars, which have been successfully used for developing new varieties with improved FHB tolerance. This study indicates that combining the QTL from related tetraploid species with native durum QTL will be useful for improving FHB resistance in durum wheat.

ACKNOWLEDGEMENT AND DISCLAIMER

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MOLECULAR MAPPING OF FUSARIUM HEAD
BLIGHT RESISTANCE IN ND2710

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ABSTRACT

ND2710 is a hard red spring wheat line developed by NDSU wheat breeding program with a very high level of resistance to Fusarium head blight (FHB). It was selected from the progeny of a cross between ND2603 (an advanced breeding line derived from the Sumai 3/Wheaton cross) and Grandin. Therefore, the FHB resistance of ND2710 is presumably derived from Sumai 3 since both Grandin and Wheaton are very susceptible to FHB. To identify and map the quantitative trait loci (QTL) for FHB resistance in ND2710, we developed a mapping population consisting of 233 recombinant inbred lines (RILs) from a cross between ND2710 and the CIMMYT spring wheat 'Bobwhite'. These RILs along with their parents and checks were evaluated for reactions to FHB in four greenhouse seasons and two field locations in 2013 and 2014. A linkage map was developed for this population using 747 SNP markers, which were distributed on 19 of the 21 wheat chromosomes spanning 1,716 cM of genetic distance. Further analyses using both phenotype and genotype data identified one major QTL on chromosome 3BS, explaining up to 27.3% of FHB severity variation in all experiments, and minor QTLs on 2A, 2B, 6A, and 6B explaining up to 10% phenotypic variation in at least two experiments. The QTL on 3BS and 6B were mapped to the same genomic regions as those harboring *Fhb1* and *Fhb2* in Sumai 3 or its derivatives. Plant maturity was not associated with FHB resistance. Three SSR markers (*Xgwm533*, *Xgwm493*, and UMN10) were also mapped to the 3BS QTL region saturated with SNP markers. These SNP markers will be further validated and used for marker-assisted selection of *Fhb1* in wheat breeding programs.

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EFFECTS OF DURUM WHEAT BACKGROUND ON THE EXPRESSION OF HEXAPLOID WHEAT-DERIVED FUSARIUM HEAD BLIGHT RESISTANCE GENES

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ABSTRACT

Multiple Fusarium head blight (FHB) resistance sources have been identified in common wheat, but an effective source of resistance to FHB has not been found in durum wheat. Significant efforts have been made toward introgression of FHB resistance from hexaploid wheat to durum wheat. However, progress has been limited due to complex inheritance patterns of the hexaploid wheat-derived FHB resistance in durum background. Here we report preliminary results on the effects of durum background on the expression of hexaploid wheat-derived FHB resistance genes. Two highly FHB-resistant hexaploid spring wheat accessions, Sumai 3 and PI 277012, were crossed to the durum cultivars ‘Langdon’ (LDN), ‘Divide’, ‘Grenora’, ‘Alkabo’, and LDN-Chinese Spring (CS) D genome substitution lines where a pair of homologous LDN A- or B-genome chromosomes were substituted by their homoeologous counterparts in CS D genome. Also, Sumai 3 was crossed to four FHB-susceptible hexaploid wheat accessions (‘2398’, ‘Choteau’, ‘AC Vista’, and ‘AC Lillian’). All F₁’s of Sumai 3 with durum exhibited a resistance level similar as or lower than their durum parents, whereas F₁’s of Sumai 3 with hexaploids exhibited a resistance level intermediate to their parents. Apparently, FHB resistance genes in Sumai 3, including *Fhb1*, were normally expressed in the F₁’s with hexaploids, but not in the F₁’s with durum. The F₁’s of PI 277012 with durum all exhibited a resistance level comparable to PI 277012, indicating complete dominance of the resistance genes in PI 277012 over the susceptible alleles in durum. Individual homoeologous substitution of D-genome chromosomes for LDN durum chromosomes 2B, 3A, 3B, 4A, 4B, 5B, 6A, 6B, and 7A all augmented resistance levels of the F₁’s between Sumai 3 and the LDN D genome substitution lines, suggesting these durum chromosomes may contain genes that suppress expression of the Sumai 3-derived FHB resistance genes in the F₁’s. Individual substitution of LDN durum chromosomes 4A, 6A, and 6B by their D-genome homoeologs lowered resistance levels in the F₁’s of PI 277012 with the LDN D genome substitution lines, whereas individual substitution of other LDN durum chromosomes did not significantly change resistance levels of their F₁’s. This suggests that LDN chromosomes 4A, 6A, and 6B may contain genetic factors required for the expression of the PI 277012-derived FHB resistance genes in the F₁’s. A wide range of segregation on FHB severity (10-90%) was observed in the F₂ generation from the crosses of Sumai 3 with durum LDN and Divide. The F₃ families derived from the most resistant F₂ segregants segregated toward more susceptible end. A similar segregation trend as the F₃ families was observed in the F₄ generation. We hardly found individuals with significantly higher levels of resistance than their durum parents in the F₄ generation. In the crosses of PI 277012 with durum, resistance also seemed to be slightly diluting over generations, but multiple resistant segregants were recovered in each generation of these crosses. Thereby, durum wheat contains multiple genetic factors on different chromosomes that positively and/or negatively

regulate expression of hexaploid wheat-derived FHB resistance genes. This has made FHB resistance introgression from hexaploids into durum a challenging task.

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OTHER

THE NEW USWBSI WEBSITE

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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. This year the site was completely rebuilt using the Drupal (<https://www.drupal.org/>) content management system (CMS). Previously, the site was run using the Xoops (<http://xoops.org/>) CMS. Unfortunately, several of the modules that were used to provide services through Xoops were no longer being updated, and presented security and stability threats. The Drupal CMS was chosen because of its extensive support, vast number of application modules, and improved content management and administrative interfaces. The new site was designed to provide all of the services previously available with Xoops plus some additional services such as a jobs posting board and a full featured image gallery. More services are expected to be offered in the future along with user interface enhancements.

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