

# **SESSION 1:**

## **VARIETY DEVELOPMENT AND HOST PLANT RESISTANCE**



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## FHB RESISTANCE QTL MAPPING USING NATIVE SOURCE OF RESISTANCE AND SNP-GBS MARKERS

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### ABSTRACT

Several wheat breeding programs across North America rely on exotic sources of resistance to FHB. Lack of adaptation and linkage drag are common problems associated with the exotic sources. DNA markers for FHB resistance, derived from adapted progenitors can increase our ability to select resistant lines with better agronomic traits. The winter wheat breeding program at the University of Illinois has identified several lines with improved levels of FHB resistance using native germplasm. One example is IL97-1828, which has been extensively used in the program, does not carry *Fhb-1*, and is one of our best breeding lines for FHB resistance. In order to map QTL associated with FHB resistance, a population of recombinant inbred lines was derived from a cross between IL97-1828 x Clark (susceptible) via SSD until the F<sub>5,6</sub> generation. Two hundred and five RILs were evaluated in two locations, Urbana-IL (2009 and 2010) and Wooster-OH (2010). Artificial inoculation and mist irrigation were applied in order to increase the inoculum pressure and favor the disease development. The Genotyping by Sequencing (GBS) protocol consisted of a two restriction-enzyme genome reduction, where a common cutter (*PstI*) and a rare cutter (*MspI*) were combined during the library preparation. Sequence data were obtained from an Illumina HiSeq2000 run. The data were analyzed with TASSEL UNEAK Pipeline and the program MapDisto was used for building a linkage map. The final version of the map was built using LOD = 3,  $r_{\max} = 0.3$  and 835 polymorphic markers: 671 “unique” SNP-GBS markers, in addition to 154 DArT and 10 SSR markers that have been previously used with this population. Single marker analysis, standard interval mapping and Haley-Knott regression were performed on R/QTL for all traits measured in the field, having heading date as a covariate in each analysis. Eight QTL were declared significant over multiple location/years: three were associated with FHB incidence (1B, 1D and 2B), three were found for severity (1B, 2B and 3B) and two were associated with FDK (2A and 3B). No major effect QTL were detected. The genetic distance between significant markers and QTL averaged 2.7 cM, ranging from 0 to 7.98cM. Fourteen extra QTL were found to be environment specific. The two-QTL scan procedure available in R/QTL revealed significant interactions between QTL for most traits. Next generation sequencing markers allowed us to map QTL that have gone undetected before, when only DArT and SSR markers have been used. This study also show that native sources of resistance like IL97-1828 can contribute to the development of wheat with improved FHB resistance.

### ACKNOWLEDGEMENT AND DISCLAIMER

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## LIFELONG LEARNING: WHAT WE HAVE LEARNED BREEDING FOR SCAB TOLERANCE

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### ABSTRACT

In the Great Plains, *Fusarium* head blight (FHB) caused by *Fusarium graminearum* Schwabe [teleomorph *Gibberella zeae* (Schwein.:Fr.) Petch] is a periodic disease which can be devastating due to disease losses and to the presence of its mycotoxin, deoxynivalenol (DON) that poses a significant threat to human health. Hard winter wheat is used roughly equally in domestic and export markets. Furthermore, bread wheat grain is increasingly used in whole wheat products. Hence our goal is to reduce the level of DON in the whole grain (as opposed to white flour) to the stricter European standard for DON in white flour. Of the two, disease losses or presence of DON, we have discovered lines (e.g. Harry) that have higher levels of DON at the same levels of disease as lower DON lines. Hence DON is the most critical measure of the lines' tolerance to FHB and its economic effects.

The Great Plains breeding programs have released lines with superior FHB tolerance (Lyman, Overland, Everest, Hitch, Art, T158, and Millennium). All of these lines are based upon native resistance. Building upon this native resistance, hundreds of crosses have been made to putative native resistance germplasm sources from other regions. Few of these crosses have led to new cultivars. Part of the low success rate may be due to the length of time it takes to release a new cultivar (12 years), but advanced experimental lines should be under testing by now. A more likely reason for the low success rate is that the germplasm was too diverse (hard to find and use the minor genes) in crosses and that our field assays were unable to separate small differences in segregating populations. Hence a new approach is needed. What has surprised us is that despite the success of *Fhb1* cultivars in the spring wheat region, relatively few have been released in the winter wheat region. A question was: does *Fhb1* or other major QTL have a negative effect on winter wheat or our selection process? In replicated studies in Nebraska using lines with and without *Fhb1* derived from a segregating population, little difference was found among the lines. This indicates that *Fhb1* does not have pleiotropic effects on agronomic performance. Furthermore a series of backcross derived lines for Wesley *Fhb1* have performed well in the field. The Wesley *Fhb1* lines have been used extensively as parents and their progeny are advancing through our selection process at a much higher rate than lines from unadapted native resistance sources. Furthermore, new backcrossing efforts are underway to put *Fhb1* into backgrounds like Overland that have native resistance in hopes the QTL and native resistance can be combined. In evaluating where we have been and how to go forward, we continue to believe in having outstanding field screens to identify native resistance and validate QTL resistance, coupling native resistance with proven FHB QTL such as *Fhb1* and newer QTL coming out of wild species (e.g. *Fhb3*) using molecular markers, and freely sharing the germplasm. The new QTL will be backcrossed in adapted germplasm to build new parent stocks. Hard winter wheat x hard spring wheat crosses and backcrosses to winter wheat

lines will continue to be used as we can recapture the needed winter adaptation relatively easily. In the future with the advent of hybrid wheat, the opportunity to study novel cytoplasm and floral biology may provide new ways of reducing FHB and DON in wheat.

Finally, we recognize that genetics can only take you so far and that a robust fungicide and crop management program is necessary to meet our goals of low DON whole grain flour.

## UNIVERSITY/INDUSTRY COLLABORATIONS: POTENTIAL WAYS TO FURTHER SCAB RESISTANCE RESEARCH

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### **ABSTRACT**

State and federal funding in the US for research into host plant resistance to scab is often limited due to budget constraints, changing budget priorities, and ever-increasing competition for state and federal funding. An excellent way to address this problem is through the formation of university/industry collaborations that leverage existing funding as well as the shared resources that are expected to be available through such collaborations. Potential research areas and types of collaborations as well as federal programs designed to foster collaboration will be discussed. Opportunities for collaborations exist across the breadth of research areas traditionally supported by the USWBSI, including educating and training the next-generation of geneticists, breeders, and pathologists to conduct scab research; identifying new host plant resistance genes; defining host genetic resistance mechanisms, host-pathogen interactions, and disease epidemiology; cloning host plant resistance genes, developing acceptable and safe transgenic approaches to host plant resistance; optimizing genetic and fungicide management practices; and limiting the production of grain toxins to safeguard the US food and feed supply. The former NIFA Plant Breeding and Education Program, a NSF Industry/University Cooperative Research Center (IUCRC) Program, and operational individual university/industry student fellowship and internship programs can serve as models to follow in furthering future scab research through collaboration. The establishment of the first IUCRC to focus on wheat genetic research was recently announced by Kansas State University and its partners. There are exciting possibilities to extend this type of IUCRC to develop collaborative projects and mitigate problems associated with scab all the way from cereal seed production, through feed and food production in the US.

ASSOCIATION OF THE EXTENT OF ANTHHER EXTRUSION  
AFTER FLOWERING WITH FIELD RESISTANCE  
TO FUSARIUM HEAD BLIGHT IN WHEAT

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**ABSTRACT**

Inspired by results from Norwegian colleagues (Skinnes et al 2008, 2011, Lu et al. 2013) we assessed several wheat populations that have also been tested for field resistance to Fusarium head blight for the extent of anther retention. FHB resistance was measured in artificially spray inoculated field trials using established methods (Buerstmayr et al. 2003). The extent of anther retention was assessed by counting the number of florets with trapped anthers among 20 florets per lot 4-6 days after pollen shed and expressed on a relative scale.

Across all populations we measured so far we found the following pattern:

- 1) The extent of anther extrusion is highly heritable, broad sense heritability was generally  $H > 0.80$ ; and
- 2) the extent of anther extrusion was significantly correlated with FHB severity, in the range of  $r > 0.60$ .

We also analysed one mapping population (Capo x Arina, 190 RILs). In this mapping population three highly significant FHB severity QTL were detected ( $LOD > 3.5$ ), two of which co-mapped with strong QTL for anther retention ( $LOD > 7$ ). These results confirm that the extent of anther retention has a pronounced influence on FHB severity under field conditions. QTL for anther retention/anther extrusion are likely at the same time passive FHB resistance QTL.

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## EFFECTS OF *FHB1* ON RESISTANCE TO WHEAT FHB IN DIFFERENT HARD WINTER WHEATS

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### ABSTRACT

Fusarium head blight (FHB) is a devastating disease that significantly reduces grain yield and quality. *Fhb1*, a quantitative trait locus on 3BS from Sumai 3, has shown the largest effect on FHB resistance. Ning 7840 is a Chinese wheat line that carries *Fhb1* from Sumai 3. Because of so many undesired traits in the Chinese lines, direct use of Chinese lines as resistant parents in U.S. hard winter wheat (HWW) breeding has not been successful. To date, none of the U.S. HWW cultivars in the Great Plains has *Fhb1*. Transferring *Fhb1* into adapted U.S. HWW cultivars may facilitate utilization of *Fhb1* to improve FHB resistance of U.S. HWW. In this study, we first transferred *Fhb1* from Ning7840 to three locally adapted HWW cultivars, Overland, Jagger, and Overley, by marker-assisted backcross, and assessed the effect of *Fhb1* on FHB resistance in these U.S HWW backgrounds. Among the three recurrent parents, Overland is moderately resistant to moderately susceptible, thus may have some indigenous minor QTL for resistance, while Overley is highly susceptible and has no any resistance QTL. Jagger is in between. A total of 227 BC<sub>3</sub> and BC<sub>4</sub> families with *Fhb1* were selected from the three backcross populations for phenotyping and marker confirmation. Both markers *Xgwm533* and *Xumn10* were used to identify *Fhb1*. FHB resistance as measured by percentage of symptomatic spikelets (PSS) in an inoculated spike was evaluated in four greenhouse experiments (spring and fall of 2011 and 2012) and one field experiment (2012-2013). *Fusarium* damaged kernel (FDK) was also scored in the field experiment. Mean PSS was highly correlated with FDK ( $r = 0.6038$ ). Mean FHB ratings were significantly different among the three populations. Overland-*Fhb1* lines had the lowest mean PSS (25.6%), ranging from 11.05 to 67.6%; Overley-*Fhb1* lines had the highest mean PSS (44.9%), ranging from 23.6 to 82.0%; and mean PSS of Jagger-*Fhb1* lines was in between (39.5%), ranging from 18.8 to 72.1%. The results indicated that the resistance levels of the recurrent parents had a large impact on the resistance of their progenies. *Fhb1* can significantly lower mean PSSs and FDK in the *Fhb1*-carrying lines selected from each population. On an average, these backcross-derived *Fhb1* lines had 7.1 to 45.9% reduction in PSS and 8.5 to 39.2% reduction in FDK compared with the lines without *Fhb1*. A combination of local minor resistance genes with *Fhb1* can significantly improve FHB resistance in U.S. HWW.

### ACKNOWLEDGEMENT AND DISCLAIMER

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## MASS SELECTION WITH OPTICAL SORTERS FOR HEAD SCAB RESISTANCE IN SOFT RED WINTER WHEAT

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### ABSTRACT

*Fusarium* head blight (FHB) or head scab, caused by *Fusarium graminearum* Schwabe [teleomorph *Gibberella zeae* (Schwein.:Fr.) Petch], is one of the most destructive diseases of wheat (*Triticum aestivum* L.) worldwide. Two important consequences of scab are a reduction in grain quality and the presence of mycotoxins. The most common mycotoxin is deoxynivalenol (DON). There is great interest among breeders in selecting for resistance to both of these traits and numerous strategies for scab resistance breeding are in use. We have tested mass selection during advancement of breeding populations using a USDA/ARS and National Manufacturing Seed Sorter System with color camera using calibrations that reflected visual differences between asymptomatic grain and grain showing FHB symptoms. In 2010, 20 bulk F<sub>3</sub> SRW wheat populations with scab resistant parents in their pedigrees were harvested by population from unreplicated plots naturally infected by a mild-moderate scab epidemic near Lexington, KY. Using this seed source the first cycle of selection was conducted by running bulk grain through the sorter and discarding rejected scabby grain. This process was repeated in 2011 using grain from C<sub>1</sub> plots that had conidial suspension applied at anthesis. In 2012, an additional cycle of selection was conducted using grain from unreplicated C<sub>2</sub> plots that were inoculated with grain spawn and sprayed with conidial suspension. In 2013, C<sub>0</sub>, C<sub>1</sub>, C<sub>2</sub> and C<sub>3</sub> selection cycles of the 20 populations, planted in a RCB experiment, in plots at Lexington and Princeton, KY and in the scab nursery at Lexington, KY, were evaluated for *Fusarium* damaged kernels (FDK) and DON concentration. Overall, no significant differences were seen between the cycles of selection. However, additional RCB studies comparing rows seeded with accepted and rejected grain separated by image-based, LED and SKNIR seed sorters were also grown in the 2013 Lexington scab nursery. Rows seeded with grain accepted by all three sorters showed reduced levels of FDK and DON compared to those seeded with rejected grain. Differences in FDK were significant ( $P < 0.005$ ) with the SKNIR sorter study. Significant differences were also seen for DON with the SKNIR ( $P < 0.05$ ) and LED ( $P < 0.1$ ) sorted material. Differences between the sorters could be due to dissimilar calibrations as well as the machines *per se*. The ability of seed sorters to improve genetic resistance to FHB is continuing to be investigated.

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## MAPPING WHEAT FUSARIUM HEAD BLIGHT RESISTANCE QTL IN THE MD01W233-06-1/SS8641 DOUBLED HAPLOID MAPPING POPULATION

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### ABSTRACT

Wheat breeding for Fusarium head blight (FHB) resistance relies on identifying and selecting for favorable quantitative trait loci (QTL). While major QTL have been identified in exotic germplasm (*Fhb1*), identifying QTL in US wheat varieties (US native resistance) is an important research area, as these are easily adaptable in breeding programs and can augment progress made with exotic material. The germplasm line MD01W233-06-1 was shown to be resistant in the 2007 and 2008 Southern and 2008 Northern Uniform Winter Wheat Scab Nurseries. It was then crossed with SS8641, a highly FHB-susceptible variety to create an F1-derived doubled haploid population with 124 lines. This population and the parents were evaluated in a single floret inoculation experiment in a greenhouse in College Park, MD in the winter of 2011 and in inoculated scab nursery field experiments in Salisbury, MD and Kinston, NC in both 2011 and 2012. Lines were phenotyped for heading date, FHB incidence (INC), FHB severity (SEV), percentage of *Fusarium*-damaged kernels (FDK), and deoxynivalenol (DON) content. The population and parents were genotyped using a variety of molecular markers including: red coleoptile morphological marker, 29 SSR microsatellite markers, 23 KASP SNP markers, and 9K iSelect Beadchip Assay. After eliminating monomorphic and markers with segregation distortion, linkage analysis was performed using QTLICIMapping v. 3.3 with 450 markers. Markers were anchored to wheat chromosomes based on data from Cavanagh et al. (2013), Somers et al. (2004), and Wilkinson et al. (2012). Linkage analysis was performed using a LOD threshold of 5.0 for linkage group construction, with the RECORD algorithm and SARF criterion for ordering and rippling, respectively. Markers mapped to 21 linkage groups, with at least 1 marker on each chromosome of the wheat genome. Phenotypic data was analyzed by location-year using PROC GLM of SAS to determine least square means for each line. QTL analysis was performed using the inclusive composite interval mapping method, with LOD threshold for significance determined by permutation test (1000 permutations, p=0.05). A total of 43 significant QTL were identified. Consistent resistance QTL were found on 2DS from 47- 64cM (2 DON and 1 FDK) and 2DL from 95-104cM (2 DON, 2INC, 2FDK, 1 SEV). The 2DS QTL were coincident with QTL for heading date, and correlation analysis showed that the resistance trait data were highly correlated with heading date, thus these resistance QTL may simply be due to disease escape. No heading date QTL mapped to the 2DL region with other resistance QTL. Additionally, several resistance QTL were identified from SS8641, where the MD01W233-06-1 alleles had a higher additive value. There were 3 such QTL regions on 3B from 56-57cM (2 DON), 75-78cM (2 FDK; 1 DON, SEV, and INC) and 93-113cM (3 SEV, 3 FDK, and 1 DON); on 1A at 129cM (2DON and 1 INC).

CONSTRUCTION OF DENSE LINKAGE MAPS “ON THE FLY” USING EARLY GENERATION PLANT BREEDING POPULATIONS TO FACILITATE MAPPING OF FUSARIUM HEAD BLIGHT RESISTANCE QTL

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**ABSTRACT**

Genetic linkage maps, consisting of marker loci ordered along chromosomes, provide the essential framework for identifying genomic regions contributing to Fusarium head blight (FHB) resistance and supporting molecular breeding efforts for varietal development. Linkage mapping has largely been confined to purpose-built mapping populations derived from a single cross. Construction of linkage maps with dense marker coverage further requires the integration of maps from several experimental populations, thus detracting from breeding efforts and delaying the application of new molecular tools. To facilitate marker assisted breeding efforts to pyramid FHB resistance QTL in our wheat breeding populations using the newly available wheat 9,000 SNP iSelect assay, as well as 26 SSR markers, we needed to construct a de novo linkage map. For this purpose, we applied multipoint linkage analysis of general pedigrees to develop a dense linkage map using our existing breeding populations consisting of 565 four-way F<sub>1</sub> plants from 28 four-way crosses. Linkage analysis was performed using the CRI-MAP version 2.504. A total of 3,880 loci were mapped, including 1,252 unique genetic bins. The estimated linkage maps covered a total genetic distance of 3,072 cM, with an average interval of 2.5 cM between genetic bins. Marker coverage was relatively poor for the D genome. Within the A and B genomes, 12 of the 14 linkage maps had rank-order correlations 0.97 to 0.99 with the locus positions in the consensus maps released for the 9,000 SNP assay. The high degree of concordance with the consensus map indicates that use of mapping algorithms for general pedigrees can be reliably adopted to develop dense linkage maps using existing segregating breeding populations. This strategy should allow researchers and breeders to develop dense linkage maps “on the fly” to accelerate marker assisted breeding efforts for host resistance to FHB.

## MULTIPLE FUSARIUM HEAD BLIGHT RESISTANCE QTL PYRAMIDED ONTO ELITE SPRING WHEAT *Fhb1* BACK- GROUNDS USING A FAMILY-BASED MAPPING APPROACH

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### ABSTRACT

Resistance to Fusarium head blight (FHB) in wheat is a complex trait, for which numerous QTL have been identified. While use of *Fhb1* for development of host resistance is commonplace in wheat breeding programs, smaller effect QTL have not been routinely exploited. Pyramiding novel sources of resistance with *Fhb1* therefore presents an opportunity to enhance FHB resistance of elite wheat germplasm. It was demonstrated that family-based linkage analysis has potential for mapping FHB resistance QTL in early generation breeding populations. Therefore, we have applied a family-based mapping approach in our breeding populations to map and pyramid multiple FHB resistance QTL onto elite hard red spring wheat (HRSW) *Fhb1* backgrounds. Segregating F<sub>1</sub> populations were developed from 44 four-way crosses among 20 HRSW founder lines, providing a total of 826 four-way F<sub>1</sub> plants. Fifteen experimental lines from the SDSU spring wheat breeding program provided the elite *Fhb1* backgrounds. Founders conferring novel sources of resistance were 2 experimental lines (MN99112 and MN99126) from the UMN spring wheat breeding program, 2 lines from a cross between 'Wheaton' and Japanese landrace PI 81791 (Sapporo Haru Komugi Jugo), and Peruvian line PI 271127 (MULT 757). Founders and four-way F<sub>1</sub> plants were genotyped for 72 SSR markers and phenotyped by spray inoculation in the greenhouse for FHB severity. F<sub>2</sub> seed collected from each four-way F<sub>1</sub> plant was used to establish selfed progeny tests of the four-way F<sub>1</sub> plants over multiple sites. Family-based linkage analysis was conducted using the software package S.A.G.E to identify FHB resistance QTL segregating in the four-way F<sub>1</sub> populations. Linkage analysis detected a QTL on the short arm of chromosome 3B corresponding with *Fhb1* in the interval between *Xbarc133* and *Xgwm493*, which was most strongly associated with *Xbarc147*. A centromeric QTL was also identified on chromosome 3B in the interval between *Xwmc787* and *Xgwm108*, roughly corresponding to a QTL reported from PI 81791. A third QTL was mapped to chromosome 7B associated with *Xbarc176*, which corresponds to a QTL reported from PI 271127. These markers are being employed to select F<sub>4.5</sub> lines from these populations with desirable QTL combinations to move forward in the breeding program.

NATIVE FUSARIUM HEAD BLIGHT RESISTANCE FROM 'LYMAN',  
'OVERLAND', 'ERNIE' AND 'FREEDOM' WHEAT CULTIVARS  
PYRAMIDED ONTO WESLEY-*FHB1* BACKGROUNDS  
USING A FAMILY-BASED MAPPING APPROACH

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**ABSTRACT**

The primary strategy for development of host resistance to Fusarium head blight (FHB) in winter wheat has been the backcrossing of *Fhb1* into elite genetic backgrounds. However, there are numerous sources of native FHB resistance in winter wheat that could be combined with *Fhb1* to produce varieties with enhanced resistance. Therefore, we have adopted a family based mapping approach to map and pyramid multiple native sources of FHB resistance onto a Wesley *Fhb1* background. Segregating F<sub>1</sub> populations were developed from 28 four-way crosses among 10 winter wheat founder lines, providing a total of 565 four-way F<sub>1</sub> plants. Founders included two Wesley *Fhb1* backcross lines (WesFHB1-BC06 and WesFHB1-BC56) that provided the *Fhb1* background for each cross. Founders conferring native sources of resistance were the hard winter wheat cultivars 'Lyman' and 'Overland', and the soft winter wheat cultivars 'Ernie' and 'Freedom.' Cultivars 'McGill', NE0645 and NI08708 were also used as founders. Founders and four-way F<sub>1</sub> plants were phenotyped in the greenhouse for FHB severity, and genotyped for 9,000 SNPs (Illumina iSelect Beadchip assay) and 26 SSR loci. F<sub>2</sub> seed collected from each four-way F<sub>1</sub> plant was used to establish selfed progeny tests of the four-way F<sub>1</sub> plants over multiple sites. A total of 39 F<sub>2</sub> lines have been selected based on phenotypic evaluations and used to derive approximately 500 F<sub>2,3</sub> lines, while 232 lines have been advanced as F<sub>3</sub> bulks for further evaluation. Family-based linkage analysis is currently being conducted to identify QTL and associated markers for FHB resistance segregating in these breeding populations. These markers will be used to select lines with desirable combinations of resistance QTL.

## MOLECULAR MAPPING OF FUSARIUM HEAD BLIGHT RESISTANCE IN GLENN, A HIGH QUALITY AND ADAPTED HARD RED SPRING WHEAT CULTIVAR

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### ABSTRACT

Fusarium head blight (FHB) is a major disease that affects both wheat yield and quality in many wheat-growing regions, including the US Northern Plains. Therefore, developing wheat cultivars with high resistance to FHB is necessary. In the last few years, North Dakota State University's spring wheat breeding program has released several FHB-resistant cultivars including high yielding and quality 'Glenn'. Based on its pedigree, Glenn resistance is supposed to be from 'Sumai3'. However, molecular analysis showed that Glenn does not possess any molecular markers associated with Sumai3-*Fhb1* locus, including umn10. Therefore, this study aimed to decipher the genetics of FHB-resistance in Glenn. To achieve this goal, a (GM) RIL population was developed from a cross between Glenn and the moderately susceptible line MN00261-4. The RIL population and parents were evaluated for different FHB traits including incidence (INC), severity (SEV), and deoxynivalenol (DON); and some agronomic traits, including height (HT). The experiments were conducted under field and greenhouse (GH) conditions in ND, and MN using artificial inoculation. Additionally, the GM population was genotyped using Diversity Array Technology (DArT). A linkage map was generated and used to identify QTL associated with FHB traits. A total of 15 QTL for SEV, 15 for INC, 7 for DON, and 8 for HT were identified. One major QTL (5B) for SEV was detected in four environments. Six QTL (1B, 5B,

6B, 7A, and 7D) for SEV and one QTL for INC (1A) were detected in two environments. The study did not identify any consistent QTL on chromosome 3BS.

### OBJECTIVES

1. Identify the genetic source of resistance in Glenn.
2. Determine if Glenn contains *Fhb1*.

### INTRODUCTION

FHB disease in the upper Midwest region of the US (ND and MN) led to hundreds of millions of dollars in losses for farmers since 1993 (Nganje *et al.*, 2001). In year 2000, \$160 million losses were recorded; reflecting the importance and need for releasing resistant varieties. Most famous markers/QTL used today in wheat breeding programs are those located on chromosomes 3BS (*Fhb1*), 5AS, and 6BS (*Fhb2*). The highest magnitude of a QTL identified to date is *Fhb1* in Sumai3. This QTL can reduce the disease on average, depending on the genetic background, by 20–25% (Pumphrey *et al.*, 2007). However, there is evidence that combining major QTLs can pyramid FHB resistance levels (Miedaner *et al.*, 2006). Recently, Liu *et al.*, (2008) identified another effective marker (*umn10*) with close linkage to the 3BS *Fhb1* locus. This new marker was widely used in many breeding programs including HRSW breeding program at NDSU, ND, USA.

Glenn, a 2005 NDSU release, developed using classical breeding methods, has very high quality, excellent agronomic performance and high levels of FHB resistance (Mergoum et al., 2006). Based on its pedigree, Glenn resistance is supposed to be from 'Sumai3'. However, molecular analysis showed that Glenn does not possess any molecular markers associated with Sumai3-*Fhb1* locus (including the Umn10).

## MATERIALS AND METHODS

*Population:* A RIL population was developed (Glenn/MN00261-4 (GM) from a cross between Glenn and the moderately susceptible line MN00261-4. The population and parent were evaluated for FHB in Prosper and Carrington (ND), MN and GH. The field experiments were inoculated artificially using dispersed diseased-kernels method; and the spore-suspension inoculation method was used in GH. All experiments were laid out in randomized complete block design (RCBD) with four replicates in ND and with two replicates in MN.

*Phenotyping and Genotyping:* Data collected for heading dates (HD), plant HT, and FHB SEV, INC, and DON. The genomic DNA was extracted from lyophilized tissue of young leaves using Qiagen DNeasy Plant mini kit (Cat# 69106) with some modifications. For each genotype 30 µl of DNA (80 ng/µl) was sent to Triticate Pty .Ltd (Canberra, Australia; <http://www.triticate.com.au>) for Diversity Array Technology (DArT) analysis (Akbari et al., 2006).

*Map construction and QTL Analysis:* all polymorphic DArT markers were converted into genotype codes according to the parental scores. Linkage maps for each chromosome were constructed using MAPMAKER v. 2.0 (Lander et al., 1987). Single-locus QTL analysis was carried out by composite interval mapping (CIM) using QTL CARTOGRAPHER v. 2.5 (Wang et al., 2007) to identify the main effect QTL for each genotype.

## RESULTS AND DISCUSSION

*QTL analysis for FHB SEV and INC:* CIM identified a total of 16 QTL for FHB severity from 6 environments (Table 1). The 16 QTL were located on 10 different chromosomes, with chromosomes 2D, 4B, 6A, 6B, and 7A carrying 1 QTL each, chromosomes 1B, 2B, 3B, and 7D carrying 2 QTL each and chromosome 5B carrying 3 QTL. Among all these QTL, 1 QTL (located on 5B) was detected in 4 environments; 6 QTL (located on 1B, 5B, 6B, 7A, 7D) were detected in 2 environments each, while the remaining 9 QTL (located on 1B, 2B, 2D, 3B, 4B, 5B, 6A) could be detected in only 1 environment. The phenotypic variation (PV) explained by individual QTL ranged from 6.4-20.5%. Among the 16 QTL, 8 showed major effects on FHB severity (PV>10%), while the remaining 6 had minor effects (PV<10%). The positive alleles for increased resistance at eight loci each were contributed by both parental genotypes. A total of 15 QTL for FHB incidence located on 10 different chromosomes (1A, 1B, 2D, 3B, 4A, 4B, 5B, 6A, 7A and 7D). The number of QTL identified in individual environments ranged from 2 (Prosper 2011) to 7 (MN 2010). Only 1 QTL for incidence (located on 1A) could be detected in 2 environments, while remaining QTL were detected in only 1 environment. The PV explained by individual QTL ranged from 4.94-16.34% (Table 1). Three QTL showed major effects, while the remaining 12 had minor effects. The alleles from Glenn contributed towards increased resistance at 8 loci and alleles from MN contributing towards increased resistance at the remaining 7 loci.

*QTL Analysis for DON and plant HT:* CIM identified a total of 7 QTL for DON; 5 in Prosper-2010 and 2 in Prosper-2011. The QTL were located on 6 different chromosomes; 2 on 2B and 1 each on 1A, 3B, 4A, 5B, and 7B. PV explained by individual QTL ranged from 7.26 to 13.88%. Four QTL had major effect of DON, while 3 showed minor effects. QTL alleles for increased DON at 4 loci were contributed by MN, while Glenn contributed alleles for the remaining 3 loci. A total of 8 QTL for HT located on 7 chromosomes, 2 on 7A and 1 each on 1A, 1B, 2A, 2B, 5B and 6A. PV explained by individual QTL ranged from 5.35-18.65%. Three QTL had major effect of HT, while 5

showed minor effects. The alleles for increased HT were contributed by both parents as 4 loci each.

*Genome A:* The QTL we identified on chromosomes 1A and 4A were associated with Type III (DON) FHB resistance while those located on 2A and 6A were associated with HT. QTL on 7A however, was associated with all above traits (Table 1). In previous studies using meta-QTL analysis (Mao et al., 2010) and 16 different FHB resistance sources including Sumai3, the QTL confirmed on 3A and 5A related to reduction in SEV was associated with HT allele *Rht*. Li et al., (2011) described a QTL on 1A, and subsequently in 2012 another QTL on 7A, both from ‘Haiyanzhong’ Chinese cultivar. However, a QTL on 7A was reported by Jayatilake et al. (2011) but from Sumai3 and for Type III resistance rather. In our study, the identified QTL on 7A was associated with INC (type I), and SEV (Type II), and HT, not with Type III (DON).

*Genome B:* Our study identified many QTL on B chromosomes (Table 1). These include 2B, 3B and 5B (DON); 1B, 2B, and 5B (HT); 1B, 3B, 4B, and 5B (INC); 1B, 2B, 3B, 4B, 5B, and 6B (SEV). Most QTL were confirmed to be from a source different from Sumai3. For instance, previous studies did not identify QTL on 1B that was associated with type SEV, INC and HT. The only report (Xu et al., (2001) that indicated a QTL on 1B was identified using a double haploid population of Sumai3, but was associated only with type II (SEV). Another study (Srinivasachary et al., 2008) identified 7 QTL including one on 1B in a Canadian hard red spring wheat cultivar derived from ‘Frontana’. Likewise, we believe that the QTL on 5B we identified in our study is novel. The only 5B QTL identified previously (Tamburic-Ilincic et al., 2009) was for resistance to *Fusarium* seedling blight in ‘Wuhan’/‘Nyubai’ population. Similarly, our study confirmed that the QTL on 7B was novel and associated with low DON. Though many studies has intensively studied QTL on 3B and 6B, our study has identified new QTL on 3B and 6B (Fig.1) different from than those well-known (*fhb1* and *fhb2*) derived from Sumai3 (Patricia et al., 2006; and 2007).

*Genome D:* We identified QTL in our study on 2D, 7D (Table 1) that were associated with Type I and II FHB

resistance. Previously, an association between a QTL on 2D and a HT gene (*Rht8c*) from ‘Aka Komugi’, a gibberellic acid GA-sensitive (J. Gilbert. and S. Haber, 2013) was established. The locus (*Rht-D1*) was reported to explain 38% of the PV for FHB Type I resistance with no effect on Type II (Lu et al., 2011). This infers that our QTL on 2D different from that identified by Lu et al., (2011). Additionally, another QTL from Sumai3 identified on 2D and associated with reduced kernel weight was also reported (Suzuki et al, 2012). However, Glenn kernel weight is high confirming that 2D FHB resistance in Glenn is may not be based on Sumai3.

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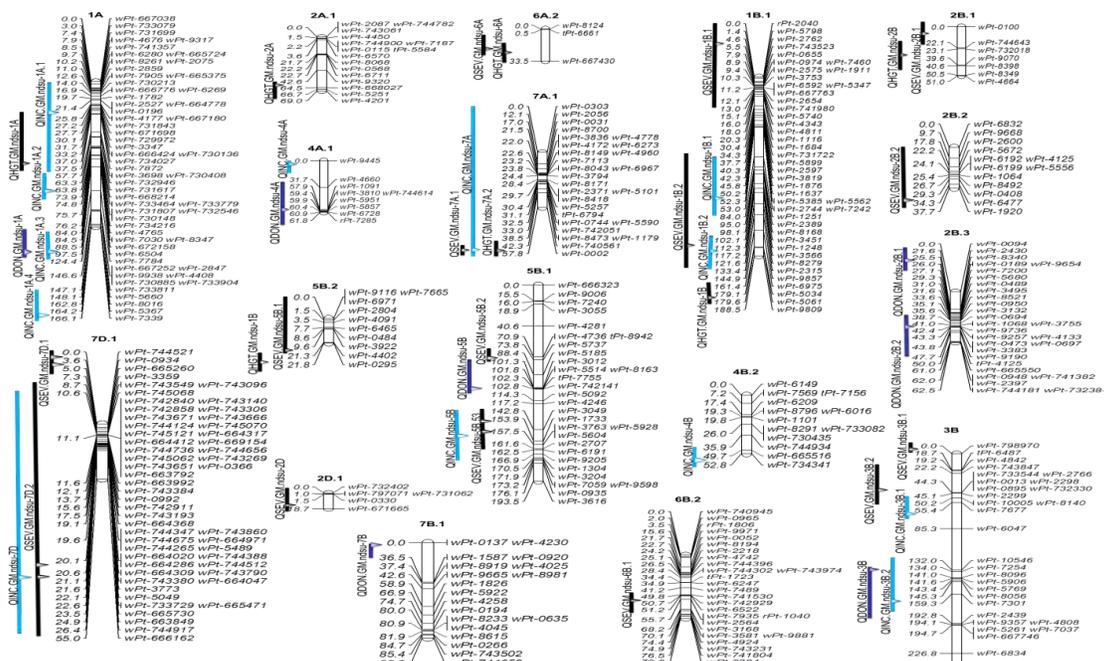
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**Fig.1.** Chromosomal map for QTL identified in (GM) population in different environments. QTL intervals shown as (lines); QTL positions shown as (Triangles); QTL for SEV, INC, DON, and HT were shown in (Black, Blue, Brown, and Purple) respectively.

**Table 1.** Important QTL repeated in more than 2 environments for SEV, INC, DON and HT.

QTL	Chrom. (group)	Environments*	LOD	QTL effect	R <sup>2</sup> (%)	Other associated traits
<b>FHB severity (SEV)</b>						
QSEV.GM.ndsu-1B.2	1B.1	Pros-2010, MN-2010	3.21	3.44	10.26	INC
QSEV.GM.ndsu-2B.2	2B.2	Pros-2011	5.79	3.27	17.12	
QSEV.GM.ndsu-2D	2D.1	Pros-2012	3.7	-4.74	13.78	
QSEV.GM.ndsu-5B.2	5B.1	Carr-2011, Pros-2012	4.09	-4.47	12.09	DON
QSEV.GM.ndsu-5B.3	5B.1	GH-2011, Pros-2010, Pros-2012, Carr-2011	7.86	6.21	20.49	INC
QSEV.GM.ndsu-6B.1	6B.2	GH-2011, Pros-2011	4.29	-6.29	12.1	
QSEV.GM.ndsu-7A.1	7A.1	Carr-2011, MN-2010	2.6	-3.09	8.61	INC, HGT
QSEV.GM.ndsu-7D.1	7D.1	MN-2010, Pros-2010	2.86	7.23	10.44	
QSEV.GM.ndsu-7D.2	7D.1	Pros-2010, MN-2010	4.04	-8.27	12.39	INC
<b>FHB incidence (INC)</b>						
QINC.GM.ndsu-1A.1	1A	MN-2010	3.37	-4.1	9.03	HGT
QINC.GM.ndsu-3B.2	3B	MN-2010	2.04	-2.95	4.94	DON
QINC.GM.ndsu-7A	7A.1	MN-2010	2	-3.05	5.17	SEV-HGT
QINC.GM.ndsu-7D	7D.1	MN-2010	5.58	-16.59	16.34	SEV
QINC.GM.ndsu-1B.2	1B.1	Carr-2011	2.72	-1.56	10.12	Sev
QINC.GM.ndsu-4B	4B.2	Pros-2011	3.99	-1.22	12.82	
QINC.GM.ndsu-5B	5B.1	Pros-2011	2.3	0.84	6.9	SEV
QINC.GM.ndsu-1A.3	1A	Pros-2012	3.05	-3.7	8.32	DON
QINC.GM.ndsu-1B.1	1B.1	Pros-2012	3.27	3.65	8.9	Sev
QINC.GM.ndsu-3B.1	3B	Pros-2012	3.24	3.81	8.96	Sev
<b>Deoxynivalenol (DON)</b>						
QDON.GM.ndsu-1A	1A	Pros-2010	2.4	-0.09	10.19	INC
QDON.GM.ndsu-2B.2	2B.3	Pros-2010	4.52	-0.17	13.88	
QDON.GM.ndsu-5B	5B.1	Pros-2010	3.24	-0.1	12.61	SEV
QDON.GM.ndsu-7B	7B.1	Pros-2010	3.93	0.1	12.07	
QDON.GM.ndsu-3B	3B	Pros-2011	2.61	-0.34	9.06	INC
<b>Plant height (HGT)</b>						
QHGT.GM.ndsu-5B	5B.2	GH-2011	3.03	-1.55	8.02	SEV
QHGT.GM.ndsu-6A	6A.2	GH-2011	4.38	-1.87	13.31	SEV
QHGT.GM.ndsu-1A	1A	GH-2012	2.12	1.42	5.89	INC
QHGT.GM.ndsu-2A	2A.1	GH-2012	4.39	2.52	18.65	
QHGT.GM.ndsu-2B	2B.1	GH-2012	3.15	-1.98	11.84	SEV-DON
QHGT.GM.ndsu-7A.2	7A.1	GH-2012	3.03	1.79	9.65	INC-SEV

\* QTL effect, indicating which parent contributes the positive or negative effect at which environment (Pros=Prosper, ND; Carr=Carrington, ND; GH=Greenhouse; MN= Minnesota State) ND; GH=Greenhouse; MN= Minnesota State)

ASSOCIATION MAPPING OF FHB RESISTANCE  
IN BARLEY UTILIZING HISTORIC NABSEN  
DATA AND GENOTYPE-BY-SEQUENCING  
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**ABSTRACT**

Fusarium head blight (FHB) is one of the most devastating diseases of barley. The most insidious part of FHB infection is the production of harmful mycotoxins, with the most important being deoxynivalenol (DON). Since DON can cause many problems for end users DON assays have become a standard specification in grain purchasing. Thus, identifying loci conferring lower DON accumulation in barley is important for marker development and breeding for resistance. A total 441 lines of elite two-rowed and six-rowed barley from The North American Barley Scab Evaluation Nursery (NABSEN) were evaluated for DON content at four sites from 2002-2012. At each site the experimental design was a randomized complete block design with three blocks. DON levels were averaged across blocks for each line at each site and square root transformed to normalize the data. These values were then used as the response variable in a mixed effects model with sites treated as fixed effects and lines as random effects. Best Linear Unbiased Predictors (BLUPs) were estimated for each line using the lme4 package in R. To utilize the BLUPs to identify DON accumulation quantitative trait loci (QTL) by association mapping with JMP genomics program, we are genotyping the entire population of 441 lines using a PCR based genotype-by-sequencing method. All barley lines are being genotyped using a 384 single nucleotide polymorphism (SNP) marker panel containing known SNP markers that are evenly distributed across the barley genome. This information will potentially allow us to identify DON accumulation QTL in elite barley germplasm from the major barley breeding programs in the Northern Great Plains.

## FHB RESISTANCE AND AGRONOMIC PERFORMANCE IN SOFT RED WINTER WHEAT

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### ABSTRACT

Local broadly adaptive cultivars have been crossed with *Fhb1* derived lines, Truman, and Jamestown to introduce FHB resistant QTL into adaptive genetic backgrounds. Elite lines with resistance from Truman, IN981359C1, or Jamestown, were evaluated in the field during 2013 for FHB resistance and agronomic performances. A double haploid line, GANC 8170, that was selected from the cross of NC-11458/Bess//SS 8641, showed a high level of FHB resistance which was similar to the resistant controls, Bess and Jamestown. The double haploid line also had low levels of resistance for FHB index, FDK, and ISK. The closely linked markers of the double haploid line were detected for the FHB resistant QTL, *Fhb\_5A\_Ning7840* and included important resistant genes for leaf and stripe rust (*Lr37/Yr17/Sr38*). In addition, an elite line, GA04151-11E26, that was evaluated in the 2013 Uniform Southern Wheat Nursery also showed a moderate level of FHB resistance and had very high grain yield. Several other lines with Jamestown, Truman, IN981359 and IN 97397 as source of resistance were identified with moderate levels 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.

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## MAPPING FHB RESISTANCE IN NATIVE SRW WHEAT CULTIVAR TRIBUTE

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### ABSTRACT

Resistance to *Fusarium* head blight (FHB) has been deployed from native and exotic sources of wheat. Native sources of resistance are important because of their efficient utilization in breeding programs without yield drag. Since resistance to FHB is governed primarily by additive genetic effects, pyramiding different sources of resistance would be an effective approach to managing FHB through resistance breeding. The objectives of the study were to identify FHB resistance QTL in the native SRW cultivar Tribute and develop diagnostic markers for use by programs in marker-assisted breeding. A total of 115 double haploid (DH) lines were evaluated for FHB incidence and FHB severity by cooperators in AR, KY, MD, NC, and VA in 2013. Grain samples from VA have been analyzed for DON content. The population was genotyped using SSR markers. The genotype-by-location interaction was significant for the population. Correlation analyses of data among locations indicate that data from AR and MD are highly correlated as were data from VA and KY. Data from NC was correlated with that from all other locations. The results from single marker analysis indicate that SSR markers on chromosomes 2A (Xgwm47), 2D (Xgwm261), 3BSc (Xgwm78, Xgwm285, Xwmc418, Xwmc471), 5A (Xwmc795), 5D (Xwmc805, Xwmc443), and 7B (Xbarc95) were significantly associated with FHB incidence and severity across different locations. The SSR marker Xgwm47 on 2A also was significantly associated with DON content. The SSR marker Xgwm261 on 2D also is linked to *Rht8* and *Ppd-D1* and therefore use of the putative QTL on 2D may not be desirable in all programs. The putative QTL on 3BSc might be in a similar region as reported for Ernie, Wangshubai, and Nyu bai. The putative QTL on 2A, 5D, and 7B may be unique to Tribute. A second year of phenotypic data and 90K SNP genotypic data will be obtained and analyzed in the population to identify and validate QTL that can be used for marker-assisted breeding.

## FUSARIUM HEAD BLIGHT RESISTANCE QTL IN THE KENYON X 86ISMN 2137 POPULATION

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### ABSTRACT

Fusarium head blight (FHB) is one of the most damaging diseases of wheat in Canada. The genetic basis of FHB resistance in Asian spring wheats has been the focus of many genetic studies. FHB resistance in Canadian spring wheat germplasm has not been studied to date. The objective of this study was to identify QTL for FHB resistance in western Canadian spring wheat with the cross Kenyon x 86ISMN 2137. Kenyon is a Canada Western Red Spring (CWRS) variety with the pedigree Neepawa\*5/Buck Manantial. Neepawa was a major CWRS variety and a parent of many current varieties. Buck Manantial was the source of the leaf rust resistance gene *Lr16* in Kenyon. The wheat line 86ISMN 2137 is of unknown origin. A RIL population from this cross was tested in six environments over 2012 and 2013 for FHB visual rating index (VRI), plant height, and anthesis date. Simple and multiple interval mapping were used for QTL analysis. The strongest FHB resistance QTL were contributed by Kenyon on chromosomes 2D and 7D. 86ISMN 2137 contributed reduced plant height and increased FHB VRI on chromosome 2D at the map location of *Rht8*. So the 2D FHB resistance QTL is likely the pleiotropic effect of *Rht8*. Kenyon also contributed a putative FHB resistance QTL on chromosome 2A. 86ISMN 2137 contributed putative FHB resistance QTL on chromosomes 2B, 4A, and 4D. The minor FHB resistance QTL on 2B mapped near *Lr16*, with *Lr16* linked in coupling with FHB susceptibility. Kenyon was consistently more susceptible to FHB than Neepawa in these field tests, which supported this finding. The QTL identified in this study provide insight into FHB resistance QTL present in Canadian spring wheat varieties.

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## COMBATING FUSARIUM HEAD BLIGHT IN THE US SPRING WHEAT REGION: 'ELGIN-ND', A NEW HARD RED SPRING WHEAT CULTIVAR WITH HIGH LEVEL OF RESISTANCE

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### OBJECTIVES

To develop new improved hard red spring wheat (HRSW) (*Triticum aestivum* L.) cultivars combining resistance to Fusarium Head Blight (FHB) disease [caused by *Fusarium graminearum* Schwabe (telomorph *Gibberella zeae* (Schwein.) Petch)] and superior grain yield and bread-making quality.

### INTRODUCTION

Hard red spring wheat is the leading crop in the Central Northern US plains. In North Dakota (ND) in particular, HRSW is the major crop with about 6 million acres annually grown with an economic value surpassing one billion dollars. The development of superior HRSW cultivars is the key element to sustain future wheat productivity and maintain economic returns to ND growers and the wheat industry as well as the wheat export market. Scab or FHB has been for decades a major constraint for wheat worldwide and in ND, causing billions of dollars in losses in productivity and quality (McMullen et al., 1997; Nganje et al., 2004). Growing adapted genetically resistant HRSW cultivars is the best strategy for an efficient, economical, and safe control of FHB disease while protecting our environment. Recently developed superior NDSU HRSW cultivars (mainly 'Glenn' (PI 639273; Mergoum et al., 2006a); 'Faller' (PI 648350; Mergoum et al., 2008); and 'Barlow' (PI 658018; Mergoum et al., 2011)) with moderate

FHB resistance are being grown on more than 50% of ND wheat acreages replacing susceptible cultivars or old very popular cultivars such as 'Alsen' (PI 615543; Frohberg et al., 2006), 1999; 'Steele-ND' (PI 634981; Mergoum et al., 2005b), 2004 and 'Howard', 2006 (PI 642367; Mergoum et al., 2006b) with the same levels of FHB resistance that dominated the spring wheat regions for years. Significant additional acreages in the neighboring states are grown to these cultivars as well. In MN for instance, Faller has been the leading cultivar since 2009 and 'Prosper' (PI 662387; Mergoum et al., 2012) was the leading in 2013 followed by Faller. The goal of the NDSU HRSW breeding program is to continue developing and releasing superior adapted cultivars to replace the susceptible cultivars grown in ND and the region.

### MATERIAL AND METHODS

'Elgin-ND' was developed using a modified bulk breeding procedure. It was selected from the "Walworth/Reeder//ND721" cross made at NDSU between 'Reeder' (PI613586), the South Dakota State University (SDSU) cultivar 'Walworth' (PI630938), and the experimental line ND721. Reeder was released by NDSU in 1999 (PVP200000211) with excellent adaptation to Western ND and Eastern MT where drought is predominant. Walworth is also a HRSW cultivar but released by SDSU in 2002 (PVP#200200108) for its high yield. ND721, is an elite HRSW line developed by NDSU from the Grandin

'/3/IAS20\*4/H567.71//'Amidon'/4/ND 674 cross. ND721 is an excellent combiner and possesses good agronomic characteristics, quality attributes and diseases resistance including FHB. Based on its pedigree, Elgin-ND resistance to FHB is not well known and could not be traced to 'Sumai 3' source.

Elgin-ND was selected from a bulk of one purified F<sub>5</sub> row-plot selected in 2002 winter nursery grown at Christchurch, NZ. Elgin-ND was first tested for agronomic and quality performance in preliminary (PYT), Intermediate (IYT), advanced (AYT) and elite yield trials (EYT) in up to four replicates at two to seven locations in ND from 2003 to 2008. From 2009 to 2012, Elgin-ND was tested as ND 818 at 22 location-years in the North Dakota Variety Trials (NDVT).

Screening of Elgin-ND for FHB was conducted from 2009 to 2011 in 8 FHB nurseries under field conditions. The FHB nursery was conducted at Prosper and Langdon, ND. The experiments were laid out in a RCBD with four replicates and inoculated with the FHB pathogen using the "Spray Inoculation Method" with overhead mist irrigation to enhance disease development. Entries were assigned to a hill plot consisting of eight to ten plants. Ten spikes from each hill were taken at random and evaluated for FHB disease incidence and severity (Stack and Frohberg, 1997).

The reaction type of Elgin-ND to the prevalent races of leaf rust (caused by *Puccinia triticina* Eriks.) was done on the basis of six field tests (RCBD, four replicates, and 1 m row-plot per replicate) and four greenhouse tests (RCBD, three replicates, and four plants per replicate) from 2009 to 2011. Similarly, Elgin was screened for major races of stem rust (caused by *Puccinia graminis* Per.:Pers. f. sp. *tritici* Eriks. & E. Henn) in 9 field and 3 greenhouse experiments. In the greenhouse experiments, Elgin-ND was specifically evaluated for resistance to the predominant stem rust pathotypes Pgt-TMLK, -QTHJ, -QFCQ, -RTQQ, -TPMK, -THTS, and -TCMJ; and leaf rust pathotypes MCDL and THBJ.

## RESULTS

Elgin-ND was tested under experimental line ND 818 and was released because it combines very high yield and agronomic traits (Table 1), resistance to FHB and leaf diseases (Table 2), and excellent end-use quality (Table 1 and 3), particularly, grain protein (Table 1).

Based on 22 site-years of testing in the NDVT and AYT, grain yield of Elgin-ND (4001 kg ha<sup>-1</sup>) was significantly higher than most released cultivars by NDSU, SDSU, the U of MN except for Barlow (3913 kg ha<sup>-1</sup>), Faller (3957 kg ha<sup>-1</sup>), and Prosper (3973 kg ha<sup>-1</sup>). These three cultivars are all released by NDSU and are the leading cultivars in ND and MN. Elgin-ND is a semi-dwarf cultivar and tends to head later than most cultivars in general, the same as Faller and Prosper.

Under severe FHB disease pressure, the average disease severity (Stack and Frohberg, 2000) recorded on Elgin-ND from the field scab nursery (28%) was lower than the very susceptible check '2398' (73%) (Table 2). In the same trials, the average FHB severities recorded for Barlow, Faller, Glenn, and ND 2710 (PI 633976) were 25, 27, 25, and 13%, respectively. Glenn is an NDSU HRSW released cultivar with FHB resistance and was the leading cultivar in ND and the spring wheat region from 2007 to 2011. Similarly, Barlow (also an NDSU release) became the most popular cultivar in ND since 2012. Based on natural FHB inoculation, visually scabby kernels of Elgin-ND (0.6%) was low compared to the susceptible checks 'Velva' (2.4%) (Mergoum et al., 2013) (Table. 1).

The seedling and adult plant screening tests conducted under greenhouse and field conditions from 2009-2011 showed that Elgin-ND possesses high level of resistance to the predominant races of leaf and stem rusts. Under greenhouse tests, Elgin-ND was resistant to pathotypes MCDL and THBJ, the predominant race of leaf rust in the region (Table 2). However, Elgin-ND seems to be medium susceptible to the new race of leaf rust that has overcome the *Lr21* gene. Elgin-ND was

also highly resistant to stem rust pathotypes Pgt TPMK, TMLK, RTQQ, QFCQ, QTHJ, THTS, and TCMJ (Table 2).

Quality parameter including Falling number (FN), Flour extraction (FE), dough and baking parameters for Elgin-ND and major grown NDSU HRSW cultivars are reported in Table 3. Mean FN and FE, mixing time and tolerance of Elgin-ND over 20 location-years in NDVT from 2009-2011 were respectively, 406 s, 68.2 gkg<sup>-1</sup>, 7.8 min, and 11 min compared to 357 s, 68.2 gkg<sup>-1</sup>, 10 min, and 15 min registered for Glenn (our best quality check); and 388 s, 70.4 gkg<sup>-1</sup>, 7 min, and 10 min for Barlow, the most popular cultivar in ND since 2012. Similarly, average Loaf volume and water absorption of Elgin-ND were respectively, 1005 ml and 66.3% compared to 1064 ml and 65.4%; 996 ml and 67.5% scored by Glenn and Barlow, respectively. Additionally, Elgin-ND has average kernel weight (28 g for the 1000 kernel weight) and with average grain volume weight (732 compared to 769 and 750 kg m<sup>-3</sup> of Glenn and Barlow, respectively) (Table 1). Grain protein of Elgin-ND was high (154 g kg<sup>-1</sup>), similar to Glenn and Barlow (Table 1).

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**Table 1.** Summary of agronomic data for Elgin-ND hard red spring wheat (HRSW) and check cultivars tested in the ND HRSW Variety Trials (2010-2012).

Variety	Days to Heading	Plant Height	Lodging <sup>†</sup>	FHB FDK	Leaf Diseases	1000 Kernel Weight	Grain		
							Volume Weight	Grain Yield	
	Day	cm	1-9	%	%	g	kg m <sup>-3</sup>	kg ha <sup>-1</sup>	%
Advance	60.3	77.9		1.0	13.8	25.5	747	3546.9	14.8
Barlow	60.0	85.9	0.8	1.4	9.0	29.1	750	3912.6	15.4
Brick	56.4	86.2	1.3	0.2	30.6	29.3	756	3697.5	14.8
Briggs	59.0	84.6	1.4	0.7	10.0	29.4	746	3645.6	15.0
Dapps	55.4	88.9	1.7	0.3	8.9	30.3	734	3296.3	15.9
<b>Elgin-ND</b>	<b>62.1</b>	<b>88.8</b>	<b>0.5</b>	<b>0.6</b>	<b>10.7</b>	<b>27.8</b>	<b>732</b>	<b>4001.4</b>	<b>15.4</b>
Faller	63.1	83.1	0.4	0.2	10.9	29.5	726	3956.7	14.5
Forefront	57.8	88.9		0.4	18.8	28.2	753	3543.6	15.4
Glenn	59.6	88.0	0.2	0.0	13.0	29.5	769	3639.7	15.4
Norden	61.6	76.9	0.2	0.7	22.2	27.2	750	3779.3	15.0
Prosper	63.6	83.1	0.7	0.2	7.5	30.3	729	3972.9	14.7
RB07	60.4	78.2	1.0	0.1	14.6	27.0	735	3788.6	15.0
Reeder	60.8	83.4	0.2	1.8	6.4	28.0	733	3627.9	15.1
Select	58.2	84.9	0.9	0.8	37.1	28.9	749	3876.3	14.8
Steele-ND	61.1	85.5	1.5	1.2	17.7	28.9	744	3831.4	15.4
Velva	63.2	80.7	0.0	2.4	11.0	28.3	715	3759.9	14.9
LSD (0.05)	2.3	3.7	0.7	1.2	5.7	2.2	12	111.3	1.1
No of environment	22	22	5	3	4	22	22	22	21

<sup>†</sup> Lodging score: 1=completely erect to 9=completely flat at harvest.

**Table 2.** Diseases reactions of Elgin-ND and hard red spring wheat (HRSW) checks tested in the ND HRSW Variety Trials (2009-2011).

Genotype	FHB <sup>†</sup>		Leaf rust		Stem rust	
	%	28	Greenhouse <sup>‡</sup>	Field	Greenhouse <sup>§</sup>	Field
Elgin-ND			R <sup>¶</sup>	tR/5R	R	R/MR
Barlow	25		R	MR/MS	MR/R	5R
Faller	27		R	MR/MS	R	R
Glenn	25		-	R	R	R
2398	73		R	S	MR/R	5R
ND2710	13		R	R	-	-
No of environment	8		4	6	3	9

<sup>†</sup> FHB (Fusarium Head blight) severity as described by (Stack and Froberg, 1997).

<sup>‡</sup> Greenhouse reactions for leaf rust races MCDL and THBJ.

<sup>§</sup> Greenhouse reactions for stem rust races Pgt TPMK, TMLK, RTQQ, QFCQ, QTHJ, THTS, and TCMJ.

<sup>¶</sup> R=resistant, MR=Moderate resistant, MS=Moderate susceptible, S=Susceptible, tR= trace/Resistant.

**Table 3.** Quality parameters of Elgin-ND and hard red spring wheat (HRSW) checks tested in 20 location-years in the ND HRSW Variety Trials (2010-2011).

Cultivar	Falling number	Flour Extraction	Mixing time	Mixing tolerance	Loaf volume	Water absorption
Elgin-ND	406	68.2	7.8	11.8	1005	66.3
Barlow	388	70.4	7.0	10.0	996	67.5
Faller	389	70.5	8.4	10.8	1005	63.9
Glenn	357	68.2	10.0	15.0	1064	65.4
Mott	373	68.8	7.0	11.0	978	64.3
Velva	404	67.4	6.8	10.6	959	64.8
Mean-Checks	382	69.1	7.8	11.5	1000	65.2
No of environment	20	20	20	20	20	20

## MAPPING FUSARIUM HEAD BLIGHT RESISTANCE QTL IN A MID-ATLANTIC-ADAPTED BREEDING POPULATION

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### ABSTRACT

Wheat host resistance to Fusarium head blight (FHB) is a quantitative trait historically mapped using biparental mapping populations. Utilizing breeding populations in quantitative trait loci (QTL) mapping will greatly minimize costs and time required for QTL of desired traits to be incorporated in commercial varieties. The goal of this project is to fine-map QTL of FHB resistance, particularly on chromosome 2D, from a tri-parental cross of the highly FHB-resistant Chinese spring wheat cultivar Ning7840 and soft red winter wheat cultivars McCormick and SS8641. At each generation of selfing, lines were selected based on flanking SSR markers of known resistance QTL from donor parent Ning7840 to target lines with recombination events near these QTL. Fifty-six of these selected lines were genotyped using the wheat 9K iSelect assay and additional SSR markers. Lines were phenotyped for FHB resistance in a greenhouse study in College Park, MD and two field trials, located in Salisbury, MD and Lexington, KY. Phenotypic data from the 2012-2013 season suggest that the putative QTL on chromosome 2D from donor parent Ning7840 has no significant effect on scab resistance in the absence of Fhb1. Further, there may be resistance contributed by local cultivars. To better elucidate these phenomena, the sample size for the 2013-2014 field trials was increased to explore resistance contributed by minor QTL in both the presence and absence of Fhb1. In total, 138 lines will be phenotyped in a greenhouse study in College Park, MD and three field locations, located in Upper Marlboro, MD; Lexington, KY; and Kinston, NC. Lines will be genotyped using the wheat 9K iSelect assay and additional KASP and SSR markers on chromosomes 2D and 3B for increased resolution.

**THE 2012-13 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 2012-13 nursery comprised 58 advanced generation breeding lines and four check cultivars, 'Ernie', 'Bess', 'Jamestown' (partially resistant) and 'Coker 9835' (susceptible). Eight U.S. public programs (Univ. of Arkansas, Univ. of Georgia, Louisiana State University, Univ. of Maryland, N.C. State Univ., VA Tech., and USDA-ARS), and two private companies (Agripro-Coker, 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 60 and 84 percent of entries had significantly better means than the susceptible check for Severity, Index, FDK, ISK and DON. Sources of resistance included Chinese, South and North American.

Copies of the full report will be available at the 2013 National Fusarium Head Blight Forum and subsequently on line at the USWBSI web site: <http://www.scabusa.org/>.

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**Table 1.** Genotypic Analyses of Regions Associated with FHB Resistance and Other Pertinent Loci.

DESIGNATION	Rht-B1	Rht-D1	Rht8	Ppd-D1a insens	Vrn-A1 short	Vrn-B1 short	Lr34/Yr18	Lr37/Yr17/Sr38	Sr2	Sr36	Sr24/Lr24	Lr9	Fhb1	Fhb Massey 3BL	Fhb 5A ERNIE	Fhb 5A Ning7840	Fhb 2DL- Wuhan1/W
1 ERNIE	b	a	no	no	yes	no	no	no	no	yes	no	no	no	yes	yes	no	no
2 COKER9835	a	b	no	yes	no	yes	no	no	no	yes	no	yes	no	no	no	no	no
3 BESS	het	het	no	het	no	no	no	no	no	no	no	no	no	no	no	no	no
4 JAMESTOWN	a	b	no	yes	yes	no	no	no	no	no	no	no	no	no	no	no	no
5 MD03W61-11-3	a	b	no	yes	no	no	no	no	no	no	no	no	yes	no	no	no	no
6 ARS07-1214	b	a	no	no	no	no	no	no	no	no	yes	no	no	no	no	no	no
7 ARS09-367	b	a	no	no	no	no	no	no	no	no	yes	no	no	no	no	no	no
8 ARS09-446	a	a	no	no	no	yes	no	no	no	yes	no	no	no	no	no	no	no
9 ARS09-643	a	b	no	yes	no	yes	no	no	no	no	no	no	no	no	no	no	no
10 LA05102C-1-2	b het	a	no	yes	het	no	no	no	no	yes?	no	no	no	no	no	no	het
11 LA05102C-8-8	a	a	no	yes	no	yes	no	no	no	yes	no	no	no	no	no	no	no
12 NC09-20986	b	a	no	no	no	no	no	yes	no	no	no	no	yes	no	no	no	no
13 AR00260-2-2	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
14 AR01044-1-1	b	a	no	yes	yes	yes	no	yes	no	no	yes	no	no	no	no	no	no
15 AR01110-3-1	b	a	no	yes	no	no	no	no	no	no	no	no	no	yes	no	no	no
16 AR01178-1-1	b	a	no	yes	het	no	no	no	no	no	no	no	no	no	no	no	no
17 ARGE05-1229-2-1	b	a	no	yes	yes	no	no	no	no	no	no	no	no	no	no	no	no
18 ARGE07-1339-10-5-8	b het	a	no	no	no	no	no	no	no	het	no	no	no	no	no	no	het
19 ARGE07-1374-17-5-4	a	b	no	yes	no	no	no	no	yes	no	no	no	no	no	no	no	no
20 ARGE07-1374-17-8-5	a	b	no	yes	no	no	no	no	no	yes	no	yes	no	no	no	no	no
21 ARS07-1073	a	b	no	no	no	yes	no	no	no	no	no	no	no	no	no	no	no
22 ARS09-082	a	b	no	yes	no	yes	no	no	no	yes	no	yes	no	no	no	no	no
23 ARS09-228	a	a	Rht8c?	yes	no	no	no	no	no	yes	no	no	no	no	no	no	no
24 ARS09-745	a	b	no	yes	no	no	no	yes	no	no	no	no	no	no	no	no	no
25 GA04494-12ES33	a	b	no	yes	no	no	no	yes	no	no	no	no	no	no	yes	no	no
26 GA051477-12ES27	a	b	no	yes	yes	no	no	no	no	no	no	no	no	no	no	no	no
27 GA051477-12ES28	a	b	no	yes	yes	no	no	no	no	no	no	no	no	no	no	no	no
28 GA051477-12ES29	a	b	no	yes	yes	no	no	no	no	no	no	no	no	no	no	no	no
29 GA051477-12ES32	a	b	no	yes	yes	het	no	no	no	no	no	no	no	no	yes	no	no
30 GANC8170-12DH7	a	b	no	yes	no	no	no	yes	no	no	no	no	no	no	no	yes	no
31 GANC8248-12DH1	a	b	no	yes	no	no	no	no	no	yes	no	no	no	no	no	no	no
32 GANCZ4-12DH21	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
33 LA05079F-P05	b	a	no	no	no	het	no	no	no	no	no	no	no	no	no	no	no
34 LA06069E-P01	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
35 LA06149C-P7	a	b	no	no	yes	yes	no	no	no	no	no	no	no	no	no	no	no
36 LA07085CW-P4	a	b	no	yes	?	no	no	yes	no	het	no	no	no	no	no	no	no
37 LA07178C-44	a	b	no	no	yes	no	no	no	no	no	no	yes	no	no	no	no	no
38 LCS19227	a	b	no	no	no	no	no	no	no	no	no	no	no	no	no	no	no
39 LCS15963	b	a	no	no	no	no	no	no	no	no	no	yes	no	no	no	no	no
40 M10-1615	b	a	nd	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
41 M10-1659	a	a	no	no	yes	no	no	no	no	yes	no	yes	no	no	no	no	no
42 MD04W249-11-12	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
43 MD04W249-11-7	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
44 MD07W272-11-5	a	b	no	yes	yes	no	no	no	no	no	no	no	yes	no	no	no	no
45 MD08-26-H2-7-12-21	a	b	no	yes	no	no	no	yes	no	no	yes	no	yes	no	no	yes	yes
46 MD08-26-H2-7-12-9	a	b	no	yes	no	no	no	yes	no	no	yes	no	yes	no	no	yes	yes
47 MDNC8248-64	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
48 MH07-7474	b	a	no	no	no	no	no	no	no	no	no	no	no	no	no	no	no
49 NC09-20768	a	b	no	yes	no	no	no	no	no	yes	no	no	no	no	no	no	no
50 NC09-22352	b	a	Rht8c?	no	no	no	no	no	no	yes	no	no	no	no	no	no	no
51 NC10-25212	a	a	no	no	no	no	no	no	no	yes	no	yes	no	no	no	no	no
52 NC8170-4-3	a	b	no	yes	no	no	no	no	no	no	no	no	yes	no	no	no	no
53 NC8170-45-2	a	b	no	yes	no	no	no	no	no	no	no	no	yes	no	no	no	no
54 NC8840-19	a	a	no	yes	no	no	no	no	no	yes	no	no	yes	no	no	no	no
55 VA10W-112	a	b	no	yes	no	no	no	yes	no	no	no	no	no	no	no	no	no
56 VA10W-118	a	b	no	yes	no	no	no	yes	no	no	no	no	no	no	no	no	no
57 VA10W-119	a	b	no	yes	no	no	no	no	no	no	no	no	no	no	no	no	no
58 VA11W-FHB110	b	a	no	yes	yes	no	no	no	no	yes	no	yes	no	het	no	no	no
59 VA11WFHB57	het	a	no	no	yes	no	no	no	no	yes	no	yes	no	no	no	no	no
60 VA11W-FHB60	a	b	no	yes	no	yes	no	no	no	no	no	no	yes	no	no	no	no
61 VA11W-FHB61	a	b	no	yes	yes	no	no	no	no	no	no	no	yes	no	no	no	no
62 VA11W-FHB75	b	a	no	yes	yes	no	no	no	no	yes	no	no	yes	no	no	yes?	yes

	DESIGNATION	1RS	H9	H13	Sbm1	Tsn1	Glu-B1	Glu-D1	Glu-A1	Sucrose Synthase	Combined Colour
1	ERNIE	non-1RS	no	no	no	no	no	2+12	Ax1_or_null	yes	RR rr rr
2	COKER9835	non-1RS	no	no	yes	no	no	2+12	Ax2*	yes	rr RR RR
3	BESS	non-1RS	no	no	yes	yes	no	het	Ax1_or_null	no	RR rr RR
4	JAMESTOWN	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR RR RR
5	MD03W61-11-3	1RS:1BL	no	no	yes	no	no	5+10	Ax1_or_null	no	rr RR RR
6	ARS07-1214	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	no	RR rr rr
7	ARS09-367	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	no	RR rr rr
8	ARS09-446	non-1RS	no	no	no	no	no	5+10	Ax2*	yes	RR rr RR
9	ARS09-643	1RS:1AL	no	no	yes	no	Bx7oe_het	2+12	Ax2*	no	RR rr RR
10	LA05102C-1-2	1RS:1BL	no	no	yes	no	no	het	Ax2*	yes	RR RR RR
11	LA05102C-8-8	non-1RS	no	no	yes	no	no	2+12	Ax2*	yes	RR RR rr
12	NC09-20986	non-1RS	no	yes	yes	no	no	2+12	Ax2*	no	RR rr RR
13	AR00260-2-2	non-1RS	no	no	yes	no	no	5+10	Ax2*	no	RR RR RR
14	AR01044-1-1	non-1RS	no	no	yes	no	no	2+12	Ax1_or_null	yes	RR RR rr
15	AR01110-3-1	non-1RS	no	no	yes	no	no	5+10	Ax2*	no	RR rr rr
16	AR01178-1-1	non-1RS	no	no	no	no	Bx7oe	5+10	Ax1_or_null	no	RR rr RR
17	ARGE05-1229-2-1	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	no	RR Rr rr
18	ARGE07-1339-10-5-8	1RS:1AL	no	no	yes	yes	no	2+12	het	het	rr rr RR
19	ARGE07-1374-17-5-4	non-1RS	no	no	yes	no	no	2+12	Ax2*	yes	Rr rr RR
20	ARGE07-1374-17-8-5	non-1RS	no	no	yes	no	no	2+12	Ax1_or_null	yes	Rr rr RR
21	ARS07-1073	1RS:1AL	no	no	yes	no	no	2+12	Ax2*	no	RR RR RR
22	ARS09-082	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	yes	RR RR RR
23	ARS09-228	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	yes	RR rr rr
24	ARS09-745	non-1RS	no	no	yes	yes	no	2+12	Ax2*	no	RR rr rr
25	GA04494-12ES33	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR RR rr
26	GA051477-12ES27	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR rr RR
27	GA051477-12ES28	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR RR RR
28	GA051477-12ES29	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR rr RR
29	GA051477-12ES32	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	rr rr RR
30	GANC8170-12DH7	1RS:1BL	no	no	yes	no	no	5+10	Ax2*	no	RR RR RR
31	GANC8248-12DH1	non-1RS	no	no	yes	no	no	2+12	Ax1_or_null	yes	RR rr RR
32	GANCZ4-12DH21	1RS:1AL	no	no	yes	no	no	2+12	Ax1_or_null	no	RR RR RR
33	LA05079F-P05	non-1RS	no	no	yes	no	no	2+12	Ax1_or_null	no	RR RR RR
34	LA06069E-P01	non-1RS	no	no	yes	no	no	2+12	Ax1_or_null	no	RR rr RR
35	LA06149C-P7	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR RR RR
36	LA07085CW-P4	non-1RS	no	no	het	no	no	2+12	Ax2*	het	Rr RR RR
37	LA07178C-44	non-1RS	no	no	yes	no	no	2+12	het	no	RR rr RR
38	LCS19227	non-1RS	no	no	yes	no	no	het	Ax1_or_null	no	RR rr RR
39	LCS15963	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	no	RR RR rr
40	M10-1615	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	RR rr rr
41	M10-1659	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	yes	RR RR rr
42	MD04W249-11-12	1RS:1BL	no	no	yes	no	no	2+12	Ax1_or_null	no	RR rr RR
43	MD04W249-11-7	1RS:1BL	no	no	yes	no	no	2+12	Ax1_or_null	no	RR rr RR
44	MD07W272-11-5	1RS:1BL	no	no	yes	no	no	het	Ax2*	no	RR rr RR
45	MD08-26-H2-7-12-21	non-1RS	no	no	yes	yes	no	2+12	Ax2*	no	RR RR RR
46	MD08-26-H2-7-12-9	non-1RS	no	no	yes	yes	no	2+12	Ax2*	no	RR RR RR
47	MDNC8248-64	1RS:1AL	no	no	yes	no	no	2+12	Ax2*	no	RR rr RR
48	MH07-7474	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	no	RR rr RR
49	NC09-20768	non-1RS	no	no	yes	no	no	2+12	Ax2*	yes	rr rr RR
50	NC09-22352	1RS:1BL	no	no	yes	yes	no	5+10	Ax1_or_null	yes	RR RR RR
51	NC10-25212	non-1RS	no	no	yes	no	no	5+10	Ax1_or_null	yes	RR rr rr
52	NC8170-4-3	non-1RS	no	no	yes	no	no	5+10	Ax2*	no	RR RR RR
53	NC8170-45-2	non-1RS	no	no	yes	no	no	5+10	Ax2*	no	RR RR RR
54	NC8840-19	non-1RS	no	no	yes	no	no	2+12	Ax1_or_null	yes	RR rr rr
55	VA10W-112	non-1RS	no	het	yes	yes	no	2+12	Ax2*	no	rr rr RR
56	VA10W-118	non-1RS	no	no	yes	yes	no	2+12	Ax2*	no	rr rr RR
57	VA10W-119	non-1RS	no	yes	yes	yes	no	2+12	Ax2*	no	RR rr rr
58	VA11W-FHB110	non-1RS	no	no	yes	no	no	2+12	het	yes	RR rr RR
59	VA11WFHB57	1RS:1BL	no	no	yes	no	no	5+10	Ax1_or_null	yes	RR rr RR
60	VA11W-FHB60	1RS:1BL	no	no	yes	no	no	2+12	Ax1_or_null	no	rr RR RR
61	VA11W-FHB61	non-1RS	no	no	yes	no	no	2+12	Ax2*	no	rr rr RR
62	VA11W-FHB75	1RS:1BL	no	no	yes	no	no	2+12	Ax1_or_null	yes	RR rr RR

## PATTERNS OF SINGLE KERNEL DEOXYNIVALENOL LEVELS IN ARTIFICIALLY INOCULATED WHEAT SPIKES AS DETECTED BY NEAR INFRARED SPECTROSCOPY

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### Abstract

Evaluation of single-kernel deoxynivalenol (DON) levels of artificially inoculated wheat spikes facilitates study of the magnitude and patterns of DON accumulation among kernels with reference to the point of inoculation. This can illustrate some modes of Fusarium Head Blight (FHB) resistance in wheat germplasm. Therefore, using FHB susceptible (S) and moderately resistant (MR) wheat cultivars, a study was conducted to estimate single-kernel DON levels in kernels extracted from artificially inoculated wheat spikes to investigate the patterns and magnitudes of DON accumulation.

Wheat spikes from cultivars 2137 (S), Overley (S), Everest (MR), and Heyne (MR) were used in this study. The 10<sup>th</sup> spikelet from the basal end of each spike was inoculated with a spore suspension (10 µl of 1X10<sup>5</sup>/ml) of *F. graminearum* (isolate 3639) at the time of flowering and the inoculated spikelet was tagged. Non-inoculated spikes grown under the same conditions were used as controls. Spikes were harvested when the kernels were mature.

Six inoculated and six non-inoculated spikes from each cultivar were randomly selected to extract kernels for DON determination. Kernels in each spikelet were manually removed beginning from the basal end upwards. The kernels in each spikelet were separately placed in cells of numbered pill boxes. To identify the position of kernels in the spike in relation to the point of inoculation, the 10<sup>th</sup> spikelet with the inoculated floret was designated as spikelet 0; those above the central spikelet and progressing towards the distal end of the spike were assigned sequential positive integers; and those below and progressing towards the basal end of the spike were assigned sequential negative integers. A single kernel from the 2-3 kernels extracted from each spikelet was randomly selected and its DON level was determined by a Single Kernel Near-Infrared (SKNIR) instrument. The average DON level and the standard error for DON level in each spikelet position were calculated.

Kernels in inoculated spikelets (Spikelet 0) had very high levels of DON compared to other kernels in the spike. In susceptible cultivars 2137 and Overley, high levels of DON were also detected in kernels extracted from spikelets away from the inoculated spikelet. However, DON levels of kernels gradually decreased as the distance from the inoculated spikelet increased in both directions. Moderately-resistant cultivars Everest and Heyne reacted in a different manner in that DON accumulation was detected mostly in the kernels in inoculated spikelets or the one below it. Kernels in non-inoculated spikes had no detectable levels of DON. These results show that cultivars Everest and Heyne not only resist the spread of FHB infection compared to cultivars 2137 and Overley but that resistance is reflected in DON accumulation patterns.

The susceptible cultivar Overlay accumulated significantly higher levels of DON in infected kernels compared to the other susceptible cultivar 2137 when grown and inoculated under similar environmental conditions. This may be due to a slightly higher resistance of 2137 for DON accumulation compared to Overlay.

This NIR spectrometric method may be useful for the evaluation of modes of FHB resistance in wheat cultivars such as resistance to spread of infection within spike (Type II resistance) and resistance to toxin (DON) accumulation (Type III resistance).

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QTL ASSOCIATED WITH FUSARIUM HEAD BLIGHT  
RESISTANCE IN THE NC-NEUSE X AGS 2000  
RECOMBINANT INBRED 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 179 random F<sub>5</sub>-derived recombinant inbred lines derived from a cross between 'NC-Neuse' and the FHB susceptible line 'AGS 2000' was evaluated for FHB resistance at one field location (3 reps) in the 2010-11 field season, at two field locations (2 reps/loc) in the 2011-12 season, and two field locations in the 2012-13 season (3 reps/loc). The FHB related traits evaluated included disease incidence (INC), severity (SEV), *Fusarium* damaged kernels (FDK), and accumulation of the mycotoxin deoxynivalenol (DON).

A linkage map developed prior to this QTL mapping study was updated with a total of 1839 polymorphic SSR, DArT and SNP markers across 27 linkage groups was utilized for mapping of QTL. QTL analysis for individual environments and across environments was conducted using Composite Interval Mapping (CIM) and then Multiple Interval Mapping (MIM) with WinQTLCart vs. 2.5. The critical LOD value to declare QTL significance was 3.2, based on 1000 permutations.

Preliminary results showed QTL associated with one or more FHB resistance traits on chromosomes 1A, 1B, 2A, 4A, 4B, 5B, and 6A. Their LOD score values ranged from 3.2 to 5.69 with R<sup>2</sup> values of 6.0% to 14.8%.

An update on pertinent results will be presented.

## PROMISING FUSARIUM HEAD BLIGHT RESISTANCE IN DURUM WHEAT

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### ABSTRACT

Cultivated tetraploid wheat, especially durum wheat (*Triticum durum*), is highly susceptible to the wide-spread disease Fusarium head blight (FHB). While many resistance QTL have been reported in hexaploid wheat (*Triticum aestivum*) the QTL identified to date in tetraploid wheat do not provide satisfactory FHB resistance. Consequently, several groups have introgressed resistance alleles from wild and cultivated relatives into durum. In this study, back-cross lines derived from crosses of *T. durum* and FHB resistance sources including *Triticum dicoccum*, *Triticum dicoccoides* and *Triticum aestivum* were used as resistant parental lines in several bi- and multi-parental crosses with *T. durum* cultivars. A large population was developed allowing the evaluation of FHB resistance derived from relatives in an agronomically acceptable durum background. This population was evaluated in 2012 and 2013 in disease nurseries at the site of the Institute for Biotechnology in Plant Production (BOKU, Austria) through artificial spray inoculation of *Fusarium culmorum* macroconidia at anthesis. The population showed a large spectrum of response for FHB resistance ranging from highly resistant to susceptible. FHB severity was significantly negatively correlated with plant height ( $r=-0.51$  in 2012 and  $r=-0.46$  in 2013) but variation for FHB resistance was observed among the short lines ( $\leq 80$  cm height) with several resistant lines. Variation for flowering date was low and FHB resistant lines were present in all maturity groups. A subset of 475 lines will be analysed through both linkage and genome-wide association mapping. The lines will be genotyped in high-density at INRA Clermont-Ferrand (France) using the GENTYANE platform and phenotyped at two locations: Florimond-Desprez in Cappelle-en-Pévèle (France) and BOKU University in Tulln. We expect to unveil QTL linked with resistance and/or increased susceptibility and to evaluate the importance of epistatic interactions for FHB resistance in durum wheat.

### ACKNOWLEDGEMENTS

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## EFFECTS OF LATE-SEASON RAIN/SIMULATED RAIN AND GRAIN DRYING ON SELECTION FOR LOW DON CONCENTRATION IN WHEAT GRAIN

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### ABSTRACT

Developing wheat cultivars with low concentrations of deoxynivalenol (DON) in grain is a high priority for breeding programs funded by the USWBSI, so it is important to understand the factors affecting DON concentration in grain. Some studies on the effects of late-season moisture found increases in DON while others found decreases due to leaching. The objectives of this study were to determine effects of late-season rain and misting on DON concentration in wheat spike tissues and to quantify the amount of DON leached from spikes. Field experiments were conducted on susceptible and moderately resistant wheat cultivars affected by FHB utilizing spike holders to catch water leaching through groups of 20 spikes, rain shelters to protect plots from rain and misting, and a rainfall simulator to apply simulated rain. Groups of spikes in spike holders were either not treated or treated with various amounts of rain/simulated rain. A sample of water that leached through the spikes was frozen at -80°C, lyophilized, and analyzed for DON. Spikes were dried in a grain dryer at 65°C, and samples of grain and chaff were analyzed for DON. A critical component of these experiments was having groups of spikes with similar levels of DON at the beginning of experiments, and methods were developed to make groups as similar as possible and to statistically test for similarity such that dissimilar groups could be eliminated to improve the accuracy of results. DON was detected in all water samples, indicating that leaching of DON is a common phenomenon. Similar percentages of DON leached from most spike samples that received a particular rain treatment, indicating that the amount leached is proportional to the amount in the sample. Chaff and scabby grain had the highest concentrations of DON and the greatest reductions in DON after rain treatments. DON concentrations in total grain were reduced with a large simulated rain event at soft dough stage and with large cumulative effects of rain and misting in unprotected plots compared to plots protected with rain shelters, indicating that late-season rain and misting reduces DON concentrations in grain. Drying wet spike samples in a grain dryer was found to degrade variable portions of DON among samples. Based on the findings of this study, several recommendations can be made to improve the accuracy of DON estimates in the inoculated and misted uniform nurseries used to evaluate breeding lines for FHB resistance. Stop mist treatments at the time DON production in grain slows considerably because misting after this time increases the probability for leaching and degradation of DON. Harvest naturally dried grain as soon as possible after entries first reach harvest dryness to avoid possible complications from leaching and degradation. If entries must be harvested at high moisture, dry as quickly as possible at moderate temperature to minimize degradation of DON. Although leaching and degradation of DON likely will continue to affect results of these screening nurseries, knowing that late-season rain and misting leaches DON and that DON is susceptible to degradation under wet conditions, especially at elevated temperatures, will allow these factors to be minimized or at least made as similar as possible across all entries in a test to improve the accuracy of the results. Evaluations done as well as possible across multiple environments should allow reliable estimates of DON concentration. For experiments requiring the most accurate estimates of DON levels in wet spikes, the spikes should be frozen at -80°C as quickly as possible and then lyophilized for DON analysis.

CHARACTERIZATION OF WHEAT RILS DERIVED FROM HIGH  
YIELDING VARIETY WL711 AND DROUGHT RESISTANT  
VARIETY C306 UNDER WATER DEFICIT STRESS  
TREATMENT USING DROUGHT SUSCEPTIBILITY  
INDEX (DSI) AS SELECTION CRITERIA

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**ABSTRACT**

Climate change is projected to have significant impacts on temperature and precipitation profiles globally which will have profound effects on crop productivity. Breeding for drought resistance is a major aim of many wheat breeding programs both nationally and internationally and requires the identification of genetic material combining adaptation with higher yield. Grain number per unit area (GN) and grain weight usually expressed as 1000-grain weight (TGW) are the major determinants of grain yield. It is the reduction in both GN and TGW that accounts for grain yield reduction in wheat under water deficit stress environments. Eight RILs from the WL711/C306 wheat RIL population were identified as drought resistant germplasm based on drought susceptibility index (DSI) from field trials conducted at IARI, New Delhi from 2007-2013 under irrigated and water deficit stress environments. The parent cultivars have contrasting differences in morpho-physiological traits, yield potential and yield stability. WL711 is a semi-dwarf, medium flowering, high yielding and drought susceptible wheat cultivar while C306 is a tall, late flowering, medium yielding and drought resistant cultivar. The eight RILs identified in this study are desirable recombinants of this population having medium to late flowering, intermediate height, higher grain number combined with high grain weight, yield higher than C306, stability for yield and yield components. The range of grain yield, biomass, harvest index, grain number and 1000-grain weight of the selected RILs was 540.0-673.6 gm<sup>-2</sup>, 1633-2375 gm<sup>-2</sup>, 31.6-35.7 %, 14302-18286 m<sup>-2</sup> and 33.5-38.0 g respectively. These RILs maintained better water relations, cooler canopies and tougher membranes under water deficit stress conditions like C306. Four of these RILs (R-7, R-61, R-19 and R-208) have been registered at National Bureau of Plant Genetic Resources (NBPGR) as drought resistant wheat germplasm and can be used as genetic material in breeding programs aimed at drought resistance in wheat.

## MAPPING AND COMBINING GENES FOR FHB RESISTANCE IN WHEAT

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### ABSTRACT

Single gene resistance to Fusarium head blight (FHB) in wheat provides only partial resistance and also the disease severity is highly influenced by environment. Consequently multiple genes are needed for effective resistance. Our hypothesis is that identifying DNA markers for Type I resistance will be very beneficial for selection, and combining Type I and Type II FHB resistance will be more effective than either type alone. The objectives of this project are to 1) characterize a RIL population from the cross INW0412 (Type I resistance)/992060 (susceptible) for frequency of initial infection and map QTLs for Type I resistance and 2) combine Type I resistance from cultivars Goldfield, INW0412 and Truman; and Type II resistance of *Fhb1* and *Qfhs.pur-7EL* backcrossed into adapted soft winter wheat lines and quantify augmentation of FHB resistance. For objective 1, a population of 198 RILs and the two parents were characterized for FHB incidence in two replicates at Lafayette, IN in 2011 and 2013. A two-enzyme genotyping by sequencing (GBS) approach is in process to identify linked markers. The RIL Type I resistance field data and the GBS single nucleotide polymorphism (SNP) marker data will be used to construct a high-density genetic map to identify QTL contributing to Type I FHB resistance. For objective 2, BC F<sub>1:2</sub> lines have been genotyped with 7 simple sequence repeat (SSR) markers for *Fhb1*, *Qfhs.pur-7EL* and low incidence. F<sub>2:4</sub> lines were phenotyped in two replicated field tests for FHB incidence and severity at Lafayette, IN in 2013 for Type I and Type II resistance separately. By using phenotypic and SSR marker selection, lines with both Type I and II FHB resistance were identified, and it was confirmed that lines with multiple markers associated with FHB resistance provide more FHB resistance than lines with few or no markers. Those lines will be beneficial for the improvement of FHB resistance in wheat.

## TRANSITIONING FROM PHENOTYPIC SELECTION TO GENOMIC SELECTION FOR LOWER DEOXYNIVALENOL IN BARLEY

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### ABSTRACT

A major challenge in barley breeding is to simultaneously increase yield and resistance to FHB while maintaining malting quality. Historically this has been accomplished by phenotypic selection (PS) for disease resistance in early generations, followed by analysis of deoxynivalenol (DON) in harvested grain samples, then evaluation of yield in more advanced lines, and finally selective assessment for a suite of malting quality traits. Information on these traits is accumulated over several years of the breeding cycle and is costly. Genomic selection (GS) methodologies use genome-wide markers to estimate the breeding value of lines early in the breeding cycle at increasingly lower costs. While the theoretical basis and methodology of GS has been the subject of many studies, still in its infancy, GS has yet to be rigorously evaluated empirically. Given their classically quantitative nature of inheritance, FHB severity, DON concentration and grain yield are excellent candidates for improvement using GS. Accurate calculations of genomic estimated breeding values (GEBVs) for any trait, in theory, should minimize cycle-to-cycle noise caused by non-genetic factors and provide means to make more consistent gains. In 2010, the University of Minnesota began replacing traditional phenotypic field screening for FHB with genomic predictions of early generation breeding material. Using disease and yield trial data from 2005 to 2013 we have characterized changes in yield and DON levels over time. This time period includes years where phenotypic and genomic selection was implemented and provides an opportunity to assess the effect of transitioning to genomic selection. We have summarized DON and (grain yield) for 6 (7) years of PS breeding and 1 (2) cycles of GS breeding. DON levels decrease steadily over years of PS and the trend continues into the first cycle of GS. Grain yields are more sporadic over the years of PS and have decreased slightly in comparison to the aggregate PS value; however they do show a promising upward trend between cycles 1 and 2 of GS. Additional data from subsequent GS cycles is required for rigorous analysis, but the initial trends are thus far supportive of the utility of GS, which is further enhanced when the economic benefits of fewer plots per trial and fewer trials are considered.

## EFFECTS OF DEOXYNIVALENOL ON THE WHEAT METABOLOME

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### ABSTRACT

The trichothecene mycotoxin deoxynivalenol (DON) is a major virulence factor of the plant pathogenic fungus *Fusarium graminearum* and plays an important role in the Fusarium head blight (FHB) disease. The presented study was carried out to investigate the effect of DON on the metabolome of six wheat genotypes which showed a varying degree of FHB resistance. A gas chromatography – mass spectrometry based targeted metabolomics approach was chosen to identify substances differentially expressed according to treatment and/or wheat genotype.

Two parent wheat lines CM-82036 (resistant) and Remus (susceptible) as well as four near isogenic wheat lines (NILs) were treated with either (i) DON or (ii) water as a control. The NILs differed in the two major resistance QTLs against FHB, namely *Qfhs.ndsu-3BS* and *Qfhs.ifa-5A*. Treated plants were sampled after 0, 12, 24, 48 and 96 hours and immediately shock frozen in liquid nitrogen. Ground wheat ears were extracted with acidified aqueous methanol and purified by liquid/liquid extraction with chloroform. The methanol/water phase was analyzed using GC-MS after an automated two step derivatisation employing methoxyamine hydrochloride (MOX) and N-methyl-N-trimethylsilyl trifluoroacetamide (MSTFA). GC-MS chromatograms were deconvoluted and further processed with the tailor made MetaboliteDetector software. Currently, the method covers more than 130 polar analytes whereof the majority are primary metabolites.

In this contribution, we aim to identify the effects of DON on the metabolites determined by our GC-MS method. We will present detailed GC-MS results of this metabolomics experiment with the goal to identify wheat metabolites, which are closely linked to the tested resistance QTLs or inoculation with DON. Thereby we intend to gain deeper insights on fungal virulence and plant resistance towards FHB.

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## CHARACTERIZATION OF FHB RESISTANCE IN SRW ROANE AND JAMESTOWN NAM POPULATIONS

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### ABSTRACT

Fusarium Head Blight (FHB), a pathogen of wheat (*Triticum aestivum* L.), frequently results in significant yield losses and accumulation of mycotoxins, such as deoxynivalenol and nivalenol in the grain. As a result, breeding programs have been working to identify resistance genes in wheat varieties known to be resistant to FHB. Some types of resistance that are particularly useful include: Type I (resistance to initial infection), Type II (resistance to pathogen spread in the spike), decrease in mycotoxin accumulation and number of *Fusarium* damaged kernels (FDK). The overall goal is to identify quantitative trait loci (QTLs) for the different resistance types and pyramid these into elite backgrounds. The objective of this study is to identify quantitative trait loci (QTL) for FHB resistance in the native soft red winter (SRW) wheat cultivars Roane and Jamestown. A total of 186 Pioneer 25R47/Jamestown (P47/JT) F<sub>5:7</sub> RILs, were evaluated for FHB incidence, severity, index, and DON concentration for two years in three environments (MD, NC, and VA). A set of 170 F<sub>5:7</sub> RILs derived from FG95195 x Jamestown (FG/JT) were evaluated for FHB incidence, severity, index, and concentrations of DON and NIV for two years in four environments (AR, LA, GA, and VA). Two Roane populations, including 33 F<sub>4:7</sub> RILs from Roane/Allegiance (R/A) and 18 F<sub>4:7</sub> RILs from Roane/KY93C-1238-17-1 (R/KY93), were grown in 5 locations (KY, MD, MO, NC, and VA) the first year and in 4 locations the second year (KY, MD, NC, and VA). The Roane populations were also evaluated for FHB incidence, severity, index, and DON concentration. Both public and proprietary single nucleotide polymorphism (SNP) markers were used to genotype 42 of the P47/JT RILs, 11 FG/JT RILs, 11 R/A RILs, and all 18 of the R/KY93 RILs at Monsanto. Bulk segregant analysis was used to select microsatellite markers (SSRs) putatively associated with FHB resistance. Linkage maps were constructed using Map Manager QTX, based on the consensus map provided by Monsanto. Windows Cartographer (WinQTLCart version 2.5) was used to identify possible QTLs. Three QTL were identified in P47/JT located on chromosomes 1B and 3B, associated with FHB severity, and on 7D, associated with FDK. In the FG/JT population, 8 QTLs were identified and almost all of these QTL were associated with FHB severity (1B, 2A, 2D, 4A, and 5D), while one was associated with incidence (2A). Of the 12 QTLs identified in the R/A population, four were associated with FHB severity (1A, 1D, 3A, 3B), six were associated with incidence (3A, 3D, 4B, 5A, 7A, and 7D) and one was associated with DON (7A). In the R/KY93 six QTLs were identified on

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chromosomes 2A, 2B, 3A, 3B, and 4A. Of the chromosomes associated with FHB in R/KY93, FDK and DON were associated with 2A and 3B respectively, and severity was associated with chromosomes 2B, 3A, and 4A. The QTL on 1B was consistently associated with decreased FHB severity in both Jamestown mapping populations and should be useful to deploy via marker assisted selection.

DETERMINE THE AUGMENTATION EFFECT OF  
FHB RESISTANCE GENES IN WHEAT

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**ABSTRACT**

Fusarium head blight (FHB) of wheat, caused mainly by *Fusarium graminearum*, causes reduced yield and lowered grain quality. Lots of QTL conferring FHB resistance have been identified in the past few years. QTL pyramiding is one of the intuitive solutions to improve the FHB resistance. A backcross population with 2 major QTL for FHB was established in this study. The population was divided into 4 genotype groups based on the combination of the presence and absence of the 2 QTL. FHB resistance was evaluated by artificial inoculation of a mixture of 4 different local *Fusarium graminearum* isolates in 4 trials in both field and greenhouse in 2011 and 2012. The results in all the trials show no augmentation of FHB resistance in different genotype groups. It indicates that these two genes don't act in an additive manner, and thus provides some evidence that QTL pyramiding is not always an effective way to seek for more durable resistance for complex disease traits.

FINE MAPPING OF THE GENOMIC REGION HARBORING  
THE FUSARIUM HEAD BLIGHT RESISTANCE QTL  
*QFHS.NDSU-3AS* IN DURUM WHEAT

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**ABSTRACT**

The FHB resistance QTL *Qfhs.ndsu-3AS* was identified on chromosome 3A in the wild emmer wheat (*Triticum dicoccoides*) accession “Israel-A” (ISA). It was positioned to a 7.4 cM chromosomal interval in a mapping population of 83 recombinant inbred chromosome lines (RICLs) in a previous study. The RICLs were derived from the cross of Langdon (LDN) durum-ISA 3A substitution line [(LDN-ISA(3A)] with LDN. Five co-segregating wheat EST-derived STS markers (*Xwgc 1188*, *Xwgc716*, *Xwgc1143*, *Xwgc1204*, and *Xwgc501*) mapped 0.6 cM proximal to the SSR marker *Xgwm2* that closely linked to the QTL peak. In the present study, we attempted to increase map resolution of the QTL region and to position the QTL to a smaller chromosomal interval. The RICL#10, which was identified to contain *Qfhs.ndsu-3AS* in the shortest *T. dicoccoides* chromosome fragment among the 83 RICLs, was crossed with LDN to generate a large F<sub>2</sub> recombination population (n>1,800) within the chromosome region. To date, we have genotyped 372 individuals of this population with *Xgwm2* and other nine STS markers proximal to *Xgwm2*, including the five co-segregating STS markers near *Qfhs.ndsu-3AS*. As a result, *Xwgc716* and *Xwgc1188* co-segregated and mapped 0.8 cM proximal to *Xgwm2*. *Xwgc1143*, *Xwgc1204*, and *Xwgc501* also co-segregated and mapped 0.1 cM proximal to *Xwgc716* and *Xwgc1188*. The other four STS markers (*Xwgc1226*, *Xwgc510*, *Xwgc1296*, and *Xwgc1301*) mapped further proximal to the above markers in a higher resolution. Homozygous recombinant lines with smaller *T. dicoccoides* chromosome fragments have been selected using the molecular markers in the F<sub>2</sub> and F<sub>3</sub> generations. We have been evaluating the recombinant lines for FHB resistance in the greenhouse. Eventually, we expect to place *Qfhs.ndsu-3AS* to a smaller chromosomal interval on 3AS and to incorporate this FHB resistance QTL into durum background with minimal *T. dicoccoides* chromatin and reduced linkage drag.