SESSION 5:

VARIETY DEVELOPMENT AND HOST RESISTANCE

Chairpersons: Gina Brown-Guedira and Mohamed Mergoum

AIR SEPARATION AND DIGITAL PHOTO ANALYSIS AS NOVEL METHODS TO MEASURE THE PERCENTAGE OF *FUSARIUM* DAMAGED KERNELS.

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ABSTRACT

One of the greatest problems in breeding for Fusarium head blight (FHB) resistance lies in the difficulty of assessing the disease. At the present time, researchers generally measure disease incidence and severity in the field, deoxynivalenol (DON) content and percentage *Fusarium* damaged kernels (FDK).

FDK is presently measured in two ways: i) visual comparison of samples with reference samples and ii) manual separation of diseased and healthy kernels. Visual comparison of samples is a quick way of assessing FDK but is arguably too subjective. On the other hand, manual separation could be less subjective but is highly time consuming. Furthermore in manual separation, due to the amount of work that it takes, only small samples (e.g. 100 kernels) can be evaluated. This may not be an adequately representative sample size. To improve the efficiency of FDK measurement we should look for a method that: (i) reduces subjectivity, (ii) reduces the amount of work and time required, and (iii) allows increased sample size. To achieve this, two new methods are being proposed: air separation and digital photo analysis.

Air separation methods have long been used in the seed industry for seed conditioning purposes and in seed labs to measure the proportion of different components of seed samples. An air separation machine was specifically developed from a Precision Machine head thresher and a Shop-Vac vacuum to separate scabby kernels from healthy ones. Once a sample is loaded into the machine air-driven elevation of the lighter portion of wheat (i.e. scabby seeds) occurs until it reaches the top of the column where is collected in a receptacle. The heavier portion of wheat (i.e. asymptomatic seeds) is suspended midair and does not reach the top of the column. Once the air is turned off, the asymptomatic seeds fall and are collected in the bottom of the column. Finally, both portions of the sample are weighed separately and FDK is calculated. Approximate time per sample is 90 seconds.

In the digital photo analysis method, samples are evaluated based in their color composition. Color histograms are generated from the digital photos of the samples by image editing software. Mean blue value appears to have a consistent correlation with the FHB damage of kernels.

The air separation and the photo analysis methods emerge as two prospective techniques to measure FDK in an efficient and objective way and, ultimately, appear as promising tools in the difficult endeavor of assessing FHB in scab breeding programs.

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USE OF MAS FOR FHB RESISTANCE: IS IT WORKING FOR WHEAT BREEDERS?

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ABSTRACT

Marker-assisted selection (MAS) is most appropriate and efficient when 1) the gene/QTL under selection has a large contribution to the phenotype; and 2) diagnostic markers are available. MAS is especially attractive to wheat breeders needing to improve FHB resistance because of the difficulties posed by phenotypic selection, including multi-genic inheritance and the need to establish inoculated, misted nurseries. Although not a substitute for established phenotypic screening methods, MAS, when used in early generations (F_2-F_4) can effectively increase the frequency of resistant genotypes. The *Fhb1* QTL on chromosome 3BS is the best known and by far the most heavily selected QTL for FHB resistance. The presence of this QTL results in 20% or more reduction of disease symptoms, making it the most potent FHB QTL mapped to date. The SSR markers *Xbarc133* and *Xgwm493* are in a 5 cM interval that flank *Fhb1*. *Xbarc133* is less than 2 cM from *Fhb1* and can be effectively used as a stand-alone marker, but is not diagnostic in all genetic backgrounds. New STS markers based on the sequences of candidate genes should be more efficient than marker *Xbarc133*. The QTL on chromosome 5AS, *Qfhs.ifa-5A*, provides only Type I (initial infection) resistance, but is complementary to *Fhb1*. However, this QTL is in a centromeric region of 5AS, making it less accessible for fine mapping and development of diagnostic markers. We have had success using *Xbarc180* to track this QTL in our germplasm. From our surveys of germplasm in the U.S. spring wheat region and some soft red winter cultivars, we discovered numerous cases of highly resistant material not having *Fhb1*, so the presence of this gene cannot be inferred based on pedigree and high levels of resistance. Robust phenotypic screening is still essential to identify resistant cultivars. Survey results regarding the use of these and other markers for FHB resistance will be presented.

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MARKER-ASSISTED TRANSFER OF 3BS QTL FOR FHB RESISTANCE INTO HARD WINTER WHEAT.

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ABSTRACT

Epidemics of Fusarium head blight (FHB) can significantly reduce wheat grain yield and quality in the central and northern Great Plains of the U.S.A. Use of resistant cultivars is the most effective measure to control the disease. However, most hard winter wheat (HWW) cultivars currently grown in this area are highly susceptible to FHB. In addition, the disease screening procedure is laborious, time consuming, and costly, the progress in breeding for resistant HWW cultivars has been relatively slow with conventional methods. We used high-throughput marker-assisted backcross method to successfully transfer the major quantitative trait locus (QTL) from Sumai 3 and its derivatives into locally adapted HWW with minor FHB-resistance QTL to develop marketable FHB resistant HWW cultivars and useful germplasm lines. Three crosses were made between Sumai 3 derived soft red wheat lines and three locally adapted hard winter wheat cultivars (Harding, Wesley and Trego). Harding and Wesley are red wheat cultivars from South Dakota and Nebraska, respectively, and Trego is a white wheat cultivar from Kansas. Using marker-assisted backcross, about 80 Bc_2F_2 plants homozygous for the 3BS QTL were selected from each backcross population based on closely linked markers to the 3BS QTL. All selected Bc_2F_3 lines were evaluated in the greenhouse for Type II resistance in the USDA Genotyping Center in the fall of 2006. The result indicated that most selected lines were either highly resistant or moderately resistant. These materials have also been planted in mist-irrigated fields for further selection of FHB resistance, winter hardiness, hard-textured grains, and other traits. Some lines with good FHB resistance and other desirable traits will be released as new germplasm or cultivars in the hard winter wheat growning region after further yield trials.

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GENETIC LINKAGE MAPPING WITH DART MARKERS TO DETECT SCAB RESISTANCE QTLS IN A 'SUMAI-3' DERIVED WHEAT POPULATION.

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ABSTRACT

Much effort has been invested in identification of molecular markers linked to quantitative trait loci (QTL) that confer Fusarium Head Blight (FHB) resistance, though levels significantly higher than those of 'Sumai-3' remain elusive. Additional resistance QTLs may exist which typically are overshadowed by partial resistance of susceptible parents used in development of mapping populations. The objectives of this research were: (1) to create a genetic linkage map using Diversity Array Technology (DArT) and Simple Sequence Repeat (SSR) markers and (2) to associate FHB resistance phenotypes with the markers. Our population was created by crossing Sumai-3 with the very susceptible 'Y1193-6' (a Tibetan accession with unknown pedigree). An $F_{2:6}$ recombinant inbred mapping population was developed. A framework map consisting of 65 polymorphic SSR markers has been used to place most of 352 DArT markers. Our report will include associations between markers and FHB resistance QTLs for disease incidence, severity, index, and FDK percentage values.

USING THE AFFYMETRIX ARRAY TO DISCOVER SINGLE NUCLEOTIDE POLYMORPHISMS IN WHEAT.

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ABSTRACT

Gene expression arrays have been used to discover single nucleotide polymorphisms (SNPs) in several crop species. This study was designed to explore the possibility of using the Affymetrix Wheat Genome Array for the discovery of SNPs in wheat. Complementary DNAs synthesized from the mRNA isolated from the seedlings of six wheat cultivars of diverse origins (Ning 7840, Clark, Jagger, Encruzilhada, Chinese Spring and Opata 85) were hybridized to Affymetrix Wheat Genome Arrays. Cluster analysis of array data selected a total of 396 genes/probe sets with a signal intensity of at least 200, p-value of <1e-10 and overall R^2 ratio >0.8 for SNP confirmation through DNA sequencing. Sequencing results confirmed that 87 probe sets had at least one SNP within the probe sequences. In addition, SNPs were also identified in 21 additional genes, but they were detected outside the probe sequences. A total of 387 SNPs were discovered from the 108 genes. One SNP was selected from each gene to design primers for SNP analysis in a mapping population using SNaPshot kit and only 62 primers were successfully designed for SNaPshot analysis. Forty-two SNP markers were further analyzed in 96 F_{8-12} recombinant inbred lines from the cross of Ning 7840/Clark and 25 markers were integrated into the existing SSR map of the population. The result shows that Affymetrix arrays can be used to discover SNP markers in wheat.

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ENHANCING HOST RESISTANCE TO FUSARIUM HEAD BLIGHT: PYRAMIDING GENES IN SPRING WHEAT.

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ABSTRACT

Spring wheat (*Triticum aestivum* L.) production in the Northern Great Plains is severely affected by Fusarium head blight (FHB). Damage due to FHB is characterized by bleached spikes with white shriveled kernels, which ultimately reduces grain yield, lowers test weight, and results in an accumulation of deoxynivalenol (DON) in the kernels. Type II resistance has been associated with genes on chromosome 3B of Sumai 3 and chromosome 3A of *Triticum turgidum* L. var. *dicoccoides*. The objective of this study was to evaluate and compare the FHB resistance of two lines. One line, designated Line 1 has the Sumai 3 source only and another line, designated Line 2 has both the Sumai 3 and *T. dicoccoides* sources. These lines were developed by crossing a synthetic hexaploid wheat having the *T. dicoccoides* resistance to Alsen, a hard red spring wheat with the Sumai 3 source of resistance. The F_1 hybrid was backcrossed twice to Alsen and then pollinated with maize to produce doubled-haploid lines. These Alsen backcross-derived doubled-haploid (BC_2F_1DH) lines were initially screened with the SSR markers *Xgwm533* and *Xgwm2* for detection of the 3B QTL of Sumai 3 and the 3A QTL of *T. dicoccoides*, respectively. Additional STS markers were later used to verify the presence of these QTL. Phenotypic evaluation of these lines was done in three greenhouse seasons. A 10 µl inoculum with 50,000 spores ml^{-1} was injected into a single floret in the middle of the spike at anthesis. FHB resistance was assessed by measuring disease severity at 7, 14, and 21 d after inoculation (dai), percent *Fusarium*-damaged kernels (FDK), and DON content of the grain. A combined ANOVA indicated a significant genotype by dai interaction for disease severity. When means among genotypes were compared at the same dai, no significant differences were observed at 7 dai. However, a significant increase in disease severity was noted at 14 and 21 dai. The disease severity of Line 1 and Line 2 was not significantly different from Alsen, but both lines exhibited significantly lower disease severity than the synthetic parent at 14 dai. At 21 dai, Line 1 exhibited significantly higher disease severity than either Line 2 or Alsen. When comparisons were made within the same genotype across different dai, Line 1 exhibited a significant increase in disease severity, progressing from 5% severity at 7 dai to 17% severity at 21 dai. However, Line 2 exhibited no significant change, progressing from 5% severity at 7 dai to 8% severity at 21 dai. Alsen and the synthetic wheat exhibited a significant change in disease severity, progressing from 5% (7 dai) to 13% (14 dai) and 5% (7 dai) to 33% (14 dai) severity, respectively. The percent FDK of Line 1 and Line 2 was not significantly different from Alsen, but the FDK of both lines was significantly lower than the synthetic wheat. There were no significant differences in DON content for either Line 1, Line 2, or Alsen across greenhouse seasons, but the synthetic wheat had a significantly higher DON content in one of the three greenhouse seasons. Line 1 and Line 2 exhibited Type II resistance in all evaluations, and although differences between them were not always significant, in some instances, the combined effect of the 3A and 3B QTL in Line 2 may have contributed to a lower expression of FHB severity, percent FDK and DON content.

QTL ASSOCIATED WITH REDUCED KERNEL DAMAGE AND RESISTANCE TO FUSARIUM HEAD BLIGHT IN WHEAT.

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ABSTRACT

Resistance to Fusarium head blight (FHB) is controlled by a number of genes, suggesting that the identification and accumulation of multiple resistance genes in a variety would result in increased resistance. Our objective in this study was to identify QTL associated with resistance to FHB, especially QTL associated with a decrease in percent *Fusarium* damaged kernels (FDK). A population of 269 recombinant inbred lines (RILs) was developed from a cross between Patton and IL94-1653, and evaluated for FHB resistance in a mist-irrigated FHB nursery in Urbana, IL in 2006 and 2007. The parent lines, Patton and IL94-1653, are moderately susceptible to FHB, and IL94-1653 appears to exhibit resistance to kernel damage. The RIL population was genotyped using SSR markers and QTL analysis was performed using MapQTL 4.0. Preliminary results identified a QTL on chromosome 4B associated with reduced percent FDK in both years. The markers associated with this region are gwm513, gwm495, and wmc47. Chromosome 4B was also associated with DON content in 2007 but not in 2006. In 2006, a QTL on 2B was associated with reduced percent FDK, severity, FHB index, and ISK index. This QTL was not significant in 2007. Further genotyping and QTL analysis is in progress to identify additional resistance QTL in this population.

RESISTANCE TO KERNEL DAMAGE CAUSED BY FUSARIUM HEAD BLIGHT IN AN RIL POPULATION.

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ABSTRACT

Fusarium head blight (FHB) infection of wheat results in *Fusarium* damaged kernels (FDK) that contain the mycotoxin deoxynivalenol (DON), reducing the value of the grain. By developing lines with resistance to kernel damage resulting in a low percent FDK, breeders may be able to reduce DON content in grain. A population of 269 recombinant inbred lines (RILs) was developed from a cross between two soft red winter wheat lines, IL94-1653 and Patton, with IL94-1653 thought to exhibit resistance to kernel damage. Both parents are moderately susceptible to FHB. The RIL population was evaluated for FHB resistance in the greenhouse in 2005 and 2006, and in field in 2006 and 2007. A wide range of FHB symptoms and DON content were observed in the RIL population. The RIL population also exhibited transgressive segregation for all measures of FHB resistance, including disease severity, percent FDK, and DON content. In the field, the correlation between percent FDK and DON content was positive, and varied by year from moderately low to medium ($r = 0.38$ in 2006; $r = 0.53$ in 2007). The ISK index, a combination of severity, incidence, and percent FDK measurements, gave a better correlation with DON content for both years ($r = 0.47$ in 2006 and $r = 0.64$ in 2007), with all correlation values significant at $p<0.0001$. However, identifying RILs with consistent resistance to kernel damage was difficult and seems to be influenced by environmental variation as well as by resistance to initial infection and resistance to spread of infection.

BARLEY CHROMOSOME 2(2H) BIN 10 FUSARIUM HEAD BLIGHT RESISTANCE QTL: MAPPING AND DEVELOPMENT OF ISOLINES.

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INTRODUCTION

Development of commercially acceptable cultivars with Fusarium Head Blight (FHB) resistance and good agronomic qualities is the goal of the barley SCAB project. One of the best FHB resistance quantitative trait loci (QTL) resides in the chromosome 2(2H) bin 10 region. Our contributions are focused on genetic and physical mapping of this region with the long-term goal of cloning the genes responsible for FHB resistance. To facilitate this, we have isolated recombinant lines with introgressed small chromosome 2(2H) bin 10 genomic segments in a susceptible genomic background. We have also developed 6-rowed recombinants in the resistant CI4196 genomic background. To further facilitate development of agronomically acceptable barley cultivars with FHB resistance, we have undertaken to modify the resistant line CI4196 by mutagenesis. Mutants with desirable traits such as semi-dwarf, early and 6-rowed are easily selected. These provide improved FHB resistant parent material that can be rapidly incorporated in breeding programs.

RESULTS AND DISCUSSION

Genetic and physical mapping

We have developed an extensive genetic map for the bins 8-10 region (markers ABC306-MWG882). This map includes 43 loci, 111 markers, and 423 bacterial artificial chromosome (BAC) clones identified in our laboratory (the Tim Close barley physical map database increases this number to over 800). The details of this map will be published elsewhere. Here we focus on the bin 10 region from MWG699 to MWG503. This region has relatively few markers and has been difficult to saturate, as it has been for others who have published maps for this region (Marcel et al., 2006; Stein et al., 2007). Nevertheless, there are nine loci with 20 markers and 125 BAC clones identified in our laboratory (Fig. 1). There are a total of 295 BAC clones identified for this region if one includes those in the Tim Close barley physical map database. Some of those BAC clones likely belong to different loci, but nevertheless the physical map is fairly saturated. Based on just the BAC clones identified in our laboratory (because we are more confident that these really belong in this region), we have identified nine BAC clone contigs. We can reasonably exclude the two BAC contigs associated with the MWG503 locus because that region is far from the FHB resistance QTL (Horsley et al., 2006). The most likely FHB resistance peak resides around marker BF265762A just below the Vrs1 locus (Fig. 1). This region is covered by a completed BAC contig that also includes the next downstream marker BI955972. The area between Vrs1 and BF265762A has been fine-mapped by Pourkheirandish et al. (2007), providing further probes for picking BAC clones to build a complete contig. This region should be sequenced as soon as possible.

Isoline development

We have been developing small introgressed CI4196 FHB resistant line chromosome 2(2H) bin 10 region fragments into a susceptible cv. Morex background in order to more accurately define the region responsible for FHB resistance (Fig. 2). We now have identified three homozygous recombinant lines from the A171 x Morex cross that integrate either the region directly below Vrs1 (07-83-11) or a slightly larger region (07-76 and 07-84). An additional five homozygous lines

were developed from the cross A80 x Morex (07-85-1; 07-87; 07-90; 07-91; 07-97). These lines contain the BG369629 to BG416977 CI4196 region, which we do not think is important in FHB resistance. They also contain various small fragments introgressed from the Vrs1 to BG343659 region that appears to be critical to FHB resistance. All of these lines are being phenotyped in China winter '07-'08. A number of less advanced recombinant lines have already been tested for FHB resistance in China and North Dakota. Among them, line 06-310-18 stands out as being reproducibly FHB resistant and about the same height as Foster. This line is six-rowed, but the spike is not very robust, probably due to the presence of the two-rowed *Int-c* allele. This allele will be replaced by the mutant *int-c* allele that we have isolated from CI4196 (see below).

Mutant selection and analysis

All of the agronomically important CI4196 mutants that were identified in the field 2006 and 2007 were confirmed as CI4196 by PCR (Boyd et al., 2006). The mutants isolated in 2006 were phenotyped in China winter '06-'07 (Table 1). The male sterile mutants show good FHB resistance. These should be useful to facilitate backcrossing and breeding efforts. Early mutants (desirable for breeding) were identified and confirmed, but only two (G07-83 and G07-105) showed FHB resistance levels comparable to CI4196. The higher FHB susceptibility may be due to longer exposure of the early-emerging spike to the disease (Nduulu, 2007), or it may be the product of other unintended mutations. Of the two semi-dwarf mutants tested, only G07-66 is about the same height as Foster with FHB resistance comparable to CI4196. This line may be suitable as an improved CI4196 parent for FHB resistance breeding. Another line of potential interest is lax spike (G07-52). This line maintains CI4196 FHB resistance and is not excessively tall. The lax spike trait may be useful in reducing the opportunities for FHB infection. The intermedium line isolated in 2006 is not very promising and may be just a distorted head mutant. Two much more promising apparently intermedium type mutants were isolated in 2007 (see below).

New mutants identified in the field 2007 from a gamma irradiated population and sent for FHB phenotyping in China winter '07-'08 include intermedium, early, premature ripening, semi-dwarf, dwarf, erectoides, anthocyanin-less, and glossy head. The two intermedium types are particularly interesting. They have reasonably good 6-rowed heads and are probably of the *int-c* type (based on phenotype). The *Vrs1* gene was sequenced from these mutants and appears to be identical to the CI4196 *Vrs1* gene. Thus, unless the mutation is in a regulatory region, they are not *Vrs1* mutants. However, they will be very useful for crossing with the 06-310-18 line described above to recover a true 6-rowed head in a mostly CI4196 line. We are also very much interested in the FHB response of the new semi-dwarf and early mutants. Since these come from a gamma irradiated population, we expect that the genome may not be as highly rearranged as in the mutants selected from fast neutron irradiated material in 2006.

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DISCLAIMER

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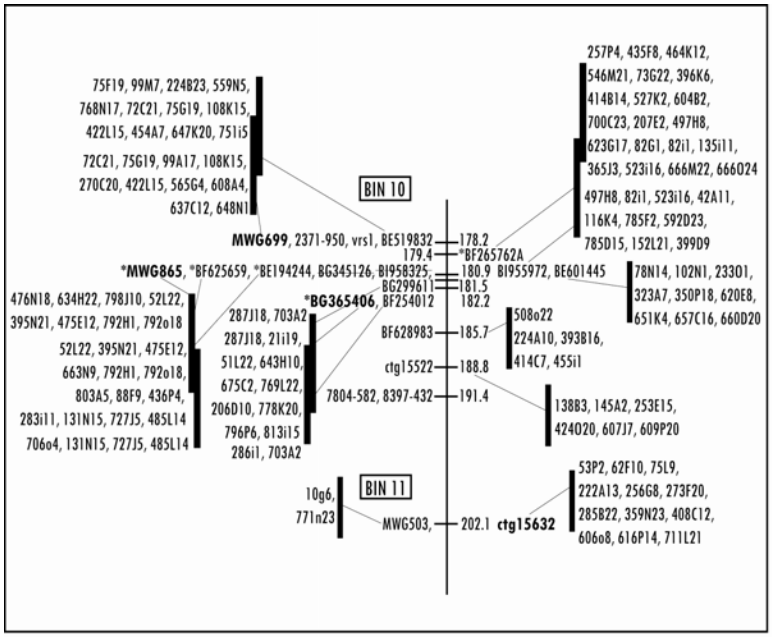

Fig. 1 Genetic map of Chr. 2(2H) bin 10 showing all markers currently mapped to the region as well as all BAC addresses picked by each marker listed next to the solid bars. Double solid bars indicate that the same contig (according to the Close database) is picked by different markers, creating a physical contig across the genetic region. Distances are in centimorgans based on a 94-line population. Bold = CAPs marker. Strikethrough = picked no BACs.

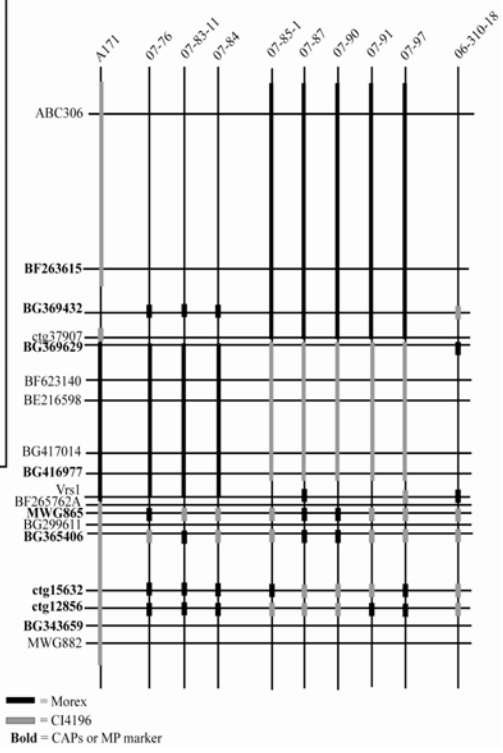

Fig. 2 Near-isogenic lines with introgression of CI4196 (gray) and Morex (black) in bin 10 (markers Vrs1 to ctg 15632). Markers in bold are either CAPs or multiplex (MP) PCR markers.

HAPLOTYPING OF KNOWN FHB RESISTANCE QTL IN PACIFIC NORTHWEST WHEAT GENOTYPES.

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ABSTRACT

This study was conducted to discern the genetic variation of Fusarium head blight resistance in PNW wheat genotypes via haplotyping of fifteen SSR and STS markers associated with six QTL for FHB resistance previously identified in known resistance sources. A total of 15 markers on six chromosome regions (2D, 3A, 3BS, 5AS, and 6B) are being used in 94 PNW wheat genotypes which have no Sumai 3 related backgrounds. Based on the preliminary results derived from haplotyping of six markers, we identified some of the known major FHB QTL present in the adapted PNW cultivars/lines. The frequency of the known QTL on 3BS and 6BS were higher than expected. The known target alleles for marker STS3B-256 on 3BS was present in 30 lines and the one for WMC 152 on 6BS was present in 45 lines out of the 94 genotypes studied. Evaluation of field FHB resistance of these genotypes is needed and they will be done in the 2007-08 growing season. This study has the potential to identify novel and adapted sources of resistance through allele size comparisons of known SSR loci associated with QTL identified in known resistance sources. Identified cultivars/lines having good field FHB resistance and/or known FHB resistance QTL can then be grown in PNW region and used as adapted resistance sources in the PNW and Great Plains breeding programs.

VALIDATION OF SIX QTLS ASSOCIATED WITH FUSARIUM HEAD BLIGHT RESISTANCE IN ADAPTED SOFT RED WINTER WHEAT.

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ABSTRACT

This study was conducted to validate molecular markers linked to six FHB resistance QTL previously identified in different bi-parental populations using elite breeding lines incorporated FHB resistance to initial infection, spread, and DON accumulation in different genetic backgrounds. A total of 129 SSRs were characterized in the 145 breeding lines. Forty-four unrelated SSRs (4 SSRs per chromosome) were used in background selection and the remaining 85 SSRs were used in validation of target QTL. The 145 wheat lines were also evaluated in yield performance trials at two locations, Blacksburg and Warsaw, VA, and for type I, type II, and DON resistance in a scab nursery at Blacksburg, VA in 2005 and 2006. Molecular markers linked to scab resistance genes located on wheat chromosomes 2BS, 2DS, 3AS, 3BS, 5AS, and 6BS were confirmed and allelic effects of associated marker loci were analyzed. Adapted resistant lines with novel alleles different from known exotic sources were characterized. Renwood 3260 and its derived lines have good overall resistance and high yield potential. These lines have unique resistance with alleles differing from those of known resistance sources W14 and Sumai 3 at marker loci Gwm429, Gwm120, Gwm261, Barc133, and Gwm186 in the chromosome 2BS, 2DS, 3BS, and 5AS QTL regions. Ernie and its derived lines also have good overall resistance but didn't produce promising grain yields in Virginia. These lines have unique resistance comprised of the same resistant alleles as Renwood 3260 at loci Gwm429, Gwm120, and Gwm261 in 2BS and 2DS QTL regions. Both the Ernie and Renwood 3260 derivatives contain the same resistant alleles as donor parent W14 at loci Wmc264, Barc133, and Barc117 in 3AS, 3BS, and 5AS QTL regions. In addition, these lines have unique resistant alleles in their background at Gwm493 and Wmc152 in 3BS and 6BS QTL regions.This is the first study validating six FHB QTL in elite breeding lines. QTL-markers validated in the current study have been used widely in parental selection, gene pyramiding, and in postulating and selection of FHB resistance of progeny derived from such newly developed FHB resistant lines. This is also the first study evaluating the effects of allelic differences and genetic backgrounds on FHB resistance. Newly developed FHB resistant lines with unique QTL/allele combinations have been used as parental lines in most of eastern wheat breeding program. Some of these lines will be released as varieties and/or adapted germplasm. The newly developed FHB resistant lines and unique QTL/marker allele profiles identified in this study will set the stage for using MAS not only for FHB resistance but also in combining FHB resistance with other important agronomic traits.

DEVELOPMENT OF SCAB RESISTANT SOFT RED WINTER WHEAT GERMPLASM USING MARKER-ASSISTED SELECTION.

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ABSTRACT

Scab of wheat, caused by *Fusarium graminearum*, is a disease that periodically strikes the US mid-Atlantic region. Breeding for resistance is an effective measure of disease control. The objective of this study was to develop scab resistant soft red winter wheat germplasm adapted to the US mid-Atlantic region using marker-assisted selection. McCormick, a genotype adapted to the Mid-Atlantic region, was used in a backcross program with the Chinese variety Ning7840. An accelerated backcross scheme was developed to incorporate scab resistance QTLs found on chromosomes 3BS, 5A and 2DL in the Chinese variety Ning7840. Two rounds of backcrossing were completed using McCormick as the female parent. Progenies from the first round of backcrossing were selected for the presence of the Ning7840 scab resistance alleles at 3BS, 5A, and 2DL, and then for a high background of McCormick alleles. Two backcross progenies had over 60% McCormick background. Using these two selected BC1F1s, 400 BC2F1s were produced in a second round of backcrossing. Additionally, the two selected BC1F1s were crossed with a wheat line with stripe rust resistance (GA96229-3A41). 800 BC2F1 seeds were screened with molecular markers to identify those with Ning7840 alleles (on 3BS, 5A and 2DL) and most McCormick background. A single BC2F2s population derived from a selected BC2F1 plant was screened with 3BS, 5AS, and 2DL markers to select those homozygous for the resistant alleles. Additionally, we derived near-isogenic lines from this F2 population to identify the effect of each QTL on scab resistance, agronomic and quality traits. We plan to test some of the BC2F3s for scab resistance in the spring of 2008. We anticipate having a small amount of seed of selected BC2F4s, containing the Ning7840 alleles in the McCormick background, available for distribution to other soft red winter wheat breeders for crossing in the fall of 2008.

APPLYING SINGLE KERNEL SORTING TECHNOLOGY TO DEVELOPING SCAB RESISTANT LINES.

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ABSTRACT

We are using automated single-kernel near-infrared (SKNIR) spectroscopy instrumentation to sort Fusarium head blight (FHB) infected kernels from healthy kernels, and to sort segregating populations by hardness to enhance the development of scab resistant hard and soft wheat varieties. We sorted 3 replicates of 192 samples into a damaged fraction yielding an average of 61.3 ppm DON, and a healthy fraction yielding an average of 0.73 ppm DON. This collaborative work with Dr. Gene Milus and Peter Horevaj investigated the resistance of soft red winter wheat lines to DON and NIV chemotypes of *Fusarium graminearum*. In another study, we also sorted the soft portion of a hard x soft cross into FHB infected and healthy fractions, and likewise sorted the hard portion into FHB infected and healthy fractions. The hard x soft crosses were separated into the hard and soft portions in 2006 where the respective portions were inoculated and planted. The 2007 scabby and healthy fractions of the hard and soft lines will be planted this fall to determine if our sorting will result in populations with FHB resistance. This work is in cooperation with Dr. Anne McKendry and Dr. Stephen Baenziger. Another project that was done in cooperation with Dr. Stephen Wegulo, Julie Breathnach and Dr. Stephen Baenziger used the automated SKNIR system to rapidly assess lines for FHB resistance by running multiple samples and obtaining a count of infected and healthy kernels. We have done this for about 300 lines and the information is being used to select resistant lines for further developement.

TUNISIAN DURUM WHEAT AS NEW SOURCES OF RESISTANCE TO FUSARIUM HEAD BLIGHT.

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ABSTRACT

There are limited sources of resistance to FHB which mostly restricted to Chinese hexaploid genotypes like Sumai3 and Wangshuibai. Therefore it is necessary to use other sources of resistance to expand the number of genes that may be used in the gene pyramiding programs. The North Dakota Durum Wheat breeding program has identified four tetraploid wheat sources of resistance from Tunisia, which were selected among a large number of lines evaluated over 55 repeated FHB trials. Since their identification, these lines have been extensively used in the breeding program to derive resistant breeding lines. We used a collection of backcross derived advanced resistant lines, susceptible sibs, and parental lines to identify markers that are associated with these novel sources of resistance. In this study we used 184 BC1F6 and 189 BC1F7 lines derived from crossing of Tun7, Tun18, Tun34, Tun36 with durum cultivars 'Ben', 'Maier', 'Lebsock' and 'Mountrail' for association studies. As Tunisian lines pedigree shows no relation to Chinese genotypes, they probably carry different genes or alleles for resistance to FHB. We checked all the parents and RILs in the greenhouse in two seasons for type II resistance to FHB by single floret injection inoculation method. The data showed that the Tunisian lines have different amount of resistance varying from 18% to 10% infection rate through the spikes. The data also showed Maier may have some minor resistance genes because it showed a moderate resistance in our greenhouse study and the crosses between Maier and different Tunisians had more transgressive resistant progenies compared with the other crosses.

To accelerate the identification of markers associated to FHB resistance, we initially screened the parents. The amounts of recombination in wheat chromosome arms are low so we picked 10-14 SSR markers per chromosome which were roughly 10cm apart and cover the whole genome. Among the 179 SSR markers that we applied on the parents about 45% showed polymorphism for at least two parents and about 8 % showed polymorphism between the whole set of Tunisian lines and susceptible cultivars. The most polymorphism was found on chromosomes 5A and 3B and the least on chromosome 6A. About 22 SSR markers that had been mentioned in different articles to be linked to FHB resistant were also applied to the parents. Among them barc117 and gwm129 from chromosome 5A showed the same pattern in Tunisian lines but not the susceptible lines. We also did the Diversity array (DArT) marker analysis to have a more complete coverage of the whole genome and to find closer markers to the genes of interest. DArT analysis used 2300 markers which showed 25% polymorphism between the parents. About 8% of the polymorphic markers were present in all the Tunisian lines but not the susceptible cultivars. The cluster analysis of the polymorphic markers revealed three distinct groups. Tun7 was in a separate group far from the other two and all the other Tunisian lines fell in a separate group from susceptible cultivars. Our data shows Tun7 and Tun18 are potential candidates for new sources of resistance which will be discussed in detail in our presentation.

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RESPONDING TO FUSARIUM HEAD BLIGHT FOR THE NORTHERN ROCKY MOUNTAINS AND WESTERN GREAT PLAINS.

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OBJECTIVES

To improve the efficiency of individual breeding programs' development of FHB resistance and minimize the impacts of associated diseases in irrigated systems.

INTRODUCTION

Fusarium head blight is a perennial disease problem for irrigated acreage in northern Rocky Mountains and western Great Plains. Montana, alone, has 150,000 acres of irrigated spring wheat with an annual production of 9.8 million bushels amounting to $50 million annually. All of this acreage is potentially impacted by this disease. Neighboring states in the northern intermountain region (Idaho, Washington, Oregon and Wyoming) have an additional 390,000 irrigated acres, all of which may be affected by FHB. FHB tolerant varieties have been utilized by producers in the past years but there are significant deficiencies which discouraged their continued use. In particular, varieties had problems with lodging, ergot and black chaff. In addition, current FHB tolerant varieties lack sawfly resistance, which is important to the western Great Plains region. The purpose of this proposal is to develop new lines specifically adapted for these areas and to determine the suitability advance FHB tolerant materials from other states for high-yielding, irrigated production.

MATERIALS AND METHODS

FHB Nursery - Wheat scab conditions were optimized with sprinkler center pivot irrigation, continuous cropping of wheat, and wheat residue serving as inoculum of *F. pseudograminearum* and *F. graminearum*. Residue management involved a fall cultivation with irrigation to germinate volunteer seed with residue remaining on the soil surface. Spring cultivation was a minimum tillage following by the planting with a no-till drill. Nursery was planted with a no-till small plot drill and seed was treated with Raxil-MD. Individual plots were 3.5 x 20 ft, 4 replications, planted May 9, 2006 and 3.0 x 12 ft, 3 replications planted May 8, 2007. Pre-plant, top dress and fertigation at soft dough was applied for potential yield of 90-100 bu. MCPA and Discover broadleaf herbicide and Quilt fungicide for leafspots during seedling stage. Best management practice to minimize FHB consisted of a Folicur fungicide (14 oz/acre) applied at anthesis (Feekes 10.5) and irrigation discontinued for nine days, July 7 to July 16. Normal sprinkler irrigation was on a daily cycle, 0.3 in/day, with 4.5 in up to flowering and then resumed on alternate days with a 4.5 in through grain development. Harvest completed with a small plot combine and chaff air volume was minimized to avoid loss of light, scabby grain. Spring wheat entries, 15 in total, were selected based on commercially available variety or advanced lines. Hank was entered twice as a susceptible check and for evalution of the uniformity of FHB distribution in the nursery.

Disease Evaluation - Scabs heads were visually determined by the pre-mature head blight and dark tan of the peduncle at ripening and hard dough stage. DON mycotoxin in the grain was processed according to standard protocols and evaluated by the NDSU Veterinary Diagnostic Lab. Germination blotter test for viability of seed and ergot quantity on percentage weight was conducted by the MSU Seed Analysis Lab. Lodging of the variety was determined as a visual assessment for the plot area at harvest.

RESULTS AND CONCLUSIONS

FHB Effects & Agronomic performance **-** The overall grain deoxynivalenol (DON) concentration was 2.2 ppm for 2006 and 2007 MSU FHB nursery. "Hank" is highly susceptible to FHB with a grain DON of 8.8 ppm, yield loss of 37%, test weight of 54 lb/bu and a head scab incidence of 58%. Other varieties without Sumai3 gene had 0.6 to 2.0 ppm grain DON, including; Vida, Howard, Granite, Explorer (HWW), Choteau and Expresso. Tolerant varieties had less than 0.5 ppm grain DON, including; Kuntz, Volt, Freyr, Granite, Knudson, Alsen, Glenn, Kelby, and an experimental line MT0550 (Choteau/ND709-9). Tolerant varieties had a 7% incidence of symptomatic scab heads as compared to 33% among varieties lacking the Sumai3 gene. Overall, varieties with the Sumai3 gene yielded 67 bu/ac or 18% higher than varieties lacking tolerance to FHB. Grain test weights were 62.3 lb/bu in the tolerant varieties and 57.8 lb/bu in the susceptible varieties. Seed germination by a blotter test was below an acceptable "92% for foundation class" on those varieties lacking tolerance to FHB. Several of the short statured varieties are adapted to high production under irrigation, but susceptible to FHB, whereas the FHB tolerant varieties will lodge under these conditions. Ergot and bacterial black chaff susceptibility of varieties are a concern for irrigated wheat production in these regions.

Breeding Lines **-**We have used molecular markers to backcross the Sumai3 QTL into Choteau, a solid-stem variety with resistance to the wheat stem sawfly, and MT0249, a variety with long green leaf duration and short stature. Molecular marker GWM 533 was utilized for selection in Choteau lines, such as MT0550, and this line is expressing tolerance to FHB. A similar marker selection line, MT0551, was removed from consideration following poor FHB tolerance in the 2006 nursery BARC 133 was used when MT0249 was a recurrent parent and there are several shorter statured lines that are under evaluation for FHB tolerance. We will have sufficient seed of for single screening and seed increase rows in the MSU FHB nursery in 2008. An FHB resistant line similar to Choteau will find utility in the western Great Plains and the northern Rocky Mountains.

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Table 1. Effects of Fusarium head blight on performance of spring wheat varieties under sprinkler irrigation in Montana during 2006 and 2007.

VARIETY	FHB reaction	Grain Yield 2 yr aver	Grain DON 2 yr aver	Test Wt 2 yr aver	Scab Heads 2 yr aver	Germ Blot Test	Lodge 1 yr 2006	Ergot 1 yr 2006
		Bu/ac	ppm	lb/bu	%	%	%	% wt
EXPLORER		53.2	2.2	55.4	13.6	83.6	37	0.00
HANK		56.7	8.8	53.5	53.9	57.3	0	0.00
HANK		59.3	8.4	54.2	48.9	57.1	0	0.00
HOWARD		65.6	1.1	60.2	21.6	86.6	64	0.04
VIDA		69.8	1.4	57.7	18.1	83.4	50	0.00
GLENN	Sumai3	70.7	0.2	63.9	3.1	96.6	31	0.01
ALSEN	Sumai3	70.7	0.2	62.2	5.9	95.6	10	0.14
GRANITE	Tolerant	75.0	0.4	62.3	16.2	93.9	5	0.07
EXPRESSO		76.7	3.6	59.9	25.8	81.1	5	0.00
MT0550	Sumai3	77.8	0.3	61.9	4.8	94.4	49	0.02
CHOTEAU*		77.9	2.8	60.8	21.5	na	na	na
KELBY	Sumai3	80.8	0.5	61.8	16.6	92.6	24	0.02
KNUDSON	Sumai3	83.8	0.4	60.3	5.6	92.6	63	0.02
KUNTZ	Sumai3	85.9	0.4	61.3	11.9	94.9	16	0.01
VOLT	Tolerant	87.6	0.5	62.4	5.6	95.6	8	0.02
FREYR	Sumai3	87.6	0.3	61.5	6.3	91.3	78	0.03
Lsd P<0.05						3.5	22	Ns
C.V.%						2.8	49	
*Choteau 2007								

RESISTANT GERMPLASM FROM SUSCEPTIBLE PARENTS: AN EVOLUTIONARY APPROACH.

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ABSTRACT

The Chinese wheat line Sumai 3 is widely considered the standard for resistance to Fusarium Head Blight (FHB). Much research effort aims to introgress its FHB resistance into germplasm with better quality, agronomic traits, geographic adaptation and resistance to other diseases. As several, or perhaps many, genes are involved in expressing the trait, it has proved difficult to combine high FHB resistance with desirable agronomic and quality traits while advancing through cycles of crosses with elite, FHB-susceptible parents. Instead of aiming to transfer the complement of genes that condition FHB resistance in Sumai 3, we pursued an alternative approach of emulating in our desired germplasm the evolution of resistance that had occurred in Sumai 3 itself as it derived from moderately susceptible parents

Evolution within a population can be speeded by enhancing variation, selection and generation of fertile progeny among its individual members, and is more readily followed using small populations that can be carefully examined and advanced through as many as 4 generations per year (3 indoors and 1 field trial). After initial crosses with lines resistant to wheat streak mosaic virus (WSMV), we identified and selected in each generation of back-crossing regimes those individuals that appeared to combine vigorous growth under pressure from virus infection with the best resemblance to the recurrent cultivar parent. After the BC2 generation, lines that consistently performed well under pressure from WSMV infection were spray-inoculated with macroconidial suspensions of *Fusarium graminearum* and the most promising individuals selected for additional backcrossing and selection.

We had chosen a Canadian amber durum spring wheat cultivar, Strongfield, to apply this novel approach of 'speeded evolution', as the germplasm in this wheat class is highly susceptible to FHB and there are no well-characterized tetraploid wheat sources of resistance that might readily be introgressed. To date, we have generated lines equivalent to BC3F4-6, and with repeated backcrosses and selection, the lines have increasingly come to resemble Strongfield in all desirable agronomic traits. In 2007, BC3F4 and BC3F5 lines were evaluated in FHB nurseries. Families of lines were observed with excellent FHB reactions (similar to the most resistant hexaploids) that were consistent with results seen in predecessor generations selected in indoor tests. The 'speeded evolution' approach allows only small quantities of seed to advance from each generation of highly selected germplasm, precluding testing for quality until disease reactions and agronomic traits are consistently good. Initial assessments, however, of gluten index of selected lines of BC3F5 and BC3F6 seed harvested from 2007 FHB nursery plots indicate several of these FHB-resistant lines have acceptable quality.

RESISTANCE OF WINTER WHEAT LINES TO DEOXYNIVALENOL AND NIVALENOL CHEMOTYPES OF *FUSARIUM GRAMINEARUM*.

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OBJECTIVES

The objectives of this research were to determine if wheat lines selected for resistance to Fusarium head blight (FHB) caused by deoxynivalenol (DON) chemotypes also have resistance to the nivalenol (NIV) chemotypes and to determine which lines appear to have resistance to mycotoxin accumulation in the grain.

INTRODUCTION

Even after several decades of intensive research on FHB of wheat caused by *Fusarium graminearum*, mycotoxins produced by this pathogen still cause serious concerns for food and feed safety. The most prevalent mycotoxins produced by *F. graminearum* are DON and NIV. Strains that produce mainly DON (DON chemotypes) predominate in the U.S. (Chandler et al. 2003). However, strains that produce mainly NIV (NIV chemotypes) were found recently in Louisiana and Arkansas (Gale et al. 2005 and 2007), and NIV is ten times more toxic to humans than DON (Ueno and Ishii 1985). Developing resistant wheat cultivars is perceived to be the most effective means for managing FHB and reducing levels of mycotoxins in grain. The presence of both DON and NIV chemotypes in the Midsouth necessitates having resistance to both chemotypes in wheat cultivars adapted to this region.

MATERIALS AND METHODS

A susceptible check and 15 resistant winter wheat lines (Table 1) were grown in the greenhouse. Heads were inoculated at flowering with two DON and two NIV chemotypes of *F. graminearum*. Inoculum (20µl at 5.10^4 cfu/µl) was injected into one floret of each head, and plants were misted for 72 hours to promote infection. The number of infected florets per head was counted 21 days after inoculation, and the percentage of infected florets (%IF) was calculated. Heads were harvested at maturity and threshed by hand to retain all of the rachis tissue and grain. Grain and rachis tissue from heads within a pot were bulked, and each bulked grain sample was separated into "healthy" and "scabby" fractions by the USDA ARS Grain Marketing and Production Research Center at Manhattan, KS. Healthy and scabby grain and rachis tissue were ground and sent to the University of Minnesota for mycotoxin analyses using GC/MS.

The statistical model was a full factorial with 16 lines, two DON chemotypes, two NIV chemotypes, and three replications (pots). Analysis of %IF was based on three experiments, and analyses of toxin concentrations were based on two experiments because there was little variation for toxin concentration in the third experiment. Data were analyzed using JMP version 7.0. Data were transformed before analyses to achieve homogeneity of variances using the most appropriate transformation suggested by the software. Means were back-transformed for presentation of results.

RESULTS AND DISCUSSION

Percentage of infected florets (%IF). The two isolates within each chemotype caused similar levels of disease, and data were pooled by chemotype for analysis. The line ´ chemotype interaction was not significant (P=0.0713), indicating that lines ranked similarly for both chemotypes. The two DON isolates averaged 16.9% IF and caused significantly more disease (P<0.0001) than the two NIV isolates that averaged 11.9% IF. All resistant lines had significantly lower %IF than the susceptible check for both chemotypes, and lines Fg 368, ARGE97-1033-10-

2 and VA04W-433 had the lowest %IF (Table 2). Wheat lines with resistance to isolates of the DON chemotype were even more resistant to isolates of the NIV chemotype, and therefore selecting lines for resistance to the DON chemotype should also select for resistance to the NIV chemotype.

Toxin concentration in grain and rachis tissue. Averaged across 16 wheat lines in experiments 1 and 2, the two isolates of the NIV chemotype primarily produced NIV and little to no DON, 3-ADON, or 15-ADON, and the two isolates of the DON chemotype primarily produced DON and little to no NIV, 3-ADON, or 15-ADON in both grain and rachis tissue (Table 3). Therefore, all comparisons of toxins only considered NIV for the NIV chemotype and DON for the DON chemotype. The two isolates within each chemotype produced similar levels of mycotoxins (Table 3), and data were pooled by chemotype for analysis.

Averaged across 16 wheat lines in experiments 1 and 2, toxin concentrations for the two NIV and two DON isolates were 0.25 ppm NIV and 0.73 ppm DON, respectively, in the healthy grain fraction and 25.71 ppm NIV and 61.30 ppm DON, respectively, in the scabby grain fraction. These results indicate that the grain sorting procedure effectively sorted grain into healthy and scabby fractions.

There were significant wheat line ´ chemotype interactions for toxin concentration in grain ($P<0.0001$) and in rachis tissue ($P=0.02$). For each line, however, the NIV concentration was always less than the DON concentration (Table 2), indicating that the interactions were due only to the magnitude of the differences between DON and NIV concentrations. Lines ARGE97-1033-10-2 and VA04W-433 had the lowest concentrations of toxin in both grain and rachis tissue and were among the most resistant lines as measured by the percentage of infected florets (Table 2). Both lines have the cultivar 'Freedom' in their pedigree, and Freedom may have contributed genes for resistance.

This report is based on a preliminary analysis of the data. Additional statistical models and transformations will be evaluated to determine which are the most appropriate, and data for healthy and scabby grain fractions will be analyzed to determine if these data are useful for characterizing resistance to FHB and mycotoxin accumulation.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

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Table 1. Winter wheat lines used in this study, their pedigree and contributors.

Line	Pedigree	Contributor
ARGE97-1033-10-2	FREEDOM/CATBIRD	Milus
ARGE97-1042-4-5	MASON / CATBIRD	Milus
ARGE97-1047-4-2	P2643 / 3 NING 7840 // PARULA / VEERY # 6	Milus
ARGE97-1048-3-6	MASON // SHA 3 / CATBIRD	Milus
ARGE97-1064-13-5	MASON//FREEDOM/SUPER ZLATNO	Milus
VA04W-433	NING 7840/PION2684//96-54-244 (CK9803/FREEDOM)	Griffey
VA04W-628	ERNIE//NING7840/ERNIE	Griffey
ROANE	VA71-54-147(CI17449)/Coker68-15//IN65309C1-18-2-3-2	Griffey
AR97002-2-1	AR396-4-2/NING 8026	Bacon
BESS	MO 11769/MADISON	McKendry
NC03-11465	NING 7804/P2643//NC95-22426	Murphy
COKER 9835	Susceptible check	Check
SZ 13	Ringo Star / Nobeoka Bozu	Mesterházy
SZ 14	Ringo Star / Nobeoka Bozu	Mesterházy
Fg 365	Sgv / Nb / MM / Sum3	Mesterházy
Fg 368	Zu / Re / Nobeoka Bozu	Mesterházy

Table 2. Toxin concentrations in grain and rachis and percentage of infected florets for each winter wheat line.

Line	Toxin concentrations (mg/kg) [1]								Infected florets (%) [1]	
	grain[2]				rachis[3]					
	DON for DON chemotype		NIV for NIV chemotype		DON for DON chemotype		NIV for NIV chemotype			
ARGE97-1033-10-2	0.54	fg	0.37	cd	75.61	d	13.97	c	6.20	e
VA04W-433	0.65	fg	0.29	cd	73.84	d	21.44	bc	6.42	e
VA04W-628	1.39	fg	0.58	bcd	119.33	cd	26.94	bc	9.43	de
AR97002-2-1	1.72	defg	0.95	bcd	136.91	bcd	30.69	bc	10.00	de
ROANE	1.76	efg	0.25	d	165.66	bcd	24.54	bc	9.52	de
BESS	1.97	cdef	0.72	bcd	138.61	bcd	28.24	bc	8.31	de
Fg 368	2.88	cdef	0.82	cd	175.37	bc	28.03	bc	5.74	de
SZ 14	7.93	bcde	0.60	cd	226.49	bc	37.86	bc	10.60	cde
Fg 365	8.57	bcd	4.69	ab	238.08	b	56.29	b	13.24	bcd
NC03-11465	11.35	bcd	0.95	bcd	192.73	bc	36.01	bc	15.57	bc
SZ 13	11.81	bcde	0.66	cd	229.72	bc	45.82	bc	9.45	bcd
ARGE97-1064-13-5	13.19	bc	1.41	bcd	223.83	bc	30.82	bc	16.90	bc
ARGE97-1047-4-2	27.41	b	3.80	abc	201.90	bc	40.91	bc	17.65	b
ARGE97-1042-4-5	31.53	b	9.96	a	217.24	bc	53.69	b	20.46	b
ARGE97-1048-3-6	40.90	b	1.40	bcd	201.11	bc	23.53	bc	15.44	b
COKER 9835	178.05	a	12.18	a	438.73	a	172.01	a	48.13	a

[1] Values within a chemotype followed by the same letter are not significantly different by a Tukey's HSD test at P=0.05.

[2] Data were transformed for statistical analyses by the formula: Log(:Tox.Con. in grain) * 1.10101230397926 ; however, values represent actual back-transformed LS means for each line and variable.

[3] Data were transformed for statistical analyses by the formula: (:Name("TOX_Rachis (ppm)_F") ^ 0.4 - 1) / 0.0350989281248155 ; however, values represent actual back-transformed LS means for each line and variable.

Table 3. Toxin concentrations in grain and rachis for each isolate of *Fusarium graminearum* DON and NIV chemotypes (averaged across 16 wheat lines and two experiments).

		Toxin concentrations in harvested grain[1, 2]				Toxin concentrations in rachis[1, 3]			
Chemotype	Isolate	DON	NIV	3-ADON	15-ADON	DON	NIV	3-ADON	15-ADON
		mg/kg							
NIV	03-29	0.06a	1.93a	0.00a	0.00a	1.74a	40.10a	0.10a	0.00a
	03-112	0.08a	3.03a	0.00a	0.00a	0.24a	43.75a	0.01a	0.00a
DON	03-57	17.69a	0.07a	0.07a	0.00a	199.78a	0.51a	13.93a	0.59a
	03-113	25.01a	0.10a	0.11a	0.00a	182.11a	0.40a	13.67a	0.46a

[1] Values within a chemotype followed by the same letter are not significantly different by an Student's t – test at P=0.05.

[2] Data were transformed for statistical analyses by the formula: Log(:Tox.Con. in grain) * 1.10101230397926 ; however, values represent actual back-transformed LS means for each isolate and variable.

[3] Data were transformed for statistical analyses by the formula: (:Name("TOX_Rachis (ppm)_F") ^ 0.4 - 1) / 0.0350989281248155 ; however, values represent actual back-transformed LS means for each isolate and variable.

CURRENT STRATEGIES FOR BREEDING FUSARIUM HEAD BLIGHT RESISTANT WHEAT IN CANADA.

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ABSTRACT

Fusarium head blight (FHB) is a destructive fungal disease of wheat that annually results in yield and grade losses for producers as well as reduced feed and end-use quality for the wheat industry. Efforts to develop FHB resistant wheat cultivars can be roughly divided into three research areas: (1)Germplasm development; (2)Molecular breeding; and (3)Line development and evaluation. Non-Canadian sources of FHB resistance, from Brazil, China, CIMMYT, Germany, Japan and the USA, are being used to develop FHB resistant material suited to Canadian growing conditions and registration requirements. FHB screening in inoculated nurseries has been established across Canada permitting evaluation of germplasm and breeding materials in multiple environments. Novel germplasm is being developed through *in vitro* selection of wheat microspores for tricothecene resistance. Molecular breeding strategies have been used to develop new breeding materials that combine resistance genes from multiple sources in improved backgrounds. Fine mapping of genes *fhb*1 and *fhb*2 has facilitated screening for FHB resistance in parental, *in vitro* selected, backcrossed, and doubled haploid lines. High throughput screening technologies such as DNA extraction robotics and multi-channel capillary electrophoresis permit the screening of multiple markers. In our wheat breeding program, lines are regularly screened for FHB resistance loci on chromosomes 3BS, 5A, 6BS, and 2D. Haplotype analyses of breeding materials at these loci permit selection of FHB resistant lines for advancement and crossing. Future breeding efforts will focus on traits which reduce deoxynivalenol content, and the mapping and deployment of non-Asian FHB resistance.

EFFECTS OF AGRONOMIC AND MORPHOLOGICAL CHARACTERS ON FHB SEVERITY, DEOXYNIVALENOL AND ERGOSTEROL CONCENTRATIONS IN NEAR-ISOGENIC LINE PAIRS OF BARLEY.

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ABSTRACT

Fusarium head blight (FHB) is a devastating disease on barley. Several agronomic traits have been shown to be associated with lower FHB severity in barley. To evaluate the relationship between different agronomic and morphological traits and FHB levels, we examined FHB severity, deoxynivalenol (DON) and ergosterol concentrations in 20 pairs of near-isogenic lines (NILs) carrying the traits two-rowed/six-rowed, lax/dense, erect/normal, club/normal spike, hulled/hulless caryopsis, fertile/infertile lateral florets, and early maturity/late maturity. These lines were planted in FHB disease nurseries at Langdon, ND, Fargo, ND, St. Paul, MN, and Crookston, MN in 1995, 2000, 2005, and 2007. Inoculations were applied either by spreading *Fusarium graminearum* infected barley kernels over the nursery for consecutive weeks starting 10 days prior to spike emergence or by spraying macroconidia inoculum once after head emergence. The FHB severity (number of FHB infected kernels out of total number of kernels per spike) was evaluated on 10 or 20 randomly selected spikes in each replicate at the mid-dough stage of development. After harvest, the concentrations of DON and ergosterol were assessed in random 3 gram grain samples of each replicate. Differences between the means of NILs for levels of FHB severity, DON and ergosterol concentrations were analyzed for statistical significance using the paired t-test. The results will be presented in the poster. Overall, few statistically significant and consistent differences were observed for the scored disease parameters (FHB severity, DON and ergosterol concentrations) on the NIL pairs. However, in general loci controlling six-rowed and six-rowed like spike phenotypes exhibited more disease symptoms.

FUSARIUM HEAD BLIGHT (FHB) RESISTANCE INTO SOFT RED WINTER WHEAT AGS2000.

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ABSTRACT

Fusarium head blight (FHB) is a potential devastating disease in the southeast region in the United States where low temperature and misted weather occurs frequently during soft red winter wheat flowering. Releasing new cultivars resistant to FHB is the most effective option to minimize the chance of FHB incidence and reducing DON contamination. Crosses were made since 2001 between AGS2000 or its derivatives and FHB resistant donor VA01-461 to introduce the exotic resistant genes into our widely local adaptive genetic background. Twelve advanced lines, 941523-E21, 991109-6E8, 991109-6A7, 991371-6E12, 991371-6E13, 031454-DH7, 031454-DH31, 031307-DH6, 031307-DH14, 031354-DH30, 981621-5E34, 951306-2E13, derived from VA01W-461, which is a derivate of Sumai3, were evaluated in scab nursery and field in 2006 and 2007 for FHB resistance and agronomy performances with Ernie and Coker 9835 as resistant and susceptible control respectively under misted conditions in Griffin-Campus, Georgia. DNA markers, XGWM533, BARC133, XGWM493, STS3B-256 for QTL on 3BS; BARC117, XGWM156, BARC186, BARC56, for QTL on 5AS; BARC18, and BARC91 for QTL on 2BS were employed to genotype 12 new lines with the donor parent of VA01W-461. Here, we reported the results of DNA genotyping and performances of our elite lines. The scab resistance and QTLs in VA01-461 are discussed in this study.

CHARACTERIZATION OF RESISTANCE TO DEOXYNIVALENOL (DON) ACCUMULATION IN DIFFERENT WHEAT LINES.

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ABSTRACT

ANOVA results suggested highly significant differences among wheat genotypes for DON accumulation (F= 22.72, P< 0.0001). As expected, disease and genotype × disease interaction for resistance to DON accumulation were also significant: F=329.66 and 15.98, respectively, P<0.0001. In the combined analysis for DON accumulation: replicate, collection date, rep × genotype, collection date × genotype, and genotype × collection date × disease interaction effects were not significant: F= 0.166-0.382, P= 0.202-0.841. Significant variation for DON content was observed between 'healthy + diseased' seeds and 'healthy' seeds. But, no significant difference was observed in the healthy seeds between resistant and susceptible genotypes. However, for the DON content in the 'healthy + diseased' samples, the FHB-resistant or moderately resistant genotypes including 0128A1, INW0411, INW0412, Bess, Freedom, and Truman, exhibited lower DON accumulation than FHB-susceptible cultivars Patterson and Pioneer2545. Patterson and Pioneer2545 both are susceptible to FHB, but, on average, the DON content for Patterson (12.4 ppm) was only approximately half of that of Pioneer2545 (21.2 ppm).

DEVELOPMENT OF CIMMYT'S 11TH SCAB RESISTANCE SCREENING NURSERY.

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ABSTRACT

CIMMYT has regularly developed and distributed a Scab Resistant Screening Nursery (SRSN) over the past decade. These nurseries have consisted of the best scab resistant material identified through CIMMYT's FHB screening trials and have been distributed to interested programs around the world upon request. The most recent nursery distributed was the 10th SRSN, which was made available in 2006. Since that time CIMMYT's method for screening FHB has been modified for more effective identification of FHB resistant germplasm. These changes have included modifications in the location of the screening nursery, isolates used for inoculation, inoculation technique and misting technology. After two years of screening a range of materials using the modified methodologies, entries for the 11th SRSN have been identified. This nursery primarily includes the best FHB resistant advanced lines developed by the CIMMYT wheat breeding programs. The 11th SRSN will be available for distribution in 2008.

PRELIMINARY EXAMINATION OF THE INFLUENCE OF GRAIN COLOR IN FHB RESISTANCE.

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ABSTRACT

Breeding programs around the world have seen noteworthy improvements in FHB resistance in recent years. However, there is the perception that white wheat lags behind red wheat in the development of resistant varieties. In our study, we asked the question whether or not grain color itself is influencing the amount of disease development. Thirty-six genotypes from fourteen sibling groups originating from different breadwheat crosses were examined. Each sibling group was comprised of at least one red and one white sibling pair. In 2006 and 2007, genotypes were screened for FHB resistance in a four replication incomplete block design at CIMMYT headquarters, Mexico. Plots were spray inoculated at anthesis and three days following, and were rated for % severity and % incidence at thirty-one days post inoculation. Post harvest, DON levels of the 2006 samples were evaluated via ELISA. Preliminary results of this study will be shared.

FHB RESISTANCE AND DON CONTAMINATION IN VIRGINIA BARLEY.

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ABSTRACT

Knowledge of FHB resistance and DON contamination in Virginia barley is essential for providing growers and producers with new and improved commercial cultivars. In 2006 and 2007, we screened 12 hulless (HLS) and 19 hulled (HLD) lines of barley in inoculated, mist-irrigated field plots at Blacksburg, VA for FHB incidence, FHB severity, and DON contamination. In 2006 and 2007, FHB incidence ranged from 40% to 80% for HLS lines and from 30% to 90% for HLD lines, and FHB severity ranged from 4.0% to 12.5% for HLS lines and from 6.1% to 33.5% for HLD lines. In 2006, DON concentrations ranged from 0.1 to 2.0 ppm for HLS lines and from 0.5 to 11.5 ppm for HLD lines; in 2007, DON concentrations ranged from 0.2 to 2.4 ppm for HLS lines and from 0.1 to 3.3 ppm for HLD lines. In 2006, FHB incidence was correlated with DON for HLS lines ($r = 0.92$, $P < 0.001$) and HLD lines ($r = 0.48$, $P < 0.05$); in 2007, FHB incidence was not significantly correlated with DON for HLS lines ($r = 0.14$, $P = 0.6$) or HLD lines ($r = -0.16$, $P = 0.5$), but FHB development was generally low in 2007 in VA. DON concentrations in 100 g kernel lots were correlated among DON testing labs in MN (Dong), ND (Schwarz), and VA (Schmale) (range of r from 0.81 to 0.89, $P < 0.001$). HLS line VA01H-125 had the lowest level of DON contamination in both years (0.2 ppm in 2006 and 2007), and HLD line VA92-42-46 had relatively low levels of DON contamination in both years (1.0 ppm in 2006, 0.36 ppm in 2007). We are continuing to develop and test new cultivars of barley in VA for FHB resistance and reduced DON potential.

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META-ANALYSES OF QTL ASSOCIATED WITH FUSARIUM HEAD BLIGHT RESISTANCE.

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OBJECTIVES

The objective of this study is to estimate the CI's of 63 QTL associated with different types of FHB resistance using meta-analyses and align them onto the consensus ITMI maps to determine if different QTL on the same chromosomes from different studies overlap.

INTRODUCTION

Many QTL have been mapped on to chromosomes of resistant sources from China, Japan, Brazil, USA and various European countries. This paper reviewed 63 QTL from 23 studies including four QTL identified for type I FHB resistance (resistance to initial infection), 50 QTL for type II resistance (resistance to spread), six QTL for type III resistance (resistance to DON accumulation) and three QTL for type IV resistance (resistance to kernel damage). Among these studies, 10 chromosomes were identified that have more than two FHB QTL regions. Each QTL explained more than 10% of the phenotypic variation in the corresponding experiment was included in the analyses. Some QTL identified from the same resistance source mapped onto the same chromosome but were not linked to the same marker loci. Furthermore, some QTL from different sources mapped in proximity to each other on the same chromosome. Differences in marker orders across studies make it very difficult to select markers linked to respective QTL from individual studies.

Meta-analyses have been used to estimate the confidence intervals (CI) of identified QTL in plants and animals (Wise et al., 1999; Guo et al., 2006). It combines the information for each individual study to estimate QTL CI which can be aligned on the consensus map. This permits the QTL position to be compared to determine if they overlap. This may provide some distinguishable common flanking markers linked to different QTL that can be used more effectively in MAS breeding.

MATERIALS AND METHODS

Estimate CI of QTL - The following formulas were used to estimate the 95% CI of each QTL:

$CI = 530 / (N*R^2)$ for backcross [1]
$CI = 530 / (N*R^2)$ for F_2 intercross [2]
$CI = 163 / (N*R^2)$ for RILs [3]
$CI = 132 / (N*R^2)$ for DH [4]

Where N is the number of lines in the mapping population and R^2 is the percentage of phenotypic variation explained by the identified QTL. Formulas [1] and [2] were deduced by Darvasi and Soller (1997) through simulation. The CI formulas [3] and [4] for RIL and DH, respectively, were derived via further detailed characterization of genetic parameters (Weller and Soller, 2004; Weller, 2007, personal communication).

Classification of FHB resistance QTL - Criteria similar to those reported by Guo et al. (2006) were used in the current study to classify QTL into three classes: (i) suggestive QTL if LOD < 4.0 (or p value > 0.0001), (ii) significant QTL if LOD e" 4.0 (or p value d" 0.0001) and, (iii) confirmed QTL if the QTL was identified in two or more separate studies (Lander and Kruglyak, 1995) and was significant in at least one study.

Meta-analyses of marker-QTL associations - Where the estimated CI of QTL regions overlapped, those QTL were grouped into one cluster. QTL alleles

within the same cluster were assumed to be the same (Guo et al., 2006). QTL were classified into different clusters if none of the estimated CI regions overlapped and were more than 20 cM apart.

RESULTS AND DISCUSSION

Significant and confirmed type II resistance QTL in the same cluster - Significant QTL from different sources but in the same cluster were distributed on chromosomes 3AS, 5A, 7AL, 3BS, 4B, 6B, and 2DS (Fig. 1). Separate mapping studies of several derivatives of Sumai3 (W14, CM82036, DH181, and Ning7840) provide evidence of a confirmed type II FHB resistance QTL located on chromosome 3BS (Fig. 1). Another confirmed type II FHB resistance QTL also located on chromosome 3BS is derived from Wangshuibai. The third confirmed QTL is on 5A of CM82036 and W14, two Sumai 3 derivatives (Fig. 1). These are confirmed and significant type II FHB resistance QTL, present in different sources and located in the same position along the respective chromosomes Markers flanking the most common regions of these QTL's CI can be applied in MAS to increase the efficiency.

Significant type II resistance QTL in different clusters - The type II resistance QTL of Renan is on chromosome 5AL, which is different from another cluster located around the centromere of chromosome 5A in other sources, such as CM82036, Ernie, Frontana and W14 (Fig. 1). Other type II resistance QTL have been located on chromosome 1BS of Fundulea 201R and 1BL of Wangshuibai and Arina (Fig. 1). A second type II resistance QTL in Renan was mapped near the centromere of chromosome 2B while the QTL in Dream is located on distal region of the same chromosome (Fig. 1). A QTL close to centromere of chromosome 3B in Ernie is in a separate cluster compared to the primary QTL located in the distal region of chromosome 3BS of Sumai 3 and its derivatives. The type II resistance QTL on chromosome 5BS of Wangshuibai does not overlap with the QTL of Arina on chromosome 5BL. These QTL belong to different clusters and, therefore, should provide different FHB resistance alleles for breeding. The flanking markers identified in the original studies should be validated to confirm their effectiveness in MAS prior to using them for pyramiding these QTL. The application of respective tightly linked markers to pyramid these QTL should be effective to breed durable FHB resistances.

Different Types of FHB Resistance QTL in the Same Sources and Clusters - Types I and II resistance QTL were found on chromosomes 3AS in Frontana, 5A in W14, and 4B in Wangshuibai. Types II, III, and IV resistance QTL were identified on chromosomes 3BSc and 5A in Ernie while types I, II, III, and IV resistance QTL were discovered on 3BS in W14, a Sumai 3 derivative (Fig. 1). QTL conferring different types of FHB resistance were identified and located in the same clusters suggesting a pleiotropic effect or association among them. FHB resistance types other than type II have been evaluated only in a limited number of resistant sources and environments with most of them being greenhouse studies. Therefore, the common QTL reported for these different types of resistance (Chen et al., 2006; Abate et al., 2007; Liu et al., 2007) need to be proved with more evaluation in additional sources and genetic backgrounds, and in studies specifically designed to assess and distinguish resistance types I, III and IV. Such studies are needed to elucidate whether these QTL have pleiotropic effects or if their interrelatedness is simply a function of the highly correlated effects that FHB assessment methods, particularly single floret point inoculation, has on multiple types of FHB resistance.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the

author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

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Figure 1a.

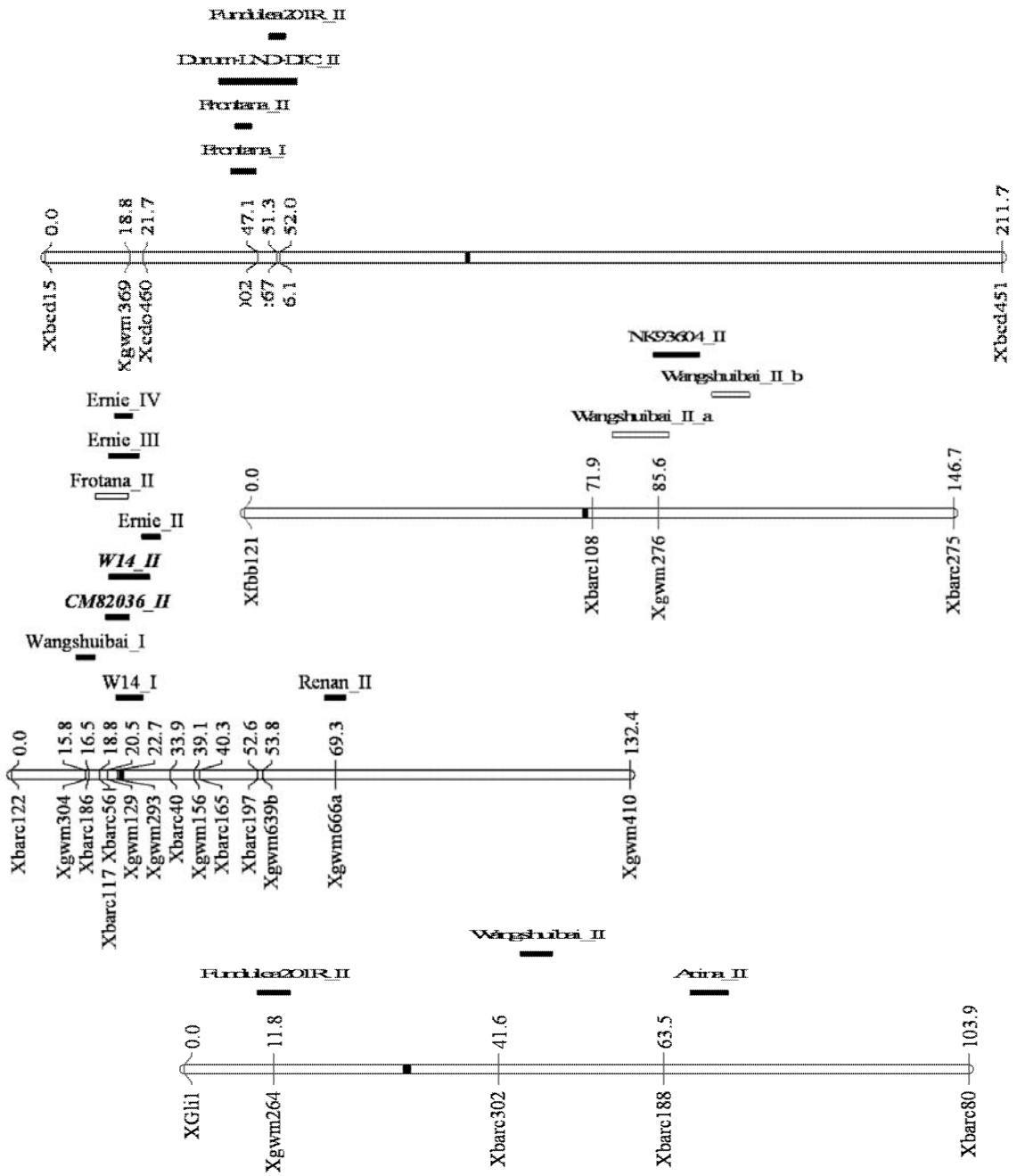
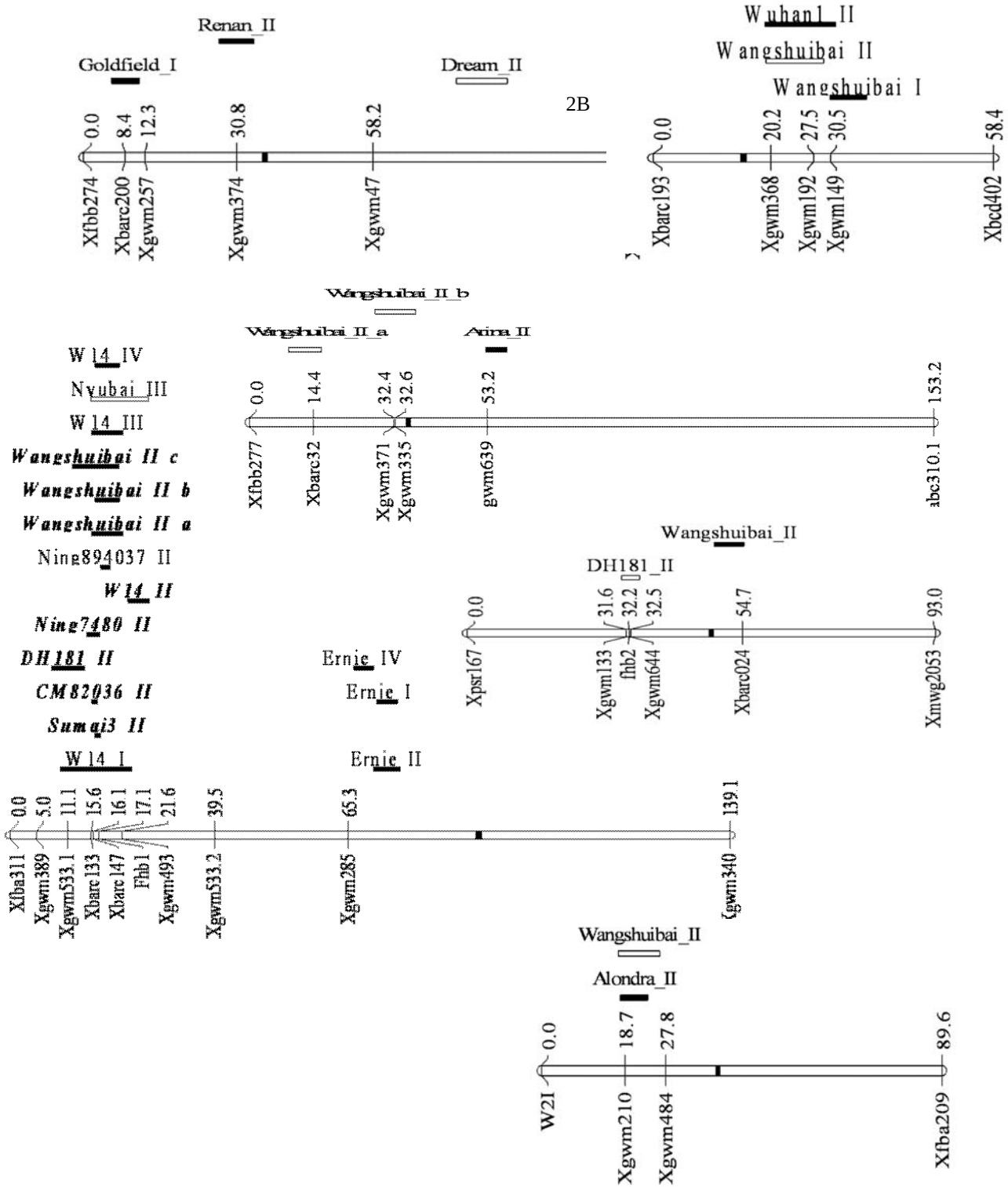


Fig. 1. The 95% confidence intervals of QTL associated with different types of Fusarium head blight (FHB) resistance. Significant QTL are represented by solid black bars (LOD > 4 or p <0.0001) above the respective chromosome region; suggestive QTL (LOD < 4 or p > 0.0001) are represented by open bars and; confirmed QTL are shown as solid black bars with bold and italic font-type. The QTL name is the source followed by an underscore and a Roman number which indicated the type of FHB resistance identified. The frame of ITMI genetic chromosome maps from Song et al. (2005) was used with the consensus map from Somers et al. (2004) as reference.

Figure 1b.


Renan_II
Goldfield_I
Dream_II
2B
Wuhan1_II
Wangshuibai II
Wangshuibai I
0.0
8.4
12.3
30.8
58.2
Xfbb274
Xbarc200
Xgwm257
Xgwm374
Xgwm47
0.0
20.2
27.5
30.5
58.4
Xbarc193
Xgwm368
Xgwm192
Xgwm149
Xbcd402
Wangshuibai_II_b
Wangshuibai_II_a
Arina_II
W14_IV
Nyubai_III
W14_III
Wangshuibai II c
Wangshuibai II b
Wangshuibai II a
Ning894037 II
W14 II
Ning7480 II
DH181 II
CM82036 II
Sumai3 II
0.0
14.4
32.4
32.6
53.2
153.2
Xfbb277
Xbarc32
Xgwm371
Xgwm335
gwm639
abc310.1
Wangshuibai_II
DH181_II
Ernie_IV
Ernie_I
0.0
31.6
32.2
32.5
54.7
93.0
Xpsr167
Xgwm133
fhb2
Xgwm644
Xbarc024
Xmwg2053
W14_I
Ernie_II
0.0
5.0
11.1
15.6
16.1
17.1
21.6
39.5
65.3
139.1
Xiba311
Xgwm389
Xgwm533.1
Xbarc133
Xbarc147
Fhb1
Xgwm493
Xgwm533.2
Xgwm285
Xgwm340
Wangshuibai_II
Alondra_II
0.0
18.7
27.8
89.6
W21
Xgwm210
Xgwm484
Xiba209

PYRAMIDING FHB RESISTANCE QTL USING MARKER-ASSISTED SELECTION IN WHEAT.

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ABSTRACT

This study was conducted to pyramid FHB resistance genes from the Chinese source Futai8944 and adapted sources Ernie and Tribute into adapted backgrounds using marker assisted selection. A three-way cross VA02W-713/Tribute//VA07W-120 was made. VA07W-120 is a backcross-derived line with the donor parent Futai8944 and the recurrent parent Ernie. This line contains QTL from both Futai8944 and Ernie and has exhibited a high level FHB resistance in both greenhouse and field tests. Marker assisted selection was applied in F_1 and F_2 generations in spring 2007 and will be applied in $F_{2:3}$ generation during winter 2007. Nineteen F_1 plants derived from the 3-way cross were selected for advancement on the basis of the presence of target alleles for 12 markers on chromosomes 2DS, 3A, 3BS, 5AS, and 6B. About 900 F_2 plants were characterized with 19 markers on 2BS, 2DS, 3A, 3BS, 5AS, and 6B. More than 200 F_2 plants having different combinations of target marker alleles for the five QTL regions were selected for advancement. All of these $F_{2:3}$ lines were planted as head rows this fall in a field scab nursery. Among the 210 $F_{2:3}$ lines, 31 having target marker alleles for two or more QTL fixed in a homozygous state also will be further evaluated and selected in greenhouse tests for plant phenotype, marker haplotypes, and Type II FHB resistance. Among 19 marker loci, these 31 progeny lines have only one to three loci in the heterozygous state and, thus, the primary goal is to identify desirable progeny having fixed target alleles in all five QTL regions. Based on the number of heterozygous marker loci in the progeny, 10 to 30 plants of each $F_{2:3}$ family will be further screened and selected using target markers. This study will assess the efficiency of marker-assisted selection for pyramiding different FHB resistance QTL.

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WINTER AND SPRING WHEAT PARENTAL DIALLEL ANALYSIS FOR SCAB RESISTANCE.

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ABSTRACT

Fusarium head blight (FHB), caused by *Fusarium graminearum*, is an important disease of wheat in South Dakota. The study was conducted to determine combining ability and gene effects in populations derived from mating among spring, winter and facultative wheat genotypes. Six genotypes consisting of susceptible winter wheat 'Nekota' and '2137', moderately susceptible winter wheat 'Harding', moderately resistant spring wheat 'ND2710' and 'BacUp' and resistant facultative wheat 'Ning7840' were crossed in a partial diallel mating design. $F_{4:5}$ lines were hand transplanted in May 2006 and 2007 and screened under mist-irrigated field conditions. Artificial inoculation consisted of corn spawn spread at jointing and inoculum suspension spray at flowering stages. Disease index percentage (incidence percentage * severity percentage/100) of the crosses was analyzed using Griffing's method 4 and model 1. General and specific combining abilities were highly significant ($P < 0.01$) for both years. The result showed that both additive and non-additive gene effects were involved in the inheritance of FHB resistance. The ratio of combining ability variation components $[2\sigma^2_{GCA}/(2\sigma^2_{GCA} + \sigma^2_{SCA})]$ was 0.85 and 0.81 in 2006 and 2007, respectively. The homogeneity of the data over two years was tested. The calculated F-value for the ratio of error variances (F = larger error MS/smaller error MS) for two years was 1.09 ($P = 0.10$, $Df_{num} = 846$, $Df_{den} = 867$). The test of homogeneity indicated that the two years data could be pooled. The pooled analysis showed that general combining ability was significant ($P < 0.01$) but not the specific combining ability ($P = 0.17$). Both the individual and pooled analysis showed that additive gene effects were more important than non-additive gene effects. Thus, progress in developing resistance in wheat can be made by selection.

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ROLE OF A PLASMA MEMBRANE CA^{2+}-ATPASE IN THE RESISTANCE OF POTATO CELLS TO *FUSARIUM SOLANI*.

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ABSTRACT

In natural conditions, plants are always under attack of numerous pathogenic micro organisms and remain, at that, resistant to most of them. To a large extent, the resistance is due to the main enzyme of plasma membrane – Ca^{2+}-ATPase, which plays an important role in the metabolism of the plant cells both in normal conditions and under pathogenesis. This research was aimed at studying the activity and some physical-chemical characteristics Ca^{2+}- ATPase of the potato varieties of different resistance level both in normal conditions and under infection of the fungus *Fusarium solani*. The research targets were the tubers and cells culture of the potato varieties *Tamasha* and *Santa*, which differ in their resistance to *Fusarium solani*. Membrane preparations were obtained by differential sucrose density-gradient centrifugation. The activity of Ca^{2+}-ATPase was evaluated by the number of Pi, obtained as a result of ATP hydrolysis. The fungus *Fusarium solani* was grown on the modified Chapek medium. The use of differential centrifugation method resulted in the isolation of pure preparation of plasma membrane, rich in ions Ca^{2+}. Maximal enzyme activity was identified at the ion concentration Ca^{2+}- 2,25mM, and pH – 7,0. The change of Ca^{2+}-ATPase activity under potation infection with the conidia of *Fusarium solani* was also studied. The increase of Ca^{2+}-ATPase transport activity is revealed in the first hours of infection with the fungus in the resistant potato varieties, where as no change of activity is observed with non-resistant potato varieties. In 24 hours, the increase of enzyme activity is observed in both resistant and non-resistant varieties. Activation of Ca^{2+}-ATPase during the fungus pathogens leads to the increased inflow of Ca^{2+} ions through plasmalemma, which results in the improvement of protective mechanisms. The studying of ATPase activity kinetics showed that for plasma membrane fractions, isolated from the resistant variety *Tamasha*, the maximum speed of hydrolysis from incubation time, was 1.5 higher than in the non-resistant variety *Santa*. The infected cells of potato, resistant to *Fusarium solani*, show the increase of K_m.

Infection leads to the change of physical-chemical enzyme parameters and, respectively, to the increased Ca^{2+} ion flow through membrane.

PROSPECTS FOR IDENTIFYING FUSARIUM HEAD BLIGHT RESISTANCE QTL BY ASSOCIATION MAPPING USING BREEDING GERMPLASM.

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ABSTRACT

In barley, there have been numerous quantitative trait loci (QTL) identified through bi-parental mapping, but very few have been utilized in marker assisted selection (MAS). A new tool, association mapping, may overcome some of the problems that accompany bi-parental mapping by: identifying QTL which are segregating in relevant germplasm, as well as give appropriate estimates of allelic effects. Use of association mapping may shorten the time to implementation of MAS. Two important considerations arise when using association mapping, the extent of linkage disequilibrium and the amount of phenotypic variation present in the mapping population. The objective of this study was to assess phenotypic variation, for FHB and DON, among representative breeding germplasm from four barley breeding programs in the upper Midwest. To accomplish this goal 768 lines were evaluated in seven environments over 2006 and 2007 with each line being assessed in at least four environments. Artificial inoculation, overhead spray and grain spawn, and mist irrigation were used to encourage disease development. Significant variation was found among lines for both traits, with heritabilities, calculated on an environment basis, of 0.45 to 0.52 (FHB) and 0.64 to 0.70 (DON). Histograms showed a range of phenotypic variation that is comparable to bi-parental mapping and therefore should be useful for mapping. Lines will be genotyped using 3000 SNP markers, which have been developed through the USDA Barley CAP. Mapping will be conducted using a mixed model approach to find significant markers.

BREEDING FOR FHB RESISTANCE IN WINTER WHEAT: WHAT'S AHEAD?

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ABSTRACT

Significant yield losses caused by *Fusarium graminearum* Schwabe (teleomorph *Gibberella zeae* (Schwein.), the pathogen known to cause Fusarium head blight (FHB), have occurred in Missouri for more than 70 years but have become more frequent in the last 15-20 years due to increased corn acreage, reduced tillage practices aimed at soil conservation, and the lack of effective cultural and/or fungicide control. Severe state-wide outbreaks in both 1990 and 1991 resulted in losses that were estimated at more than $250 million. In addition to the direct yield losses, test weights were reduced and associated deoxynivalenol (DON) accumulation in the grain prevented harvested grain from being marketed. In 1993, FHB resistance was included as a major breeding objective in the Missouri wheat breeding program and systemic screening of both Missouri breeding lines and germplasm that had been introduced through collaborations with CIMMYT became a routine part of the breeding effort. In 1998, germplasm screening efforts were augmented through U.S. Wheat and Barley Scab Initiative funding. Despite evaluating more than 10,000 genotypes from targeted regions globally between 1993 and 2005 we discovered that some of the best sources of resistance were in our own program in genetic backgrounds that were adapted in much of the soft red winter wheat region. It was clear that this 'native' resistance would lead to the most rapid release of resistant cultivars. Since 1994, three FHB resistant cultivars have been released from the Missouri breeding program including: 'Ernie' released in 1994; Truman, released in 2003; and Bess, an early maturing full-sib of Truman, released in 2005. All have been widely accepted in Missouri and Bess and Truman, which are more widely adapted than Ernie, are being grown on significant acreage outside of the state. The identification of native sources of resistance within the Missouri program has enabled us to have a productive pipeline of FHB resistant germplasm in adapted backgrounds and has led to our focus on this source of resistance. Truman and its early maturing full-sib Bess, have good to excellent levels of types I and II resistances coupled with low DON and good kernel quality retention. They are unique in that this high level of resistance is in an agronomic background that couples excellent yield and test weight with broad geographic adaptation. Haplotype data using known FHB resistance markers suggests that resistance alleles in Truman and Bess probably differ from those in other widely-used sources of resistance. Coupled with its potentially unique resistance alleles, Truman has excellent combining ability (both general and specific) for FHB resistance, producing progeny populations with a high percentage of agronomically desirable, FHB-resistant offspring. This paper will explore opportunities for further enhancing FHB resistance in winter wheat cultivars and accelerating their release by building on the broad-based native resistance available in the winter wheat region. It will focus on the use Truman and its early maturing, full sib, Bess.

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SCAB EPIDEMIC IN NEBRASKA.

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ABSTRACT

Fusarium head blight (FHB), caused by *Fusarium graminearum*, is an important disease of wheat. Natural epidemics of the disease may result in severe yield losses, reduction in quality, and contamination of the harvested grain by mycotoxins. Deoxynivalenol (DON) is the most important mycotoxin that affects all sectors of the wheat industry and it has serious food safety implications in marketing, exporting, processing, and feeding scabby grain. FHB is an episodic disease in the hard winter wheat region of the Great Plains that is known for its diverse and highly variable climate. In eastern Nebraska, the predominant rotation is corn-soybeans, but wheat acreage is increasing as wheat price increases, and wheat continues to be an important winter annual rotational crop. In the 2007 cropping season, a severe epidemic of FHB occurred in the eastern, southeastern, south central, and southwestern parts of Nebraska starting from Omaha to Ogallala (>435 km; >320, 000 ha of wheat). To gain an understanding of the impact of the disease, a sample of grain from each of sixty elite hard winter wheat experimental lines grown in four different locations (Lincoln, Mead, Clay Center, and North Platte) were tested for DON content. The overall mean DON level at each location ranged from <0.5 ppm to 2.3 ppm, the average across locations being 0.8 ppm. The level of DON was highest at Clay Center (3.9 ppm) followed by Lincoln (2.5 ppm), Mead (2.2 ppm), and North Platte (1.2 ppm). Of the sixty experimental lines NE05568, NE05418, and Overland had consistently low DON levels (a mean of <0.5 ppm) at all four locations and also in our mist-irrigated nursery at Mead. Two of the elite lines (NE04653 and Harry) which had the highest DON levels (a mean of >2 ppm) at all locations also showed elevated levels of DON in the mist-irrigated field nursery. This year's scab epidemic impacted many wheat growers in Nebraska. The ongoing FHB research will help in developing adapted FHB-resistant/tolerant cultivars.

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'FALLER': A NEW HARD RED SPRING WHEAT CULTIVAR WITH HIGH YIELD AND QUALITY ADDED TO COMBAT FUSARIUM HEAD BLIGHT DISEASE.

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OBJECTIVES

To develop a new improved hard red spring wheat (HRSW) cultivar which combines resistance to Fusarium Head Blight (FHB) disease and superior grain yield and bread-making quality.

INTRODUCTION

Scab or FHB has been a serious threat to wheat production throughout the world (Schroeder and Christenson, 1963; Bai and Shaner, 1994; McMullen et al., 1997; Stack, 2003). In North America, FHB is caused mainly by *Fusarium graminearum Schwabe* [telomorph *Gibberella zeae* (Schwein.)] (Bai and Shaner, 1994; McMullen et al., 1997). In the spring wheat region, FHB has been a major disease for HRSW produced in North Dakota and neighboring states since 1993. The most recent economic report (Nganje et al., 2004) estimate combined direct and secondary economic losses caused by FHB for all crops were at $7.7 billion. ND and MN account for about 68% ($5.2 billion) of the total dollar losses. Direct losses from 1993 trough 2001 for wheat only were estimated to $2.492 billion (Nganje et al., 2004). The use of genetically resistant cultivars is believed to be the most efficient and economical method of controlling this FHB in wheat. This has been demonstrated in 2002, 2003, and 2004 when 'Alsen', a moderate FHB resistance cultivar derived from the Chinese source 'Sumai 3', released in 2000 by NDSU (with the support of the scab initiative funds) was planted on more than 2.1, 2.4, and 1.9 million acres representing 30.8, 37.4, and 28.9% of ND wheat acreages, respectively (N.D. Agricultural Statistics Service, USDA. 2002; 2003; 2004). Similar scenario was repeated in 2007 when 'Glenn', the 2005 NDSU release jumped from about 2% to more than 20% of total HRSW grown in ND (N.D. Agricultural Statistics Service, USDA. 2006; 2007). The rapid increase in acreage planted to both Alsen and Glenn indicates the desire of ND wheat growers to produce such HRSW cultivars.

MATERIAL AND METHODS

Faller was developed using a modified bulk breeding procedure. It was selected from the "ND2857/ ND2814" cross made at NDSU in the fall of 1997. ND2857 (ND2709/ND688) is a hard red spring experimental lines that has good resistance to FHB originating from ND2709 line derived from the cross involving 'Sumai3' (PI 481542). Sumai3, a spring wheat from China, is arguably the most used source of resistance to FHB in the world. Both ND 2709 and ND688 are HRSW experimental lines developed by the NDSU breeding program. ND2814 ('KITT' (PI 518818)/'AMIDON' (PI 527682)//'GRANDIN' (PI 531005) /'STOA S' (PI 520297)) is a HRSW line developed by NDSU HRSW breeding program. Kitt is HRSW cultivar released in1975 by the Minnesota Agricultural Experiment Station and the USDA-ARS while Amidon, Grandin and Stoa are HRSW cultivars released by NDAES in 1988, 1989, 1984, respectively.

Faller was selected from a bulk of one purified F5 row-plot selected in 2001 at Christchurch, NZ. Faller was initially put in PYT in the summer of 2001. Subsequently, Faller was tested in the advanced yield trials (AYT) and elite yield trials (YET) at four locations in ND in 2002 and 2003, respectively. Faller was tested as ND 805 at 21 location-years in the North Dakota Variety Trials (NDVT) from 2004 to 2006 and in the HRSW Uniform Regional Nursery (URN) (18 locations) in 2005. The URN is conducted in the

states of North Dakota, Minnesota, South Dakota, Nebraska, Montana, Wyoming, Washington, and Manitoba, Canada. The first seed increase of Faller was grown in Prosper, ND in the summer of 2004.
Faller was tested for its reaction to different races of tan spot, leaf and stem rusts, SNB, STB, and FHB in the greenhouse and in the field during the period of 2001- 2006. The SNB, STB and tan spot are the major components of the leaf spotting disease complex of wheat in North America. A complex of these diseases occurs in nature. Hence managing leaf spots is difficult; however, resistant cultivars are the most effective and economical means of controlling leaf spot.

RESULTS

Faller was tested under experimental line ND 805 and was released because it combines very high yield (Table 1), resistance to FHB and leaf diseases (Table 2), and very good end-use quality (Table 3). The name of Faller was chosen as recognition to late James Faller, a former technician in the HRSW breeding program for almost three decades.

Based on 27 site-years of testing in the NDVT and AYT, grain yield of Faller (4467 kg ha-1) was significantly ($p<0.05$) higher than all previously NDSU released cultivars including Alsen (3763 kg ha-1), Glenn (3743 kg ha-1), 'Parshall' (3607 kg ha-1), 'Steele-ND' (4052 kg ha-1), 'Reeder' (3625 kg ha-1), and 'Howard' (3943 kg ha-1) (Table 1). In 19 site-years of testing in the URN trials conducted in 2006, Faller yielded 4055 kg ha-1 compared to 3631, 4095, and 2952 kg ha-1 for 'Keene', 'Verde', and 'Chris', respectively. Other agronomic traits including kernel weight, heading date, plant height and straw strength of Faller and other HRSW cultivars are reported in Table 1.

Quality parameter including Falling number, Flour extraction, dough and baking parameters for Faller and major grown NDSU HRSW cultivars are reported in Table 2. Mean grain volume weight of Faller (757 kg m-3) over 26 site-years in NDVT was similar to Reeder (753 kg m-3) and 'Dapps' (756 kg m-3), but significantly ($p<0.05$) lower than Glenn (797 kg m-3) and Howard (778 kg m-3) (Table 1). Similarly, grain protein of Faller (150 g kg-1) was comparable to Reeder (155 g kg-1) and Parshall (156 g kg-1), but lower ($p<0.05$) than Alsen (157 g kg-1) and Dapps (165 g kg-1) (Table 1).

The seedling and adult plant screening tests conducted under greenhouse conditions from 2003-2006 showed that Faller posses high level of resistance to pathotype THBL, the predominant race of leaf rust (caused by *Puccinia triticina* Eriks.) in the region (Table 3). Faller was also evaluated for resistance to stem rust (caused by *Puccinia graminis* Per.:Pers. f. sp. tritici Eriks. & E. Henn) and was found to be highly resistant to pathotypes Pgt-QCCJ, -QTHJ, -RTQQ, -TMLK, -TPMK, and –HPHJ (Table 3). Faller was screened in the greenhouse for *Septoria nodorum* [caused by *Stagonospora nodorum* (Berk.) Castellani & E.G. Germano] and tan spot [caused by *Pyrenophora tritici-repentis* (Died.) Drechs]. On a scale of 1 to 5 where 1 is resistant and 5 susceptible, Faller had average scores of 1.7, 2.1, 3.7, 2.7, and 3.1 in reaction to tan spot race, 1, 2, 5, *Septoria tritici*, and *Spetoria nodorum*, respectively (Table 3).

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Table 1. Summary of agronomic data for Faller hard red spring wheat (HRSW) and check cultivars tested in the ND HRSW Variety Trials (2003-2006).

Cultivar	Grain yield	Grain protein	Thousand kernel weight	Grain volume weight	Heading date	Height	Straw strength
	kg ha^{-1}	%	g	kg m^{-3}	days from 06/1	cm	score[†]
Faller	4467c[†]	15.0a	29.8cd	757a	29	85	0.6
Howard	3943b	15.3ab	29.0cd	778b	28 (<0.05)[‡]	85(<0.10)	0.5 (1.00)
Glenn	3743a	15.7b	30.2d	797c	28 (<0.05)	83 (<0.50)	0.5 (1.00)
Steele-ND	4052b	15.3ab	28.4bc	773b	28 (<0.05)	85 (<0.10)	2.0 (<0.01)
Dapps	3646a	16.5c	29.5c	756a	26 (<0.01)	93 (<0.01)	0.5 (1.00)
Alsen	3763ab	15.7b	26.9ab	777b	27 (<0.05)	80 (<0.05)	0.3 (0.50)
Parshall	3607a	15.6b	26.5a	770b	27 (<0.05)	86 (1.00)	0.2 (0.50)
Reeder	3625a	15.5ab	26.7a	753a	29 (1.00)	80(<0.05)	1.0 (0.50)
Observations	27	26	26	26	26	26	7

[†] Lodging score: 1=completely erect to 9=completely flat at harvest.

[‡] P values (in parentheses) represent the significance of the comparison between Faller and the respective check cultivar based on a Student's paired *t*-test procedure (SAS-JMP version 6.0.3, SAS Institute Inc., Cary, NC).

Table 2. Quality parameters for Faller hard red spring wheat (HRSW) and check cultivars tested in the ND HRSW Variety Trials (2003-2006).

Cultivar	Falling number	Flour Extraction	Mixing time	Mixing tolerance	Loaf volume	Water absorption
	sec	g kg^{-1}	min	min		%
Faller	423	71.1	8.1	15.8	1042	64.3
Howard	427	69.7	8.2	12.2	1007	64.5
Glenn	401	67.6	9.3	20.6	1102	64.9
Steele-ND	425	70.1	8.5	13.5	1011	
Alsen	412	68.6	9.0	16.2	1057	64.8
Parshall	415	69.2	8.3	14.9	1081	64.7
Reeder	431	67.8	7.0	12.0	1002	63.8
Observations	21	21	21	21	21	21

Table 3. Diseases reactions of Faller hard red spring wheat (HRSW) and check cultivars tested in the ND HRSW Variety Trials (2003-2006).

Cultivar	FHB[†]	Leaf rust		Stem rust		Tan spot			Septoria tritici	Septoria nodorum
		Greenhouse[‡]	Field	Greenhouse[§]	Field	Race 1	Race 2	Race 5		
	%					1-5[#]	1-5	1-5		
Faller	27	R[¶]	R	R	tR	1.7	2.1	3.7	2.7	3.1
Alsen	22	R	MR/MS	MR/R	5R	-	-	-	2.7	4.4
Traverse	-	R	MR/MS	R	R	-	-	-	2.9	2.6
Knudson	-	-	R	R	R	-	-	-	2.2	1.6
Reeder	55	R	S	MR/R	5R	-	-	-	2.9	2.2
Baart	-	-	S	S	50MS	-	-	-	-	-
Tatcher	-	S	-	-	-	-	-	-	-	-
Glenlea	-					4.3	2.0	1.9	2.4	3.7
Salamouni	-					1.4	1.4	1.3	1.7	1.7
6B662	-					1.7	1.7	4.0	1.9	1.4
6B365	-					1.7	4.1	1.9	2.1	1.7
Observations		9	5	4	9	6	6	6	4	4

[†] FHB (Fusarium Head blight) severity as described by (Stack and Frohberg, 2000).
[‡] Greenhouse reactions for leaf rust races MCDL and THBJ.
[§] Greenhouse reactions for stem rust races Pgt TPMK, TMLK, RTQQ, QFCQ, QTHJ, THTS, and TCMJ.
[¶] R=resistant, MR=Moderate resistant, MS=Moderate susceptible, S=Susceptible, tR= trace/Resistant.
[#] The 1-5 scale developed by Lamari and Bernier (1989) was used to score the genotype

PUTATIVE FHB RESISTANCE COMPONENTS RESISTANCE TO KERNEL INFECTION AND TOLERANCE IN THE SSRWW NURSERY, 2005-2007.

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ABSTRACT

The Southern Soft Red Winter Wheat Nursery has been tested in Hungary since 2003 with four isolates of *F. graminearum* and *F. culmorum* separately in three replicates, so four epidemic situations could be studied at the same time. Three bagged groups of heads served as controls. From 2005 we applied one inoculation date as the flowering differences were not larger than 4-5 days, and so an important source of mistake could be neutralized and the data become more suitable to estimate resistance components. After spraying the heads of 15-20 in a group with *Fusarium* suspension, polyethylene bags over 48 hrs secured the high humidity to initiate infection. As the members of the nursery changed from year to year, a repeatability of the Szeged results could not be tested over years, however, we provide a way to estimate the presence of the resistance to kernel infection and tolerance, which can be applied on the multilocated data set of the nursery. The statistical evaluation is as follows: A linear regression slope will be counted between FHB and FDK data. By using the function, the data points of FDK data for all FHB data points will be developed. From the original FDK data the counted FDK values will be extracted, so for each genotype we receive a difference. When this difference is larger than the LSD 5 % from the ANOVA of the FDK values, we will have two sorts of deviations. When the value will be negative, e. g. the predicted FDK is larger than the original data, we speak about the presence of the kernel infection resistance component, e. g. the kernel infection is lower than would be calculated from the function. On the opposite, for positive number the predicted value is smaller than the original data, we speak about an extra sensitivity. Both are important and this is one reason why FHB alone does not sufficiently describe resistance behavior of the given genotype. In 2007 for example, six genotypes of 45 provided resistance and seven revealed extra susceptibility. The estimation of tolerance has the same pattern. We think that such additional analyses of the data will help to evaluate additional resistance components and understand better behavior of the genotypes under different epidemic conditions.

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EVALUATION OF FHB PROFILES OF ADVANCED WHEAT BREEDING LINES TREATED WITH A FUNGICIDE.

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ABSTRACT

In evaluating FHB resistance of wheat breeding lines, breeders strive to estimate, as accurately as possible, the genetic component of resistance. The possible benefits of management tools, e.g., fungicides, are often ignored by the breeder. The purpose of this study was to evaluate the FHB profile of a set of advanced breeding lines in the presence and absence of the fungicide Prosaro. The study was conducted at Princeton and Lexington, KY. The Princeton location was non-irrigated and inoculated with a single application of scabby corn at Feeke's growth stage 9 and two conidial sprays (1×10^6 spores ml^{-1}) at flowering and one week later. The Lexington location was irrigated and inoculated with scabby corn at Feeke's growth stage 9. Rainfall across Kentucky was inadequate for FHB development, but measurable levels of disease were achieved in both nurseries. Diseases other than FHB were present in Lexington, but at very low levels.

A factorial design with 3 replications was used at each location. At Princeton the experimental unit was a conventional 6 row yield plot, 15 ft. long; at Lexington the experimental unit was a 4 row plot, 4 ft long, planted with a headrow planter. Plots at each location were treated at flowering with a tank mix of Prosaro fungicide (6.5 fl. oz. $acre^{-1}$) with Induce (0.125% w/v). Three replicates were left untreated for comparison. FHB symptoms were evaluated 21 days after flowering using a 5 point visual rating scale that encompasses both severity and incidence. After harvest, percentage *Fusarium* damaged kernels (FDK), deoxynivalenol concentration (DON), yield and test weight were measured.

In Princeton, where rain was a limiting factor, there was no significant difference between fungicide-treated and control plots for rating, FDK, DON, and test weight. There was a significant difference in yield, with the average yield of the control plots 11.5 % less than the average yield of the treated plots. In the irrigated Lexington nursery, FHB rating, FDK and DON were all significantly lower in treated than in controls. Yield and test weight were significantly higher for the treated plots than for the control plots. Particularly interesting was the DON level reduction in Lexington. Fungicide x genotype interaction was apparent. For instance, in the control plots, KY99C-1205-06-1 had the lowest DON with 15.4 ppm, but it was third lowest in the treated plots with 13.2 ppm, a 2.2 ppm reduction. In KY98C-1324-01-3 the reduction was 21.4 ppm. The study suggests that advanced breeding lines should routinely be screened with a fungicide as part of the candidate variety evaluation process.

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THE 2006-07 SOUTHERN UNIFORM WINTER WHEAT SCAB NURSERY.

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ABSTRACT

Most components of Fusarium Head Blight (FHB) resistance are greatly influenced by genotype by environment interaction which limits the heritability of resistance estimated by a single program in any given year. 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 and Bess. In addition, the nursery facilitates the sharing of the best resistant materials throughout the breeding community.

The 2006-07 nursery comprised 42 advanced generation breeding lines and three check cultivars, 'Ernie' and 'Bess' (partially resistant) and 'Coker 9835' (susceptible). Six U.S. public programs (Univ. of Arkansas, Univ. of Georgia, Louisiana State University, Univ. of Maryland, N.C. State Univ. and VA Tech.), and one private company, Agripro-Coker, submitted entries. The nursery was distributed to 11 U.S., one Hungarian, and one Romanian cooperator for field and / or greenhouse evaluations. In addition three USDA-ARS laboratories conducted evaluations for Hessian Fly resistance, milling and baking quality and haplotypes based on established SSR markers.

Several nights of uncharacteristic freezing temperatures during the April 6th to April 9th, 2007 period severely damaged the wheat crop throughout the southern US. As a result, no data were obtained by our Agripro-Coker and University of Georgia cooperators. Partial data were provided by the Univ. of Illinois and N.C. State Univ. Copies of the full report will be available at the 2007 National Fusarium Head Blight Forum and subsequently on line at the USWBSI web site: http://www.scabusa.org/.

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HAPLOTYPE STRUCTURE AND GENETIC DIVERSITY AT FUSARIUM HEAD BLIGHT RESISTANCE QTLS IN SOFT WINTER WHEAT GERMPLASM.

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INTRODUCTION AND OBJECTIVES

Several quantitative trait loci (QTLs) for resistance to Fusarium head blight (FHB) have been mapped in wheat. Among these, three were mapped in the Chinese cultivar Sumai 3 and its derivatives on chromosomes 3BS, 5AS, and 6BS (Anderson et al. 2001; Buerstmayr et al. 2002; McCartney et al. 2004; Yang et al. 2003; Zhou et al. 2002). The 3BS FHB resistance QTL (designated *Fhb1*) has been by far the most deployed by breeding programs worldwide (McCartney et al. 2004). Other FHB resistance QTLs have been mapped in Wuhan 1 on chromosomes 2DL and 4BL, (Somers et al 2003) and in the soft red winter wheat cultivar Ernie on chromosomes 4BL, 5A and 3BSc near the centromere (Liu et al. 2007). Haplotyping strategies make use of previous QTL mapping and molecular marker information. In our study we selected markers reported to be near FHB resistance QTL mapped in Sumai 3, Wuhan 1 and Ernie to haplotype a large set of eastern SW wheat lines submitted by breeders .

The objectives of this research were to (1) determine the genetic relationship among soft winter (SW) wheat lines with native and exotic sources of resistance using SSR markers data, (2) compare the SSR marker haplotypes of SW wheat lines with those of Sumai 3, Wuhan 1, and Ernie at known FHB resistance QTLs, and (3) identify lines with novel sources of FHB resistance.

MATERIALS AND METHODS

Two hundred forty-five SW wheat lines with moderately low to strong FHB resistance from native and/or exotic sources, including susceptible and resistant checks, were grown in screening nurseries at Wooster, OH; Urbana, IL; Lexington, KY; and Blacksburg, VA in 2005 to test for type I and type II resistance to FHB. Some exotic FHB resistant accessions (Sumai 3, Ning 7840, Futai 8944, W14, Wuhan 1, F201R, and VR95B717) were planted only for the purpose of obtaining DNA for the molecular marker analysis because they were in the pedigrees of some of the entries screened for FHB and they served as reference for alleles sizes for markers linked to FHB resistance. Forty-seven SSR markers and one STS marker that map near or at FHB-resistance QTLs on chromosome 2DL and 4BL based on Wuhan 1; 3BS, 5AS, and 6BS based on Sumai 3; and 3BS and 5A based on Ernie were selected.

Statistical analysis of phenotypic data was performed using the SAS software package (SAS Institute Inc., NC, USA), and wheat accessions were classified as resistant (R), moderately resistant (MR), moderate (M), moderately susceptible (MS), and susceptible (S). Scoring of polymorphic DNA fragments generated by SSR markers at each locus was conducted by using GeneMarker v1.5 (SoftGenetics LLC, State College, PA). The freeware package PowerMarker version 3.25 (Liu and Muse 2005) was used to perform the phylogenetic analysis and to obtain genetic

diversity estimates; polymorphism information content (PIC) values; and a total number of allele at each SSR marker locus. Haplotype numbers were calculated (1) on the basis of shared alleles at all loci to detect putative novel QTLs in germplasm lines, and (2) on the basis of the allelic distribution of SSR markers linked to 2DL and 4BL QTLs in Wuhan 1; 3BS, 5AS, and 6BS in Sumai 3; and 3BSc and 5A in Ernie.

RESULTS AND DISCUSSION

The table with haplotypes and the dendrogram of all the wheat lines/cultivars included in this study were too large to be included in these proceedings; this information can be found at http://www.cropsci.ncsu.edu/sggenotyping/index.htm.

Phenotypic Evaluation

Reaction of the SW entries evaluated was skewed toward resistance, with 59 lines classified as resistant, 116 moderately resistant, and 28 intermediate. Only 12 and 18 lines were considered moderately susceptible and susceptible, respectively. Of the resistant lines, 24 have exotic sources of resistance in the pedigree and the remaining resistant lines had only SW germplasm in their pedigrees. The SW wheat cultivars NC-Neuse and Truman were resistant to FHB. Of the seventy-three entries in the experiment having exotic sources of FHB resistance in their pedigrees, index scores ranged from resistant (VA 01W-476, IND = 4.1) to susceptible (VA41W-495, IND = 41.9). The exotic sources of resistance in pedigrees of lines in this study included Sumai 3, Ning 7840, Ning 8026, Ermai 9, Futai 8944, ZM10782, W14, Catbird, F201R, and VR95B717. The most common SW wheat cultivars in pedigrees with moderate resistance were Roane, Freedom, Patton, and Ernie.

Marker Diversity

The forty-eight SSR markers evaluated had PIC values that ranged from 0.175 (*Xgwm113*) to 0.922 (*Xcfd233*) with a mean value of 0.639. Only ten SSR markers had PIC values of less than 0.50. Two alleles were observed for STS marker *Xsts3B-256* that is closely linked to the *Fhb1* gene. The number of alleles observed for SSR markers ranged from two (*Xgwm508*) to 22 (*Xgwm601*) with a mean number of 10.42 alleles per locus. The 3BS QTL region had the lowest mean number of alleles detected by SSR markers (6.3), which resulted in the lowest mean PIC value of 0.493. In constrast, the 3BSc QTL interval had the highest mean number of alleles detected by SSR markers (12.1) and the highest mean PIC value of 0.7. A total of 251 haplotypes were detected by the 48 SSR markers, indicating that although there were several full-sib lines in included in the study, no entries had the exact same alleles at all loci.

Cluster Analysis

Entries were grouped into 16 clusters that were generally based on breeding program or geographic origin of lines. The Chinese wheat cultivars having the Sumai 3 haplotype at *Xbarc75*, *Xgwm533*, *Xgwm133*, *Xsts3B-256*, *Xbarc147*, and *Xgwm493*; and therefore the *Fhb1* resistance gene, were grouped separately from all other entries. One exception was inclusion of entry VA01W-476 (W14 x Roane) in this cluster. VA01W-476 had the highest level of resistance of all entries evaluated in the study.

Soft white winter wheat accessions were grouped together and with the resistant Chinese cultivar Wuhan 1, and the susceptible cultivars Madison and Pioneer 2545. In general, entries from breeding programs in the Corn Belt (IN, OH, MO and IL) were in different clusters than entries from the Southeast. The French line VR95B717 that was used as a resistance source by the Virginia Tech. breeding program was included in a cluster of resistant to moderately resistant Corn Belt entries.

Comparison of haplotypes of SW wheat with Sumai 3, Wuhan 1 and Ernie

Markers linked to the *Fhb1* locus had the lowest genetic diversity and linkage disequilibrium was observed between markers across the interval. Markers *Xgwm533* and *Xsts3B-256* proved to be the best available for identifying wheat lines with the *Fhb1* gene derived from Chinese sources. The *Fhb1* resistance gene was not present in all entries derived from crosses

with donor Chinese lines. However, all eight entries in this study that have the *Fhb1* resistance gene based on haplotype data were resistant in the field evaluation.

At the 6BS interval, no entries matched the Sumai 3 haplotype. The most common haplotype on this chromosome region was *Xgwm518* (224 bp), *Xgwm508* (Null), *Xwmc398* (168 bp), *Xgwm133* (131 bp), *Xwmc397* (178 bp), *Xwmc152* (Null), and *Xgwm219* (205 bp), which was shared by a group of moderately resistant to susceptible lines of diverse origin. The two moderately resistant SRW wheat cultivars Patton and Goldfield, while different from Sumai 3, had the most closely related haplotypes in this interval, with matching alleles at six of the seven SSR marker loci assayed.

Resistance QTLs located on chromosome 5A have been identified in a number of lines, including Sumai 3, Ning 7840 and Ernie. Chinese lines Futai 8944, Shaan 85-2, and Ning 7840 had the Sumai 3 haplotype across the ten loci at the 5AS interval analyzed. No SW wheat line had the complete Sumai 3 haplotype at all ten markers. Sumai 3 alleles at SSR marker loci *Xbarc117*, *Xgwm304*, *Xbarc186* and *Xgwm415* reported near the *Qfhs.ifa-5A* QTL peak were common in the SW entries evaluated, with frequencies of 0.42, 0.39, 0.31 and 0.26, respectively. However, when these four marker alleles were considered to form a Sumai 3 haplotype, only controls Ning 7840, W14, Shaan 85-2 and Futai 8944, and four resistant and moderately resistant SW lines derived from crosses with ZM10782 (OH904 and OH902), and with Ning 8026 (AR9700-2-1 and AR9700-2-2) had the donor parent haplotype.

No lines had the complete Ernie haplotype at all ten marker loci on 5A. Liu et al. (2007) reported the location of the *Qfhs.umc-5A* FHB resistance QTL between markers *Xbarc56* and *Xbarc40*. The alleles observed in Ernie at SSR marker loci *Xbarc56*, *Xbarc165*, *Xbarc40*, and *Xgwm156* were common in the SW germplasm in this study having allele frequencies of 0.53, 0.41, 0.34, and 0.40, respectively. When these alleles were considered as a haplotype, nine resistant SW wheat lines, 11 moderately resistant and 2 susceptible lines were identical to Ernie.

Wuhan 1 was not in the pedigree of any of the lines in this experiment. None of the SW wheat entries shared the complete haplotype of Wuhan 1 for markers in the 2DL or 4BL QTL intervals. The highest degree of similarity in the 2DL region was observed in soft white winter wheat lines from Michigan and New York. The most similar accession to Wuhan 1 was the susceptible soft white winter wheat cultivar Geneva. Wuhan 1 alleles for proximal SSR markers *Xwmc245*, *Xwmc144*, and *Xwmc601* were common among the soft white wheat lines. These haplotypes were not observed among the soft red winter wheat entries.

Resistance QTL were located on chromosome 4B in cultivars Ernie and Wuhan 1. The *Qfhs.umc-4BL* QTL peak in Ernie was located near marker *Xgwm495*. Only three backcross-derived lines had the Ernie haplotype for SSR markers spanning this interval (*Xwmc238, Xgwm165* and *Xgwm495*). Eighteen resistant to moderately resistant and 2 susceptible lines had the Ernie haplotype for markers *Xgwm165* and *Xgwm495*. Released cultivars Pat, Superior, Freedom, and Truman, as well as the French line VR95B717, shared this haplotype.

Wheat cultivars Freedom and Patton, and the SW wheat line VA04W-608 had the Ernie haplotype at all eight loci in the 3BSc QTL interval, and they showed moderately resistance reactions to FHB. Thirty-six moderately resistant to resistant lines had a partial Ernie haplotype for SSR markers *Xwmc625*, *Xgwm285*, *Xwmc307*, and *Xwmc418*, including the French line VR95B717, the soft red winter wheat cultivars Freedom, Roane, Patton, and their backcross-derived lines.

CONCLUSIONS

The *Xsts3B-256* and *Xgwm533* markers can be clearly used to identify lines with the *Fhb1* resistance gene. However, there is a need for fine mapping other regions in which FHB resistance QTLs have been located. This seems to be particularly important for resistance from Ernie. Allele sizes of Ernie at 5A and

4BL QTL intervals are common among Eastern soft wheat germplasm. The haplotypes at these loci, along with the 3BSc region, suggest that SW wheat cultivars such as Patton, Freedom, and Roane, that have been considered important sources of native FHB resistance, may share resistance QTL with Ernie. However, fine mapping and marker enrichment of these intervals could provide closer and better molecular markers to be used in germplasm characterization as well as marker-assisted selection programs.

There were 59 wheat entries with high levels of FHB resistance in this study. Many of the most resistant lines had the *Fhb1* QTL combined with native resistance. However, more than half of the resistant lines had no exotic parentage, including the cultivars NC-Neuse and Truman. Neither of these cultivars had the complete haplotype of the donor sources for any of the interval examined in this research. A number of other SW wheat breeding lines did not share any haplotype at known QTLs evaluated in this study. These lines likely carry novel sources of FHB resistance. In contrast, similarity of marker alleles in the 3BSc and 4BL regions suggests that resistance in the line VR95B717 may be due to the QTL identified in Ernie. These data are useful in prioritizing germplasm for QTL mapping and identifying diverse sources of resistance that can be combined to further increase the level of FHB resistance.

DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

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SOLVING THE FHB PROBLEM: GROWERS, EXPORT MARKET AND WHEAT COMMODITY PERSPECTIVES.

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ABSTRACT

The Fusarium Head Blight (FHB) disease of cereal crops has become a very important disease in recent years. The favorable environmental conditions including wet weather during flowering and grain filling and major changes in cropping system (introduction of Maize and minimum tillage) has favored disease development. All classes of wheat produced in the United States, as well as barley, have been impacted by this disease, some regions and classes more than others with the most pronounced impacts in regions that tend to have higher precipitation during the growing season or a greater concentration of Maize in the crop rotation such the US Northern Plains.

In the Northern Plains region, FHB had a devastating impact on the wheat and barley production in 1993, and has continued to cause minor to major annual impacts on cereal crops since. The impact of FHB has not only caused significant economic loss to producers in the region but has also impacted domestic and export customers by threatening the quality reputation; particularly the accumulation of mycotoxins such as DON; and reliability of the Northern Plains in its ability to supply their demands. The Northern Plains has a strong history of being a quality source for hard red spring wheat (HRSW) and durum wheat, and malting and feed barley. Customers in the United States and in numerous export markets have come to rely on the regions wheat and barley production for making specialty breads, premium pasta and couscous and well known brands of beer.

Solving the FHB problem is a top priority for wheat, durum and barley research programs in the region, including developing varieties with higher levels of tolerance, research on optimal crop rotation and management practices that incorporate fungicide applications, and studying ways to help millers and processors handle the inherent quality problems that FHB has on the raw wheat and barley and their products. While notable gains have been made in wheat, durum and barley varieties during the past 15 years, further advancements in varietal tolerance and other tangent production research is needed. The significant financial contributions from the US Wheat and Barley Scab Initiative (USWBSI) have been complemented with State dollars and direct contributions from producers themselves through wheat and barley check-off programs, to help producers in the region and the customers they serve reduce the impacts from FHB.

The successful incorporation of resistance genes to FHB from the Chinese source 'Sumai 3' wheat and other sources such as wheat wild relative *Triticum dicoccoides* into hard red spring and other market classes wheat varieties like Alsen, Glenn, Steele-ND, Freyr, Truman, Neuse, Tribute, McCormick, and others has given producers some attractive options to ward off FHB pressures. An added bonus of many of these varieties is that they also maintain the end-use quality standards demanded by our domestic and international customers. For example, Glenn, released by NDSU and the mostly grown cultivar in the spring wheat region (1.3 millions in 2007) is currently the HRSW quality standard accepted by the US wh eat quality industry and for the regional breeding programs. Worldwide, Glenn has received numerous top quality ratings from key customers participating in the U.S. Wheat Associates Overseas Variety Analysis (OVA) program. In addition,

major improvements in forecast models, available fungicides and application methods have allowed producers to more effectively manage the disease in the field.

Development of tolerant varieties has been more of a challenge with barley and durum due to smaller germplasm pools. However, successful incorporation of tolerance has accelerated in recent years due to new breeding techniques. Advanced lines of both are showing more promise, and it now looks possible to have varieties available for commercial production within the next two to three years that could have the same level of tolerance as is found in some classes of red wheat.

Producers and end-users have already received significant dividends from the enhancement to breeding programs through the USWBSI and other complementary funding. Additional improvements are still needed however, and solving the FHB threat remains a top priority and will likely be for the foreseeable future for small grain producers and researchers in the Northern Plains and the entire US. Domestic and international customers need the additional research to ensure they are able to draw from quality cereal crop production to continue making premium priced, safe and wholesome food products. The producers in the United States and around the world need the additional research to help them maintain viable cereal crop options in their farming operations.

ASSOCIATION MAPPING OF COMPLEX TRAITS IN A DIVERSE DURUM WHEAT POPULATION.

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OBJECTIVE

To assess the potential of association mapping to complement existing mapping efforts in durum wheat.

INTRODUCTION

In plants, the traditional approach to quantitative trait loci (QTL) mapping is to develop a set of random lines derived from crossing two inbred lines that vary in phenotypic values for a particular trait of interest. Although this method has proven effective, there are a number of inherent limitations. First, QTL resolution is generally poor due to the low number of recombination events sampled (Nordborg et al. 2002). This limits potential for marker assisted selection and prevents identification of candidate genes coincident with the QTL. Second, allele variation is restricted to the alleles present in the two parents and QTL localization is restricted to loci segregating between the two parents. This prevents identification of novel alleles and QTL outside of the mapping population (Buckler and Thornsberry, 2002).

Association mapping (AM) mapping is employed in medical studies, and is expected to be a complementary strategy for describing associations between genotype and phenotype in crop plants. The most notable attributes of AM mapping are an improved level of molecular polymorphism as multiple alleles are detected at each locus. Second, accessions or breeding lines have been derived over many generations of meiotic events and this increases the number of crossover events in a defined chromosome interval which is expected to improve the resolution of trait/genotype associations. In plants, two strategies have been used for AM mapping: a) genome scans where the entire genome can be analyzed with molecular markers of sufficient density to localize the QTL (Kraakman et al. 2004; Rafalski 2002) or b) where a candidate gene has been identified, an association of polymorphic markers within the gene and a trait are examined (Thornsberry et al. 2001).

An attractive feature of AM is the ability to perform marker trait associations in well phenotyped breeding populations and locally adapted varieties. However, population structure, due to selection and high levels of co-ancestry, is expected in pedigree-based breeding programs, resulting in a high probability of identifying of spurious associations (Pritchard et al. 2000). However, computational methods have been developed to account for population structure and relatedness to reduce identification of spurious associations (Yu et al. 2005).

Studies on association mapping have been conducted in a number of crop species including barley (Kraakman et al. 2004) and wheat (Tommasini et al. 2007). However, most studies in wheat have focused on specific chromosomes where major QTL have previously been reported and further research to assess the potential of genome-wide association mapping for complex traits in wheat is needed. In this study, we performed genome-wide AM and report on a) the agreement of identified associations with previously published QTL b) and identification of novel QTL yet to be reported in the literature.

MATERIALS AND METHODS

AM Population and Molecular Analysis – Ninety-six (96) diverse durum wheat cultivars and breeding lines collected from breeding programs in Canada (25), Argentina (5), Australia (9), France (3), Italy (18), Germany (2), Mexico (3), Morrocco (3), United States (12), New Zealand (1), Russia (1), Iran (4), Spain (9), as well as one line of unknown origin formed the AM population. Genotyping was performed on an ABI 3100 capillary electrophoresis using M13-labeled microsatellites. A total of 241 microsatellite (SSR) markers were used to amplify 245 loci.

Trait Analysis and AM Mapping – Two traits were selected for genome wide AM study. Grain yellow pigment content (YP; mg kg^{-1}) was assessed on a plot basis using AACC approved method 14-50 on samples collected from replicated trials grown at Saskatoon and Swift Current, Saskatchewan, Canada in 2005 and 2006. Resistance to stem rust race TTKS (UG99) was also assessed in 2007 at an endemic nursery in Kenya. Prior to AM analysis, population structure was assessed using a selection of 147 SSRs were selected at 2 cM intervals within the program STRUCUTRE v.2 (Pritchard et al, 2000). Structure parameter settings were: linkage model, allele frequencies correlated, burn-in length 10,000, and 10,000 repetitions. The highest likelihood was observed for K (no. sub-populations)=5, but little difference was observed between K=3 and K=5. Therefore, the Q matrix was estimated as the average of five runs for K=3. Marker-trait associations were determined using a general linear model in TASSEL version 2.0.1 with the Q-matrix as covariates. Pair-wise linkage disequilibrium (LD) of each SSR with allele specific CAPS markers for phytoene synthase genes *Psy1-A1* and *Psy1-B1* was estimated as the squared allele frequency correlation (r^2) within TASSEL. In cases of multiple alleles, a weighted average of r^2 between loci was calculated.

RESULTS AND DISCUSSION

We first examined YP as a validation trait for AM as the genetics of this trait are well understood and QTL for have been well documented in the literature (Pozniak et al. 2007). Variation for YP was large, ranging from less than 5 mg kg^{-1} to greater than 12 mg ha^{-1}, regardless of testing environments. Heritability estimates were high, ranging from 0.95 to 0.99. Using the sub-population Q-matrix as covariates in a general linear model, marker associations for YP were identified on five chromosomes (Fig. 1) and were statistically significant ($P<0.01$) in all four environments. These associations were coincident with previously published QTL (Fig. 1). Additional markers on chromosomes 2B, 3A and 3B were identified, but these were not consistent among testing environments (data not shown).

Phytoene synthase (*Psy*) is the first critical enzyme in the biosynthesis of lutein, the major xanthophyll responsible for yellow pigment. Genes coding for *Psy1* have been mapped in durum, and *Psy1-B1* co-segregates with a QTL for YP on chromosome 7B (Pozniak et al. 2007). In genetic mapping studies, *Psy1-B1* mapped approx. 4 cM from *gwm146* and Pair-wise LD analysis revealed a CAPs marker for *Psy1-B1* was in strong LD with that SSR (Fig. 2). AM confirmed that this gene was also associated with variation in YP (Fig. 1).

Psy1-A1 was in disequilibrium with *cfa2257* on 7A and genetic studies have confirmed that this gene is linked to *cfa2257*. Using AM, *Psy1-A1* was associated with YP in the durum population (Fig. 1), and localizes to a region on 7A previously associated with YP (Elouafi et al., 2001). These results suggest that LD analysis can be used to correctly position genes in the durum wheat genome, and to determine their association with phenotypic variation. Interestingly, both genes were in LD with a region on 1A where a putative QTL for YP has been identified.

Given the apparent success of genome wide AM mapping for YP, similar analyses were performed for resistance to stem rust race TTKS. Nearly half of the durum wheat accessions evaluated in the 2007 Kenya nursery were scored as moderately to high resistant. The remaining lines possessed intermediate resistance (n=25), or were scored as being moderately susceptible (n=21) or susceptible (n=10). Marker associations for TTKS using numerical severity ratings were

identified on chromosomes 1B, 4B, and the group 5 and 7 chromosomes (Fig. 3).

Of these, only two regions identified are known to house mapped *Sr* resistance genes. Two regions were identified on chromosome 7A, one distal to the centromere, and a second at *gwm276* and *cfa2257* (Fig. 3). *Sr22* is linked to *gwm276*, and that gene is effective against TTKS (Jin et al., 2007). Linked markers on 4B, including the lipoxygenase gene *Lpx-B1.1*, were significantly associated with variation in disease resistance. Lipoxygenase is known to play a role in disease resistance and enzyme activity has been reported to increase in wheat treated with a rust fungal elicitor (Bohland et al. 1997). Sr gene *Tmp* from winter wheat cultivar 'Triumph 64' is effective against TTKS (Jin et al., 2007) and that gene is believed to reside on 4B. Marker associations were identified for *gwm291* and *wmc727* near the distal end of 5A and linked markers *wmc537* and *Cdu1* on 5B, were also significant. Although the association on 5B is in a region were *Sr* genes have yet to be identified, this region has recently been associated with stem rust resistance in hexaploid wheat using AM (Crossa et al., 2007). Interestingly *Psy1-B1* was associated with TTKS resistance (Fig. 3). Although linkage with leaf rust resistance with YP has been reported on 7A, linkage with stem rust has not been reported on 7B

CONCLUSIONS

Our results experimentally validate the potential of genome-wide AM to detect marker associations for complex traits in durum wheat. In the case of YP, marker associations were in agreement with previously identified QTL, suggesting that AM for that trait was effective in this population. For TTKS resistance, novel regions not associated with previously mapped *Sr* resistance genes were identified, and further validation of these marker-trait associations is a high priority. In particular, the association with *Lpx-B1.1* will need to be confirmed, as high enzyme activity results in undesirable colour loss in pasta products. Given the apparent success of AM in this population, we are currently in the process of performing additional genotyping and more detailed phenotyping for mapping of other important agronomic, end-use quality and disease resistance traits, including Fusarium head blight (FHB) resistance. Only a few FHB resistance QTL have been reported in durum wheat, but given the results of this study, AM could be used for identification of novel chromosome regions and alleles that confer some degree of resistance. Although FHB resistance has yet to be evaluated in balanced field trials in this population, sufficient variation for resistance is known to exist in and efforts to perform AM will be pursued. Inclusion of additional FHB resistant breeding lines known to carry similar resistance (i.e. progeny of known lines with improved tolerance) is being considered to improve statistical power for detecting relevant marker associations.

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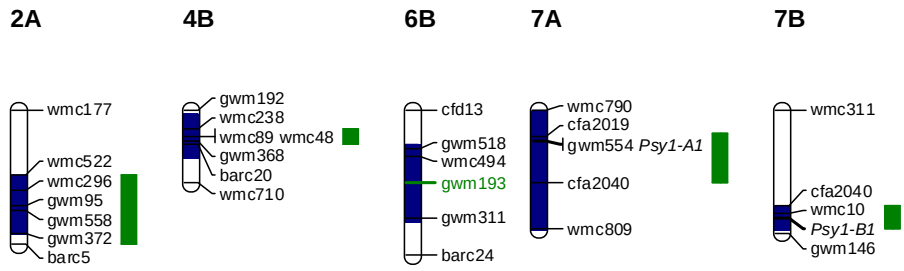

Figure 1. Marker associations for YP in the durum wheat association mapping population. Bars to the right of the linkage group regions where significant associations ($p<0.001$) were identified at all four testing environments. Highlighted regions in the centre of the linkage groups are QTL regions previously identified using bi-parental mapping populations. Only relevant regions of the linkage groups are shown.

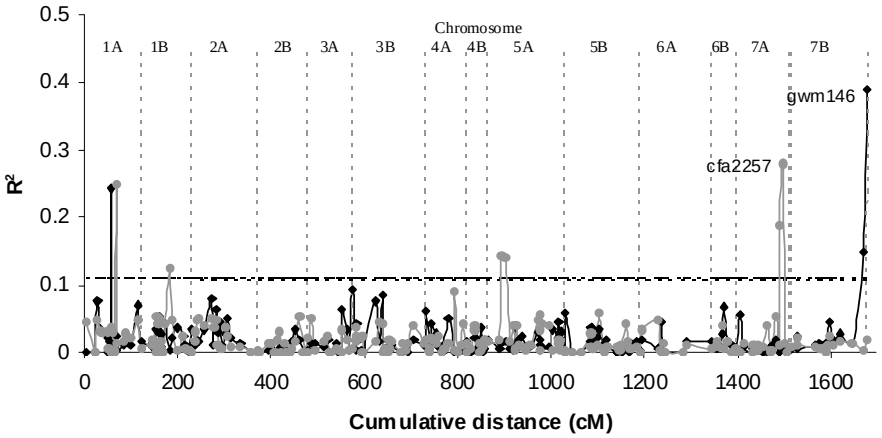

Figure 2. Pairwise LD (r^2) with *Psy1-B1* (—) and *Psy1-A1* (—). The dashed line represents where the cumulative frequency of genome wide r^2 reached 95%.

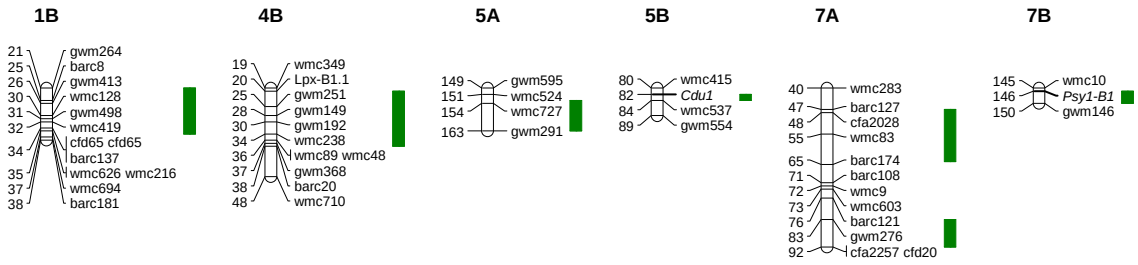

Figure 3. Marker associations for TTKS in the durum wheat association mapping population. Bars to the right of the chromosomes represent regions where significant associations ($p<0.001$) were identified. Only relevant regions are presented.

MOLECULAR CHARACTERIZATION OF A WHEAT-*LEYMUS* COMPENSATING TRANSLOCATION LINE CONFERRING RESISTANCE TO FUSARIUM HEAD BLIGHT.

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ABSTRACT

Fusarium head blight (FHB) resistance was identified in the alien species *Leymus racemosus* (syn *Elymus giganteus*). Several wheat-*Leymus* translocation lines with FHB resistance were developed using radiation treatment or gametocidal gene action. However, all of them are noncompensating, preventing their use in cultivar improvement. We have further screened 58 wheat-*Leymus* introgression lines from 14 siblings for their resistance to spread of infection within spikes. Of 24 lines with high levels of resistance to FHB, we determined that three lines (T01, T09, and T14) were lacking Sumai 3-type alleles at marker loci linked to *Fhb1*, indicating that the FHB resistance in these lines was most likely derived from *L. racemosus*. Previous cytogenetic data revealed that line T01 has a translocation, T4BS·4BL-7Lr#1S, and line T14 has a translocation, T6BL·6BS-5Lr#1L. Line T09 has an unknown wheat-*Leymus* translocation chromosome. A total of 33 RFLP markers selected from seven homoeologous groups of wheat were used to screen T09. Only short arm markers of group-7 detected *Leymus* specific fragments in line T09. The 7AS-specific RFLP fragments were missing in T09, indicating that this line has a compensating Robertsonian translocation involving the long arm of wheat chromosome 7A and the short arm of *Leymus* chromosome 7Lr#1. C-banding and genomic in situ hybridization (GISH) analyses using *Leymus* genomic DNA as probe confirmed the RFLP results. RFLP analysis was further conducted in lines T01 and T14, as well as the wheat-*Leymus* disomic addition lines of DA5Lr#1 and DA7Lr#1, with 11 group-5 markers (five on the short arm and six on the long arm) and eight short arm markers of group-7 chromosomes. The results revealed that both lines T01 and T14 were complex translocations involving the short arms of *Leymus* chromosomes 5Lr#1 and 7Lr#1. These two lines have similar segments from 5Lr#1S. However, the length of 7Lr#1S segment in T01 and T14 is different. Line T01 contains an almost complete short arm of 7Lr#1, whereas about 50% of the distal portion of 7Lr#1S is transferred to the translocation chromosome in the line T14. Three translocation lines and the disomic addition 7Lr#1 were consistently resistant to FHB, whereas the disomic addition 5Lr#1 was susceptible. Because three translocation lines share a common distal segment of 7Lr#1S, a novel scab resistance gene from *Leymus* most likely resides in the distal region of the short arm of chromosome 7Lr#1. Three PCR-based markers, BE586744-STS, BE404728-STS, and BE586111-STS, were developed to accommodate marker-assisted selection in breeding programs. Development of wheat-*Leymus* compensating recombinant lines with smaller alien segments that retain FHB resistance is underway by *ph1b*-induced homoeologous recombination.

FHB RESISTANCE OF WHEAT LINES NEAR-ISOGENIC FOR FIVE DIFFERENT FHB RESISTANCE QTLS.

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ABSTRACT

To continue improving FHB resistance in hard red spring wheat (HRSW), it is imperative that new FHB resistance genes from wheat and its relatives be introduced into the HRSW germplasm base. In 2001 we initiated a program to use marker-assisted backcrossing to individually introgress five confirmed or postulated FHB resistance QTLs from diverse germplasm sources into three FHB-susceptible HRSW backgrounds (Norm, Wheaton, Apogee). The QTLs include two from Sumai 3 (*Fhb1* and *Qfhs.ifa-5AS*) to serve as reference QTLs, one from the soft red winter wheat Freedom reportedly on chromosome arm 2AS, one from the Brazilian wheat Frontana on chromosome arm 3AL, and one from chromosome 3A of wild emmer (*Qfhs.ndsu-3A*). This individual QTL introgression permits evaluation of the effect of each QTL while simultaneously performing prebreeding introgression into HRSW germplasm. The development of the BC_4-derived QTL near-isogenic lines (QTL-NILs) is now complete. For each genetic background/QTL combination, 4 to 5 independent resistant and susceptible NIL pairs were developed, except for the QTL *Qfhs.ndsu-3A*, for which genetic barriers prevented introgression into Norm and Apogee. These lines have been subjected to comparative FHB resistance evaluations both in the field and greenhouse. Results of these evaluations reveal the effect of these QTLs on FHB resistance improvement in the different HRSW genetic backgrounds. Evidence from greenhouse and/or field evaluations has accrued to indicate enhancement of FHB resistance by each of the QTLs introgressed, with both genetic background and the efficacy of the molecular markers contributing to the outcomes. The QTL-NIL series we have developed represent new germplasm for HRSW FHB resistance breeding efforts, and they are a useful resource for additional research on FHB resistance. For instance, we currently are using the lines to examine epistatic interactions between these QTLs to determine which combinations most effectively reduce FHB symptoms. Additional scientific uses include exploring the molecular basis of the wheat-*F. graminearum* interaction as well as the biological basis both of type I and type II resistance.

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FAMILY BASED MAPPING OF FHB RESISTANCE QTLS IN HEXAPLOID WHEAT.

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ABSTRACT

Traditionally quantitative trait loci (QTL) mapping and marker aided selection programs have been two different ventures. For application in breeding programs, novel QTLs must be mapped, though mapping populations are generally not highly useful to breeders because the bi-parental cross is often composed of an unadapted parent. In contrast, breeding programs obtain the most success by creating a large number of breeding populations, or families, using adapted parental germplasm. We combined data from many breeding populations to map QTLs initially, and subsequently used markers of interest for selection. This single step approach is quick, simple, and employs pedigree information and variance component based linkage analysis to place QTLs with substantial effects. Experiments were conducted to validate the approach by mapping *Fhb1*. As part of an ongoing spring wheat breeding program, forty-five susceptible spring wheat genotypes were crossed in different combinations with at least one founder (i.e., a parent containing *Fhb1*). Eighty-three unique families were generated and 793 individuals were screened for resistance in the greenhouse using a point inoculation method. Genotyping was done using mapped simple sequence repeat markers. The QTL was placed in the same position as previous studies with a high probability value. These results demonstrate the usefulness of this approach to quickly map QTLs with relatively large effects, and should allow for marker aided selection as generations are advanced.

FACING THE FHB CHALLENGES TO MALTING BARLEY AND BREWING THROUGH BARLEY BREEDING.

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INTRODUCTION

Fusarium head blight has posed the biggest challenge to the malting and brewing industries, possibly since Prohibition. While disease has lead to reduced yield and quality problems for farmers, DON has caused scarcity of high quality malting barley and relocation of production areas farther from the bricks and mortar (malt houses and breweries) of the industry. In response, BARI Seed Research integrated breeding for resistance into its domestic 6 rowed spring malting barley program with an emphasis on resistance to DON accumulation.

THE CHALLENGES

All malting barley cultivars grown in the 1990s were susceptible to FHB. Breeding programs had little or no resistance in their up-and-coming lines. The Challenge - *Where to get resistance?*

Even under high disease pressure, it was risky to rely on natural infection in the field. Field trials are expensive and require many location years. Greenhouse inoculations, while giving high levels of disease and DON, overwhelmed the low levels of resistance available. The Challenge - *How to insure good screening?*

FHB is difficult to evaluate and DON is costly to determine. The Challenge - *How to measure DON in thousands of breeding lines?*

Meeting and overcoming these challenges has required a collaborative effort involving the entire global barley scientific community. What is reported here includes results from that global effort.

SOURCES OF RESISTANCE

Sources of resistance have been identified from the National Small Grains Collection (NSGC), Composite lines, ICARDA/CIMMYT breeding program, Swiss landraces, the Vavilov Collection in Russia and others. Screening of the NSGC was done by Skoglund and Menert at BARI (Skoglund and Menert, 2002) from 1998-2001 and Steffenson and Scholz at NDSU (Steffenson and Scholz, 2001) from 1999-2001. Over 8100 6 rowed spring barley accessions were screened by one or both groups. Screening was carried out in the field and in the greenhouse. The BARI group identified 15 accessions for possible breeding and the NDSU group identified 10. Only one accession, CIho 6613 (Seed Stocks 1148-1) was identified by both groups.

BARI began collaboration with the ICARDA/CIMMYT barley breeding program in 1999. Since then there has been 1) exchange of germplasm, 2) crossing of elite malting germplasm to resistant sources and 3) screening of various germplasm in nurseries run by BARI and collaborators in North Dakota and Minnesota. Some of the best parents from ICARDA/CIMMYT are listed in Table 1.

Dr. Brian Steffenson (University of Minnesota) has been instrumental in identifying and distributing sources of resistance from other, less accessible sources. These include Composite Cross XXX, Swiss landraces, the Vavilov Collection in Russia and Nordic Gene Bank (Steffenson, 2003; Steffenson and Dahl, 2003: Steffenson, Dahl and Luskutov, 2005). Table 2 has a list of the accessions used by BARI.

FHB NURSERIES AND COLLABORATIVE SCREENING

A number of misted, inoculated nurseries have been established around the world (Table 3). Data collected usually include FHB severity and, in some cases, DON concentration. BARI has collaborated with these institutions to have breeding lines tested. These nurseries have proven invaluable as insurance against low DON years in our yield trials.

There are a number of collaborative screening trials in which BARI participates, some intended for FHB only and others that are primarily for yield and agronomic characteristics. The North American Barley Scab Evaluation Nursery (NABSEN) is an international screening nursery that includes six breeding programs and is planted in 8-10 locations, both dryland and irrigated. Data collected includes FHB incidence and severity, DON and any other (heading date, etc.). The Mississippi Valley Barley Nursery and the Midwest Coop are examples of trials that are grown by multiple collaborators in multiple locations. Where feasible, these are evaluated for resistance to FHB and DON.

DON TESTING

For the past 8-10 years, we have placed high priority on screening for DON accumulation in the BARI breeding program. This has been facilitated by the establishment and/or expansion of a number of facilities (Table 4). We primarily rely on the NDSU DON Testing Lab. Also, BARI Seed Research has collaborated with Dr. Nick Hill, Agrinostics Inc, in testing an ELISA-based technique for quantifying *Fusarium graminearum* mycelium in grain.

WHERE ARE WE NOW?

Progress has been painfully slow due to lack of major genes with large effects in a background close to that needed for brewing. Legacy (accepted in 2001) has reduced DON accumulation to about 30% of that typically found in Robust. Lines in the program now reduce DON by 30-50% of Robust. This progress is the result of incremental integration of genes available within the BARI germplasm as well as UM and NDSU germplasm. Overall, this approach has been a matter of integrating multiple genes with minor effects into an elite line that has acceptable malt quality – not an easy task.

Many additional sources of resistance to FHB/DON are in use at BARI. Projects are currently underway at various universities to determine their genetic diversity. Meanwhile, the most advanced of these were planted as F6 headrows in summer 2007. Selections from these were planted in the nursery in China in Fall 2007. Some of these should advance to first year yield trial status in 2008. These include crosses to the following resistant parents:

- COMP 351 & 355
- HV 746, 779 and 531
- B2027/5/Ataco/Bermejo//Higo/3/CLN/ Gloria/Copal/4/Chevron
- Legacy/Chamico
- Merit//Canela/Zhedar#2
- Merit/MSEL
- Merit/4/Gob/Humai10//Canela/3/Aleli
- Arupo/K8755//Mora/3/Gob/ Humai10/4/ Shyri

Seed Stocks 1148-1 (CIho 6613) was crossed with Legacy in 2000. Though several selections did well in field trials, they failed to advance through our selection process. HV 527, VIR 16537, 28797 and 28807 and NGB9443 are in the current crossing block.

THE FINAL CHALLENGE

Most elite lines never make it through AMBA testing and Anheuser-Busch acceptance. The Final Challenge - *How to get resistant cultivars into the beer?*

BARI continues to submit our best resistant lines to AMBA for testing, as do NDSU and UM. At this time, two lines have made it through AMBA and will be tested in brewing trials with the 2008 crop. These are the UM line M122 and the NDSU line ND20448.

CONCLUSION

Malting barley breeding programs are making progress and meeting the challenges faced by industry. There have been incremental improvements in DON accumulation in advanced lines using a variety of genetic resources. These lines are slowly progressing through the testing and acceptance procedures on their way to farmers' fields and the brew house.

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Table 1. Two rowed and six rowed spring barley lines developed by ICARDA/CIMMYT with resistance to Fusarium head blight and other diseases.

LINE	TYPE
SVANHALS-BAR/MSEL//AZAF/GOB24DH	2 rowed
GOB/HUMAI10/3/ATAH92/ALELI	2 rowed
TOCTE//GOB/HUMAI10/3/ATAH92/ALELI	2 rowed
CANELA/ZHEDAR#2	2 rowed
ATACO/BERMEJO//HIGO/3/CLN/GLORIA/COPAL/4/CHEVRON	2 rowed
CHAMICO	6 rowed
MADRE SELVA	2 rowed
PENCO/CHEVRON	6 rowed

Table 2. New sources of resistance to Fusarium head blight utilized in the Busch Agricultural Resources, Inc. barley breeding program.

ACCESSIONS	TYPE	YEAR	SOURCE	STATUS
COMP 351	6 rowed	2003	Composite Cross	Active
COMP 355	6 rowed	2003	Composite Cross	Active
HV 746	6 rowed	2003	Swiss Landrace	Active
HV 779	6 rowed	2003	s Swiss Landrace	Active
HV 527	2 rowed	2003	Swiss Landrace	Active
HV 531	2 rowed	2003	Swiss Landrace	Dropped
VIR 20738	6 rowed	2004	Vavilov Collection	Dropped
VIR 20733	2 rowed	2004	Vavilov Collection	Dropped
VIR 16537	2 rowed	2007	Vavilov Collection	Active
VIR 28797	6 rowed	2007	Vavilov Collection	Active
VIR 28807	2 rowed	2007	Vavilov Collection	Active
NGB 9443	6 rowed	2007	Nordic Gene Bank	Active

Table 3. Misted and inoculated Fusarium head blight nurseries available to barley breeding programs.

INSTITUTION	LOCATIONS	# BARLEY ENTRIES
NDSU	Osnabrock, ND	21000 hills, 1200 rows, 90 plots
UM	St. Paul, Morris and Crookston, MN	12000 rows
BARI	Fort Collins, CO	350 rows
AAFC	Brandon, MB	18000 rows
ZHEJIANG UNIVERSITY	Hangzhou, China	10000 half rows
CIMMYT	El Batan, Mexico	3000-3500 rows

Table 4. DON testing facilities available to barley breeders.

INSTITUTION	# BARLEY SAMPLES	USWBSI FUNDING	COMMENT
NDSU	18000 in 2006 12000 in 2007 (est)	Yes	Up from 4000 samples in 2000
UM	5000 in 2006/2007	Yes	
AAFC	7000-9000 samples/yr	No	Private lab - 3000 samples
CIMMYT	unknown	No	Est. by Dr. Lucy Gilchrest

DEVELOPMENT OF BARLEY VARIETY CANDIDATE M122WITH ENHANCED RESISTANCE TO FHB.

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ABSTRACT

M122 is a barley variety candidate with enhanced FHB resistance, good malting quality and yield performance similar to currently grown varieties in the Midwest. This line was developed from the sources of resistance Chevron and Zhedar 1 over three breeding cycles. Based on 27 field experiments conducted between 2002 and 2007, M122 has FHB severity and DON levels that are 47% and 52% of the common variety check Robust, respectively. M122 was developed using field-based screening for FHB resistance and low DON in harvested grain. Simultaneous selection was imposed for agronomic performance and malting quality. M122 does not appear to contain resistance source alleles at known and validated QTL for FHB or DON.

Development

M122 is an experimental breeding line, with enhanced FHB resistance, from the University of Minnesota that has potential as a new six-rowed malting variety for the Upper Midwest barley production area. M122 is an F5 derived selection from the cross FEG18-20 / M110 (Figure 1). FEG18-20 was a line selected for its FHB (Fusarium Head Blight) tolerance over several years of disease screening from the cross MNBrite / SI4-29. SI4-29 was also a line selected for its FHB tolerance from the cross Zhedar 1 / Stander // Foster, which was given to us by Rich Horsley as F2 seed in 1994. M122 was advanced by single seed descent from the F1 thru F4 generations. It was selected from an FHB tolerant population (FEG65) in 2002 as an F4.5 line, then seed from a single F5 plant was increased in New Zealand for preliminary yield testing and malt quality evaluations in 2003. M122 was included in the Mississippi Valley Nursery in 2005 (limited locations), 2006 and 2007. M122 was entered into AMBA Pilot Malt evaluations with the 2005 and 2006 crops. M122 is scheduled to be evaluated in plant-scale brewing evaluations with the 2008 crop. From 2002 – 2007, M122 was evaluated for FHB resistance and DON level in numerous FHB nurseries.

FHB Screening and Performance

Screening for resistance that lead to the development of M122 was conducted entirely in inoculated and mist-irrigated field nurseries in Minnesota or in an off-season nursery in China. Disease evaluation in the barley improvement project is a large collaborative effort that involves personnel from the barley breeding project, Ruth Dill-Macky and Char Hollingsworth's pathology programs, and staff at the UM Research and Outreach Centers at Morris and Crookston. Off-season screening in Hangzhou China was done in collaboration with Dr. Bingxin Zhang at Zhejiang University in Hangzhou. Disease screening of M122 began 2002 in replicated F4:5 plots. M122 has been evaluated for resistance to FHB and DON in 25 field experiments conducted between 2002 and 2007. M122 has FHB severity and DON levels that are 47% and 52% of the common variety check Robust, respectively.

Agronomic Performance

In Minnesota State Variety Trials (2004-2007) based on the 12 location mean, M122 was the top yielding variety (Table 1). In on-farm trials conducted in Minnesota, M122 was among the higher yielding lines although had slightly more lodging (Table 2). In North Dakota trials in 2007, M122 yielded better than Ro-

bust but not as high as Lacey, Tradition and Legacy (Table 3).

Graphical Genotype

We assessed M122 and all of the parents in the pedigree (Figure 1) with DArT markers (Wenzl et al., 2006). A total of 884 markers provided a good signal in the assay and 165 of those were polymorphic between the parents of M122. We examined chromosome 2H and 6H since these chromosomes have been implicated in FHB resistance in mapping studies with Chevron, Zheddar 2 and other sources of resistance(de la Peña et al., 1999; Dahleen et al., 2003; Canci et al., 2004). In the two QTL regions that have been mapped and validated in Chevron, M122 appears to carry the susceptible parent allele. We were also able to confirm that M122 does not carry the Zhedar 1 allele in these regions. This indicates that we should be able to improve resistance in M122 via MAS for the Chevron or Zhedar 1 alleles at these QTL.

ACKNOWLEDGEMENT

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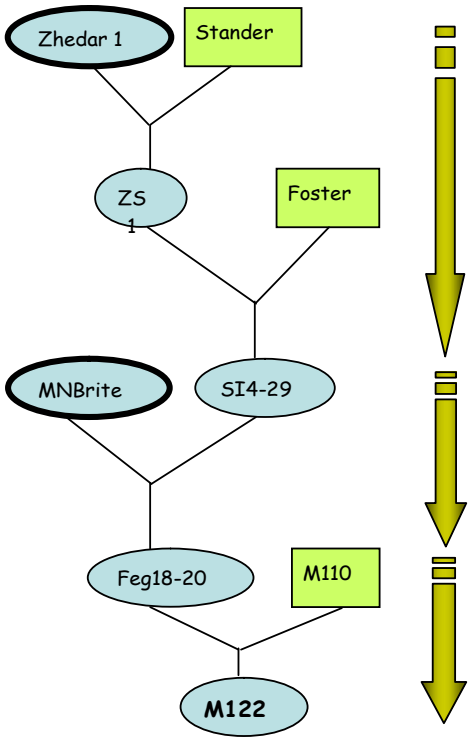

Figure 1. M122 was developed over three breeding cycles and has two resistant sources in its pedigree (Zhedar 1 and MNBrite) MNBrite derives its resistance from Chevron. Lines or varieties in boxes are susceptible to FHB and those in ovals have enhanced resistance to FHB.

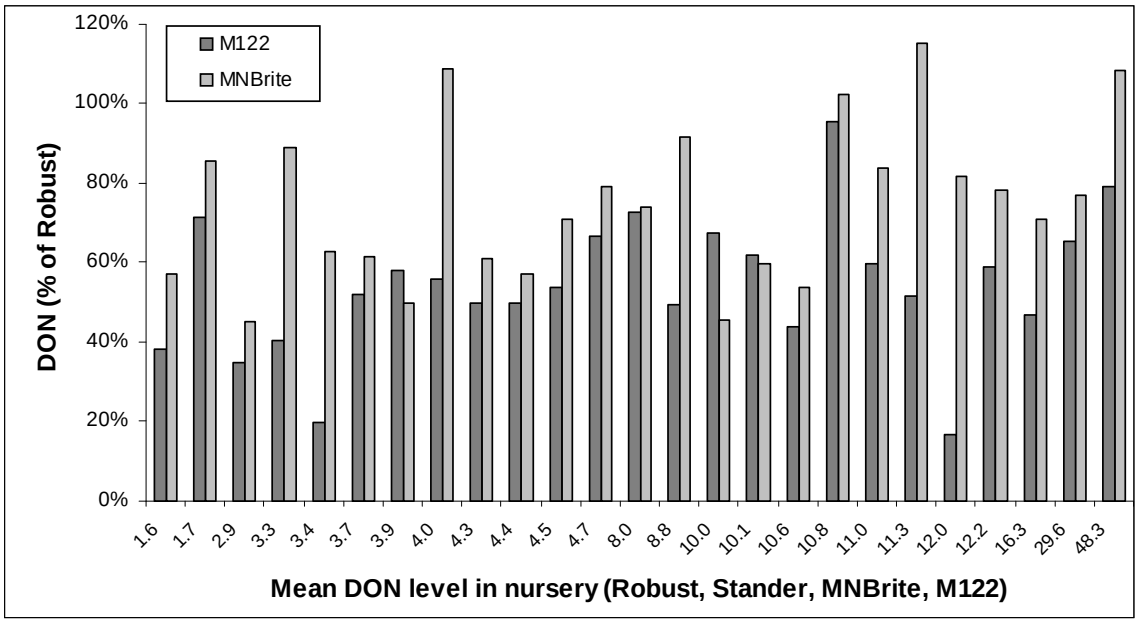

Figure 2. DON levels as a percent of Robust for M122 and MNBrite in 25 trials representing a wide range of disease pressure.

Table 1. Yield comparisons of M122 compared to check varieties 2005-2007 in Minnesota State Variety Trials (mean of 12 trials).

	Crookston		Morris		Stephen		St.Paul		Roseau		
	3 yr.		3 yr.		2 yr.		2 yr.		2 yr.		**12 Sta.Yr.**
Variety	Ave.		Ave.		Ave.		Ave.		Ave.		Ave.
Robust	91		71		84		100		86		85
Stander	94		77		86		98		92		88
MNBrite	92		70		86		93		74		83
Lacey	94		79		99		96		76		88
Drummond	93		72		92		106		84		88
Stellar ND	96		66		94		90		84		85
Legacy	95		79		103		106		85		92
Tradition	96		80		101		95		88		91
Conlon	97		69		96		81		73		83
M122	**103**		**77**		**96**		**113**		**81**		**93**

Table 2. Yield comparisons of M122 compared to check varieties 2006-2007 in Minnesota On-Farm trials (mean of 5 locations).

Variety/Line	Yield (Bu/A)	Test Weight	Protein	Plump	Plant Height	Lodging*
Drummond	107.5	43.9	13.2	72.5	32.6	1.0
Lacey	105.5	45.6	13.6	73.9	31.4	1.0
Legacy	99.1	41.5	13.0	61.8	31.2	1.3
Robust	100.1	44.6	13.8	69.4	32.4	1.8
Stellar	107.2	44.2	12.7	77.1	31.9	1.7
Tradition	101.7	44.5	13.2	74.7	31.7	1.0
M122	**105.5**	**43.4**	**13.3**	**68.7**	**32.6**	**3.2**

Data provided courtesy Jochum Wiersma
*data from 2007 only

Table 3. Yield comparisons of M122 compared to check varieties 2007 in North Dakota State Variety Trials (mean of 5 trials).

Variety / Line	Days to Heading (June)	Plant Height (inches)	Lodging (%)	Stem break (1-5)	Yield (Bu/A)
Robust	25.0	33	33	2.5	75.7
Lacey	24.7	32	30	2.5	84.0
Drummond	24.4	32	25	1.9	78.1
Stellar-ND	24.3	32	26	2.6	76.7
Legacy	26.5	32	34	2.2	82.8
Tradition	25.2	32	31	1.5	80.5
M122	**24.9**	**31**	**31**	**2.3**	**79.0**

Data provided courtesy Richard Horsley

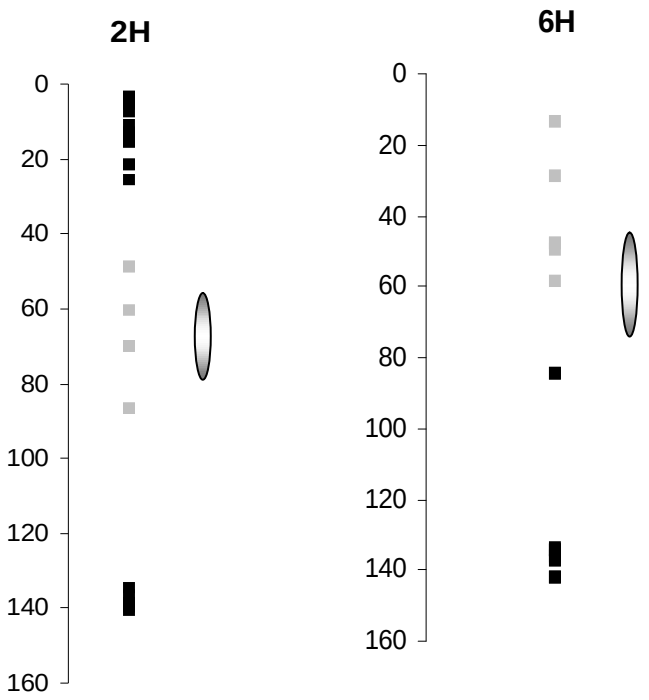

Figure 3. Graphical genotype of DArT markers for M122 on chromosomes 2H and 6H. Numbers are centimorgans. Dark squares indicate that M122 carries the resistant parent (Feg18-20) allele at the marker locus and grey squares indicate M122 carries the susceptible parent allele (M110). Ovals indicate position of QTL for FHB.

REPORT ON THE 2006-07 NORTHERN UNIFORM WINTER WHEAT SCAB NURSERIES (NUWWSN AND PNUWWSN).

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OBJECTIVES

This is a summary of the report on the 2006-2007 Northern Uniform Winter Wheat Scab Nursery (NUWWSN) and the Preliminary Northern Uniform Winter Wheat Scab Nursery (PNUWWSN). A full report will be available on the USWBSI web site prior to the 2007 forum. The objective of these tests is to screen winter wheat genotype adapted to the northern portion of the eastern US for scab resistance.

MATERIAL AND METHODS

The traits assessed and locations that reported data are listed in Table 1. Entries for the NUWWSN came from 13 programs while the PNUWWSN entries came from nine programs (Table 2).

RESULTS

There are eight FHB traits for each trail. Entries with means that were not significantly different than the lowest mean for six or more FHB traits are shown in Tables 3 and 4 (eg entries with at least 6 "l"s). Only three entries had DON < 2 ppm (entries 5, 22, and 26 in the PNUWWSN, see Tables 4 and 5).

Table 1. Traits assessed in the 2006-07 PNUWWSN and NUWWSN tests.

Code	Trait	Description	PNUWWSN Locations*	NUWWSN Locations*
SEV	Disease severity from field tests	% of infected spikelets in an infected head.	IL,KY,MI,MO,ON,VA	IL,KY,MD,MI,MO,NE,NY,OH,ON, VA
INC	Disease incidence	% of heads with at least one infected spikelets	IL,KY,MI,MO,ON,VA	IL,KY,MD,MI,MO,NE,NY,OH,ON, VA
IND	Disease index	IND = (SEVxINC)/100	IL,IN,,KY,MI,MO,OH,ON, VA	IL,IN,KS,KY,MD,MI,MO,NE,NY, OH,ON,RO,VA
KR	Kernel rating	A visual assessment of the percent infected kernels	IL	IL,KS
PSS	Percent scabby seed	Percent of scabby seed by weight	KY,MO	KY,MD,MO,NE,RO
ISK	Composite of head and kernel traits	ISK Index = .3 (Severity) + .3 (Incidence)+.4 (% FDK or PSS)	IL,KY,MO	IL,KY,MD,MO,NE
DON	DON (vomitoxin)	PPM of vomitoxin in grain	IL,KY,VA	IL,KS,KY,MD,NE,NY,VA
GH	Greenhouse severity	Same as SEV except from greenhouse	IL	IL,MO

* ON and RO indicate Ontario Canada, and Romania, respectively

Table 2. Entries in the 2006-07 PNUWWSN and NUWWSN.

NAME	PNUWWSN PEDIGREE
ERNIE	Moderate Resistant Check
TRUMAN	Mod Resistant/Resistant Check
FREEDOM	Moderate Resistant Check
PIONEER 2545	Susceptible Check
P.981129A1--17	92829A1/Patton
P.99751RA1--94	92212/961331/5/92212/4/F201R/3/9547//Patterson/Ernie
P.0128A1-44-1-7	981129A1/981312A1
P.03528A1-10	INW0315/9895C1/3/INW0301/INW0304//981542A1
P.03630A1-18	99751RA1/INW0315//981358C1/97462
SE981089-34	P25R57/SE1694-12
SE91 1492-4	TAISHANG1/GR863//CARDINAL
SE94-1012-25	T814/L880119
M04-4843	KM2186-92/M94*1649//Patton
M04-4788	Pio26R61/Patton
M04*5109	VA94-54-479/Pio2628
M04-4258	Madison/Roane
M04-4393	M94*1586-1/Roane
RCUOGGold.Val	N/A
RCUOGL15	ACRONxSVP/R/FR.#1
RCUOGL4	2737W x EX9806/TF13
RCUOGL17	SVPx ACRON/TF18
RCUOG10/18	ACRON x R/FR. #1
IL03-18438	IL97-3574 / IL95-4162
IL03-15452	IL96-2526 / IL97-3574
IL03-453	Ernie/ IL95-4162
IL01-34159	IL84-2191 / IL87-2834 // IL90-6364 / IL96-24851
IL79-002T-B-B	IL94-6727 / IL96-6472
KY99C-1298-08-1	KY 89C-804-11/KY 89C-225-5//2540
KY99C-1051-03-1	2552/2684//2540
KY99C-1176-02-1	NC96 BGT 6/2552//25R26
MO 050600	Kingraze/Bess 'S'
MO 050699	950016/3/950016//90X54-1-1/MO 91-1009
MO 050917	Truman 'S'/MO 960815
MO 050921	Ernie/Truman 'S'
VA06W-598	P89118RC1-X-9-3-3-1/TRIBUTE//M94-1069
VA06W-557	IL 94-1549/AGS 2000,F8
VA06W-595	P88288C1-6-1-2-8/VAN98W-346//RC STRAT.
VA06W-608	FREEDOM/NC96-13374//RC STRATEG, F7
VA06W-627	IL 94-1549/VA97W-375//COKER 9025, F7
OH03-183-32	15497 /897A
OH03-235-2	OH552 /HOPEWELL
OH03-41-45	IL91-14167 /OH599
OH03-97-6	P88288C1-6-1-2 /OH536
OH03-75-58	HOPEWELL /OH655

NAME	NUWWSN PEDIGREE
ERNIE	Moderate Resistant Check
TRUMAN	Mod Resistant/Resistant Check
FREEDOM	Moderate Resistant Check
PIONEER 2545	Susceptible Check
NY88046-8138	Susquehanna/Harus
NY93285SP-7343	SuMei Comp: 92002
NY93285-7110	SuMei Comp: 92002
NY91028SP-9082	Harus/4/CS/A.Curvif//Glenn/3/Ald/Pvn(M-30)
NY93306-7091	18cc-59/Pio2548

NAME	NUWWSN PEDIGREE (CONTINUED)
KS04HW47-3	X921012-A-7-1/TGO
KS04HW101-3	98HW423/98HW170
P.011035A1-71	981128A1/981477A1//92145E8
P.011036A1-14	981128A1/97462A1//92145E8
P.02444A1-23-6	981129A1/99793RE2//INW0301/92145E8
P.03647A1-1	981477A1/INW0315//981517A1/97462A1
P.04287A1-10	INW0315*2/5/INW0304/4/9346/CS5A//91202/3/INW0301/INW0315
NE01643	NE94482 (=ARA/ABILENE//NE86488)/ND8974
HARRY	NE90614 /NE87612
NI04421	NE96644//PAVON/*3SCOUT66/3/NE94653
NE04653	N97S084//W96-500W/N95L158
NE03490	WI90-540W/NE93554
MD01W233-06-11	MCCORMICK/CHOPTANK
M03-3002	Winter/Winter FHB Bulk
M03-3104	Hopewell / M94-1107
M03-3616	Hopewell / Patton
M03*3877	T8141 / D93-6093
M03*3861	Pio2552 / M94-1407
RCUOG19/21	AC Ron/WEK0609H3xACRon
RCUOGF110202D/4	SD97060 x Ringo Star
RCUOGF111202A/3	Freedom x Harding
RCUOGDHACF1109O2D	SD97060 x Freedom
RCUOGNS984-1	Not available
IL00-8530	IL89-1687 // IL90-6364 / IL93-2489
IL01-11445	IL87-2834-1 / IL95-678
IL01-11934	IL90-6364 / IL94-1909
IL02-19463	Patton / Cardinal // IL96-2550
IL02-23168	IL94-1909 / Pioneer25R26 // IL95-4162
KY97C-0540-01-03	COKER 9803/L910097//2552
KY97C-0554-03-06	VA94-54-549/Roane//Kristy
KY97C-0554-04-05	VA94-54-549/Roane//Kristy
KY97C-0508-01-01A-1	FFR 555W/VA94-52-25//2568
KY97C-0554-03-02	VA94-54-549/Roane//Kristy
MO 040165	Bess RS, earlier
MO 050101	Bess RS, same
MO 050143	Bess RS, shorter
MO 050197	MO 12278/Pioneer 2552
MSU Line E3023	CALEDONIA/NY85020-395
MSU Line E5015	CALEDONIA/PIONEER_25W33
MSU Line E6001	PIONEER_25W60/CJ9306
MSU Line E6002	VA96W-403WS /CJ9403
MSU Line E6003	VA96W-403WS /W14
VA06W-600	P89118RC1-X-9-3-3-1/TRIBUTE//M94-1069,F7
VA06W-602	P89118RC1-X-9-3-3-1/TRIBUTE//M94-1069, F7
VA06W-587	ROANE//OH 552/AGS 2000, F7
VA06W-594	P88288C1-6-1-2-8/VAN98W-346//RC STRATEGY
VA06W-585	Roane / Ernie//McCORMICK,F8
OH02-15978	PATTERSON/HOPEWELL
OH02-12678	FOSTER/HOPEWELL//OH581/OH569
OH02-12686	FOSTER/HOPEWELL//OH581/OH569
OH02-13567	OH581/IN83127E1-24-5-2//5088B-D-32-1/OH601

Table 3. Best entries (top) and worst (bottom) from the 2006-07 NUWWSN. Summary statistics are for all 60 entries.

	NAME	SEV		INC		IND		KR		PSS		ISK		DON		GHSEV		#l	#h
2	TRUMAN	11.2	l	26.7	l	6.1	l	17.5	l	12.6	l	16.6	l	3.9	l	3.4	l	8	0
51	MSU Line E6003	14.0	l	18.3	l	6.1	l	24.4	l	17.4	l	12.6	l	4.8	l	8.4	l	8	0
43	MO 040165	20.2	l	34.7		12.2	l	16.2	l	15.4	l	25.6	l	3.1	l	4.6	l	7	0
45	MO 050143	19.5	l	38.1		12.2	l	20.6	l	7.1	l	26.7	l	3.9	l	6.2	l	7	0
44	MO 050101	21.4	l	40.4		12.7	l	26.2	l	7.3	l	26.3	l	3.9	l	4.5	l	7	0
50	MSU Line E6002	17.5	l	27.1	l	12.9	l	35.6		13.9	l	26.7	l	4.6	l	25.5	l	7	0
59	OH02-12686	20.2	l	35.5		14.6	l	25.0	l	17.1	l	26.0	l	2.1	l	24.0	l	7	0
46	MO 050197	17.7	l	33.7		11.6	l	17.9	l	8.6	l	21.3	l	3.7	l	39.1		6	0
31	RCUOGDHACF 1109O2D	18.3	l	21.4	l	12.1	l	47.1		22.7	h	26.7	l	3.9	l	25.4	l	6	1
49	MSU Line E6001	19.1	l	41.5		14.2	l	33.1		14.4	l	25.0	l	4.6	l	4.9	l	6	0
29	RCUOGF110202D/4	22.9		25.7	l	14.7	l	32.1		11.1	l	23.5	l	1.7	l	10.7	l	6	0
58	OH02-12678	19.4	l	39.5		17.0		15.3	l	11.2	l	25.2	l	4.2	l	13.3	l	6	0
5	NY88046-8138	38.6	h	54.1	h	24.8	h	44.6		28.0	h	48.5	h	7.8		46.6	h	0	6
32	RCUOGNS984-1	38.0	h	61.1	h	28.3	h	45.6		26.6	h	42.6	h	8.2		46.9	h	0	6
24	M03-3104	31.4	h	64.3	h	25.5	h	50.9	h	22.6	h	49.5	h	8.4		59.1	h	0	7
4	PIONEER 2545	39.0	h	62.3	h	30.0	h	49.8	h	18.5	lh	50.3	h	11.6		50.5	h	1	7
10	KS04HW47-3	41.5	h	62.7	h	34.1	h	65.0	h	21.2	h	46.1	h	16.6	h	70.6	h	0	8
	AVERAGE	24.1		40.4		17.0		33.3		16.2		30.5		5.7		26.1		4.8	2.1
	LSD	11.2		14.3		10.0		17.6		14.8		14.1		3.9		28.8			
	R2	0.53		0.73		0.52		0.79		0.48		0.52		0.8		0.71			
	CV	46.7		34.7		66.2		27.6		64.9		32.6		60.9		57.8			
	n	10		10		13		2		5		5				2			

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

Table 4. Best entries (top) and worst (bottom) from the 2006-07 PNUWWSN. Summary statistics are for all 44 entries.

	NAME	SEV		INC		IND		KR		PSS		ISK		DON		GHSEV		#l	#h
2	TRUMAN	16.9	l	38.4	l	7.3	l	13.0	l	9.4	l	17.8	l	2.2	l	12.3	l	8	0
26	IL01-34159	18.9	l	40.1	l	8.0	l	8.0	l	5.5	l	14.9	l	0.3	l	3.2	l	8	0
31	MO 050600	21.7	l	39.1	l	10.0	l	20.0	l	9.8	l	18.9	l	1.8	l	18.0	l	8	0
34	MO 050921	19.9	l	42.7	l	10.4	l	15.0	l	6.4	l	17.2	l	2.7	l	4.0	l	8	0
27	IL79-002T-B-B	19.4	l	40.3	l	11.1	l	11.0	l	4.0	l	11.8	l	3.4	l	21.1	l	8	0
40	OH03-183-32	19.5	l	46.1	l	12.2	l	15.0	l	7.7	l	19.7	l	2.0	l	32.9	l	8	0
23	IL03-18438	23.1	l	54.7	l	13.3	l	18.0	l	5.1	l	20.2	l	4.1	l	12.5	l	8	0
5	P.981129A1--17	23.7	l	47.0	l	13.4	l	23.0	l	19.2	l	22.4	l	0.4	l	3.8	l	8	0
6	P.99751RA1--94	23.3	l	47.9	l	15.2	l	20.0	l	15.6	l	23.8	l	3.9	l	4.1	l	8	0
37	VA06W-595	25.0	l	49.9	l	16.7	l	25.0	l	13.7	l	22.3	l	3.4	l	37.5	l	8	0
8	P.03528A1-10	16.9	l	43.2	l	9.6	l	30.0		5.6	l	22.2	l	3.6	l	28.8	l	7	0
43	OH03-97-6	18.8	l	47.7	l	10.0	l	33.0		18.6	l	24.1	l	4.0	l	37.8	l	7	0
24	IL03-15452	22.0	l	40.0	l	11.0	l	12.0	l	7.6	l	15.1	l	3.7	l	58.2		7	0
9	P.03630A1-18	21.0	l	50.8	l	11.2	l	27.0		10.5	l	24.0	l	2.6	l	3.4	l	7	0
32	MO 050699	22.6	l	50.8	l	12.6	l	25.0	l	10.6	l	22.5	l	4.9		6.0	l	7	0
22	RCUOG10/18	31.0		48.0	l	16.8	l	11.0	l	6.8	l	16.9	l	1.3	l	17.4	l	7	0
1	ERNIE	22.8	l	44.5	l	12.9	l	33.0		12.7	l	23.6	l	4.9		28.6	l	6	0
44	OH03-75-58	23.6	l	50.0	l	13.3	l	60.0		12.5	l	32.5		3.5	l	40.8	l	6	0
25	IL03-453	24.3	l	57.5		15.8	l	25.0	l	3.0	l	23.5	l	3.7	l	59.3		6	0
33	MO 050917	24.3	l	55.7	l	16.0	l	37.0		6.9	l	31.0		4.4	l	27.5	l	6	0
16	M04-4258	29.4	l	56.8		17.9	l	22.0	l	9.1	l	24.9		4.1	l	4.8	l	6	0
13	M04-4843	32.3		42.5	l	20.4		15.0	l	10.8	l	20.0	l	2.6	l	36.1	l	6	0
4	PIONEER 2545	38.1		69.0	h	30.9	h	47.0		28.6	h	39.8	h	6.5	h			0	5
12	SE94-1012-25	49.6	h	60.0		33.5	h	63.0	h	32.0	h	43.5	h	6.7	h	62.1	h	0	7
10	SE981089-34	47.8	h	64.4	h	36.4	h	80.0	h	42.8	h	48.0	h	9.9	h	38.8	l	1	7
18	RCUOGGoldenValue	53.3	h	80.4	h	38.9	h			40.1	h	46.7	h	8.3	h	85.4	h	0	7
	Average	26.5		50.3		16.3		31.7		13.6		24.9		3.8		36.7		4.8	1.2
	LSD	13.9		17.5		11.6		17.8		19.6		12.8		4.3		38.6			
	R2	0.6		0.6		0.6		0.8		0.7		0.8		0.9		0.4			
	CV	42.5		42.5		65.5		33.4		65.3		29.4		55.3		90.1			
	# Locations	6		6		8		1		2		3		3		1			

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

Table 5. Summary of results of the 2006-076 PNUWWSN.

	NAME	SEV	INC	IND	KR	PSS	ISK	DON	GHSEV	#l	#h
1	ERNIE	22.8 l	44.5 l	12.9 l	33.0	12.7 l	23.6 l	4.9	28.6 l	6	0
2	TRUMAN	16.9 l	38.4 l	7.3 l	13.0 l	9.4 l	17.8 l	2.2 l	12.3 l	8	0
3	FREEDOM	28.3 l	47.5 l	15.6 l	37.0	12.5 l	26.2	6.0 h	5.0 l	5	1
4	PIONEER 2545	38.1	69.0 h	30.9 h	47.0	28.6 h	39.8 h	6.5 h		0	5
5	P.981129A1--17	23.7 l	47.0 l	13.4 l	23.0 l	19.2 l	22.4 l	0.4 l	3.8 l	8	0
6	P.99751RA1--94	23.3 l	47.9 l	15.2 l	20.0 l	15.6 l	23.8 l	3.9 l	4.1 l	8	0
7	P.0128A1-44-1-7	27.6 l	56.9	17.1 l	40.0	17.1 l	30.3	3.0 l	4.9 l	5	0
8	P.03528A1-10	16.9 l	43.2 l	9.6 l	30.0	5.6 l	22.2 l	3.6 l	28.8 l	7	0
9	P.03630A1-18	21.0 l	50.8 l	11.2 l	27.0	10.5 l	24.0 l	2.6 l	3.4 l	7	0
10	SE981089-34	47.8 h	64.4 h	36.4 h	80.0 h	42.8 h	48.0 h	9.9 h	38.8 l	1	7
11	SE91 1492-4	31.7	55.0 l	21.5	32.0	13.0 l	28.7	4.9	44.5	2	0
12	SE94-1012-25	49.6 h	60.0	33.5 h	63.0 h	32.0 h	43.5 h	6.7 h	62.1 h	0	7
13	M04-4843	32.3	42.5 l	20.4	15.0 l	10.8 l	20.0 l	2.6 l	36.1 l	6	0
14	M04-4788	38.4	42.1 l	16.4 l	37.0	20.1 l	29.2	2.8 l	100.0 h	4	1
15	M04*5109	25.0 l	53.7 l	16.0 l	50.0	4.6 l	25.6	5.8 h	21.5 l	5	1
16	M04-4258	29.4 l	56.8	17.9 l	22.0 l	9.1 l	24.9	4.1 l	4.8 l	6	0
17	M04-4393	24.7 l	59.7	14.7 l	35.0	14.0 l	29.1	5.6 h	65.4 h	3	2
18	RCUOGGoldenValue	53.3 h	80.4 h	38.9 h		40.1 h	46.7 h	8.3 h	85.4 h	0	7
19	RCUOGL15	29.8 l	52.1 l	17.9 l	43.0	23.1	35.6 h	4.6 l	76.2 h	4	2
20	RCUOGL4	37.8	53.8 l	22.6	47.0	13.9 l	31.1	6.9 h	40.3 l	3	1
21	RCUOGL17	31.6	51.8 l	17.7 l	40.0	11.2 l	25.6	4.5 l	66.7 h	4	1
22	RCUOG10/18	31.0	48.0 l	16.8 l	11.0 l	6.8 l	16.9 l	1.3 l	17.4 l	7	0
23	IL03-18438	23.1 l	54.7 l	13.3 l	18.0 l	5.1 l	20.2 l	4.1 l	12.5 l	8	0
24	IL03-15452	22.0 l	40.0 l	11.0 l	12.0 l	7.6 l	15.1 l	3.7 l	58.2	7	0
25	IL03-453	24.3 l	57.5	15.8 l	25.0 l	3.0 l	23.5 l	3.7 l	59.3	6	0
26	IL01-34159	18.9 l	40.1 l	8.0 l	8.0 l	5.5 l	14.9 l	0.3 l	3.2 l	8	0
27	IL79-002T-B-B	19.4 l	40.3 l	11.1 l	11.0 l	4.0 l	11.8 l	3.4 l	21.1 l	8	0
28	KY99C-1298-08-1	30.0 l	50.4 l	19.9	35.0	13.7 l	24.3 l	7.8 h	40.9 l	5	1
29	KY99C-1051-03-1	33.3	67.8 h	25.0	43.0	14.8 l	33.6	9.1 h	52.1	1	2
30	KY99C-1176-02-1	31.7	63.3 h	19.7	37.0	28.8 h	34.4	7.5 h	62.2 h	0	4
31	MO 050600	21.7 l	39.1 l	10.0 l	20.0 l	9.8 l	18.9 l	1.8 l	18.0 l	8	0
32	MO 050699	22.6 l	50.8 l	12.6 l	25.0 l	10.6 l	22.5 l	4.9	6.0 l	7	0
33	MO 050917	24.3 l	55.7 l	16.0 l	37.0	6.9 l	31.0	4.4 l	27.5 l	6	0
34	MO 050921	19.9 l	42.7 l	10.4 l	15.0 l	6.4 l	17.2 l	2.7 l	4.0 l	8	0
35	VA06W-598	28.9 l	51.4 l	22.0	30.0	11.3 l	25.7	4.0 l	60.7	4	0
36	VA06W-557	35.5	62.4	27.1	50.0	26.3 h	30.5	8.4 h	42.4	0	2
37	VA06W-595	25.0 l	49.9 l	16.7 l	25.0 l	13.7 l	22.3 l	3.4 l	37.5 l	8	0
38	VA06W-608	30.9	62.6	17.8 l	35.0	16.0 l	30.8	5.6 h	32.5 l	3	1
39	VA06W-627	35.1	62.7	27.8 h	38.0	19.6 l	34.4	7.2 h	66.0 h	1	3
40	OH03-183-32	19.5 l	46.1 l	12.2 l	15.0 l	7.7 l	19.7 l	2.0 l	32.9 l	8	0
41	OH03-235-2	28.7 l	61.4	19.6	38.0	27.3 h	30.8	6.3 h	61.5 h	1	3
42	OH03-41-45	34.3	56.6	21.6	30.0	14.5 l	29.4	4.7	52.7	1	0
43	OH03-97-6	18.8 l	47.7 l	10.0 l	33.0	18.6 l	24.1 l	4.0 l	37.8 l	7	0
44	OH03-75-58	23.6 l	50.0 l	13.3 l	60.0	12.5 l	32.5	3.5 l	40.8 l	6	0
	Average	28.5	52.7	17.8	31.7	14.9	26.9	4.6	36.7	4.8	1.2
	LSD	13.9	17.5	11.6	17.8	19.6	12.8	4.3	38.6		
	R2	0.6	0.6	0.6	0.8	0.7	0.8	0.9	0.4		
	CV	42.5	42.5	65.5	33.4	65.3	29.4	55.3	90.1		
	# Locations	6	6	8	1	2	3	3	1		

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

Table 6. Summary of results of the 2006-07 NUWWSN (l,h indicate a mean that is not significantly different than the lowest (l) or highest (h) mean in the col.).

	NAME	SEV	INC	IND	KR	PSS	ISK	DON	GHS	#l	#h
1	ERNIE	27.1	45.7	16.8	40.6	4.3 l	30.7	6.2	18.0 l	2	0
2	TRUMAN	11.2 l	26.7 l	6.1 l	17.5 l	12.6 l	16.6 l	3.9 l	3.4 l	8	0
3	FREEDOM	24.0	47.2	15.2 l	38.4	7.1 l	34.9	5.8	20.7 l	3	0
4	PIONEER 2545	39.0 h	62.3 h	30.0 h	49.8 h	18.5 lh	50.3 h	11.6	50.5 h	1	7
5	NY88046-8138	38.6 h	54.1 h	24.8 h	44.6	28.0 h	48.5 h	7.8	46.6 h	0	6
6	NY93285SP-7343	25.6	33.3	18.4	24.4 l	33.2 h	32.6	4.4 l	22.3 l	3	1
7	NY93285-7110	34.1 h	32.7	15.8 l	26.3 l	24.4 h	29.5	4.2 l	32.8	3	2
8	NY91028SP-9082	29.9	58.7 h	20.4	36.9	25.1 h	42.2 h	11.6	42.6 h	0	4
9	NY93306-7091	24.2	52.2 h	17.1	45.6	31.4 h	37.9 h	10.1	42.6 h	0	4
10	KS04HW47-3	41.5 h	62.7 h	34.1 h	65.0 h	21.2 h	46.1 h	16.6 h	70.6 h	0	8
11	KS04HW101-3	33.8 h	49.2	24.5 h	41.3	31.8 h	35.4	8.8	40.5	0	3
12	P.011035A1-71	29.7	53.9 h	23.4	31.5	21.4 h	43.4 h	4.2 l	22.1 l	2	3
13	P.011036A1-14	32.6 h	46.9	23.0	32.7	18.6 lh	39.0 h	4.9 l	10.6 l	3	3
14	P.02444A1-23-6	24.7	46.2	17.1	17.1 l	16.1 l	39.8 h	3.9 l	19.8 l	4	1
15	P.03647A1-1	23.6	38.7	13.8 l	22.9 l	17.8 l	27.6	3.4 l	10.2 l	5	0
16	P.04287A1-10	30.4 h	51.2 h	23.8	39.4	13.2 l	34.7	6.6	21.8 l	2	2
17	NE01643	22.5	44.1	19.2	41.6	10.9 l	30.3	6.7	21.4 l	2	0
18	HARRY	23.2	51.1 h	17.6	57.1 h	16.4 l	35.9	11.2	14.1 l	2	2
19	NI04421	28.5	56.8 h	20.3	46.6	25.7 h	39.2 h	10.3	19.5 l	1	3
20	NE04653	27.2	51.5 h	20.6	45.6	20.6 h	31.1	8.8	19.7 l	1	2
21	NE03490	29.1	49.8	21.8	41.0	16.8 l	37.5 h	7.2	25.9 l	2	1
22	MD01W233-06-11	27.2	48.3	19.1	25.2 l	24.4 h	31.1	3.7 l	25.2 l	3	1
23	M03-3002	33.1 h	50.8 h	26.0 h	36.5	12.7 l	43.7 h	6.1	35.6	1	4
24	M03-3104	31.4 h	64.3 h	25.5 h	50.9 h	22.6 h	49.5 h	8.4	59.1 h	0	7
25	M03-3616	28.2	45.2	20.5	26.5 l	27.0 h	36.5 h	4.9 l	21.7 l	3	2
26	M03*3877	27.5	61.8 h	23.3	37.9	20.2 h	37.8 h	8.5	26.7 l	1	3
27	M03*3861	27.6	61.6 h	21.9	44.1	11.7 l	34.1	8.7	20.3 l	2	1
28	RCUOG19/21	28.6	44.9	15.8 l	34.0	14.2 l	29.5	7.8	31.2 l	3	0
29	RCUOGF110202D/4	22.9	25.7 l	14.7 l	32.1	11.1 l	23.5 l	1.7 l	10.7 l	6	0
30	RCUOGF111202A/3	35.4 h	58.5 h	21.9	32.5	22.4 h	42.5 h	5.9	25.5 l	1	4
31	RCUOGDHACF1109O2D	18.3 l	21.4 l	12.1 l	47.1	22.7 h	26.7 l	3.9 l	25.4 l	6	1
32	RCUOGNS984-1	38.0 h	61.1 h	28.3 h	45.6	26.6 h	42.6 h	8.2	46.9 h	0	6
33	IL00-8530	30.3 h	48.4	20.0	18.4 l	19.9 h	30.9	3 l	14.7 l	3	2
34	IL01-11445	32.5 h	39.2	22.3	15.9 l	7.0 l	34.0	3.5 l	11.1 l	4	1
35	IL01-11934	24.0	40.2	18.0	15.0 l	11.9 l	26.3 l	3.1 l	11.6 l	5	0
36	IL02-19463	34.7 h	37.4	22.5	21.0 l	9.7 l	34.8	4 l	25.3 l	4	1
37	IL02-23168	28.8	45.3	20.5	24.1 l	19.1 lh	32.4	4 l	31.8 l	4	1
38	KY97C-0540-01-03	35.4 h	57.9 h	25.6 h	30.0	14.5 l	40.3 h	7.1	39.6	1	4
39	KY97C-0554-03-06	23.7	55.7 h	16.0 l	38.8	22.5 h	42.9 h	4.4 l	12.1 l	3	3
40	KY97C-0554-04-05	24.0	48.6	16.8	23.5 l	30.4 h	42.3 h	4.1 l	9.8 l	3	2
41	KY97C-0508-01-01A-1	23.8	42.8	17.9	45.9	27.7 h	31.9	6.4	28.5 l	1	1
42	KY97C-0554-03-02	20.9 l	50.7 h	16.4	22.9 l	15.5 l	29.2	3.5 l	8.8 l	5	1
43	MO 040165	20.2 l	34.7	12.2 l	16.2 l	15.4 l	25.6 l	3.1 l	4.6 l	7	0
44	MO 050101	21.4 l	40.4	12.7 l	26.2 l	7.3 l	26.3 l	3.9 l	4.5 l	7	0
45	MO 050143	19.5 l	38.1	12.2 l	20.6 l	7.1 l	26.7 l	3.9 l	6.2 l	7	0
46	MO 050197	17.7 l	33.7	11.6 l	17.9 l	8.6 l	21.3 l	3.7 l	39.1	6	0
47	MSU Line E3023	35.4 h	50.0 h	22.0	28.1	12.7 l	38.7 h	7.3	50.7 h	1	4
48	MSU Line E5015	27.2	55.3 h	20.8	29.1	23.6 h	33.9	10.7	17.1 l	1	2
49	MSU Line E6001	19.1 l	41.5	14.2 l	33.1	14.4 l	25.0 l	4.6 l	4.9 l	6	0
50	MSU Line E6002	17.5 l	27.1 l	12.9 l	35.6	13.9 l	26.7 l	4.6 l	25.5 l	7	0
51	MSU Line E6003	14.0 l	18.3 l	6.1 l	24.4 l	17.4 l	12.6 l	4.8 l	8.4 l	8	0
52	VA06W-600	30.1	45.1	27.3 h	29.4	22.5 h	42.2 h	4.1 l	39.5	1	3
53	VA06W-602	32.9 h	49.6	27.7 h	19.0 l	18.5 lh	38.1 h	4.1 l	26.8 l	4	4
54	VA06W-587	33.7 h	55.4 h	23.6	20.9 l	23.4 h	43.1 h	4 l	34.4	2	4
55	VA06W-594	25.9	52.3 h	18.3	24.4 l	13.8 l	35.8	4.8 l	18.8 l	4	1
56	VA06W-585	21.2 l	47.4	18.6	9.4 l	15.1 l	34.2	3.5 l	19.1 l	5	0
57	OH02-15978	25.7	41.5	17.5	37.9	14.1 l	31.3	5.7	50.2 h	1	1
58	OH02-12678	19.4 l	39.5	17.0	15.3 l	11.2 l	25.2 l	4.2 l	13.3 l	6	0
59	OH02-12686	20.2 l	35.5	14.6 l	25.0 l	17.1 l	26.0 l	2.1 l	24.0 l	7	0
60	OH02-13567	18.8 l	43.8	12.8 l	28.5	15.8 l	29.6	5.2 l	12.3 l	5	0
	AVERAGE	26.9	46.2	19.2	31.9	18.0	34.1	5.9	24.9	3.1	1.9
	LSD	11.2	14.3	10.0	17.6	14.8	14.1	3.9	28.8		
	R2	0.53	0.73	0.52	0.79	0.48	0.52	0.8	0.71		

DEOXYNIVALENOL ACCUMULATION AND FUSARIUM HEAD BLIGHT SEVERITY IN WINTER WHEAT AFTER SPRAY-INOCULATION WITH MIXTURE OR SINGLE ISOLATES OF *FUSARIUM GRAMINEARUM*.

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OBJECTIVES

1) to compare aggressiveness of four *F. graminearum* isolates and their mixture based on FHB severity and DON accumulation in grain, after spray-inoculation of winter wheat cultivars with known FHB resistance and 2) to test the influence of isolates, wheat cultivar, year and their interactions on level of FHB symptoms and DON accumulation to ensure that there was no isolate-specific resistance from different wheat cultivars with respect to DON accumulation.

INTRODUCTION

Fusarium head blight (FHB), caused by *Fusarium graminearum* (Schwabe), is an important wheat disease. In Canada FHB is caused primarily by *Fusarium graminearum* (Schwabe) [teleomorph: *Gibberella zeae* Schw. (Petch)]. Apart from yield and quality losses, the most serious concern associated with FHB infection is the contamination of the harvested crop with mycotoxins. Deoxynivalenol (DON) is the mycotoxin most commonly recovered from wheat grain in Canada. In Ontario, losses in winter wheat production from yield and quality reductions, were more than $ 100 million CAD in 1996 (Schaafsma , 2002).

Host specific strains have not been demonstrated and resistance to FHB in wheat is generally considered horizontal (race-nonspecific) (Van Euwijk et al., 1995).

When breeding for FHB and DON resistance, a breeder often has to rely on FHB symptoms rather than to perform costly DON analysis. However, FHB severity and DON content are not always well correlated; we have experienced correlations, from as low as r=0.18, to as high as r=0.70, depending on the year and level of FHB resistance in cultivars/lines tested in *F. graminearum* inoculated nurseries (unpublished data). Resistance to FHB infection and DON content may be controlled by independent loci or genes (Somers et al., 2003; Tamburic-Ilincic et al., 2002).

MATERIALS AND METHODS

Winter wheat cultivars were planted in mid October in 2003, 2004 and 2005 at Ridgetown, Ontario, Canada in single rows, 4 m long, spaced 17.8 cm apart, containing 270 seeds each. The experiments were arranged in a 5 x 4 factorial randomized complete block design with four replications. There were four cultivars (FHB susceptible (S) 'AC Ron' and three FHB moderately resistant (MR): 'Wisdom', 'Vienna' and 'AC Morley') and four isolates of *F. graminearum* (DAOM178148, DAOM234041, DAOM234042 and DAOM234043) and a mixture of the four.

Spray-inoculations of each row with a suspension of 50-ml of macroconidia (50,000 spores/ml) of each of four *F. graminearum* isolates (1-4) and their mixture (5) were done, when primary wheat heads were at 50% anthesis for each cultivar (Zadoks Growth Stage, ZGS 65) (Zadoks et al., 1974) using a back-sprayer. The suspension was produced in liquid shake culture using Bilay's medium. *F. graminearum* isolate #1 (DAOM178148) was obtained from Agriculture Canada (ECORC, Ottawa, Ontario, Canada) and was isolated from wheat. Isolates #2 (DAOM234041), 3 (DAOM234042), and 4 (DAOM234043), were isolated from spring barley varieties 'Chapais', 'AC Stephen', and 'C231-0141', respectively in 2000 at Elora Research Station, Guelph, Ontario, Canada by

Tamburic-Ilincic. After isolation the isolates were kept in liquid nitrogen and they have been submitted to the Canadian collection of fungal cultures in Ottawa and are available as DAOM234041, DAOM234042 and DAOM234043. All the plots were misted daily beginning after the first inoculations with about 7.5 mm of water each day. The mist system was engaged until three days after the last cultivar was inoculated. In each year, cultivars were assessed for visual symptoms when the early dough stage (ZGS 83) was reached. Disease levels were estimated as Fusarium head blight severity on a scale of 1-9 where 1 was disease free and 9 was total. The entire grain sample for each cultivar was harvested in mid July in 2004, 2005 and 2006 and finely ground through a ROMER mill (Model 2A, Romer Labs, Inc. Union, MO). Deoxynivalenol (DON) content was estimated from the three replications with highest mean FHB severity using a quantitative fluorometric test-FluoroQuan (Romer® Labs, Inc, Union MO).

PROC UNIVARIATE (SAS Institute Inc., 2003) was used to test ANOVA assumptions to determine if transformations were needed. DON content was transformed by log (x+0.5). The data was analyzed using PROC GLM (SAS Institute Inc., 2003) where year, cultivar, isolate and their interactions were sources of variation.

RESULTS AND DISCUSSION

FHB severity and DON accumulation in this experiment depended on year, isolate and cultivar and the interaction between year and isolate and year and cultivar, but there was no significant interaction between isolates and cultivars (Table 1), suggesting that variation in resistance in these winter wheat cultivars was independent of the variation in aggressiveness of the *F. graminearum* isolates tested. The results from the present study are also in agreement with Mesterhazy (1997) who also reported that DON level in wheat depends on both cultivar and isolate. FHB severity and DON content in winter wheat cultivars after inoculation with four isolates and their mixture during three years are shown in Fig. 1 and Fig. 2.

Aggressiveness of F. graminearum isolates

High isolate x year interaction was reported in the present study (Table 1). Average FHB severity after spray-inoculation with isolates #3 (DAOM234042), #4 (DAOM234043) and the mixture of all (5) four across the cultivars was higher in 2004 than in 2005 or 2006 (Fig. 1a). Average DON accumulation across the cultivars was also higher in 2004 than in 2005 or 2006 (Fig. 1b). In 2004, after spray-inoculation with isolate #4 (DAOM234043) the DON level across the cultivars was significantly higher than after inoculation with the three other isolates or the mixture of all four (Fig. 1b). The summer of 2005 and 2006 was hot and dry and was not favorable to DON accumulation in winter wheat in Ontario. DON content measured in a survey of winter wheat in Ontario in 2005 and 2006 ranged from 0.1 ppm to 1.0 ppm and from 0.1 ppm to 0.3 ppm, respectively, which was much lower than found in a similar survey in 2004, where the highest level of DON was 4.9 ppm (Tamburic-Ilincic et al., 2006).

FHB susceptibility and DON accumulation in winter wheat cultivars

In 2004 and 2005, the average FHB severity was the highest for the FHB-susceptible cv. 'AC Ron' and significantly higher than three FHB MR cultivars (Fig. 2a). In 2006, cv. 'AC Ron' had significantly higher FHB severity than 'AC Morley' and 'Wisdom' (Fig. 2a).

'AC Ron' had the highest average DON level after inoculation in all three years; while the relative ranking of the three MR cultivars was slightly different in each year (Fig. 2b).

In 2004, 'Wisdom' had the lowest DON level (Fig. 2b); 'Wisdom' and 'Vienna' accumulated the least DON in 2005 (Fig. 2b). There was no significant difference in DON accumulation among MR cultivars in 2006 (Fig. 2b).

In conclusion, it is cautiously suggest that perhaps the selection of isolates may be less important when

screening for FHB symptoms than when screening for the propensity to accumulate DON. Relatively similar FHB ratings between years resulted in very different DON levels between years. Although inconclusive the data suggest that a mixture of isolates might contribute some stability in results over years. This possibility would need further study with more cultivars under more environments and years. It seems from the results that high DON-producing isolates would allow for differentiation among cultivars which differ in their propensity to produce DON, and are recommended for screening wheat cultivars for simultaneous FHB resistance and DON accumulation. A highly pathogenic isolates of *F. graminearum*, that simultaneously produce a high level of FHB symptoms and DON, or a mixture of isolates is recommended for breeding programs targeting FHB resistance and reduced DON production in winter wheat.

ACKNOWLEDGEMENTS

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Table 1. Analysis of variance for the effect of year, *Fusarium graminearum* isolate, winter wheat cultivar, and interactions on: a) FHB severity (1-9) and b) transformed (log (x+0.5)) deoxynivalenol (DON) content (ppm) in winter wheat. Ridgetown, ON, 2003-2004, 2004-2005 and 2005-2006.

a) FHB severity (1-9)

Source	df	Mean square	*F*	*P>F*
Year	2	6.254	19.16	<.0001
Isolate	4	4.235	12.98	<.0001
Cultivar	3	1.249	3.83	0.0098
Year*Isolate	8	4.192	12.84	<.0001
Year*Cultivar	6	3.249	9.95	<.0001
Isolate*Cultivar	12	0.474	1.45	0.1459
Year*Isolate*Cultivar	24	0.547	1.68	0.0310

b) DON accumulation

Source	df	Mean square	*F*	*P>F*
Year	2	90.034	427.24	<.0001
Isolate	4	1.584	7.52	<.0001
Cultivar	3	5.027	23.86	<.0001
Year*Isolate	8	1.710	8.11	<.0001
Year*Cultivar	6	2.125	10.09	<.0001
Isolate*Cultivar	12	0.202	0.96	0.4912
Year*Isolate*Cultivar	24	0.195	0.93	0.5671

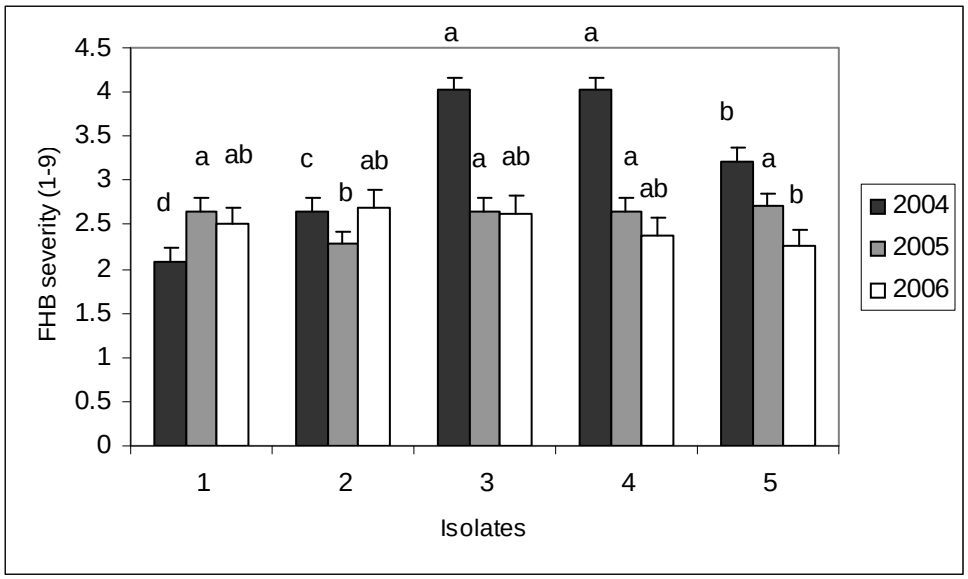

Figure 1 a

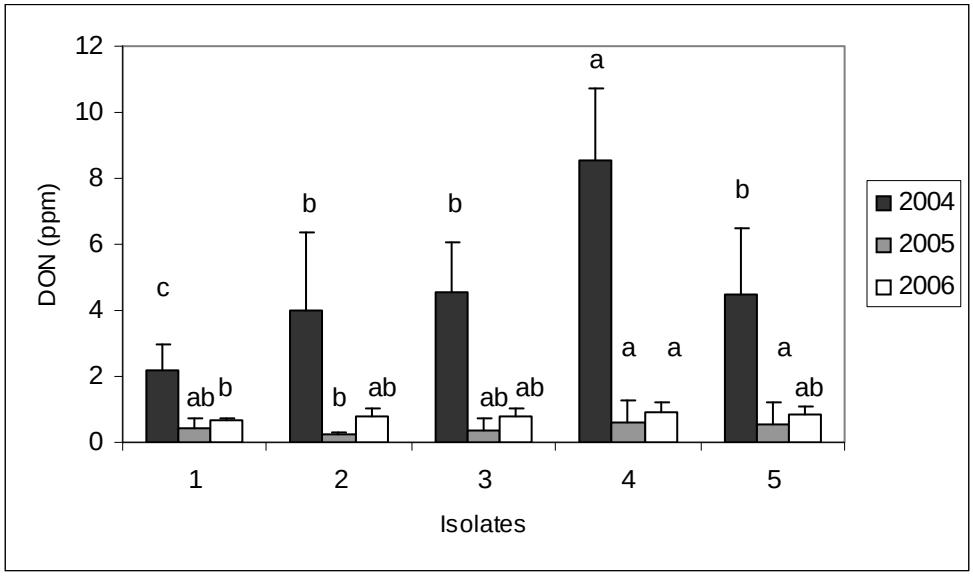

Figure 1 b

Figure. 1. The effect of *Fusarium graminearum* isolates (1-4) and their mixture (5) (± SE) on: a) FHB severity (1-9) and b) deoxynivalenol (DON) content (ppm) across winter wheat cultivars. Ridgetown, ON. Means within years followed by the same letter are not different according to Fisher's protected least significant difference test (P= 0.05).

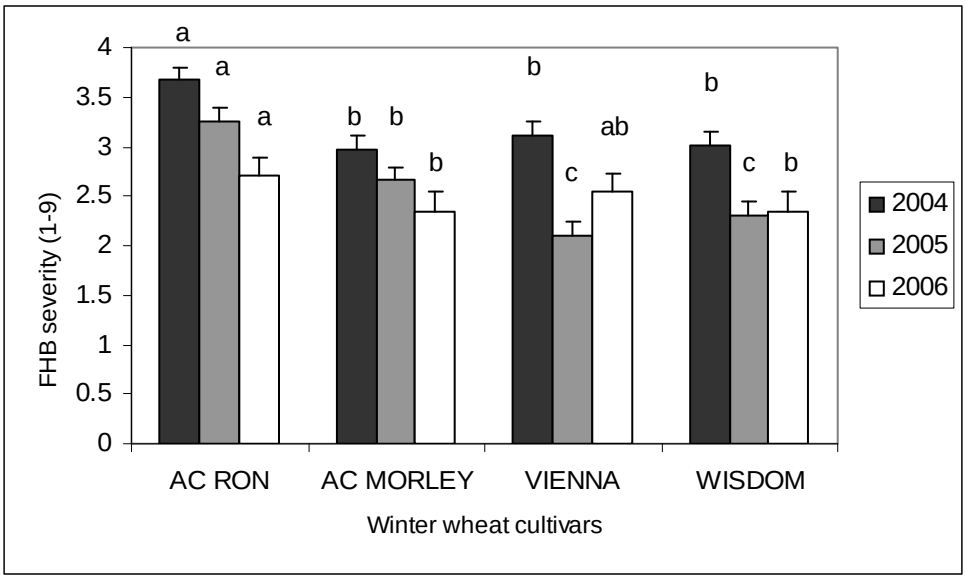

Figure 2 a

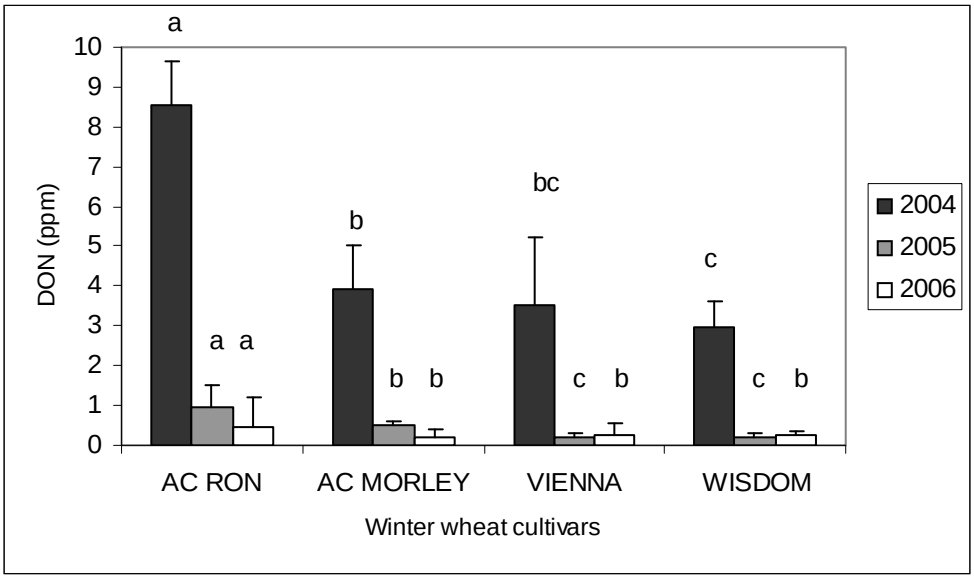

Figure 2 b

Figure. 2. The effect of winter wheat cultivars ('AC RON', 'AC Morley', 'Vienna' and 'Wisdom') on: a) FHB severity (1-9) and b) deoxynivalenol (DON) content (ppm) (± SE) after spray-inoculation across *Fusarium graminearum* isolates. Ridgetown, ON. Means within years followed by the same letter are not different according to Fisher's protected least significant difference test (P= 0.05).

THE EFFECT OF *FUSARIUM SOLANI* METABOLITES ON PEROXIDASE ACTIVITY IN POTATO.

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ABSTRACT

The fungus *Fusarium solani* cause the dry rot of potato tubers. The disease advances in tubers during storage. It leads to great losses of harvest. The reveal of qualities of host- pathogen interaction and natural biochemical mechanisms of tolerance is an actual scientific problem. It is known that so-called "oxidative burst" is one from early defense reaction of plant cells to attack of pathogens. The formation reactive oxygen species such as hydrogen peroxide, superoxide radicals etc. to result from "oxidative burst". The many enzymes involved into cascade of oxidative reaction. The peroxidase (POD, EC.1.11.1.7) is one antioxidant enzymes which play important role in defense reaction. POD to oxidize substances with use hydrogen peroxide or molecular oxygen and participate in biosynthesis of toxic compounds for pathogen and processes of lignification or suberinization for forming barriers against pathogens. The various POD activate on different stages of protective mechanism. The role of soluble and cell-wall bound forms of POD in protective reactions of host-pathogen system "Solanum tuberosum – *Fusarium solani*" was studied. The changes of enzyme activity in tubers and *in vitro* cultivated cells of potato by additional different fractions of fungal metabolites were investigated. Changes showed response reactions of cells depended from plant's sensitivity, localization of enzyme and chemical composition of fungal isolates. The effect of proteins and non-proteins isolates of cultural filtrate and mycelium on the disks of tubers was studied. The proteins of cultural filtrate stimulated rapid induction of soluble (in 2-2,5 time) and especially cell-wall bound (in 5,5-8 time) forms of POD as in tolerance as in sensitivity sorts. The non-proteins isolates also insignificantly induced soluble PODs (in 1,8-2 time). Induction of cell-wall bound PODs depend from resistible of plant. In tolerance sort was more significant increase (in 4-6 time) of enzyme activity than in sensitivity (in 1,2-1,5 time). The influence of metabolites of cultural filtrate and mycelium on POD activity in suspension cells was studied. The metabolites of cultural filtrate induced activity only of soluble forms POD. The magnitude of induction depended from filtrate concentration. The low concentrations (dilution 1:50) increased POD in sensitivity, but high (dilution 1:5) – in resistible cells. Metabolites of mycelium along with cytotoxic effect to suspension cells caused exchanges as soluble as cell-wall bound PODs. The metabolites of ethanol fraction of mycelium induced rapid increase of soluble forms in 2 time, but metabolites of acetone and water-soluble fractions increased activities only cell-wall bound PODs in 2-3 and 3-5 time accordingly and independently from initial sensitivity of cells to fungus.

SEARCHING FOR NEW SOURCES OF FHB RESISTANCE IN THE RELATIVES OF WHEAT.

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ABSTRACT

Epidemics of Fusarium head blight (FHB), caused mainly by *Fusarium graminearum* Schwabe, have threatened the production of bread wheat (*Triticum aestivum* L.) and durum wheat (*T. turgidum* L., subsp. *durum*) in North America in recent years. Deployment of FHB-resistant cultivars has been considered the most efficient and cost-effective strategy to combat this disease. However, only limited sources of FHB resistance are currently available, especially in durum, which makes the development of FHB-resistant varieties difficult. In an effort to identify novel sources of FHB resistance, we have screened about 900 accessions of wheat relative species and their derived lines for Type II resistance in greenhouse and in field nurseries (Fargo and Langdon, ND) during the past four years. A number of accessions and derived lines of the relative species have exhibited resistance or moderate resistance to FHB in these screening experiments. Resistant lines include 16 *T. carthlicum* and 20 *T. dicoccum* accessions, two synthetic hexaploid wheat lines, one *T. timopheevi*-derived hexaploid line, one 'Fukuhokomuji'-*Elymus rectisetus* disomic addition line, and two 'Chinese Spring'- *Thinopyrum junceum* disomic addition lines. These materials likely represent new sources of FHB resistance for durum and bread wheat. The resistant tetraploid wheat accessions are currently utilized for developing durum wheat germplasm resistant to FHB. Introgression of FHB resistance from the derived lines of wild species is currently in progress.

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COMPARISON OF BARLEY SEED PROTEOMIC PROFILES ASSOCIATED WITH FUSARIUM HEAD BLIGHT REACTION.

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ABSTRACT

Plants have evolved a complex array of chemical and enzymatic defenses expressed both constitutive and inducible, that influence pathogenesis and disease resistance. To better understand the constitutive molecular mechanisms associated with the differential reactions to FHB, mature seed of barley genotypes and sister lines differing in FHB reactions were subject to proteome analysis using two dimensional gel electrophoresis (2DE). A total of 38 protein spots were correlated with FHB resistance and susceptibility. Several of these proteins were previously identified as important for disease resistance or as pathogen related proteins (PR-protein). Aldehyde dehydrogenase (BIS1) upregulated in resistant lines has been previously identified as a PR-protein upregulated in barley during stem infection. Protein spot #155 also upregulated in resistant sister lines, was identified as aconitate hydratase. Aconitate hydratase is important for Pto-mediated plant defense response. Putative NADP Malic enzyme found elevated in resistant barley lines, has been previously identified in EST libraries prepared from the barley lemma, palea and FHB infected spikes. Alpha amylase inhibitor (BDAI-1) also upregulated in resistant lines is also known to have antifungal activities. Several proteins were identified to ESTs or proteins, for which their functions were unknown. Selecting for constitutively expressed proteins or enzyme activities that correlate with enhanced FHB resistance may allow the development of new *in vitro* tissue test that could be used to select for improved FHB resistance in new barley lines.

NOVEL FUSARIUM HEAD BLIGHT RESISTANCE IN *TRITICUM AESTIVUM* REVEALED BY HAPLOTYPING WITH DNA MARKERS ASSOCIATED WITH A KNOWN RESISTANCE QTL.

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ABSTRACT

Fusarium head blight (FHB) is a devastating disease of wheat (*Tritcium aestivum* L.) that causes reduced grain yield, and the fungus, *Fusarium graminearum*, produces a mycotoxin, deoxynivalenol (DON), that renders infected wheat grain to be unfit for food or feed. Several sources of resistance in wheat and certain related grass species have been identified. However, research to date indicates that several resistance genes need to be combined to result in highly effective resistance. Thus, there is need to identify additional novel sources of resistance. Xing 117, a wheat line obtained from China that has FHB resistance, the seven other wheat lines Ning 7840, Frontana, Wangshuibai, Arina, Renan, F201R, and Chokwang, all previously identified as having partial FHB resistance, as well as P9762 and P9774 that are susceptible to FHB, were haplotyped at marker loci that are associated with FHB resistance of the seven partially resistant wheat lines. Xing 117 was polymorphic at all previously identified marker loci of the seven wheat lines with partial FHB resistance, as well as these marker loci of P9762 and P9774, indicating that the FHB resistance of Xing 117 is likely novel compared to the resistances of the seven wheat lines in this study.