

# Host Plant Resistance Genes for Fusarium Head Blight: Sources, Mechanisms, and Utility in Conventional Breeding Systems

J. C. Rudd,\* R. D. Horsley, A. L. McKendry, and E. M. Elias

## ABSTRACT

**Fusarium head blight (FHB), caused by *Fusarium graminearum* Schwabe [teleomorph *Gibberella zeae* (Schwein.)], also known as scab, is a destructive disease of wheat (*Triticum aestivum* L.; *T. durum* L. var *durum*) and barley (*Hordeum vulgare* L.). Host resistance has long been considered the most practical and effective means of control, but breeding has been hindered by a lack of effective resistance genes and by the complexity of the resistance in identified sources. This paper will provide an overview of progress in developing host plant resistance for FHB, primarily in the USA, by review of the sources of resistance in wheat and barley, and their utilization in breeding programs. Although there are no reported sources of immunity, considerable genetic variability exists for resistance in both wheat and barley. Sources of resistance in durum, however, are limited. The strategy of breeding programs is to recombine different types and sources of resistance steadily through traditional breeding strategies. To facilitate selection, artificial inoculation techniques are used in both the field and greenhouse. This enables breeders to select simultaneously for resistance and desirable agronomic characteristics. Incremental increases in resistance are being reported in hexaploid wheat and to a lesser extent in barley and durum wheat. It is anticipated that the development of molecular markers will improve the efficiency of developing FHB wheat and barley cultivars.**

**FUSARIUM GRAMINEARUM** Schwabe [teleomorph *Gibberella zeae* (Schwein.)], also known as Fusarium head blight or scab is a destructive disease of wheat and barley in warm and humid wheat growing regions of the world. FHB has recently gained prominence as a national research problem in the USA because of the more frequent U.S. epiphytotic thought to be the result of the increased emphasis on conservation tillage (Bai and Shaner, 1994; Wilcoxson et al., 1988), rotations with corn (*Zea mays* L.) (Windels and Kommedahl, 1984), the lack of effective cultural and/or fungicide control (McMullen et al., 1997), above average precipitation and/or humidity during flowering and early grain fill (McMullen et al., 1997), and the lack of effective sources of genetic resistance. In addition to reduced kernel density and discoloration at harvest, associated deoxynivalenol (DON) accumulation prevents grain from being marketable. Host resistance has long been considered the most practical and effective means of control (Martin and Johnston, 1982; Schroeder and Christensen, 1963), but breeding has been hindered by a lack of effective resistance genes and by the complexity of the resistance in identified sources (Mesterházy, 1997). No

source of complete resistance is known, and current sources provide only partial resistance.

The United States Department of Agriculture (USDA) currently ranks FHB as the worst plant disease of wheat and barley since the stem rust (caused by *Puccinia graminis* Pers.:Pers.) epidemics of the 1950s (Wood et al., 1999). FHB epidemics have been documented in 26 states and five Canadian provinces. Yield losses in wheat since 1990 have exceeded 13 Tg (500 million bushels) with economic losses estimated at \$2.5 billion (Windels, 2000). Wheat yields in 1993 were reduced by about 50% in northeastern North Dakota and 40% in northwestern Minnesota compared with 1992 (National Agricultural Statistics Service, 1993–1999). In barley, losses have been equally devastating with estimated losses from 1993 to 1999 totaling in excess of \$400 million (Windels, 2000). Since 1993, North Dakota, South Dakota, and Minnesota have lost 73% of their malting barley market with losses in Minnesota alone approaching 95% (Windels, 2000).

The resurgence of FHB has united scientists, producers, industry, and politicians in an effort to find a solution to this disease. Nearly \$10 million in new public and private funding has been generated to support FHB research and education in the USA (Windels, 2000). In 1999, the U.S. Wheat and Barley Scab Initiative supported research efforts of 66 principal scientists working on 111 research projects at 19 Land Grant Universities and the USDA Agricultural Research Service (U.S. Wheat and Barley Scab Initiative news release, <http://www.scabusa.org/newpage13.htm>, 1 Dec. 2000).

Scab is not a new problem. As early as 1891, Arthur (1891) stressed the importance of breeding for resistance to head blight in wheat. In the 1920s, plant pathologists and breeders observed that wheat genotypes differed in their susceptibility to FHB; however, differences in maturity made it hard to separate genotypic differences in susceptibility from disease escape (Immer and Christensen, 1943). Much of our present knowledge about FHB originates from extensive research that was done at the University of Minnesota from the 1920s through the 1950s (Hanson et al., 1950; Schroeder and Christensen, 1963). A severe rust epidemic, however, changed the focus of wheat research to stem rust and U.S. work on FHB was discontinued (Wilcoxson, 1993).

Worldwide, resistance to FHB is a major focus of wheat and barley breeding programs. In China, FHB research began in the 1950s and continues today (Liu and Wang, 1990). Japanese scientists initiated a major

J.C. Rudd, Plant Science Dep., South Dakota State Univ., Brookings, SD 57006; R.D. Horsley and E.M. Elias, Dep. of Plant Sciences, North Dakota State Univ., Fargo, ND 58105; A.L. McKendry, Dep. of Agronomy, Univ. of Missouri, Columbia, MO 65211. Received 28 March 2000. \*Corresponding author (Jackie\_Rudd@sdstate.edu).

**Abbreviations:** AFLP, amplified fragment length polymorphism; DON, deoxynivalenol; FHB, Fusarium head blight; QTL, quantitative trait locus; RFLP, restriction fragment length polymorphism; RIL, recombinant inbred line; SSR, single sequence repeat.

research effort in the 1960s (Nisikado, 1959) and currently have a strong FHB resistance effort in barley (Takeda and Heta, 1989). FSB resistance is also a major focus of breeding programs in several European countries, notably Hungary, Poland, Austria, Germany, and the Netherlands (Mesterházy, 1995; Meidaner, 1997; Snjders, 1990).

Several reviews have been written recently that address genetic resistance to FHB (Bai and Shaner, 1994; Mesterházy, 1995; Parry et al., 1995; Miedaner, 1997). Although this paper will contain some review of the literature, much of it has been compiled from an informal 1999 survey of 25 U.S. breeding programs working on FHB resistance. It also reports on recent progress in the germplasm and breeding programmatic areas of the U.S. Wheat and Barley Scab Initiative. These progress reports are available in full at the initiative website <http://www.scabusa.org> (verified January 12, 2001).

### Sources of Resistance in Wheat

The spring wheat cultivar Sumai 3, including derived lines such as 'Ning 7840', is arguably the most widely used source of resistance to FHB in the world and is certainly the best characterized. It has been used in Chinese breeding programs for at least 20 yr (Liu, 1984) and since introduction into the USA, it has been used extensively by both spring wheat and winter wheat breeding programs (Wilcoxson, 1993). Sumai 3 has been rated as resistant or highly resistant by most of the programs in which it is used. Breeders have found this source of resistance to be more heritable, stable, and consistent across environments than resistance from most other sources. Problems associated with the use of Sumai 3 as a parent, however, include susceptibility to other diseases and shattering. A caution about the use of Sumai 3 is that there appear to be different selections in circulation that differ for FHB resistance as well as other agronomic traits.

Although Sumai 3 has been widely used by hexaploid wheat breeders, durum breeders have been less successful in using it as a source of resistance. This lack of success initially led researchers to believe that the resistance genes from Sumai 3 might be on the D genome and therefore would not recombine in tetraploid durum (AABB) where the D genome is absent. Since several molecular marker studies have in fact mapped resistance genes from Sumai 3 to the A and B genomes (Kolb et al., 2001), breeders now believe that the genetic background of the elite germplasm used by some durum breeding programs may be suppressing the expression of the Sumai 3 resistance (E. M. Elias, 1999, personal communication).

Other sources of resistance that have been widely used include 'Frontana' from Brazil and 'Nobeokabouzu' from Japan (Dubin et al., 1997). In addition, many incidental sources of resistance to FHB have been identified through routine screening of elite germplasm in breeding programs. Although the resistance is often only intermediate, these sources are attractive to breeders because they produce segregating populations that

are adapted, have FHB resistance, and have acceptable agronomic and end-use quality characteristics. Some of these incidental sources of resistance include '2375' used primarily, in the USA spring wheat region, and 'Ernie' (McKendry et al., 1995) and 'Freedom' (Gooding et al., 1997) used in the eastern soft red winter wheat region of the USA. Although some variability for FHB resistance has been identified in elite durum germplasm, the level of resistance is much lower than that found in hexaploid wheat programs (E. M. Elias, 1999, personal communication).

Several programs have screened wild relatives for FHB resistance, but these efforts have been met with limited success. Wan et al. (1997) evaluated 1463 accessions of 85 species belonging to 17 genera of the *Triticeae*. Variation was found among species and within species. Although no accessions were immune, accessions from 18 species were found to be resistant or highly resistant to both initial infection and the spread of FHB in the spike. The genus *Roegneria* had the highest level of resistance with 67 of the 69 accessions tested being resistant to both initial infection and spread. Additionally, the wide-crossing program of CIMMYT has reported resistance in some synthetic hexaploid wheats, suggesting that some accessions of *Aegilops tauschii* Coss. may be resistant (Gilchrist et al., 1999). These sources of resistance have been difficult to use in breeding programs because of the well known problems associated with the direct use of alien genes including (i) lack of pairing between alien and wheat chromosomes, (ii) the quantitative nature of the resistance, and (iii) the agronomic inferiority of their hybrid progenies (Chen et al., 1997). The wild species may offer some hope to durum breeders, who have struggled to find acceptable levels of resistance in adapted germplasm (Jauhar and Peterson, 1999; Gilbert, 1998). Gilbert (1998) screened 96 accessions of *Triticum dicoccoides* (Koern. ex Asch. & Graebner) Aarons. and found six accessions with a useful level of resistance. Crosses are currently being made to transfer this resistance into elite durum germplasm.

With funding from the U.S. National Wheat and Barley Initiative, aggressive worldwide searches are now underway to identify new sources of resistance to FHB in wheat and barley. Centers have been established at the Univ. of Missouri, North Carolina State Univ., South Dakota State Univ., and North Dakota State Univ. for the introduction, screening, and distribution of new sources of resistance in winter, spring, and durum wheat and in barley (McKendry et al., 1999; Murphy et al., 1999; Zhang et al., 1999; Elias, 1999; Scholz et al., 1999). Researchers at these centers evaluated a total of 6269 accessions of wheat and barley in 1998 and 1999. Resistances identified will be confirmed in further tests and distributed to breeders either through the established regional nursery systems or by direct contact with the program leaders. Complete data for these sources of resistance by commodity group can be found under the germplasm programmatic area at the website of the U.S. National Wheat and Barley Scab Initiative (<http://www.scabusa.org>).

### Sources of Resistance in Barley

Unlike Sumai 3 in wheat, no single barley cultivar or accession is being used on a wide scale as a source of FHB resistance. 'Chevron', a six-rowed, nonmalting barley originating from Switzerland has been used most frequently. The University of Minnesota Barley Breeding Program has used it extensively since the early 1970s as a source of resistance genes for prevention of kernel discoloration (Gebhardt et al., 1992) that can be caused by several fungi, including *F. graminearum*. Chevron has the highest level of FHB resistance and lowest DON content of six-rowed genotypes evaluated to date (Prom et al., 1996). The cultivar MNBrite (Rasmusson et al., 1998), is a Chevron-derived cultivar with FHB resistance intermediate to 'Stander' and Chevron. Chevron-derived progenies with resistance similar to Chevron have been identified; however, they were not released because they were tall, had weak straw, late maturity, and/or thin kernels.

Additional sources of resistance in barley are the two-rowed accessions CIho4196, Zhedar 1, and Zhedar 2 from China; 'Fredrickson' from Japan; 'Harrington' and 'AC Oxbow' from Canada; 'Kitchin' from the USA; and 'Shyri' and 'Atahualpa' from Ecuador but developed and released by the ICARDA/CIMMYT Barley Breeding Program in Mexico. Barley breeders are screening their own elite germplasm for genotypes that may have inherently better FHB resistance and/or lower concentrations of DON than cultivars currently grown. Although moderately resistant genotypes have been identified that may be different from those from Europe and Asia, the levels of FHB resistance and DON accumulation in these genotypes are not as good as those observed in the accessions previously mentioned. Screening of all six-rowed spring barley accessions in the USDA-ARS National Small Grains Collection for FHB resistance is currently underway. About 50 accessions with FHB resistance similar to Chevron were identified in 1999. Seed from these accessions was harvested and DON concentration will be determined. The wild relatives of barley including *H. bulbosum* L., *H. jubatum* L., and *H. spontaneum* K. Koch are also being screened for resistance to FHB; however, no accessions have been identified that have acceptable FHB resistance.

### Resistance Types and Disease Assessment in Wheat and Barley

Resistance types are generally classified as either morphological or physiological. Head anatomy or positioning that contributes to higher humidity around the spikelets is often associated with more disease. Generally, awned genotypes with a short peduncle and a compact spike have faster disease spread than genotypes that are awnless, have a long peduncle, and a lax spike. In addition, short statured genotypes with a long grain-filling duration generally get more disease than tall genotypes that have rapid grain fill (Meidaner, 1997; Mesterházy, 1995). These morphological characteristics contribute to resistance, but are often considered nuisance factors in screening nurseries. They can confound

the data and must be considered when making selections. It is generally agreed that these morphological characteristics are of minor significance compared with physiological resistance discussed below.

Morphological traits have also been associated with FHB resistance in barley. Two-rowed barley is more resistant to FHB than six-rowed barley (Xihang et al., 1991; Takeda and Heta, 1989; Gocho and Hirai, 1987) and in crosses between six-rowed and two-rowed genotypes two-rowed progenies were most resistant, followed by genotypes heterozygous for spike type. Six-rowed types were most susceptible (Takeda, 1990). Lateral floret size in two-rowed barley has also been associated with FHB resistance. Correlations between lateral floret size and Type II resistance, and lateral floret size and DON concentration were 0.63 and 0.54, respectively. A quantitative trait locus (QTL) affecting all three traits has been mapped to the centromeric region on chromosome 2H; however, it was not determined if genes for these three traits were linked or pleiotropic (Zhu et al., 1999).

Mesterházy (1995) described five types of physiological resistance, expanding on the two types described by Schroeder and Christensen (1963). These included (I) resistance to initial infection, (II) resistance to spread within the spike, (III) kernel size and number retention, (IV) yield tolerance, and (V) decomposition or non-accumulation of mycotoxins. In wheat, Type II resistance is most commonly assessed and Sumai 3 is the most commonly used source (Wagester et al, 1999; Liu and Wang, 1990; Dubin et al, 1997). It is measured by observing symptoms due to disease spread after some type of point inoculation. Type II resistance is not measured as frequently in barley as it is in wheat. In barley germplasm from the Midwest, the spread of FHB up and down the spike is not often observed; this has led breeders in that region to conclude that their germplasm may inherently have this type of resistance. Variation for Type II resistance in barley does exist in other regions. The ICARDA/CIMMYT barley breeding program screens for Type II resistance, and a QTL controlling this trait has been mapped to the centromeric region of chromosome 2H (Vivar et al., 1999). Type I resistance is usually measured by spraying plants with a conidial suspension and then counting infected spikelets 7 to 21 d post inoculation. It has also been measured by injecting inoculum into multiple spikelets per spike and assessing the percentage of infected spikelets at maturity (Wan et al., 1997). Accurate assessment of Type I resistance is difficult because the amount of inoculum actually applied is difficult to quantify and disease assessment is confounded by the Type II resistance of the germplasm being evaluated. A genotype must have Type II resistance before Type I resistance can be accurately measured. Type III resistance is measured by threshing infected spikes and observing the damage to the kernels. Kernel number reduction, kernel weight, test weight, or visual estimates of *Fusarium*-damaged kernels (tombstones) are common measurements used to assess Type III resistance. Type IV resistance or yield tolerance can be assessed by measuring grain yield of naturally or

artificially inoculated spikes or plots and comparing the data with spikes or plots that do not show disease symptoms. Several programs have reported variation among cultivars in yield reduction at a given level of disease symptoms on the spikes. From a practical breeding perspective, the measurement of grain yield under heavy disease pressure should be a valuable tool for breeders who have both grain yield and FHB resistance as breeding objectives. Finally, Type V resistance, which is important from a grain utilization perspective, is identified by measuring DON concentration at a given level of FHB.

### Genetics of Resistance in Wheat

Reports on the genetics of known sources of FHB resistance have been inconsistent but all suggest that inheritance is complex. Several studies on Type II resistance in Sumai 3 suggest the presence of two to three resistance genes (Bai et al., 1989; Bai and Shaner, 1994; Zhou et al., 1987). Singh et al. (1995) reported three genes condition resistance in Frontana while van Ginkel et al. (1996) showed that Ning 7840 and Frontana each carry two different dominant genes for resistance. Additive effects are generally found to be greater than non-additive effects (Bai and Shaner, 1994; Snijders, 1990; Zhuang and Li, 1993). This suggests that the accumulation of resistance genes from diverse sources could enhance resistance. Finally, adding to the complexity of the genetics of FHB resistance is the large environmental variance component. Campbell and Lipps (1998) estimate this is often as high as that for grain yield.

There is considerable evidence to support transgressive segregation for FHB resistance (Waldron et al., 1999; Jiang et al., 1994; Snijders, 1990). Sumai 3 was derived from a cross of two lines with intermediate levels of resistance and Ernie was selected from progeny of two moderately susceptible lines (McKendry et al., 1995). van Ginkel et al. (1996) reported that Frontana and Ning 7840 each had two dominant genes and that some of the progeny from the cross of these two lines had better resistance than either parent.

Molecular markers associated with FHB resistance have been identified by several laboratories (Kolb et al., 2001). Waldron et al. (1999) used restriction fragment length polymorphism (RFLP) markers to identify five genomic regions significantly associated with FHB in recombinant inbred lines (RILs) derived from the spring wheat cross Sumai 3 × 'Stoa' while Bai et al. (1999) have successfully used amplified fragment length polymorphism (AFLP) analyses to identify markers associated with FHB resistance in RILs of the cross 'Ning 7840' × 'Clark'. Our understanding of the genetics of FHB resistance will greatly increase as markers, particularly single sequence repeats (SSRs) become more readily available and the mapping of genes for different types and sources of resistance is completed.

### Genetics of Resistance in Barley

Published reports on identification of loci controlling FHB resistance and DON accumulation in barley are

more limited. In a mapping population developed from a cross between Chevron and the elite breeding line M69, de la Pena et al. (1999) identified QTLs associated with FHB resistance, DON content, and kernel discoloration on six of the seven barley chromosomes. QTLs explaining 10% or more of the variation in FHB severity were found in chromosomes 2H and 7H while QTLs each accounting for 10% of the variation in DON accumulation were found in three chromosomes, 2H, 5H, and 7H. Low DON concentration was associated with Chevron for all three QTLs.

QTLs for FHB resistance and DON concentration in mapping populations developed using the resistant parents Zhedar 1 and Zhedar 2 were found in chromosomes 2H and 7H (L. Dahleen, 1999, personal communication). These chromosomal locations agreed with those identified by de la Pena et al. (1999) in Chevron. Efforts to identify QTL associated with DON concentration in these mapping populations are ongoing. Finally, as discussed earlier in this report, a QTL associated with Type II resistance of FHB was identified in chromosome 2H (Vivar et al., 1999). Ultimately, these QTLs will be critical to the incorporation of genes for multiple sources and types of resistance into a single cultivar.

### Screening for Fusarium Head Blight Resistance in Wheat

Screening techniques for FHB can be as diverse as the programs that utilize them. Project goals, level of precision needed, number of lines under evaluation, and available resources are all important considerations when choosing a technique. Common to all techniques are inoculation at anthesis and provision of a favorable environment for infection and disease development. Although several species of *Fusarium* can induce FHB, *F. graminearum* is the main species responsible for recent epidemics in the USA, Canada, and China. In Northern Europe, *F. culmorum* (Wm. G. Sm.) Sacc. is most prevalent. Host specificity has not been shown (Snijders and van Eeuwijk, 1990; van Eeuwijk et al., 1995; Stack et al., 1997). After studying the FHB reaction of 25 European genotypes to 17 strains of *F. culmorum*, *F. graminearum*, and *Microdochium nivale* (Fr.) Samuels & I.C. Hallett (= *F. nivale* Ces. ex Berl. & Voglino), van Eeuwijk et al. (1995) concluded, "Any reasonable aggressive strain, should be satisfactory for screening purposes." Some breeding programs use a single aggressive isolate in their screening programs but since isolate aggressiveness has been shown to be affected by environment, most programs use a mixture of isolates.

### Point Inoculation

Type II resistance is typically evaluated in the greenhouse by inoculating a single central spikelet of a spike at anthesis and measuring the progression of disease symptoms from the point of inoculation. Four to 10 spikes per accession are usually inoculated. Various methods of inoculation have been developed. Typically, a single central floret is inoculated at first anthesis with 5 to 10 mL of a macroconidial spore suspension concen-

trated to 50 000 macroconidia/mL. Although this concentration is common, concentrations ranging from 10 000 to 100 000 macroconidia/mL have been reported. Inoculum is delivered into a floret with a repeat dispensing syringe, needle, a small piece of inoculum-soaked cotton (Bekele, 1985), or a colonized millet [*Setaria italica* (L.) P. Beauv.] kernel (Jin et al., 1999). Humidity is maintained to facilitate infection and disease development by covering the individual spike or the entire plant with plastic wrap or with a misting system. Where the latter is used, misting periods range from 12 to 72 h. Disease progress is recorded by counting the number of diseased spikelets after inoculation. Both the frequency and timing of data collection vary from program to program. In some programs, several observations are made and area under the disease progress curve (AUDPC) is calculated. In others, symptoms are recorded only once, typically 14 to 21 d after inoculation depending on disease progress in the susceptible check. Some programs then harvest and thresh the inoculated spikes at maturity to assess kernel quality.

### Spray Inoculation

Breeding programs often use spray inoculation to evaluate large amounts of material in the field, developing unique protocols to fit their individual needs. Plants in individual rows (typically 1.5 m long with three replications) that are at 50% anthesis are sprayed with a conidial suspension of 50 000 spores/mL. They are often sprayed again 1 wk later to catch spikes that were not in anthesis during the first inoculation. In most nurseries overhead mist irrigation is used during the evening, night, or early morning to enhance disease development. Misting is started when the earliest material in the nursery is inoculated and is often continued until the latest material in the nursery is evaluated. Segregation by maturity group is helpful to avoid long periods of misting, but this is not always possible because the heading date of germplasm accessions is often not known. Symptoms begin to appear 7 to 10 d after inoculation. Most breeders make disease evaluations approximately 21 d after inoculation but these may be earlier or later depending on disease progression. Incidence (percentage of spikes with symptoms), severity (percentage of diseased spikelets on the infected spikes), and disease index (incidence  $\times$  severity) are determined in 20 to 30 spikes per row. Some researchers associate incidence with Type I resistance and severity with Type II resistance; others consider the data as an estimate of a combination of Type I and II. Inoculum concentrations and misting intervals must be adjusted to obtain the desired disease level. Some programs try to obtain 100% incidence, while others want much less. Although Sumai 3 levels of resistance can be identified even at very high disease pressure, intermediate levels of resistance can be overwhelmed (Zhang, 1999).

Sources of variation in field-based evaluations are due to differences in anthesis dates which subject lines being evaluated to different environmental conditions, variable misting patterns in the field, and uneven inocu-

lum distribution. The use of resistant and susceptible checks that differ in maturity is critical to assess both disease levels in the nursery and relative resistance of the lines being evaluated. Four checks are commonly used: early resistant, early susceptible, late resistant, and late susceptible. These checks are seeded at many locations in the field and are often seeded on different dates to check environmental variability.

At maturity, rows are harvested and grain is evaluated for diseased or tombstone kernels and DON concentration. Although these kernel evaluations (except tombstone kernels) can be confounded by the presence of other diseases and environmental conditions, they have proven to be useful to breeding programs and many researchers associate them with Type III and IV resistance. Grain yield, test weight, 1000-kernel weight, and percent shriveled kernels also provide estimates of these two types of resistance. DON concentration provides an estimate of Type V resistance.

### Grain Spawn

Grain spawn inoculation is another method used for the evaluation of large amounts of material in field nurseries. The protocols developed for this method are similar to those developed utilizing spray inoculation with the exception that the inoculum comes from colonized grain that has been spread throughout the field. In most programs, the grain spawn is produced in the laboratory using wheat or corn, but some programs simply use FHB-infected wheat. The grain is normally spread in the field around the boot stage of plant development and then at weekly intervals thereafter. In mist-irrigated nurseries, irrigation is started soon after the grain is spread so that perithecia will be formed by anthesis. Disease assessment is the same as that described for spray inoculation. This method probably comes closest to simulating natural epidemics.

### Screening for FHB Resistance in Barley

Although some protocols exist for greenhouse screening for FHB in barley, most screening is done in the field because of the low correlation between greenhouse and field data. In field nurseries in North Dakota and Minnesota, inoculation is done by either the spray or grain spawn method. Additional mist-irrigated nurseries containing germplasm from these states and Busch Agricultural Resources, Inc. are grown each winter in Hangzhou and Shanghai, China. These nurseries have been grown since 1994 and are overseen by Professor Zhang Bingxing at Zhejiang University (Hangzhou) and Professor Liu Zongzhen at the Shanghai Academy of Agricultural Sciences. Of primary importance to barley breeders are data on FHB severity and DON concentration, since these are the traits that most severely affect the marketing of grain for malting. Scoring for FHB severity is generally done at the hard dough stage in both the greenhouse and field. Fusarium head blight severity is determined as it is in wheat. Facilities for DON concentration are available at NDSU and at the Univ. of Minnesota. Over the last 2 yr, more than 8000

assays for DON concentration have been run by gas chromatography and/or mass spectroscopy methods (Schwarz et al., 1995).

### Breeding for FHB Resistance in Wheat

Most breeders attempt to improve FHB resistance by recombining different sources and types of resistance and simultaneously selecting for resistance and desirable agronomic performance. Most, if not all, have found genetic variability for FHB resistance in their existing germplasm. The level of resistance will increase in this adapted germplasm pool as programs actively screen for FHB resistance. In most breeding programs, highly susceptible lines are eliminated except where they are retained for specific traits. Significant gains have been reported without the use of Asian sources of resistance. These improved lines are being put back into the crossing block where genes they contain are recombined with other sources of resistance.

Most breeders realize that the Type II resistance of Suami 3 is not enough to protect adequately against severe FHB epidemics; hence the strategy is to combine Type II resistance with Type I and kernel retention. Many programs have introgressed the resistance from Sumai 3 into their adapted germplasm and then crossed these lines with other more adapted lines. The recent germplasm release from North Dakota State Univ., ND2710, was developed by this stepwise procedure. A selection from the cross Sumai 3/'Wheaton' was identified that had resistance comparable to Sumai 3 but had better agronomic performance. That selection was then crossed with 'Grandin' to create the population from which ND2710 was selected. Breeding programs can now use ND2710 as a parent instead of Sumai 3.

Traditional breeding methods such as the pedigree method and single seed descent are being used for improving FHB in wheat; however, backcross breeding has not proven to be effective because genetic background appears to influence expression of FHB resistance. Recurrent selection has proven successful (Jiang et al., 1994) and could be useful to accumulate resistance genes conditioning different types and sources of resistance. Finally, to accelerate breeding efforts, doubled haploids are being used to more rapidly achieve homozygosity in selected populations. As well, off-season nurseries and greenhouses are used extensively by spring wheat and barley breeders to accomplish the same objective. Although not a common practice in winter wheat programs, Ohm (1999) has also reported on the use of an off-season nursery for winter wheat.

### Breeding for FHB Resistance and Low DON Concentration in Barley

The history of breeding for FHB resistance in barley in the upper Midwest is more recent, beginning in 1993. The first sources of resistance used were the breeding lines 'Gobernadora' from ICARDA/CIMMYT in Mexico and Zhedar 1 and Zhedar 2 from China. All three lines had the two-rowed spike morphology and were not adapted for production in Minnesota or North Dakota

because of weak straw and susceptibility to other foliar diseases. Prior to the FHB epidemic of 1993, the Univ. of Minnesota's barley breeding program had been breeding for resistance to kernel discoloration using Chevron as a source of kernel discoloration resistance genes. Chevron and Chevron-derived lines from this project were observed to have FHB resistance and this germplasm has subsequently been used by other barley breeders in the upper U.S. Midwest as a genetic base for scab resistance.

A goal of midwestern six-rowed barley programs has been to transfer FHB resistance from two-rowed accessions into elite midwestern six-rowed malting barley germplasm. In crosses between six-rowed malting barley cultivars and Zhedar 1 and Zhedar 2; however, no six-rowed progenies have been identified with FHB resistance similar to that in the two-rowed parent. The lack of success in transferring the resistance from two-rowed to six-rowed barley was thought to be due to insufficient population size, unfavorable linkages between genes controlling row type and FHB resistance, and/or pleiotropy. A cross between 'Foster', a six-rowed malting barley cultivar, and CIho 4196, a two-rowed FHB resistant accession from China, suggested that these negative linkages could be broken when six-rowed progenies were identified with resistance similar to CIho 4196.

Finally, the biggest challenge that barley breeders face is the development of cultivars with the low to zero detectable concentrations of DON necessary for the malting and brewing industry. DON has been found to carry through the malting and brewing processes into the finished beer and has also been associated with gushing in beer (Schwarz et al., 1996). Under high disease pressure, even the most resistant barley accessions accumulate unacceptable levels of DON. Parent building and traditional breeding methodologies are being used to combine different types and sources of resistance to solve this problem. Breeding techniques and strategies are similar to those used in wheat.

In conclusion, the U.S. National Wheat and Barley Scab Initiative, a collaborative, cooperative national research initiative aimed at reducing the devastating losses associated with FHB in both wheat and barley is funding an aggressive attack on this disease in all classes of wheat and barley. Research is ongoing in six major areas including cultivar development, germplasm introduction and introgression, biotechnology, biological and chemical control, epidemiology, and food safety and toxicology. This initiative now drives the FHB research agenda in the USA and promises to enhance breeding for FHB resistance in wheat and barley through the identification of new sources of resistance, genetic analyses of those sources to identify resistances in which alleles differ from those currently known, the development of transgenic cultivars that carry effective antifungal genes, and the development of molecular markers that will enable breeders to combine efficiently and effectively genes conditioning both different sources of resistance and different types of resistance in single cultivars. Over 20 breeding programs in the USA alone are conducting research relative to FHB. Information is

shared at national and international forums. Germplasm centers are identifying and distributing new sources of resistance. Elite lines are being shared via regional testing programs and an international FHB nursery will be in place by 2000. This multi-faceted and cooperative approach should enable breeders to develop highly resistant wheat and barley cultivars thereby, significantly reducing if not eliminating the devastating effects of FHB both nationally and worldwide.

## REFERENCES

- Arthur, J.C. 1891. Wheat scab. *Indiana Agric. Exp. Stn Bull.* 36:129–132.
- Bai, G., F.L. Kolb, G. Shaner, and L.L. Domier. 1999. Amplified fragment length polymorphism markers linked to a major quantitative trait locus controlling scab resistance in wheat. *Phytopathology* 89:343–348.
- Bai, G., and G. Shaner. 1994. Scab of wheat: Prospects for Control. *Plant Dis.* 78:760–766.
- Bai, G., Q. Xiao, and J. Mei. 1989. Studies on the inheritance of scab resistance in six wheat varieties. *Acta Agriculturae Shanghai* 5:17–23.
- Bekele, G.T. 1985. Head scab screening methods used at CIMMYT. p. 169–173. *In* *Wheats for more tropical environments*. CIMMYT, Mexico, D.F., Mexico.
- Campbell, K.A.G., and P.E. Lipps. 1999. Allocation of resources: sources of variation in *Fusarium* head blight screening nurseries. *Phytopathology* 88:1078–1086.
- Chen, P., D. Liu, and W. Sun. 1997. New countermeasures of breeding wheat for scab resistance. p. 59–65. *In* Dubin et al. (ed.) *Fusarium head scab: Global status and future prospects*. CIMMYT, Mexico, D.F., Mexico.
- de la Pena, R.C., K.P. Smith, F. Capettini, G.J. Muehlbauer, M. Gallo-Meagher, R. Dill-Macky, D.A. Somers, and D.C. Rasmusson. 1999. Quantitative trait loci associated with resistance to *Fusarium* head blight and kernel discoloration in barley. *Theor. Appl. Genet.* 99: 561–569.
- Dubin, H.J., L. Gilchrist, J. Reeves, A. McNab (ed.). 1997. *Fusarium head scab: Global status and future prospects*. CIMMYT, Mexico, D.F., Mexico.
- Elias, E.M. 1999. Evaluation of durum germplasm for resistance to *Fusarium* head blight. p. 127. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Gebhardt, D.J., D.C. Rasmusson, and R.D. Wilcoxson. 1992. Cyclic breeding used to incorporate kernel discoloration resistance into malting barley. *Crop Sci.* 32:352–356.
- Gilbert, J. 1998. Comparison of inoculation methods for screening tetraploid wheat lines for reaction to *Fusarium* head blight. p. 71–72. *In* P. Hart et al. (ed.) Proc. 1998 National FHB Forum. University Printing, East Lansing, MI.
- Gilchrist, L., C. Velazquez, and A. Mujeeb-Kazi. 1999. Resistance to *Fusarium* head blight in synthetic hexaploid wheats (2n=6x=42, AABBDD). p. 162–164. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Gocho, H., and T. Hirai. 1987. Varietal resistance to scab in barley. p. 625–630. *In* S. Yasuda and T. Konishi (ed.) Proc. Int. Barley Genet. Symp., 5th. Okayama, Japan. 6–8 Oct. 1986 Sanyo Press.
- Gooding, R.W., H.N. Lafever, K.G. Campbell, and L.D. Herald. 1997. Registration of 'Freedom' wheat. *Crop Sci.* 37:1007.
- Hanson, E.W., E.R. Ausemus, and E.C. Stakman. 1950. Varietal resistance of spring wheats to fusarial head blight. *Phytopathology* 40:902–914.
- Immer, F.R., and J.J. Christenson. 1943. Studies on the susceptibility of varieties and strains of barley to *Fusarium* and *Helminthosporium* kernel blight when tested under muslin tents or in nurseries. *J. Am. Soc. Agron.* 35:515–522.
- Jauhar, P.P., and T.S. Peterson. 1999. Alien gene transfer into durum wheat: cytological and molecular characterization of scab-resistant hybrid derivatives. p. 83. *In* *Agron. abstr.* ASA, Madison, WI.
- Jiang, G., Z. Wu, and D. Huang. 1994. Effects of recurrent selection for resistance to scab (*Gibberella zeae*) in wheat. *Euphytica* 72:107–113.
- Jin, Y., X. Zhang, R. Rudd, and J. Rudd. 1999. A point inoculation method for evaluating scab resistance in wheat. p. 128. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Kolb, F.L., G.H. Bai, G.J. Muehlbauer, J.A. Anderson, K.P. Smith, and G. Fedak. 2001. Host plant resistance genes for *Fusarium* head blight: Mapping and manipulation with molecular markers. *Crop Sci.* 41:xxx–xxx (this issue).
- Liu, Z.Z., 1984. Recent advances in research on wheat scab in China. p. 174–181. *In* *Wheats for more tropical environments*. CIMMYT, Mexico, D.F., Mexico.
- Liu, Z.Z. and Z.Y. Wang. 1990. Improved scab resistance in China: Sources of resistance and problems. p. 178–188. *In* D. A. Sanders (ed.) *Wheat for the nontraditional warm areas*. Proc. Int. Conf. CIMMYT, Mexico, D.F., Mexico.
- Martin, R.A., and H.W. Johnston. 1982. Effects and control of *Fusarium* diseases of cereal grains in the Atlantic Provinces. *Can J. Plant. Pathol.* 4:210–216.
- McKendry, A.L., J.E. Berg, D.N. Tague, and K.D. Kephart. 1995. Registration of 'Ernie' soft red winter wheat. *Crop Sci.* 35:1513.
- McKendry, A.L., K.S. Salzman, and S. Liu. 1999. Evaluation of Asian, Italian and Brazilian winter wheat germplasm for type II and III resistance to *Fusarium* head blight. p. 129–132. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- McMullen, M., R. Jones, and D. Gallenberg. 1997. Scab of wheat and barley: A re-emerging disease of devastating impact. *Plant Dis.* 81:1340–1348.
- McMullen, M.P., B. Schatz, R. Stover, and T. Gregoire. 1997. Studies of fungicide efficacy, application timing, and application technologies to reduce *Fusarium* head blight and deoxynivalenol. *Cereal Res. Commun.* 25:779–783.
- Mesterházy, Á. 1995. Types and components of resistance to *Fusarium* head blight of wheat. *Plant Breed.* 114:377–386.
- Mesterházy, Á. 1997. Methodology of resistance testing and breeding against *Fusarium* head blight in wheat and results of selection. *Cereal Res. Commun.* 25:631–637.
- Miedaner, T., 1997. Review: Breeding wheat and rye for resistance to *Fusarium* diseases. *Plant Breed.* 116:201–220.
- Murphy, J.P., R.A. Navarro, and S. Leath. 1999. Evaluation of diploid, tetraploid, hexaploid, and synthetic wheats for type II resistance to *Fusarium* head blight. p. 133–136. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- National Agricultural Statistics Service. 1993–1999. Annual reports: Minnesota agriculture statistics, Minnesota Office, St. Paul, MN, and North Dakota Agriculture Statistics, North Dakota Office, Fargo, ND. USDA, Washington, DC.
- Nisikado, Y. 1959. Studies on the wheat scab, caused by *Gibberella zeae* (Schw) Petch, and its control. *Berichted.* Ohara Inst. Landw. Biol., Okayama Univ. 11(2):1–165 (English summary p. 141–165.)
- Ohm, H.W. 1999. 'Winter nursery' for winter wheat. p. 181–182. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Parry D.W., P. Jenkinson, and L. McLeod. 1995. *Fusarium* ear blight (scab) in small grain cereals-A review. *Plant Pathol.* 44:207–238.
- Prom, L.K., B.J. Steffenson, B. Salas, T.G. Fetch Jr., and H.H. Casper. 1996. Evaluation of selected barley accessions for resistance to *Fusarium* head blight and deoxynivalenol concentration. p. 764–766. *In* G. Scoles and B. Rosnagel (ed.) Proceedings of the 5th International Oat Conference and the 7th International Barley Genetics Symposium. University Extension Press University of Saskatchewan, Saskatoon, SK, Canada.
- Rasmusson, D.C., R.D. Wilcoxson, R. Dill-Macky, E.L. Schiefelbein, and J.V. Wiersma. 1998. Registration of MNbrite barley. *Crop Sci.* 39:290.
- Scholz, U., B. Steffenson, C. Urrea, and R. Horsley. 1999. Evaluation of six-rowed spring barley accessions for resistance to *Fusarium* head blight. p. 137–139. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Schroeder, H.W., and J.J. Christensen. 1963. Factors affecting resistance of wheat to scab caused by *Gibberella zeae*. *Phytopathology* 53:831–838.

- Schwarz, P.B., S. Beattie, and H.H. Casper. 1995. The fate and development of naturally occurring Fusarium mycotoxins during malting and brewing. *J. Am. Soc. Brew. Chem.* 53:121–127.
- Schwarz, P.B., S. Beattie, and H.H. Casper. 1996. Relationship between Fusarium infestation of barley and the gushing potential of malt. *J. Inst. Brew.* 102:93–96.
- Singh, R.P., H. Ma, and S. Rajaram. 1995. Genetic analysis of resistance to scab in spring wheat cultivar Frontana. *Plant Dis.* 79:238–240.
- Snijders, C.H.A. 1990. Genetic variation for resistance to Fusarium head blight in bread wheat. *Euphytica* 50:171–179.
- Snijders, C.H.A., and E.A. van Eeuwijk. 1990. Genotype x strain interactions for resistance to Fusarium head blight caused by *F. culmorum* in winter wheat. *Theor. Appl. Genet.* 81:239–244.
- Stack, R.W., R.C. Froberg, and H. Casper. 1997. Reaction of spring wheats incorporating Sumai#3-derived resistance to inoculation with several *Fusarium* species. *Cereal Res. Comm.* 25:667–671.
- Takeda, K. 1990. Selection response and parent-offspring correlation of the resistance to Fusarium head blight in barley. *Res. Inst. Bioresources, Okayama Univ.* 40:100.
- Takeda, K., and H. Heta. 1989. Establishing the testing method and a search for resistant varieties to Fusarium head blight in barley. *Jap. J. Breed.* 39:203–216.
- van Eeuwijk, E.A., A. Mesterházy, Ch.I. Kling, P. Ruckenbauer, L. Saur, H. Bürstmayr, M. Lemmens, L.C.P. Keizer, N. Maurin, C.H.A. Snijders. 1995. Assessing non-specificity of resistance in wheat to head blight caused by inoculation with European strains of *Fusarium culmorum*, *F. graminearum* and *F. nivale* using a multiplicative model for interaction. *Theor. Appl. Genet.* 90:221–228.
- van Ginkel, M., W. Van der Schaar, Y. Zhuping, and S. Rajaram. 1996. Inheritance of resistance to scab in two wheat cultivars from Brazil and China. *Plant Dis.* 80:863–967.
- Vivar, H., L. Gilchrist, L. Marquez, C. Velazquez, and P. Hayes. 1999. Using morphological markers in breeding barley for Fusarium resistance. p. 16–18. *In* P. Bregitzer (ed.) Proc. American Barley Researchers Workshop, 16th. USDA-ARS National Small Grains Research Facility, Aberdeen, ID.
- Wagester, J., R. Ward, L.P. Hart, S.P. Hazen, J. Lewis, H. Borden (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Waldron, B.L., B. Moreno-Sevilla, J.A. Anderson, R.W. Stack, and R.C. Froberg. 1999. RFLP mapping of QTL for Fusarium head blight resistance in wheat. *Crop Sci.* 39:805–811.
- Wan, Yong-Fang, Chi Yen, Jun-Liang Yang, and Deng-Cai Lin. 1997. The diversity of resources resistant to scab in *Triticeae* (*Poaceae*). Wheat Information Service 84:7–12.
- Wilcoxson, R.D. 1993. Historical overview of scab research, p. 1–5. *In* Proc. (1st) Regional Scab Forum, Moorhead, MN. Publ. Minn. Wheat Res. & PromCouncil, Red Lake Falls, MN.
- Wilcoxson, R.D., T. Kommedahl, E.A. Oxmon, and C.E. Windels. 1988. Occurrence of Fusarium species in scabby wheat from Minnesota and their pathogenicity to wheat. *Phytopathology* 78:586–589.
- Windels, C.E. 2000. Economic and social impacts of Fusarium head blight: Changing farms and rural communities in the northern great plains. *Phytopathology* 90:17–21.
- Windels, C.E., and T. Kommedahl. 1984. Late season colonization and survival of *Fusarium graminearum* group II in corn stalks in Minnesota. *Plant Dis.* 68:791–793.
- Wood, M., D. Comis, D. Harden, L. McGraw, and K.B. Stelljes. 1999. Fighting Fusarium. Agricultural Research. June issue. USDA-ARS, Beltsville, MD.
- Xihang, Z., C. Mingsheng, and L. Xunyi. 1991. Screening and testing of barley varieties for scab resistance. *Acta Phytopythologica Sinica* 18(3):265.
- Zhang, X., Y. Jin, R. Rudd, J. Rudd, and H. Bockelman. 1999. Screening of spring wheat scab resistance from the USDA germplasm collection. p. 140–142. *In* J. Wagester et al. (ed.) Proc. 1999 National FHB forum. University Printing, East Lansing, MI.
- Zhou, C.R., S.S. Xua, C.M. Qian, G.C. Yao, and J.X. Shen. 1987. On the problem of breeding wheat for scab resistance. *Scientia Agri. Sinica.* 20:19–25.
- Zhu, H., L. Gilchrist, P. Hayes, A. Kleinhofs, D. Kudrna, Z. Liu, L. Prom, B. Steffenson, T. Toojinda and H. Vivar. 1999. Does function follow form? Principal QTLs for Fusarium head blight (FHB) resistance are coincident with QTLs for inflorescence traits and plant height in a doubled haploid population of barley. *Theor. Appl. Genet.* 99:1221–1232.
- Zhuang, Q.S., and Z.S. Li. 1993. Present status of wheat breeding and related genetic study in China. *Wheat Inf. Serv.* 76:1–15.