# New Fusarium Head Blight Resistant Spring Wheat Germplasm Identified in the USDA National Small Grains Collection

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#### **ABSTRACT**

Fusarium head blight (FHB; caused by Fusarium graminearum Schwabe) is one of the most destructive wheat (Triticum aestivum L.) diseases worldwide. Sources of FHB resistance are limited. The objectives of this study were to screen selected spring wheat accessions in the USDA National Small Grains Collection for FHB reactions using FHB index, visual scabby kernel (VSK), and deoxynivalenol (DON) content. A total of 1035 spring wheat accessions were initially screened in unreplicated field evaluation nurseries in 1998 and 1999. Accessions with low FHB were selected as putative resistant materials and were tested in replicated trials from 1999 to 2002. After three or more years of evaluation, 73 accessions with resistance were identified, including 10 accessions previously reported as resistant to FHB. Selections from Europe had the highest percentage of resistance to VSK, followed by selections from South America and Asia. We concluded that there is diversity for FHB resistance in wheat. Fusarium head blight resistance identified from Europe appeared to be unique in that these accessions normally displayed a moderate level of disease in the field, but a higher level of resistance based on VSK and DON. The discovery of diverse resistant sources will provide diversity so that higher levels of resistance could be developed. The novelty and types of FHB resistance in these selections should be further characterized using molecular markers and different inoculation techniques.

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**Abbreviations:** DON, deoxynivalenol; FHB, Fusarium head blight; MR, moderately resistant; MS, moderately susceptible; NSGC, National Small Grains Collection; PDA, potato dextrose agar; R, resistant; S, susceptible; VR, very resistant; VSK, visual scabby kernel.

 $\Gamma_{graminearum}^{USARIUM\ HEAD\ BLIGHT\ (FHB)}$ , primarily caused by Fusarium graminearum Schwabe in North America, is a devastating disease throughout the spring wheat and eastern soft winter wheat (Triticum aestivum L.) production regions in the United States. Losses due to FHB in wheat include reduced yields, discolored and shriveled "tombstone" kernels, contamination of the infected grains with mycotoxins, and seedling blight when infected and infested grain is used as seed (McMullen et al., 1997; Schroeder and Christensen, 1963). The use of host resistance is the most economically and environmentally sound solution to this problem. The reaction to inoculation of wheat by Fusarium species capable of inciting FHB is complex. Different types or components of disease measurements have been used to reflect the expressions or manifestations of FHB resistance (Snijders, 1990; Stack, 2000). Two types of resistance were described by Schroeder and Christensen (1963): Type 1 (resistance to initial infection) and Type 2 (resistance to spread of the pathogen within wheat spike) and have been widely used (Bushnell, 2002). So far only Type 2 resistance can be directly measured, as the spread of blight after

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inoculating a single floret around the center of a wheat spike. Other types or components of FHB resistance have been proposed including resistance or tolerance to FHB expressed on developing kernels (Mesterhazy, 1995) and reduced accumulation of mycotoxins (Miller et al., 1985; Snijders and Perkowski, 1990). Despite the disagreement among researchers on the definition and measurements of the latter types and components of resistance mechanisms (Bushnell, 2002; Shaner, 2002), different components of resistance should be considered to thoroughly evaluate the FHB resistance of a given line. Besides the complexity of resistance mechanisms, significant environment effects on FHB development require screening over several environments to characterize the true FHB resistance potential of a particular genotype (Fuentes et al., 2005).

A few sources of resistance to FHB have been identified in wheat previously. Reported sources of FHB resistance in spring wheat include a few landraces; 'Sumai 3' and its derivatives from China; 'Nobeoka Bozu' and 'Sin Chunaga' and its relatives from Japan; and 'Frontana' and 'Encruzilhada' from Brazil (Ban, 2000; Ban and Suenaga, 2000; Liu and Wang, 1991; Mesterhazy, 1987; Schroeder and Christensen, 1963; Yu et al., 2006). Winter wheat cultivars Arina, Renan, and Praag-8 with FHB resistance were reported in European wheat germplasm (Gervais et al., 2003; Ruckenbauer et al., 2001; Snijders, 1990). Fusarium head blight resistance in 'Ernie' and '2375' may not be related to the aforementioned sources of resistance (Rudd et al., 2001), likely representing resistance from native U.S. germplasm. Novel FHB resistance was also postulated to be present in several recently released cultivars, including Truman (McKendry et al., 2005), Steele-ND (Mergoum et al., 2005), and Glenn (Mergoum et al., 2006). Sumai 3, including its derived lines, is the most widely used source of FHB resistance (Rudd et al., 2001). The resistance in Sumai 3 is partial, and cultivars with this resistance can sustain substantial damage from FHB when environmental conditions are conducive to the disease. Identification and characterization of additional sources of resistance is important for enhancing the level of resistance and for introducing genetic variation to the breeding materials.

The National Small Grains Collection (NSGC) maintains a large collection of common wheat and represents gene pools from all major wheat growing regions of the world. This worldwide collection of germplasm is a valuable gene pool for genetic improvement of modern wheat cultivars. The FHB resistance in this collection has not been assessed systematically. The objectives of this research were to assess the FHB reaction in selected spring wheat accessions in the NSGC and to identify and characterize FHB resistant germplasm based on measurements of FHB index, visual scabby kernel (VSK), and deoxynivalenol (DON) concentration.

#### MATERIALS AND METHODS

#### **Plant Materials**

In 1998, 424 accessions of spring wheat from the NSGC were evaluated for FHB reaction. The accessions were selected from regions where FHB resistance is known to be present (Table 1). In 1999, an additional 999 NSGC accessions were evaluated for FHB with an emphasis on germplasm from southern Europe (Table 1). Spring wheat cultivars BacUp (PI 596533) and ND2710 (PI 633976) were used as resistant checks. ND2710, like its parent Sumai 3, is highly resistant to FHB based on disease incidence, severity, VSK, and reduced levels of DON (Frohberg et al., 2004). BacUp has been a good FHB resistance check because of its high level of FHB resistance (Fuentes et al., 2005). ND 2710 was later maturing than BacUp. 'Sonalika' (CItr 15392) and 'Wheaton' (PI 469271) were used as susceptible checks. Sonalika has the earliest maturity among the checks and is highly susceptible to FHB. Wheaton is an intermediate maturing spring wheat cultivar and has been routinely used as one of the most susceptible checks in FHB studies.

# FHB Screening Nursery Design Nonreplicated FHB Evaluation Nursery

The field screening nurseries were conducted between 1998 and 2002 in Brookings, SD. The nursery was planted into a field with a soybean [Glycine max (L.) Merr.] crop in the previous season and fertilizer was not used. Accessions from the NSGC were planted into nonreplicated single 1-m rows spaced 30 cm apart using a plot planter. The four check cultivars were planted next to each other after every 35 test entries. A postemergence herbicide was applied when wheat was between three- and four-leaf stage. Preliminary selections were made by accession (row) or heads within a row having low FHB indices or low kernel damage. Seed of the selections was hand harvested and increased in an off-season nursery in Christchurch, New Zealand. The crop season in the New Zealand nursery was from October to early April. Thus, we were able to use seed harvested from the New Zealand nursery for FHB nursery planting in late April to early May in Brookings, SD.

#### Replicated FHB Evaluation Nursery

Heading date data collected from the nonreplicated screening nursery were used to group the FHB resistant selections into three maturity groups: early, intermediate, and late. The replicated FHB evaluation nursery was planted and managed in the same manner as described above. Maturity groups were planted into separate blocks and treated as separate experiments for nursery management and data analysis. Fusarium head blight resistant selections were planted with three replications in a randomized complete block design, and evaluated for at least three consecutive years. Selections initially made from the 1998 and 1999 nonreplicated FHB evaluation nursery were planted in replicated trials for four years (1999–2002) and three years (2000–2002), respectively.

### **Inoculum Preparations**

To generate consistent and maximum disease pressure in the field, corn (*Zea mays* L.) kernels colonized with *F. graminearum* and conidial suspension of the fungus were applied in the FHB screening nurseries. The macroconidial inoculum consisted of

Table 1. Origin and number of accessions of spring wheat germplasm from the National Small Grains Collection evaluated for Fusarium head blight resistance (FHB) in nonreplicated evaluation nurseries in 1998 and 1999, number of accessions identified with FHB resistance based on replicated evaluation nurseries between 1999 and 2002, and geographical distribution of FHB resistance selections based on measurements of FHB index, percentage visual scab kernel (VSK), and deoxynivalenol (DON) concentration.

	No. acc	essions	No.			N	o. Acc	essic	ons in each FHB measurement‡ group								
Origin <sup>†</sup>	evalı	uated	accessions with FHB			Index					VSK				D	ON	
	1998	1999	resistance	VR	R	MR	MS	S	VR	R	MR	MS	S	VR	R	MR	MS
Asia	131	26	15	2	6	6	1		3	1	1	7	3	2	5	7	1
China	66	15	3		1	1	1			1	1	1			1	2	
Taiwan		8	0														
Japan	65	3	12	2	5	5			3			6	3	2	4	5	1
Europe	93	498	26		1	12	11	2	4	9	10	3			7	16	3
Austria		93	0														
Belarus		2	0														
Bosnia and Herzegovina		11	0														
Bulgaria		8	1			1			1							1	
Croatia		4	0														
Czech Republic		7	0														
Czechoslovakia		12	0														
Greece		20	3			3				1	2				2	1	
Hungary		25	3			1	2			1	2					3	
Italy	93	7	5		1	1	3		2		1	2				3	2
Macedonia		7	0														
Poland		26	1			1				1							1
Romania		23	3			1	2			2	1					3	
Slovakia		1	0														
Slovenia		1	0														
Switzerland		198	5			2	3		1	2	1	1			3	2	
Ukraine		15	3			1		2		1	2				2	1	
Yugoslavia		38	2			1	1			1	1					2	
South America	164	123	32	1	6	13	10	2	2	8	15	7		3	8	16	5
Argentina		89	15		2	5	8		1	6	7	1			5	9	1
Brazil	164	11	12	1	1	7	1	2	1	2	4	5		3	1	4	4
Paraguay		4	1				1					1			1		
Uruguay		19	4		3	1					4				1	3	
Total	388	647	73	3	13	31	22	4	9	18	26	17	3	5	20	39	9

†Data source: National Plant Germplasm System (http://www.ars-grin.gov/npgs/acc/acc\_queries.html).

10 F. graminearum isolates isolated from infected wheat spikes collected in several wheat fields from eastern South Dakota in 1996 and 1997. These isolates, each derived from a single-spore culture, were selected from a collection of 30 isolates based on two criteria: producing abundant macroconidia when cultured on acidic potato dextrose agar (PDA) in petri plates and producing typical symptoms on the susceptible check cultivar Wheaton. After incubation for 7 d, agar of well-colonized plates was cut into 1-cm<sup>2</sup> cubes and dried under a flow hood for 72 h. The dried fragments of cultures were stored in a sterile glass vial at -80°C as stock cultures. This method of culture maintenance reduces the need for frequent transfers. For mass production of conidia, pieces of the dried stock culture were plated onto modified acidic PDA medium (19.5 g potato dextrose, 10 g agar, 1 L water, 2.0 mL lactic acid after autoclaving). After 5 to 7 d of incubation at 23°C, 2 mL of sterile water was added to each

culture plate and macroconidia and mycelial fragments were suspended using a hockey stick-shaped sterile glass rod. The culture suspension was transferred to new plates by streaking the suspension evenly onto new plates. Compared to transferring agar blocks, we observed that streaking suspension to cover the entire agar surface appeared to promote the production of macroconidia while suppressing mycelium growth. Macroconidia were harvested after 3 to 5 d incubation by flooding the plates with sterile water and gently rubbing culture surface with a sterile glass rod. The spore suspension was poured off and strained through two layers of sterile cheesecloth to remove hyphal fragments. Macroconidia could be harvested for a second time after an additional incubation period of 3 to 5 d after the first harvest as long as a strict sterile procedure is followed. Spore suspensions for equal numbers of petri plates of 10 isolates were mixed and adjusted to 70,000 conidia mL<sup>-1</sup>. The macroconidial inoculum

<sup>&</sup>lt;sup>‡</sup>VR, very resistant; R, resistant; MR, moderately resistant; MS, moderately susceptible; S; susceptible.

suspension was prepared 2 to 4 h before field inoculation and maintained in an ice tray until use.

Stainless steel trays (53 cm long by 33 cm wide by 7 cm high) and 4.5-L milk jars were used to increase F. graminearum on corn kernels. About one-half (2.5-3.8 cm) of the stainless steel tray was filled with red or yellow dent corn. Then water was added until about three-quarters of the tray was filled with water and corn. The grain was soaked in the water for 18 to 24 h and excess water was drained. After washing the soaked corn with clean water, water was added until it was about 0.32 to 0.64 cm above the corn surface. The tray was covered with two layers of aluminum foil and autoclaved for 2 h. The tray was cooled at room temperature for about 5 h, drained if excessive water was present, and inoculated with two petri plates of a single isolate of F. graminearum. Then the tray covered with two layers of aluminum foil was incubated on a laboratory bench at 20 to 22°C. The tray was shaken vigorously every 24 h. Seven to ten days after inoculation, the corn kernels were fully colonized. The colonized corn kernels were dried on a greenhouse bench, and stored at 4°C for up to a month before field application. To increase colonized corn kernels in the milk jars, about one-third of the jar was filled with corn. After being soaked in water for 18 to 24 h, the corn was washed clean, and excess water was drained. Fresh water was added until about 0.32 to 0.64 cm above the corn surface. The milk jar was sealed with a cotton plug and autoclaved for 45 min. After the jar was cooled, one petri dish of F. graminearum culture was added into the jar and mixed evenly with the corn in a flow hood. The sealed milk jar was incubated at room temperature, and shaken every 24 h. Seven days after incubation in the milk jar, the corn was fully colonized and ready to be dried or used immediately in the field. Equal amount of F. graminearum colonized corn grains (corn spawn) measured by weight of each isolate was mixed and used for field inoculation.

#### **Nursery Inoculation**

Beginning at the jointing stage of the earliest maturing test entries, each row was inoculated with 7 to 8 g of corn spawn weekly for three consecutive weeks. To promote production of ascospores, the plots were daily mist-irrigated for 3 min with a 30-min recess between 2000 and 0800 hours. On hot and windy days, irrigation was extended to 5 min with a 30-min recess. Mist-irrigation was continued until the last disease recording. At the heading stage, each plot was marked with colored plastic tape so that the plot could be identified for inoculation and disease scoring at the appropriate time. The field was tagged three times a week. At the heading stage, each plot was spray-inoculated with approximately 40 mL of macroconidial suspension using a backpack sprayer. A second spray inoculation was applied 5 d later. All spray inoculations were conducted after 1900 hours when the environment was conducive for infection.

#### FHB Evaluation and Selection

Disease severity of each plot was rated by evaluating 20 randomly selected spikes 3 wk after the first spray inoculation. Fusarium head blight incidence was recorded as percentage of infected spikes of the 20 spikes used for the FHB severity assessment (i.e., incidence = 100 × number of nonzero spikes/20). The FHB index of each plot was derived by the product of average FHB severity and FHB incidence multiplied by 100. A preliminary selection for reduced kernel damage was made

in the nonreplicated FHB evaluation nursery by tactile estimation of kernel filling, independent of FHB index. At the hard dough stage of plant development, each plot was assessed for kernel damage by repeatedly squeezing several handfuls of spikes throughout the plot. Because little kernel filling occurred in susceptible lines due to high and consistent disease pressure in the nursery, a seed set of moderate level could be identified regardless of visual disease symptoms. This method was particularly useful for identifying plants that were resistant to kernel damage but appeared to have high FHB index. Individual plots were also carefully inspected visually to identify individual plants or spikes that might be showing resistance among susceptible plants.

Entries or individual heads within an entry having a low FHB index and/or good seed set in the nonreplicated FHB evaluation nursery were harvested by hand. Selections of entire rows (row selections) were first threshed by a plot combine at minimum air pressure, then threshed and cleaned with a single head thresher at minimum air pressure. Heads selected within an entry (head selections) were bulked and threshed with a single head thresher at minimum air pressure. Single plants of low disease within an entry were selected as single plant selections. Percentage of VSK of each selection was recorded. Accessions with high VSK were discarded.

The replicated FHB evaluation nursery was hand harvested and immediately threshed with a plot combine at minimum air pressure in the field. Because a high proportion of diseased spikelets with chaffs were often retained in the initial threshing with the combine at a low air pressure, the field-threshed materials were threshed two to three times again at low air pressure using a single plant thresher to remove chaffs. Percentage of VSK of each entry was determined by counting 200 random seeds from each sample. A 15-g seed sample from each plot was analyzed for DON concentration. In 2000, the seed samples of some selections were tested by Dr. Paul Schwarz (Dep. of Plant Sciences, North Dakota State University, Fargo, ND). In 2001 and 2002, the seed samples were tested for DON concentration by Ms. Beth Tacke (Dep. of Veterinary Diagnostic Services, North Dakota State University, Fargo, ND). Each year, the lines with high FHB index and high DON levels were discarded. Final selections from the 2002 replicated FHB evaluation nursery constituted the final list of FHB resistant germplasm.

#### **Data Presentation and Analysis**

To simplify interpretation of the data, FHB index, VSK, and DON of the resistant selections were classified into five groups using the criteria presented in Table 2. The criteria for the classification were based on the mean values and ranges of the four standard checks. Under moderate to low disease pressure, FHB levels of ND 2710 and BacUp are similar. Our nursery disease pressure was consistently high enough to differentiate the FHB levels between ND 2710 and BacUp. Therefore, accessions with less FHB than ND 2710 were grouped into the very resistant (VR) category. Measurements of FHB index, VSK, and DON of the test entries in each year were analyzed using GLM procedures of SAS 9.1 (SAS Institute, Inc., Cary, NC) by maturity group. To detect effects of year and year\*entry in each maturity group, 3-yr data of the entries in the 2002 replicated screening nursery were used for analysis of variance.

### **RESULTS**

### **Nonreplicated FHB Evaluation Nurseries**

Disease pressure in the nonreplicated screening nurseries was very high and uniform in both years. In 1998, FHB incidence ranged from 80 to 100% and 91% of the plots had 100% FHB incidence. Field visual FHB reactions could be best estimated by FHB index because it is the product of severity and incidence (Wilcoxson et al., 1992). Average FHB index of ND 2710 was 26%, of BacUp was 35%, of Sonalika was 90%, and of Wheaton was 80%. In 1999, disease incidence in the nonreplicated nursery ranged from 83 to 100% with 94% of the plots having 100% FHB incidence. In 1999, average FHB index of ND 2710 was 22%, of BacUp was 24%, of Sonalika was 83%, and of Wheaton was 77%.

In 1998, 36 of the 424 accessions failed to germinate and provide at least five plants for FHB evaluation. In 1999, 352 of the 999 accessions failed to germinate or produced less than five plants for FHB evaluation. The complete dataset of the FHB index of the remaining 388 accessions evaluated in 1998, and 647 accessions evaluated in 1999 were submitted to the Germplasm Resources Information Net-(http://www.ars-grin.gov/cgi-bin/npgs/html/desc. pl?65066). Those accessions displayed a wide range of FHB disease indices from highly resistant to highly susceptible (Fig. 1). In 1998, 11 accessions had FHB indices of 0 to 30%, and 156 accessions had disease indices more than 70%. In 1999, seven accessions had FHB indices of 0 to 30%, and 402 accessions had disease indices more than 70%. Accessions with low FHB index or good seed set were selected for evaluation of FHB reaction in the replicated trials in the following year. Based on the FHB index and our visual selection for seed set,

48 accessions were selected from the 1998 nonreplicated FHB evaluation nursery and 123 accessions from the 1999 nonreplicated evaluation nursery as putative sources of FHB resistant germplasm. We observed heterogeneous reactions to FHB in some of the accessions. The 1998 selections consisted of 32 head- or single-plant selections and the remainder was whole row selections. The 1999 selections consisted of head-selections from 78 accessions and the remainder was whole row selections. Those selections were planted in the replicated screening nursery in the following year.

## FHB-Resistant Accessions Selected through Replicated FHB Evaluation Nursery

After two years of evaluation for the 1999 selections and three years evaluation for the 1998 selections, the 2002 replicated FHB nursery consisted of 94 accessions

Table 2. Assignment of categories of wheat germplasm for Fusarium head blight (FHB) reaction based on FHB index, percentage visual scab kernel (VSK), and deoxynivalenol (DON) evaluated between 1999 and 2002 in Brookings, SD.

FHB reaction	Range	FHB index <sup>†</sup>	VSK‡	DON§
				μg g <sup>-1</sup>
Very resistant (VR)	Mean ≤ ND 2710	0–14	0-27	0-5.1
Resistant (R)	Mean > ND 2710 and ≤ BacUp	15–28	28–33	5.2-8.1
Moderately resistant (MR)	·	29-45	34-45	8.2-12.5
Moderately susceptible (MS)		46-60	46-60	12.6-16.0
Susceptible (S)		>60	>60	>16.0

 $^\dagger$ Mean FHB index of 1999–2002 or 2000–2002 on 20 spikes with three replicated plots per year.  $^\dagger$ Mean VSK of 1999–2002 or 2000–2002 on seed harvested from the whole plot with three

§Mean DON of 2000–2002 of seed samples from one replicate in 2000, two replications in 2001, and three replications in 2002.

with 29 in the early maturity group, 37 in the intermediate maturity group, and 28 in the late maturity group. Analysis of variance of FHB index and VSK of each maturity group over years using the entries in the 2002 replicated FHB nursery revealed significant effects of accession, and accession  $\times$  year interaction (P < 0.001) in each maturity group. The replicate within year effect was not significant, indicating that each year disease pressure was uniform across the screening field.

The susceptible checks Sonalika and Wheaton had the highest FHB index, VSK, and DON (Table 3). ND 2710 was more resistant than BacUp across the maturity groups and years (Table 3). ND 2710 had lower FHB indices and DON than BacUp in every maturity group and year, and lower VSK than BacUp in all the years except in 2002.

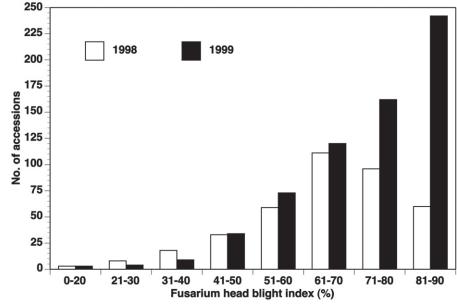


Figure 1. Distribution of Fusarium head blight (FHB) index of spring wheat germplasm in the National Small Grains Collection tested in unreplicated FHB evaluation nursery in 1998 and 1999. Fusarium head blight index = average FHB severity of 20 heads  $\times$  FHB incidence  $\times$  100.

Table 3. Means (over three replicates) of Fusarium head blight (FHB) index, percentage visual scabby kernel (VSK), deoxynivalenol (DON) concentration, plant height, and days to heading of FHB resistant germplasm evaluated in field FHB screening nursery in Brookings, SD, in 2000–2002.

Accession		Inc	dex			V	SK			D	ON	Plant	Days to	
Accession	2000	2001	2002	Mean	2000	2001	2002	Mean	2000†	2001‡	2002§	Mean	height¶	heading
				9	%					µg	g <sup>-1</sup>		cm	
Early maturit	•													
ND2710	13	10	15	13	20	12	43	25	5.4	2.3	5.4	4.4	106	56
BacUp	23	23	28	25	30	17	42	30	7.7	9.0	7.5	8.1	90	54
Sonalika	87	86	86	86	80	57	87	75	37.0	26.1	25.4	29.5	76	53
Wheaton	87	90	90	89	95	58	80	78	24.3	18.0	14.9	19.1	74	59
PI 382161	10	6	10	9	22	12	33	22	1.9	2.0	2.3	2.1	107	57
PI 382140	15	18	18	17	38	47	57	47	3.3	4.2	3.7	3.7	91	57
PI 182568	22	16	28	22	47	30	70	49	7.8	5.7	6.3	6.6	108	57
Cltr 12002	26	21	22	23	42	37	47	42		9.3	5.6	7.4	112	57
PI 519790	20	29	25	25	40	35	57	44		8.5	10.8	9.7	103	55
PI 345731	27	19	30	25	17	23	45	28	12.0	6.3	6.6	8.3	93	53
PI 434987	22	23	31	26	57	16	63	45	9.4	10.7	11.3	10.5	105	56
PI 182586	30	21	27	26	50	57	70	59	14.9	7.8	8.0	10.2	79	56
PI 411132	28	24	34	29	77	67	73	72	9.4	11.8	7.2	9.5	64	52
PI 182591	37	29	31	32	67	41	63	57	8.2	6.0	4.2	6.1	70	56
Cltr 12021	32	32	34	33	42	25	53	40		8.4	13.0	10.7	108	57
PI 163429	27	32	40	33	30	29	40	33	12.0	7.9	6.9	8.9	106	56
PI 351743	45	65	22	44	28	30	33	31	9.1		7.8	8.5	122	56
PI 163439	59	25	47	44	40	28	50	39	0.1	9.0	10.2	9.6	104	57
PI 185383	42	43	51	45	50	30	33	38		10.2	5.3	7.7	101	53
PI 584926	43	56	37	45	43	33	70	49		14.0	14.9	14.5	89	57
	43 57	23		45	43 25	33 24	63	37		9.4	11.1		110	57 55
Cltr 2492 Pl 168716		23 53	56 32	45 47		31	23	30	13.0		9.1	10.2	109	55 57
	55				35				13.0	15.0		12.4		
PI 344467	48	51	49	49	38	32	50	40	40.4	5.2	7.8	6.5	105	55
PI 344454	50	82	30	54	28	30	33	31	13.1	15.2	7.1	11.8	108	55
PI 104138	59	53	52	55	57	40	50	49		9.7	14.9	12.3	106	56
PI 184512	77	38	53	56	33	19	33	29	18.5	11.1	12.2	13.9	115	56
PI 192498	55	57	62	58	50	42	43	45			10.9	10.9	103	55
PI 352062	63	59	57	59	50	37	43	43			7.1	7.1	94	57
Mean	38	34	35	36	40	32	49	40	9.3	8.4	8.0	8.6	101	56
LSD				16.5				15.1				4.6		
Intermediate	maturity	′												
ND2710	13	13	13	13	18	17	43	26	5.4	5.2	4.6	5.1	111	56
BacUp	22	17	30	23	25	30	40	32	7.7	10.6	6.0	8.1	89	54
Sonalika	87	86	86	86	80	57	87	75	37.0	26.1	25.4	29.5	76	53
Wheaton	87	90	90	89	95	58	80	78	24.3	18.0	14.9	19.1	74	59
PI 382154	14	9	12	12	15	15	33	21	1.5	2.8	2.2	2.2	111	58
PI 382153	17	10	12	13	13	16	33	21	4.8	3.5	1.7	3.3	102	58
PI 182561	20	22	22	21	85	77	77	79	15.5	13.2	6.7	11.8	94	58
PI 462151	19	30	19	22	22	20	53	32	4.2	7.3	5.8	5.8	101	59
PI 182583	19	40	23	27	82	75	80	79	7.1		8.1	7.6	73	61
PI 285933	36	31	22	30	27	22	43	31	21.8	11.7	5.8	13.1	125	61
PI 351816	32	29	33	31	70	33	70	58	9.0	7.8	7.6	8.1	88	58
PI 351256	21	36	38	32	42	19	77	46	7.6	8.4	8.0	8.0	94	59
PI 351221	34	35	30	33	20	20	40	27		9.9	5.6	7.8	119	60
Cltr 13136	43	35	32	37	50	24	53	42	18.4	6.7	9.4	11.5	108	59
PI 104131	36	34	32 45	38	22	24 14	33	23	7.6	3.9	9.4 7.4	6.3	121	59 58
PI 192634	54	23	49	42	42	35	53	43	11.4	7.6	8.0	9.0	109	58

Table 3. Continued.

		Inc	dex			VS	SK			DON			Plant	Days to
Accession	2000	2001	2002	Mean	2000	2001	2002	Mean	2000†	2001‡	2002§	Mean	height <sup>¶</sup>	heading
				9	6					μg	g <sup>-1</sup>		cm	
PI 185843	58	40	30	43	40	23	37	33	13.9	7.6	7.4	9.7	114	60
PI 182565	32	57	39	43	53	53	73	60	15.8		8.7	12.2	106	60
PI 233207	53	49	28	44	33	53	33	40			9.9	11.9	109	58
PI 264998	44	48	41	44	30	27	53	37		8.2	7.8	8.0	120	61
PI 264940	55	48	30	44	42	16	57	38		9.3	9.9	9.6	98	59
PI 168727	37	28	69	45	25	20	57	34	20.0	5.7	7.6	11.1	95	58
PI 256958	42	59	46	49	20	27	53	33	11.7	11.9	8.9	10.8	119	60
PI 132856	48	42	60	50	37	30	70	45		12.0	14.1	13.1	105	61
PI 351993	54	57	39	50	30	25	53	36		6.8	6.2	6.5	120	61
PI 351748	44	69	39	51	30	43	53	42		10.2	8.2	9.2	117	60
PI 264946	50	35	67	51	53	38	73	55		10.2	7.7	9.0	101	60
PI 349534	54	67	34	52	27	20	43	30	11.6	11.9	9.6	11.0	118	61
PI 272348	40	61	58	53	27	63	40	43			9.2	9.2	110	60
PI 344465	48	63	61	57	37	30	50	39		7.8	4.3	6.0	97	58
PI 203083	63	54	59	59	33	65	57	52			7.4	7.4	116	61
PI 584934	63	69	58	63	42	32	50	41		11.8	9.1	10.5	99	58
Mean	42	45	41	43	40	35	55	43	12.5	9.0	8.0	9.4	107	59
LSD				16.5				17.7				5.6		
Late maturity	,													
ND2710	15	11	15	14	23	15	43	27	5.4	3.6	4.6	4.5	107	56
BacUp	31	22	30	28	30	28	40	33	7.7	11.3	5.3	8.1	91	54
Sonalika	85	87	88	87	77	93	87	85	37.0	39.0	18.8	31.6	80	53
Wheaton	88	90	84	87	92	100	90	94	24.3	25.2	13.8	21.1	79	59
Cltr 5103	14	27	35	26	19	23	43	29	16.5	7.2	6.2	9.9	122	62
PI 81791	24	40	16	27	22	18	40	26	18.0	12.6	11.8	14.1	127	64
PI 192660	39	20	24	28	23	19	33	25	16.4	5.2	9.8	10.5	123	63
PI 185380	36	25	26	29	30	16	33	26	11.2	5.1	8.6	8.3	119	62
PI 382167	11	29	50	30	23	18	57	33	5.5	4.3	5.4	5.1	119	67
PI 382144	30	37	36	34	45	33	63	47		8.0	8.2	8.1	111	64
PI 294975	25	67	17	36	20	20	27	22	11.4	8.4	9.5	9.8	125	62
PI 264927	32	48	30	37	17	21	50	29	5.6	3.6	6.3	5.2	117	62
PI 83729	51	50	18	40	40	23	33	32	10.5	7.5	7.5	8.5	121	62
Cltr 17427	35	44	45	41	33	24	67	41	19.4		9.4	14.4	111	59
PI 360869	40	41	46	42	40	63	70	58			9.7	9.7	101	66
PI 362437	36	52	43	44	33	22	47	34	8.9	7.3	10.2	8.8	123	64
PI 214392	16	82	65	54	30	55	57	47	16.3		12.9	14.6	113	68
PI 351476	56	75	35	56	25	40	33	33	13.4		8.1	10.7	122	63
PI 192219	49	79	44	58	37	25	47	36		8.7	8.3	8.5	114	70
PI 57364	45	87	44	59	55	53	43	51			10.6	10.6	119	65
PI 213833	45	65	68	59	63	50	60	58	13.5	22.1	7.1	14.2	95	64
Oltr 11215	40	84	54	59	35	29	27	30	9.2	10.5	5.2	8.3	116	66
PI 113949	63	71	51	62	38	25	33	32		7.4	6.8	7.1	118	63
PI 113948	78	88	62	76	30	43	47	40	7.1		7.4	7.2	118	62
Cltr 12470	74	75	82	77	45	57	70	57	21.9	7.9	12.8	14.2	114	63
Mean	40	57	42	46	34	32	47	37	12.8	8.4	8.7	9.9	117	64
LSD	-			21.8	- •			15.7				7.5	**	

 $<sup>^{\</sup>dagger}\text{DON}$  concentration of the first replication.

<sup>&</sup>lt;sup>‡</sup>Mean DON concentration of the first and second replications.

<sup>§</sup>Mean DON concentration of three replications.

<sup>¶</sup>Plant height in the 2001 FHB nursery.

During the 2002 FHB inoculation period daily temperatures were above normal and most of the days were windy. ND 2710 had very high percentage of discolored and plump kernels. The discoloration might have resulted from heat stress instead of direct damage by FHB. The VSK ratings in 2002 could be partly inflated due to heat stress in the disease nursery as indicated by a higher mean VSK in 2002 in each maturity group than the means in the other years (Table 3). This hypothesis is supported by the similar DON concentration of ND 2710 and many resistance selections in 2002 as in 2000 and 2001. Seventythree accessions were selected as putative sources of FHB resistance after evaluations in replicated field trials in three to four consecutive years. Among these, FHB resistance in 10 accessions has been reported in previous studies. The 10 accessions of known FHB resistance were CItr 12470 (Frontana), PI 113948 (Kooperatorka), PI 132856 (Mentana), PI 182561 (Sin Chunaga), PI 182591 (Norin 61), PI 213833 (Funo), PI 382144 (Encruzilhada), PI 382153 (Nobeoka Bozu), PI 382154 (Nyu Bai), and PI 462151 (Shu Chou Wheat No. 3) (Tables 3 and 4) (Ban, 2000; Ban and Suenaga, 2000; Liu and Wang, 1991; Snijders, 1990). Because FHB resistance was first determined by disease index in the field, we ranked the resistance selections by FHB indices in each maturity group in Table 3. Most of the resistant selections had much lower FHB index, VSK frequency, and DON content than the susceptible checks Sonalika and Wheaton except Kooperatorka (late maturity) and two well-known FHB resistant lines, Frontana (late maturity) and Sin Chunaga (intermediate maturity). Thirty-six selections were head selections. The seed of the head selections were deposited in the NSGC with new accession numbers. For simplicity, we used the accession number and name of the original seed source throughout the report. Detailed selection type and new accession numbers for the head selections are presented in Table 4, including the origin, pedigree of the original seed source, maturity group, and the FHB reactions of the resistant selections.

#### FHB Resistance Measured by FHB Index

Mean FHB index of the 73 resistant selections ranged from 9 to 77% (Table 3) with 16 accessions classified as VR to resistant (R), 53 accessions classified as moderately resistant (MR) to moderately susceptible (MS), and four accessions classified as susceptible (S; Table 1). PI 382161 (Tokai 66) in the early maturity group and PI 382153 (Nobeoka Bozu) and PI 382154 (Nyu Bai) in the intermediate maturity group were classified as VR (Table 4). Similar to ND 2710, the FHB index of the three VR lines was consistently low across the three years of replicated tests (Table 3). Selections with an FHB index comparable to BacUp but higher than ND 2710 (R to FHB based on index) were CItr 12002, PI 182568, PI 182586, PI 345731, PI 434987, and PI 519790,

in the early maturity group; PI 182561, PI 182583 and PI 462151 in the intermediate maturity group; and CItr 5103, PI 192660, and PI 81791 in the late maturity group. Among the 31 lines classified as MR based on FHB index (Table 4), CItr 12021, PI 163429, PI 182591, and PI 411132 in the early maturity group; PI 285933, PI 351256, and PI 351816 in the intermediate maturity group; PI 185380 and PI 382167 in the late maturity group had average FHB indices of 31 to 35% (Table 3). Most of those lines had FHB indices less than or equal to the FHB index of BacUp in at least one of the four years. Among the 22 lines classified as MS based on FHB index, 10 accessions, PI 184512, PI 192498, and PI 352062 in the early maturity group; PI 203083 and PI 344465 in the intermediate maturity group; and CItr 11215, PI 192219, PI 213833, PI 351476, and PI 57364 in the late maturity group had mean FHB indices of 56 to 60%. Funo (PI 213833) belonged to this group of selections. Funo has been characterized as an MS type following point inoculation in the greenhouse (Yu et al., 2006) and in field evaluations (Liu and Wang, 1991). Therefore, we believe that the 10 accessions with the highest FHB indices, which were classified as MS, had some level of FHB resistance. The FHB index cut-off (60% of disease) between MS and S was reasonable for materials evaluated in our nurseries because the disease pressure was high and uniform throughout the evaluation years.

Accessions PI 584934, of intermediate maturity, and CItr 12470, PI 113948, and PI 113949, of late maturity, had FHB indices greater than 60% and were classified as S (Tables 3 and 4). PI 113948 (Kooperatorka) and CItr 12470 (Frontana) were as susceptible as the susceptible checks Sonalika and Wheaton (Table 3). Kooperatorka and Frontana have been reported to have lower FHB indices than Sumai 3 and Nobeoka Bozu (Snijders, 1990). In our disease nursery, the former two lines were consistently very susceptible as measured by FHB index. Their susceptibility in our nursery could be due to the seed source, or perhaps these lines responded differently in our testing environment.

### FHB Resistance Measured by VSK

Mean VSK of the 73 FHB resistant selections ranged from 21 to 79% (Table 3). We classified 27 accessions as VR to R, 26 accessions as MR, 17 accessions as MS, and three accessions as S (Table 1). Accessions with VSK comparable to ND 2710 and rated VR were PI 382161 (Tokai 66), of early maturity; PI 104131 (Excelsior), PI 382153 (Nobeoka Bozu), and PI 382154 (Nyu Bai), of intermediate maturity; and PI 185380, PI 192660 (Prodigio Italiano) and PI 81791 (Sapporo Haru Komugi Jugo), of late maturity. Accessions with VSK within the range of BacUp and classified as R included six accessions of early maturity, six accessions of intermediate maturity, and six accessions of late maturity (Tables 3 and 4). The VSK measurements in the R class were stable and consistently low across years.

Seven accessions, including PI 213833 (Funo) and CItr 12470 (Frontana), were classified as MS and had some of the highest values of VSK of 56 to 60% among the selections from this study. Field-based evaluation of VSK on Funo has not been reported. The VSK level of Frontana in our study was much higher than that reported by Mesterhazy et al. (2005). There were four accessions classified as S, including PI 182561 (Sin Chunaga), and two accessions derived from Sin Chunaga (PI 411132, Gogatsu-Komugi and PI 182586, Norin 43), and PI 182583 (Chuko) (Table 4). These four lines were rated as R to MR according to FHB index (Table 3). Seeds from those lines grown in the FHB nursery were frequently discolored and classified as MS to S. However, most of the discolored kernels were plump and light weight, and lacked the pink color that is characteristic of FHB damage. The discoloration of the seed was most likely due to frequent mist-irrigation and other environmental stresses during kernel development. This hypothesis was supported by relatively low DON on these lines (Table 3).

# FHB Resistance Measured by DON Concentration

Average DON of the resistant selections ranged from 2.1 to 14.5 µg g<sup>-1</sup> (Table 3), which was much lower than means of DON for the susceptible checks. We selected for FHB resistance primarily based on FHB index and VSK; DON was a secondary criterion. Lower DON in the FHB resistant selections suggested that selection based on head blight intensity and visible seed damage successfully selected for resistance to DON. Twenty-five accessions had DON levels in the VR to R classes, 39 had DON levels in the MR class, and only nine accessions had DON in the MS class (Table 1). The four accessions rated VR were PI 382161 (Tokai 66) and PI 382140 (Abura) in the early maturity group, and PI 382154 (Nyu Bai) and PI 382153 (Nobeoka Bozu) in the intermediate maturity group (Tables 3 and 4). The four VR accessions consistently had DON levels lower than that of ND 2710 in the three years of the replicated evaluation nursery (Table 3). Among the 21 accessions classified as R, PI 382167 and PI 264927 in the late maturity group had DON levels similar to ND 2710. Of the 43 accessions with DON levels classified as MR, PI 345731 and PI 351743, in the early maturity group, and PI 185380, PI 83729, PI 192219, and CItr 11215, in the late maturity group, had 8.3 to 8.5 µg g<sup>-1</sup> DON, which was only 0.2 to 0.4 μg g<sup>-1</sup> higher than BacUp (Table 3). Nine accessions were rated MS, including known resistant sources Frontana (CItr 12470), Mentana (PI 132856), and Funo (PI 213833). Mesterhazy et al. (2005) reported that Frontana had about 69 to 263% of the DON concentration of Nobeoka Bozu. In our study the DON level of Frontana was about seven times higher than that of Nobeoka Bozu. The higher level of DON of Frontana in our study could be due to differences in seed source or environment.

# Correlation among FHB Index, VSK, and DON Concentration

Among the 73 selections, DON concentration was significantly correlated with FHB index (r = 0.40, P < 0.01) and VSK (r = 0.24, P < 0.05) (Table 5); however, FHB index and VSK was not correlated. We believe a group of germplasm with relatively low FHB index but high VSK, represented by PI 182561 (Sin Chunaga), PI 182583 (Chuko), PI 182586 (Norin 43), and PI 41132 (Gogatsu-Komugi), contributed to the poor correlation between FHB index and VSK. For example, when these four lines were removed from the correlation analysis, the correlation between FHB index and VSK became significant (r =0.33, P < 0.01), while the correlation coefficients between the other traits were similar to that when these lines were not removed. A positive correlation between FHB index and plant height (r = 0.22, P < 0.05) and a negative correlation between VSK and plant height (r = -0.66, P <0.001) were observed. Although a wide range of maturity was observed, days-to-heading was not correlated with any of the FHB measurements, indicating that the disease pressure was consistent during the evaluation period.

#### DISCUSSION

Evaluation of FHB resistance is complex and subject to environment (Parry et al., 1995). It has been suggested that FHB evaluation should be performed over a period of several years (Christensen et al., 1929; Fuentes et al., 2005; Hanson et al., 1950). We followed these recommendations by screening these traits (i.e., FHB index, VSK, and DON) over several years. To ensure constant and adequate disease pressure for evaluating unadapted germplasm of wide ranging maturity, we inoculated with both corn spawn and conidial suspensions and adjusted the misting period according to the daily weather (specifically wind and temperature) conditions during the inoculation period. The FHB index, VSK, and DON for the susceptible checks were consistently high over years and maturity groups. Lines with high FHB indices will likely be susceptible in other environments and not worthy of further evaluation. However, accessions with a moderate or low index but not recommended for FHB resistance in this report should be retested over several years if one is interested in using those accessions as resistance sources.

Out of the 1045 accessions of spring wheat from Asia, South America, and Europe evaluated in this study, 73 accessions were selected as putative FHB resistant sources. Among these, FHB resistance in 63 of the 73 accessions has not been reported previously. Resistant accessions were identified from all the geographic regions evaluated. Thirty-two resistant accessions were from four countries in South America,

Table 4. Original National Small Grains Collection (NSGC) seed information and new NSGC accession in Fusarium head blight (FHB) resistant selections, their maturity, and reactions to FHB measured by FHB index, percentage visual scabby kernel (VSK), and concentration of deoxynivalenol (DON) in 73 spring wheat accessions selected for FHB resistance from the NSGC.

	Original NSGC seed information				Selection		ISGS seed rmation <sup>‡</sup>		Re meas		
Accession	ID	Pedigree	Status	Country	type <sup>†</sup>	Accession	ID	Maturity§	Index	VSK	DON
Cltr 11215	Belgrade 4		Cultivated	Yugoslavia	Head	PI 644114	Cltr11215-sel-fhb	Late	MS	R	MR
Cltr 12002	Renacimiento	Americano 25C open pollinated	Cultivar	Uruguay	Row			Early	R	MR	R
Cltr 12021	Centenario	selection from landrace	Cultivar	Uruguay	Head	PI 644115	Cltr12021-sel-fhb	Early	MR	MR	MR
Cltr 12470	Frontana	Fronteira/Mentana	Cultivar	Brazil	Head	PI 644116	Cltr12470-sel-fhb	Late	S	MS	MS
Cltr 13136	Rio Negro	Supresa/Centenario	Cultivar	Brazil	Row			Intermediate	MR	MR	MR
Cltr 17427	16-52-2		Breeding	Brazil	Row			Late	MR	MR	MS
Cltr 2492	White Russian		Landrace	China	Head	PI 644113	Cltr2492-sel-fhb	Early	MR	MR	MR
Cltr 5103	274		Landrace	Argentina	Row			Late	R	R	MR
PI 104131	Excelsior	Arminda/Virtue	Cultivar	Argentina	Row			Intermediate	MR	VR	R
PI 104138	Klein Triunfo	Americano 25C/Pelon 33C1	Cultivar	Argentina	Row			Early	MS	MS	MR
PI 113948	Kooperatorka	selection from Krymka	Cultivar	Ukraine	Row			Late	S	MR	R
PI 113949	Stepnjachka	selection from Banatka Khersonskaya	Cultivar	Ukraine	Row			Late	S	R	R
PI 132856	Mentana	Rieti/Wilhelmina//Akagomughi	Cultivar	Italy	Head	PI 644118	PI132856-sel-fhb	Intermediate	MS	MR	MS
PI 163429			Cultivated	Argentina	Head	PI 644119	Pl163429-sel-fhb	Early	MR	R	MR
PI 163439			Cultivated	Argentina	Head	PI 644120	Pl163439-sel-fhb	Early	MR	MR	MR
PI 168716	Klein Condor	Standard F.C.S./Sud Oeste F.C.S.	Cultivar	Argentina	Head	PI 644121	Pl168716-sel-fhb	Early	MS	R	MR
PI 168727	Bahiense	Klein Sinmarq/Eureka F.C.S.	Cultivar	Argentina	Head	PI 644122	Pl168727-sel-fhb	Intermediate	MR	MR	MR
PI 182561	Sin Chunaga		Cultivated	Japan	Row			Intermediate	R	S	MR
PI 182565	Haya Komugi		Cultivated	Japan	Row			Intermediate	MR	MS	MR
PI 182568	Norin 34	Shinchunaga/Eshimashinriki	Cultivar	Japan	Head	PI 644123	PI182568-sel-fhb	Early	R	MS	R
PI 182583	Chuko		Landrace	Japan	Head	PI 644124	PI182583-sel-fhb	Intermediate	R	S	R
PI 182586	Norin 43	Shiromansaku/Akakomugi 3// Shichunaga	Cultivar	Japan	Head	PI 644125	Pl182586-sel-fhb	Early	R	MS	MR
PI 182591	Norin 61	Fukuoka 18/Shinchunaga	Cultivar	Japan	Head	PI 644126	Pl182591-sel-fhb	Early	MR	MS	R
PI 184512	H 51	Americano 25e/Favorito//Universal	Breeding	Argentina	Head	PI 644127	Pl184512-sel-fhb	Early	MS	R	MS
PI 185380	Prodigio Italiano	)	Cultivated	Italy	Head	PI 644128	Pl185380-sel-fhb	Late	MR	VR	MR
PI 185383	3084		Cultivated	Argentina	Row			Early	MR	MR	R
PI 185843	Surpresa	Polyssu/Alfredo Chaves 6-21	Cultivar	Brazil	Row			Intermediate	MR	R	MR
PI 192219	Hatvani		Cultivar	Hungary	Head	PI 644129	Pl192219-sel-fhb	Late	MS	MR	MR
PI 192498	lv C 390 1/2 10132		Cultivated	Argentina	Head	PI 644130	PI192498-sel-fhb	Early	MS	MR	MR
PI 192634	Trintecinco	Alfredo Chaves 3–21/Alfredo Chaves 4–21	Cultivar	Brazil	Row			Intermediate	MR	MR	MR
PI 192660	Prodigio Italiano		Cultivated	Italy	Head	PI 644131	PI192660-sel-fhb	Late	R	VR	MR
PI 203083	Wabian		Cultivated	Paraguay	Head	PI 644132	Pl203083-sel-fhb	Intermediate	MS	MS	R
PI 213833	Funo	Duecentodieci/Damiano	Cultivar	Italy	Head	PI 644133	Pl213833-sel-fhb	Late	MS	MS	MS
		1 Colonista/Frontana Erythrospermum 7623-1/	Breeding	Brazil	Row			Late	MS	MS	MS
PI 233207	Odesskaja 13	Lutescens 62 Selection from Romanian	Cultivar	Ukraine	Row			Intermediate	MR	MR	MR
PI 256958	Academia 48	land variety	Cultivar	Romania	Head	PI 644134	Pl256958-sel-fhb			R	MR
PI 264927	220		Landrace	Greece	Head	PI 644135	Pl264927-sel-fhb	Late	MR	R	R
PI 264940	111a		Landrace	Greece	Head	PI 644136	Pl264940-sel-fhb			MR	MR
PI 264946	1032		Landrace	Italy	Head	PI 644137	Pl264946-sel-fhb			MS	MR
PI 264998	628		Landrace	Greece	Head	PI 644138	Pl264998-sel-fhb			MR	R
PI 272348	Lontoi		Cultivated	Hungary	Row			Intermediate		MR	MR
PI 285933	Chudoskaja		Cultivated	Poland	Row	DI 0 4 44 0 0	DI004075 1.55	Intermediate		R	MS
PI 294975	Artemowska	Solo 50//01/19/19/01/01/01/19	Cultivated	Bulgaria	Head	PI 644139	Pl294975-sel-fhb	Late	MR	VR	MR
PI 344454	Buck Austral	Sola 50//Quivira/Guatrache/3/Massaux No. 1/Buck Quequen 2-2-11	Cultivar	Argentina	Row			Early	MS	R	MR

	Oriç	ginal NSGC seed information		Selection		ISGS seed rmation <sup>‡</sup>			Reactions measured by <sup>¶</sup>		
Accession	ID	Pedigree	Status	Country	type <sup>†</sup>	Accession	ID	Maturity§			
PI 344465	Laureano Alvarez Laah	Benvenuto Inca/Klein 157	Cultivar	Argentina	Row			Intermediate	MS	MR	R
PI 344467	Oncativo Inta	Thatcher/Sinvalocho M.A.// Beckman 1971	Cultivar	Argentina	Row			Early	MS	MR	R
PI 345731	Tezanos Pintos Precoz	Frontana//Thatcher/Sinvalocho	Cultivar	Argentina	Row			Early	R	R	MR
PI 349534	533b		Landrace	Switzerland	Head	PI 644140	Pl349534-sel-fhb	Intermediate	MS	R	MR
PI 351221	Newthatch Selection		Cultivated	Switzerland	Head	PI 644141	Pl351221-sel-fhb	Intermediate	MR	VR	R
PI 351256	Japon 2		Cultivated	Japan	Head	PI 644142	Pl351256-sel-fhb	Intermediate	MR	MS	R
PI 351476	Vaulion		Cultivar	Switzerland	Head	PI 644143	Pl351476-sel-fhb	Late	MS	R	MR
PI 351743	Cluj 49-926		Cultivar	Romania	Head	PI 644144	Pl351743-sel-fhb	Early	MR	R	MR
PI 351748	Jasi 10t		Cultivar	Romania	Head	PI 644145	Pl351748-sel-fhb	Intermediate	MS	MR	MR
PI 351816	Froment Du Japon		Cultivated	Switzerland	Row			Intermediate	MR	MS	R
PI 351993	Z.88.54		Breeding	Switzerland	Head	PI 644146	Pl351993-sel-fhb	Intermediate	MS	MR	R
PI 352062	Vivela Mar		Cultivated	Argentina	Row			Early	MS	MR	R
PI 360869	Fujimi Komugi	Norin 67/2*Norin 26	Cultivar	Japan	Head	PI 644147	Pl360869-sel-fhb	Late	MR	MS	MR
PI 362437	lii/14-B		Landrace	Yugoslavia	Row			Late	MR	MR	MR
PI 382140	Abura		Cultivar	Brazil#	Row			Early	R	MS	VR
PI 382144	Encruzilhada	Fortaleza/Kenya Farmer	Cultivar	Brazil	Head	PI 644148	PI382144-sel-fhb	Late	MR	MS	R
PI 382153	Nobeoka Bozu		Landrace	Japan	Row			Intermediate	VR	VR	VR
PI 382154	Nyu Bai		Landrace	Japan	Row			Intermediate	VR	VR	VR
PI 382161	Tokai 66		Cultivar	Brazil#	Row			Early	VR	VR	VR
PI 382167	16-52-9	Red Hart/PG 1	Breeding	Brazil	Row			Late	MR	R	R
PI 411132	Gogatsu-Komuç	giGokuwase 2/Norin 61	Cultivar	Japan	Row			Early	MR	S	MR
PI 434987	Estanzuela Young	Bage/4/Thatcher/3/Frontana// Kenya 58/Newthatch	Cultivar	Uruguay	Row			Early	R	MR	MR
PI 462151	Shu Chou Wheat No. 3		Cultivated	China	Row			Intermediate	R	R	R
PI 519790	274-1-118	Bage/Tehuacan/3/Frontana/Kenya 58/Newthatch/RL 4151	Breeding	Uruguay	Row			Early	R	MR	MR
PI 57364	Cltr 7175		Landrace	China	Head	PI 644117	PI57364-sel-fhb	Late	MS	MS	MR
PI 584926	Pantaneiro	Sonora 63*2/Lagoa Vermelha	Cultivar	Brazil	Row			Early	MR	MS	MS
PI 584934	Whestphalen	CNT 10/Burgas 2//Jacui	Cultivar	Brazil	Row			Intermediate	S	MR	MR
PI 81791	Sapporo Haru Komugi Jugo		Cultivar	Japan	Row			Late	R	VR	MS
PI 83729	Magyarovar 81		Cultivar	Hungary	Row			Late	MR	R	MR

<sup>†</sup>Row: in the unreplicated FHB evaluation nursery of 1998 or 1999, the whole plot planted from the NSGC seeds were harvested, the harvested seeds were multiplied in Christchurch, NZ, in crop season 1998–1999 or 1999–2000, and used in the replicated FHB evaluation nursery in 1999–2002. Head: In unreplicated disease screening nursery 1998 or 1999, a few head of low FHB were selected from plants of the NSGC seeds, the harvested seeds were multiplied in Christchurch, NZ, and used in the replicated FHB evaluation nursery in 1999–2002.

26 accessions were from nine countries in Europe, and 15 accessions were from China and Japan (Table 1). Therefore, FHB resistance of European origin consisted of 36% of FHB resistant selections in this study. Most of the accessions from China and Japan had a VR to MR reaction based on FHB index. Only one European accession, PI 192660 (Prodigio

Italiano), was classified as R based on FHB index, while the remaining European lines were MR to S. Selections classified as VR to R based on VSK consisted of 13 accessions from Europe, 10 accessions from South America, and four accessions from Asia. More selections from Europe fell in the groups VR to R based on VSK, compared to selections

<sup>&</sup>lt;sup>‡</sup>The seed of the head selections made from the original NSGC was deposited in the NSGC with a new accession number and ID.

<sup>§</sup>Early, mean days from planting to heading equal or less than 57 d; Intermediate, mean days from planting to heading 58–61 d; Late, mean days from planting to heading more than 61 d.

<sup>¶</sup>VR, very resistant; R, resistant; MR, moderately resistant; MS, moderately susceptible; S, susceptible.

<sup>\*</sup>This wheat line is most likely introduced from Japan to Brazil (T. Ban, personal communication, 2006).

Table 5. Correlation coefficient among Fusarium head blight (FHB) index, percentage visual scab kernel (VSK), and deoxynivalenol (DON), plant height, and days to heading of the 73 selections of FHB resistant germplasm based on means of replicated field nurseries from 2000 to 2002 in Brookings, SD.

	VSK	DON	Plant height	Days to heading
Index	0.09	0.40**	0.22*	0.10
VSK		0.24*	-0.66**	-0.08
DON			0.08	0.10
Plant height				0.49**

<sup>\*</sup>Significant at P < 0.05.

from South America and Asia. Low DON concentration was more common in selections from China and Japan than from Europe and South America. We believe that the European spring wheat FHB resistance gene pool appeared to be unique in that selected accessions were moderately resistant in the field and produced sound seed as measured by VSK and reduced DON contamination.

Among the 63 accessions of putative new FHB resistant sources, known FHB resistant cultivars Frontana and Shin Chunaga appeared in the pedigrees of seven accessions (Table 4), and 11 accessions were landraces. The landraces and modern breeding lines without known resistance in the pedigrees may represent new genes for resistance. For example, the two new Chinese resistant accessions, CItr 2492 and PI 57364, were landraces from Heilongjiang Province in northeastern China. Those lines might have FHB resistance different from that in modern cultivars represented by Sumai 3 and its relatives from the spring wheat regions in the middle and lower Yangtze River Valley and South China (Liu and Wang, 1991; Wan et al., 1997; Yu et al., 2006).

There was a wide range of FHB reaction in terms of FHB index, VSK, and DON. Accessions of the best FHB resistance were PI 382153 (Nobeoka Bozu), PI 382154 (Nyu Bai), and PI 382161(Tokai). Accession PI 382140 (Abura) was rated as R based on FHB index, MS based on VSK, but VR based on DON. Brazil was the seed source of PI 382161 and PI 382140 in NSGC. However, the names Tokai 66 and Abura suggest that they were originally from Japan and may have been introduced to Brazil (T. Ban, personal communication, 2006). Molecular marker data results suggest that the above four accessions might have the Sumai 3 FHB resistance gene Fhb1 on the chromosome 3BS (Liu and Anderson, 2003; S. Liu, personal communication, 2007). In our work these lines were more resistant to DON contamination than Sumai 3 and should be valuable for introducing a higher level of resistance to DON in wheat cultivars.

In our study, selections with low VSK were generally associated with low DON contaminations. Nine of the 27 accessions classified as VR to R based on VSK, nine

were also classified as VR to R and 16 were classified as MR, based on DON. However, this was not always the case. PI 184512, PI 285933, and PI 81791 were classified R for VSK, but MS for DON accumulation. Resistance measured based on FHB index did not necessarily indicate resistance expressed as a low frequency of VSK. Significant correlation of index and VSK with DON indicated that selection based on field visual estimate and kernel appearance generally would result in lower DON levels, but the extent of DON reduction based on the index and VSK varies among genotypes. The discovery of potentially diverse FHB resistance sources of spring wheat germplasm in this study will provide diversity and may produce higher levels of resistance when different sources are combined in breeding. The diverse sources of resistance will also provide materials for studies of mechanisms of FHB resistance in wheat. The addition of European spring wheat germplasm for FHB resistance is encouraging for future discovery of resistance in nontraditional FHB resistance gene pools. The novelty and types of FHB resistance in these selections should be further characterized in the greenhouse by point inoculation and by haplotyping using closely linked and diagnostic molecular markers.

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#### References

Ban, T. 2000. Studies on the genetics of resistance of resistance to Fusarium head blight caused by *Fusarium graminearum* in wheat.
p. 82–93. *In* W.J. Raupp et al. (ed.) Proc. Int. Symp. on Wheat Improvement for Scab Resistance, Suzhou and Nanjing, China. 5–11 May 2000. Michigan State Univ., East Lansing.

Ban, T., and K. Suenaga. 2000. Genetic analysis of resistance to Fusarium head blight caused by *Fusarium graminearum* in Chinese wheat cultivar Sumai 3 and the Japanese cultivar Saikai 165. Euphytica 113:87–99.

Bushnell, W.R. 2002. Designating types of scab resistance: A discussion. p. 200. *In Proc. National Fusarium Head Blight Forum*, Erlanger, KY. 7–9 Dec. 2002. Michigan State Univ., East Lansing.

<sup>\*\*</sup>Significant at P < 0.01.

- Christensen, J.J., E.C. Stakman, and F.R. Immer. 1929. Susceptibility of wheat varieties and hybrids to Fusarium head blight in Minnesota. Minnesota Agric. Exp. Stn. Tech. Bull. 59.
- Frohberg, R.C., R.W. Stack, and M. Mergoum. 2004. Registration of spring wheat germplasm ND2710 resistant to Fusarium head blight. Crop Sci. 4:1498–1499.
- Fuentes, R.G., H.R. Mickelson, R.H. Busch, R. Dill-Macky, C.K. Evans, W.G. Thompson, J.V. Wiersma, W. Xie, Y. Dong, and J.A. Anderson. 2005. Resource allocation and cultivar stability in breeding for Fusarium head blight resistance in spring wheat. Crop Sci. 5:1965–1972.
- Gervais, L., F. Dedryver, J.Y. Morlais, V. Bodusseau, S. Negre, M. Bilous, C. Groos, and M. Trottet. 2003. Mapping of quantitative trait loci for field resistance to Fusarium head blight in an European winter wheat. Theor. Appl. Genet. 106:961–970.
- Hanson, E.W., E.R. Ausemus, and E.C. Stakman. 1950. Varietal resistance of spring wheats to Fusarium head blight. Phytopathology 40:902–914.
- Liu, S., and J.A. Anderson. 2003. Marker assisted evaluation of Fusarium head blight resistant wheat germplasm. Crop Sci. 43:760-766.
- Liu, Z.Z., and Z.Y. Wang. 1991. Improved scab resistance in China: Sources of resistance and problems. p. 178–188. *In* D.A. Saunders (ed.) Wheat for the nontraditional warm areas. CIMMYT, Mexico, D.F.
- McKendry, A.L., D.N. Tague, R.L. Wright, J.A. Tremain, and S.P. Conley. 2005. Registration of 'Truman' wheat. Crop Sci. 45:421–423.
- 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.
- Mergoum, M., R.C. Frohberg, J.D. Miller, and R.W. Stack. 2005. Registration of 'Steele-ND' wheat. Crop Sci. 45:1163–1164.
- Mergoum, M., R.C. Frohberg, J.D. Miller, R.W. Stack, T. Olsona, T.L. Friesen, and J.B. Rasmussen. 2006. Registration of 'Glenn' wheat. Crop Sci. 46:473–474.
- Mesterhazy, A. 1987. Selection of head blight resistant wheats through improved seedling resistance. Plant Breed. 98:25–36.
- Mesterhazy, A. 1995. Types and components of resistance to Fusarium head blight of wheat. Plant Breed. 114:377–386.
- Mesterhazy, A., T. Bartok, G. Kaszonyi, M. Varga, B. Toth, and

- J. Varga. 2005. Common resistance to different *Fusarium* spp. causing Fusarium head blight in wheat. Eur. J. Plant Pathol. 112:267–281.
- Miller, J.D., J.V. Young, and D.R. Sampson. 1985. Deoxynivalenol and Fusarium head blight resistance in spring cereals. Phytopathol. Z. 113:359–367.
- Parry, D.W., P. Jenkinson, and L. McLeod. 1995. Fusarium ear blight (scab) in small grain cereals—A review. Plant Pathol. 44:207–238.
- Ruckenbauer, P., H. Buerstmayr, and M. Lemmens. 2001. Present strategies in resistance breeding against scab (*Fusarium* spp.). Euphytica 119:121–127.
- Rudd, J.C., R.D. Horsley, A.L. McKendry, and E.M. Elias. 2001. Host plant resistance genes for Fusarium head blight: Sources, mechanisms, and utility in conventional breeding systems. Crop Sci. 41:620–627.
- Schroeder, H.W., and J.J. Christensen. 1963. Factors affecting resistance of wheat to scab caused by *Gibberella zeae*. Phytopathology 53:831–838.
- Shaner, G. 2002. Resistance in hexaploid wheat to Fusarium head blight. p. 208–211. *In* Proc. National Fusarium Head Blight Forum, Erlanger, KY. 7–9 Dec. 2002. Michigan State Univ., East Lansing.
- 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 J. Perkowski. 1990. Effects of head blight caused by *Fusarium culmorum* on toxin content and weight of wheat kernels. Phytopathology 80:566–570.
- Stack, R.W. 2000. Return of an old problem: Fusarium head blight of small grains. Available at www.apsnet.org/education/feature/FHB/. Plant Health Prog. DOI: 10.1094/PHP-2000-0622-01-RV.
- Wan, Y.F., C. Yen, and J.L. Yang. 1997. Sources of resistance to head scab in *Triticum*. Euphytica 94:31–36.
- Wilcoxson, R.D., R.H. Busch, and E.A. Ozmon. 1992. Fusarium head blight resistance in spring wheat cultivars. Plant Dis. 76:658–661.
- Yu, J.B., G.H. Bai, S.B. Cai, and T. Ban. 2006. Marker-assisted characterization of Asian wheat lines for resistance to Fusarium head blight. Theor. Appl. Genet. 113:308–320.