Reaction of Elite Wheat Genotypes from the Northern Great Plains of North America to Septoria Diseases

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ABSTRACT

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Stagonospora nodorum blotch (SNB), caused by *Phaeosphaeria nodorum*, and Septoria tritici blotch (STB), caused by *Mycosphaerella graminicola*, are the main pathogens of the Septoria disease complex of wheat (*Triticum aestivum*) in North America. This study was conducted to determine the disease reaction of 126 elite hard red spring, white, and durum wheat cultivars and advanced breeding lines collected from the northern Great Plains of the United States and Canada to SNB and STB. Seedlings of the 126 wheat genotypes were evaluated for resistance to SNB and STB under controlled environmental conditions. Moreover, these 126 wheat genotypes also were infiltrated with culture filtrate of *P. nodorum* isolate Sn2000. Based on disease reactions, three cultivars (McNeal, Dapps, and Oklee) and 12 advanced breeding lines (CA-901-580W, 97SO254-8-1, MN03291, MN03308, WA007925, MT0245, ND756, ND801, ND803, ND808, ND809, and ND811) adapted to the northern Great Plains were found to be resistant to both Septoria diseases and insensitive to the culture filtrate. Additionally, eight genetically diverse lines and cultivars, including two tetraploid wheat genotypes, were identified to be resistant to both Septoria diseases. These results suggest that the wheat genotypes contain a broad genetic base for resistance to the Septoria diseases in the northern Great Plains of the United States and Canada, and the resistant sources identified in this study may be utilized in wheat-breeding programs.

Stagonospora nodorum blotch (SNB), caused by *Phaeosphaeria nodorum* (E. Müll.) Hedjar. (anamorph: *Stagonospora nodorum* (Berk.) E. Castell. & Germano), and Septoria tritici blotch (STB), caused by *Mycosphaerella graminicola* (Fückel) J. Schröt. (anamorph: *Septoria tritici* Desm.), are the major components of the Septoria disease complex in North America (12,23). In combination with tan spot, caused by *Pyrenophora tritici-repentis* (Died.) Drechsler, Septoria diseases form the leaf spot complex, which is a major limiting biotic factor for wheat (*Triticum aestivum*) production in the northern Great Plains of North America (12,29). Septoria diseases can cause yield losses of 10 to 15%; however, under conditions favorable for disease development, it can be as high as 50% (16). Foliar disease reduces photosynthetic area of the leaves, resulting in a decrease in the amount of carbohydrates available during seed fill. Septoria diseases also

cause significant loss in grain quality by grain shriveling, red smudge, and black point (8,22).

Intensive wheat production, changes in cultural practices (including shifts from conventional tillage and stubble burning to reduced tillage), shorter crop rotations, and growing of wheat cultivars susceptible to leaf spots have resulted in development of leaf spot epidemics worldwide (7,8). A complex of these diseases occurs in nature; thus, managing leaf spot diseases is difficult. A number of management practices are available and can be useful in controlling leaf spot diseases. These include the use of nonhost crops in the rotation and destruction and avoidance of infested straw and stubble by either burning or burying (7,8). In recent years, no or minimum tillage and lack of stubble burning have been gaining popularity for reasons ranging from the economics of production to protection of environment. Although the application of fungicides is effective in controlling leaf spot diseases, their use may not be cost effective due to low yield potential or low wheat prices. The recent discovery of strobilurin (QoI) fungicide-resistant leaf spot pathogens (10) has further increased interest in breeding and growing resistant wheat cultivars. However, most of the widely grown wheat cul-

tivars in the northern Great Plains of North America are susceptible to leaf spot diseases.

Confounding factors that hamper breeding for host resistance for Septoria diseases include lack of continuous selection pressure, low heritability, sporadic epidemics, scarcity in resistance sources, and, most importantly, lack of knowledge on the virulence spectrum in the pathogens (8). There have been conflicting reports regarding the existence of specific virulence in the wheat-*M. graminicola* pathosystem. Some researchers believe that no true differences in virulence of isolates exist (31), whereas others (15,21) reported that isolate-specific resistance of wheat to STB follows a gene-for-gene relationship. However, the total number of races of *M. graminicola* is unknown. Although variation in aggressiveness in *P. nodorum* populations has been observed (17,27), no pathogenic races of this fungus have been convincingly reported. Some level of gene-for-gene interaction has been demonstrated in studies with selected host genotypes inoculated under controlled environments with selected isolates (4,9). It has been reported further that a single aggressive isolate might be more reliable than a mixture of isolates in screening for resistance to SNB (4) and STB (33).

Knowledge of genetics of resistance is crucial to develop strategies for breeding for resistance to Septoria diseases. Wheat has both isolate-nonspecific, polygenic resistance and isolate-specific, monogenic resistance to Septoria diseases. Twelve major genes, *Stb1* to *Stb12*, for resistance to STB have been identified and mapped (1-3,21,30). Among these, only two genes, *Stb1* and *Stb4*, have been used extensively to develop the wheat cultivars resistant to STB in the United States (1,2,30). Quantitative trait loci (QTL) conferring resistance to STB also have been identified (3,6,34). Similarly, inheritance of resistance to SNB was reported to be qualitative (9,18) and quantitative (19,32).

Evaluating wheat for field resistance to SNB and STB is hampered by the variation in environmental conditions, amount of natural inoculum, and competition between pathogens causing foliar diseases for healthy tissue to infect and colonize (13,20). In addition, mixed infection of

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leaf spot diseases that produce similar lesions has been observed (5,20), thus making it difficult to evaluate reaction of wheat genotypes to individual races or diseases. Evaluation of seedlings for resistance to *Septoria* diseases under controlled conditions provides accurate and repeatable results (8). Screening wheat seedlings by artificial inoculation in controlled conditions also permits examination of large genotypes for resistance under uniform disease pressure (28). Additionally, a positive correlation was found between assessments of resistance at the seedling stage and ratings of adult plants under conditions of natural infection in the field (14,25,30).

Singh et al. (28) evaluated the reaction of 126 elite wheat genotypes developed in the northern Great Plains of North America to individual virulent races of *P. tritici-repentis* and its toxins. As a part of our effort to evaluate resistance to other leaf spot diseases, this study was conducted to determine the reaction of the same 126 wheat genotypes to SNB and STB. The wheat genotypes were further screened with culture filtrate of *Phaeosphaeria nodorum* isolate Sn2000 to determine whether culture filtrate can be surrogate to spore inoculation.

MATERIALS AND METHODS

Plant material. A collection of 126 elite wheat genotypes was evaluated in greenhouse studies. This collection contained hard red spring wheat, white wheat, and durum cultivars along with advanced breeding materials from both private and public wheat-breeding programs of the Northern Great Plains of the United States and Canada. The wheat genotypes used in this study, their pedigree, and source of origin are given in Table 1.

SNB. Inoculum production. Inoculum of *P. nodorum* isolate Sn2000 (18,19) was produced using a modified method of Liu et al. (19). Cultures were grown at room temperature on V8-potato dextrose agar (PDA) medium (150 ml of V8 juice, 30 g of sucrose, 10 g of agar, 3 g of calcium carbonate, and 10 g of Difco PDA in 850 ml of distilled water) in 10-cm-diameter petri dishes under continuous illumination provided by 25-watt cool-white fluorescent tubes. Fresh cultures of *P. nodorum* were prepared by rubbing a 0.5-cm-diameter plug of stock cultures on fresh V8-PDA medium. Pink pycnidia were observed on the surface of the V8-PDA medium after 7 days. A pycnidiospores suspension was prepared by flooding each petri dish with sterile distilled water and scraping the medium surface with a flamed glass slide to liberate the pycnidiospores. The resulting suspension was filtered through four layers of cheesecloth. The spore concentration was measured with a hemacytometer and adjusted to 1.0×10^6 spores/ml prior to inoculation. Two drops of Tween 20 (poly-

oxyethylene sorbitan monolaurate) were added per 100 ml of spore inoculum.

Disease screening. The seedlings of the 126 wheat genotypes, including resistant check 'Salamouni' and susceptible checks 'ND495,' 'Conway,' and 'Coulter,' were grown in the greenhouse at an average temperature of 21°C with a 16-h photoperiod. Seed were planted in plastic cones (3.8 cm in diameter and 20 cm in length) filled with Fison sunshine blend no. 1 (Fison Horticulture, Vancouver, BC, Canada). The seedlings were watered and fertilized as needed. Plants at the two-leaf stage were sprayed with the spore suspension of *P. nodorum* isolate Sn2000 until runoff using a CO₂-pressurized hand sprayer. Following inoculation, the seedlings were incubated for 24 h under continuous leaf wetness in a mist chamber to enhance the infection process and then placed on benches in a growth chamber set at 22 and 17°C (day and night) temperatures with a 16-h photoperiod. Plants were rated for SNB 8 days after inoculation using a 1-to-5 scale (9). The disease classes of the 1-to-5 rating scale were defined as 1 = pinpoint dark-brown lesions without chlorosis; 2 = small lesions with very little necrosis or chlorosis; 3 = chlorotic lesions or necrotic lesions completely surrounded by a chlorotic ring, lesions generally not coalescing; 4 = lesions completely surrounded by chlorotic zones, some of the lesions coalescing; and 5 = extensive chlorosis and large necrotic lesions, extensive coalescing of lesions. Ratings of 1 and 2, indicating restricted development of necrotic lesions with little or no chlorosis, were considered resistant, whereas those greater than 2 were considered susceptible because, in these reactions, there was little or no restriction of the development of chlorosis or necrosis.

Culture filtrate production and infiltration. Culture filtrate containing toxins was produced from *P. nodorum* isolate Sn2000 using the procedures of Liu et al. (18). We used 10-fold diluted filtrate in our bioassay instead of partially purified toxin. Briefly, fresh culture of *P. nodorum* isolate Sn2000 was initiated by placing its mycelial plugs (two plugs/plate), previously stored at -20°C, on fresh V8-PDA plates. The plates were incubated at 22°C in the dark for 4 days to induce mycelial growth and then placed under continuous light for 5 to 6 days to enhance pycnidial formation. Pycnidiospores were obtained by adding 25 to 30 ml of sterile distilled water into individual plates and gently scraping the plate surface. The spore suspension was passed through three layers of cheesecloth and then quantified with the hemacytometer to about 1×10^6 spores/ml. To obtain culture filtrate of the fungus, 200 µl of spore suspension was added into a 250-ml flask containing 100 ml of Fries medium (5.0 g of ammonium tartrate, 1.0 g of ammonium nitrate, 0.5 g of magnesium sul-

fate, 1.3 g of potassium phosphate [dibasic], 2.6 g of potassium phosphate [monobasic], 1.0 g of yeast extract, and 30 g of sucrose dissolved in 1,000 ml of sterile distilled water). The flasks were placed on a shaker for 48 h in the dark at room temperature and then moved to an incubator in the dark at 21°C for 3 weeks. The filtrate was passed through filter paper (Whatman no. 1) and then through 0.45-µm sterilized membrane (Gelman Sciences, Ann Arbor, MI). The filtrate was diluted 10-fold with sterile distilled water prior to infiltration. Approximately 25 µl of the culture filtrate was infiltrated into the 2-week-old wheat seedlings using a 1-ml syringe without needle. The infiltrated second leaf was marked by a nontoxic permanent marker. Plant reactions to the culture filtrate infiltration were recorded 4 days after infiltration. The presence or absence of necrosis at the site of infiltration indicated sensitivity or insensitivity, respectively, to the culture filtrate. Infiltration of Fries medium and sterile distilled water into resistant check Salamouni and susceptible checks ND495, Conway, and Coulter were used as controls.

STB. Inoculum production. A single-spored isolate, Ma04-9-4, of *M. graminicola* collected from diseased wheat leaf in northeastern North Dakota was used for inoculation. Inoculum of *M. graminicola* was produced using a modified method of McCartney et al. (21). Fresh V8-PDA plates were streaked with a pycnidiospore suspension of *M. graminicola* and plates were placed under continuous light at room temperature for 5 days to produce pycnidiospores. Approximately 30 ml of sterile distilled water was added to each plate and pycnidiospores were collected using a flamed wire loop. The spore suspension was counted with a hemacytometer and adjusted to 2.0×10^7 spores/ml prior to inoculation.

Disease screening. Seedlings of the 126 wheat genotypes, including resistant check Salamouni and susceptible checks ND495, Conway, and Coulter, were inoculated with a spore suspension at the two-leaf stage using a CO₂-pressurized hand sprayer until runoff. After inoculation, the plants were kept in the mist chamber under continuous leaf wetness for 48 h. Subsequently, the plants were moved to a growth chamber bench with the same light and temperature regime as for SNB screening. Plants were rated for STB infection 14 days after inoculation using the 0-to-5 lesion-type disease scale (21), where 0 = immune without visible symptoms; 1 = highly resistant with small flecks; 2 = resistant with small chlorotic or necrotic lesions; 3 = moderately susceptible, characterized by coalescence of necrotic or chlorotic lesions normally toward the leaf tip and, to a lesser extent, elsewhere on the leaf blade; 4 = susceptible, with coalesced necrotic lesions; and 5 = highly susceptible, with

large necrotic lesions extensively coalesced. Ratings of 0 to 2 were considered resistant and 3 to 5 were considered susceptible.

Experimental design and statistical analysis. To evaluate the 126 wheat genotypes, two independent experiments were conducted in the greenhouse in spring and

fall of 2006 for each disease. Each experiment was conducted in a randomized complete block design with three replicates. Each replicate consisted of the 126 wheat

Table 1. Wheat type, origin, pedigree, and disease reaction of 126 elite wheat genotypes to spore inoculum and culture filtrate of *Phaeosphaeria nodorum* (Stagonospora nodorum blotch [SNB]) and *Mycosphaerella graminicola* (Septoria tritici blotch [STB])

Genotype	Type-origin ^a	Pedigree	SNB		STB
			Spore ^b	Culture	Spore ^c
Oxen	HRSW-SD	SDY366A/SDZ004A	4.11 S	Sensitive	2.79 S
Ingot	HRSW-SD	Butte86/SD3004/Dalen	3.92 S	Sensitive	2.08 S
Norpro	HRSW-CO	N93-0119	3.58 S	Sensitive	2.54 S
Parshall	HRSW-ND	Keene//Grandin*2/Glupro	3.38 S	Sensitive	2.96 S
Reeder	HRSW-ND	IAS20*4/H567.71//Stoa/3/ND 674	2.38 S	Insensitve	2.88 S
Alsen	HRSW-ND	ND 716 (Sumai3/Wheaton//Grandin)	4.38 S	Sensitive	2.71 S
Knudson	HRSW-CO	Karl/Krona/3/Bergen//Erik/MN73167	1.58 R	Insensitve	2.21 S
Hanna	HRSW-CO	N93-2424/AC DOMAIN	3.67 S	Sensitive	2.54 S
Briggs	HRSW-SD	BW114/Bergen/SD3097	2.75 S	Sensitive	2.71 S
Granite	HRSW-MT	Unknown	1.67 R	Insensitve	2.88 S
Oklee	HRSW-MN	2375/SBF0670	1.54 R	Insensitve	1.81 R
Dapps	HRSW-ND	Kitt/Amidon//GR/Stoa sib	1.45 R	Insensitve	1.98 R
Steele-ND	HRSW-ND	Parshall/ND706	3.54 S	Sensitive	2.75 S
Fryer	HRSW-CO	Sonja/Vance//Sumai3/Dalen	1.54 R	Insensitve	2.29 S
Trooper	HRSW-MT	Keystone/Ivan	1.63 R	Insensitve	2.32 S
Banton	HRSW-MN	Buck Antorcha/HJ 9	1.33 R	Insensitve	2.88 S
Saturn	HRSW-MN	ANB'S//HEGE312-75-262/CHAT'S	1.92 R	Insensitve	3.25 S
Polaris	HRSW-MN	Lona/4/Kent/B564//SO/3/Kent/2*B564//SAP	1.39 R	Insensitve	2.56 S
2375	HRSW-ND	Olaf//Era/Suqamuxi68/3/Cis/ND487//Lark	2.63 S	Sensitive	2.67 S
Verde	HRSW-MN	MN7663/SBY35A	1.31 R	Insensitve	2.17 S
Keene	HRSW-ND	Stoa's'3/IAS20*4//H567.71//Amidon	1.54 R	Insensitve	2.32 S
Butte 86	HRSW-ND	Butte*2/3/ND551//Butte*2/ND507	3.64 S	Sensitive	2.21 S
McNeal	HRSW-MT	PI125000/Centana//PK176/Frontiera/3/Glenman	1.33 R	Insensitve	1.63 R
Walworth	HRSW-SD	SD 3116/OXEN	2.58 S	Sensitive	2.83 S
Dandy	HRSW-MN	Unknown	3.64 S	Sensitive	1.96 R
Granger	HRSW-SD	SD8070/SD3165	3.75 S	Sensitive	3.29 S
SD3618	HRSW-SD	SD8119/SD3236	4.08 S	Sensitive	3.04 S
SD3635	HRSW-SD	SD8070/SD3118//SD3225	4.10 S	Sensitive	2.64 S
SD3668	HRSW-SD	SD8089/SD8070	4.04 S	Sensitive	2.96 S
SD3687	HRSW-SD	SD3305/KS91W005-1-4//SD8089	4.29 S	Sensitive	2.96 S
SD3746	HRSW-SD	SD93380/SD3335//Oxen	3.22 S	Sensitive	2.33 S
SD3827	HRSW-SD	W94-320/SD3367//SD3335	3.29 S	Sensitive	2.50 S
SD3851	HRSW-SD	ND 2897/SD3219//SD3414	4.04 S	Sensitive	2.88 S
SD3854	HRSW-SD	ND 2897/SD3219//SD3414	4.50 S	Sensitive	2.53 S
SD3860	HRSW-SD	AZ99FHB-RC	1.54 R	Insensitve	3.08 S
SD3868	HRSW-SD	ND 2897/SD3479//SD3411	3.00 S	Sensitive	2.92 S
SD3870	HRSW-SD	FHBCOMP	3.63 S	Sensitive	2.79 S
BZ 998-447W	WW-MT	Spillman/WestBred 906R	1.71 R	Insensitve	3.00 S
CA-902-701	OW-MT	Keystone/Granite	1.29 R	Insensitve	2.60 S
CA-901-580W	WW-MT	White Sharpshooter/Lars	1.92 R	Insensitve	1.78 R
N99-0241	HRSW-CO	N93-0090/Hamer	1.46 R	Insensitve	2.31 S
96S0404-03	HRSW-CO	Hamer//RHT21/Lars	1.26 R	Insensitve	2.10 S
98S0051-1	HRSW-CO	N93-0243//CHISCAB#7/N93-0338	3.96 S	Sensitive	3.33 S
98S0127-06	HRSW-CO	N97-0214/3/N93-0338//Sumai3/Dalen	3.54 S	Sensitive	3.46 S
97S0254-8-1	HRSW-CO	N94-0236/IVAN//Verde	1.31 R	Insensitve	1.94 R
MN97369	HRSW-MN	MN97369-98*4/Pavon	1.45 R	Insensitve	2.08 S
MN97448	HRSW-MN	HJ98/SHARP	4.03 S	Sensitive	2.71 S
MN97695	HRSW-MN	MN92387/SBE0303-23	2.81 S	Sensitive	2.71 S
MN99017	HRSW-MN	Fujian 5114/Grandin	4.35 S	Sensitive	1.79 R
MN99436-6	HRSW-MN	Norlander/HJ98	3.67 S	Sensitive	2.43 S
MN00232-2-2	HRSW-MN	MN2540W/KULM//ND2710	3.54 S	Sensitive	2.79 S
MN00258-2	HRSW-MN	INGOT/HJ98	3.71 S	Sensitive	2.08 S
MN00261-4	HRSW-MN	MN95286/MN94155//Verde	3.46 S	Sensitive	2.06 S
MN01261-8	HRSW-MN	MN95229/ND694	1.38 R	Insensitve	2.21 S
MN01281	HRSW-MN	SD-AD B9/MN93434(2375/Grandin)	3.50 S	Sensitive	2.54 S
MN01333-A	HRSW-MN	MN97008/MN97695	3.04 S	Sensitive	1.71 R
MN01108-6	HRSW-MN	MN95229//MF9606/Aberdeen 9	3.42 S	Sensitive	2.71 S
MN01116	HRSW-MN	MN95211(PAR11(MRNC/BUC//BLD/PSN)/MN90124)//LizCimmyt9/MN95023	3.70 S	Sensitive	2.90 S
MN01180-4	HRSW-MN	LizCimmyt17/Parshall//HJ98	1.46 R	Insensitve	3.40 S
MN01278-7	HRSW-MN	MN94200/SD-AD B61	3.32 S	Sensitive	1.58 R

(continued on next page)

^a Types of wheat: HRSW, OW, and WW = hard red spring wheat, other wheat, and white wheat, respectively. Origins of the different genotypes studied: CAN, CO, KS, MEX, MN, MT, ND, SD, and WA stand for Canada, Colorado, Kansas, Mexico, Minnesota, Montana, North Dakota, South Dakota, and Washington, respectively.

^b Plants were rated on a 1-to-5 rating scale where 1 and 2 were considered resistant (R) and 3 to 5 susceptible (S).

^c Plants were rated on a 0-to-5 rating scale where 0 to 2 were considered resistant (R) and 3 to 5 susceptible (S).

^d LSD = least significant difference.

genotypes planted in cones. Four plants planted in a cone formed the experimental unit. For the reaction of 126 wheat genotypes to the culture filtrate, an experiment

including 12 plants per genotype (4 plants and three replicates) was infiltrated with culture filtrate produced by *P. nodorum* isolate Sn2000.

An analysis of variance for each disease was performed using the general linear model (PROC GLM) (Statistical Analysis System version 8.2) (26). The existence of

Table 1. (continued from preceding page)

Genotype	Type-origin ^a	Pedigree	SNB		STB
			Spore ^b	Culture	Spore ^c
MN02255	HRSW-MN	ND721/MN97448	1.54 R	Insensitve	2.33 S
MN02258	HRSW-MN	ND724/MN97448	3.38 S	Sensitive	2.65 S
MN02296-A	HRSW-MN	Verde/MN93415//HJ98	3.15 S	Sensitive	2.54 S
MN03098	HRSW-MN	MN97803-10/DANDY-1	1.54 R	Insensitve	2.96 S
MN03119	HRSW-MN	Alsen-1/MN97803-10	1.88 R	Insensitve	2.89 S
MN03130	HRSW-MN	MN97695-4/MN97448-17	2.58 S	Sensitive	3.50 S
MN03148	HRSW-MN	MN99390-1/MN97803-10	1.50 R	Insensitve	2.92 S
MN03239	HRSW-MN	MN98543/MN97448	1.17 R	Insensitve	2.61 S
MN03243	HRSW-MN	01X019-3/MN97803-10	3.00 S	Insensitve	2.13 S
MN03279	HRSW-MN	MN98224/MN97448	1.28 R	Insensitve	2.89 S
MN03291	HRSW-MN	MN98389/MN98021	1.38 R	Insensitve	1.83 R
MN03308	HRSW-MN	MN95002//Parshall/MN97518	1.25 R	Insensitve	1.33 R
MN03328	HRSW-MN	Alsen-1/MN97448-17	3.67 S	Sensitive	1.67 R
WA007925	OW-WA	Spillman/WPB906R//Sunstar II	1.54 R	Insensitve	1.71 R
WA007931	WW-WA	PI591045/3/Tanager 'S'/Torim 73//Spillman	3.54 S	Sensitive	2.29 S
MT 0245	HRSW-MT	MT9433/Reeder	1.32 R	Insensitve	1.86 R
MT 0249	HRSW-MT	Reeder/MT9433	1.25 R	Insensitve	2.40 S
ND 747	HRSW-ND	ND2831//Parshall/ND706	4.13 S	Sensitive	2.20 S
ND 751	HRSW-ND	ND2709/3/GR*3//Ramsey/ND622/4/ND688/ND674	1.29 R	Insensitve	2.08 S
ND 752	HRSW-ND	ND2891/ND721	1.92 R	Insensitve	2.17 S
ND 753	HRSW-ND	ND721/Parshall	1.72 R	Insensitve	2.92 S
ND 755	HRSW-ND	ND2849/ND721	3.67 S	Sensitive	2.50 S
ND 756	HRSW-ND	ND2849/ND721	1.83 R	Insensitve	1.95 R
ND 800	HRSW-ND	ND741 SELN (Parshall/ND706)	3.63 S	Sensitive	2.17 S
ND 801	HRSW-ND	ND2849/ND721	1.46 R	Insensitve	1.88 R
ND 802	HRSW-ND	ND721/ND725	1.33 R	Insensitve	2.58 S
ND 803	HRSW-ND	ND2831/FO.2808	1.13 R	Insensitve	1.54 R
ND 804	HRSW-ND	ND744 SELN	3.32 S	Sensitive	2.25 S
ND 805	HRSW-ND	ND2857/3/KITT/Amidon//GR/Stoa SIB	3.14 S	Insensitve	2.75 S
ND 806	HRSW-ND	SD3348/Reeder//ND721	1.54 R	Insensitve	2.38 S
ND 807	HRSW-ND	ND2831//Parshall/ND706	3.92 S	Insensitve	1.63 R
ND 808	HRSW-ND	ND2857/3/Kitt/Amidon//GR/Stoa SIB	1.54 R	Insensitve	1.79 R
ND 809	HRSW-ND	ND2831//Parshall/ND706/3/ND721	1.51 R	Insensitve	1.83 R
ND 810	HRSW-ND	ND694/GR*2/RL4137//Amidon	1.33 R	Insensitve	2.46 S
ND 811	HRSW-ND	ND724/2*Reeder	1.38 R	Insensitve	1.54 R
NDSW0207	WW-ND	9734/AC Vista	1.31 R	Insensitve	2.25 S
NDSW0217	WW-ND	Sunstate/9734	4.21 S	Sensitive	2.13 S
NDSW0345	WW-ND	MT9904/Alsen	4.07 S	Sensitive	2.28 S
NDSW0347	WW-ND	Reeder//IDO560/9712	4.21 S	Sensitive	2.13 S
NDSW0431	WW-ND	BaiHuo/Kanto107//Express/3/MTHW9420	4.04 S	Sensitive	3.13 S
Erik	HRSW-KS	Kitt/2/Waldron/Era	1.29 R	Insensitve	2.40 S
Glenlea	OW-CAN	Pemina*2/Bage/2/CB100	3.63 S	Sensitive	2.39 S
6B-365	OW-CAN	Unknown	1.74 R	Insensitve	2.13 S
6B-662	OW-CAN	Unknown	1.50 R	Insensitve	1.86 R
ND 495	HRSW-ND	Justin*2/3/ND259/Conley//ND112	4.54 S	Sensitive	3.67 S
Conway	HRSW-CAN	Chris/Siete-Cerros-66//Neepawa/Opal	4.08 S	Sensitive	3.17 S
Katepwa	HRSW-CAN	Neepwa*6/RL-2938/3/Neepawa*6//CI-8154/2*Frocor	4.13 S	Sensitive	2.79 S
Kenyon	HRSW-CAN	Neepawa*5/Buck Manantial	4.50 S	Sensitive	2.36 S
Coulter	Durum-CAN	DT-188/Dt-224//DT-182	3.96 S	Sensitive	3.79 S
4B-160	OW-CAN	Unknown	1.63 R	Insensitve	1.54 R
<i>T. turgidum</i> no. 283	OW-CAN	PI352519	1.17 R	Insensitve	1.04 R
Intros no. 7	OW-MEX	Unknown	1.76 R	Insensitve	1.22 R
92 MREHTR28B	OW-CAN	Car853/Coc//Vee'S'/3/E7408/Pam'S'//Hor	1.17 R	Insensitve	1.14 R
2000 Spelt no. 20	OW-CAN	PI348771/Oberkum	1.24 R	Insensitve	1.21 R
CIMMYT L no. 18	OW-MEX	Unknown	1.33 R	Insensitve	1.18 R
Salamouni	OW-CAN	Unknown	1.54 R	Insensitve	1.74 R
Mountrail	Durum-ND	D8479/Renville	1.79 R	Insensitve	3.17 S
Ben	Durum-ND	D8024/Monroe	3.35 S	Sensitive	3.00 S
Lebsock	Durum-ND	Munich/D8469	2.33 S	Sensitive	3.00 S
Pierce	Durum-ND	D86117/D88289	2.83 S	Insensitve	2.71 S
Maier	Durum-ND	D8193/D8335	3.29 S	Sensitive	2.61 S
Plaza	Durum-ND	DT606/D8291	3.07 S	Sensitive	1.54 R
Dilse	Durum-ND	Maier/D88273	3.50 S	Sensitive	3.36 S
Grenora	Durum-ND	D901260/D901419	3.54 S	Sensitive	1.42 R
Divide	Durum-ND	Ben/D901282//Belzer	3.04 S	Sensitive	3.07 S
Alkabo	Durum-ND	D901247/D89263	2.71 S	Sensitive	3.04 S
LSD _(0.05) ^d	...	...	0.53	...	0.49

significant differences ($P < 0.05$) in disease reaction among genotypes was determined using Fisher's protected least significant differences (LSD) at $P < 0.05$ level. The 126 genotypes were considered having fixed effects whereas the two greenhouse experiments had random effects.

RESULTS AND DISCUSSION

Analysis of variance of the disease data revealed significant ($P < 0.05$) differences among genotypes in all experiments for the two Septoria diseases. There was no significant ($P < 0.05$) genotype experiment interaction for any of the diseases studied. The results of statistical analysis permitted combining the two experiments for each disease, and mean disease score of each genotype with respect to SNB and STB is given in Table 1.

In all, 57 genotypes showed resistant reactions to spore suspension of *P. nodorum* isolate Sn2000 whereas 62 genotypes were insensitive to the culture filtrate. Only 31 genotypes were resistant to STB. Combined results reveal that 23 of the 126 genotypes tested showed resistance to SNB and STB and insensitivity to culture filtrate of *P. nodorum* isolate Sn2000 (Table 1). Preliminary results of artificially inoculated field evaluation of adult plant reaction of these 126 genotypes to SNB indicates that the resistance sources identified in greenhouse tests are also good in field conditions (Singh et al., unpublished).

Among the resistant sources identified, three cultivars (McNeal, Dapps, and Oklee) and 12 advanced breeding lines (CA-901-580W, 97SO254-8-1, MN03291, MN03308, WA007925, MT0245, ND756, ND801, ND803, ND808, ND809, and ND811) are well adapted to the Northern Great Plains of the United States and Canada and thus can be used successfully and directly in wheat-breeding programs. To broaden the genetic base of resistance to Septoria diseases, the resistance from lines 6B-662, Intros no. 7, 2000 Spelt no. 20, 92 MREHTR28B, CIMMYT L no. 18, and cv. Salamouni also can be used. Some of these resistant sources were found to be resistant to leaf spot diseases in a previous study (29). All durum lines were susceptible; however, two tetraploid accessions, *T. turgidum* no. 283 and 4B-160, exhibited high levels of resistance to the two diseases and insensitivity to culture filtrate of isolate Sn2000. The pedigree of resistant sources identified in this study indicate that they come from diverse genetic backgrounds and possibly may possess different resistance genes that can be utilized in gene pyramiding and developing cultivars with broad and durable resistance to Septoria diseases.

Singh et al. (28), studying the same set of 126 wheat genotypes, identified resistant sources to races 2, 3, and 5 and toxins Ptr ToxA and Ptr ToxB of *Pyrenophora*

tritici-repentis. Combined results from this study and those of Singh et al. (28) reveal that cv. Oklee and breeding line MN03291, adapted to the northern Great Plains of North America, are resistant to tan spot, SNB, and STB. Additionally, hexaploid genotypes 92MREHTR28B, CIMMYT L no. 18, Intros no. 7, Salamouni, and 2000 Spelt no. 20 and tetraploid genotype *T. turgidum* no. 283 (PI352519) were found to be resistant to the three leaf spot diseases tested. These resistant sources also come from a diverse genetic background and potentially possess different genes for resistance to leaf spotting diseases. Hence, incorporation of resistance from these sources will result in development of cultivars with broad, effective, and durable resistance to leaf spot diseases.

The highly resistant reaction of the newly identified sources of resistance was due to a strong hypersensitive reaction of the resistant genotypes that resulted in just black spot at the point of infection and no lesion expansion. Evaluation of genotypes under strong selection pressure for disease development, which is achieved in greenhouse tests, tends to select for highly resistant genotypes possessing major genes for resistance (24,27). Preliminary genetic studies (Singh et al., unpublished) indicate that the newly identified sources of resistance possess major genes for leaf spot diseases. This further enhances the utility of these genotypes in wheat-breeding programs as donors for leaf spot resistance.

Host-selective toxins are important factors in many host-pathogen interactions, including wheat-*P. nodorum*. The *P. nodorum* isolate Sn2000 is reported to produce multiple toxins and culture filtrate possessing multiple toxins, and is able to differentiate between resistant and susceptible genotypes (11,18). This is the first report to evaluate a large collection of elite wheat genotypes using culture filtrate of *P. nodorum*. Although we cannot determine the susceptibility of a genotype to a specific toxin, this study indicates that culture filtrate can be used as a surrogate for spore inoculation because 96% of the genotypes tested gave similar responses to spore inoculation. The time required for wheat genotype evaluation by culture filtrate infiltration (4 days) is half the duration required by spore suspension (8 days). Reaction to the culture filtrate is clear and can differentiate between sensitive and insensitive plants, and easily can be combined with screening for resistance to other leaf spot diseases (28). This study reveals that culture filtrates can be used for large-scale evaluation of early generation breeding material; however, final confirmation of SNB resistance has to be done by spore inoculum to avoid selection of lines which are insensitive to toxins but susceptible to spore inoculum. Five (4%) wheat genotypes were susceptible to spore suspension but resistant to culture filtrate. There may

be factors involved in pathogenicity other than the toxins in the wheat-*P. nodorum* pathosystem. Presently, genetic studies with these genotypes are being conducted to determine the exact cause of differential response to spore inoculation and culture filtrate infiltration.

An association has long been observed between tall stature, late maturity, and resistance to Septoria diseases (8,13). In the last two decades, wheat breeders in the northern Great Plains of the United States have been breeding for early maturing and semidwarf cultivars with resistance to rusts and Fusarium head blight (FHB), caused by *Fusarium graminearum* Schwabe. Among the best source for FHB resistance is the Chinese line Sumai 3, which also has acceptable levels of resistance to Septoria diseases (13). As success is achieved in developing rust-resistant and FHB-moderately resistant cultivars, the emphasis on breeding for leaf spot resistance is increasing. In the last decade, a major emphasis on leaf spot breeding in the northern Great Plains of North America has been carried out, and the results of this study indicate the success of the effort. The resistant sources identified in this study have good agronomic traits and high-quality characteristics; therefore, these genetic resources can be used directly in wheat-breeding programs.

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