Performance of Four New Leaf Rust Resistance Genes Transferred to Common Wheat from *Aegilops tauschii* and *Triticum monococcum*

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ABSTRACT

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The objective of this study was to test the performance of four new wheat leaf rust resistance genes previously transferred from wild relatives of common wheat. Leaf rust resistance gene *Lr43*, in wheat germplasm line KS92WGRC16, was originally from *Aegilops tauschii*. A second resistance gene, in line KS92WGRC23, was transferred from *Triticum monococcum* var. *monococcum*. Two other genes, in lines KS93U3 and KS96WGRC34, were obtained from *T. monococcum* var. *boeoticum*. In greenhouse tests, the typical low infection types produced by these lines were fleck (;), immune (0), fleck with chlorosis (;C), and heterogeneous (X–) for KS92WGRC16, KS92WGRC23, KS96WGRC34, and KS93U3, respectively. In field tests in Kansas and Texas, KS92WGRC23 and KS92WGRC16 were highly resistant. KS93U3 was moderately resistant in Kansas but moderately resistant to moderately susceptible in Texas. KS96WGRC34 was moderately resistant in Kansas but moderately resistant to susceptible in Texas. Greenhouse adult-plant tests with race PBJL of *Puccinia recondita* f. sp. *tritici* indicated that KS92WGRC16, KS92WGRC23, and KS96WGRC34 were highly resistant, but KS93U3 gave a moderately resistant reaction. Growth-chamber studies in different environments (12, 16, 20, and 24°C) showed slight temperature effects on the expression of resistance in KS96WGRC34 but not in the other lines. Tests with nine races of *P. recondita* f. sp. *tritici* indicated that only KS92WGRC16 was resistant to all the races. Races PNML and PNMQ were virulent on KS92WGRC23, and race TFGL was virulent on both KS93U3 and KS96WGRC34. The genes in the four germplasm lines should be used in combination with other resistance genes to prolong their usefulness.

Additional keywords: alien germplasm, durability, *Triticum aestivum*, *T. tauschii*

Genetic resistance is the cheapest, most effective, and most environmentally sound method of controlling leaf rust of wheat (*Triticum aestivum* L.). Unfortunately, development and management of durably resistant cultivars in the central Great Plains has not been entirely successful (1,15,19, 25). New resistant cultivars frequently are overcome by new or previously undetected races of the leaf rust fungus (*Puccinia recondita* Roberge ex Desmaz. f. sp. *tritici* (Eriks. & E. Henn.) D.M. Henderson) after several years of large-scale production. For example, cvs. Abilene, Karl, and Newton were classified as resistant when originally released but are now susceptible to prevalent races.

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Evolution of new races of *P. recondita* f. sp. *tritici* in the Great Plains is favored by many factors, including (i) large populations of inoculum produced on highly susceptible, widely grown cultivars like TAM 107; (ii) frequent overwintering of the pathogen on juvenile winter wheat; (iii) oversummering on volunteer wheat; (iv) commercial production of certain cultivars over a wide range of latitude corresponding to the migration route of the pathogen; (v) reliance on a relatively small pool of resistance genes in popular cultivars (18); and (vi) use of only one or a few effective resistance genes in each cultivar. Development of new sources of resistance genes could ameliorate the last two problems and possibly slow the evolution of new pathogen races.

Wild relatives of cultivated wheat are a valuable source of new resistance genes for improving wheat cultivars (5,6,9,12,13). The Wheat Genetics Resource Center at Kansas State University, Manhattan, recently released wheat germplasm line KS92WGRC16, which contains a leaf rust resistance gene (designated *Lr43*) from *Aegilops tauschii* Coss. (synonym *T. tauschii*) (5). Diploid *A. tauschii* (DD) is the D-genome progenitor of common wheat (AABBDD) and is widely distributed in countries surrounding the Caspian Sea, including Turkey, Iran, Pakistan, Afghanistan, Azerbaijan, Armenia, southern Russia (Dagestan), Georgia, Tashkent, and Turkmenia (9,11). Kihara et al. (11) collected a large number of varieties and strains of this wild species of wheat in 1965.

Three other genes now in wheat lines KS92WGRC23, KS93U3, and KS96WGRC34 were transferred from cultivated einkorn wheat (*T. monococcum* L. var. *monococcum,* AA*)* and its wild progenitor *T. monococcum* var. *boeoticum*. *T. monococcum* is the A-genome donor of common wheat. *T. monococcum* var. *monococcum* is still cultivated in the mountainous areas of southern Europe and Turkey (24).

Study of the characteristics and performance of the four new leaf rust resistance genes may help optimize their use in breeding programs. For instance, determination of the typical infection types (ITs) will facilitate recognition of the resistance genes in segregating populations. This information will supplement leaf rust resistance gene IT data recently compiled by McIntosh et al. (17). Leaf rust ITs are often affected by temperature (2–4). For example, Pretorius et al. (21) detected adult-plant wheat leaf rust resistance gene *Lr13* in seedlings of wheat at 25.5°C but not at 18°C. Temperature sensitivity could affect both the screening and deployment of leaf rust resistance genes.

One of the major questions in the evaluation of new resistance genes is whether they are stable across locations and years. Variable results may be caused by direct environmental effects on the interaction phenotype. Inconsistent results also may be obtained if a virulent pathogen race is present in some cases but absent in others. Field tests conducted over several locations and years may help reveal environmental sensitivity or the existence of rare virulent races. Seedling tests to an array of leaf rust fungal isolates collected from different ecological zones also will help to check whether there is preexisting virulence to these genes.

The current study was undertaken with the following objectives: (i) to characterize the typical low ITs and any other unique characteristics associated with these genes; (ii) to test how well the genes perform under field conditions in Kansas and Texas; (iii) to determine whether any of these

genes are temperature sensitive; and (iv) to determine whether there is preexisting virulence to these genes in the leaf rust fungal population.

MATERIALS AND METHODS

Plant materials. Hard red winter wheat germplasm lines containing the new leaf rust resistance genes investigated in this study are listed in Table 1. Crosses of resistant germplasm lines with susceptible wheat cultivars gave F_2 segregation ratios of 3:1 (resistant/susceptible) when tested with standard races of *P. recondita* f. sp. *tritici* (5,7,10). This suggested that each germplasm line contained a single dominant resistance gene. However, an approximate 12:3:1 (immune/intermediate/susceptible) ratio was produced with the combination of KS92WGRC23 and race PBJL. This suggested that an additional unidentified resistance gene with an intermediate IT is present in KS92WGRC23. The unidentified gene was not detectable in the following studies, because it was masked by the stronger resistance gene in KS92WGRC23 or the unidentified gene was ineffective against the isolates tested.

Several wheat cultivars and germplasm lines were used as controls, including Wrangler (PI477288), TAM 107 (PI495594), Karl 92 (PI564245), TAM 200 (PI578255), Wichita (CI11952), Century (PI502912), KS86WGRC2 (PI504517), KS90WGRC10 (PI549278), KS91WGRC11 (PI566668), and KS92WGRC15 (PI566669).

Leaf rust races. Single uredinial isolates of *P. recondita* f. sp. *tritici* and their avirulence/virulence phenotypes were provided by D. L. Long (USDA-ARS Cereal Rust Laboratory, St. Paul, MN) and M. G. Eversmeyer (USDA-ARS Plant Science and Entomology Research Unit, Manhattan, KS). Each isolate was assigned to a race according to the nomenclature of Long and Kolmer (14). The avirulence/virulence phenotypes of these races are given in Table 2. Races were increased on cv. Trison and stored in liquid nitrogen.

Field studies. Field nurseries were established at the Ashland experimental field, Manhattan, KS, during the growing seasons of 1992 to 1993, 1993 to 1994, and 1994 to 1995. The wheat germplasm lines were planted in three-row plots with 20 cm between rows. Plots were 2.7-m long by 0.6-m wide with no border between plots. Experiments were arranged in a randomized complete block design with four replications.

To enhance buildup of rust epidemics, test entries were inoculated uniformly when they were between boot and early heading stages of development with a mixture of urediniospores of different leaf rust fungal isolates collected from the Ashland field during previous years. Urediniospores were suspended in lightweight mineral oil (Phillips Petroleum Company, Bartlesville, OK) at a rate of 200 mg of spores per 600 ml of oil and were sprayed with a battery operated, fine-nozzle sprayer between 6:00 and 7:00 p.m.

Disease severity (percentage of leaf area affected by rust) in each line was assessed in Kansas twice a week between the early milk (73 on Zadoks' scale) (26) and hard dough stages (87 on Zadoks' scale) of development, using the modified Cobb scale (20). The host response to infection was scored, using R to indicate resistance or miniature uredinia; MR to indicate moderate resistance, expressed as small uredinia; MS to indicate moderate susceptibility, expressed as moderate sized uredinia (smaller than the fully compatible type); and S to indicate full susceptibility (23).

In Texas nurseries, single-row entries of 3 m each were planted at the Texas Agricultural Experiment stations at Prosper and Beeville during 1994 to 1995. These nurseries were exposed to natural infection. Rust severity estimations were made at the watery ripe (Zadoks 71) to milk (Zadoks 75) developmental stages.

Growth-chamber studies. Seedlings of the four germplasm lines with new leaf rust resistance genes, as well as parental lines and susceptible checks, were tested at different temperatures, using three races (CBBQ, CDBL, and MFBL) collected in the Great Plains area. Fifteen to twenty-five seeds of each germplasm line were planted in 6- to 7-cm-diameter plastic pots filled with vermiculite. Each pot represented an experimental unit, and treatments were replicated three times in a completely randomized design. Urediniospores of the three races were removed from storage in liquid nitrogen and were heat shocked at 40°C for 5 min. Primary leaves of each line were inoculated just before emergence of the second leaf, using a suspension of urediniospores in lightweight mineral oil (23). After inoculation, the plants were misted with tap water and placed in a refrigerated (16°C), moist chamber (100% relative humidity [RH]) overnight. After 16 h of incubation, plants were allowed to dry slowly and were moved to 12 ± 2, 16 ± 2, 20 ± 2, or 24 ± 2°C growth chambers. Seedlings were illuminated at about 200 µmol m^{-2} s^{-1} for 12 h each day with light from cool white fluorescent tubes suspended about 1 m above the plants. ITs were evaluated when they appeared to be fully developed for each temperature and were classified by the Stakman scale as modified by Roelfs et al. (23). The experiment was performed twice.

Race specificity was tested with nine races representing diverse virulence phenotypes from North America. ITs greater than 2+ (on a 0 to 4 scale) were considered susceptible. Wheat lines carrying resistance genes transferred to common wheat from *A. tauschii* in earlier studies, including R.L.6043 (*Lr21*), R.L.6044 (*Lr22a*), R.L.5497-1 (*Lr32*), KS86WGRC2 (*Lr39*), KS90WGRC10 (*Lr41*), and KS91WGRC11 (*Lr42*) (5,8,17), also were tested against these nine races. Plant growth, inoculation, and evaluation techniques were as described above. The experiment was performed twice.

Greenhouse studies. For adult-plant evaluations in the greenhouse, five to six seeds were sown in 6- to 7-cm-diameter plastic

Table 1. Pedigrees of four leaf rust-resistant wheat germplasm lines

Resistant hexaploid line	Pedigree	Source of resistance gene
KS92WGRC16	Triumph 64/3/KS8010-71/ TA2470//TAM 200	TA2470 *(Aegilops tauschii)*
KS92WGRC23	Karl*3//PI266844/PI355520	PI266844 *(Triticum monococcum* var. *monococcum)*
KS96WGRC34	'TAM 107'/TA749//'Wrangler'	TA749 *(T. monococcum* var. *boeoticum)*
KS93U3	Wrangler//Mustang*2/TA213	TA213 *(T. monococcum* var. *boeoticum)*

Table 2. Avirulence/virulence phenotypes of leaf rust races

Race[a]	Effective/ineffective host *Lr* genes
CBBQ	1, 2a, 2c, 3ka, 9, 11, 16, 17, 24, 26, 30/3a, 10, 18
CDBL	1, 2a, 2c, 3ka, 9, 11, 16, 17, 18, 26, 30/3a, 10, 24
KDBL	1, 3ka, 9, 11, 16, 17, 18, 26, 30/2a, 2c, 3a, 10, 24
MBRL	2a, 2c, 9, 16, 17, 18, 24, 26/1, 3a, 3ka, 10, 11, 30
MCDL	2a, 2c, 3ka, 9, 11, 16, 18, 24, 30/1, 3a, 10, 17, 26
MCRL	2a, 2c, 9, 16, 17, 18, 24/1, 3a, 3ka, 10, 11, 26, 30
MFBL	2a, 2c, 3ka, 9, 11,16, 17, 18, 30/1, 3a, 10, 24, 26
MGML	2a, 2c ,9, 11, 17, 18, 24, 26/1, 3a, 3ka, 10, 16, 30
PBBL	2a, 3ka, 9, 11, 16, 17, 18, 24, 26, 30/1, 2c, 3a, 10
PBJL	2a, 3ka, 9, 16, 18, 24, 26, 30/1, 2c, 3a, 10, 11, 17
PNML	2a, 11, 16, 17, 18, 26/1, 2c, 3a, 3ka, 9, 10, 24, 30
PNMQ	2a, 11, 16, 17, 26/1, 2c, 3a, 3ka, 9, 10, 18, 24, 30
TBGL	3ka, 9, 16, 17, 18, 24, 26, 30/1, 2a, 2c, 3a, 10, 11
TFGL	3ka, 9, 16, 17, 18, 30/1, 2a, 2c, 3a, 10, 11, 24, 26

[a] Race nomenclature and first three differential sets based on Long and Kolmer (14). The fourth differential set consisted of *Lr10* and *Lr18*.

pots filled with vermiculite. The seedlings were vernalized at 10°C for 7 weeks. At the end of vernalization, two seedlings per pot were transplanted into 15-cm-diameter plastic pots filled with a 1:1:3 peat moss/perlite/soil mixture. Approximately 0.3 g per pot of 15-30-15 (N-P-K) fertilizer was applied weekly. Seedlings were grown at 21 ± 5°C, with a 12-h photoperiod, in the greenhouse. Each pot represented an experimental unit, and treatments were replicated three times in a completely randomized design.

Plants were inoculated with race PBJL when they were at the flowering (Zadoks 65) to watery ripe stages of development. After inoculation, plants were incubated overnight in an enclosed plastic chamber with 100% RH for 15 h. Plants then were returned to a greenhouse with conditions similar to those described above. Disease severity was assessed on flag leaves of five randomly selected tillers 15 to 20 days after inoculation, using the scales and scoring procedures described for seedling tests. The experiment was performed twice.

RESULTS

Field reactions to leaf rust. The level of leaf rust infection in the field varied among locations and years (Table 3). The 1992 to 1993 crop year was particularly favorable for overwintering of the leaf rust fungus in Kansas, resulting in higher than average severity on susceptible cultivars (1). Rust severity also was high in Texas in 1995. KS92WGRC23 and KS92WGRC16 were highly resistant to leaf rust in both Kansas and Texas. KS93U3 was moderately resistant in Kansas but moderately resistant to moderately susceptible in Texas. KS96WGRC34 was moderately resistant in Kansas but moderately resistant to susceptible in Texas. TAM 200 (a parent of KS92WGRC16), Karl 92 (the recurrent parent of KS92WGRC23), and Wrangler (a parent of KS96WGRC34 and KS93U3) all had high ITs in the field tests.

Adult-plant reactions to leaf rust. Adult-plant tests conducted in the greenhouse with race PBJL showed levels of resistance similar to those observed under field conditions (Table 3). The ITs on flag leaves of KS92WGRC16, KS92WGRC23, and KS96WGRC34 were fleck (;), zero (0), and fleck with extra chlorosis (;C), respectively. KS93U3 was moderately resistant, producing a typical heterogeneous (X–) IT.

Effects of temperature on seedling reactions. KS92WGRC23 had a 0 IT and KS92WGRC16 had a ; IT at all temperatures with the three races (Table 4). KS96WGRC34 had a ;C IT with all of the races at 12 and 16°C. At 20 and 24°C, however, a mixture of flecks and minute pustules with extra chlorosis was observed, indicating a slight temperature effect on this line. KS93U3 had an X– IT with all three races at all temperatures.

Race specificity. The wheat germplasm line KS92WGRC23 was immune to seven and susceptible to two leaf rust races (Table 5). Contrary to the field data, susceptibility of this line to two races indicates that virulence already exists in the pathogen population for this particular resistance gene. KS92WGRC16 produced a ; IT with seven of the races. Races PNML and PNMQ, however, produced a slightly higher IT (;1C) on this genotype. None of the races were considered virulent on KS92WGRC16.

KS96WGRC34 expressed a ;C IT with most of the races. On this line, MCRL and

Table 3. Flag leaf reactions to *Puccinia recondita* f. sp. *tritici* of field- and greenhouse-grown adult plants of wheat germplasm lines and cultivars

	Flag leaf reaction					
	Manhattan, KS[a]			Prosper, TX[a]	Beeville, TX[a]	
Cultivar or line	1993	1994	1995	1995	1995	Greenhouse[b]
KS91WGRC16	0	0	0	0	0	;
KS92WGRC23	0	0	0	0	0	0
KS96WGRC34	10 MR	0	1 MR	10 MR	40–50 S	;C
KS93U3	20 MR	2 MR	2 MR	5–10 MR	10–20 MS	X–
Wrangler	30 S	55 S	28 S	80 S	80 S	3+
TAM 107	30 S	18 S	23 S	50 S	70 S	3+
Karl 92	50 S	25 S	55 S	60 S	70 S	2+
Wichita	60 S	55 S	28 S	40 S	80 S	NT[c]
TAM 200	30 S	1 S	1 S	80 S	80 S	;
Century	30 S	9 S	9 S	NT	NT	12–
KS86WGRC2	5 MR	1 MR	1 MR	NT	NT	NT
KS90WGRC10	5 MR	8 MR	0	1 R	0	NT
KS91WGRC11	0	0	0	1 R	0	NT
KS92WGRC15	5 MR	0	0	0	0	NT

[a] Field infection responses are based on the modified Cobb scale (20) and include two components: terminal disease severity and infection type; e.g., 1 = 1% severity, 5 = 5% severity, etc.; and 0 = immune; R = resistant; MR= moderately resistant; MS = moderately susceptible; and S = susceptible infection type (IT).

[b] The greenhouse adult plant ITs are 0 = no uredinia or other macroscopic sign of infection, ; = no uredinia but small hypersensitive necrotic or chlorotic flecks present, 1 = small uredinia surrounded by necrosis, 2 = small to medium uredinia surrounded by necrosis or chlorosis (green island may be surrounded by necrotic or chlorotic border), 3 = medium uredinia with or without chlorosis, 4 = large uredinia without chlorosis, X = heterogeneous (random distribution of variable-sized uredinia on a single leaf), C = more chlorosis than normal for the IT, + = uredinia somewhat larger than normal for the IT, and – = uredinia somewhat smaller than normal for the IT. A range of variation between ITs is recorded by indicating the range, with the most prevalent IT listed first (e.g., 0;, 23, or ;1C) (23).

[c] NT = not tested.

Table 4. Seedling infection types[a] (ITs) at four temperatures of wheat germplasm lines and cultivars inoculated with three races of *Puccinia recondita* f. sp. *tritici*

	12°C			16°C			20°C			24°C		
Cultivar or line	CBBQ	CDBL	MFBL	CBBQ	CDBL	MFBL	CBBQ	CDBL	MFBL	CBBQ	CDBL	MFBL
KS92WGRC16	;	;	;	;	;	;	;	;	;	;	;	;
KS92WGRC23	0	0	0	0	0	0	0	0	0	0	0	0
KS96WGRC34	;C	;C	;C	;C	;C	;C	;1C	;1C	;1C	;1C	;1C	;1C
KS93U3	X–	X–	X–	X–	X–	X–	X–	X–	X–	X–	X–	X–
Karl 92	2–	3	3	2	3+	3+	2	3+	3+	2	3+	3+
TAM 107	3	3	3	3	3	3	3	3	3	3	3	3+
TAM 200	0;	0;	0;	0;	0;	0;	0;	0	;1	0	0	;1
Wichita	3+	3+	3+	4	4	4	4	4	4	4	4	4
Wrangler	3	3	3	3	3	3+	3	3	3+	3	3	3+

[a] The seedling ITs are 0 = no uredinia or other macroscopic sign of infection, ; = no uredinia but small hypersensitive necrotic or chlorotic flecks present, 1 = small uredinia surrounded by necrosis, 2 = small to medium uredinia surrounded by necrosis or chlorosis (green island may be surrounded by necrotic or chlorotic border), 3 = medium uredinia with or without chlorosis, 4 = large uredinia without chlorosis, X = heterogeneous (random distribution of variable-sized uredinia on a single leaf), C = more chlorosis than normal for the IT, + = uredinia somewhat larger than normal for the IT, and – = uredinia somewhat smaller than normal for the IT. A range of variation between ITs is recorded by indicating the range, with the most prevalent IT listed first (e.g., 0;, 23, or ;1C) (23).

KDBL produced ITs of ;1C and ;2C, respectively. Race TFGL, however, was virulent on this line, producing a 3– IT. KS93U3 had moderately resistant ITs, ranging from ;2 to X– with most races. One race, TFGL, was virulent on this genotype.

Lines containing resistance genes *Lr21*, *Lr22a*, *Lr32*, *Lr39*, *Lr41*, and *Lr42*, which were transferred earlier to common wheat from *A. tauschii*, also were tested with the same nine races of the leaf rust fungus. In this experiment, our goal was to determine the reactions of these *Lr* genes to the races that were virulent on KS92WGRC23, KS93U3, and KS96WGRC34. The line carrying adult-plant resistance gene *Lr22a* (8) and cv. Wichita were seedling-susceptible to all the races (Table 6). PNML and PNMQ were virulent on the line with *Lr41*. The race specificity of this line was similar to KS92WGRC23, except KDBL and MCDL gave an intermediate IT on the *Lr41* line. Lines with *Lr21*, *Lr32*, *Lr39*, and *Lr42* were resistant to all the races tested.

DISCUSSION

New rust resistance genes in germplasm lines KS96WGRC34 and KS93U3 from *T. monococcum* var. *boeoticum* provided good levels of resistance in Kansas and north Texas field tests. However, KS96WGRC34 and KS93U3 were moderately susceptible to susceptible in south Texas at Beeville in 1995. Texas generally is considered a "hot spot" for leaf rust (13,19). Therefore, the difference in performance of resistant lines between locations is likely due to greater diversity of *P. recondita* f. sp. *tritici* races in south Texas. Isolates from south Texas can have different virulence combinations than isolates found in other parts of Texas (16). Greenhouse tests indicated that both KS96WGRC34 and KS93U3 are susceptible to race TFGL. The field tests are most easily explained by the presence of TFGL or similar races at Beeville, TX, but not at Prosper, TX, or Manhattan, KS. TFGL is scattered around the United States, and its population currently is increasing (D. Long, *personal communication*).

Although the genes in KS96WGRC34 and KS93U3 both are derived from *T. monococcum* var. *boeoticum* and both appear to have the same race specificity, they are not identical. The low IT for the gene in KS96WGRC34 is typically a fleck with extra chlorosis (;C). In contrast, the low IT for KS93U3 is typically heterogeneous (X–) with a mixture of pustule types. The gene in KS96WGRC34 also showed slight temperature sensitivity, whereas the gene in KS93U3 did not. These two genes segregated independently of each other, indicating they are not allelic (T. S. Cox, *unpublished data*).

The resistance gene transferred from *T. monococcum* var. *monococcum* to line KS92WGRC23 conferred very high resistance in all field tests. Greenhouse tests with standard races also had very low ITs, ranging from immune (0) to a few flecks (0;). However, races PNML and PNMQ were virulent on KS92WGRC23 in greenhouse tests. PNML and PNMQ also recently were found to be virulent on lines with *Lr41* (D. Long, *personal communication*; Table 6). PNMQ was first detected in 1993 on wheat cv. Coker 9877, which possesses *Lr9* plus *Lr24*. In 1993, PNMQ constituted 15% of the *P. recondita* f. sp. *tritici* isolates collected from the southern United States (22).

KS92WGRC16, containing *Lr43* from *A. tauschii*, is the only line of the four that was resistant in all field and greenhouse tests. Temperature did not appear to affect the reaction, and the gene was equally effective in seedlings and adult plants. The IT of *Lr43* was very low, ranging from fleck (;) to flecks with a few small pustules

Table 5. Seedling infection types[a] (ITs) of wheat germplasm lines and cultivars inoculated with nine races of *Puccinia recondita* f. sp. *tritici*

	Race								
Cultivar or line	**MGML**	**MCDL**	**TBGL**	**MBRL**	**KDBL**	**PNMQ**	**PNML**	**TFGL**	**MCRL**
KS92WGRC16	;	;	;	;	;	;1C	;1C	;	;
KS92WGRC23	0;	0;	0;	0;	0;	3	3	0;	0;
KS96WGRC34	;C	;C	;C	;C	;2C	;C	;C	3–	;1C
KS93U3	X–	;2	;2	X–	X–	;2C	;2C	3	X–
Karl 92	2;	;2	2+	3+	X–	;2	;2–	3	4
Wichita	3+	4	4	4	4	4	4	3+	4

[a] The seedling ITs are 0 = no uredinia or other macroscopic sign of infection, ; = no uredinia but small hypersensitive necrotic or chlorotic flecks present, 1 = small uredinia surrounded by necrosis, 2 = small to medium uredinia surrounded by necrosis or chlorosis (green island may be surrounded by necrotic or chlorotic border), 3 = medium uredinia with or without chlorosis, 4 = large uredinia without chlorosis, X = heterogeneous (random distribution of variable-sized uredinia on a single leaf), C = more chlorosis than normal for the IT, + = uredinia somewhat larger than normal for the IT, and – = uredinia somewhat smaller than normal for the IT. A range of variation between ITs is recorded by indicating the range, with the most prevalent IT listed first (e.g., 0;, 23, or ;1C) (23).

Table 6. Seedling infection types[a] (ITs) of wheat lines containing *Lr* genes transferred to common wheat from *Aegilops tauschii* inoculated with nine races of *Puccinia recondita* f. sp. *tritici*

	Race								
Cultivar or line	**MGML**	**MCDL**	**TBGL**	**MBRL**	**KDBL**	**PNMQ**	**PNML**	**TFGL**	**MCRL**
Lr21[b]	;1C	2;	;2–	;1	;1+	2;	;2	;2–	;1
Lr 22a[b]	3	3	3	3	3	3+	3	3–	3
Lr32[b]	;1C	2	2	2;	2+	2	2;	;2–	2;
Lr39[c]	;1C	;1+	;1–	;1–	;1	;1	;1	;1–	;1
Lr41[d]	;C	;,2	0;	0;	;,3	3+	4	0;	0;
Lr42[e]	;	;1–	0;	0;	2–C	;1C	12C	2–C	0;
Karl 92	2;C	;2	3–	3+	;2	;2	;1+	3	3
Wichita	3+	4	4	4	4	4	4	3+	4

[a] The seedling ITs are 0 = no uredinia or other macroscopic sign of infection, ; = no uredinia but small hypersensitive necrotic or chlorotic flecks present, 1 = small uredinia surrounded by necrosis, 2 = small to medium uredinia surrounded by necrosis or chlorosis (green island may be surrounded by necrotic or chlorotic border), 3 = medium uredinia with or without chlorosis, 4 = large uredinia without chlorosis, X = heterogeneous (random distribution of variable-sized uredinia on a single leaf), C = more chlorosis than normal for the IT, + = uredinia somewhat larger than normal for the IT, and – = uredinia somewhat smaller than normal for the IT. A range of variation between ITs is recorded by indicating the range, with the most prevalent IT listed first (e.g., 0;, 23, or ;1C) (23).

[b] Backcross lines of cv. Thatcher with the indicated gene.

[c] Line KS86WGRC2.

[d] Line KS90WGRC10.

[e] Line KS91WGRC11.

associated with chlorosis (;1C). Interestingly, races PNML and PNMQ were the only ones able to sporulate on KS92WGRC16. Nevertheless, pustules were tiny and few, and the reaction was still very resistant. The line with *Lr43* differed from lines with previously transferred leaf rust resistance genes from *A. tauschii* in ITs and race specificity (Tables 5 and 6). *Lr43* is nonallelic with previously described *A. tauschii* genes (5).

If these new genes are used singly, their usefulness may vary. For example, *Lr43* is highly effective and no preexisting virulence has been detected yet. On the other hand, the resistance genes in germplasm lines KS96WGRC34 and KS93U3 may not be as useful where race TFGL is prevalent. It is difficult to predict which of these two would be more useful when used singly. The gene in KS96WGRC34 provides a lower IT against races other than TFGL, but it also produces excess chlorosis that could affect yields. The resistance gene in KS92WGRC23, although it currently provides excellent resistance in Kansas and Texas, may not be useful where PNML and PNMQ races are prevalent. There is also a risk of increasing the local prevalence of preexisting virulent races if these resistance genes eventually occupy large production areas.

The utility of these genes may be increased greatly if they are used in combinations rather than singly. For example, a combination of the genes in KS92WGRC23 plus KS96WGRC34 should be resistant to both PNMQ and TFGL, whereas separately these lines are defeated by these races. The gene in KS92WGRC23, which confers near immunity, also might prevent the excess chlorosis produced by the gene in KS96WGRC34. Using genes from both KS96WGRC34 and KS93U3 in combination might not be effective, because both lines are vulnerable to TFGL. It would be interesting to know whether the virulence in race TFGL against these lines is conditioned by one or two loci. If there are two loci, genes in KS96WGRC34 and KS93U3 might be used very effectively in combinations against other races.

The utility of *Lr43* also could be optimized by combination with other *Lr* genes. Even though preexisting virulence has not been detected yet for *Lr43*, the pathogen presumably only needs to mutate at one locus to become virulent. However, if *Lr43* were combined with several other effective resistance genes, such as *Lr21*, *Lr39*, *Lr41*, *Lr42*, or the genes in KS92WGRC23 and KS96WGRC34, more durable resistance could result. Durability probably would be enhanced if the genes were not deployed singly in any other cultivars. Release of cultivars with single resistance genes could allow the rust to defeat the genes in a stepwise manner.

Unfortunately, identifying progeny with a three-way combination, such as *Lr43* plus the genes in KS92WGRC23 and KS96WGRC34, would be difficult. Races TFGL and PNMQ could be used to eliminate segregating progeny that contain only KS92WGRC23 or KS96WGRC34. However, progeny with only *Lr43* or any two- or three-way combinations would all appear highly resistant. To construct a combination of several highly effective resistance genes, marker-assisted selection will be needed. We did not detect any useful morphological markers for our four new genes. Therefore, we have initiated gene-mapping studies to eventually identify useful molecular markers.

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