Identification of New Pathogenic Races of Common Bunt and Dwarf Bunt Fungi, and Evaluation of Known Races Using an Expanded Set of Differential Wheat Lines

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Abstract

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Pathogenic races of *Tilletia caries* and *T. foetida*, which cause common bunt of wheat (*Triticum aestivum*), and *Tilletia contraversa,* which causes dwarf bunt of wheat, have been identified previously by their reaction to 10 differential wheat lines, each containing single bunt resistance genes *Bt1* through *Bt10*. The reactions of races to the differential wheat lines follow the classic gene-for gene system for host–pathogen interactions. The pathogens are closely related and resistance to both diseases in wheat is controlled by the same genes. To better define pathogenic races, six additional wheat lines containing the genes *Bt11* through *Bt15* and a wheat line with a resistance factor designated as *Btp* were added to the set of 10 differentials and tested with all named U.S. races of common bunt and dwarf bunt. In addition, new isolates of dwarf bunt, and common bunt from hybrids and field collections, were tested with all 16 differentials for race identification. Six new races of *T. caries*, five new races of *T. foetida*, and two new races of *T. contraversa* were identified. Races of common bunt virulent to *Bt8* or *Bt12*, and dwarf bunt races virulent to the combinations of *Bt11* and *Bt12*, and *Bt8*, *Bt9*, *Bt10*, *Bt11*, and *Bt12,* were identified for the first time. Comparison of the reactions of the common bunt races with the *Bt14* and *Bt15* differentials grown in different environments after initial infection showed that these genes are temperature sensitive, indicating they should be excluded from the set of differential lines to avoid ambiguity in determining virulent or avirulent reactions. In the previous list of bunt races, there were races that had the same reaction to the set of 10 differentials but were designated as different races. These races were not differentiated further with the six additional differentials, indicating that the duplicate races should be dropped from the list of pathogenic races. The new races of common bunt and dwarf bunt identified have unique patterns of virulence that allow specific targeting and elucidation of bunt resistance genes in wheat and will aid the development of bunt-resistant wheat cultivars.

Common bunt of wheat (*Triticum aestivum* L.) caused by *Tilletia caries* (DC.) Tul. & C. Tul. (=*T. tritici*) and *T. foetida* (Wallr.) Liro (=*T. laevis*), and dwarf bunt of wheat caused by *T. contraversa* J.G. Kühn, have a classic gene-for-gene relationship between the host and pathogen (17,25,34). The three bunt species have not been differentiated at the genetic level when a sufficient number of isolates is compared (5,6). Additionally, the close genetic relationship of these three species is indicated by similar polypeptide patterns produced by two-dimensional polyacrylamide gel electrophoresis (29). The three bunt fungi are very closely related, to the extent that virulence in these fungi is regulated by the same set of bunt resistance genes in wheat (25,34). Thus, resistance to common bunt also confers resistance to dwarf bunt. Common bunt is often used by breeders in initial screening of segregating lines for resistance to dwarf bunt because common bunt can be induced more reliably due to less stringent environmental requirements that are easily managed (17).

Despite the close relationship of the three *Tilletia* spp., common bunt and dwarf bunt are distinct diseases that have completely different etiologies and climatic requirements (17). *T. contraversa* teliospore germination requires several weeks of stable cool temperatures that are provided by continuous snow cover. Soilborne teliospores of *T. contraversa* germinate at the soil surface beneath snow during midwinter and produce hyphae that infect emerged seedlings. Seedborne teliospores of *T. contraversa* are essentially inconsequential to development of dwarf bunt (22). In contrast, common bunt is typically induced by seedborne teliospores that contaminate seed. Teliospores germinate within a few days after planting and produce hyphae that infect wheat coleoptiles prior to seedling emergence. Soilborne teliospore inoculum can also be important in common bunt infection. Inoculum can remain viable for 2 to 3 years; thus, sufficient time is needed between wheat crops in rotations to eliminate teliospore viability.

Even though common bunt and dwarf bunt can be effectively controlled with seed treatment fungicides, incorporating resistance into cultivars is still important in many wheat-breeding programs worldwide. Currently, host resistance has gained renewed interest with the increase in organic wheat production (33). Knowledge of bunt resistance genes in wheat and their reaction to races of the pathogens is essential to this effort.

A set of differential wheat lines has been used since the mid-1940s to evaluate virulence characteristics of common bunt and dwarf bunt pathogens (36). More recently, pathogenic races of these bunt fungi are identified by the virulent or avirulent reactions to a set of putative monogenic differential wheat cultivars developed by R. J. Metzger and J. A. Hoffmann (17,25,34). Races of *T. caries*, *T. foetida*, and *T. contraversa* are designated by the letters "T", "L", and "D", respectively, followed by a number to indicate the specific race. Thus far, there have been 30 races of *T. caries*, 10 races of *T. foetida*, and 17 races of *T. contraversa* identified based on their reactions to a set of 10 wheat differential cultivars containing the resistance genes *Bt1* through *Bt10* (25,34).

The set of 10 differential lines used by Hoffmann and Metzger (25) has evolved and expanded over the years to include five additional wheat lines developed by R. J. Metzger that carry the resistance genes *Bt11* through *Bt15* (17). The *Bt1* to *Bt13* differentials are winter hexaploid wheat, whereas the *Bt14* and *Bt15* differentials are spring tetraploid (durum) wheat. In addition, the winter landrace PI 173437, which has a reaction to races that is independent of the other differentials, has been utilized and given the designation *Btp* (R. J. Metzger, *personal communication*).

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The set of 15 differential cultivars (*Bt1* to *Bt15*) has been used throughout the world to evaluate the virulence characteristics of local wheat bunt isolates (1–4,7–9,14,26,27,30,31,38,42). Virulence characteristics of common bunt fungi that are different than those in the United States have been reported occasionally (1,8,27,38) and new pathotype designations have been suggested; however, these were based on limited testing or undefined methods (1,4,9,27,31). The most notable difference between U.S. and non-U.S. isolates of common bunt is the virulence detected to *Bt8* in Indian isolates (38) whereas no known natural collections of common bunt in the United States have virulence to this gene. Also, there are reports of some isolates that lack virulence to any of the differentials (1,4). Finci (13) used a different set of differential cultivars and reported considerable variation of virulence characteristics in common bunt isolates from Turkey. Recently, a new virulence combination in *T. caries* has been identified in North America using the set of differentials with the resistance genes *Bt1* to *Bt13* (32).

The last reports of pathogenic races of common bunt and dwarf bunt fungi in the United States were published in the 1970s and were based on the set of 10 differentials (25,34). A summary of an unpublished preliminary study of the reaction of the common bunt races to the newer differentials *Bt11* through *Bt15* that was conducted by R. J. Metzger has been presented (17) but there is no information on the reaction of dwarf bunt races to these newer differentials, or on the reaction of common bunt races to *Btp*.

Testing new dwarf bunt and common bunt isolates in the United States for race identification declined after the retirement of J. A. Hoffmann and R. J. Metzger in mid-1980. However, the analysis of bunt isolates has continued in an effort to find isolates that have virulence characteristics that can assist in the elucidation of resistance genes effective against both common bunt and dwarf bunt. The objectives of this study were to analyze the reaction of the previously named pathogenic races of *T. caries*, *T. foetida*, and *T. contraversa* to an expanded set of bunt differentials, and to describe unique patterns of virulence of new bunt isolates to these differentials.

Materials and Methods

Wheat lines. Sixteen differential wheat lines that contain the bunt resistance genes *Bt1* to *Bt15* and *Btp* were used in this study (Table 1). The differentials with genes *Bt1* through *Bt13* and *Btp* are winter hexaploid wheat, whereas the differentials with *Bt14* and *Bt15* are spring tetraploid (durum) wheat. Seed of the wheat originated from R. J. Metzger, who was primarily responsible for the development of the differentials. The winter wheat 'Heines VII' and spring wheat 'Red Bobs' were used as universally susceptible lines (designated in previous work as *Bt0*) in fall-planted and spring-planted field nurseries, respectively, to indicate the disease pressure in each experiment. Red Bobs was most often used for tests conducted in the greenhouse because it is better adapted for growth under greenhouse conditions.

Table 1. Wheat lines with specific bunt resistance genes (*Bt*) used to determine pathogenic races of common bunt and dwarf bunt fungi, and wheat lines with no known resistance genes that were used as positive controls

Wheat line[a]	Resistance gene	CI or PI number
Red Bobs	None	CI 6255
Heines VII	None	PI 209794
Sel 2092	*Bt1*	PI 554101
Sel 1102	*Bt2*	PI 554097
Ridit	*Bt3*	CI 6703
CI 1558	*Bt4*	PI 11610
Hohenheimer	*Bt5*	CI 11458
Rio	*Bt6*	CI 10061
Sel 50077	*Bt7*	PI 554100
PI 173438/Eg	*Bt8*	PI 554120
Elgin/PI 178383	*Bt9*	PI 554099
Elgin/PI 178383	*Bt10*	PI 554118
Elgin/PI 166910	*Bt11*	PI 554119
PI 119333	*Bt12*	PI 119333
Thule III	*Bt13*	PI 181463
Doubbi	*Bt14*	CI 13711
Carleton	*Bt15*	CI 12064
PI 173437	*Btp*	PI 173437

[a] Wheat lines are hexaploid winter growth habit except Red Bobs, which is a spring habit hexaploid, and Doubbi and Carleton, which are spring habit tetraploid (durum).

Pathogens. The bunt isolates used in this study consisted of (i) named pathogenic races of common bunt fungi T-1 through T-30 (*T. caries*) and L-1 through L-16 (*T. foetida*), and of dwarf bunt fungi D-1 through D-17 (*T. contraversa*), originating from the collections of R. J. Metzger, J. A. Hoffmann, and the author; (ii) domestic and international isolates of dwarf bunt and common bunt fungi from field collections; and (iii) isolates of common bunt fungi that were developed by R. J. Metzger from hybridizing known pathogenic races (Tables 2–5). The hybrids were developed by inoculating wheat seed with teliospores or, in some cases, compatible haploid cultures of two pathogenic races followed by selection for new combinations of virulence (25,34). Some hybrids were virulent to certain bunt resistance genes even though neither parent was virulent to the genes. The mechanism for this has not been studied.

R. J. Metzger provided isolates of common bunt that had unique virulence characteristics in initial analyses; 38 originated from the hybrids and 12 were from field collections that originated in Turkey. Twelve of the hybrids and five isolates from field collections that indicated virulence characteristics valuable for elucidating resistance genes in wheat were chosen for further analysis. Six of these were excluded from further tests after showing the same reaction to the differentials as that of known races or other test isolates (Table 3). These common bunt isolates were tested periodically over a 10-year period on all the differentials in fall-planted field nurseries. The isolates selected for analysis were also included in spring-planted field nurseries and in tests conducted in a greenhouse.

Two isolates from field collections of *T. contraversa* that had unique and valuable virulence combinations in initial tests were

Table 2. Named pathogenic races and other isolates of *Tilletia contraversa* from field collections throughout the northwestern United States that were tested for reaction to wheat lines containing bunt resistance genes *Bt11* through *Bt15* and *Btp*

Accession number	Race	Origin
32	D-1	Nephi, UT
29	D-2	Pullman, WA
379	D-3	Kalispell, MT
22	D-4	Genesee, ID
36	D-5	Mt. Hope, WA
245	D-6	Antone, WA
40	D-7	Logan, UT
44	D-8	Preston, ID
288	D-9	Tetonia, ID
108	D-10	Blind Spring, UT
260	D-11	Hill City, ID
36	D-12	Mt. Hope, WA
202	D-13	Paris, ID
265	D-14	Petersboro, UT
375	D-15	Preston, ID
216	D-16	Preston, ID
161	D-17	Blind Spring, UT
638	Unknown	San Juan County, UT
688	Unknown	Hayden, CO
694	Unknown	Benington, ID
695	Unknown	Driggs, ID
697	Unknown	Bozeman, MT
701	Unknown	Reardon, WA
703	Unknown	Creston, MT
715	Unknown	Worley, ID
756	Unknown	Haines, OR
759	Unknown	Flora, OR
761	Unknown	Farmington, WA
770	Unknown	Paradise, OR

evaluated further with the differentials. One isolate originated from Turkey and the other originated from Franklin County, ID and showed virulence to cultivars that were resistant to other known races (20).

The named races and other isolates of common bunt and dwarf bunt fungi were maintained as diseased spikes stored at room temperature. Teliospores in sori maintained under these conditions can retain viability for over 10 years. Pathogenic races that were less than 10 years old were increased on the differentials to validate the correct race reaction. Diseased spikes were collected from the differential that best discriminated the race from others. For example, if only one race is virulent to a specific resistance gene, or if the race is virulent to a gene that rarely confers susceptibility to other races, the bunted spikes were harvested from the differential that carries that gene.

Inoculation—common bunt. Seed for all common bunt tests was inoculated with a concentrated suspension of teliospores in 5% aqueous methyl cellulose. The methyl cellulose adhered the spores to the seed, which helped avoid cross-contamination of adjacent plots during planting. For small quantities of seed (up to 10 g), a few drops of the teliospore suspension was mixed with seed in a glass vial using a vortexer. For larger quantities of seed, the suspension was shaken with seed by hand in an Erlenmeyer flask until the suspension was taken up by the seed, and the seed was evenly and visibly darkened with teliospores.

Field tests—common bunt. Spring-planted and fall-planted field tests with common bunt fungi were performed at the University of Idaho Research and Extension Center in Aberdeen, ID. Fields that had not been planted with wheat for at least 3 years were used to ensure that the soil was free of common bunt teliospores, which lose viability under field conditions within that time period.

Fall-planted common bunt field nurseries were seeded during the second to third week of October, when soil temperatures are typically ideal for infection (approximately 5 to 10°C). Seed (5 g) was planted by hand approximately 5 cm deep into 1.5-m rows. Each pathogenic race and isolate of common bunt was tested to all the differentials for 2 to 10 seasons. Data on the reaction of differentials *Bt11 to Bt15* and *Btp* to the named races of common bunt were obtained during routine pathogen increases in field nurseries, where the race was inoculated to all the differentials to confirm correct virulence reaction patterns. In tests where infection in the susceptible control line Heines VII had more than 70% diseased spikes resulting from inoculation with a particular race or isolate, the disease pressure was considered adequate to determine a definitive reaction on the differentials. The virulence reaction of a race or isolate to a particular differential can often be determined even if the positive control has disease levels considerably less than 70% but avirulent reactions under lower disease pressure are questionable. Data from each season were averaged to determine the avirulent–resistant (0 to 10% spikes infected) or virulent–susceptible (11 to 100% spikes infected) reaction of each race or isolate–differential wheat line interaction in accordance with the methods of Hoffmann and Metzger (25). Data from 2 to 8 seasons were used for the analysis of each named race and isolate.

The spring tetraploid differentials 'Doubbi' (*Bt14*) and 'Carleton' (*Bt15*) are not adapted to field conditions during winter, which often resulted in poor or no plant stands in fall-planted nurseries, making it difficult to determine disease reaction. Thus, all named common bunt races and the 17 isolates from R. J. Metzger mentioned above were tested to the *Bt14* and *Bt15* differentials and Red Bobs in spring-planted field nurseries using the same methods as described above for the fall-planted nurseries. Tests of each combination of pathogen isolate and wheat differential were conducted in two replicate 1.5-m rows for four consecutive seasons beginning in 2004. Seed for all the spring-planted nurseries was inoculated in the spring of 2004 and separated into subsets for subsequent tests. Inoculated seed was stored at 5°C to preserve seed and teliospore viability. The spring nurseries were planted as soon as fields were dry enough to cultivate, which occurred approximately the first week of April, to take advantage of cool soil temperatures that are conducive to disease development. Data from three to four seasons

Table 3. Reaction of isolates of common bunt fungi to wheat differentials with specific bunt resistance genes (*Bt*) that were tested in fall-planted field nurseries[a]

			Diseased spikes (%) for each bunt resistance gene														
Isolate[b]	Years[c]	Data[d]	Control[e]	*Bt1*	*Bt2*	*Bt3*	*Bt4*	*Bt5*	*Bt6*	*Bt7*	*Bt8*	*Bt9*	*Bt10*	*Bt11*	*Bt12*	*Bt13*	*Btp*	New[f]
R-36	5	Mean	83.0	0.0	76.0	0.4	3.0	0.0	3.7	66.0	45.0	0.5	0.2	0.0	0.1	84.8	0.0	L-17
T. foetida	...	SD	7.6	0.0	18.5	0.1	3.1	0.0	6.4	24.8	26.2	0.5	0.5	0.0	0.1	19.8	0.0	...
R-40	5	Mean	93.6	70.0	79.0	38.6	7.0	71.3	1.8	60.0	0.1	2.0	0.0	0.0	0.1	1.0	40.1	T-31
T. caries	...	SD	3.5	19.6	20.4	30.0	5.0	23.2	1.5	27.6	0.1	3.1	0.0	0.0	0.1	1.2	23.9	...
R-43	5	Mean	93.7	0.0	54.0	0.1	0.8	47.5	1.3	68.0	68.3	0.2	0.5	0.0	0.0	97.2	0.1	L-18
T. foetida	...	SD	4.2	0.0	32.3	0.1	0.8	13.3	1.8	19.9	16.6	0.4	0.8	0.0	0.0	2.4	0.1	...
R47	3	Mean	83.3	53.3	80.0	22.3	56.7	0.0	66.7	80.0	0.3	60.0	0.0	0.0	0.0	0.0	0.0	T-32
T. caries	...	SD	7.6	10.4	5.0	9.3	15.3	0.0	5.8	5.0	0.6	8.7	0.0	0.0	0.0	0.0	0.0	...
R-53	6	Mean	91.6	0.0	0.0	1.2	1.3	0.3	0.7	77.8	0.0	67.5	0.2	0.0	65.8	0.1	67.0	L-19
T. foetida	...	SD	4.9	0.0	0.0	2.9	1.0	0.8	0.5	23.9	0.0	16.0	0.4	0.0	10.7	0.1	22.2	...
R-54	5	Mean	92.2	63.3	79.2	0.5	83.3	72.5	84.2	64.0	0.0	73.3	67.5	0.0	0.0	0.5	0.0	T-33
T. caries	...	SD	7.2	16.0	13.2	0.8	8.2	13.3	6.6	24.3	0.0	16.9	13.2	0.0	0.0	0.5	0.0	...
R-55	8	Mean	98.0	61.9	81.1	50.0	81.3	0.0	80.0	79.4	0.4	74.4	80.1	0.0	0.6	1.9	46.9	T-34
T. caries	...	SD	1.9	24.3	14.6	14.4	11.9	0.0	10.0	13.5	0.4	15.5	14.5	0.0	0.7	3.0	24.9	...
R-59	3	Mean	96.3	68.3	83.3	40.0	85.0	0.0	66.7	53.3	0.2	80.0	0.0	0.0	0.1	0.0	45.0	T-35
T. caries	...	SD	5.5	29.3	11.5	17.3	10.0	0.0	15.3	15.3	0.1	10.0	0.0	0.0	0.1	0.0	5.0	...
Iso-41	3	Mean	87.0	0.0	0.0	0.0	80.3	0.0	55.0	86.5	0.0	66.7	0.0	0.0	0.0	0.3	0.0	L-20
T. foetida	...	SD	13.9	0.0	0.0	0.0	20.6	0.0	17.3	13.9	0.0	11.5	0.0	0.0	0.0	0.6	0.0	...
Iso-44[g]	2	Mean	77.5	0.0	0.0	0.0	3.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	99.5	0.0	T-36
T. caries	...	SD	3.5	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.5	0.0	...
Iso-51	4	Mean	85.0	50.0	0.0	0.1	3.5	0.0	0.3	47.5	5.3	47.5	0.0	0.1	0.0	0.1	0.0	L-21
T. foetida	...	SD	7.1	14.1	0.0	0.1	4.3	0.0	0.5	29.6	2.6	22.2	0.0	0.1	0.0	0.1	0.0	...

[a] Unique virulence combinations were identified and given new pathogenic race designations.
[b] Isolates of *Tilletia foetida* or *T. caries* labeled "R" are selections from hybrids of known races, and those labeled "Iso" are field-collected isolates originating from Turkey (Iso 41 and Iso 44) and the United States (Iso 51).
[c] Years tested.
[d] Mean and standard deviation (SD).
[e] Heines VII, which has no known resistance genes, was used for the positive control.
[f] New race designation.
[g] An additional isolate tested for 2 years had the same virulence–avirulence pattern and aggressiveness to *Bt13*.

were used to determine the virulent–susceptible or avirulent–resistant reaction of each race and isolate to the *Bt14* and *Bt15* differentials in the spring-planted nurseries.

Greenhouse tests—common bunt. Additional tests were performed to compensate for poor winter survival of the spring wheat by growing inoculated Red Bobs, *Bt14*, and *Bt15* in a greenhouse, where temperatures were adjusted to simulate mild winter conditions. A subset of the inoculated seed prepared for the spring-planted field nurseries was used for the greenhouse tests. Inoculated seed of each differential was sown approximately 4 cm deep into two 15-cm-diameter plastic pots that were maintained in a greenhouse at approximately 5 to 10°C for a minimum of 8 weeks during the winter. The greenhouse was gradually warmed during early spring to approximately 30°C during the day and 18°C during the night. Plants were grown to maturity and the percent diseased spikes in each of the two pots was averaged. The experiment was repeated in each of three separate seasons.

Table 4. Avirulent (A) or virulent (V) reaction of previously named pathogenic races of *Tilletia caries* (T races) and *T. foetida* (L races) to wheat differentials containing the resistance genes *Bt11*, *Bt12*, *Bt13*, and *Btp*[a]

	Bunt resistance gene			
Race	*Bt11*	*Bt12*	*Bt13*	*Btp*
T-1	A	A	*	A
T-2	A	A	A	A
T-3	A	A	*	A
T-4	A	A	A	A
T-5	A	A	A	A
T-6	A	A	A	A
T-7	A	A	A	A
T-8	A	A	A	A
T-9	A	A	A	A
T-10	A	A	A	A
T-11	A	A	A	A
T-12	A	A	A	A
T-13	A	A	V	A
T-14	A	A	V	A
T-15	A	A	A	A
T-16	A	A	A	A
T-17	A	A	A	A
T-18	A	A	V	A
T-19	A	A	*	V
T-20	A	A	V	A
T-21	A	A	V	A
T-22	A	A	V	A
T-23	A	A	A	A
T-25	A	A	A	A
T-26	A	A	A	A
T-27	A	A	*	A
T-28	A	A	A	A
T-29	A	A	*	A
T-30	A	A	A	A
L-1	A	A	A	A
L-2	A	A	A	A
L-3	A	A	A	A
L-4	A	A	A	A
L-5	A	A	A	A
L-7	A	A	*	A
L-8	A	A	A	A
L-9	A	A	A	A
L-10	A	A	V	V
L-16	A	A	A	A

[a] Reaction based on a minimum of two experiments in field nurseries during separate seasons. A = 0–10% infected spikes and V = 11–100%. Race T-24 was lost and, thus, is not included; an asterisk (*) indicates that *Bt13* was not included in sufficient tests to determine reaction.

Greenhouse tests—dwarf bunt. An artificial inoculation procedure was used for *T. contraversa* tests in the greenhouse (17) to avoid complications encountered in field tests. Inoculation in typical dwarf bunt field-screening nurseries is done by spraying a water suspension of teliospores onto the soil surface after planting. Using these methods to test different races of *T. contraversa* in the field is not possible due to potential cross contamination from adjacent soil-inoculated plots. There could also be residual inoculum in soils contaminated with long-lived teliospores in disease-conducive areas. Moreover, inoculating seed with *T. contraversa* teliospores prior to planting (as is done to induce common bunt) has very little or no effect on the induction of dwarf bunt (22). The artificial inoculation procedure that was used to induce dwarf bunt involved placing approximately 3 g of seed of each differential into a 9-cm petri dish containing teliospores germinating on agar, followed by adding approximately 3 ml of water and then mixing with a clean finger until seed was uniformly wet with a suspension of germination products consisting of fused primary sporidia, secondary sporidia, and hyphae. Inoculated seed was sown 5 cm deep into moist vermiculite in an 11-cm-diameter plastic pot which was then covered with foil. Pots were incubated at 10°C for approximately 2 weeks; then, about 12 seedlings were transplanted into soil in each of two 15-cm-diameter pots (17). Plants were maintained in the greenhouse at 5 to 10°C during winter for a minimum of 8 weeks. The cool greenhouse temperatures simulated mild winter conditions and vernalized the winter wheat. The greenhouse was gradually warmed to 30°C during the day and 18°C during the night during early spring. Plants were grown to maturity, after which the percent diseased spikes in each of the two pots was averaged.

The artificial inoculation method was used to test the reaction of the susceptible control Red Bobs and the *Bt11* to *Bt15* differentials to all the named pathogenic races of *T. contraversa* (D-1 to D-17). In addition, 11 isolates of *T. contraversa* from field collections that originated from the widest possible geographic area in the northwestern United States (Table 2) were included in the same tests to examine the potential virulence of additional isolates to the newer differentials. Tests were conducted over five winter seasons. Data from a minimum of three seasons where infection in Red Bobs was 70% or higher were averaged to determine the avirulent or virulent reaction of the races and isolates to *Bt11* to *Bt15*. Infection of the *Btp* differential induced by the named races was recorded after artificial inoculation in a minimum of two tests performed during race increases with the differentials, as described for the common bunt race increases.

Table 5. Reaction of isolates of *Tilletia contraversa* to wheat differentials with specific bunt resistance genes (*Bt*) after artificial inoculation and growth in a greenhouse adjusted to simulate mild winter conditions[a]

			Diseased spikes (%) for each bunt resistance gene														
Isolate[b]	Years[c]	Data[d]	Control[e]	*Bt1*	*Bt2*	*Bt3*	*Bt4*	*Bt5*	*Bt6*	*Bt7*	*Bt8*	*Bt9*	*Bt10*	*Bt11*	*Bt12*	*Bt13*	*Btp*	New[f]
Prom	5	Mean	64.2	54.2	91.7	30.7	37.8	0.5	77.5	41.7	16.2	63.0	68.3	1.7	1.5	21.8	62.5	D-18
	…	SD	15.0	20.8	8.1	13.7	21.4	1.2	14.7	25.7	11.9	19.1	23.8	1.9	1.5	20.4	19.9	…
TR29-1	4	Mean	48.3	0.0	76.8	42.5	65.0	23.3	86.3	51.7	85.0	38.3	73.8	38.3	66.3	85.5	48.3	D-19
	…	SD	12.6	0.0	25.5	15.0	12.9	11.0	4.8	14.4	5.8	17.3	6.3	2.9	11.8	9.9	36.2	…

[a] Unique virulence combinations were identified and given new race designations.
[b] Isolates originated from field collections in Idaho (Prom) and Turkey (TR29-1).
[c] Years tested.
[d] Mean and standard deviation (SD).
[e] Heines VII, which has no known resistance genes and is poorly adapted to greenhouse conditions, was used for the positive control.
[f] New race designation.

In initial tests, two isolates of dwarf bunt from field collections indicated unique patterns of virulence to the differentials. These isolates were analyzed further on all of the differentials in tests over a period of 4 to 5 years. Infection data from each test were averaged to determine the virulent or avirulent reaction of the isolates to the differentials.

Uninoculated seed of each wheat line tested was included in all field and greenhouse tests to ensure that any bunt produced was a result of the inoculation. In all tests, a reaction was considered avirulent when 10% or less of the spikes were diseased, and virulent if the disease exceeded 10% (25).

Results and Discussion

Reaction of *Bt14* and *Bt15* to common bunt isolates. Comparison of the results of common bunt tests of the spring tetraploid differentials Doubbi (*Bt14*) and Carleton (*Bt15*) with the named races and the additional isolates in fall-planted and spring-planted field tests and greenhouse tests demonstrated that the environment often influenced bunt incidence in these differentials. The same overall level of disease pressure occurred in spring and winter tests, where the mean bunt infection in the Red Bobs and Heines VII positive control lines inoculated with all isolates averaged 60.0 and 60.2% bunted spikes, respectively. However, of the 51 isolates tested, 24 isolates which were avirulent to either *Bt14* or *Bt15* or both *Bt14* and *Bt15* in the spring field tests were virulent to the differentials when tested under simulated winter conditions in the greenhouse and in fall-planted field tests. One isolate had virulence to *Bt15* in spring-planted nurseries but was avirulent in the simulated winter conditions. The remaining 26 isolates had the same avirulent or virulent rating in spring and greenhouse winter tests. For 15 of the 51 isolates tested in the fall-planted field nurseries, there was insufficient data to determine the virulent or avirulent reaction of the isolates to *Bt14* and *Bt15* due to poor plant stands in these spring wheat lines. In tests with the remaining 36 isolates that had adequate plant stands in fall-planted nurseries, the virulent or avirulent reactions to *Bt14* and *Bt15* always matched those from the simulated winter conditions in the greenhouse. This indicates the resistance genes in Doubbi (Bt14) and Carleton (Bt15) have sensitivity to temperature or other environmental factors that renders them less effective under winter conditions with specific bunt races or isolates.

Environmental influences on the effectiveness of bunt resistance has been reported previously, where it was shown that resistance in certain wheat lines is less effective when plants are grown in extensive cool conditions (15,23,37,43). Similarly, in recent race tests conducted by Matanguihan and Jones (32) with common bunt isolates in fall- and spring-planted nurseries, there was too much variability from year to year to accurately determine the reaction of the isolates to *Bt14* and *Bt15*.

The initial tests with common bunt conducted by R. J. Metzger in relatively mild winter field environments near Corvallis, OR showed that only race L-7 was virulent to *Bt14* and only races T-2 and L-2 were virulent to *Bt15* (17). In the current tests, conducted in the colder winter environments of southeastern Idaho, 16 of the 51 isolates tested had virulence to *Bt14* and 48 isolates had virulence to *Bt15*. Environmental variation might account for this difference. Although the resistance factors in Doubbi (*Bt14*) and Carleton (*Bt15*) may indeed represent valid resistance genes, the reaction seems too variable in different environments to accurately assess virulence or avirulence. It is not known if the *Bt14* gene named in 'Eryth-5221' by Liatukas and Ruzgas, (30) is the same as the *Bt14* gene of Doubbi used here. Pathogenic race tests with Doubbi and Carleton that may be conducted in the different areas of the world, and variable environments within them, can make race determination with these two differentials unreliable. In addition, utilizing these spring wheat lines makes comparison with the remainder of the differentials in the same field experiments problematic, because the rest are winter wheat lines. Thus, although the *Bt14* and *Bt15* differentials were included in all tests conducted here with common bunt and dwarf bunt fungi, the specific ratings of these differentials to the races are not presented to avoid giving potentially misleading information. Further, it is proposed that Doubbi and Carleton be dropped from the set of bunt differentials.

Common bunt field screening of new isolates. Testing the 17 hybrids and isolates of common bunt fungi provided by R. J. Metzger to all the differential wheat lines under high disease pressure in fall-planted field nurseries demonstrated that 11 of them had unique patterns of virulence to the differentials. Six new pathogenic races of *T. caries* and five new races of *T. foetida* were identified. These isolates were given new race designations that expanded the list of named pathogenic races (Table 3). The remaining six isolates had the same virulence pattern as known races or other test isolates after 2 to 3 years of tests, and were excluded from further analysis. The new races will assist the elucidation of resistance genes by examining the reaction of resistant wheat to several races.

None of the previously named races of common bunt fungi were virulent to *Bt8*, *Bt11*, or *Bt12* (17,25,34). However, in the current tests, two of the new races, L-17 and L-18, had virulence to *Bt8* and one, L-19, had virulence to *Bt12*. *Bt11* remained uncompromised by any of the new common bunt races identified in this study. *Bt8* and *Bt12* are the primary genes responsible for resistance to bunt among cultivars that are commonly grown in the Pacific Northwestern United States *Bt8* originated from PI 178383 and *Bt12* originated from CI 14106 (16,21). The new races virulent to *Bt8* and *Bt12* will enable specific targeting of these resistance genes in wheat screening tests.

The new race T-34 had virulence to nine of the differentials (Table 3). This represents the highest number of genes compromised by any common bunt race reported. Race T-34 had the same pattern of virulence and avirulence to the resistance genes *Bt1* through *Bt13* as that described recently for an isolate of *T. caries* identified in the northwestern United States (32). Similar to T-34, this isolate also showed virulence to *Btp* based on limited tests (Matanguihan, *personal communication*), indicating that the isolate is the same race as race T-34 described here. Race T-35 had virulence to one fewer gene (*Bt10)* than T-34. Screening wheat lines with these two races would indicate the presence of *Bt10*, if the wheat is susceptible to T-34 but resistant to T-35.

Race T-36 had virulence to only one differential, *Bt13*. An additional isolate, Iso47 (*data not shown*), was tested for 2 years during these experiments and had the same pattern of aggressive virulence, with an average of 77.5% bunted spikes in the susceptible Heines VII, 97.5% in *Bt13*, and 0 to 1.6% in the other differentials. Of the common bunt disease reactions between individual race–differential combinations that occurred in tests with the 11 new common bunt races identified, the virulent or avirulent reaction to the *Bt13* differential was more extreme than the reaction in the other differentials (Table 3). Depending on the race of the inoculum, *Bt13* was either highly effective, causing complete or nearly complete disease resistance, or ineffective, which resulted in 100% diseased spikes in many individual tests. The average mean bunt infection of *Bt13* produced by the virulent races was 93.8%, whereas the avirulent races produced an average mean infection of 0.5%. In the same tests, Heines VII averaged 84.7 and 90.8% diseased spikes when inoculated with races having virulence and avirulence, respectively, to *Bt13*, demonstrating a near equal level of disease pressure. The level of the virulence of the other races to individual *Bt* genes other than *Bt13* was less severe and had an average bunt infection that ranged between 36.3 and 75.9% diseased spikes. The extremely divergent reaction of these new races to *Bt13* indicates that the interaction would be a good candidate for studies of the microbiology or molecular biology of resistance gene expression and function in host–pathogen interactions. The best race for such studies might be T-36, given that the race has virulence only to *Bt13*, eliminating the potential influence of other virulence factors. This could be compared with the reaction with races such as T-1, T-14, or L-1 that are avirulent to *Bt13* and virulent to only one of the other *Bt* genes.

In the tests conducted here, the universally susceptible Heines VII had a relatively consistent high level of infection in tests over several years whereas, in the same tests, some race–differential interactions had considerable year-to-year variation in disease severity, as indicated by a high standard deviation (SD) relative to the mean percent infected spikes (Table 3). This suggests that certain race–host combinations are more susceptible to environmental influences than other combinations, which had a low SD.

Reaction of previously named races of common bunt fungi to *Bt11, Bt12*, *Bt13*, and *Btp*. The reaction of the named races of common bunt fungi to *Bt11*, *Bt12*, *Bt13*, and *Btp* that was recorded in a minimum of 2 years of fall-planted field tests during pathogen increases are presented as avirulent or virulent in Table 4. Like the preliminary tests (17), no previously named race of common bunt was virulent to *Bt11* or *Bt12* in the current experiments. However, in the previous tests, only a single common bunt race, T-13, was virulent to *Bt13* whereas, in the current experiments, virulence to *Bt13* was detected in T-13, T-14, T-18, T-20, T-21, T-22, and L-10. Thus, the reaction of the previously named races of common bunt fungi to *Bt13* has been updated (Table 4). Data that lead to conclusions of the avirulence or virulent reaction of *Bt13* in the previous report (17) have not been presented and were based on a personal communication (R. J. Metzger). Thus, the reason for the discrepancy in the current and previous report can only be speculative. However, the current experiments describe disease reactions of *Bt13* to the races in repeated experiments under high disease pressure, which should be conclusive.

Table 6. Virulence formula summary of the reaction of 14 wheat lines with specific bunt resistance genes (*Bt*) to pathogenic races of *Tilletia* caries ("T" races), *T. foetida* ("L" races), *and T. contraversa* ("D" races)

Race	Virulence or avirulence to *Bt* genes[a]
T-10	5/1,2,3,4,6,7,8,9,10,11,12,13,p
L-1	7/1,2,3,4,5,6,8,9,10,11,12,13,p
T-1	7/1,2,3,4,5,6,8,9,10,11,12,13*,p
T-36	13/1,2,3,4,5,6,7,8,9,10,11,12,p
T-2, L-4	1,7/2,3,4,5,6,8,9,10,11,12,13,p
T-14	1,13/2,3,4,5,6,7,8,9,10,11,12,p
T-11	2,3/1,4,5,6,7,8,9,10,11,12,13,p
L-3	2,7/1,3,4,5,6,8,9,10,11,12,13,p
T-3	2,7/1,3,4,5,6,8,9,10,11,12,13*,p
T-9	5,7/1,2,3,4,6,8,9,10,11,12,13,p
D-1	7,10/1,2,3,4,5,6,8,9,11,12,13,p
L-9	1,2,3/4,5,6,7,8,9,10,11,12,13,p
T-5, L-5, D-4	1,2,7/3,4,5,6,8,9,10,11,12,13,p
T-20	1,2,13/3,4,5,6,7,8,9,10,11,12,p
T-12	1,5,7/2,3,4,6,8,9,10,11,12,13,p
L-21	1,7,9/2,3,4,5,6,8,10,11,12,13,p
T-25	1,7,10/2,3,4,5,6,8,9,11,12,13,p
T-17, D-2	4,6,7/1,2,3,5,8,9,10,11,12,13,p
T-13	1,2,3,13/4,5,6,7,8,9,10,11,12,p
T-15	1,2,5,7/3,4,6,8,9,10,11,12,13,p
T-26	1,2,7,10/3,4,5,6,8,9,11,12,13,p
L-10	2,3,7,13,p/1,4,5,6,8,9,10,11,12
D-10	2,3,7,p/1,4,5,6,8,9,10,11,12,13
L-17	2,7,8,13/1,3,4,5,6,9,10,11,12,p
L-20	4,6,7,9/1,2,3,5,8,10,11,12,13,p
D-11	4,6,7,10/1,2,3,5,8,9,11,12,13,p
L-19	7,9,12,p/1,2,3,4,5,6,8,10,11,13
T-19	1,2,3,7,p/4,5,6,8,9,10,11,12,13*
D-8	1,2,3,7,p/4,5,6,8,9,10,11,12,13
L-16, D-6	1,2,4,6,7/3,5,8,9,10,11,12,13,p
T-29	1,2,7,9,10/3,4,5,6,8,11,12,13*,p
D-5	1,4,6,7/2,3,5,8,9,10,11,12,13,p
D-12	1,4,6,7,10/2,3,5,8,9,11,12,13,p
T-18	1,4,6,7,13/2,3,5,8,9,10,11,12,p
T-16	2,4,5,6,7/1,3,8,9,10,11,12,13,p
T-28, L-8	2,4,6,7,9/1,3,5,8,10,11,12,13,p
T-22	2,4,6,7,13/1,3,5,8,9,10,11,12,p
L-18	2,5,7,8,13/1,3,4,6,9,10,11,12,p
T-21	1,2,4,6,7,13/3,5,8,9,10,11,12,p
T-31	1,2,3,5,7,p/4,6,8,9,10,11,12,13
T-23	1,2,4,6,7,9/3,4,8,10,11,12,13,p
T-27	1,2,4,6,7,10/3,5,8,9,11,12,13*,p
D-14	2,3,4,6,7,10,p/1,5,8,9,11,12,13
D-17	2,3,7,9,13,p/1,4,5,6,8,10,11,12
T-32	1,2,3,4,6,7,9/5,8,10,11,12,13,p
D-3	1,2,3,4,6,7,p/5,8,9,10,11,12,13
D-16	1,2,3,7,9,13,p/4,5,6,8,10,11,12
T-30	1,2,4,6,7,9,10/3,5,8,11,12,13,p
T-33	1,2,4,5,6,7,9,10/3,8,11,12,13,p
D-13	1,2,4,6,7,10,13/3,5,8,9,11,12,p
T-35	1,2,3,4,6,7,9,p/5,8,10,11,12,13
T-34	1,2,3,4,6,7,9,10,p/5,8,11,12,13
D-15	1,2,3,4,6,7,10,13,p/5,8,9,11,12
D-7[b]	2,3,4,6,7,11,12,13,p/1,5,8,9,10
D-18	1,2,3,4,6,7,8,9,10,13,p/5,11,12
D-19	2,3,4,5,6,7,8,9,10,11,12,13,p/1

[a] An asterisk (*) indicates that the avirulent reaction of *Bt13* needs confirmation.

[b] Virulent reaction to *Bt11* and *Bt12* with race D-7 was slight.

The reaction of the previously named common bunt races to *Btp* was identified for the first time and showed that only two races, T-19 and L-10, had virulence to *Btp* (Table 4). Of the new common bunt races, 4 of the 11 had virulence to *Btp*. Other common bunt isolates tested to *Btp* outside the United States have not detected virulence to this differential (26,42). Similarly, virulence to *Bt11* and *Bt12* has not been detected in common bunt isolates that have been tested to these differentials in various regions of the world (1,4,9,12,26,28, 30,31,42), indicating the potential broad applicability of these genes in resistance breeding. The resistance gene *Bt11* in the wheat PI 166910 that was used to develop the *Bt11* differential continues to be the most difficult bunt resistance gene to overcome. The resistance from PI 166910 has not been utilized for dwarf bunt or common bunt resistance in cultivar development even though it appears to have considerable value for such purpose. Only a single race of common bunt (new race L-19) had virulence to *Bt12*, and it was developed through artificial hybridization. *Bt12* has been highly effective for control of dwarf bunt in winter wheat cultivars in the United States that are grown in areas that are prone to the disease. *Bt12* was introgressed from either CI 14106 or PI 476214 that have been demonstrated to carry the gene (B. J. Goates, *unpublished*).

In the last update of pathogenic races of common bunt in 1976 (25), some races were designated as different races, even though they had the same pattern of virulence to the 10 differentials containing *Bt1* to *Bt10*, indicating that they may be the same pathogenic race. These duplicate races can be separated into four groups with identical virulence–avirulence patterns: (i) T-2, T-4, and T-6; (ii) T-5, T-7, and T-8; (iii) L-1 and L-2; and (iv) L-5, L-6, and L-7. In the current study, testing these races with the four additional differentials containing the *Bt11*, *Bt12*, *Bt13*, and *Btp* genes also did not differentiate the races within the groups from each other, as is shown in a summary of the reaction of the differentials to all the races in Table 6. Thus, it seems appropriate to retain the race designations T-2, T-5, L-1, and L-5 and drop their duplicates (T-4 and T-6, T-7 and T-8, and L-2, respectively) from the list of common bunt races unless new differentials can be identified to differentiate these isolates from each other. Reactions for L-6 are not included in the results here because race L-6 was dropped as a duplicate race prior to these experiments. The reaction of L-7 to *Bt13* was inconclusive; thus, it should be retained and considered a duplicate to L-5 until the reaction to this gene can be determined.

Race identification of new *T. contraversa* isolates. The two *T. contraversa* isolates that were tested with the differentials showed unique virulence–avirulence patterns and were given the new race designations D-18 and D-19 (Table 5). D-18 had virulence to the combination of *Bt9* and *Bt10*, which is the first documentation of this virulence combination in a race of *T. contraversa*. The virulence of D-18 to *Bt9* and *Bt10* in addition to virulence to *Bt3* appears to be responsible for overcoming the dwarf bunt resistance reported previously (19) in cultivars such as 'Hansel', 'Manning', 'Promontory', and 'Utah 100' that had been highly resistant since the form of resistance in these wheat lines was first released in cultivars during the mid-1970s and early 1980s (10,11). The weak

virulent reaction of D-18 to *Bt8* varied considerably in different tests, as indicated by the high SD in infection rates. The virulence of D-18 to *Bt8* does not appear to be important in the susceptibility of the previously resistant cultivars to D-18 because the new common bunt race T-34 produced a high level of disease in these cultivars but lacks virulence to *Bt8* (20). Testing breeding lines with race T-34 has been valuable for eliminating susceptibility to D-18 (B. J. Goates and D. Hole, *unpublished*).

The new race D-19 was virulent to all the *Bt* genes, with the exception of *Bt1*. This is by far the highest level of virulence recorded for any race of *T. contraversa*, *T. caries*, or *T. foetida*. D-19 also produced high levels of disease on the differentials that carry the genes *Bt8*, *Bt11*, and *Bt12* that are either rarely compromised (*Bt8* and *Bt12*) or not compromised (*Bt11*) by common bunt races (Table 5). This is the first time any race of common bunt or dwarf bunt has shown significant virulence to *Bt11*, which had an average infection rate of about 40% after inoculation with D-19. The landrace wheat PI 178383 has the resistance genes *Bt8*, *Bt9*, and *Bt10* and unidentified factors (24). PI 178383 is commonly included in all common bunt and dwarf bunt race evaluations and screening nurseries conducted by the author and in previous tests conducted by Hoffmann and Metzger and has been extremely resistant or immune to all bunt races and isolates in numerous tests (B. J. Goates, *unpublished*). However, PI 178383 was included in the current race evaluations and was usually susceptible to race D-19. In one test, PI 178383 had 40% infected spikes (*data not shown*). This is the first time that this wheat has shown susceptibility to common bunt or dwarf bunt fungi. The new races of *T. controversa* have been valuable for use in disease screening tests to elucidate the genes of resistant wheat and to discover new sources of resistance (B. J. Goates, *unpublished*). The Heines VII positive control in the dwarf bunt greenhouse tests had a relatively low mean infection of about 50 to 60%. However, the high levels of infection among other differentials in the tests indicated that the moderate disease reaction of Heines VII may be due to poor agronomic performance under the greenhouse conditions, previously noted, rather than disease escape. Numerous additional dwarf bunt isolates have been taken through preliminary pathogenic race analyses and have indicated new virulence combinations (B. J. Goates, *unpublished*), suggesting that there is still considerable diversity of virulence among *T. controversa* collections yet to be described.

Reaction of named races and isolates of *T. contraversa* to *Bt11*, *Bt12*, *Bt13*, and *Btp*. A high level of disease pressure occurred in greenhouse tests when the previously named *T. contraversa* races and the additional isolates were tested to the newer differentials, which clearly defined their virulent or avirulent reactions to the newer differentials (Table 7). *Bt11* and *Bt12* were resistant to all of the previously named races and isolates of *T. contraversa* except race D-7, which induced a weak virulent reaction with a mean of 14.5 and 13.0% infected spikes, respectively. This is compared with the reaction of *Bt11* and *Bt12* to the new race D-19, which had a fairly strong virulent reaction to *Bt11* (mean 38.3%) and *Bt12* (mean 66.3%). The average bunt infection produced by D-7 to *Bt11* and *Bt12* just exceeded the defined infection level of 10% for a virulence rating. A considerable amount of variation occurred in the disease reaction of *Bt11* when inoculated with D-7 in the five experiments performed with this race, as is reflected in the high SD. Some reactions of *Bt11* in tests with D-7 were well above that required for a virulence rating. In two tests, infection exceeded 30% but, in two other tests, the infection was 0 and 2%, which indicated avirulence. Repeated testing was required to elucidate the virulent reaction even though all the tests had 80 to 100% infection in the Red Bobs positive control. As mentioned by Matanguihan et al. (33), year-to-year variation in reaction to resis-

Table 7. Disease reaction of pathogenic races (D) and numbered isolates of *Tilletia contraversa* to wheat differentials with specific resistance bunt genes (*Bt*) after artificial inoculation

	Diseased spikes (%) for each bunt resistance gene[a]								
	Control[b]		*Bt11*		*Bt12*		*Bt13*		
Race, isolate	Mean	SD	Mean	SD	Mean	SD	Mean	SD	*Btp*[c]
D-1	89.4	11.1	0.0	0.0	0.0	0.0	2.5	3.8	A
D-2	97.8	1.9	0.0	0.0	0.0	0.0	2.5	1.9	A
D-3	93.6	7.9	0.0	0.0	0.0	0.0	1.0	1.2	V
D-4	85.0	11.7	0.0	0.0	0.0	0.0	3.0	3.8	A
D-5	89.8	10.5	0.0	0.0	0.0	0.0	2.3	3.3	A
D-6	86.3	7.5	0.0	0.0	0.0	0.0	2.0	3.5	A
D-7	86.7	8.2	14.5	14.7	13.0	2.6	27.8	24.5	V
D-8	96.3	4.2	1.0	1.7	0.0	0.0	2.3	2.5	V
D-9	91.3	7.5	0.0	0.0	0.0	0.0	0.8	1.5	V
D-10	86.3	15.1	0.5	1.0	0.0	0.0	0.5	1.0	V
D-11	78.8	8.5	0.0	0.0	0.0	0.0	4.3	2.6	A
D-12	92.3	8.4	0.0	0.0	0.0	0.0	3.8	5.7	A
D-13	90.0	7.1	0.0	0.0	0.0	0.0	39.0	29.7	A
D-14	91.0	10.8	0.0	0.0	0.3	0.5	1.3	1.5	V
D-15	81.3	8.5	0.0	0.0	0.0	0.0	31.0	24.3	V
D-16	80.0	14.7	0.0	0.0	0.0	0.0	36.2	27.7	V
D-17	78.8	12.5	0.0	0.0	0.3	0.5	37.3	21.6	V
638	96.0	1.7	0.0	0.0	0.0	0.0	15.3	3.1	…
688	82.5	11.9	0.0	0.0	0.0	0.0	0.3	0.6	…
694	93.8	8.0	0.0	0.0	0.3	0.6	17.0	8.7	…
695	96.2	1.6	0.0	0.0	0.0	0.0	0.0	0.0	…
697	95.8	1.5	0.0	0.0	0.0	0.0	4.0	4.0	…
701	88.8	9.4	0.0	0.0	0.4	0.5	20.0	7.6	…
703	92.2	9.7	0.0	0.0	1.0	1.7	0.3	0.6	…
715	96.4	1.3	0.0	0.0	0.0	0.0	26.0	12.7	…
756	97.0	4.0	0.0	0.0	0.0	0.0	14.3	4.0	…
759	90.8	7.9	0.0	0.0	0.0	0.0	30.3	25.9	…
761	93.5	4.4	0.0	0.0	0.0	0.0	3.0	5.2	…
770	85.0	20.4	0.0	0.0	0.7	1.2	25.7	8.1	…

[a] Mean = mean percent infected spikes in a minimum of three seasonal experiments and SD = standard deviation.
[b] Red Bobs, which has no known resistance genes, was used as the positive control.
[c] Avirulent (A) and virulent (V) reaction to *Btp* was determined during race increases in a minimum of two separate tests; 0–10% infected spikes = A and 11–100% = V. The reaction to *Btp* was not determined for the numbered isolates.

tance genes can occur, requiring repeated testing with high disease pressure before a definitive reaction to the differentials can be determined. Year-to-year variation of the reactions of differentials in race tests has also been observed by Hoffmann (24).

About half of the *T. contraversa* races and isolates had virulence to *Bt13* (Table 7). The extreme disease reaction of *Bt13* that occurred in interactions with the new common bunt races virulent to *Bt13* described above was not apparent in the tests with dwarf bunt races. Although the reaction of *Bt14* and *Bt15* is not listed in the table for reasons given above, all races and isolates were virulent to *Bt14* except D-4, D-8, and D-10; and all were virulent to *Bt15* except D-4 and isolate number 688. The dwarf bunt tests were conducted only in simulated mild winter conditions in the greenhouse; therefore, it is not known whether post-infection temperature influences dwarf bunt infection in *Bt14* and *Bt15* as was shown in the tests with common bunt. The reaction of the named races to *Btp,* which was recorded in separate tests during pathogen increases, showed that about half the races were virulent to this gene (Table 7). It should be kept in mind that the dwarf bunt tests performed here and elsewhere are all based on an artificial inoculation procedure, and that results from the procedure have not been compared directly with tests under natural field conditions after inoculating the soil surface with teliospores.

In the previous analysis of dwarf bunt races (25), the *Bt5* differential used was a hybrid of Elgin and Hohenheimer. However, it was determined later that this hybrid did not fully express the *Bt5* resistance of Hohenheimer, and that U.S. dwarf bunt races lacked virulence to *Bt5* (24). In later studies, workers reverted back to using Hohenheimer as the *Bt5* differential in race tests. The two dwarf bunt races D-3 and D-9 were differentiated from each other in the previous analysis only by their reaction to the *Bt5* hybrid but both are avirulent to Hohenheimer (17). In the current study, these two races also had the same reactions to *Bt11* through *Bt15* and *Btp*. Thus, it is suggested that the designation D-3 be retained and D-9 dropped unless additional differentials are identified that can differentiate D-3 and D-9 from each other.

Concluding remarks. A common set of differential cultivars that is used internationally is essential for comparison of virulence characteristics of bunt isolates and for a common understanding of useful resistance genes in wheat in various areas of the world that can benefit the development of local bunt-resistant cultivars. The winter habit differential cultivar set *Bt1* through *Bt13* and *Btp* developed by R. J. Metzger and J.A. Hoffmann that was used for the current study has a wide international distribution and has been valuable for such purposes. An advantage of using these differentials is that they are morphologically distinguishable from each other for the most part (42), reassuring workers of correct readings. An additional benefit is that the differentials are readily available to international researchers via the United States Department of Agriculture–Agricultural Research Service National Small Grains Collection (NSGC) in Aberdeen, ID. New, useful differentials that might be discovered can be added to those currently used to better define the resistance factors in wheat.

This report illustrates the remarkable level of disease control that can be conferred by single resistance genes where disease can be reduced from over 90% severity to 0%. The vertical resistance offered by certain single bunt-resistance genes in cultivars has been durable under commercial conditions in the United States. In the 1960s, dwarf bunt was a serious production problem in areas where the disease was most prevalent in the United States (24). The problem was solved when breeders utilized the genes from PI 178383 in new cultivar releases in the early 1970s, where the resistance comes primarily from *Bt8* (35,39). This was followed by the release of several other resistant cultivars based on this same resistance, and also the *Bt12* resistance gene that originated from CI 14106 or SM22 (18). Cultivars that have resistance genes *Bt8* or *Bt12* are still highly effective today for the control of bunt and have eliminated losses caused by dwarf bunt where they are commonly grown. However, the narrow genetic basis for resistance in the United States indicates a vulnerability to new races that might develop.

Screening of more than 20,000 winter wheat accessions from the NSGC in annual dwarf bunt screening nurseries over numerous years has identified several wheat landraces that are highly resistant to a composite of pathogenic races of *T. contraversa* (41). Similarly, over 25,000 accessions from the NSGC have also been screened for common bunt resistance using different composites of pathogenic races of common bunt fungi (40). The few lines that remain highly resistant in repeated experiments are then screened to individual races of common bunt and dwarf bunt fungi in an attempt to elucidate the genes responsible for conferring the resistance (21). Such tests have identified wheat lines that carry known and unknown resistance genes or new combinations of resistance genes that could be useful in cultivar development, including wheat lines that are resistant to the highly virulent races described here (B. J. Goates, *unpublished*). The resistance characteristics in these wheat lines will be the subject of a future report. There is no doubt that many virulence factors in these bunt fungi and resistance factors in wheat are unknown. The races of common bunt and dwarf bunt fungi described in these studies that have virulence to previously uncompromised individual resistance genes or gene combinations are particularly valuable for identifying resistance factors in wheat. In addition, examination of the response of resistant wheat to the defined pathogenic races may assist the elucidation of unknown virulence factors in the pathogens.

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