Occurrence of *Puccinia graminis* subsp. *graminicola* in Chewings Fescue in Oregon

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ABSTRACT

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Stem rust was found on Chewings fescue throughout seed production areas of the Willamette Valley of Oregon in 1993. Stem rust developed in greenhouse-grown seedlings of cultivar Jamestown when inoculated with isolates of stem rust collected from three locations in the Willamette Valley. No difference in plant response was observed among isolates. Twelve cultivars of Chewings fescue and four other species of cool season grasses used for turf (red fescue, tall fescue, sheep fescue, and perennial ryegrass) were inoculated with the same mixture of isolates of stem rust and incubated for infection in a controlled environment chamber. Among the cultivars of Chewings fescue inoculated, stem rust pustules developed on 624 of 673 seedlings; 49 seedlings remained free of stem rust. This is believed to be the first report of *Puccinia graminis* subsp. *graminicola* on Chewings fescue in Oregon. *Puccinia g.* subsp. *graminicola* infected 28 of 60 seedlings of red fescue, which was less susceptible than Chewings fescue. Five of 46 seedlings of sheep fescue and one of 60 seedlings of perennial ryegrass became infected by *P. g.* subsp. *graminicola*. Pustules of stem rust did not develop in tall fescue inoculated with this collection of isolates.

Chewings fescue (*Festuca rubra* L. var. *commutata* Gaudin) is a fine-textured, open-pollinated, perennial grass adapted to cool, humid regions of the U.S. It requires moderate to good drainage and grows well on poor, droughty sites and in acid soils (3). It is widely used for turfgrass in open sun and moderate shade and forms a dense turf when seeded heavily. Production of certified seed of Chewings fescue in Oregon in 1991 to 1993 averaged 677 kg/ha on 6,714 ha with a farm value of $5.7 million.

Stem rust occurs commonly in perennial ryegrass (*Lolium perenne* L.) (4) and tall fescue (*Festuca arundinacea* Schreb.) (10) grown for seed in the Willamette Valley, but no cases of stem rust occurring on Chewings fescue are recorded in disease clinic files at Oregon State University, nor in the Index of Plant Diseases (7) or Fungi on Plants and Plant Products in the United States (2). Search of literature in BIOSIS, CAB, and AGRICOLA between 1984 and 1994 produced no reports of stem rust on Chewings fescue. The purpose of this paper is to report the occurrence of stem rust in Chewings fescue in Oregon, compare virulence among isolates of *P. g. graminicola*, and assess stem rust infection percentages for seedlings among selected cultivars of Chewings fescue. Four other species of cool-season grasses grown for seed in the Willamette Valley were inoculated with these isolates of stem rust. When severe, stem rust can reduce seed yields of perennial ryegrass and tall fescue by more than 90% (4,8).

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Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by USDA implies no approval of the product to the exclusion of others that may also be suitable.

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MATERIALS AND METHODS

Pathogenicity and a comparison of virulence among isolates. Urediniospores of stem rust collected from leaves and culms of an unknown cultivar of Chewings fescue growing in a commercial field in May 1993 were used to inoculate greenhouse-grown plants of cv. Jamestown Chewings fescue. A second collection of urediniospores of stem rust was made in June and July 1993 from Chewings fescue plants growing in three locations in the Willamette Valley, two in the central part of the valley (designated as isolates B-1 and F-2), and one from the north end of the valley, designated as isolate M-3. Urediniospores from the second collection were dried over calcium chloride for 3 h at 20°C, sealed in air-tight vials, and frozen at –70°C until used. Jamestown Chewings fescue was inoculated with each of these three isolates to evaluate their disease response on a common host cultivar.

Plant growth procedures. Seed of cv. Jamestown was placed on blotter paper moistened with 0.1% KNO_3, incubated at 4°C in the dark for 1 week, then moved to a controlled environment chamber to provide 25°C with 8 h light (50 $\mu E\ s^{-1}\ m^{-2}$) and 15°C with 16 h dark. After 17 days, individual plants were transplanted into single cone-shaped plastic containers (2.9 by 12.3 cm) containing fine-grade vermiculite. The containers were placed in a mist chamber in a greenhouse for 1 week to promote rooting and then moved to an incubation chamber at 25°C with 16 h light (500 $\mu E\ s^{-1}\ m^{-2}$) and 15°C with 8 h dark for 5 weeks. Plants were watered daily and fertilized weekly with 2.4 g/liter of 20-20-20 (N-P-K) liquid fertilizer (Peters) supplemented with Peters Compound 111 soluble trace element mix at the rate of 0.06 g/g of Peters 20-20-20 to maintain vigorous growth.

Inoculation and infection. In the first inoculation, urediniospores fresh from the field were used. In the second inoculation, frozen urediniospores of each isolate were heat shocked at 45°C for 2 min and suspended in Soltrol 170 oil, a highly refined, nonphytotoxic oil frequently used in rust inoculations of cereals (5). Urediniospore suspensions were adjusted to a concentration of 6.0, 5.5, and 5.5×10^6 per ml, for isolates B-1, F-2, and M-3, respectively. Urediniospores were sprayed evenly onto leaves of 9-week-old plants of cv. Jamestown. Clean glass microscope slides were placed within the plant canopy to trap urediniospores. Urediniospores were counted in a 1 × 25 mm band (four bands per slide × four slides per plant rack × 2 replications) to determine the density of inoculum and compare uniformity of inoculation. Urediospore counts per 1 mm^2 for isolate B-1, F-2, and M-3 in replication 1 were 4.8, 3.6, and 2.8, respectively. Viability of urediniospores was determined by spraying a portion of the urediniospore suspension onto the surface of 2% water agar in a petri dish. After the Soltrol evaporated, dishes were incubated at 20°C for 23 h and urediniospores were examined at ×100 to determine percent viability. The percentage of germination (based on 100 to 125 urediniospores) for isolate B-1 was 66%, for isolate F-2 was 39%, and for isolate M-3 was 61%.

Inoculated plants were placed in a dew chamber programmed to provide 17 h at 19°C in the dark followed by 5 h at 25°C in light that ranged from 110 $\mu E\ s^{-1}\ m^{-2}$ in the front to 30 $\mu E\ s^{-1}\ m^{-2}$ in the back of

the chamber. Halfway through the light cycle, plant racks were rotated 180° to provide light to two sides of plants. Fifty plants were inoculated with each of three isolates, with two replications (i.e., 100 plants per isolate). Following the infection period, plants were returned to the incubation chamber previously described for 14 days at 25°C with 16 h light and 15°C with 8 h dark.

Disease assessment. Leaves were rated for stem rust infection 14 days after inoculation. A plant was considered infected if sporulating pustules of stem rust developed on one or more inoculated leaves. Usually, stem rust pustules developed in 10 or more leaves, with 4 to 6 pustules per leaf, in susceptible plants. Percent infection was based on the number of inoculated plants with pustules. Plants without pustules were considered to be resistant to stem rust in the seedling stage.

A comparison of Chewings fescue cultivars and test plants. Following the comparison of virulence among stem rust isolates, urediniospores were collected from Jamestown Chewings fescue, combined, frozen as before, and used as inoculum. Young plants of twelve cultivars of Chewings fescue, one cultivar of creeping fescue (*Festuca rubra* L. var. *rubra,* cv. Crestlawn), one cultivar of sheep fescue (*Festuca ovina* L., cv. Bighorn), one cultivar of tall fescue (Bonanza), and one cultivar of perennial ryegrass (Palmer) were inoculated as described previously. When inoculated, fine fescues were 9 weeks old, tall fescue was 5 weeks old, and perennial ryegrass was 6 weeks old. Planting dates of different species were staggered to allow inoculation on the same day. Except for perennial ryegrass and tall fescue, seed was requested from the company developing the cultivar. Seed of tall fescue and perennial ryegrass were from a source of certified seed. Cultivars Bonanza and Palmer are highly susceptible to stem rust (9,10). Procedures used to prepare inoculum, inoculate plants, and assess disease were the same as those used in the first experiment.

The position of each entry in a plant rack was randomized and up to 10 seedlings per entry were included for each of three replications in each of two runs. The study was repeated on successive days (run 1 = 26 April 1994; run 2 = 27 April 1994). Plants were incubated in the dark in the dew chamber at 18°C for 17 h in run 1 and for 15 h in run 2. Inoculum contained 6.1 (run 1) or 4.9 (run 2) × 10^6 urediniospores per ml; urediniospores deposited on glass slides within the plant canopy (mean of 4 slides, 2 observations per slide, 3 replications) were 2.9 (run 1) or 3.2 (run 2) per mm^2; urediniospore germination after 24 h on 2% water agar was 56% (run 1) or 55% (run 2).

Data analysis. The study was arranged to assess stem rust in each of 10 plants, for each of 16 entries, with three replications in each of two runs. The study was planned to include 960 plants (16 cultivars × 10 plants × 6 replications), but fewer seedlings of some cultivars were inoculated because of low germination. Some seedlings died during the test from causes other than stem rust, usually due to a failure to become established in the mist chamber after transplanting. When the study was completed, the number of plants in the study was reduced to 899. The percent infected plants and the mean and standard deviation were calculated for each entry (six replications).

RESULTS AND DISCUSSION

Pathogenicity and a comparison of virulence among isolates. Uredinial pustules typical of *P. g.* subsp. *graminicola* Urban (1,6) erupted in leaves of greenhouse-grown Chewings fescue 10 days after inoculation with urediniospores collect from the field. This is believed to be the first report of stem rust on Chewings fescue in Oregon. When young plants of cv. Jamestown were inoculated with three isolates of *P. g.* subsp. *graminicola*, the percent infected plants in replication 1 was 95% for B-1, 98% for F-2, and 94% for M-3, and in replication 2 was 94% for B-1, 96% for F-2, and 98% for M-3. Based on inoculating 300 plants of Jamestown, 288 plants (96%) were infected by stem rust, with no differences in infection percentages among isolates and between replications.

Comparison of cultivars of Chewings fescue and test plants. When mean percent stem rust–infected plants of Chewings fescue were compared by cultivar, all were susceptible (Table 1). Twenty-eight seedlings of red fescue cv. Crestlawn were infected and this species had fewer stem rust–infected seedlings than the 12 cultivars of Chewings fescue. Despite infection of five seedlings of sheep fescue and one seedling of perennial ryegrass by these isolates in environmentally controlled conditions, these hosts are probably not susceptible to *P. g.* subsp. *graminicola* in field conditions. None of the seedlings of tall fescue were infected by *P. g.* subsp. *graminicola* in this study.

Forty-nine stem rust–free plants from 12 cultivars of Chewings fescue (Table 1), about 7% of 673 plants inoculated, were transplanted into 3.8-liter plastic pots containing sand:soil:peat:pumice (1:1:1:1, vol/vol/vol/vol) adjusted to pH 6.4 with lime ($CaCO_3$). Plants were placed in a greenhouse at 15 ± 3°C with 12 h of supplemental light, watered daily and fertilized weekly to maintain vigorous growth, and transplanted on 29 October 1994 into field plots on a farm near Corvallis.

Plants were assessed for stem rust beginning the first week of May 1995. Stem rust pustules were observed in primary and secondary branches of panicles in eight plants on 10 May. The cumulative number of stem rust–infected plants increased to 15 on 15 May, 23 on 17 May, 40 on 2 June and 45 on 6 June. On June 23, three plants were free of stem rust and remained rust-free until seeds were harvested 26 June 1995. Stem rust severity in susceptible plants was assessed by the modified Cobb scale and ranged from 20 to 100%. Conditions in the field were different from those

Table 1. Seedling reactions of five grass species inoculated with *Puccinia graminis* subsp. *graminicola* isolated from Chewings fescue.

Species and cultivar	Number of plants[a]		Mean infection		
	Noninfected	Infected	Percent[b]	SD[c]	Plant total[d]
Chewings fescue					
Jamestown II	1	59	98	4	60
Performer	2	58	97	5	60
Victory	2	52	96	6	54
Banner II	3	56	95	6	59
Jamestown	2	58	95	5	60
Tiffany	3	56	95	8	59
Molinda	5	55	92	10	60
Camero	5	54	92	8	59
SR 5100	6	51	90	9	57
Shadow	6	42	88	12	48
HYOB	7	53	88	15	60
Longfellow	7	30	72	23	37
Red fescue					
Crestlawn	32	28	47	16	60
Sheep fescue					
Bighorn	41	5	10	13	46
Perennial ryegrass					
Palmer	59	1	2	4	60
Tall fescue					
Bonanza	60	0	0	0	60

[a] Number of stem rust–infected and noninfected plants in each entry; total for six replications.
[b] Mean percent stem rust–infected plants divided by plants inoculated; mean for six replications of each entry.
[c] Standard deviation of the mean for six replications of each entry.
[d] Number of plants inoculated; total for six replications.

that occurred during the inoculation of 9-week-old plants in the greenhouse. Obviously, questions remain about how the host, pathogen, environment, and time interact to influence development of stem rust in Chewings fescue.

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LITERATURE CITED

1. Cummins, G. B. 1971. The Rust Fungi of Cereals, Grasses, and Bamboos. Springer-Verlag. New York.
2. Farr, D. F., Bills, G. F., Chamuris, G. P., and Rossman, A. 1989. Fungi on Plants and Plant Products in the United States. American Phytopathological Society, St. Paul, Minn.
3. Hanson, A. A., Juska, F. V., and Burton, G. W. 1969. Species and varieties. Pages 370-409 in: Turfgrass Science. A. A. Hanson and F. V. Juska, eds. Am. Soc. Agron. Madison, Wis.
4. Meyer, W. A. 1982. Breeding disease-resistant cool-season turfgrass cultivars for the United States. Plant Dis. 66:341-344.
5. Rowell, J. B. 1984. Controlled infection by *Puccinia graminis* f. sp. *tritici* under artificial conditions. Pages 291-332 in: The Cereal Rusts. Vol. 1 Origins, Specificity, Structure, and Physiology. W. R. Bushnell and A. P. Roelfs, eds. Academic Press, Orlando, Fla.
6. Savile, D. B. O. 1984. Taxonomy of cereal rust fungi. Pages 79-112 in: The Cereal Rusts. Vol. 1. Origins, Specificity, Structure, and Physiology. W. R. Bushnell and A. P. Roelfs, eds. Academic Press, Orlando, Fla.
7. U.S. Department of Agriculture. 1960. Index of plant diseases in the United States. Agric. Handbook No. 165. U.S. Government Printing Office. Washington D.C.
8. Welty, R. E., and Azevedo, M. D. 1993. Application of propiconazole in management of stem rust in perennial ryegrass grown for seed. Plant Dis. 78:236-240.
9. Welty, R. E., and Barker, R. E. 1992. Evaluation of resistance to stem rust in perennial ryegrass grown in controlled and field conditions. Plant Dis. 76:637-641.
10. Welty, R. E., and Barker, R. E. 1993. Reaction of twenty cultivars of tall fescue to stem rust in controlled and field conditions. Crop Sci. 33:963-967.