# *Septoria spraguei, Pyrenophora trichostoma,* and *Cochliobolus sativus* Incidence on Russian Wildrye Grass Leaves and *S. spraguei* Host Range

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**ABSTRACT**

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Of 112 samples of Russian wildrye (*Elymus junceus*) leaves collected in 1982 in North Dakota, 57% were infected with *Septoria spraguei*, 51% with *Pyrenophora trichostoma*, and 68% with *Cochliobolus sativus*. All three fungi were also isolated from other *Elymus* spp. *S. spraguei* has not been previously reported on beardless wildrye (*E. triticoides*), Altai wildrye (*E. angustus*), or European dune wildrye (*E. arenarius*), and *P. trichostoma* has not been previously reported on field collections of basin wildrye (*E. cinereus*), beardless wildrye, or European dune wildrye. Five isolates of *S. spraguei*, two from Russian wildrye and one each from basin wildrye, Siberian wildrye, and beardless wildrye, were pathogenic on a wide range of grass species and could be reisolated from each. Among 25 inoculated grass species, leaf necrosis was most severe on Russian wildrye and beardless wildrye, followed by Siberian wildrye and tetraploid crested wheatgrass (*Agropyron desertorum*). Lesions were fewer and smaller on spring wheat than on Russian wildrye.

Additional key words: forage grasses

Many pathogens have been identified in North American grasslands (5,8,9). *Cochliobolus sativus* (Ito & Kurib.) Drechs. ex Dastur (asexual state *Helminthosporium sativum* P.K. & B. = *Bipolaris sorokiniana* (Sacc. in Sorok.) Shoemaker) and *Pyrenophora trichostoma* (Fr.) Fckl. (syn. *Pyrenophora tritici-repentis* (Died.) Drechs.; asexual state *Helminthosporium tritici-repentis* Died. = *Drechslera tritici-repentis* (Died.) Shoem. = *H. tritici-vulgaris* Nisikado) are two pathogens that have been reported on a wide range of grasses (1–3,5–9). In the northern Great Plains, *C. sativus* has been isolated from 79 species of Gramineae and is common on species of *Elymus, Bromus, Agropyron,* and *Hordeum* (8). *C. sativus* was the most common leaf spot pathogen isolated from *Elymus* spp. in Alberta, Canada (1). *P. trichostoma* has been considered the most common leaf spot pathogen in Saskatchewan native prairie dominated by thickspike wheatgrass (*Agropyron dasystachyum* (Hook.) Scribn.) and western wheatgrass (*A. smithii* Rydb.) (6). *P. trichostoma* has produced a high level of symptom expression when inoculated on *Elymus* spp. in the glasshouse (3).

*Septoria spraguei* Uecker & Krupinsky has recently been described as a pathogen on *Elymus* spp. and has been reported on only a few grasses: Russian wildrye (*Elymus junceus* Fisch.), basin wildrye (*E. cinereus* Scribn. & Merr.), and Siberian wildrye (*E. sibericus* L.) (10).

This study was conducted to determine the incidence of *S. spraguei, P. trichostoma,* and *C. sativus* in seeded pastures and in various experimental breeding nurseries of Russian wildrye. A second objective was to ascertain the host range of *S. spraguei*, which is frequently found in experimental plots and pastures seeded to *Elymus* spp.

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## MATERIALS AND METHODS

**Survey.** In 1981, a pasture of the Russian wildrye cultivar Vinall at Mandan, ND, was severely affected by a leaf spot disease. Sections from 38 leaves collected at random from established plants were plated out for identification of the pathogen. Sixteen severely affected seedlings were also collected. In 1982, leaves were collected on three dates from established plants in at least four random locations in two Vinall pastures in the Mandan area. In addition, samples were collected from 20 random locations in one of the Vinall pastures near the end of the growing season.

In 1982, samples were taken from 81 Russian wildrye experimental plots located in four different field performance tests at Mandan. A Western Canada Cooperative Test for Russian wildrye that included six cultivars (Idaho 100, Swift, Vinall, Mayak, Cabree, Sawki) and six experimental strains (NRG-711, SCR-3753, SCR-3751, SCR-3752, SCR-17020, Mdn 2 Cl) was sampled. A more recent Western Canada Cooperative Test containing 13 entries, nine of which were common to the first test, was sampled; the four additional strains were SCR-3762, SC-17040, SCR-3793, and SCR-3792. Four plant introductions (PI 314082, PI 314668, PI 314675, PI 434241), two cultivars (Vinall, Sawki), and one experimental strain (ND-692) were sampled from Russian wildrye plots maintained by the Soil Conservation Service at Mandan. In addition, 49 clones of Russian wildrye were sampled from clonal evaluation plots.

At Dickinson, ND, three Russian wildrye cultivars (Mayak, Sawki, Vinall), Altai wildrye (*E. angustus* Trin.), basin wildrye, mammoth wildrye (*E. giganteus* Vahl), and Canada wildrye (*E. canadensis* L.) were sampled. *Elymus* spp. other than Russian wildrye collected at Mandan were Altai wildrye, mammoth wildrye, beardless wildrye (*E. triticoides* Buckl.), basin wildrye, Siberian wildrye, and European dune wildrye (*E. arenarius* L.).

Leaf sections approximately 2 cm long from eight leaves from each sample were surface-sterilized for 3 min in a 1.05% sodium hypochlorite solution containing two drops of a surfactant (Tween 20) per 100 ml. The leaf sections were rinsed in sterile distilled water, plated on water agar in plastic petri dishes, sealed with Parafilm, and incubated at 20 ± 1 C with illumination by cool-white fluorescent tubes. On the fifth day, lights were turned off for 24 hr to promote sporulation of fungi that require a dark period for sporulation, such as *P. trichostoma*. After 7 days, leaf sections were checked for the presence of fungi, particularly *S. spraguei*, and the conidial states of *P. trichostoma* and *C. sativus*. Pycnidiospores from pycnidia on the leaf sections were examined microscopically to determine the presence of *S. spraguei*.

**Host range of *S. spraguei*.** Seeds of 25 grass species (Table 1) were planted in a glasshouse in plastic Cone-tainers (4 cm diam, 21 cm deep) containing a peat moss-vermiculite mixture (1:1) and fertilized with a balanced macronutrient and micronutrient fertilizer. Sodium vapor lamps (400 W) were used to supplement the natural photoperiod and to maintain a 12-hr light-dark cycle. Temperatures ranged from 13 ± 3 C during the dark period to 24 ± 4 C during the light period.

Five isolates of *S. spraguei* were obtained from leaf samples collected from experimental plots and pastures at Mandan. Two (79-2759 and 81-4460) were from Russian wildrye and one each was from basin wildrye (80-3499), Siberian wildrye (80-3517), and beardless wildrye (80-3527). To obtain cultures, cirrhi produced on pycnidia on the leaf sections were transferred to V-8 juice agar (18% V-8 juice, 2 g $CaCO_3$ per liter, 2% agar) in petri plates and incubated at 21 ± 1 C under cool-white fluorescent tubes. Each isolate was used twice to inoculate all the grasses evaluated, and all inoculations were completed within 2 wk.

Inoculum of the five isolates was produced on V-8 juice agar. Fungal growth was scraped from the surface of the agar plates, blended for 60 sec with distilled water, and filtered through four layers of cheesecloth. The inoculum from each isolate for all inoculations contained a comparable number of spores per ml ($4.6–5.6 \times 10^4$). Two drops of Tween 20 per 100 ml were added to the inoculum before it was sprayed on the plants. Two inoculations were conducted with five individual isolates on 25 species of grass to form group 1. Two inoculations were also made with the five isolates of *S. spraguei* on 12 wheat (*Triticum aestivum* L.) cultivars (Angus, CI 17744; Butte, CI 17681; Coteau, CI 17749; Fortuna, CI 13596; James, CI 17791; Kitt, CI 17297; Len, CI 17790; Alex, CI 17910; Olaf, CI 15930; Sinton, CI 17573; Eureka, CI 17738; Waldron, CI 13958), two barley (*Hordeum vulgare* L.) cultivars (Park, CI 15768; Glenn, CI 15769), one oat (*Avena sativa* L.) cultivar (Kelsey, CI 8171), and the Russian wildrye cultivar Vinall to form group 2.

After inoculation, the plants were maintained for 48 hr in a high-humidity chamber (4) in a glasshouse and shielded from direct sunlight by shade cloth. The inoculated plants were kept on a glasshouse bench after being removed from the chamber and rated twice for disease reaction 7–12 days after inoculation. Leaves were visually assessed for percent necrosis, number of lesions, and lesion size. Percent necrosis ratings were recorded as the percentage of leaf blade tissue that was necrotic. The rating for number of lesions was 0 = none, 1 = very few, 2 = few, 3 = intermediate, and 4 = numerous. The rating for lesion size was 0 = no lesion, 1 = very small, 2 = small, 3 = medium, and 4 = large.

After disease ratings were made, leaves were harvested and allowed to dry. The procedure described for obtaining the original isolates was also used to detect and reisolate the fungus. Four leaf sections from each inoculated grass species for each of two inoculations with isolate 81-4460 from Russian wildrye were plated on water agar. These eight leaf sections from each species were assayed for the presence of *S. spraguei* 7–9 days later. Pycnidiospores from cirrhi from at least one leaf of each grass species were checked microscopically for positive identification.

**Table 1.** Gramineous hosts infected by *Septoria spraguei*

| Host | Common name | Cultivar or experimental name |
|---|---|---|
| *Agropyron cristatum* | Crested wheatgrass | Fairway |
| *A. desertorum* | Crested wheatgrass | Nordan |
| *A. intermedium* | Intermediate wheatgrass | Mandan 759 |
| *A. smithii* | Western wheatgrass | Rodan (Mandan 456) |
| *A. spicatum* | Bluebunch wheatgrass | Secar |
| *A. spicatum* f. *inerme* | Beardless wheatgrass | Whitmar |
| *Alopecurus arundinaceus* | Creeping foxtail | Garrison |
| *Andropogon gerardii* | Big bluestem | SD-27 |
| *Andropogon hallii* | Sand bluestem | ND-329 |
| *Andropogon scoparius* | Little bluestem | ND-130 |
| *Bouteloua gracilis* | Blue grama | Mandan local seed collection |
| *Bromus inermis* | Smooth bromegrass | Mandan 404 |
| *Calamovilfa longifolia* | Prairie sandreed | ND-95 |
| *Dactylis glomerata* | Orchardgrass | Kay |
| *Elymus angustus* | Altai wildrye | Prairieland |
| *E. arenarius* | European dune wildrye | PI 348865 |
| *E. cinereus*[a] | Basin wildrye | P-15590 |
| *E. giganteus* | Mammoth wildrye | Mandan accession |
| *E. junceus*[a] | Russian wildrye | Vinall |
| *E. sibericus*[a] | Siberian wildrye | PI 325315 |
| *E. triticoides* | Beardless wildrye | Shoshone |
| *Panicum virgatum* | Switchgrass | Switchgrass 98 |
| *Phalaris arundinacea* | Reed canarygrass | Pop. A 74 |
| *Sorghastrum nutans* | Indiangrass | PM-ND-444 |
| *Stipa viridula* | Green needlegrass | Lodorm |
| *Triticum aestivum* | Spring wheat | Angus, Fortuna, James, Kitt |

[a] Previously reported as host (10).

## RESULTS AND DISCUSSION

**Survey.** Thirty-six of the 38 leaf sections plated out from the 1981 collections of Russian wildrye leaves from established plants showing leaf spot symptoms and 13 of the 16 leaf sections from seedlings were infected with *S. spraguei*. Thus, *S. spraguei* caused a leaf spot disease in Vinall Russian wildrye pastures at Mandan in 1981. The presence of *S. spraguei* on recently killed seedlings may help explain the difficulty of interseeding Russian wildrye into a Russian wildrye pasture to improve the stand or why thin stands of Russian wildrye often do not thicken even though seed is produced by existing plants. The diseased stand of established plants apparently serves as a source of inoculum to emerging seedlings and may be the main factor contributing to their poor survival.

Of the 112 samples of Russian wildrye leaves collected from experimental plots and pastures in 1982, 57% were infected with *S. spraguei*, 68% with *C. sativus*, and 51% with *P. trichostoma* (Table 2). Two or three of the fungi were present on some leaf samples. *S. spraguei* was isolated from all of the forage evaluation plots, most of the pastures, but few of the clonal plots (Table 2), suggesting that the selections in the clonal plots were more resistant to *S. spraguei* than the surveyed cultivars and experimental strains. Plants from the source population with symptoms similar to those produced by *S. spraguei* were eliminated when selections were made for further clonal evaluation. In contrast, *P. trichostoma* was identified twice as often on the single-plant selections in the clonal plots as on the cultivars or experimental strains in the forage evaluation plots or on Vinall in the pastures (Table 2). *C. sativus* was identified on more samples from experimental and clonal plots than from the Vinall pasture. All three fungi must be considered when evaluating Russian wildrye germ plasm in a breeding program, because each has the potential to cause serious disease when environmental conditions are favorable.

Though not previously reported on Altai wildrye, beardless wildrye, or European dune wildrye, *S. spraguei* was found on these species and also on Siberian wildrye. *S. spraguei* was not isolated from any species sampled at Dickinson. *P. trichostoma* was identified on Altai wildrye, basin wildrye, Canada wildrye, mammoth wildrye, European dune wildrye, and beardless wildrye. *P. trichostoma* has not been previously reported on field collections of basin wildrye, beardless wildrye, or European dune wildrye. Considering the wide host range of *P. trichostoma* and the high level of symptom expression in inoculated *Elymus* spp. (3), the presence of *P. trichostoma* on a wide range of *Elymus* spp. is to be expected. *C. sativus* was identified on Altai wildrye, basin wildrye, Canada wildrye, mammoth wildrye, European dune wildrye, and beardless wildrye. Since *C. sativus* is a very common pathogen on grasses (1,8,9), its presence on *Elymus* spp. is to be expected.

Other pathogens that might cause leaf spot symptoms on *Elymus* spp. were identified infrequently and considered to be of minor importance. These included *Septoria nodorum* (Berk.) Berk., *S. avenae* Frank. f. sp. *triticea*, *Ascochyta* sp., *Selenophoma* sp., *Fusarium* sp., and *Curvularia* sp.

**Host range of *S. spraguei*.** Although the amount of necrosis produced and the number and size of lesions on different hosts varied slightly, the most susceptible and the most resistant grasses reacted similarly to all five isolates. In the first group inoculated, Russian wildrye and beardless wildrye averaged 61 and 50% necrosis, respectively (Table 3). They were followed by Fortuna spring wheat, Siberian wildrye, and tetraploid crested wheatgrass (*Agropyron desertorum* (Link) Schult.) with 21, 14, and 13% leaf necrosis, respectively (Table 3). All other grasses averaged 10% necrosis or less.

*Elymus* spp. had the most and the largest lesions of all species surveyed (Table 3). Of the *Elymus* spp., European dune wildrye showed the least damage, with a few small lesions. Only four other grasses had an appreciable number of small to medium-sized lesions: western wheatgrass, beardless wheatgrass, smooth bromegrass (*Bromus inermis* Leyss.), and spring wheat cultivars James and Fortuna. Most of the remaining grasses developed very few and very small lesions. Two grasses, little bluestem (*Andropogon scoparius* Michx.) and prairie sandreed (*Calamovilfa longifolia* (Hook.) Scribn.), developed no visible symptoms throughout all the studies (Table 3), even though *S. spraguei* was reisolated from all grasses inoculated (Table 3).

Russian wildrye averaged 50% necrosis in the group of grasses that also included wheat, barley, and oats. Wheat cultivars with the most damage were Fortuna, James, Waldron, and Eureka, with 9,7,7, and 6% necrosis, respectively. All other wheat cultivars and the barley and oat cultivars showed less than 5% necrosis. Wheat cultivars Kitt and Angus showed no necrosis. Russian wildrye had numerous large lesions, and wheat

**Table 2.** Fungi isolated from Russian wildrye grass leaves in 1982

| Location | Number of collections | *Septoria spraguei* No. | *Septoria spraguei* % | *Pyrenophora trichostoma* No. | *Pyrenophora trichostoma* % | *Cochliobolus sativus* No. | *Cochliobolus sativus* % |
|---|---|---|---|---|---|---|---|
| Pastures of cultivar Vinall | 31 | 27 | 87 | 12 | 39 | 15 | 48 |
| Experimental plots | 32 | 32 | 100 | 11 | 34 | 26 | 81 |
| Clonal evaluation plots | 49 | 5 | 10 | 34 | 69 | 35 | 71 |
| Total | 112 | 64 | 57 | 57 | 51 | 76 | 68 |

**Table 3.** Susceptibility of 25 species of forage grasses and four cultivars of wheat to *Septoria spraguei* isolated from *Elymus cinereus*, *E. junceus*, *E. sibericus*, and *E. triticoides*

| Host | Percent necrosis[a] | Number of lesions[b] | Lesion size[c] | *S. spraguei* on leaf pieces[d] |
|---|---|---|---|---|
| *Agropyron cristatum* | 8 | 0.3 | 0.5 | 8/8 |
| *A. desertorum* | 13 | 1.0 | 1.2 | 6/8 |
| *A. intermedium* | 4 | 0.8 | 1.0 | 7/8 |
| *A. smithii* | 7 | 3.1 | 2.3 | 8/8 |
| *A. spicatum* | 5 | 1.3 | 1.7 | 7/8 |
| *A. spicatum* f. *inerme* | 10 | 1.8 | 2.3 | 8/8 |
| *Alopecurus arundinaceus* | 2 | 0.8 | 1.0 | 8/8 |
| *Andropogon gerardii* | 0 | 0.1 | 0.3 | 2/8 |
| *Andropogon hallii* | 0 | 0.3 | 0.3 | 4/8 |
| *Andropogon scoparius* | 0 | 0 | 0 | 8/8 |
| *Bouteloua gracilis* | 4 | 0.9 | 1.1 | 7/8 |
| *Bromus inermis* | 5 | 1.9 | 2.4 | 8/8 |
| *Calamovilfa longifolia* | 0 | 0 | 0 | 2/8 |
| *Dactylis glomerata* | 1 | 0.5 | 0.6 | 7/7 |
| *Elymus angustus* | 4 | 3.3 | 3.2 | 8/8 |
| *E. arenarius* | 4 | 1.7 | 2.1 | 8/8 |
| *E. cinereus* | 4 | 2.5 | 2.2 | 7/8 |
| *E. giganteus* | 4 | 3.4 | 2.4 | 8/8 |
| *E. junceus* | 61 | 3.8 | 4.0 | 8/8 |
| *E. sibericus* | 14 | 2.3 | 3.0 | 8/8 |
| *E. triticoides* | 50 | 4.0 | 4.0 | 8/8 |
| *Panicum virgatum* | 1 | 0.4 | 0.5 | 7/7 |
| *Phalaris arundinacea* | 1 | 0.2 | 0.2 | 7/8 |
| *Sorghastrum nutans* | 2 | 0.5 | 0.6 | 6/8 |
| *Stipa viridula* | 1 | 1.2 | 1.1 | 8/8 |
| *Triticum aestivum* 'Angus' | 1 | 0.7 | 1.0 | 7/8 |
| *T. aestivum* 'Fortuna' | 21 | 3.2 | 2.2 | 7/8 |
| *T. aestivum* 'James' | 8 | 2.2 | 2.0 | 7/8 |
| *T. aestivum* 'Kitt' | 1 | 0.5 | 0.7 | 8/8 |
| LSD (0.05) | 5 | 0.5 | 0.5 | |

[a] Average of 20 observations (10 inoculations, two observations per inoculation).
[b] 0 = none, 1 = very few, 2 = few, 3 = intermediate, 4 = numerous.
[c] 0 = no lesion, 1 = very small, 2 = small, 3 = medium, 4 = large.
[d] Number of leaf pieces with fungus/number of leaf pieces plated.

cultivars James and Fortuna had an intermediate number of small lesions. The rating for number of lesions was below 2 for all other cultivars. Wheat cultivars Fortuna, James, Olaf, Sinton, Eureka, and Waldron and the barley cultivar Park scored 2 or less for lesion size. Wheat cultivars Angus, Kitt, and Butte had the fewest lesions, and these were very small.

*S. spraguei* was reisolated from all inoculated cereal hosts and could be considered pathogenic on them under artificial conditions. The fungus is not likely to be a problem on wheat, however, because it has never been isolated from collections of field-grown wheat and because virulence on the wheat cultivars artificially inoculated in this study was low.

Symptoms were most severe on *Elymus* spp., even though all five isolates were pathogenic on a wide range of hosts. The similar reaction of isolates from various hosts suggests a lack of pathogenic races. *S. spraguei* should be considered a potential problem on the wildryes, particularly in monoculture. The perennial nature of the wildrye grasses would facilitate overwintering and survival of *S. spraguei.* The low virulence of *S. spraguei* on other genera of perennial grasses suggests that the pathogen will not become a serious problem for these forage grasses.

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