Quantitative Disease Assessment of Wheat Seedling Leaves
Inoculated with *Fusarium roseum* 'Culmorum'

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ABSTRACT


Wheat seedling leaves were wounded and inoculated with agar disks containing microconidia of *Fusarium roseum* 'Culmorum.' Seedlings were grown in a growth chamber under moisture stress conditions of -5 or -10 bars for 3 wk before inoculation. Relative comparisons between cultivar pairs were made by measuring lesion length 1 wk after inoculation. There were no differences in lesion size among seedlings of the same cultivar grown under the two different soil matric potentials. Significant lesion size differences occurred between cultivars in all paired comparisons when seedlings were grown under moisture stress and lesions were measured 7 days after inoculation. This quantitative method of measuring disease reaction between wheat cultivars may be useful in identifying general resistance (horizontal resistance sensu Vanderplank). Identification of general resistance could significantly aid breeders in selecting for increased resistance to *Fusarium* foot or root rot of wheat.

MATERIALS AND METHODS

Fort Collins clay loam (10) was air-dried 3 days, sifted through a 4-mm mesh screen, and added at the rate of 508 g per 15-cm-high x 11-cm-diameter plastic pot. Matric potentials of -5 or -10 bars were established in the soil by adding water. The amount of water needed to establish the matric potentials was determined from the matric potential versus soil moisture curve for Fort Collins clay loam (10). The matric potential was maintained by weighing the pots every 2 days and adding the amount of water needed to reestablish the original weight. Six wheat (*Triticum aestivum* L.) seeds were sown in each pot. Each pot was covered with a clear plastic bag to enhance seed germination. Pots were placed in a Percival plant growth chamber (Percival Refrigeration and Manufacturing Co., Boone, IA 50036) programmed for 21 ± 1 C and 16 hr of both fluorescent and incandescent light (~5,000 lx) each day. After 5 days, the plastic bags were removed and the seedlings were thinned to four plants per pot. The plants were inoculated when they were ~3 wk old.

A culture of *F. roseum* 'Culmorum' (supplied by R. J. Cook, USDA, ARS, Pullman, WA) was maintained on potato-dextrose agar (PDA) by single-sporo transfers. Off types were discarded after each transfer. The original culture was preserved in sterile soil at 3-4 C and periodically cultured for comparison with the cultures in use. Inoculum was prepared by calibrating a suspension to contain 10⁶ macroconidia per milliliter of distilled water and adding 1 ml of this spore suspension to 20 ml of warm, melted (50 C), sterilized water agar in petri dishes. Disks were cut from the solidified agar medium with a 5.5-mm-diameter sterilized cork borer. Agar disks, each containing ~35 conidia, were used to inoculate seedlings. A sterilized dissecting needle was used to puncture the leaf and an agar disk was secured over the wound with cellophane tape. Two leaves per plant were inoculated for a total of eight inoculations per pot. Infection was enhanced by individually covering the pots with plastic to maintain high humidity, and the pots were returned to the growth chamber. The plastic was removed after 5 days, and the lesion length was measured 2 days later. No infection occurred in 10-30% of the inoculations, and there was only a very small necrotic area surrounding the wounds. *F. roseum* 'Culmorum' routinely could be isolated from surface-disinfested lesions cultured on PDA.

Each randomized experiment was designed to compare two cultivars and consisted of: 10 pots of cultivar 1 at -5 bars; 10 pots of cultivar 2 at -5 bars; 10 pots of cultivar 2 at -5 bars; and 10 pots of cultivar 2 at -10 bars. There were four experiments comparing cultivars Scout and Baca, and one experiment each comparing

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cultivars Scout and Calvin, Newton and Vona, Baca and Calvin, and Vic and Calvin. Lesions were measured 5, 6, and 7 days after inoculation in one of the Scout and Baca comparisons. A separate experiment was done to compare Scout and Baca under −5 bars and under no water stress (ample moisture). The data for each experiment were analyzed with a two-way analysis of variance (14).

**RESULTS**

The first experiment compared Scout and Baca under matric potentials of −5 and −10, and the analysis of variance of these data revealed highly significant differences. Differences between cultivars were evident, whereas differences within the same cultivar under the two matric potentials were slight. To determine where significant differences existed, each combination of the cultivar and matric potential factors was analyzed by a one-way analysis of variance. There were no significant differences between identical cultivars under different matric potentials, but all differences between cultivars were significant. The disease measurements of each cultivar under the different matric potentials were pooled and the comparison was made between cultivars. Matric potential also had little effect on disease rating in all the other experiments. Therefore, the lesion size measurements of both matric potentials for each cultivar were pooled, and these comparisons were analyzed for each experiment. The analyzed data are presented in Table 1.

Significant differences between cultivars in lesion size were found at 7 but not at 5 and 6 days after inoculation. No significant lesion size differences between Scout and Baca were found when seedlings were grown under adequate moisture. The lesion sizes of both cultivars were significantly larger when seedlings were grown under moisture stress and the difference between cultivars was significant.

**DISCUSSION**

Results of previous studies (12,13) demonstrated that *F. roseum 'Culmorum,*’ in similar stem-wound inoculation experiments, produced larger lesions under −5 and −10 than under −1/3 and −1 bars. These lower matric potentials (−5 and −10 bars) were used in this leaf-wound study because more disease (larger lesions) was consistently observed under these conditions. No significant lesion size differences between cultivars occurred when plants were grown with ample moisture. The lesions on the nonstressed plants were significantly smaller than on the moisture stressed plants. This is consistent with results from inoculations in stem wounds (12,13) and Cook and Papendick’s (5) observations that moisture stress enhances infection and disease severity caused in cereals by foot and root rotting *Fusarium* species.

Lesion size was recorded 5, 6, and 7 days after inoculation during one experiment with cultivars Scout and Baca. The 5- and 6-day comparisons revealed no significant differences. This indicates significant lesion size differences may be expressed only at least 7 days after inoculation. Lesion size variation between experiments on the same cultivars was observed, which demonstrates the necessity of making cultivar comparisons only within and not among experiments.

Cook (3) reported that wheat plants can sustain mild root and culm rot due to infection by *F. roseum 'Culmorum,*’ but without moisture stress no severe foot rot occurred. Physical factors, such as depth of rooting, leaf area, and number of stomata per unit area, may affect the rate of soil water use. These factors would help determine the relative moisture stress a cultivar sustains under various environmental conditions. Wheat cultivars having the ability to maintain high leaf water potentials may sustain the least Fusarium foot rot (3). Variation in field “resistance” may only be due to variation of stress factors, or an interaction of these factors with resistance to the pathogen. For these reasons, resistance identified by the leaf inoculation method may or may not correlate with field resistance. Additional studies are needed to determine if this resistance identified in the laboratory is effective in the field and can be correlated with yield loss.

Calvin and Vic are durum wheat cultivars reported to be susceptible and resistant, respectively, to foot and root rot in North Dakota field tests (J. S. Quick, personal communication). Foot and root rot losses in North Dakota may be due more to *C. sativus* than *Fusarium* species, although *Fusarium* species are involved in the disease complex (15). The lesion size comparisons of cultivar Calvin and Vic correlated with their reported field reactions. This does not necessarily indicate that resistance to *C. sativus* and *F. roseum 'Culmorum* are under the same genetic control. *Fusarium* species, or specifically *F. roseum 'Culmorum,*’ may be more involved in the North Dakota disease complex than previously thought.

Cultivar variation affecting expression of disease caused by *F. roseum 'Culmorum,*’ was demonstrated in the laboratory with a leaf inoculation method and moisture stress conditions. The small cultivar differences that were observed may be due to resistance mechanisms under quantitative genetic control. Moisture stress conditions simulate natural water stress in the field and are necessary for the expression of cultivar differences in lesion size. This method is less expensive and faster than tests in the field. Another advantage is that the plant survives and can be grown to maturity as a seed source. The leaf inoculation method may be adaptable to breeding programs since it would enable workers to identify and select for increased resistance to Fusarium foot and root rot causal agents. Such a quantitative method of measuring resistance would be extremely helpful in developing commercial cultivars with increased resistance to Fusarium foot and root rot of wheat.

**LITERATURE CITED**


