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The Influence of Common Root Rot on Net Blotch of Winter Barley

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ABSTRACT

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Field plots were established with different severity levels of common root rot (primarily *Bipolaris sorokiniana*) on the barley cultivar Maury. Soil fumigation, chemical seed treatment, and soil infestation with *B. sorokiniana* were used to induce root rot severities ranging from 0 to 47%. At growth stage 9 (Feekes scale) plants were evaluated for common root rot and net blotch (*Drechslera teres*) severity. With increasing root rot levels, net blotch severity increased significantly. The results indicate that common root rot can predispose barley plants to the net blotch disease.

The concept of predisposition has been well described (16), and many examples of pathogen-pathogen interactions related to this concept have been documented (10). Wheat (*Triticum aestivum* L.), oats (*Avena sativa* L.), and barley (*Hordeum vulgare* L.) have been predisposed to fungal infection by viruses (1,3,9) and by other fungi (2,5,8,13–15). The work of Jenkins and Jones (5) is unique in that a soilborne pathogen predisposed wheat to infection by a foliar pathogen. They used a detached leaf technique, and the authors urged caution in the extrapolation of their findings to possible effects in the field. However, they indicated that there was sufficient evidence to suggest that these effects warranted further examination.

Two of the major barley diseases in Pennsylvania are net blotch, a foliar disease caused by *Drechslera teres* (Sacc.) Shoem. (teleomorph *Pyrenophora teres* Drechs.), and a root rot complex in which the principal pathogen is *Bipolaris sorokiniana* Sacc. in Sorok. Shoem. (teleomorph *Cochliobolus sativus* (Ito & Kurib.) Drechs. ex Dast.). In a preliminary experiment conducted in Pennsyvlania during the 1979–1980 growing season, it appeared that with increasing root rot severity, there was a concurrent increase in net blotch severity (4). The purpose of this study was to examine the relationship between root rot and net blotch on barley.

MATERIALS AND METHODS

A preliminary field experiment was conducted in the fall of 1979 to establish an inoculum level to be used in future root rot experiments. Conidia of four barley isolates of *B. sorokiniana* from Pennsylvania were pooled and grown on sterilized rye grains for 4 wk. The infested grain was then air dried, distributed on the soil surface of specific plots with a fertilizer spreader at five different rates, and incorporated into the soil by raking lightly.

Plots were located near University Park, PA, in an area planted with corn the previous year. All plots were fumigated with methyl bromide at a rate of 393 kg/ha 2 wk before infestation and planting. The fumigant was injected, and the area was covered with a plastic tarp, which was removed after 4 days. Plots were planted 5 days after tarp removal. The plots were 0.9×3.6 m, and the barley cultivar Maury (certified seed) was seeded with a 5-row cone-type planter at a rate of 167 kg/ha in rows spaced 178 mm. Plots were fertilized with 672 kg/ha of 5-10-10 (N-P-K) in the fall and topdressed with NH_4NO_3 (67.2 kg N/ha) as early as possible the following spring. The plot design was a randomized complete block with five levels of soil infestation and four replications. The entire experiment was duplicated in the same field, and spores of *D. teres* were applied to the foliage with a backpack sprayer when the first node of the stem was detectable on the main shoot, which is growth stage (GS) 6 Feekes scale (6). The pathogen was grown on low-carbon potato-dextrose agar (PDA) (0.5% dextrose) for 10 days, spores were collected in distilled water, and a spore suspension of approximately 5,000 spores per milliliter and 50 ml per plot was used for the inoculation.

Net blotch and root rot evaluations were made when the flag leaf ligule and auricles were visible (GS-9). Twenty-five main tillers were selected at random from each plot. Plants were removed from the soil with care taken to preserve the integrity of the root system. The foliage was evaluated for net blotch severity by estimating the percentage of leaf area infected on the flag leaf and the two leaves below the flag of each tiller. These three values were averaged to provide an average severity per tiller. The 25 severities were averaged to provide an average severity per plot. These plot values were used for statistical analyses. This same procedure was followed for spot blotch, the foliar phase of *B. sorokiniana*.

After the foliar disease evaluations were completed for each plot, the root systems were removed from the plants, and the 25 root systems from the plants evaluated for net and spot blotch were evaluated for root rot, so that root rot severity could be related to the foliar disease severity. Soil was removed from the roots by washing with a stream of water, and root systems were refrigerated until they could be placed in a Plexiglas root washer, where they were washed under high pressure mist nozzles for 16 hr (chamber designed by L. W. Burgess, Department of Plant Pathology and Agricultural Entomology, University of Sydney, Australia). The subcrown internodes were examined, and the percentage of the subcrown internode area covered with lesions was estimated.

Reduction in total root area is a component of the root rot syndrome and should contribute to yield loss (12), so this factor was added to the disease evaluation. The percent reduction in the total root system for each plant was assessed visually by comparing the total size of the root plume to healthy root plumes (five root systems selected randomly from the healthy plants in adjacent buffer rows) and estimating the percent reduction in size. The root reduction estimate was added to the estimate for percentage of subcrown internode lesion area to provide the root rot severity for each plant. The severities of the 25 root systems for each plot were averaged. All data were subjected to analyses of variance.

Nonlinear regression analysis was performed, using root rot, net blotch, and spot blotch as dependent variables. Whereas the net blotch severity was greater in the foliar-inoculated test, the slope of the regression line was similar to that in the uninoculated test for all three variables, based on a Student's t test, and the data from the two tests were pooled to provide eight replications for further analyses. The increments between the levels of the independent variable (inoculum of *B. sorokiniana*) were equal; therefore, orthogonal polynomial coefficients were used to compute the regression equations (7).

Each dependent variable was tested for goodness of fit in a stepwise fashion through the third-degree polynomial using the coefficients as coded X values. The best-fitting curve was selected by analysis of variance and the deviations from the linear, quadratic, and cubic components.

During 1980 and 1981, four different factors with two levels of each factor were used in combinations to provide different levels of root rot in field plots. The combinations included infested or clean seed, chemical treated or nontreated seed, fumigated or nonfumigated soil, and infested or noninfested soil. The infested seed were obtained from field plots in Pennsylvania where the barley plants had a high incidence of *B. sorokiniana*, based on the root rot and foliar (spot blotch) symptoms. These seed were tested for the presence of seedborne pathogens by placing 1,000 seed on PDA. Fifty-eight percent of the seed was infested with *B. sorokiniana*, and 7% of the seed was infested with *Fusarium graminearum* Schwabe based on microscopic identification. Both of these pathogens may be involved in a root rot complex (12). The clean seed were obtained from Aberdeen, ID, where barley was grown under irrigation. These seed also were tested, and *D. teres* was not detected on the infested or clean seed. The seed treatment was a commercial combination of carboxin (17% a.i.) and thiram (17% a.i.). The application rate was 2.5 ml of product per kilogram of seed. The fumigated plots were treated with methyl bromide at a rate of 393 kg/ha. Soil was infested by incorporating *B. sorokiniana* on rye grain substrate (138 g/m^2) into the seed bed just before planting as described previously. The amount of rye grain added was based on results obtained in 1979 and 1980.

Plots in the fall of 1980 were located near University Park, PA, in soil planted with wheat in the previous year. The plots were 0.9 × 3.6 m, and the barley cultivar Maury was seeded with a 5-row cone-type planter at a rate of 167 kg/ha in rows spaced 178 mm. Plots were fertilized with 672 kg/ha of 5-10-10 (N-P-K) in the fall and topdressed with NH_4NO_3 as early as possible in the spring. A preliminary experiment was conducted in 1979 to determine the fertilizer rate to be used in fumigated plots. The highest N rate that allowed growth without lodging was selected. The fumigated plots received 33.6 kg of N per hectare, whereas nonfumigated plots were topdressed with 67.2 kg. The additional N was added to nonfumigated plots, since fumigated plots receive additional N, which is released after fumigation (11). The plot design in 1980 was a split plot with the fumigation factor (fumigated and nonfumigated soil) assigned to the whole plots and arranged in a randomized block with four replications. Three factors with two levels each (infested or noninfested soil, fungicide treated or nontreated seed, and infested or clean seed) were assigned to subplots in all possible combinations in a completely randomized treatment arrangement.

The presence of root rot was substantiated at late tillering or the leaf sheath lengthening stages (GS-3 to GS-4) in the spring by removing plants from the soil and examining the subcrown internodes. Net blotch symptoms were not widely distributed until GS-7 when the second node was detectable.

Net blotch and root rot evaluations were made when plants reached GS-9. Twenty main tillers were selected at random from each of the 64 plots. Disease evaluations were conducted as described previously. All data were subjected to analyses of variance.

RESULTS

In 1980, there was an increase in root rot with increasing inoculum concentrations up to an inoculum level of 185 g/m^2 (Fig. 1). Net blotch severity also increased in a similar manner. While the response curves for net blotch and root rot were similar, the response curve for spot blotch was different. This disease was not visible until the inoculum levels of *B. sorokiniana* reached 185 g, the same level at which root rot and net blotch severities appeared to peak. There was a strong positive correlation between root rot and net blotch severities ($r = 0.96$), but the correlation between root rot and spot blotch (0.62) or net blotch and spot blotch severities (0.47) was weak.

In 1981, root rot severities ranged from 0 to 47% depending on the treatment combination, while net blotch severities ranged from 5 to 28% (Table 1). The effect of chemical seed treatment was nonsignificant on all diseases; therefore, only the significant main treatments (soil fumigation, seed infestation, and soil infestation) are presented in this table. Soil fumigation did not affect net blotch severity but did decrease the root rot severity. Soil and seed infestations had significant effects on all three diseases, and infested seed contributed to the largest increase in disease severity for all diseases. Spot blotch did not appear on the top three leaves until root rot severities were approximately 15%, and it increased as root rot increased. Some net blotch was present in the absence of root rot, but its severity increased as root rot severities increased. However, at root rot severities greater than 32%, net blotch severity did not continue to increase, and actually decreased as root rot severity increased to 47%. There was a positive correlation between root rot and net blotch ($r = 0.83$) and between root rot and spot blotch ($r = 0.73$) severities. The correlation between net blotch and spot blotch was weak ($r = 0.47$).

All significant treatment interactions with regard to root rot and spot blotch severities were related to a difference in the magnitude of the response. In the second-order interaction for both of these variables, the effect of fumigation was greater with noninfested soil. The infestation process added *B. sorokiniana* to soil following fumigation, thereby reducing the effectiveness of the fumigation. With regard to net blotch, the infested seed by infested soil interaction was related to a change in the magnitude of the response. However, the first- and second-order interactions that involved soil fumigation and soil infestation treatments were related to a difference in the direction of the response. The net blotch severity was higher in the fumigated soil when the soil was infested with *B. sorokiniana*. However, in nonfumigated soil the severity of net blotch was greater when the soil was not infested with the pathogen. This can in part be explained by the competition for leaf area by the net blotch and spot blotch pathogens. When the treatments included infested seed and infested soil, there was a decrease in net blotch severity in the nonfumigated plots with a concomitant increase in spot blotch severity.

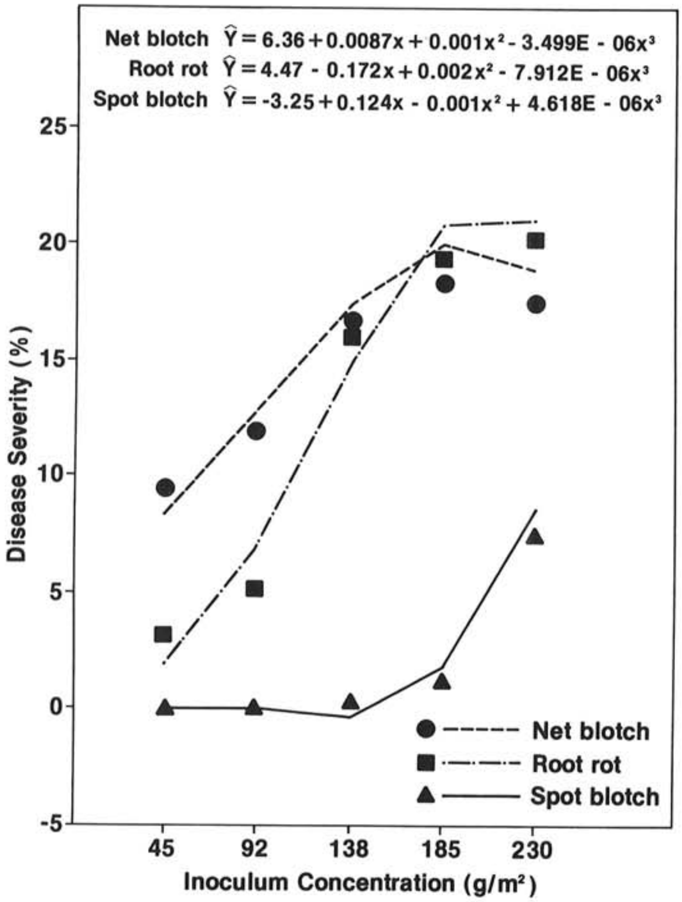

Fig. 1. The effect of inoculum concentrations of *Bipolaris sorokiniana* on subsequent severities of root rot and spot blotch, and net blotch caused by *Drechslera teres* based on nonlinear regression analysis.

DISCUSSION

The results from the 1980 study indicate that net blotch severity increased as root rot severity increased. However, at a specific level of inoculum of *B. sorokiniana*, the root rot severity stabilized and the net blotch severity subsequently peaked or declined. One might be tempted to relate the net blotch severity solely to the severity of root rot, but the role of spot blotch cannot be ignored. In both years, the spot blotch phase of *B. sorokiniana* was evident when inoculum levels were high. The decrease in net blotch could be associated with the appearance of spot blotch and a possible competition for leaf area. In 1981, the net blotch severity decreased from 28.7 to 18.1%, while root rot severity increased from 41.4 to 46.8%. At the highest inoculum levels it might be more appropriate to relate root rot severity to total leaf blotch severity (net blotch and spot blotch).

Because of the large number of root samples that were processed in the root washer, it was not feasible to evaluate the root system of each plant in relation to its foliar disease rating. The system we used calculated an average disease severity per plot with 25 tillers per plot in 1980 and 20 tillers in 1981. The root rot incidence for 1981 is presented in Table 1 so that one also can compare root rot incidence to net blotch severity. Root rot incidence or severity relates in a similar manner to net blotch severity. By calculating severity as we have, disease incidence and intensity are represented in one value.

In the case of nonfumigated soil and also infested seed in 1981, *F. graminearum* may have contributed to the overall root rot syndrome. In another study in an adjacent field using these same treatments, isolations were made from infected subcrown internodes taken from plots where soil was not fumigated (J. A. Frank, *unpublished*). *B. sorokiniana* was isolated from 59% of the subcrown internodes, and *F. graminearum* was isolated from 45%

TABLE 1. The effect of the three significant main factors on severity of net blotch caused by *Drechslera teres*, and root rot and spot blotch caused by *Bipolaris sorokiniana* on winter barley cultivar Maury

Factors			Net blotch severity (%)	Root rot severity (%)	Spot blotch severity (%)	Root rot incidence (%)
Seed[a]	Soil[b]	Fum[c]				
0	0	1	5.4[d]	0.0[e]	0.0[d]	0.0[f]
0	0	0	5.8	1.6	0.0	6.8
0	1	1	9.6	14.8	0.0	35.0
0	1	0	12.9	18.6	0.1	43.1
1	0	1	16.2	21.3	0.1	48.7
1	0	0	28.7	32.0	1.0	76.2
1	1	1	28.7	41.4	5.9	93.7
1	1	0	18.1	46.8	16.9	96.8
Seed × soil			**[g]	NS[h]	**	NS
Seed × fum			NS	**	**	**
Soil × fum			**	**	**	**
Seed × soil × fum			**	**	**	**

[a] Noninfested seed = 0, infested seed (primarily *Bipolaris sorokiniana*) = 1.
[b] Noninfested soil = 0, infested soil (rye grain artificially infested with *Bipolaris sorokiniana* and incorporated into the soil) = 1.
[c] Nonfumigated = 0, fumigated methyl bromide, 393 kg/ha before soil infestation) = 1.
[d] Mean severity of the top three leaves, based on percent leaf area infected for 20 tillers per plot and four replications at growth stage 9 (Feekes scale). The values represent the means for specific treatments pooled over seed treatments.
[e] Mean (percent reduction for root system) plus (percent infection on subcrown internodes) for 20 tillers per plot and four replications at growth stage 9 (Feekes scale). The values represent the means for specific treatments pooled over seed treatments.
[f] Percent plants out of 20 with root rot symptoms.
[g] ** = Significant at the $P = 0.01$ level based on analysis of variance.
[h] NS = Nonsignificant.

of the subcrown internodes. Although *F. graminearum* may be responsible for at least part of the common root rot that developed in this study, it would not interfere with the interpretation of results, which are based on overall root rot severity.

Because *D. teres* may be seedborne, there was the possibility that the infested seed treatment may have contributed directly to the increase in net blotch severity rather than increasing root rot severity which, in turn, predisposed the plant to net blotch. However, *D. teres* was not isolated from either seed source. While this does not completely rule out the possibility that seedborne *D. teres* may have been present, it strongly suggests that the infested seed had a significant effect on root rot severity and that this root rot was primarily responsible for the increase in net blotch severity. The increase in net blotch severity that occurred when plots were infested with *B. sorokiniana* strongly supports this. These results lend support to the findings of Jenkins and Jones (5), which suggested that a soilborne pathogen may predispose a plant to foliar infections.

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