Leaf Infection and Yield Losses Caused by Brown Spot and Bacterial Blight Diseases of Soybean

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ABSTRACT

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Yield reductions caused by *Septoria glycines* and *Pseudomonas glycinea* (*P. syringae*) on soybeans were measured in the field on plants inoculated with either pathogen and by both in combination. The disease index based on percentage defoliation and percentage of remaining leaf area diseased on samples of 25 inoculated plants per plot ranged 65–78% for *S. glycines*, 60–69% for both combined, 46–62% for *P. glycinea*, and 4–24% of the uninoculated check plants. Yield reductions of 17.9% for *P. glycinea*, and 17.4% for *S. glycines*, and 14.1% for both organisms combined were observed. Both yield and infection of inoculated plants differed statistically from uninoculated plants.

Brown spot (caused by *Septoria glycines* Hemmi) and bacterial blight (caused by *Pseudomonas glycinea* Coerper) (=*P. syringae*) are two of the most common diseases of soybean (*Glycine max* [L.] Merrill) in the major growing areas (1,2,7,10,11,13). Seed and infected plant debris are the sources of primary inoculum for both pathogens (3,5,6,10,13,14) and both are spread under conditions of high moisture and strong winds (4,13). Development of bacterial blight is favored by cool temperatures 22–26 C (2) while brown spot develops best under warmer temperatures 20–34 C (2,11). The cool nights, warm days, and wind-driven rain characteristic of the northern soybean growing region of the United States, provides good conditions for the development of these diseases.

Dunleavy et al (8) determined that bacterial blight reduced soybean yield as much as 22% under favorable disease conditions. Recently, in similar yield trials with brown spot disease, yield reductions approached 18% (15). In this study, yield losses associated with these two pathogens acting together or alone were determined in an area free of other soybean pathogens.

MATERIALS AND METHODS

Field plots. Field plots were located 10 km north of Waukon, in northeastern Iowa. Natural inoculum was avoided by placing plots at a site on which soybeans had never been grown and 16 km from any soybean fields.

Treatments consisted of soybeans (cultivar Corsoy) inoculated with *S. glycines* alone, *P. glycinea* alone, *S. glycines* and *P. glycinea* together, or water (control). Treatments were replicated five times and arranged in a randomized complete-block design. Each plot consisted of seven rows, 75 cm apart and 8 m long and were separated by three rows of corn. Seeds were sown 4.5 cm apart on 22 May 1978. Before planting, seeds were soaked in 0.5 percent NaOCl for 20 min, air-dried, soaked in 1 mg/ml tetracycline HCl solution 20 min, and then air-dried to reduce seed-borne contamination. Germination was not affected by this procedure as determined by seed germination trials (authors *unpublished*).

Inoculum. The *S. glycines* isolate was obtained from S. M. Lim (Illinois) and maintained by periodic subculturing on potato dextrose agar (PDA) at 26–28 C. *S. glycines* was inoculated onto soybean leaves and reisolated from lesions every 60–90 days to maintain pathogenicity. Inoculum was harvested from PDA in petri dishes by adding 10 ml of sterile distilled water to each plate and rubbing a glass rod over the surface to dislodge fungal spores. Inoculum concentrations were adjusted to 10^5 conidia per milliliter. Inoculum was applied wih a hand sprayer (Solo Handjet 455 [mfr.—Solo, 0-7032, Box 20, Sindelfingen 6, West Germany; dealer—Solo Co., P.O. Box 5030, 5100 Chestnut Avenue., Newport News, VA 23605]) at 3.07 kg/cm^2 pressure by using a flat-tip spray tip.

The culture of *P. glycinea* was obtained from J. M. Dunleavy (Iowa) and maintained on trypticase soy agar (TSA) at 24–26 C. *P. glycinea* cells used for inoculum were harvested from a 48-hr-old trypticase soy broth shake culture; cells were centrifuged at 16,500 *g* for 10 min, resuspended in sterile distilled water, adjusted to 10^8 cells per milliliter, and applied with a hand sprayer tip similar to those used for *S. glycines*. Fungus and bacterial inocula were not mixed in spray containers, but were applied separately.

Inoculation. Plants were inoculated five times at 10- to 15-day intervals starting 2 August (full bloom, R-2, [9]). An average of 1.2 L of inoculum was applied per plot during evening hours to assure favorable infection conditions (1,11,13).

Disease and yield assessment. Disease assessments were made 2 wk before and 4 wk after the first inoculations. The percentage of diseased leaf tissue and defoliation was measured on 25 plants per plot trial. Defoliation was measured from the top 25 nodes of the plants and the percentage of nodes without foliage was calculated. The percentage of leaf area infected was determined visually according to the scale developed by James (12) for leaf spot of red clover. A disease index for each pathogen acting alone or in combination was calculated by adding the percent defoliated (no. of leaves defoliated × 100% severity) to the percent leaf area infected (no. of leaves remaining ×% severity) and dividing by 25 (the number of leaves per plant per plot). Defoliated leaves were considered 100% diseased and the remaining disease of foliage was measured based on leaf area which was necrotic and chlorotic.

Two 6.25-m-long rows from each plot were harvested and threshed manually on 8 October 1978. The total seed per plot was weighed, moisture content was determined by GAC II grain analysis computer (Dickey John Corp., Box 10, Auburn, IL 62615), and yield was calculated at 13% moisture. Yield loss percentage was calculated by subtracting the mean yield of inoculated plots for each treatment from the mean yield of uninoculated plots, dividing by the latter, and multiplying by 100 ([control−diseased/control] × 100).

Yields of the various treatments were compared by analysis of variance and least significant difference tests. Correlation of

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diseases with yield and yield reduction per percent leaf area affected within treatments were determined by correlation and regression analysis.

RESULTS

Soybean plants developed quite rapidly after sowing, with continuous rains and moderate temperatures (15–26 C avg.) prevailing during the first 10 wk of plant growth. The lack of disease development under these weather conditions favorable for disease indicated that soil debris and seeds were relatively free of contaminating inoculum. Only trace levels of bacterial blight and no brown spot were observed preceding the first inoculations. Weather was less favorable for disease development for the last 10 wk of the season, with temperatures averaging 26–36 C and the precipitation averaging less than half that of the first half of the season.

Disease development was extensive 4 wk after the first inoculations in all plots. The percentage of infected leaf tissue was greatest for plants inoculated with *S. glycines,* 65–78%; followed by plants inoculated with both organisms, 60–69%; and least for plants inoculated with *P. glycinea,* 46–62% (Table 1). Uninoculated plants were noticeably less infected, with 4–24% diseased foliage, the principal pathogen being *S. glycines.* A similar amount of brown spot was detected in bacterial blight plots when compared with controls.

Defoliation of plants inoculated with *S. glycines* alone or with *S. glycines* and *P. glycinea* together was extensive; the level of defoliation of plants in plots treated with *P. glycinea* alone was 10–15% less. Defoliation associated with each treatment was positively related to infection.

No other foliar diseases were detected in late season. Pod and stem blight (*Diaporthe phaseolorum* Cke. & Ell. *sojae* Wehm.) was detected late and was randomly distributed in plots at harvest.

An analysis of variance for both yield ($P = 0.05$) and disease prevalence ($P = 0.01$) revealed significant differences among treatments. Least significant differences values (LSD) showed significant disease and yield differences, between uninoculated plots and plots inoculated with *S. glycines* alone, *P. glycinea* alone, or *S. glycines* and *P. glycinea* together (Table 1). Differences between inoculated plots, however, were significant for disease, but not for yield.

Yields were noticeably reduced in inoculated plots when compared with controls, with mean yield reductions for *S. glycines* alone and *P. glycinea* alone of 17.4 and 17.9%, respectively, and 14.1% combined (Table 1). Yield was highly correlated with diseased leaf tissue for *S. glycines* ($r = -0.95$; $P = 0.01$) and moderately correlated for *P. glycinea* ($r = -0.73$; $P = 0.10$) but was not correlated for plots inoculated with both pathogens ($r = -0.61$; $P = 0.10$). Yield reduction in kg/ha per percent of foliage affected, as indicated by regression coefficients (b), was $b = -0.49$ for *P. glycinea* alone, $b = -0.50$ for *S. glycines* alone, and $b = -0.62$ for *S. glycines* and *P. glycinea* combined. Because of differing amounts of contamination in treated plots, primarily *S. glycines* in plots inoculated with *P. glycinea,* it was not possible to make a test for significance among regression lines for disease.

DISCUSSION

Disease severity and yield loss in plots inoculated with both pathogens did not differ significantly from disease severity and yield loss in plots inoculated only with *S. glycines.* This suggests some degree of antagonism between the two pathogens such that an increase of each is inhibited by the other under the high temperature and low precipitation conditions present during the last half of this study.

Yield was associated with the amount of defoliation in plots, primarily foliage infected with *S. glycines.* Defoliation in turn was related to the level of brown spot observed. The effect of brown spot on defoliation and yield with bacterial blight present was noticeably reduced (18 and 19%, respectively) over plants infected with *S. glycines* alone. In plots inoculated with *P. glycinea,* defoliation was reduced 13% and yield 21% over plants diseased with both brown spot and bacterial blight.

TABLE 1. Mean disease[a] and yield assessments for soybean plants infected with *Septoria glycines, Pseudomonas glycinea,* alone or combined

Inoculum	Defoliation[b] (%)	Remaining leaf area diseased[c] (%)	Disease index[d]	Yield[e] (kg/ha)	Yield reduction (%)
S. glycines	58	32	71	2,185	17.4
P. glycinea	42	19	53	2,174	17.9
P. glycinea + *S. glycines*	48	32	65	2,274	14.1
Controls	10	2	12	2,647	
LSD ($P = 0.05$)			4	293	
LSD ($P = 0.01$)			6	412	

[a] Disease assessments were made on 29 August 1978.
[b] Average percentage of the top 25 leaves abcised per plant (basis, 25 plants).
[c] Percentage of remaining leaf area diseased = remaining 25 leaves × % infected.
[d] Disease index = ([% defoliation × 25 leaves × 100% severity] + [proportion of remaining 25 leaves × % leaf area diseased]) / 25 leaves.
[e] Yield based on 13% moisture.

Premature defoliation was regularly associated with 10% and 25% or more *S. glycines*-infected leaf area for lower and upper leaves, respectively. Plants treated with *S. glycines* alone exhibited 25 – 50% of this defoliation in the upper half while plants treated with *P. glycinea* and *S. glycines* together had less than 25% defoliation of the upper foliage. Plants inoculated with *P. glycinea* alone and contaminated with *S. glycines* exhibited less than 12% random brown spot symptoms and associated defoliation. *P. glycinea* was confined primarily to middle and lower parts of the plants where defoliation was associated more with senescence. Uninoculated plants contaminated with *S. glycines* exhibited less than 5% random brown spot symptoms. Defoliation due to natural senescence as detected in uninoculated plots was limited to lower foliage and averaged 10%.

Further research on yield loss caused by disease complexes is needed. Disease components must be studied separately from any interaction of herbicides, other diseases, insects, and unusual physical conditions. Once a baseline for potential yield loss is determined for each pathogen, the effect of two or more pathogens on a host can be determined under different environmental conditions. Such research in the future will provide realistic information to aid in deciding what control measures to apply.

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