# Seasonal Progress of Brown Stem Rot and Its Impact on Soybean Productivity

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## ABSTRACT

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The seasonal progress of brown stem rot was measured for two growing seasons in plots irrigated at growth stages VC–R1, R1–R8, VC–R8, and in nonirrigated plots. Disease was assessed by disease incidence based on stem symptoms and the isolation frequency of *Phialophora gregata*, and disease severity based on proportion of internal stem discoloration, severity of foliar symptoms, and loss of symptomless leaf area. Each measurement of disease was related to yield loss, which was estimated as the percentage difference in yield between resistant and susceptible cultivars. The pathogen was recovered from roots and stems in advance of internal stem symptoms. Internal stem symptoms were minimal during vegetative growth but rapidly developed after flowering. Although isolation of the pathogen and progress of internal stem symptoms were less during periods of no irrigation, disease incidence and proportion of internal stem symptoms were similar regardless of irrigation regime by growth stage R7. Foliar symptoms did not develop until growth stage R4 and were most severe on plants in plots that received postflowering irrigation. The loss of symptomless leaf area was correlated with the severity of foliar symptoms as measured by the Horsfall-Barratt system. Yield loss with brown stem rot ranged from 13 to 30%. The extent of yield loss was affected by severity of brown stem rot, which in turn was affected by irrigation at specific stages of growth. Yield loss was greatest if both stem and foliar symptoms were present. The severity of foliar symptoms accounted for more yield loss variation than did stem symptoms, especially if multiple point models or an area under the disease progress curve was used to assess disease. Growth stage R5 was an optimum period for a single-point assessment of stem and foliar symptoms.

Brown stem rot of soybean (*Glycine max* (L.) Merrill), caused by *Phialophora gregata* (Allington & Chamberlain) Gams (9), is a prevalent disease in North America (1,2,13,16,19,24). This vascular disease commonly is associated with soybean management systems that call for limited rotation with nonhost crops (5,16). The agronomic impact of this disease has been disputed by many soybean scientists despite reported yield loss estimates ranging from 9 to 44% for individual fields (1,5,11,12,16,24,28). The availability of brown stem rot resistant soybean lines has provided a means to assess the agronomic impact of this vascular disease (19,23,24). Comparing resistant and susceptible cultivars of similar maturity in fields naturally infested with *P. gregata* has resulted in yield loss estimates similar to those derived by crop rotation studies (5,16), comparative yield in infested and uninfested field plots (28), and artificial inoculation in the field (11,12).

*P. gregata* causes a progressive necrosis and browning of vascular and pith tissues (1,2). Approximately 20–30 days before physiological maturity, sudden interveinal chlorosis occurs, followed by necrosis, curling, and wilt of leaves (1,2). Although both stem and foliar symptoms are caused by *P. gregata*, occurrence of internal stem symptoms at maturity has been used most to assess disease incidence (5,12,16,24,28) and severity based on the height of internal stem discoloration (24). It is only recently that foliar symptoms have been used to differentiate resistant and susceptible genotypes in the field (19,23). Limited data are available on the relationship between agronomic performance of soybean and brown stem rot incidence and severity, especially with seasonal progress of the disease (12). The relationship between brown stem rot severity at specific growth stages and over time and agronomic performance of soybean needed further investigation to better understand the economic importance of this disease.

The objectives of this study were to: determine the effect of soil moisture on the seasonal progress of brown stem rot and its impact on agronomic performance of soybean, compare methods to measure the incidence and severity of brown stem rot in the field, and determine the critical stage(s) in plant development at which disease measurements best predict the effect of brown stem rot on agronomic performance.



## MATERIALS AND METHODS

**Experimental locations.** Field studies were performed in plots naturally infested with *P. gregata* at the University of Wisconsin Research Stations at Hancock and Arlington. Soil at Hancock was a Plainfield sandy loam (Typic Udipsamments) and that at Arlington was a Plano silt loam soil (mesic Typic Agriudolls). Plots were planted between 15 and 20 May each year at a rate of 30 seeds per meter of row. Fertilizer (6-24-24) was broadcasted at 10.9 q/ha before planting. Alachlor and trifluralin were applied for weed control at Hancock and Arlington, respectively.

**Plot design and treatments.** A split-split plot design with six replications was employed at the Hancock in 1982 and 1983. Each subplot consisted of one cultivar grown in four rows 9 m long and spaced 0.9 m apart. Experimental variables at Hancock included: the resistant cultivar BSR 201 (25) and the susceptible cultivar Corsoy 79 (19); irrigation during specific growth stages of VC–R1 (preflower application), R1–R8 (postflower application), VC–R8 (season-long application), or no irrigation during the growing season; and six sampling dates, 21, 42, 63, 76, 97, and 119 days after planting, which corresponded to growth stages VC, V4, R2, R4, R5, and R7, respectively (7). Irrigation water was applied based on evapotranspirational losses. Irrigation regimes were not replicated.

Plots of Corsoy 79 and BSR 201 at the Arlington were arranged in a randomized complete block design with four replications. Plots were four rows 9 m long, each row spaced 0.76 m apart, and were not irrigated.

At Hancock in 1983, the agronomic performance of Corsoy 79 and BSR 201 was compared in a second field where brown stem rot had not been observed in previous years and was located 100 m from the infested field. Cultivars were arranged in a randomized complete block design with four replications. Plots were four rows 9 m long, each row was spaced 0.9 m apart, and plots were irrigated at all stages of growth.

**Environmental data.** Environmental data were recorded at the Hancock location with a CR-21 micrologger (Campbell Scientific Inc., Logan, UT) equipped with the appropriate sensing devices. Hourly measurement of a wide range of soil water potential over the growing season was achieved with a gypsum moisture block, and a lower range (zero to −1 bars) of daily soil water potential was measured using tensiometers placed to a depth of 30 cm. The water potential values recorded from the tensiometers were compared to

values recorded by the CR-21 micrologger. Soil water content was determined 21, 42, 63, 76, 97, and 119 days after planting, which correspond to the VC, V4, R2, R4, R5, and R7 soybean growth stages, respectively. The weight of the dried soil sample was recorded, and the water content of the soil was related to the voltmeter reading in the CR-21 micrologger for the date and the time the soil was sampled. A soil water characteristic curve was generated by plotting display readings vs. soil water content. Total water (irrigation water and precipitation) was recorded with rain gauges placed at two sites in each irrigation regime. Air temperature and relative humidity were measured using thermistors and electric hygrometers connected to a CR-21 micrologger.

**Disease assessment.** Ten plants were sampled randomly per replication at each date and each plant was assessed for: the proportion of internal stem discoloration obtained by splitting the stem longitudinally, measuring the greatest advancement of discoloration attributed to brown stem rot, and dividing by the stem height; severity of foliar symptoms based on the Horsfall-Barratt (14) scale of 0–11. Ratings were converted to standardized percentage values using conversion tables provided by Elanco Products Company (a division of Eli Lilly and Company, Indianapolis, IN). Foliar symptoms were rated for severity on a weekly basis from the date of symptom onset and disease incidence based on percentage of plants with internal stem discoloration symptomatic of brown stem rot. Disease incidence also was determined by the percentage of plants from which *P. gregata* was isolated from stem or root tissue. Twenty stem and root pieces were excised from each of five plants per replication. Pieces of tissue were removed 5 cm above and below the soil line and were surface disinfested by immersion in a 0.25% NaOCl solution for 2 min and were blotted for 1 min. Stem and root tissues from each plant were plated separately on acidified potato-dextrose agar and incubated for 10 days at 22 C.

An area under the disease progress curve (AUDPC) was calculated for severity of foliar symptoms, internal stem discoloration, incidence based on percent isolation of *P. gregata* from stems, and incidence based on internal stem discoloration using the following formula (26,27):

$$\text{AUDPC} = \sum_{i=1}^{n-1} [(X_{i+1}+X_i)/2]\ [t_{i+1}-t_i]$$

in which $X_i$ = cumulative disease incidence or severity, expressed as a proportion at the $i$th observation, $t_i$ = time (days after planting) at the $i$th observation, and $n$ = total number of observations of brown stem rot severity or incidence.

**Leaf area measurement.** Leaf area of symptomless foliage of each cultivar was determined by removing all leaflets from five plants per replication and passing them through a Licor leaf area meter (Li-Cor L1-3100, Li-Cor, Lincoln, NE). Each leaf was examined before being passed through the leaf area meter, and chlorotic and necrotic leaves or portions of leaves were discarded. Symptomless leaf area was measured at 3-wk intervals for each cultivar, and the difference in leaf area between Corsoy 79 and BSR 201 was expressed as a percent leaf area reduction for Corsoy 79.

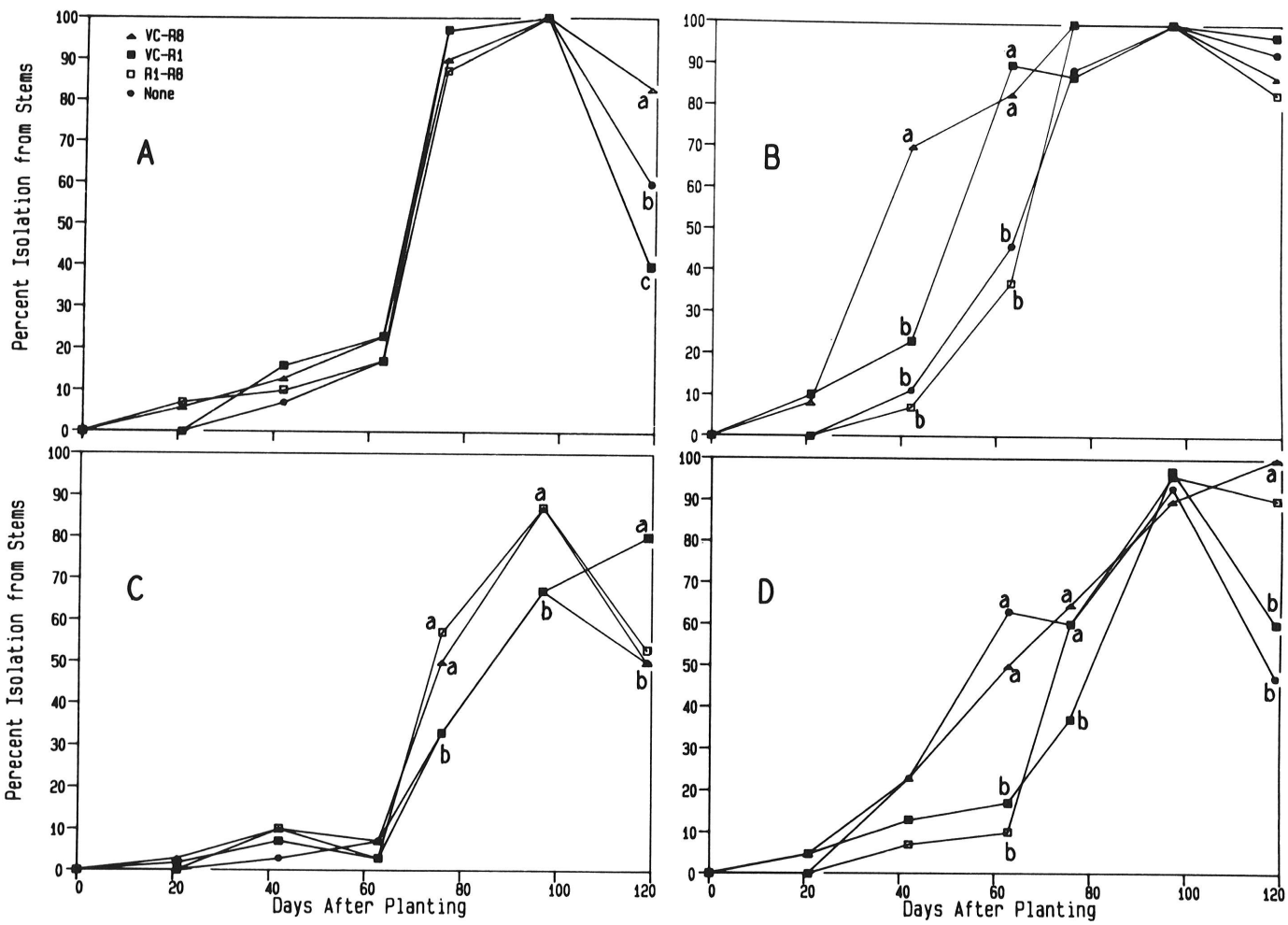


**Fig. 1.** Percentage of plants from which *Phialophora gregata* was isolated from soybean stems of Corsoy 79 in **A,** 1982 and **B,** 1983; and BSR 201 in **C,** 1982 and **D,** 1983. Plants were irrigated at the following growth stages: VC–R8, R1–R8, VC–R1, or none and were sampled 21, 42, 63, 76, 97, and 119 days after planting, which corresponded to growth stages VC, V4, R2, R5, and R7, respectively. Means at each sampling date followed by the same letter are not significantly different from each other ($P = 0.05$) based on Fisher's least significant difference test.

**Yield, yield components, and yield loss estimates.** Yield was obtained at Hancock and Arlington by harvesting 6.1 m of the two center rows of each four-row plot with a plot combine. Total seed weight and 300-seed weight were determined after harvested seed were dried to 12% moisture content. Percent yield loss was derived by the following formula: ([yield of BSR 201 − yield of Corsoy 79]/[yield of BSR 201]) × 100.

**Data analysis.** Data were first analyzed using a standard analysis of variance procedure and Fisher's least significant difference test for mean comparisons. Regression analyses were performed by fitting one line to combined data from all irrigation regimes in an attempt to establish a relationship between the predictor variable (disease) and the dependent variable (percent yield loss). Statistics used to evaluate the predictive value of a regression model were the coefficient of determination ($r^2$) and $F$ and $s$ tests. Calculations were performed using the Statistical Analysis System statistical package (SAS Users Guide, 1979 edition, SAS Institute, Raleigh, NC). A general linear model and the maximum $r^2$ improvement were used to arrive at the above statistics. Critical single point and multiple point models and AUDPC were used in relating disease measurements to estimates of soybean yield loss.

## RESULTS

**Environmental data.** Both irrigation and precipitation were combined to determine the total amount of water each plot received during each growing season. In 1983, plots not irrigated before flowering received less total cumulative water during this period compared with 1982. In 1983, the season-long and postflower irrigated plots received more total water than the preflower and nonirrigated regimes. Because water potential is expressed in negative values, higher (more positive) water potential implies wetter, and lower (more negative) water potential implies drier conditions wherever it is mentioned. The daily mean water potential (−bars) monitored over the entire 1982 season remained higher than −0.5 bar for all irrigation regimes except for 90–96 days after planting (growth stage R5). In 1983, soil water potential during irrigated periods remained higher than −0.5 bars except 64–68 days after planting, when soil water potential dropped to −2 bars. During the period from 50 to 90 days after planting, soil water potential ranged from −2 to −12 bars in plots that did not receive irrigation water. Daily mean air temperatures were 22 and 25 C in 1982 and 1983, respectively. The mean relative humidity measured between R4–R7 growth stages for all irrigation regimes was 85% in 1982, whereas in 1983 it was 85% under season-long and postflowering irrigations and 70% during periods of preflowering and no irrigation.

**Seasonal isolation of *P. gregata*.** The frequency with which *P. gregata* was recovered from stems of Corsoy 79 was influenced by irrigation (Fig. 1A and B). In 1982, a year of higher soil water potential, no differences in isolation frequencies occurred between irrigation regimes until 119 days after planting (Fig. 1A). However, in 1983, isolation of the pathogen during the vegetative phase of growth was greatest from plants grown in irrigated plots (Fig. 1B), but differences subsided during the reproductive phase (after 63 days) of plant development. BSR 201 responded to irrigation much like Corsoy 79 in 1982, but isolation frequencies from BSR 201 were inconsistent with irrigation treatment before flowering in 1983 (Fig. 1C and D).

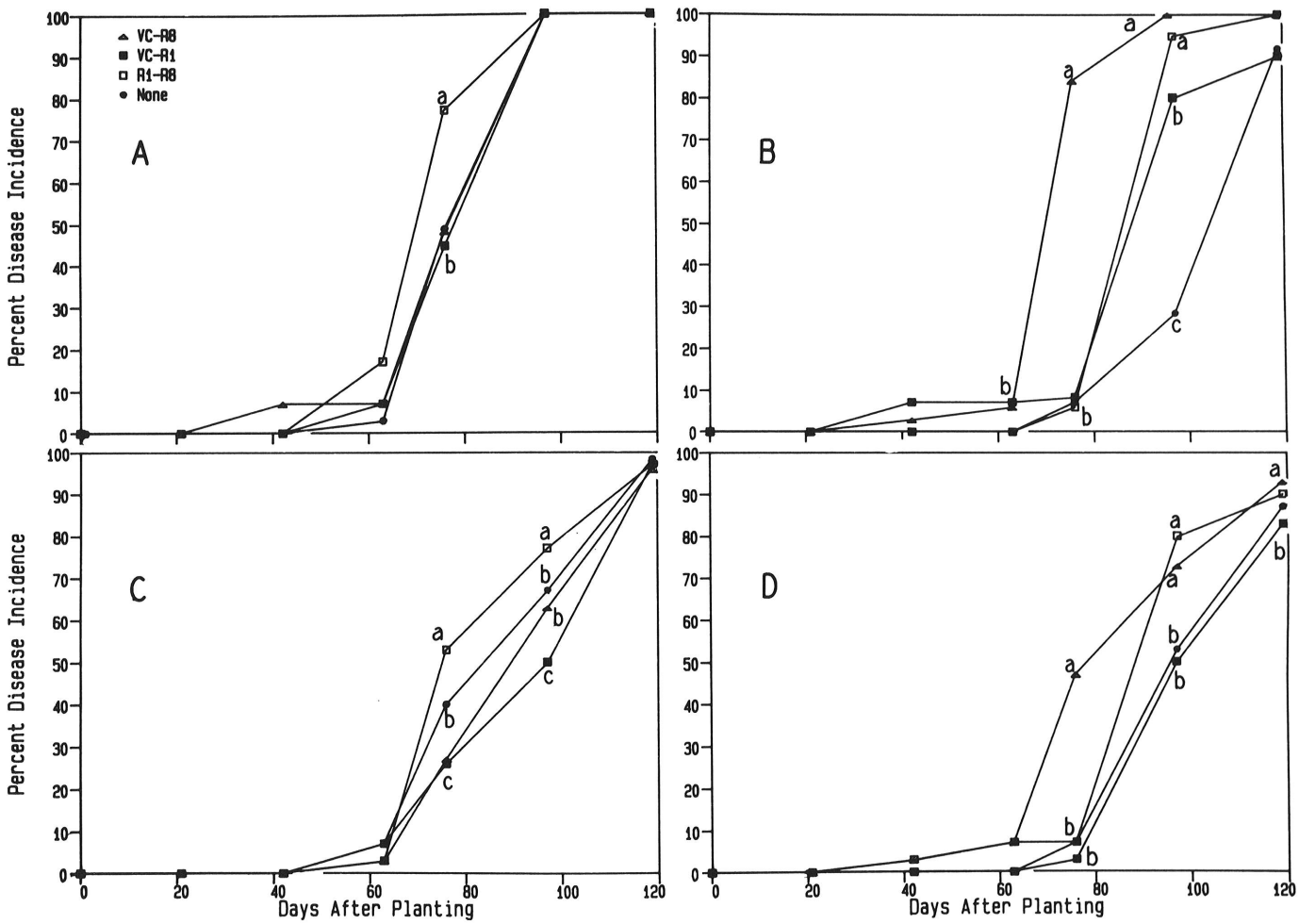


**Fig. 2.** Progress of disease incidence of brown stem rot for soybeans not irrigated or irrigated at growth stages VC–R8, R1–R8, and VC–R1. Plants were sampled 21, 42, 63, 76, 97, and 119 days after planting (growth stages VC, V4, R2, R4, R5, and R7, respectively). **A,** Corsoy 79 in 1982; **B,** Corsoy 79 in 1983; **C,** BSR 201 in 1982; and **D,** BSR 201 in 1983. Means at each sampling date followed by the same letter are not significantly different from each other ($P = 0.05$) based on Fisher's least significant difference test.

**Disease incidence.** Onset of internal stem symptoms for Corsoy 79 was earliest (growth stage V4) in plots that received preflower irrigation (Fig. 2A and B). In 1983, the onset of internal stem symptoms was delayed 34 days for Corsoy 79 grown in plots that did not receive preflower irrigation compared with plots irrigated during the VC–R1 stages (Fig. 2B). Disease incidence was greater for Corsoy 79 than for BSR 201 at 76 and 97 days after planting in plots irrigated at stages R1–R8 (Fig. 2A–D). However, disease

TABLE 1. Area under the disease progress curves (AUDPC)[x] for different measurements of brown stem rot for BSR 201 and Corsoy 79 soybeans grown under four irrigation regimes at Hancock, WI, in 1982

| | Disease measurement criteria[y] | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Disease severity | | | | Disease incidence (%) | | | |
| | Foliar symptoms | | PISD | | Isolation from stems | | Internal stem symptoms | |
| Growth stage irrigation applied | BSR 201 | Corsoy 79 | BSR 201 | Corsoy 79 | BSR 201 | Corsoy 79 | BSR 201 | Corsoy 79 |
| VC–R1 | 80 c[z] | 1920 a | 6 c | 15 b | 27 c | 49 a | 27 d | 41 b |
| R1–R8 | 110 c | 1350 b | 6 c | 19 a | 37 b | 48 a | 37 bc | 48 a |
| VC–R8 | 80 c | 1660 b | 8 c | 20 a | 31 bc | 53 a | 30 d | 42 ab |
| None | 90 c | 1340 b | 7 c | 14 b | 36 b | 46 a | 33 cd | 41 b |

[x] AUDPC = $\sum_{i=1}^{n-1} [(X_i+1+X_i)/2]\,[t_i+1-t_i]$, in which $X_i$ = cumulative disease incidence or severity, expressed as a proportion of the $i$th observation, $t_i$ = time (days after planting) at the $i$th observation, and $n$ = total number of observations BSR severity or incidence (per unit) at the $i$th observation, $t_i$ = time (days) of the $i$th observation, and $n$ = total number of observations.

[y] Disease measurement criteria: severity of foliar symptoms, percent internal stem discoloration (PISD); disease incidence based on recovery of *Phialophora gregata* and presence of internal stem symptoms.

[z] Means of AUDPC followed by the same letter within cultivar and irrigation regimes are not significantly different from each other ($P = 0.05$) based on Fisher's least significant difference.

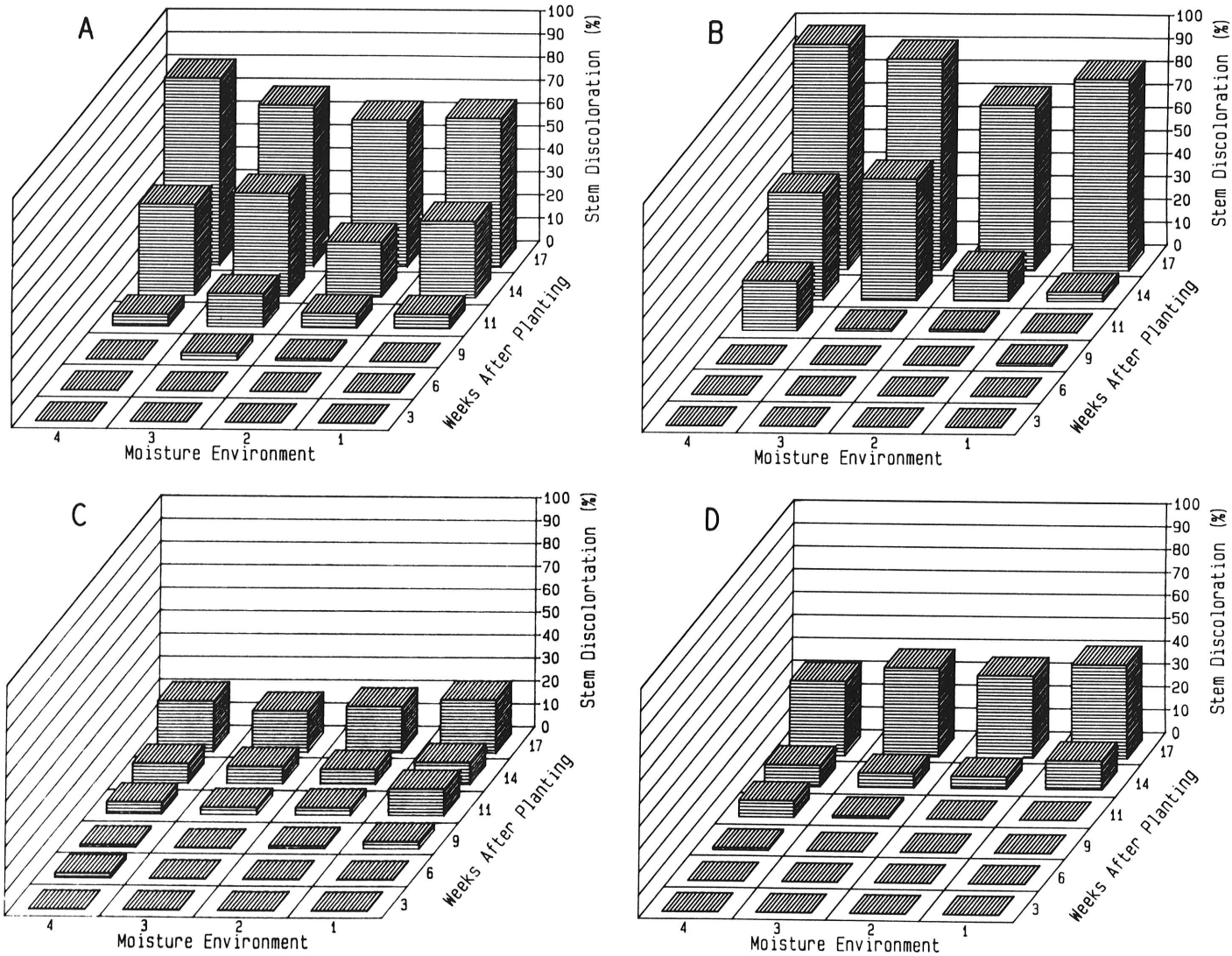


**Fig. 3.** Progress of percent internal stem discoloration for soybeans grown under four irrigation regimes at Hancock, WI; the X-axis denotes soil moisture environments: 1 = no irrigation; 2 = irrigation at VC–R1; 3 = irrigation at R1–R8; and 4 = irrigation at VC–R8. The Z-axis indicates weeks after planting: 3, 6, 9, 11, 14, and 17 correspond to growth stages VC, V4, R2, R4, R5, and R7, respectively. **A,** Corsoy 79 in 1982 and **B,** 1983; **C,** BSR 201 in 1982, and **D,** 1983.

incidence was similar for both cultivars at 119 days (growth stage R7) for all irrigation regimes. Disease incidence was greater for both cultivars after flowering when postflower irrigation was applied in 1983, a year that greater differences in soil water potential developed between irrigated and nonirrigated plots (Fig. 2B and D).

**Proportion of internal stem discoloration.** The advancement of internal stem discoloration progressed rapidly after the

TABLE 2. Area under the disease progress curves (AUDPC)[x] for different measurements of brown stem rot for BSR 201 and Corsoy 79 soybeans grown under four irrigation regimes at Hancock, WI, in 1983

| Growth stage irrigation applied | Disease measurement criteria[y] | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Disease severity | | | | Disease incidence (%) | | | |
| | Foliar symptoms | | PISD | | Isolation from stems | | Internal stem symptoms | |
| | BSR 201 | Corsoy 79 | BSR 201 | Corsoy 79 | BSR 201 | Corosy 79 | BSR 201 | Corsoy 79 |
| VC–R1 | 50 b[z] | 100 b | 5 e | 12 c | 30 d | 68 a | 21 c | 31 b |
| R1–R8 | 80 b | 1340 a | 8 d | 23 b | 44 cd | 60 b | 30 b | 33 b |
| VC–R8 | 90 b | 1380 a | 7 de | 26 a | 55 b | 77 a | 29 b | 48 a |
| None | 50 b | 700 b | 6 de | 11 c | 45 bc | 70 a | 23 c | 18 c |

[x] $AUDPC = \sum_{i=1}^{n-1} [(X_i + 1 + X_i)/2]\ [t_i + 1 - t_i]$, in which $X_i$ = cumulative disease incidence or severity, expressed as a proportion of the $i$th observation, $t_i$ = time (days after planting) at the $i$th observation, and $n$ = total number of observations BSR severity or incidence (per unit) at the $i$th observation, $t_i$ = time (days) of the $i$th observation, and $n$ = total number of observations.

[y] Disease measurement criteria: severity of foliar symptoms, percent internal stem discoloration (PISD); disease incidence based on recovery of *Phialophora gregata* and presence of internal stem symptoms.

[z] Means of AUDPC followed by the same letter within cultivar and irrigation regime are not significantly different from each other ($P = 0.05$) based on Fisher's least significant difference.

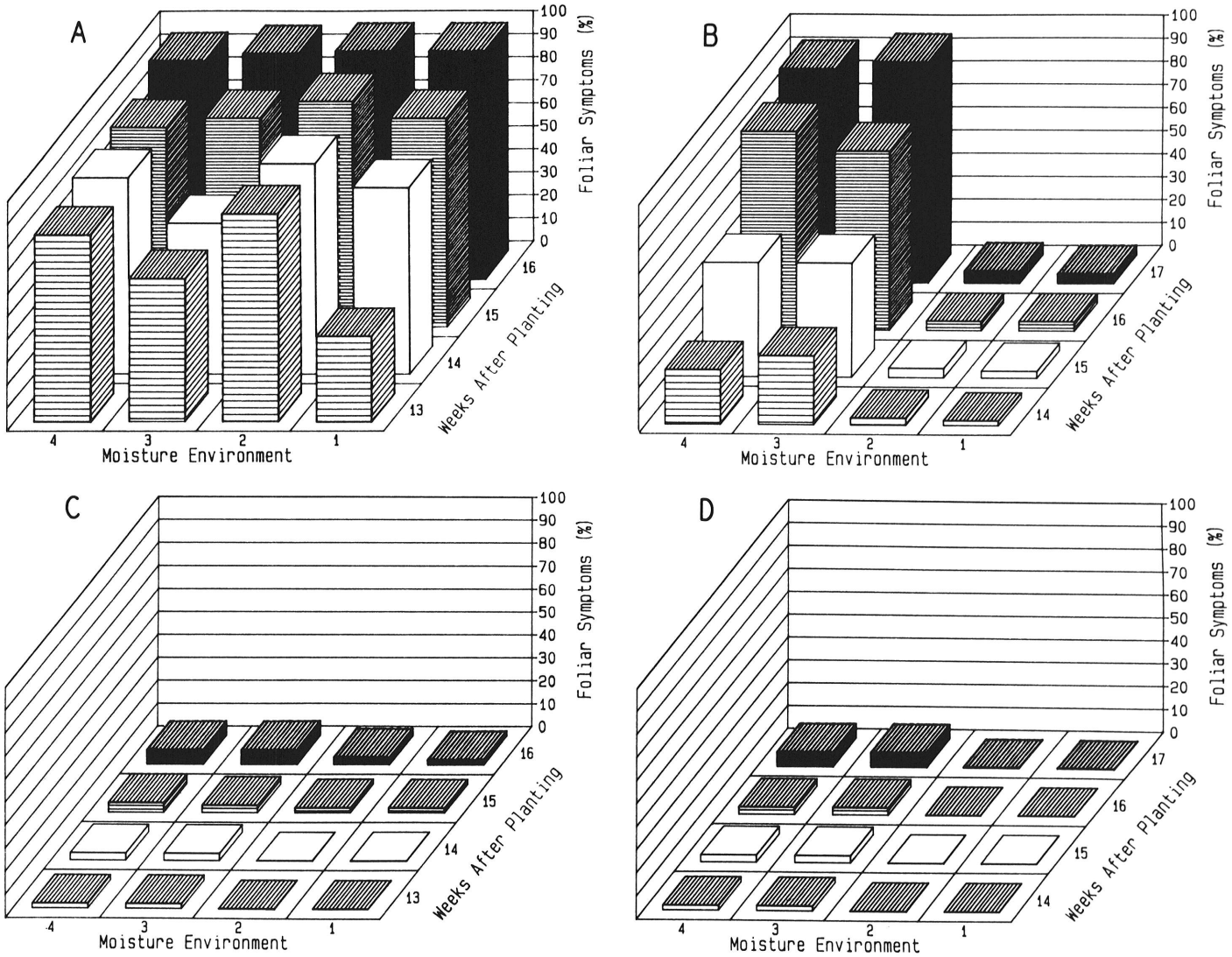


**Fig. 4.** Severity of foliar symptoms for Corsoy 79 grown under four irrigation regimes at Hancock, WI; **A,** 1982 and **B,** 1983. The X-axis denotes soil moisture environments: 1 = no irrigation; 2 = irrigation at VC–R1; 3 = irrigation at R1–R8; and 4 = irrigation at VC–R8. The Z-axis indicates weeks after planting: 13, 14, 15, 16, and 17 correspond to growth stages R5, R5, R6, R6, and R7, respectively. **A,** Corsoy 79 in 1982 and **B,** 1983; **C,** BSR 201 in 1982, and **D,** 1983.

reproductive phase of growth (week 9) and was affected by irrigation regimes. Internal stem discoloration for Corsoy 79 was significantly greater at weeks 14 and 17 (growth stages R5 and R7) in plots receiving postflower irrigation in both years but was most evident in 1983 (Fig. 3A and B). Cultivar differences were greatest at stages R5 and R7, but differences between cultivars were modified by irrigation regimes (Fig. 3A–D). For example, in 1983, the cultivars did not differ for internal stem discoloration through growth stage R5 (97 days) in plots not receiving postflower irrigation (Fig. 3B and D). The mean percent internal stem discoloration for Corsoy 79 and BSR 201 over all irrigation regimes at stage R7 (week 17) was 70 and 21%, respectively, in 1982; and 86 and 37%, respectively, in 1983 (Fig. 3A–D). Internal stem discoloration for BSR 201 was not greatly affected by irrigation regimes (Fig. 3C and D).

**Severity of foliar symptoms.** Differences between Corsoy 79 and BSR 201 for susceptibility to brown stem rot were magnified when severity of foliar symptoms was used to measure disease. Foliar symptoms were not observed until the R4 and R5 growth stages in 1982 and 1983, respectively (Fig. 4A and B). BSR 201 expressed a maximum of 6% foliar necrosis by growth stage R7 in both years (Fig. 4C and D). In contrast, 90–96% of the foliage was necrotic for Corsoy 79 by the R7 growth stage (Fig. 4A and B). In 1982, plants grown in plots irrigated before flowering expressed greater severity of foliar symptoms at growth stages R5 and R6 (13 and 14 wk) for Corsoy 79, but severity of foliar symptoms was similar by growth stage R7 (17 wk) for all irrigation regimes. In 1983, severity of foliar symptoms expressed at growth stages R5 and R6 (14 and 16 wk) was significantly greater in plots that received postflower irrigation, but, in contrast to 1982, severity of foliar symptoms remained minimal up to maturity in plots not receiving postflower irrigation. Also, Corsoy 79 and BSR 201 did not differ in severity of foliar symptoms when grown in plots not irrigated after flowering in 1983 (Fig. 4B and D).

**Area under the disease progress curve.** An AUDPC was computed for severity of foliar symptoms, internal stem discoloration, disease incidence based on isolation of *P. gregata* from stems, and disease incidence based on percentage of plants that expressed stem symptoms (Tables 1 and 2). Except for disease incidence (based on stem symptoms) in 1983, AUDPC readily differentiated both cultivars for reaction to brown stem rot. Although there were significant differences between cultivars for most measures of AUDPC for the different irrigation regimes, differences were smaller in 1982 than in 1983 (Tables 1 and 2). Significantly greater AUDPC for most measures of disease was obtained for Corsoy 79 and BSR 201 grown in postflower and season-long irrigation regimes when compared with other regimes, but the magnitude of differences was much less for BSR 201.

**Leaf area reduction.** Significant reduction of symptomless leaf area for Corsoy 79 occurred at the R5 growth stage, which corresponded to the onset of foliar symptoms. In 1982, leaf area reduction at the R7 (119 days) growth stage for Corsoy 79 was not affected by irrigation regimes (Fig. 5A) but was increased in postflower irrigation regimes in 1983 (Fig. 5B). In 1982, leaf area was reduced 86–99% by the R7 (119 days) growth stage for all irrigation regimes (Fig. 5A). In contrast in 1983, percent leaf area reductions at the R7 (119 days) growth stage were 97, 76, 45, and 30% for season-long, postflower only, preflower only, and

TABLE 3. Yield, yield components, and plant height of two soybean cultivars grown under four irrigation regimes in a field plot naturally infested with *Phialophora gregata* at Hancock, WI, in 1982 and 1983

| Growth stages irrigation applied | Cultivar | Yield (q/ha) | | 300-seed weight (g) | | Pods per plant (no.) | | Plant height at maturity (cm) | |
|---|---|---|---|---|---|---|---|---|---|
| | | 1982 | 1983 | 1982 | 1983 | 1982 | 1983 | 1982 | 1983 |
| VC–R1 | BSR 201 | 20.4 | 23.0 | 48.6 | 58.2 | 28.6 | 27.4 | 92 | 71 |
| | Corsoy 79 | 14.2 | 19.6 | 45.0 | 50.4 | 26.8 | 25.6 | 107 | 88 |
| R1–R8 | BSR 201 | 21.4 | 30.6 | 47.2 | 55.7 | 38.4 | 43.7 | 96 | 104 |
| | Corsoy 79 | 15.7 | 25.3 | 41.5 | 47.9 | 32.5 | 42.3 | 113 | 109 |
| VC–R8 | BSR 201 | 22.0 | 30.3 | 49.8 | 54.3 | 37.8 | 39.7 | 95 | 103 |
| | Corsoy 79 | 16.2 | 25.0 | 43.1 | 46.8 | 33.6 | 40.0 | 112 | 109 |
| None | BSR 201 | 21.8 | 19.0 | 50.6 | 57.1 | 44.1 | 27.3 | 97 | 65 |
| | Corsoy 79 | 15.7 | 18.3 | 45.2 | 53.0 | 37.2 | 25.6 | 112 | 72 |
| FLSD[x] | | 1.4 | 2.3 | 3.9 | 5.6 | 1.0 | 2.2 | 1.5 | 2.3 |
| FLSD[y] | | 1.0 | 1.5 | 5.8 | 3.5 | 1.5 | 1.4 | 2.2 | 1.5 |

[x] Fisher's least significant difference ($P = 0.05$) between irrigation regimes for the same cultivar.
[y] Fisher's least significant difference ($P = 0.05$) between cultivars for the same irrigation regime.

TABLE 4. Critical point models for yield loss (dependent variable) and measures of brown stem rot (independent variable) at Hancock, WI, in 1982 and 1983

| Independent[a] variable | 1982 | | | | | 1983 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Intercept | Regression coefficient | Standard error of dependent variable | $F$ stat.[b] | $r^2$ adj. (%) | Intercept | Regression coefficient | Standard error of dependent variable | $F$ stat.[b] | $r^2$ adj. (%) |
| X1 PISD(R2) | 28.5 | 0.12 | 2.05 | 0.38 | 3.8 | 2.3 | 0.01 | 9.70 | 1.02 | 3.7 |
| X2 PISD(R4) | 30.6 | −0.13 | 3.47 | 0.54 | 3.2 | 13.1 | 0.17 | 2.50 | 0.67 | 3.0 |
| X3 PISD(R5) | 44.9 | −0.44 | 6.97 | 5.56 | 17.0 | 1.2 | 0.31 | 3.41 | 9.10 | 22.0* |
| X4 PISD(R7) | 31.4 | −0.04 | 11.68 | 0.22 | 4.3 | 1.4 | −0.14 | 5.38 | 1.53 | 11.3 |
| X5 AUDPC(PISD) | −38.0 | −0.53 | 9.30 | 0.99 | −8.3 | −6.2 | 1.05 | 0.23 | 20.80 | 42.0* |
| X6 SFS(R5) | 15.6 | 0.21 | 4.74 | 3.30 | 25.0*[b] | 11.6 | 0.18 | 3.03 | 1.41 | 6.0 |
| X7 SFS(R6) | 3.4 | 0.29 | 10.37 | 2.68 | 18.0 | 1.5 | 0.23 | 3.03 | 12.50 | 31.0* |
| X8 SFS(R7) | −43.1 | 0.68 | 66.44 | 1.08 | 5.0 | 11.9 | 0.04 | 3.17 | 0.89 | 3.9 |
| X9 AUDPC(SFS) | 4.6 | 1.55 | 7.89 | 9.87 | 28.0* | 2.3 | 1.06 | 0.38 | 7.70 | 52.0* |
| X10 PLAR(R7) | −12.3 | 0.45 | 14.25 | 8.47 | 25.0* | −8.0 | 0.32 | 3.09 | 41.70 | 58.0** |

[a] Variables X1 through X4 represent disease based on percent internal stem discoloration (PISD) at growth stages R2, R4, R5, and R7, respectively; X5 represents AUDPC based on PISD; X6 through X8 represent disease based on severity of foliar symptoms (SFS) at growth stages of R5, R6, and R7, respectively; X9 represents AUDPC based on SFS; and X10 represents percent leaf area reduction (PLAR) at growth stage of R7.
[b] The $F$-values significant at $P = 0.05$ are indicated by one asterisk and $P = 0.01$ by two asterisks.

nonirrigated environments, respectively (Fig. 5B). Severity of foliar symptoms at growth stage R6 was significantly correlated with percent leaf area reduction at the R7 growth stage ($P = 0.05$, $r = 0.43$ and 0.71 in 1982 and 1983, respectively). Leaf area reduction was not correlated with AUDPC for internal stem discoloration in 1982, but was correlated ($P = 0.05$, $r = 0.60$) in 1983.

**Yield differences between susceptible and resistant cultivars.** Corsoy 79 and BSR 201 were grown in a noninfested plot at Hancock to compare their relative yield potential in the absence of brown stem rot. Yields of 30 and 35 q/ha were obtained for BSR 201 and Corsoy 79, respectively, but differences in yield between the cultivars were not statistically significant ($P = 0.05$). When yield of Corsoy 79 in each replicate plot was regressed against the corresponding yield of BSR 201, a regression coefficient of 0.97 and $r^2$ value of 90% was obtained.

In infested plots, the yield of Corsoy 79 was 28 and 13% less than that of BSR 201 (Table 3) when averaged across all irrigation regimes at Hancock in 1982 and 1983, respectively. The 300-seed weight for Corsoy 79 was 11 and 12% less than for BSR 201 for both years. Corsoy 79 plants had fewer pods per plant than BSR 201, and these differences were greatest in 1982. Based on 300-seed weight data, seed numbers accounted for more of the yield difference between cultivars.

**Agronomic performance of soybeans grown under different irrigation regimes.** In 1982, yield of Corsoy 79 was 26–30% lower than BSR 201 across all irrigation regimes (Table 3). Soil water potential remained on the average higher than −0.5 bars in all plots regardless of the irrigation regime in 1982. Thus, the desired range of soil water potentials was not achieved because of frequent precipitation. In 1983, however, a soil moisture gradient among plots was achieved by selective irrigation, which was likely due to less precipitation than in 1982. Soil water potential remained higher than −0.5 bar in plots during an irrigation period compared with frequent water potentials of −12 bars in plots during periods of no irrigation. Yields of both cultivars were greatest in plots that received postflower irrigation (Table 3). The lack of preflower irrigation did not affect yield if postflower irrigation was employed. The yield of BSR 201 was greater than Corsoy 79 in all irrigation regimes except for the nonirrigated plots. Yields of both cultivars in nonirrigated plots were low and not significantly different from each other.

**Prediction of agronomic performance of soybean from measures of brown stem rot.** Percent yield loss estimates were regressed against specific measurements of brown stem rot. Data were fitted to a single critical point model, a multiple point model, and an AUDPC model. The best predictors of yield loss were disease measurements associated with foliar symptoms. In these models, AUDPC for severity of foliar symptoms explained 28 and 52% of the total variation for yield loss in 1982 and 1983, respectively (Table 4). Severity of foliar symptoms at some specific growth stages predicted yield loss but accounted for less of the variation compared with an AUDPC (Table 4) or multiple point model (Table 5). Percentage of leaf area reduction at growth stage R7 explained 25 and 58% of the total variation in yield loss in 1982 and 1983, respectively. The coefficient of determination ($r^2$) improved when data for the 2 yr were combined. AUDPC for severity of foliar symptoms (Fig. 6A) and percent leaf area reduction at R7 growth stage (119 days) (Fig. 6B) explained 59 and 73% of the variation in yield loss, respectively, and both measures of disease were linearly related to yield loss.

In general, stem symptoms were less predictive of yield loss than were foliar symptoms. Internal stem discoloration recorded at growth stage R5 was acceptable as a predictor of yield loss, but a seasonal AUDPC was more predictive of yield loss. A multiple point model for internal stem discoloration at the R5 and R7 growth stages explained more variation for yield loss than a single critical point model for each growth stage (Tables 4 and 5). Percent disease incidence based on stem symptoms at specific growth stages or calculated over time (AUDPC) was not a good predictor of yield loss.

**Results at Arlington.** *P. gregata* was isolated at low frequencies from roots and shoots 42 days after planting. Isolation frequency of *P. gregata* from stems was greater for Corsoy 79 than for BSR 201, particularly during the R2–R5 growth stages, but equalized by growth stage R7. *P. gregata* was readily isolated from roots early in the growing season but decreased as plants approached flowering. Internal stem symptoms were first observed for Corsoy 79 at the R4 growth stage. Cultivars were not differentiated based on internal stem discoloration at growth stages R2–R4, but Corsoy 79 and BSR 201 differed for internal stem symptoms at R5 and R7. Foliar symptoms did not appear in both years. Yield between cultivars was not statistically different at the Arlington Experimental Station; yield for Corsoy 79 was 20.9 and 22.3 q/ha and 21.6 and 23.5 q/ha for BSR 201 in 1982 and 1983, respectively.

TABLE 5. Multiple point models for yield loss and measures of brown stem rot in 1982 and 1983 at Hancock, WI

| Independent[a] variable | Regression equation | $F$ stat.[b] | $r^2$ adj. |
|---|---|---|---|
| X1 SFS | 2.6 + 0.16(−R5) + 0.21(−R6) − 0.30(−R7) | 7.02 | 43.0 |
| X2 SFS | 2.35 + 0.18(−R5) + 0.23(−R6) | 4.75 | 41.0 |
| X3 SFS | 2.09 + 0.2(−R5) + 0.50(−R6) − 0.10(−R7) | 6.79 | 49.0 |
| X4 PISD | 1.40 + 0.18(−R5) − 0.04(−R7) | 3.27 | 24.0 |

[a] Variable X1 represents severity of foliar symptoms (SFS) at growth stages of R5, R6, and R7, in 1982; variable X2 represents severity of foliar symptoms at growth stages of R5 and R6, in 1982; variable X3 represents severity of foliar symptoms at growth stages of R5, R6, and R7, in 1983; and variable X4 represents percent internal stem discoloration (PISD) at growth stages of R5 and R7, in 1983.
[b] All $F$-values are significant at $P = 0.05$.

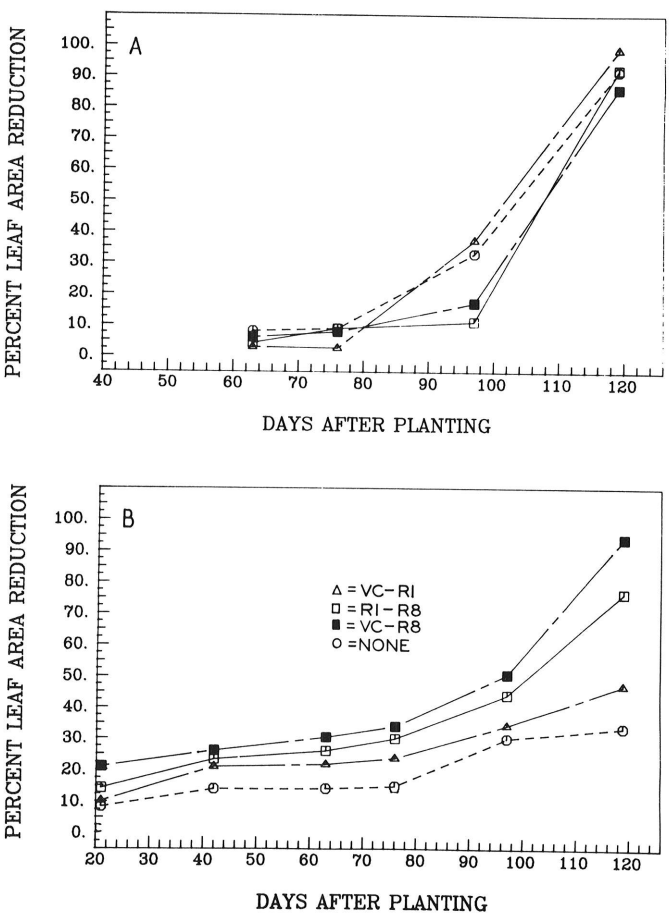


**Fig. 5.** Percent leaf area reduction for Corsoy 79 grown under four irrigation regimes in a naturally infested field plot at Hancock, WI in: **A,** 1982 and **B,** 1983. Days after planting: 21, 42, 63, 76, 97, and 119 correspond to growth stages VC, V4, R2, R4, R5, and R7, respectively.

## DISCUSSION

*P. gregata* was isolated from field-grown plants 3 wk after planting and 21 days earlier than previously reported (12,21). Isolation of the pathogen increased as the crop developed and occurred in advance of internal stem symptoms. Gray and Sinclair (12) did not isolate the pathogen before the occurrence of internal stem symptoms. However, Weber et al (28) speculated that since pod and seed abortion was a major effect of the disease, the pathogen must be affecting the plant before internal stem browning. Although resistance was first reported in 1968 (3), the resistance mechanisms operating in soybean against *P. gregata* have not been determined. Resistant and susceptible plants were similar for disease incidence as determined by isolations and presence of internal stem symptoms, but the advancement of necrosis up the stem and the expression of foliar symptoms were restricted within plants of the resistant cultivar. The same situation has been reported for other resistant genotypes (19).

Dry soil conditions during the preflower period were associated with less recovery of the pathogen and lower disease incidence, and postflower moisture deficits were associated with restricted advancement of internal stem browning and expression of foliar symptoms. This finding is contrary to earlier reports (1,20), which suggested that soil moisture deficits during the reproductive phase favor the expression of symptoms of brown stem rot. Our results are consistent with the hypothesis that fungal pathogens of the vascular system advance upward in the stem largely by spores carried in the transpirational stream, and, therefore, symptom severity is greatest if soil water is readily available (4). This concept is supported by the report of El-Zik (6), who reported an increased severity of both foliar and stem symptoms of Verticillium wilt of cotton with increased applications of irrigation water.

We agree with Sebastian et al (22,23) that foliar symptoms can be used to assess the severity of brown stem rot. Although internal stem symptoms should not be abandoned, assessment of foliar symptoms provided a less labor-intensive measurement of the disease, which resulted in a greater contrast between resistant and susceptible soybean genotypes. Foliar symptoms were correlated with leaf area reduction, particularly at the R7 growth stage. This finding supports the Horsfall-Barratt system to assess subjectively the severity of foliar symptoms, although leaf area reduction was a more quantitative measure for estimating leaf destruction associated with brown stem rot.

Yield loss attributed to brown stem rot was not always associated with a high incidence and severity of internal stem symptoms by growth stage R7. Although disease incidence and severity of internal stem discoloration were high in most environments by growth stage R7, the progress of disease was delayed when drier soil conditions were present. Thus, the seasonal progress of disease development should be considered in brown stem rot assessment. Brown stem rot has been traditionally assessed only once during the growing season and usually at growth stage R7 or later (24). Our results indicate that disease assessments at a specific growth stage do relate to agronomic performance, but multiple disease readings expressed as an AUDPC or by multiple point models (15) improved the predictive relationship between both stem and foliar symptoms and agronomic performance. This does not preclude the importance of disease severity at a specific growth stage, but a multiple point assessment provides a more complete assessment of the epidemic's impact on soybean productivity. The use of an AUDPC using internal stem discoloration was a valuable assessment method under conditions where foliar symptoms were not expressed.

Foliar symptoms have been described for brown stem rot but previously have not been related to its affect on agronomic performance of soybean (1,2). The importance of foliar symptoms was studied by the use of critical and multiple point models that showed measurements of leaf symptoms explained more of the variation for yield loss than did stem symptoms. Similar results have been reported for Verticillium wilt of cotton, a disease with many similarities to brown stem rot (6,17). In our study, data pooled for both years showed that soybeans could withstand a 14% loss in leaf area by growth stage R7 before yield was affected. This finding relates to studies in which foliage was physically removed during the reproductive phase of growth, and foliage loss was related to yield loss (8). Regression of data combined for both years predicted that brown stem rot can reduce yield by 8% in the absence of foliar symptoms. This appears very possible in that much greater yield losses have been reported with no mention of foliar symptoms (5,11,12,16,24,28). However, our results showed that greater yield losses occurred when both foliar and stem symptoms develop in contrast to when only internal stem symptoms are expressed. This is especially the case for results at the Arlington Experiment Station where foliar symptoms were not expressed in both years and a yield difference between cultivars was not detected. Postflower soil moisture deficits were associated with minimal expression of foliar symptoms and a lesser impact of brown stem rot on soybean yield. Future studies could investigate how soil moisture patterns during the postflower phase of plant development affect symptom expression and the impact of brown stem rot on soybean productivity. Future investigations also could involve the relationship between postflower soil moisture and pathotypes within *P. gregata* and other inhabitants of the soybean vascular system (10,11,18).

The approaches for brown stem rot assessment proposed from this study could provide a more quantitative relationship between disease and yield parameters than previously reported. The seasonal progress of brown stem rot can portray a very different

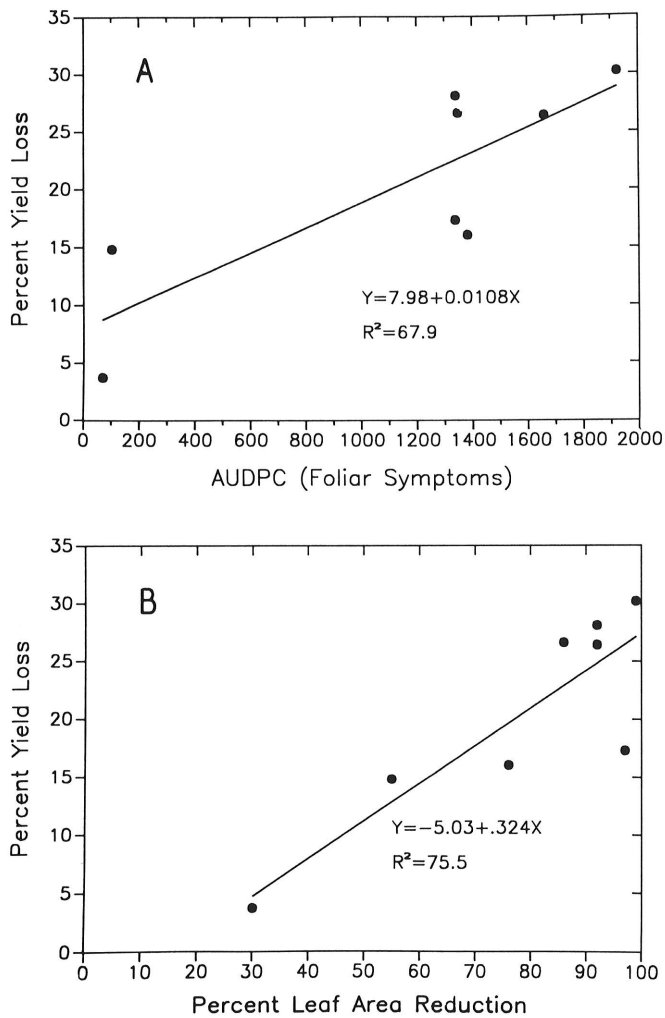


**Fig. 6.** The relationship between measures of leaf disease and percentage of yield loss for data combined for years, 1982 and 1983: **A,** Area under the disease progress curve (AUDPC) using severity of foliar symptoms and percent yield loss ($P = 0.05$) and **B,** percent leaf area reduction at R7 growth stage and percent yield loss ($P = 0.05$).

picture compared with a single-point disease assessment, especially if disease assessments are made after growth stage R6. Multiple point assessments are more common for foliar diseases (15,27), but this approach could lead to a better understanding of disease caused by soilborne pathogens as well (26).

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