Responses to *Septoria glycines* of Soybeans Nearly Isogenic Except for Seed Color

S. M. Lim

Research plant pathologist, Agricultural Research Service, U.S. Department of Agriculture, and Department of Plant Pathology, University of Illinois, Urbana 61801.

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ABSTRACT

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Two distinct types of chlorotic and nonchlorotic lesions on the leaves of soybean plants infected with *Septoria glycines* are associated with plants grown from yellow and green seeds, respectively. This research was undertaken to determine the effects of two lesion types on the development of brown spot and subsequent effect on yield of three derivatives of Clark soybeans nearly isogenic except for seed color (Clark-L-1 [yellow], L64-2545 G d_1d_2, and L62-1027 *cyt*-G [green]) in the field in 1978 and 1979. Treatments included inoculation and control (protected with benomyl application) in both years and a check (natural infection) was added in the 1979 experiments. Brown spot severity was rated three to five times during the reproductive growth stages. Defoliation was also determined by counting the number of defoliated nodes at each disease rating. In both years, there were no significant differences in brown spot severity, apparent infection rate, or number of defoliated nodes between chlorotic and nonchlorotic lesion types of Clark lines inoculated with *S. glycines*. Yield reductions in these isogenic lines ranged from 8 to 11% at Urbana in 1978, 12 to 14% at Urbana in 1979, and 9 to 10% at Brownstown in 1979 compared to yields of the same lines protected with benomyl. However, differences in yield reductions among lines producing two lesion types were not significant for either years. Therefore, resistance and tolerance to brown spot cannot be characterized by these two lesion types associated with seed color.

Additional key words: brown spot, disease reaction, *Glycine max*, soybean disease, soybean yield loss.

Brown spot of soybeans (*Glycine max* (L.) Merr.), which is caused by *Septoria glycines* Hemmi, is one of the most prevalent foliar diseases in the Illinois and other soybean-growing areas of the U.S. (2,8,12). Young and Ross (14) reported that the cultivar Essex, maturity group V (MG V), inoculated with *S. glycines* yielded 17% less than uninoculated Essex soybeans. Williams and Nyvall (11) also reported yield reductions of 17% in the inoculated cultivar Corsoy (MG II) compared with uninoculated Corsoy plants. Yield reduction in two cultivars, Wells (MG II) and Williams (MG III), ranged 12–34%, compared to the yield of plants protected with benomyl (7). The range occurred when different levels of brown spot epidemics were established by inoculating soybean plants at various growth stages.

Cultivars resistant to *S. glycines* are not currently available, although there are differences in the susceptibility of cultivars and germ plasm strains (6). Two distinct types of brown spot lesions on soybeans infected with *S. glycines* have been described (6,13). Angular reddish brown spots surrounded by a yellow area (chlorotic) are associated with plants grown from yellow seeds, and angular dark-brown spots without the surrounding yellow area (nonchlorotic) are associated with plants grown from green seeds (6).

Young and Ross (13) stated that selection for the nonchlorotic lesion type would limit loss of photosynthetic area to the disease. However, chlorophyll retention (green seeds) is an unacceptable characteristic in the processing industry. They considered that the nonchlorotic lesion is a resistant reaction, but that it cannot be utilized in breeding for brown spot resistance unless it is shown to be controlled by genes other than those that control chlorophyll retention in the seeds. The most significant effect of the chlorotic lesions is to cause defoliation of the infected leaves. In a previous study (6), percentage of leaf area diseased of nonchlorotic lesions was lower than chlorotic lesions at R1 growth stage of soybeans (one flower at any node). However, at late R7 (physiological maturity), all plants were severely diseased and there was no difference in brown spot severity between the two lesion types. Additional evaluations of brown spot between the two growth stages would provide information on the rate of disease development for the two lesion types. In some instances (1,4,9,15), tolerance to plant disease is expressed by the differences in yield losses between cultivars with equivalent disease severity. The objectives of this study were to determine the effects of the two lesion types of brown spot on the disease development in the field during two growing seasons, and its effect on yield of Clark soybean lines nearly isogenic except for yellow (chlorotic lesions) or green seed color (nonchlorotic lesions).

MATERIALS AND METHODS

Three Clark soybean lines, nearly isogenic except for yellow seed (Clark-L-1) and green seed (L64-2545 G d_1d_2) controlled by two recessive genes, and green seed (L62-1027 *cyt*-G) controlled cytoplasmically, were studied in 1978 and 1979 at the Plant Pathology Farm, Urbana, IL, on Drummer silty clay loam soil (Typic Haplaquolls) that had been planted to corn (*Zea mays*) the previous year. In 1979, an additional location at Brownstown on Cisne silt loam soil (Mollic Albaqualf) was included.

Four-row plots, 6.1 m in length, were planted in 76-cm row widths on 29 May 1978 and 18 May 1979 at Urbana, and 22 May 1979 at Brownstown at a rate of eight seeds per 30 cm of row. The experiments were replicated four times in a split-plot arrangement of a randomized complete block design in which treatments were main plots and soybean lines were subplots. A check (natural infection) was included as an additional main plot in the 1979 experiments. For field inoculation, inoculum was prepared from 2- to 3-wk-old cultures of *S. glycines* (ATCC 38699) grown on potato-dextrose agar. Cultures were blended in tap water for 2 min and adjusted to a final concentration of 10^5 spores per milliliter. Soybean plants with four to six nodes were sprayed with a spore

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suspension until runoff with a handgun sprayer at 5.6 kg/cm^2 (80 psi). In 1978, control plots were protected with four applications of benomyl 50% WP applied at a rate of 1.1 kg active ingredient per hectare during the reproductive stage (flowering to physiological maturity). There were two applications of benomyl in 1979.

During the reproductive growth stage, disease severity in each plot was rated three times in 1978, four times in 1979 at Urbana, and five times at Brownstown by using the Horsfall and Barratt scale (5). The percent severity (percentage of leaf area infected) was calculated by conversion of disease ratings with Elanco conversion tables (Elanco Products Co., Indianapolis, IN 46140). The soybean growth stage (3) was recorded on each disease rating date. Defoliation was determined by counting the number of defoliated nodes at each disease rating. The regression of disease progress over time for each soybean line was estimated with logit transformation of disease severity (10) based on linear regression. Regression coefficient of logit-transformed values over time is the same as Vanderplank's (10) apparent infection rate (r).

TABLE 1. Brown spot severity, defoliation, and yield of three Clark soybeans nearly isogenic except for seed color

Location, year, and isoline	Treatment	Severity (%) at R6[a]	Defoliated nodes (no.) at R6	Yield q/ha	Yield Loss (%)
Urbana, IL (1978)					
Clark-L-1	IN[b]	62.5	9.3	29.3	8.7
	PT	4.5	7.7	32.0	···
L64-2545 G d_1d_2	IN	64.5	9.7	25.6	10.7
	PT	5.0	8.3	28.7	···
L62-1027 *cyt*-G	IN	64.6	9.7	26.2	8.0
	PT	4.7	8.3	28.4	···
F.L.S.D. ($P = 0.05$)		6.4	1.4	1.6	NS
Urbana, IL (1979)					
Clark-L-1	IN	56.2	9.2	27.2	13.7
	CK	49.9	8.7	28.6	9.5
	PT	22.9	8.5	31.6	···
L64-2545 G d_1d_2	IN	56.2	8.7	24.1	12.0
	CK	56.2	9.0	25.0	9.0
	PT	24.9	8.2	27.4	···
L62-1027 *cyt*-G	IN	56.2	9.0	24.4	12.2
	CK	49.9	9.5	25.7	8.7
	PT	25.7	9.0	27.8	···
F.L.S.D. ($P = 0.05$)		11.7	NS	1.9	4.8
Brownstown, IL (1979)					
Clark-L-1	IN	48.4	7.7	25.7	10.5
	CK	43.7	6.7	26.5	7.7
	PT	17.9	6.2	28.7	···
L64-2545 G d_1d_2	IN	34.4	7.2	22.4	9.7
	CK	32.9	6.7	23.2	6.0
	PT	11.0	6.2	24.8	···
L62-1027 *cyt*-G	IN	39.0	7.7	24.0	9.5
	CK	35.9	6.5	24.5	7.5
	PT	10.1	6.0	26.6	···
F.L.S.D. ($P = 0.05$)[c]		11.5	0.9	1.6	NS

[a]Severity expressed as the percentage of the total leaf area diseased.
[b]IN = inoculated with *Septoria glycines* (ATCC 38699) at growth stages V4–V6. PT = protected with benomyl applications at 1.1 kg active ingredient per hectare during the reproductive growth stage; CK = natural infection.
[c]Fisher's least significant difference.

The center two rows of all plots were trimmed to 4.6 m in length before harvest. Soybean seed was harvested and dried to 7% moisture at 38 C for 72 hr. Seed yields were adjusted to 13% moisture. Percent seed yield reductions were determined by comparisons of seed yields of diseased plots to seed yields obtained from protected plots. The results of 1978 and 1979 were not compared statistically because of the differences in the number of treatments and disease ratings.

RESULTS

The development of brown spot on the three nearly isogenic Clark soybean lines that were tested in inoculated plots in 1978 is given in Fig. 1. The apparent infection rate for brown spot during the reproductive growth stage of R1 (18 July—one flower at any node) through R6 (29 August—pod containing full-size green beans at one of the four uppermost nodes) were not significantly different among lines. Brown spot severity (62–65%) at R6 in inoculated plots were similar among Clark isolines, and defoliation ranged from nine to 10 nodes (Table 1). All three isogenic lines in plots protected with benomyl had brown spot severity of 5%, and seven to eight nodes were defoliated. There were significant differences in disease severity, number of defoliated nodes, and yield between inoculated and protected plants for each isogenic line. Inoculated plots had yield reductions of 8–10.7% when compared with yields in protected plots. No significant differences in yield reductions were found among inoculated isogenic lines.

In 1979, the progress of brown spot was similar among the three isogenic lines in the inoculated plots at each of the two locations (Figs. 2 and 3). Brown spot severity in inoculated and check plots was similar for each isoline at the R6 stage, and there were no significant differences at either location (Table 1). Brown spot severity in both inoculated and check plots of all isogenic lines ranged from 50 to 56% at Urbana and 33 to 48% at Brownstown. Brown spot severity of protected plants was significantly lower than that of inoculated and check plants. At Urbana, no significant difference in severity was found among isogenic lines in inoculated or check plots. There were no significant differences in number of defoliated nodes among isogenic lines and treatments. However, at Brownstown disease severities of L64-2545 G d_1d_2 in both inoculated and check plots were significantly lower than those of Clark-L-1. Also, disease severity of L62-1027 *cyt*-G in the check plot was significantly lower than that on Clark-L-1 in the inoculated plot. Number of defoliated nodes of all three isogenic lines in inoculated

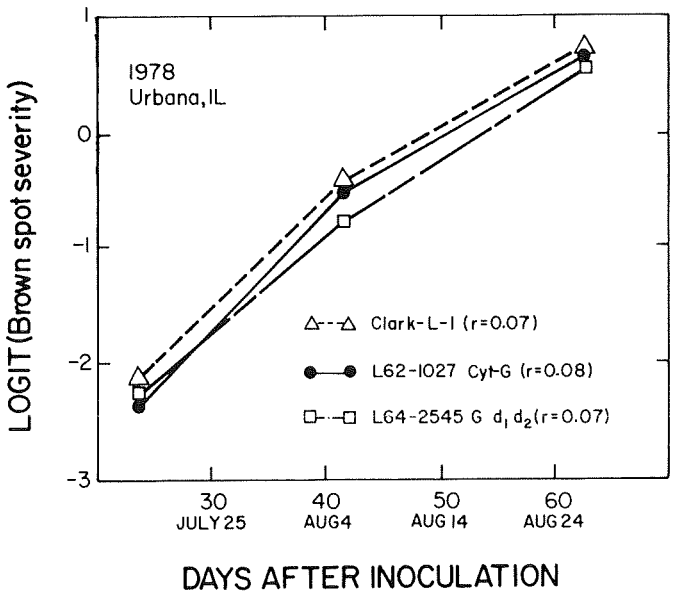

Fig. 1. Progress of *Septoria glycines* on three Clark soybeans nearly isogenic except for seed color at Urbana, IL, in 1978. The r values are apparent infection rates, per unit per day.

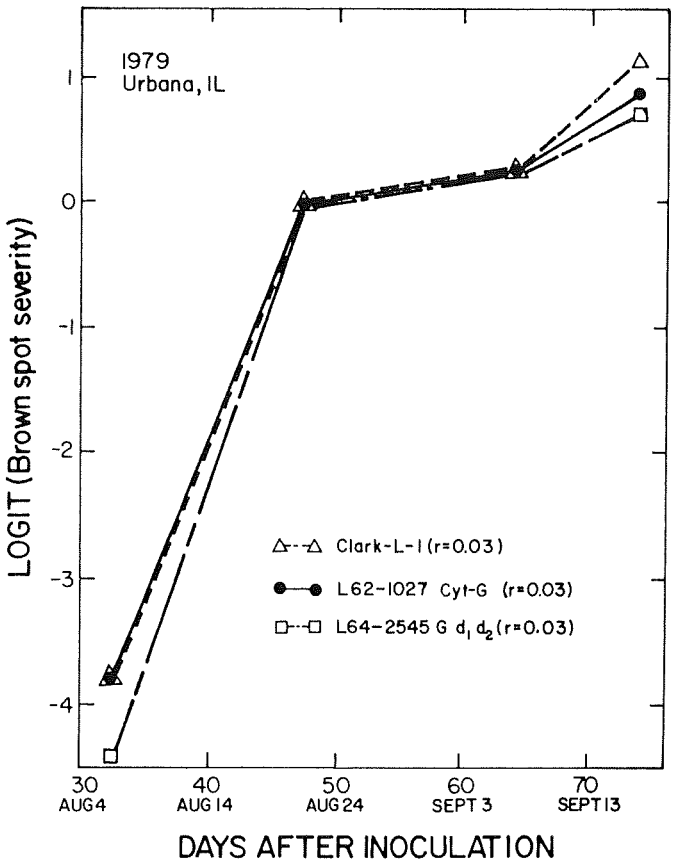

Fig. 2. Progress of *Septoria glycines* on three Clark soybeans nearly isogenic except for seed color at Urbana, IL, in 1979. The *r* values are apparent infection rates, per unit per day.

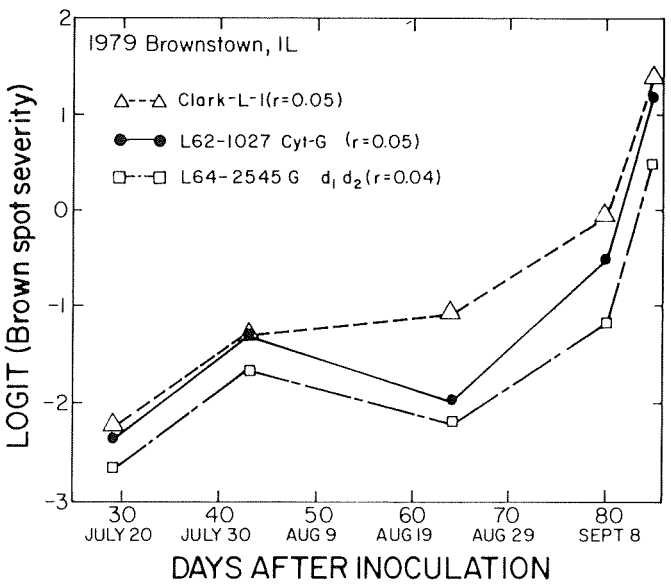

Fig. 3. Progress of *Septoria glycines* on three Clark soybeans nearly isogenic except for seed color at Brownstown, IL, in 1979. The *r* values are apparent infection rates, per unit per day.

plots was significantly greater than that of plants in protected plots. Differences in number of defoliated nodes between inoculated and check plots were significant for Clark-L-1 and L62-1027 *cyt*-G lines. There were no significant differences in defoliated nodes between check and protected plots of any isogenic lines.

Yields in plots of both inoculated and check plants of each isogenic line were significantly less than yields in plots of protected plants (Table 1). Differences in yields of inoculated and check plots were not significant for each isogenic line. Compared with yields of protected plots, yield reductions in both inoculated and check plots of all isogenic lines ranged from 8.7 to 13.7% at Urbana, and 6.0 to 10.5% at Brownstown. There were no significant differences in yield reductions between inoculated or check plots of isogenic lines at either location.

DISCUSSION

Brown spot development on three Clark isogenic lines of soybeans was influenced by weather conditions between years and locations. In 1978, rainfall was adequate at Urbana in June and early July, but there was little precipitation and frequent temperatures above 27 C during late July and early August (flowering and podding stages). Rainfall occurred during late August and September (pod-filling stages) and brown spot increased steadily. In 1979, lack of rainfall at both Urbana and Brownstown during the pod-filling stages probably resulted in little or no increase of brown spot. Consequently, apparent infection rates at both locations in 1979 were lower than those in 1978 at Urbana. Brown spot severities on all three Clark isogenic lines at Brownstown were generally low during the entire growing season of 1979. At a low disease severity, the brown spots surrounded by yellow tissue are more visible on the leaves than the brown spots without the yellow area. The absence of yellowing may have resulted in lower disease ratings on plants with nonchlorotic lesions at Brownstown. As brown spot progresses, the tissue surrounding the nonchlorotic spots becomes wilted following the appearance of a greyish green area. Leaf drop often is similar to that observed for plants with leaves that develop chlorotic spots surrounded by yellow area. The greyish necrotic areas of leaves with nonchlorotic spots and yellow areas of leaves that developed chlorotic brown spots were included in the disease ratings. At Urbana, brown spot severities in all the inoculated plots exceeded 50% at the R6 stage, and there were no significant differences in the severity between chlorotic and nonchlorotic lesion types.

In both years, yield reductions of all isogenic lines in inoculated plots were significant compared with yields of plants protected with benomyl, which allowed comparisons of yield reductions among the three nearly isogenic lines of Clark soybeans. Yields of a Clark line that produced chlorotic spots were significantly higher than those of two other Clark lines that produced nonchlorotic spots for all the treatments: inoculated, naturally infected, and protected. Apparently, the three Clark lines used were not truly isogenic for yielding ability. However, there were no significant differences in yield reductions among the three lines. Disease tolerance, defined as cultivars that appear to be equally susceptible to infection, but do sustain less yield reduction, was not found in this study. The results indicate that resistance and tolerance in soybeans to *S. glycines* can not be distinguished by the chlorotic or nonchlorotic lesion types, which have been shown (6,13) to always be produced on infected plants grown from yellow or green seeds, respectively.

Since 1977, over 6,000 soybean plant introductions (USDA-ARS, Northern Soybean Germ Plasm Collection) have been evaluated for brown spot resistance (S. M. Lim, *unpublished*). Differences in severity have been observed among soybean entries at an earlier growth stage, but such differences are not detectable at later growth stages when all entries are severely attacked. Because of the importance of the disease and lack of resistance to *S. glycines* in commercial cultivars, a search for any form of resistance in the Soybean Germ Plasm Collections is continuing.

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