

# Response of Soybeans and Soybean Pathogens to Soil Fumigation and Foliar Fungicide Sprays

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## ABSTRACT

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Soil fumigation with sodium methylthiocarbamate (metham sodium) and foliar fungicide sprays with methyl 1-(butylcarbonyl)-2-benzimidazolecarbamate (benomyl) were effective in controlling the major soybean pathogens encountered in this study. Fumigation reduced populations of *Macrophomina phaseolina* in residue and of *M. phaseolina*, *Mycoleptodiscus terrestris*, and *Fusarium* spp. in roots. Fumigation also reduced vascular discoloration in stem and roots and increased infection rate of *Septoria glycines* on leaves compared with check plots. Fungicide sprays reduced brown spot severity and *S. glycines* infection rate. Spraying also reduced sporulation on pods and stems of *Phomopsis sojae*, reduced vascular discoloration in root and stems, reduced rate of leaf loss, and increased seed quality compared with control plots. During 3 yr, neither treatment consistently increased yield. When the soil fumigation and fungicide spray treatments were combined, however, there was a large yield increase, averaging 26%, compared with the untreated control.

Additional key words: *Phialophora gregata*

Research on the role of pathogens in crop production has generally been limited to a single host-pathogen interaction. Information generated from such studies is extremely useful in understanding the specifics of the host-pathogen-environment interaction, but has limited application when considering field situations where there may be several pathogens coexisting. Moreover, in studies where interaction of pathogens has been investigated, they are primarily limited to interaction of pathogens that attack the same region of the host, as in combinations of foliar (2,5) or soilborne pathogens (11,12,16), but not the combination of foliar and soilborne pathogens.

Foliar and soilborne pathogens of soybeans may interact in the field. For instance, *Septoria glycines* Hemmi, which causes brown spot of soybeans, is

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prevalent in the Midwest (13), as is *Macrophomina phaseolina* (Tassi) Goid., which causes charcoal rot (19). Stem diseases, such as brown stem rot caused by *Phialophora gregata* (Allington and Chamberl.) W. Gams and pod and stem blight caused by *Phomopsis sojae* Leh., are both common soybean diseases in Illinois (3,7).

The objectives of this study were to control foliar and soilborne pathogen populations with chemicals; to evaluate the effect of these controls, separately and in combination, on foliar and soilborne pathogen populations; and to evaluate host response to changes in pathogen populations resulting from control measures used separately and in combination.

## MATERIALS AND METHODS

Field plots were established in Urbana, IL, in 1977, 1978, and 1979 on a silt loam soil (Aquic Agriudoll) that had been in continuous soybeans for 4 yr. Each plot was 2.7 × 3.3 m and was surrounded by a wooden frame to facilitate securing of the plastic used in fumigation. A soil test showed a pH of 6.7, a phosphorus level of 74 kg/ha, and a potassium level of 430 kg/ha. The plots were fertilized with the equivalent of 336 kg/ha of a 0-30-60 fertilizer each spring.

Plots were fumigated in the spring when soil temperatures were about 15.5 C at 7.5 cm depth with sodium methylthiocarbamate (metham sodium) at 128 ml/m<sup>2</sup>. The fumigant for each plot was diluted with 16 L of water, spread uniformly over the plot, and incorporated

to a depth of about 10 cm with a rotary cultivator. Immediately after the fumigant was incorporated, plots were covered with 6-mil polyethylene plastic, which was secured to the wooden frame to prevent rapid loss of fumigant. Plastic was removed 14 days later and each plot was again rototilled to promote aeration.

Immediately after aeration, soybean residue samples collected from each plot were air-dried and ground in a Wiley mill (#30 screen). A small amount (about 2 mg) of residue was spread uniformly with a microspatula over plates of potato-dextrose agar containing tetracycline at 500 ppm. Plates were incubated for 7 days at 25 C, and colonies of *M. phaseolina* and other fungi were counted. Counts of total fungi and of *M. phaseolina* were expressed as colonies per gram of residue, based on the average weight of residue plated in 10 random blanks.

Each five-row plot contained rows 3 m long planted 51 cm apart. Soybeans (cultivar Wells) were planted on 17 May 1977, 29 May 1978, and 31 May 1979 at the rate of 25 seeds per meter of row and thinned after emergence to the best 15 plants per meter to establish uniform stands. Seeds were coated with commercial *Rhizobium japonicum* (Kirchner) Buchanan inoculum just before planting.

The experiment was arranged in a randomized complete block design with four replications. The treatments were untreated, fumigated, sprayed, and fumigated and sprayed. In 1979, two additional treatments were included: fumigated plus soybean residue and fumigated plus residue with foliar fungicide spray. The amount of residue added to these plots was 2.56 kg each.

At first flower (stage R1 [4]), soil samples 8 cm in diameter and 10 cm deep were removed between plant rows with a tulip-bulb planter. Soybean roots were washed from the soil samples, surface sterilized for 60 sec in 0.5% sodium hypochlorite, and plated onto the potato-dextrose agar medium with tetracycline. Twenty 1-cm-long root sections from each plot were plated onto each of two culture dishes. After 7 days at 25 C, colonies of *M. phaseolina* were counted.

Five fungicide sprays were applied at 10-day intervals beginning at early flower (R2). The fungicide spray consisted of 35 g of methyl 1-(butylcarbonyl)-2-benzimidazolecarbamate (benomyl) in

**Table 1.** Influence of soil fumigation and foliar fungicide sprays on brown spot severity at an early pod stage (R4) of soybean

Treatment	Leaf area diseased (%)			
	1977	1978	1979	3-yr mean
Check	22.7 a <sup>a</sup>	13.2 a	24.2 a	20.0 a
Metham sodium fumigated	18.8 a	12.1 a	14.5 b	15.1 a
Benomyl sprayed	1.6 b	2.3 b	1.2 c	1.7 b
Fumigated and sprayed	1.9 b	2.3 b	1.2 c	1.8 b

<sup>a</sup> Values in columns with the same letter are not significantly different ( $P = 0.05$ ) according to Duncan's multiple range test.

**Table 2.** Effect of soil fumigation and foliar fungicide sprays of soybean on the infection rate of *Septoria glycines* in 1979 based on the regression of disease severity with time

Treatment	Infection rate (slope b) <sup>a</sup>	Coefficient of determination ( $r^2$ )
Check	0.074 b <sup>b</sup>	0.83
Metham sodium fumigated	0.106 a	0.93
Benomyl sprayed	0.026 c	0.62
Fumigated and sprayed	0.026 c	0.62
Fumigated plus residue	0.067 b	0.92
Fumigated plus residue and sprayed	0.018 c	0.30

<sup>a</sup> Regression analysis.

<sup>b</sup> Values in columns with the same letter are not significantly different ( $P = 0.05$ ) according to Duncan's multiple range test.

100 L of water applied at a pressure of 4.2 kg/cm<sup>2</sup> until foliage was wet.

Foliar disease was assessed at an early pod stage (R4) using a modified Horsfall-Barratt grading system (10). Disease severities were based upon an Elanco conversion of these ratings (15). In 1979, additional emphasis was placed upon the foliar disease aspect based on previous observations. The first foliar disease rating was made on 21 July and continued at 7-day intervals until maturity.

Rate of leaf loss and total leaf weight for each treatment were determined by collecting all attached yellow leaves in a 2-m row at 2- or 3-day intervals. It was found from preliminary work that few leaves were lost when a 2- or 3-day schedule was used to collect the leaves. Leaflets and petioles were dried at 70 C to constant weight. Sampling started on 18 August each year and continued until all leaves in the sample row had been collected. The number of nodes defoliated before the start of sampling and the total number of nodes at harvest were determined for each plot.

Rate of pod fill was determined in 1978 and 1979 by sampling three plants from each plot of the check and the fumigated sprayed treatments starting at early pod stage (R3) and continuing at 7-day intervals until maturity. Pods were removed, counted, and weighed after

drying to constant weight. The last three sampling dates were combined for pod number comparisons.

Stem and root disease assessments of five plants randomly selected from the unharvested rows in each plot were taken when 50% of the leaves were yellow (R7). The assessments were based upon the following subjective scale: 1 = no discoloration of taproot and stem vascular tissue, 2 = slight discoloration of either, 3 = slight discoloration of both, 4 = moderate discoloration of either or both, 5 = severe discoloration of either or both, 6 = severe discoloration of vascular tissue and chambering of the pith in the stem.

The amount of fungal sporulation on the third to sixth nodes of stems was measured 1 day before harvest using a subjective scale of 1 to 5: 1 = no fungal fruiting bodies; 2 = slight sporulation on stems, <10% of the stem nodal tissue covered by fungal stromatic tissue; 3 = moderate sporulation, 10–25% of stem nodal tissue involved; 4 = pronounced sporulation, 25–50% of stem nodal tissue involved; and 5 = very dense sporulation on stems, >50% of stem nodal tissue involved. Five randomly selected plants were rated in each plot.

Yield was based on the harvest of two rows 2 m long from each plot, and the weight of 300 seeds was determined. A computer-based system of analysis of variance and Duncan's multiple range test were used (1). Student's *t*-test was performed when only two treatments were compared.

## RESULTS

No sign of phytotoxicity was observed in any fumigated plot. Fumigation resulted in a highly significant reduction each year in the total number of fungal colonies that grew from residue samples, compared with unfumigated plots. The 3-yr means for untreated and fumigated plots were  $3.8 \times 10^4$  and  $5.1 \times 10^3$  fungal colonies produced per gram of residue, respectively. The fumigated plots had no colonies of *M. phaseolina*, and the untreated plots averaged  $1.2 \times 10^3$  colonies per gram.

Fumigation also resulted in a highly significant reduction each year in the percentage of the root sections infected by *M. phaseolina* compared with

untreated plots at first flower (R1). The 3-yr means for untreated and fumigated plots were 17.5 and 0.8% of roots infected, respectively. Fumigation also reduced the levels of *Mycoleptodiscus terrestris* (Gerd.) Ostazeski and various isolates of *Fusarium* spp. observed on plates.

Foliar disease evaluation showed that brown spot was the most prevalent disease. Only trace levels of bacterial blight (*Pseudomonas glycinea* Coerper), downy mildew (*Peronospora manshurica* (Naoum.) Syd. ex Gaum.), and powdery mildew (*Microsphaera diffusa* Cke. & Pk.) were observed. The severity of brown spot in sprayed and fumigated-sprayed plots was significantly less than in other plots (Table 1).

The logit of brown spot severity was plotted over time, and the regression lines were evaluated (Table 2). Disease severity in plots with soybean residue added after fumigation was similar to that in control plots, which suggests that soybean residue in the soil is the source of initial inoculum of *S. glycines*. The fumigated plots had the steepest slope of any treatment and the highest brown spot infection rate. All sprayed treatments had low infection rates; control and fumigated plus residue plots had intermediate infection rates.

Soybeans produced significantly more leaf dry matter in the fumigated-sprayed treatment than other treatments over all 3 yr (Table 3). By regression analysis, the control plots had a significantly higher rate of leaf loss than either sprayed treatment (Table 3). The pattern of overall leaf loss, however, was similar. The number of nodes defoliated at the onset of leaf collection and the total number of nodes per plant at maturity, which averaged 5.4 and 15.8, respectively, were not significantly different. Final plant height, which averaged 75.1 cm, did not differ significantly between treatments.

The rate at which pods increased in mass did not differ significantly between control and fumigated-sprayed treatments. There was, however, a difference between years, but no difference between the two treatments on any one sampling date. The difference ( $P = 0.05$ ) between the 2-yr means of the control and fumigated-sprayed treatments was significant, averaging 36.8 and 41.3 pods per plant, respectively.

The 3-yr mean rating for root and stem discoloration was significantly higher ( $P = 0.05$ ) for plants from the control plot (4.1 mean rating) than for plants from the fumigated, sprayed, or fumigated-sprayed treatments, averaging 2.6, 3.0, and 2.3, respectively. When treatments are compared for the incidence of plants that were rated severely discolored (ratings 5 and 6), the difference between treatments becomes more apparent. The percentage of plants that were severely discolored was 32.9, 17.5, 11.1, and 7.8

**Table 3.** Influence of soil fumigation and foliar fungicide sprays on the total leaf dry matter and rate of soybean leaf loss (3-yr mean)

Treatment	Leaf dry matter (g/m of row)				Rate of leaf loss (slope b) <sup>a</sup>	Coefficient of determination (r <sup>2</sup> )
	1977	1978	1979	3-yr mean		
Check	64.0 bc <sup>b</sup>	88.6 b	59.4 c	70.7 b	4.40 a	0.72
Metham sodium fumigated	59.0 c	88.4 b	69.3 bc	72.2 b	4.28 ab	0.81
Benomyl sprayed	70.4 ab	89.3 b	78.5 b	79.5 b	4.07 b	0.71
Fumigated and sprayed	80.8 a	104.6 a	90.0 a	91.8 a	4.05 b	0.72

<sup>a</sup>Regression analysis.<sup>b</sup>Values in columns with the same letter are not significantly different ( $P = 0.05$ ) according to Duncan's multiple range test.**Table 4.** Influence of soil fumigation and foliar fungicide spray on soybean yields

Treatment	Yield (g/m of row)			
	1977	1978	1979	3-yr mean
Check	155.6 b <sup>a</sup>	177.1 b	143.2 b	158.6 b
Metham sodium fumigated	152.2 b	197.2 ab	172.5 a	174.0 ab
Benomyl sprayed	196.2 a	175.8 b	143.2 b	171.7 ab
Fumigated and sprayed	208.0 a	213.8 a	176.8 a	199.5 a

<sup>a</sup>Values in columns with the same letter are not significantly different ( $P = 0.05$ ) according to Duncan's multiple range test.

for control, sprayed, fumigated, and fumigated-sprayed, respectively. The percentage of plants with healthy roots and stems was inversely correlated—30, 21, 15, and 0% for fumigated-sprayed, fumigated, sprayed, and control treatments, respectively.

*Phomopsis sojae* was the predominant fungus sporulating on stems. The 3-yr mean rating for sporulation on stems and pods was highest for plants in the control (4.4) and fumigated plots (3.4) and significantly lower ( $P = 0.05$ ) in the sprayed (1.3) and fumigated-sprayed plots (1.2). However, a significantly ( $P = 0.05$ ) greater portion of plants from the control plots had dense sporulation (rating 5) than did plants from fumigated plots, averaging 45.0 and 15.6%, respectively. In addition, control plots had significantly fewer plants with no to slight sporulation (ratings 1 or 2) than did fumigated plots averaging 0.0 and 17.2%, respectively.

The fumigated-sprayed treatment had the highest 3-yr mean yield. Fumigated and sprayed treatments had intermediate yields compared with the control plots (Table 4). No significant difference between treatments in the 300-seed weights, which averaged 48.6 g, was observed over the 3-yr test period. Fungicide sprays delayed maturity of treated plots and improved seed quality. A significant interaction between year and treatment occurred for all parameters

evaluated.

## DISCUSSION

High brown spot pathogen pressure was expected in unsprayed plots, based on previous monitoring studies in Illinois (13). Benomyl has previously been reported effective in controlling *S. glycines* (17), the major foliar disease in this study, and *Phomopsis sojae*, which causes pod and stem blight (9,14). The latter was the major component of sporulation examined on soybean stems. Even though yield reduction and loss of seed quality have been attributed to these pathogens (8,20), no overall yield increase resulted from benomyl sprays alone. This is consistent with other work (17) that shows that yield increase in benomyl-sprayed plots can only be expected if foliar disease is the most limiting factor. Increase in seed quality and delayed maturity have previously been observed in soybeans sprayed with benomyl (14,17). The work of Horn et al (9) shows that this is the result of disease control.

Metham sodium applied as a soil fumigant provided good control of soilborne pathogens, as found earlier (6). Fumigation alone can increase yield (6), but under some situations it can reduce soybean yields. These reduced yields have been attributed to phytotoxicity (12) or loss of mycorrhizal fungi in fumigated soil (18).

This study points out the need to be aware of foliar and soilborne disease interactions, especially when trying to define the impact of a single pathogen.

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