

## Interaction Among a Nematode (*Heterodera glycines*), an Insect, and Three Weeds in Soybean<sup>1</sup>

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**Abstract:** A 2 × 3 × 4 factorial field experiment was established to determine the interaction among a nematode, an insect, and three weed species on soybean in 1983-86. Low (nematicide treated) or high (untreated) population densities of the soybean cyst nematode (SCN), *Heterodera glycines*, and 0, 30, or 70% main stem girdling by the threecornered alfalfa hopper (TCAH), *Spissistilus festinus*, were combined with no weeds, one common cocklebur (CC), *Xanthium strumarium*, one sicklepod (SP), *Cassia obtusifolia*, or one pitted morningglory (PMG), *Ipomoea lacunosa*, per meter of row in all possible combinations. Most of the losses from the pests were significant ( $P \leq 0.05$ ) and additive. The high population density of SCN suppressed soybean seed yield by 14%. Girdling of 30 and 70% by TCAH suppressed yields by 10 and 25%, respectively. One CC, SP, or PMG per meter of row suppressed yield by 22, 14, and 12%, respectively. The addition of loss predictions for each pest was approximately the actual treatment losses recorded. The pests did not have an evident interactive effect on yield losses; however, the losses attributed to each pest were additive.

**Key words:** *Cassia obtusifolia*, common cocklebur, *Glycine max*, *Heterodera glycines*, *Ipomoea lacunosa*, pest complex, pitted morningglory, sicklepod, soybean, soybean cyst nematode, *Spissistilus festinus*, threecornered alfalfa hopper, *Xanthium strumarium*, yield loss.

Soybean pests usually occur in combinations that exert biological stresses on soybean, *Glycine max* (L.) Merr., plants by direct injury or by competition for space, moisture, or nutrients. These stresses often result in plant damage and seed yield loss. Damage or competitive thresholds have been established individually for many major soybean pests acting alone or with little regard to interacting or competing organisms. Much information is needed on multiple pest situations.

Nematodes parasitic to soybean, and insects that feed on soybean cause direct injury to the plant that may result in suppression of seed yield, whereas weeds compete for space, moisture, and nutrients and thereby suppress yield. Soybean yield losses caused by the soybean cyst nematode (SCN), *Heterodera glycines* Ichinohe, are documented (8,10,13,17-19). In Arkansas, SCN is the major nematode pest of soybean (17). Many insects also suppress soybean yields (11,14,22). The threecornered alfalfa hopper (TCAH), *Spissistilus*

*festinus* Say, is a common pest of soybean in Arkansas. Many weeds suppress soybean yield through competition for water, nutrients, and sunlight (3,5,9,12,21,23). Three weeds often found in Arkansas soybean fields are common cocklebur (CC), *Xanthium strumarium* L., sicklepod (SP), *Cassia obtusifolia* L., and pitted morningglory (PMG), *Ipomoea lacunosa* L.

In Arkansas, the estimated soybean seed yield loss attributed to SCN has ranged from 3.5 to 6.7% (15,20). Mueller and Jones (14) determined that early season girdling of more than 65% of the main stems (density of 26 plants per meter of row) by TCAH significantly reduced soybean seed yield. In Arkansas one CC, one SP, or one PMG plant per meter of row suppressed soybean seed yield 34, 9, and 6%, respectively (3,5,9). Even though research has shown that each of these pests alone can cause significant soybean yield loss, their interactive effects, if any, have not been characterized. Research was initiated in 1983 to evaluate the interaction among SCN, TCAH, and the three weed species, CC, SP, and PMG, on soybean seed yield potential.

### MATERIALS AND METHODS

The 4-year study initiated in 1983 was conducted at the Cotton Branch Experi-

Received for publication 14 November 1989.

<sup>1</sup> Published with the approval of the director of the Arkansas Agricultural Experiment Station. This work was supported in part by grants from the Arkansas Soybean Promotion Board.

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ment Station, Marianna, Arkansas. The experimental design was a  $2 \times 3 \times 4$  factorial with four replications. The factors evaluated were low (nematicide treated) and high (untreated) levels of SCN; 0, 30, 70% of main stem girdling by TCAH; and no weeds, one CC, one PMG, or one SP plant per meter of row. Bedded plots were  $1 \times 1$  m with a 2-m alley between replications and a border row between plots. The soil was a Calloway silt loam (glossaquic fragiudalfs, 3% sand, 84% silt, 13% clay; < 1% organic matter) with less than 1% slope.

Plots were established in May each year 2–5 weeks before planting. Low population densities of SCN second-stage juveniles (J2) were obtained by soil treatment with 1,3-dichloropropene (1,3-D) at 65.5 liters/ha (injected with a handgun to a depth of 20 cm in the seed row and 30 cm to either side) 25 May 1983, 22 May 1984, 7 May 1985, and 15 May 1986. All plots, alleys, and borders were planted to 'Forrest' soybean with a commercial grain drill with rows spaced 1 m apart. Weed seeds were planted simultaneously in designated plots within 5 cm of the drill row at mid-plot. Planting dates were 8 June 1983, 19 June 1984, 11 June 1985, and 3 June 1986. Two weeks after emergence, soybean plants were thinned to 20 and weeds to one per plot. At the V1 to V3 soybean growth stage (7), fourth-stage or fifth-stage TCAH nymphs (one per plant) were placed on 0, 6, or 14 plants in the appropriate plots. Each nymph was confined to the plant stem by placing it in a clear plastic container (28.4 g) with a lid. The cup-shaped container was modified by cutting opposing 0.5-cm wide slits from the lid opening to halfway down the sides. Two pieces of foam rubber were placed around the stem, then inserted into the slits to prevent nymphal escape and to protect the plant from any sharp edges on the container. After the stem was completely girdled, the nymph and container were removed.

Plots were planted and maintained according to standard agronomic practices recommended by the Arkansas Extension

Service (16). Natural rainfall was the only source of water. Eight 2-cm-d soil cores were taken 15–20 cm deep from the plant row and composited for each subplot at the time of treatment, at planting, 6 weeks after planting, and at harvest. From these samples a 236.5-cm<sup>3</sup> subsample was assayed for nematodes and converted to numbers per 473 cm<sup>3</sup>. Active nematodes were extracted from the soil by the sieving–roiling Baermann funnel technique (6), and the number of SCN J2 per sample was determined. Plots were maintained free of undesirable weeds. Annual soybean seed yield data were subjected to an analysis of variance, and least significant difference at the 5% probability level was used to separate means.

#### RESULTS AND DISCUSSION

Soybean yields (hl/ha) in the pest-free control plots (Table 1), while not as high as desired, were average for dry land soybean in the area. The pest effects were generally significant, but interactions were not, indicating that the pests acted independently with respect to soybean yield.

The soybean seed yields in the nematicide-treated plots were greater ( $P \leq 0.05$ ) than in untreated plots in 3 of the 4 years (Table 1). The 4-year average in the untreated plots resulted in a 14% (LSD 0.05 = 12) yield suppression when compared with the nematicide-treated plots, regardless of weed species or insect density. The loss is approximately twice the Arkansas state average reported by the nematode crop loss committee (19).

The average SCN J2 population densities in the soil during the growing season for each of the 4 years give an indication of the effect of the nematicide (Table 2). Before nematicide treatment, the average J2 density in the soil was not significantly different between treated (low) and untreated (high) levels, except in 1985. The average early season (at planting and at treatment) J2 densities in 1983 were very low, resulting in yield losses greater than expected. Early season J2 densities in the other 3 years were relatively high and yield

TABLE 1. Annual soybean yield (hl/ha) for main effects: soybean cyst nematode (SCN), threecornered alfalfa hopper (TCAH), and weeds—common cocklebur (CC), pitted morningglory (PMG), and sicklepod (SP)—across all plots, 1983–86.

Main effects	1983	1984	1985	1986
No Pests†				
Control	15.38	11.42	22.17	22.08
SCN				
Nematicide				
Treated	12.42	9.32	17.12	15.81
Untreated	10.06	10.21	14.41	12.28
LSD (0.05)	1.33	NS	1.3	1.66
TCAH				
Girdling				
0	12.28	10.88	16.30	17.96
30%	11.11	10.07	14.36	16.24
70%	10.32	8.40	11.41	13.08
LSD (0.05)	NS	1.30	1.59	2.03
Weeds				
Species (density)				
None	12.71	10.78	18.05	16.23
SP (1/m)	11.91	9.29	15.49	13.34
PMG (1/m)	10.37	10.52	16.28	13.73
CC (1/m)	9.95	8.54	13.49	12.88
LSD (0.05)	1.88	1.51	1.83	2.35

Data are means of four replications.

† For comparative purposes only.

losses were as expected. In 1984 rainfall was low, yields were very low in all plots, and no differences due to SCN were found. The reductions in SCN J2 numbers by the nematicide treatment were 77% at planting and 79% 6 weeks after planting in 1983, 88% and 87% in 1984, 62% and 18% in 1985, and 94% and 71% in 1986. Reductions in J2 population densities in nematicide-treated plots at planting were about as expected, except in 1985. For unknown reasons, the nematicide was not as effective in 1985 as in other years. The population densities at 6 weeks in treated samples were much lower (13–29%) than in the untreated samples, except in 1985 when the nematode population showed a much more rapid recovery (82%). In 1985, even though nematode population densities in the treated plots were high at planting and 6 weeks after planting, yield increase was similar to increases observed in 1983 and 1986. The J2 populations in the soil at harvest were

TABLE 2. Soybean cyst nematode second-stage juveniles (av. no/473 cm<sup>3</sup> soil) from nematicide treated and untreated plots across all treatments at the time of nematicide treatment, at planting, 6 weeks after planting, and harvest, 1983–86.

	At treatment	At planting	6 weeks after planting	At harvest
1983				
Treated	53	13**	306**	33**
Untreated	62	56	1,461	139
1984				
Treated	583	45**	150**	909**
Untreated	616	368	1,143	588
1985				
Treated	798*	187**	2,343	93
Untreated	643	492	2,847	94
1986				
Treated	630	25**	448**	120**
Untreated	509	452	1,555	264

Data are means of four replications.

\*, \*\* = significant difference from untreated at  $P \leq 0.05$  and  $P \leq 0.01$ , respectively.

much lower than the population levels at 6 weeks in all years, except 1984 which was a very dry year. Observations (unpubl.) in Arkansas strongly suggest that the J2 population increase at 6 weeks is due primarily to the hatch of eggs from female egg masses produced in the current season, all of which are assumed to hatch quickly. Late-season declines in the J2 population are likely because of the production of far fewer late-season females and egg masses, maturation of early-season females into cysts with cessation of egg deposition into egg masses, starvation because of failure to infect suitable host roots, and (or) J2 death caused by nematode parasites and predators. The cysts usually enter into a period of dormancy lasting through the winter months. The next spring the eggs of many of these cysts are triggered to hatch by unknown mechanisms. This dormancy and later hatch has been observed in microplots infested with only eggs and J2 (unpubl.).

The J2 is the infective stage of SCN; therefore J2 counts were used to define 1) the infective population densities at the time of treatment, 2) the decrease caused

TABLE 3. Average actual and predicted percentage of soybean seed yield losses† attributed to weeds—common cocklebur (CC), sicklepod (SP), and pitted morningglory (PMG)—threecornered alfalfa hopper (TCAH), and soybean cyst nematode (SCN), 1983–86.

Weed	TCAH girdling (%)	SCN	
		Treated‡	Untreated
None	0	0 (0)	6 (14)
	30	10 (10)	27 (24)
	70	28 (25)	38 (39)
CC	0	27 (22)	31 (36)
	30	26 (32)	41 (46)
	70	45 (47)	56 (61)
SP	0	24 (14)	26 (28)
	30	22 (24)	41 (38)
	70	38 (39)	44 (53)
PMG	0	13 (12)	23 (26)
	30	20 (22)	35 (36)
	70	30 (37)	51 (51)

† The actual percentages of seed yield losses were calculated as follows:  $100\% - \left( \frac{\text{total yield of each treatment over all years}}{\text{total yield of no pest treatment over all years}} \times 100 \right)$ . The predicted percentages of seed yield losses (in parentheses) were calculated by addition of the appropriate losses for each treatment.

‡ 1,3-dichloropropene at 65.5 liters/ha.

by fumigation, 3) the relative increases in the treated and untreated plots near mid-season, and 4) the normal late-season decline. Egg counts are useful for estimating SCN population density (2,4). Egg counts may or may not have been as useful as the J2 counts in our study. However, the reduction of egg numbers probably would not have been as dramatic as the J2 counts. Baermann funnel J2 counts give a measure of the expected hatch and the resulting J2 population level. Differentiation between live and dead eggs is difficult and the total egg counts would be misleading. The fact that J2 were not found in 3% of the samples in the study by Barker et al. (2) suggests erratic distribution. The presence of J2 in the soil of all plots in our study suggests a more uniform distribution. Second-stage juveniles in the soil survive the winter readily in some areas (4). These survivors may be in a starved state and thus would not be as infective as the freshly hatched J2. Where high J2 survival occurs, it may interfere with the reliability of J2 for threshold studies. The J2 in our study probably resulted

primarily from cyst egg hatch; this premise is supported by the fact that winter counts in Arkansas are usually near zero. Thus, J2 that hatch near the time of treatment should be reasonable indicators of infective potential of the SCN population.

There are many problems inherent with nematicide use for attaining low numbers of nematodes in experimental plots, but it was the best option available to us. The 1,3-D was applied at least 14 days before planting but there was still a possibility of phytotoxicity. However, 1,3-D was the most effective nematicide available for our conditions and its use has not resulted in yield increases in the absence of the nematode (1).

Girdled plants respond in five major ways (14); however, in our study the plant responses were combined into three categories relative to yield. The most severe plant response was death, with the plants dying usually within 2–3 weeks after girdling and not contributing to yield. The 4-year average of the death response to TCAH girdled plants was 33%. The second response resulted in suppressed plant yield; this included plants that continued to grow but became weak and spindly, producing few or no pods. Plants that partially broke at the girdle site and lodged on the ground were also included in this response. The tips of the lodged plants continued to grow vertically and usually produced a few pods. An average of 12% of the girdled plants during this 4-year study fit this response. The third response included plants that remained upright, appeared to recover, and produced a full or nearly full complement of pods. This girdled plant response average was 55%.

The 30% TCAH main stem girdling caused a yield loss in 1986 only, whereas the 70% girdling caused a loss ( $P \leq 0.05$ ) in all years except 1983. The 4-year average yield losses of 10 and 25% (LSD 0.05 = 12) occurred at 30 and 70% TCAH main stem girdling, respectively. In irrigated soybean, significant loss was not detected until TCAH had girdled the mainstem of over 77% of the plants (14). The yield re-

ductions at lower levels found in our study may be attributed in part to a lower original seedling density (20 vs. 26 plants per meter) and the reduced ability of undamaged and girdled but upright plants to compensate under nonirrigated conditions. Vigorously growing soybean plants will compensate for injured or dead plants by filling in the vacant areas if moisture and nutrients are not limiting.

The three weed species showed different levels of competitiveness. The most competitive, CC, caused yield suppression ( $P \leq 0.05$ ) each year, whereas the less competitive SP and PMG suppressed yield in only 2 of the 4 years (Table 1). One CC, SP, or PMG per meter of row suppressed soybean yield an average of 22, 14, and 12% (LSD 0.05 = 12), respectively. The vining PMG caused almost as much yield suppression as the open, upright growing SP. The bushy, more vigorously growing CC caused more yield suppression than either PMG or SP. In previous studies (3,12), yield suppression caused by CC ranged from 18 to 34%. In our study, both SP and PMG caused more yield loss than previously reported (5,9); however, increased competitiveness might be expected under the nonirrigated conditions of this study.

Interactions among the three types of pest organisms on soybean seed yield were not significant (Table 1). This suggests that each pest caused a suppression in yield and that the effects were additive. Predictions of average yield loss for the test period were determined by using an additive model which computed percentage loss attributed to each pest present in a treatment (Table 3). The actual losses are approximately those predicted. This further suggests that the effects of the pests in our test are additive. The results of our study indicate that the soybean producer will need to control each pest independently according to the established threshold levels for each pest.

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