Interactions of Herbicides and Nematicides with Root Diseases of Snapbean and Southern Pea

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ABSTRACT

In greenhouse experiments treating soil with trifluralin + dinoseb increased foliage wt and decreased root discoloration of snapbean, but treating soil with ethoprop had the opposite effect. Plants grown in soil treated with all three pesticides were similar to plants grown in nontreated soil. DBCP, trifluralin + dinoseb, and ethoprop reduced growth and increased damping-off of snapbean in soils infested with *Pythium myriotylum*. In contrast, treatment with ethoprop alone increased root rot in soil infested with *P. irregulare*, but treatment with trifluralin + dinoseb reduced root rot. Root rot of snapbean was most severe in soil infested with *Rhizoctonia solani*, or a combination of several fungi, and only treatment with dimethyl tetrachloroterephthalate increased root rot while none of the pesticide treatments decreased root rot. Root disease in snapbean was increased by treating with ethoprop in soils infested with *Fusarium roseum* and foliage wt was reduced in soil infested with *F. solani*. Effects of pesticides on snapbeans in soil infested with *F. oxysporum* or *Sclerotium rolfsii* were variable. Pesticides did not significantly affect root disease and foliage wt in southern pea. Inoculum density of *F. solani* was significantly increased in soil by treatment with trifluralin + dinoseb 1 day after snapbeans were planted, but there were no significant differences when plants were harvested 27 to 28 days later. Pesticides did not significantly influence inoculum density of *F. oxysporum* or *F. roseum*.

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Additional key words: Vigna sinensis, Phaseolus vulgaris, soil temperature.

Multiple pest control is becoming increasingly important in modern agriculture (4), and in many crops herbicides, insecticides, nematicides, and fungicides, separately or in combination, are applied as soil treatments. However, herbicides and nematicides are known to interact with soil fungi and affect root diseases in many crops (1, 2, 14, 17). Trifluralin increased Rhizoctonia root rot in cotton (6, 15), and stimulated respiration of *Sclerotium rolfsii* Sacc (19). Dinoseb inhibited growth of *S. rolfsii* and *F. oxysporum* f. sp. *lycopersici* and *conglutinans* in vitro (7), reduced Fusarium wilt of tomato in nutrient-sand culture (18), and reduced peanut stem rot caused by *S. rolfsii* (10).

Damping-off of cotton seedlings, caused by *P. ultimum,* was reduced by DBCP in greenhouse tests, and the nematicide inhibited growth of *P. ultimum* at 10 μg/g and was fungicidal at 100,000 μg/g in vitro (3). In addition, DBCP inhibited growth of *P. irregulare* and *P. aphanidermatum* at 5,000 μg/g in vitro and reduced populations for 14 days in field plots treated with 3.7 and 9.2 liters/hectare (ha) (5). Inoculum potential of *R. solani* was also reduced by DBCP up to 106 days after planting in field plots of susceptible Spanish peanut, and less damping-off of tomato occurred in infested soil in greenhouse tests (2). In a sandy loam soil incubated in the laboratory, ethoprop had no significant effect on populations of bacteria and fungi, ammonification, or sulfur oxidation, but did stimulate O_2 consumption (26).

My observations on field plots of snapbeans (*Phaseolus vulgaris* L.) and southern peas [*Vigna sinensis* (Torner) Savi] in the sandy soils of the coastal plain of Georgia, indicate that commercially recommended herbicides and nematicides may influence root diseases and plant growth. Root diseases are severe on snapbeans in the summer and fall crops, and to a lesser extent in the spring crops. Southern peas are not as susceptible to root rot as snapbeans, and root injury is infrequent in commercial fields. This research was initiated to determine if soil pesticides have a differential effect on fungi associated with root discoloration and decay in snapbean and southern pea. A preliminary report was previously published (23).

MATERIALS AND METHODS.—Dothan loamy sand (ca. 85% sand, 7.5% silt, and 7.5% clay) from a fallow field was collected in 1972 and stored in cans in a greenhouse 2-8 mo before it was used in two experiments. In a third experiment, a mixture of air-dry loamy sand soils stored indoors 2-3 yr was combined with methyl-bromide-treated Dothan loamy sand. Air-dry soil (ca. 1% moisture) was sieved (10 mm openings) just prior to each experiment. There were < 10 stylet-bearing nematodes per 150 cc of soil. The soil pH was 5.8 and the soil contained ca. 1.6% organic matter. Bulk density was ca. 1.4 g/ml.

Fungi were cultured in flasks of autoclaved cornmeal-sand (CMS) 3:100 w/w. Soil was infested by blending with CMS inoculum 1:720 to 1:1000, v/v in a concrete mixer for 5-10 min. Where several fungi were used, equal volumes of each were combined for one additional treatment. Soil used as a control was not amended. Fertilizer was added to soil as needed to give ca. 20, 100, and 200 μg/g of N, P_2O_5, and K_2O, respectively, including residual fertility.

Pesticide dosages were based on manufacturer's field recommendations, and are stated in active ingredient per hectare: trifluralin - 0.56 kg plus dinoseb - 1.68 kg; ethoprop - 8.97 kg; 1,2-dibromo-3-chloropropane (DBCP)-4.9 kg; and dimethyl tetrachloroterephthlate (DCPA)-8.97 kg. Trifluralin, dinoseb, and DCPA are herbicides and ethoprop and DBCP are nematicides. One kg/ha is equivalent to 0.5 μg/g of soil.

Split-plot experiments with a randomized complete block design were used. Fungi were whole plots and pesticides were subplots. Soil was mixed with the CMS inoculum, then 1,500 ml (2,100 g) was placed in pots 15 cm in diam for controls, DBCP, or DCPA treatments, and 800 ml in the bottom of each pot for subsequent trifluralin treatments. Infested soil was removed from the mixer (700 ml/pot) and retained for later treatment with trifluralin. Soil remaining in the mixer was treated with 4 μg/g ethoprop and 1,500 ml placed in each pot for the ethoprop and 800 ml in each pot for the trifluralin + dinoseb + ethoprop treatments. The ethoprop-treated soil remaining in the mixer was treated with trifluralin and 700 ml placed over the ethoprop-treated soil. Soil saved for the trifluralin treatment was then returned to

TABLE 1. Effect of soil pesticides on foliage wt of 23- to 27-day-old snapbeans and southern peas. Each wt is a mean of one noninfested soil and six to eight soils separately infested with one or more soil-borne fungal pathogens

	Foliage wt (g) by experiment[a]				
	Exp. 1		Exp. 2		Exp. 3
Pesticide[b]	GV 50	PHPE	GV 50	PHPE	A & TG
None	5.1 CD	10.4 A	7.5	9.4	17.1 AB
Ethoprop (E)	3.8 D	8.7 AB	5.3	9.0	16.8 B
Trifluralin (T) + dinoseb (D)	6.7 BC	10.7 A	6.5	8.9	19.6 A
T + D + E	6.4 BC	11.0 A	5.7	8.6	17.1 AB
DBCP			NS		15.8 B
DCPA					15.9 B

[a]GV 50 = Gallatin Valley 50 snapbean; PHPE = Purple Hull Pinkeye southern pea; A = Astro snapbean; TG = Tendergreen snapbean. Numbers followed by the same letter are not significantly different, $P = 0.05$. NS = no significant differences.

[b]Formulations and rates (active ingredient per gram of soil) of pesticides were: ethoprop, 10% granules, 4 μg/g; trifluralin, 44.5% emulsifiable concn (EC), 0.25 μg/g; dinoseb, 51% EC, 0.75 μg/g; DBCP, 70% EC, 2.2 μg/g; and DCPA, 54.7% flowable liquid, 4 μg/g.

TABLE 2. Percent of snapbean plants with < 10% root and hypocotyl discoloration grown in a greenhouse in field soils artificially infested with one or more soil-borne fungi. Mean of three experiments

Fungi	Pesticide			
	Ethoprop (E)	Trifluralin (T) + dinoseb (D)	E + T + D	None
None	41	69	70	48
Fusarium oxysporum	43	58	45	38
F. roseum	26	68	45	52
F. solani	55	53	60	53
Rhizoctonia solani	2	12	18	3
Sclerotium rolfsii[a]	50	36	42	47
Pythium irregulare[a]	42	72	60	42
P. myriotylum[b]	15	12	66	34
Combination of several fungi[b]	5	6	4	3

[a]Used in only two experiments.

[b]Soil was infested with equal volumes of *F. oxysporum, F. roseum, F. solani,* and *R. solani* in each experiment; *P. irregulare, P. myriotylum,* and *S. rolfsii* were included in only two experiments.

TABLE 3. Effect of soil pesticides on foliage wt of 27-day-old snapbeans grown in Dothan loamy sand separately infested 1:1,000, v/v with cornmeal-sand cultures of several soil-borne fungi, or a mixture of fungi. Soil temp 2-3 cm deep were 19-28 C

Pesticide	Foliage wt (g)					
	Control	*Rhizoctonia solani*	*Sclerotium rolfsii*	*Fusarium roseum*	*Pythium myriotylum*	Mixture[a]
None	17.8	13.1 AB	8.9 B	22.3 A	23.5	9.7 AB
Trifluralin (T) + dinoseb (D)	17.0	15.8 A	18.0 A	24.2 A	18.4	17.6 A
Ethoprop (E)	13.1	9.7 AB	17.5 AB	20.6 AB	15.6	15.4 AB
T + D + E	18.3	12.4 AB	18.5 A	12.1 B	20.9	12.5 AB
DBCP	18.5	14.3 A	14.1 AB	17.7 AB	15.5	9.1 AB
DCPA	14.4	4.4 B	15.7 AB	20.8 AB	19.0	6.7 B
	NS				NS	

[a]Soil was infested with *Fusarium oxysporum, F. solani,* and *Pythium irregulare* in addition to the fungi listed in the table. Numbers followed by the same letter are not significantly different, $P = 0.05$. NS = no significant differences.

the mixer and treated with 0.25 μg/g trifluralin and 700 ml placed over the 800 ml of nontreated soil in each pot. Thus, trifluralin was only incorporated into the top 5 cm of soil allowing roots to grow into 6 cm of nontreated soil. Just prior to emergence, dinoseb (0.7 μg/g) was sprayed on the surface of trifluralin-treated soil. In DCPA treatments, 4 μg/g was sprayed on the surface immediately after planting and in DBCP treatments 2.2 μg/g was injected ca. 8 cm deep with a syringe 1 day before planting. Ethoprop was applied in 10% granules, DBCP as a 70% emulsifiable concentrate (EC), trifluralin as a 44.5% EC, DCPA in a 54.7% flowable liquid, and dinoseb in a 51% EC.

Fusarium oxysporum Schlecht, *F. solani* (Mart.) Appel & Wr. f. sp. *phaseoli* (Burk.) Synd. & Hans., *F. roseum* Link, *P. irregulare* Buis., *Sclerotium rolfsii* Sacc., and *Rhizoctonia solani* Kuehn were isolated from snapbean or southern pea, and *P. myriotylum* Drechs. was isolated from peanut.

Snapbean cultivars Tendergreen, GV 50, and Astro, and southern pea cultivar Purple Hull Pinkeye were planted 2.5 - 3.0 cm deep, five seeds per pot. Astro seeds were treated with 1.8 g captan-dieldrin (4:1)/kg of seed, Tendergreen with 1.2 g/kg arasan, and GV 50 with 1.8 g/kg thiram-dieldrin (4:1) and 1.6 g/kg chloroneb. All were commercially treated seed lots. Plants were grown in a greenhouse for 23-27 days and sidedressed with NH_4NO_3 (33% N), 32 μg/g of soil, 17-20 days after planting. Three experiments were run in the greenhouse from August 1972 through March 1973. Average minimum and maximum soil temp 2-3 cm deep were 22 and 32, 16.5 and 28.5, and 22.5 and 25.5 C, respectively.

Stand counts were recorded every 5-7 days. The total fresh wt of foliage in each pot was determined 23-27 days after planting when the third trifoliate leaf was expanding and secondary roots were well developed. Soil was carefully washed from roots and hypocotyls and they were rated according to the percentage of surface area that was discolored or watersoaked, as follows: None =< 2%, slight = 3-10%, moderate = 11-50%, and severe => 50%. Isolations were made from hypocotyls and roots in selected treatments in the first experiment.

All data were analyzed by the least squares analysis of variance, linear correlation, and multiple regression.

In the third experiment, populations of several soil-borne pathogens were assayed by soil dilutions on selective media 1-5 days after soils were infested, and again after plants were harvested 27-28 days later. Populations of the Fusaria were determined with peptone-PCNB agar as modified by Papavizas (16), *Pythium* spp. were assayed on gallic acid media (9), and *Rhizoctonia solani* was assayed with gallic acid media modified with 0.1 ml Lorvek [Pyridine: 2-chloro-6-methyoxy-4-(trichloromethyl pyridine)-]/liter (Dow

Chemical Co., Midland, Michigan 48640). Populations of *Pythium* spp. and *R. solani* were determined only in soil not treated with pesticides, but separately infested with *P. irregulare, P. myriotylum, R. solani,* infested with a combination of equal volumes of several fungi; or noninfested. Nontreated soil and soils treated with ethoprop, trifluralin + dinoseb, trifluralin + dinoseb + ethoprop, DBCP, or DPCA and infested with *F. solani, F. oxysporum, F. roseum,* or a combination of fungi were assayed for *Fusaria*.

The day after planting, 2-3 ml samples of soil collected in each of five areas from the top 3-cm of soil in each pot were combined and mixed. Then 1.5 - 5.0 g of each sample was used for each dilution on selective media. After plants were harvested, the roots were separated from the soil, and the remaining soil mass, with adhering root fragments, was thoroughly mixed. Five samples of 4-6 ml each were randomly collected from each pot, combined, mixed, and 1.5 - 5.0 g assayed on selective media.

RESULTS.—*Snapbeans.*—Ethoprop reduced foliage wt and increased root discoloration in all experiments. In contrast, trifluralin + dinoseb increased foliage wt and decreased root discoloration in two out of three experiments, as compared to controls. Foliage wt of plants from soil treated with trifluralin + dinoseb were greater than plants from soil treated with ethoprop in all experiments. Differences were significant in two experiments (Table 1). Foliage wt of plants grown in soil treated with both the herbicide and the nematicide were usually intermediate between soils separately treated with each.

There were fungi-pesticide interactions (Table 2). Root discoloration and damping-off was increased by *P. myriotylum* when the soil was treated with DBCP, ethoprop, and trifluralin + dinoseb, but it decreased when the soil was treated with trifluralin + dinoseb + ethoprop as compared with nontreated soil. Treating soil with ethoprop did not significantly decrease foliage wt and increase root discoloration in soils infested with *P. irregulare*. In contrast to *P. myriotylum,* trifluralin + dinoseb increased foliage wt in soil infested with *P. irregulare,* and in one experiment significantly decreased root discoloration as compared with nontreated soil.

Root injury and damping-off caused by *F. roseum* was not significantly increased nor foliage wt decreased by ethoprop. Foliage wt was significantly reduced by trifluralin + dinoseb + ethoprop at a soil temp range of 19-28 C (Table 3), but not in the two experiments where soil temp maxima were frequently 28-34 C. However, no research was done on the effects of soil temp on pesticide-root disease interactions.

Root deterioration caused by *F. solani* was not severe in nontreated soil in any of the experiments, but foliage wts were reduced an avg of 51% in soils by ethoprop in the two experiments with avg soil temp maxima of 28.5 and 32 C. Nevertheless, root and hypocotyl discoloration was only increased by ethoprop in one experiment. DCPA treatments significantly increased root discoloration caused by *F. solani* but did not reduce foliage wt.

Pesticide treatments did not increase root rot or reduce foliage wt in soils infested with *F. oxysporum*. Results were variable, but trifluralin + dinoseb + ethoprop did significantly decrease root rot in one experiment in contrast with nontreated soil, and trifluralin + dinoseb increased foliage wt an avg of 40%.

Pesticides had a variable effect on *S. rolfsii*. Trifluralin + dinoseb and trifluralin + dinoseb + ethoprop reduced damping-off and significantly increased foliage wt in infested soils at avg soil temp maxima of 25.5 (Table 3), but not at 28. 5C. Other pesticides did not significantly influence damage by *S. rolfsii*.

The most severe root rot and reduction in foliage wt was caused by *R. solani*. In two of the three experiments *R. solani* significantly increased root rot and decreased foliage wt more than the other fungi. Also, when all fungi in each experiment were mixed together in soil, the root disease symptoms and reductions in foliage wt were similar to those caused by *R. solani* alone. In soil infested with *R. solani,* or all fungi in combination, DCPA significantly reduced foliage wt and increased root discoloration more than trifluralin + dinoseb, but foliage wt in the two pesticide treatments were not significantly different from the control (Table 3). Other pesticides did not significantly influence root damage and foliage wt in soil infested with *R. solani*.

A linear correlation and multiple regression analysis was run on a combination of all data from the snapbean experiments. Pesticide treatments were significantly correlated with numbers of live plants at 23-27 days ($r = 0.24$) and foliage wt ($r = 0.23$) but not with the number of plants with $< 10\%$ root and hypocotyl discoloration. Pesticide treatments did not significantly contribute to R^2 (proportion of the total sum of squares attributable to regression) in snapbean.

Southern pea.—Pesticides did not influence foliage growth (Table 1) or root deterioration in southern pea as much as in snapbean. In the first experiment, trifluralin + dinoseb significantly increased foliage wt more than ethoprop in soil infested with *F. solani,* but neither was significantly different from foliage wt of plants grown in nontreated soil. None of the fungi significantly increased root rot or decreased foliage wt in nontreated soil. In contrast, *R. solani* and the combination of several fungi, significantly increased root discoloration and decreased foliage wt in the second experiment. Root rot symptoms caused by *R. solani* alone were similar to those caused by a mixture of fungi, indicating that *R. solani* was the only fungus that was pathogenic on southern pea. There were no significant fungus-pesticide interactions and pesticides did not effect root discoloration and foliage wt.

Southern pea produced significantly more foliage than snapbean, and there was significantly less root discoloration and damping-off in southern pea than in snapbean in both experiments.

Inoculum density.—Populations of *F. solani, F. oxysporum,* and *F. roseum* averaged 14,130, 23,950, and 10,490 propagules/g of oven-dry soil (PG), respectively, 1 day after planting in soils infested with each fungus, and 27-28 days later populations of each were 14,300, 4,620, and 3,350, respectively. Populations of *F. solani* were significantly increased by trifluralin + dinoseb + ethoprop 1 day after planting (12,970 vs. 6,410), but there were no other significant pesticide effects. No significant differences in populations of the three species of *Fusaria* were induced by different pesticide treatments at harvest (27 days). The populations of the Fusaria were 2- to 3-fold greater than found in naturally infested coastal plain soils (24).

There were no significant differences in populations of *Pythium* spp. in the nontreated soils. Inoculum density varied from an avg of 66-224 PG 1 day after planting to 55-104 PG when the plants were harvested. In nontreated soils infested with *R. solani,* or a mixture of *R. solani* and other fungi, 0.5 - 0.8 PG of *R. solani,* was recovered 1 day after planting, but *R. solani* was not detected when plants were harvested. However, 0.8 PG was recovered in noninfested soil and soils separately infested with *P. irregulare* and *P. myriotylum* when plants were harvested.

Rhizoctonia solani and *F. solani* were frequently isolated from roots and hypocotyls of plants grown in soils separately infested with each fungus, respectively, or a combination of fungi, but less frequently from controls. *Pythium* spp. were irregularly isolated from plants grown in both infested and noninfested soil, but *F. oxysporum, F. roseum,* and *S. rolfsii* were rarely isolated.

DISCUSSION.—Numerous researchers have studied the interactions of herbicides (1, 6, 14, 15, 22) and nematicides (2, 3, 17) with root diseases, but much of the research has been in vitro or in sterile soil, and few workers have studied the interactions of soil pesticides with individual fungi in nonsterile field soil. My research indicates the specific effects that soil pesticides and fungi interacting with the host can have on root disease symptoms, growth of the host, and populations of fungi.

Romig and Sasser found increased root disease in snapbeans grown in *R. solani*-infested fields treated with trifluralin + dinoseb (20), but I found that soil treatment with trifluralin + dinoseb decreased root disease and increased foliage wt in soil infested with either *R. solani* or a mixture of several fungi. I am unaware of other reports of specific interactions of nematicides and herbicides with the fungi which cause root diseases of snapbean or southern pea. In the coastal plain of Georgia *Pythium* spp. are most frequently isolated from hypocotyls and roots of snapbean in the spring and *R. solani* and *F. solani* in the fall (24). *F. oxysporum* is commonly isolated in both seasons, but *F. roseum* and *S. rolfsii* are infrequently isolated. *S. rolfsii* may cause severe root rot in snapbeans and polebeans, but the role of *F. roseum* and *F. oxysporum* in the root rot complex in Georgia coastal plain soils has not been studied.

Trifluralin (25) increased chlamydospore production in *F. oxysporum* f. sp. *vasinfectum,* but I found no increase in inoculum density of *F. oxysporum* or *F. roseum,* and only a temporary increase in *F. solani* in soil treated with trifluralin + dinoseb.

In my tests, trifluralin + dinoseb and trifluralin + dinoseb + ethoprop reduced or increased damage by *S. rolfsii* when avg soil temp maxima were 25.5 C and 28.5 C, respectively. I have only observed southern blight, caused by *S. rolfsii,* on snapbeans in the field in the late summer when avg soil temp maxima 5-cm deep frequently range from 30-35 C.

My research suggests that DCPA may increase root diseases in snapbeans. Other research has shown snapbean seedlings grown in soil treated with DCPA were less susceptible to root-knot nematode (21), but DCPA treatments had no significant influence on total populations of bacteria, actinomycetes, and fungi in field soil (8, 27).

In my tests, DBCP decreased damping-off caused by *P. irregulare* or *R. solani,* but increased damping-off caused by *P. myriotylum*. Nevertheless, differences were not significant with any of the soil fungi. In other research at Tifton in field tests DBCP gave significant control of *Fusarium* wilt of okra (12), but not southern blight of tomato caused by *S. rolfsii* (17). Dosages of DBCP that control nematodes frequently do not control soil fungi (4). Interactions of DBCP with soil fungi merits further consideration, since DBCP is commonly used by growers as a nematicide for snapbeans and polebeans in the coastal plain.

Ethoprop increased root discoloration and damping-off of snapbean more than any pesticide tested, and had the most depressing effect on foliage wt. Nevertheless, ethoprop increased yields of snapbeans 18-26% in field plots of Tifton loamy sand naturally infested with root-knot nematodes (13). Yield of southern pea was also significantly increased by ethoprop, even in the absence of plant parasitic nematodes (11).

The most frequently-used herbicides in snapbeans and southern peas in the coastal plain of Georgia are a preplant incorporation of trifluralin 5-cm deep followed by a surface spray of dinoseb a few days later just prior to emergence. However, some growers prefer to use DCPA as a pre-emergence spray rather than treating with trifluralin + dinoseb. In addition, many growers also use DBCP if pathogenic nematodes are a potential problem. Thus, soil pesticides could increase or decrease the severity of root diseases in fields, depending on the indigenous populations of microorganisms.

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