Antagonistic Interaction of *Heterodera schachtii* Schmidt and *Fusarium oxysporum* (Woll.) on Sugarbeets[1]

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Abstract: In a field experiment, nematicides controlled the disease of sugarbeets caused by *Heterodera schachtii* and *Fusarium oxysporum*. Biocides that were both fungicidal and nematicidal also controlled the disease, but sugar yields were no higher than those obtained with the plain nematicides. In greenhouse experiments, the interaction between *H. schachtii* and *F. oxysporum* was disadvantageous to the nematode. Damage to sugarbeets was less when the fungus and the nematode were present than when only the nematode was present. The fungus inhibited nematode invasion and development in sugarbeet seedlings, thereby decreasing the number of nematodes that matured about 3-fold. *Key Words: Heterodera schachtii, Fusarium oxysporum,* Antagonism, Interaction, Sugarbeet.

Numerous reports of *Heterodera*-fungus interactions (2, 3, 4, 5, 6, 7, 9, 10, 11, 15) have been published but only three of these deal with *H. schachtii* (9, 10, 15) and only two with sugarbeets (9, 10). Positive (2, 3, 4, 5, 15) and negative (7) interactions were reported. *Rhizoctonia* (2, 3, 5, 9, 10) and *Fusarium* (4, 11, 15) were the fungi most often involved with *Heterodera*; *Phytophthora* (15), *Colletotrichum* (2), *Oospora* (3), and grey sterile fungus, GSF (7, 12), were less often implicated.

This is a report on interactions of *H. schachtii* and a mutant of *Fusarium oxysporum* on sugarbeets in field and greenhouse experiments.

METHODS AND RESULTS

One field and five greenhouse experiments were conducted for a study of the *H. schachtii/F. oxysporum* interaction on sugarbeet. The first four greenhouse experiments were conducted to assess the influence of the interaction of *H. schachtii* and *F. oxysporum* on growth of sugarbeet. An additional greenhouse experiment was conducted to determine the intensity of attack of *H. schachtii* on sugarbeet as influenced by *F. oxysporum*.

FIELD EXPERIMENT: Plots were established on Welby fine sandy loam which had been in sugarbeet production 12 of the preceding 14 years. Each year beets were grown the soil was fumigated to control sugarbeet nematode. Besides the high nematode population, there was a moderate level of a mutant form of *F. oxysporum*. Individual plots 3.05 × 15.24 m (10 × 50 ft) were established in a randomized, complete-block design with four replications of each chemical treatment. The common or code name, chemical composition and dosage rates of the pesticides used are included in Table 1. All chemicals were applied 14 days before planting as overall treatments metered through a tractor-mounted, pressure-orifice system and injected 20 to 25 cm (8 to 10 in.) deep with chisels spaced 30 cm (12 in.) apart. Some of the plots were covered with polyethylene film (tarped) for 72 hr after application (Table 1).

Plots were harvested 150 days after planting. The beets from the center two rows of each plot were dug and weighed, and a 10-beet random sample was washed and weighed to determine tare. Sugar percentage was determined from each 10-beet sample. Weights were converted to metric tons per hectare and adjusted to exclude tare.

MBR-CP-PBR, a broad spectrum pesti-

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TABLE 1. Yield of sugarbeets as influenced by preplanting soil treatment with broad spectrum pesticides and standard nematicides.

Common name or Code name[1]	Treatment Chemical composition	Dosage/ha	Stand beets/30 m row	Metric tons/ha Beets	Sugar
1,3-D	1,3 dichloropropene & related chlorinated hydrocarbons	234 liters	55	35.4[2] cdef	5.67 bcdefg
		234 liters tarped	59	42.1 abc	6.57 abc
1,3-D-PBC	80% 1,3-dichloropropene & related chlorinated hydrocarbons, 15% chloropicrin, 5% propargyl bromide	94 liters	57	30.9 def	4.77 defgh
		187 liters	54	37.7 bcd	5.92 bcde
		187 liters tarped	68	44.2 ab	6.81 ab
MBR-CP-PBR	61% methyl bromide, 31% chloropicrin, 8% propargyl bromide	1090 kg tarped	59	37.7 bcd	6.03 abcd
		1635 kg tarped	69	46.8 a	7.26 a
DD	a mixture of 1,2 dichloropropane & 1,3 dichloropropene	234 liters	55	36.1 cde	5.71 bcdef
Control		—	54	22.4 g	3.59 h

[1] Experimental quantities of 1,3-D, 1,3-D-PBC, MBR-CP-PBR were supplied by The Dow Chemical Company; DD by the Shell Chemical Company.
[2] Figures followed by the same letter do not differ significantly at P 0.05.

cide, at 1090 and 1635 kg/ha (200 and 300 lb/acre) tarped; 1, 3-D-PBC at 187 liters/ha (20 gal/acre) tarped; and 1, 3-D, a standard nematicide, at 234 liters/ha (25 gal/acre) tarped gave the highest yields. Differences in sugar yield between these four treatments were not statistically significant. Tarping obviously improved the efficiency of these materials. Improved efficiency not only resulted in higher yields, but good weed control was achieved in all tarped plots, these being nearly free of weeds; non-tarped, treated plots appeared to have as many weeds as the untreated controls. The yield increases in all tarped treatments were apparently caused by improving nematode and weed control; because if improved control of *F. oxysporum* had been the only cause for the yield increases, yields from the fungicidal broad spectrum treatments would have significantly exceeded all others (Table 1).

GREENHOUSE EXPERIMENTS: *Influence of the nematode and fungus interaction on growth of sugarbeet.*—Four greenhouse experiments were conducted to assess the influence of the interaction of *H. schachtii* and *F. oxysporum* on growth of sugarbeet.

In the first experiment of this series, a sandy loam soil heavily infested with *H. schachtii* (150 larvae/cc) was thoroughly mixed and divided into two lots. One lot was fumigated. *F. oxysporum* inoculum, produced in mass culture on nutrient-saturated vermiculite (14), was added to half of each

TABLE 2. Influence of *Heterodera schachtii* and *Fusarium oxysporum* on sugarbeet yield in 15-cm clay pots of soil.

Treatment	Fresh top wt (g)	Dry top wt (g)	Fresh root wt (g)	Dry root wt (g)	% Dry Matter Tops (g)	% Dry Matter Roots (g)
H. schachtii, (app. 150 larvae/cc soil)	43.8[1]	3.79	6.8	1.89	.09	.28
H. schachtii + *F. oxysporum*	52.1	4.46	6.6	1.60	.09	.24
F. oxysporum, (5 cc vermiculite)	69.9	6.22	13.3	2.75	.09	.21
Control	110.5	9.69	22.8	4.66	.09	.20
LSD .05	5.46	.48	1.34	.29		
.01	7.34	.65	1.80	.39		

[1] Mean of 12 replications.

lot of soil at the rate of one part vermiculite to 100 parts soil. Twelve 15-cm pots were filled with soil from each of the four treatments, and sugarbeet seed was planted in each pot. Pots, arranged in randomized blocks, were supported in moist sand in polyethylene pans immersed in a temperature-controlled, waterbath at 25 C. After 10 weeks, fresh and dry weights of roots and tops were obtained.

When *F. oxysporum* was added to soil infested with *H. schachtii*, damage to sugarbeets caused by *H. schachtii* was reduced and fresh plant weights increased. Fresh root weights of nematode only and nematode + fungus treatments were similar, but differences in fresh and dry top weights show that when the fungus was present, weight was reduced less than when the nematode alone attacked the sugarbeets. Percentage dry matter was higher in the roots of beets from the nematode-only treatments than in nematode + fungus treatments. This also shows that the nematode caused more damage alone than it did when competing with the fungus (Table 2).

In a second experiment 64 (1.5-liter) polyethylene containers were filled with fumigated, sandy loam soil. *F. oxysporum* inoculum (10 cc vermiculite/ container) was mixed into the top 5 cm of soil in 32 of the containers before planting. The nematode inoculum consisted of 100 fresh *H. schachtii* cysts placed near the sugarbeet seed at planting. As in the previous experiment, four treatments, nematode only, nematode + fungus, fungus only, and untreated control were used. The containers were arranged in a randomized, complete-block design on a greenhouse bench, and the plants were grown for 18 weeks at 25 ± 3 C after which fresh root weights were obtained.

A third experiment was similar to the second, except that *F. oxysporum* inoculum was applied at 20 cc vermiculite/container and *H. schachtii* at 5000 hatched larvae/container. The plants were grown for 19 weeks in the greenhouse at a mean temperature of 25 C, and then fresh root weights were obtained.

Results of these two experiments were similar to the first, but more definitive. Both experiments showed that each parasite is capable of reducing yields of sugarbeets. Fresh root weights were reduced most by *H. schachtii* alone, whether cysts or larvae were applied. Even though *F. oxysporum* reduced root weights when applied alone, it inhibited *H. schachtii* when the two parasites were applied together. The fresh root weights from the nematode + fungus treatment were higher than those from the nematode-only treatment and lower than the fungus-only treatment. Thus a negative, antagonistic interaction existed, with *F. oxysporum* inhibiting *H. schachtii* in essentially the same way GSF inhibits *H. rostochiensis* (7, 12) (Table 3). It

TABLE 3. Fresh root weights of sugarbeets as influenced by greenhouse infection of *Heterodera schachtii* and *Fusarium oxysporum*.

Parasite	In 1.5 liter container		Fresh root wt (g)	
	Expt. II	Expt. III	Exp. II	Exp. III
H. schachtii	100 cysts	5000 larvae	13.98[1]	12.98[2]
H. schachtii + *F. oxysporum*	100 cysts + 10 cc vermiculite	5000 larvae + 20 cc vermiculite	19.19	20.04
F. oxysporum	10 cc vermiculite	20 cc vermiculite	23.59	24.13
Control	—	—	26.11	27.35
LSD .05			3.70	2.23
.01			4.93	3.07

[1] Mean of 16 replications.
[2] Mean of 12 replications.

is a negative interaction because damage to the plant was less when the two parasites were co-inoculated than when only the nematode, the primary pathogen, is inoculated. The interaction may be termed antagonistic because the fungus interferes with the normal activity of the nematode.

In the fourth experiment of this series, sugarbeets were grown for 16 weeks in water baths at 15, 20, 25, and 30 C. There were four replications of each treatment for each temperature. The *F. oxysporum* added at the rate of 10 cc vermiculite/1.5-liter container was mixed into the upper 5 cm of soil and the nematode at the rate of 10.7 *H. schachtii* larvae/cc soil was added near the seed at planting.

In this experiment, also, *H. schachtii* reduced yields more alone than in combination with *F. oxysporum* at all four temperatures. The fungus-only yields were lower than those from the untreated controls at only one temperature, 15 C (Table 4). Greatest yield reductions occurred at temperatures (25–30 C), which were near optimum for the nematode and fungus. Yields were reduced less at the lower temperatures (15–20 C), which favored the host more than the parasites. Thus, growing sugarbeets at different temperatures did not alter the negative antagonistic relationship between *H. schachtii* and *F. oxysporum*.

Influence of the interaction on nematode invasion of sugarbeet seedlings.—A final greenhouse experiment was conducted to determine the intensity of attack of *H. schachtii* of sugarbeet as influenced by competition with *F. oxysporum*. Sugarbeet seed was washed with running tap water 6 hr, rinsed with distilled water, then placed in fumigated soil in 350-cc styrofoam containers. When the seedlings were 3 weeks old they were thinned to one per container and *H. schachtii* was added at the rate of 6000 larvae/100 cc soil. *F. oxysporum* was added at the rate of 1 cc vermiculite/100 cc soil. At 5, 10, and 20 days after inoculation, seven plants from each treatment were washed, weighed, and stained, and the larvae in the roots were counted.

TABLE 4. Influence of temperature on the yield of sugarbeets infected by combinations of *Heteroda schachtii* and *Fusarium oxysporum*.

Parasite combinations	Fresh Root wt (g)			
	15°	20°	25°	30°
H. schachtii	10.05[1]	10.05	6.35	7.09
H. schachtii + *F. oxysporum*	14.64	12.64	6.95	12.41
F. oxysporum	19.13	25.30	17.10	16.45
Control	19.28	25.23	16.84	15.68
LSD .05	3.00	2.79	3.85	1.90
.01	4.19	3.90	5.39	2.66

[1] Mean of four replications.

TABLE 5. Effect of *Fusarium oxysporum* on the invasion of sugarbeet seedlings by *Heterodera schachtii*. (All differences highly significant.)

Sampling time (days)	Nemas per gram fresh roots	
	Heterodera schachtii	*H. schachtii* + *Fusarium oxysporum*[1]
5	602[2]	214
10	7125	1810
20	1566	544

[1] Inoculated with 1 cc vermiculite per 100 cc soil + 6000 *H. schachtii* per 100 cc soil.
[2] Mean of seven replications.

TABLE 6. Effect of *Fusarium oxysporum* and *Heterodera schachtii* on growth of *Beta vulgaris* seedlings.

Sampling time (days)	Whole plant weight (mg)			
	H. schachtii	*H. schachtii* + *F. oxysporum*	*F. oxysporum*	Control
5	98[1]	205	266	292
10	233	209	814	529
20	210	510	2010	2200

[1] Mean of seven replications.

Approximately three times more *H. schachtii* larvae invaded sugarbeet seedlings that were inoculated with only the nematode than invaded seedlings inoculated with both organisms (Table 5).

Seedling growth was reduced by *H. schachtii* but not by *F. oxysporum* (Table 6).

DISCUSSION

The interaction is similar to that described by DuCharme (1), who found that secondary fungal invaders diminished the severity of damage caused by the burrowing nematode in citrus roots. James (7) found that fungi lowered the hatch of *Heterodera rostochiensis,* decreased the number of cysts produced by the nematode and larval invasion of tomato roots.

Growth reduction of infected plants, as compared with noninfected plants, is the universal symptom of nematode disease (8). Most comparisons of sugarbeet yields decrease with increase in nematode numbers (13).

Although the interaction between *H. schachtii* and *F. oxysporum* on sugarbeets is disadvantageous to *H. schachtii,* it is not sufficient for useful economic suppression of the nematode in sugarbeets. The greenhouse experiments that demonstrate the negative interaction also help explain why the plots treated with 1,3-D nematicide yielded as much as those treated with 1,3-D-PBC and MBR-CP-PBR. The nematode is an obligate, primary plant pathogen, and is therefore vulnerable to the competition from the secondary fungal invader. It is unnecessary to use fungicidal pesticides for economic control of the disease because the nematode is the primary pathogen.

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