

## Influence of *Pratylenchus brachyurus* on the Incidence of Fusarium Wilt in Cotton

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### ABSTRACT

Inoculation of Fusarium wilt-susceptible and -resistant cotton cultivars with *Pratylenchus brachyurus* and *Fusarium oxysporum* f. sp. *vasinfectum* alone and in combination with each other, resulted in wilting of the susceptible, but not the resistant, cultivar. A greater percentage of the plants wilted when the nematode and fungus were applied simultaneously than when the nematode was added 2 weeks prior to the fungus or when the fungus alone was used. *Fusarium* was isolated from all

plants of both cultivars treated with the fungus, but colonization was most extensive in plants simultaneously treated with the nematode-fungus combination. *Pratylenchus brachyurus* reproduced on both cultivars of cotton, and populations were significantly higher on the susceptible cultivar when the nematode and fungus were applied at the same time.

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*Additional key words:* *Gossypium hirsutum* L., nematode-fungus interaction.

The root-lesion nematode, *Pratylenchus brachyurus* (Godfrey) Filipjev & Stekhoven parasitizes cotton throughout Georgia, South Carolina, Louisiana, and Alabama (2, 5). Endo (1) reported that this nematode has a good reproductive potential on cotton. Fusarium wilt of cotton, caused by *Fusarium oxysporum* f. sp. *vasinfectum* (Atk.) Snyd. & Hans. is a disease of major economic importance in Georgia responsible for annual losses of 2 to 10% of the crop (6). Nematode-fungus interactions involving *Fusarium* spp. have been reported for a number of nematodes (4), but *Pratylenchus* spp. have not been shown to increase the incidence of Fusarium wilt. Evidence is presented here to show that *P. brachyurus* is capable of increasing the incidence of Fusarium wilt in cotton.

**MATERIALS AND METHODS.**—Fifty seedlings each of the Fusarium-wilt-susceptible cotton cultivar, *Gossypium hirsutum* L. 'Deltapine Smoothleaf' (DPSL), and the resistant cultivar, Auburn 56 (A56), were grown for 3 weeks in a steam-treated mixture of sand and peatmoss (1:1,v/v) in 6.25-cm clay pots with two seedlings/pot. Seedlings were transplanted with the root-ball intact into 20-cm clay pots with the same soil mixture containing one of the following: noninfested control (C); *F. oxysporum* f. sp. *vasinfectum* (F); *P. brachyurus* (N); *P. brachyurus* and *F. oxysporum* f. sp. *vasinfectum* (N + F); or *P.*

*brachyurus* with *F. oxysporum* f. sp. *vasinfectum* added 2 weeks later (N + 2W + F). We infested soil with *P. brachyurus* by pouring a 10-ml aqueous suspension containing 150 to 300 nematodes into a depression made in the soil immediately before transplanting seedlings. Infestation with *F. oxysporum* f. sp. *vasinfectum* was accomplished by a thorough mixing of 200 cc of a 6-week-old sand-cornmeal (4:1,v/v) culture of the fungus with the soil. Each treatment was replicated 10 times on a greenhouse bench, and arranged in a completely random design. Plants in treatment N + 2W + F were initially transferred to nematode-infested soil, and 2 weeks later, to fungus-infested soil. Plants were examined daily; when Fusarium wilt symptoms appeared, the affected plant was harvested and assayed for the presence of *P. brachyurus* and *F. oxysporum*. After 104 to 114 days, nonwilted plants were removed and assayed.

Nematode assays were made by randomly selecting 10 lateral roots from each plant and staining with 0.05% cotton blue-lactophenol for 1 to 3 min. Fifty 5-cm root sections were examined for nematode density, and recorded as the number of nematodes per 250 cm of root tissue. Isolations were made from the following zones to determine the presence of *F. oxysporum*: top 2 cm of stem; 1 to 3 cm of stem above the soil line; 1 to 3 cm of stem below the soil

TABLE 1. Location of the fungus and the percentage of wilting in two cultivars of cotton inoculated with *Fusarium oxysporum* f. sp. *vasinfectum* and *Pratylenchus brachyurus*

Treatments	Mean number of sections infected with the fungus <sup>a</sup>				
	Plants wilted (%)	Top 2 cm of stem <sup>b</sup>	1-3 cm of stem above soil line	1-3 cm of stem below soil line	Bottom 2 cm of taproot
Deltapine Smoothleaf					
Check	0	0.0	0.0	0.0	0.0
Fungus alone	20	0.9	1.6	2.0	1.7
Nematode alone	0	0.0	0.0	0.0	0.0
Nematode + Fungus	40	2.1	1.9	2.9	2.2
Nematode 2 weeks prior to Fungus	10	0.0	2.2	2.3	1.0
Auburn 56					
Check	0		0.0	0.0	0.0
Fungus alone	0		1.8	1.2	0.4
Nematode alone	0		0.0	0.0	0.0
Nematode + Fungus	0		3.1	0.9	1.1
Nematode 2 weeks prior to Fungus	0		1.6	0.3	0.7

<sup>a</sup> Mean of 10 plants (four sections/plant/zone); LSD (.05) = 1.1 and LSD (.01) = 1.5 for comparisons within rows or columns.

<sup>b</sup> No isolations made from this zone with the Auburn 56 cultivar.

TABLE 2. Percentage of 5-cm root sections infected with *Pratylenchus brachyurus*, and mean number of nematodes per 250 cm of root tissue in two cultivars of cotton

Treatments	No. plants examined	Sections infected with nematodes <sup>a</sup> (%)	Mean no. nematodes per 250 cm of root tissue
Deltapine Smoothleaf			
Check	10	0.0	0
Fungus alone	6	0.0	0
Nematode alone	10	32.4	173
Nematode + Fungus	4	44.5	507
Nematode 2 weeks prior to Fungus	8	24.5	116
Auburn 56			
Check	10	0.0	0
Fungus alone	10	0.0	0
Nematode alone	10	32.8	234
Nematode + Fungus	10	30.0	163
Nematode 2 weeks prior to Fungus	10	42.0	232
LSD (.05)		NS <sup>b</sup>	209
LSD (.01)		NS	279

<sup>a</sup> Based on 50 5-cm root sections/plant.

<sup>b</sup> NS = not significant.

line; and bottom 2 cm of taproot. The upper zone of A56 was not analyzed for the presence of *F. oxysporum*. Four 4-mm sections of tissue from each zone were surface-sterilized in a 0.52% sodium hypochlorite solution for 1 to 3 min, placed on water agar, and incubated at 23 to 25 C. After 4 to 6 days, culture plates were examined under a microscope, and sections with *F. oxysporum* growth were

recorded. Twenty lateral roots from one of the wilted plants in treatment N + 2W + F were examined to determine whether a correlation existed between nematode invasion and fungus recovery.

RESULTS.—Exposure of cotton plants to *P. brachyurus* 2 weeks prior to *F. oxysporum* f. sp. *vasinfectum*-infested soil resulted in 10% wilting of DPSL, as compared to 20 and 40%, respectively, for

the fungus alone or the nematode and fungus combined simultaneously (Table 1). No wilting occurred in A56 with any of the fungus treatments. Nematode population density was significantly higher on DPSL when the nematode and fungus were applied simultaneously than on any other nematode treatment in either cotton cultivar (Table 2).

*Fusarium oxysporum* was isolated from all plants of both DPSL and A56 exposed to the test fungus, and was most frequently isolated from plants simultaneously treated with the nematode-fungus combination. Examination of 20 lateral roots from a wilted plant in treatment N + 2W + F revealed that *F. oxysporum* was present in 6 of the 14 roots containing nematodes, but absent in roots without nematodes. No statistical correlation could be made between nematode invasion and fungus recovery in these roots. The fungus was recovered from the top 2 cm of the stem in 6 of 7 wilted plants, but from 3 of 23 infected but nonwilted plants.

**DISCUSSION.**—Most known nematode-*Fusarium* interactions have increased wilting on both susceptible and resistant cultivars (4). In this study, a low inoculum level of *P. brachyurus* was ineffective in decreasing the resistance of A56 to *F. oxysporum* f. sp. *vasinfectum*, but it did increase the incidence of Fusarium wilt symptoms on DPSL. This suggests that *P. brachyurus* is either not very important in increasing the incidence of wilt, or that a higher inoculum level is required. Nematode counts from stained roots showed that both cultivars were suitable hosts for the nematode.

Porter & Powell (3) reported a higher incidence of Fusarium wilt on flue-cured tobacco when *Meloidogyne* spp. were applied 2 or 4 weeks prior to the fungus, suggesting that some change in host

physiology is necessary before the fungus can become established. This does not appear to be the case with *P. brachyurus* on cotton, since a decrease in wilting and fungus recovery occurred when nematodes were applied 2 weeks prior to the fungus. Thus, the relationship in the current study may be mechanical whereby nematode wounding enhances fungal entry. Our data suggests that *P. brachyurus* is capable of increasing the incidence of Fusarium wilt on susceptible cotton cultivars, but is probably of lesser importance than species of *Meloidogyne*. With simultaneous inoculation of Fusarium-wilt-susceptible cotton with the fungus and nematode, the subsequent enhancement of nematode reproduction is a significant observation which warrants further investigation.

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