Influence of Plant-Parasitic Nematodes on Longleaf Pine Seedlings

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Abstract: Seedlings of longleaf pine (*Pinus palustris*) were grown in 20-cm pots for 5 to 7 months in the greenhouse following inoculation with a high or low level of one of seven species of plant-parasitic nematodes. *Belonolaimus longicaudatus* and *Helicotylenchus dihystera* had no effect on seedling growth. High inoculum densities of *Hoplolaimus galeatus* and *Tylenchorhynchus claytoni* caused a significant reduction of fresh weight of seedling roots. Root and top weights of seedlings grown in soil infested with *Meloidodera floridensis* or *Pratylenchus brachyurus* were significantly less than those of seedlings in noninfested soil. Root growth of seedlings was stimulated by the higher inoculum density of *Scutellonema brachyurum*. *Key Words:* host-parasite relation.

Soil fumigation controlled nematodes and improved growth of longleaf pine (*Pinus palustris* Mill.) seedlings in a south Georgia plantation (7) where more than a dozen species of plant-parasitic nematodes were found in the soils. Since the nematicides used in that study controlled all nematodes, the effect of individual species on growth and development of longleaf pine seedlings was not determined. This information is needed in order to recognize and diagnose problems and recommend control in the field.

The complexity of field conditions makes pot experiments under controlled conditions a better method of defining, measuring and evaluating quantitative relationships between nematodes and host seedlings (9). Of the species collected from the plantation and successfully reared in the greenhouse on various host plants, nine fed and reproduced on longleaf pine seedlings (5, 8), but only one, *Trichodorus christiei* Allen, was subsequently proven pathogenic (6).

Because the results from the plantation study mentioned above suggested that certain plant-parasitic nematodes restricted growth of longleaf pine seedlings, the present greenhouse study was designed to measure the influence of each of seven nematode species on seedling growth.

MATERIALS AND METHODS

Six species of nematodes remaining from pot cultures used to supply inoculum for previous host-range studies (5, 8) were used in the present study: *Helicotylenchus dihystera* (Cobb) Sher, *Hoplolaimus galeatus* (Cobb) Thorne, *Meloidodera floridensis* Chitwood, Hannon and Esser, *Pratylenchus brachyurus* (Godfrey) Filipjev and Schuurmans Stekhoven, *Scutellonema brachyurum* Steiner and *Tylenchorhynchus claytoni* Steiner. In addition, *Belonolaimus longicaudatus* Rau, recovered from forest soils within the natural range of longleaf pine in north Florida and reared on St. Augustine grass (*Stenotaphrum secundatum* Kuntze), was used. Adults and larvae of *H. galeatus, M. floridensis* and *P. brachyurus* in the pot cultures were extracted from infected roots in a mist chamber (2). The other four species were obtained from soil in the pot cultures by a combination elutriator-cottonwool filter method (3).

Stratified and surface-sterilized longleaf pine seed from a mixed seedlot were planted in 20-cm clay pots, each containing 2480 cc of steamed fine sandy loam. One to 2 months after germination, the seedlings were thinned to five/pot and inoculated with water suspensions of nematodes poured into several holes in the soil. Inoculum densities of 1000 and 10,000 nematodes/pot were used for all but two species. Since sufficient inoculum of *S. brachyurum* and *T. claytoni* was not available, inoculum densities of *S. brachyurum* were 1000 and 5700 nematodes/pot; and of *T. claytoni*, 500 and 1500. Water from extraction pans after nematode removal was combined with water from the tubes in the mist chamber and added at 50-ml volumes to the control pots.

All treatments were replicated five or six times. Just prior to inoculation, 0.4 g of commercially available NPK fertilizer (23:19:17) was added to the soil of each pot. The pots were placed on benches in a greenhouse with controlled temperatures ranging from 20 to 32 C and watered as required.

After 5 to 7 months, the seedlings were removed from the pots and the soil was gently

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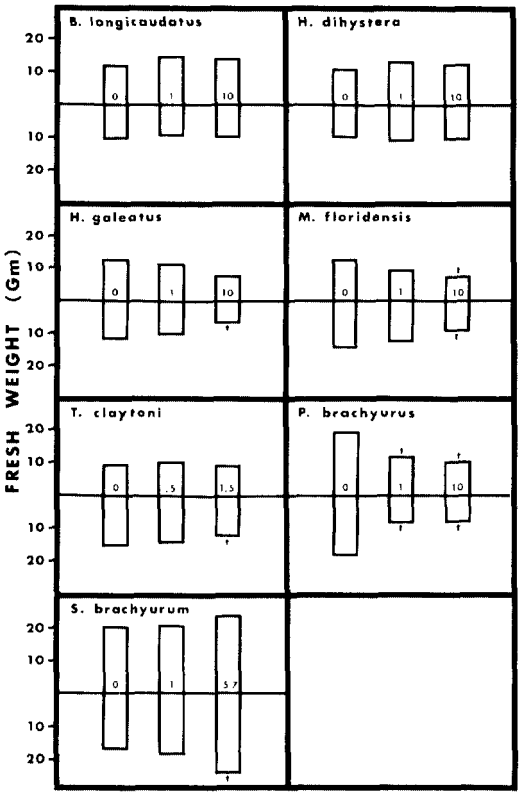

FIG. 1. Fresh weights of the tops (above medians) and roots (below medians) of longleaf pine seedlings 5 to 7 months after inoculation with seven species of plant-parasitic nematodes. Numbers within bars indicate initial inoculum density expressed as thousands, and letters outside bars indicate that group means are significantly different ($P \leq .05$) from the controls by Student's t-test.

shaken from the roots. The roots from each pot were rinsed separately in 4 liters of tap water. Soil remaining in each pot was mixed thoroughly and a 100-cc sample taken. The root washings and soil samples were assayed separately for nematodes by Jenkins' centrifugal-flotation technique (1). Root samples (15 g/pot) were processed for 10 days in a mist chamber to recover nematodes embedded in the roots (2).

Fresh and oven-dry weights (75 C for 24 hr) of the tops and roots of seedlings from each pot were recorded. The stem diameter of each seedling was measured with a caliper just below the needle cluster.

RESULTS AND CONCLUSIONS

Data indicating seedling response to nematodes were basically the same whether assessed as fresh or dry weight of seedlings; therefore, only fresh weights and stem diameters are presented in this paper. *B. longicaudatus* and *H. dihystera* had no effect on top or root growth (Fig. 1). Mean root weights were significantly reduced by 39% in seedlings inoculated with the higher density of *H. galeatus* (10,000/pot) and by 23% in those inoculated with the higher density of *T. claytoni* (1500/pot). Neither of these nematodes reduced top weight. *M. floridensis* at the higher inoculum density (10,000/pot) caused a significant reduction of 39% in top and 36% in root weights (Fig. 1, 2). Plants inoculated with *P. brachyurus* at either density had significantly less top and root weight than did noninoculated plants. At the higher density, this nematode caused the greatest reduction in plant growth—48% in top weight and 56% in root weight—of any of the nematodes tested. The high density of *S. brachyurum* caused a stimulation rather than a reduction in root growth of longleaf pine, since seedlings inoculated with the higher density (5700/pot) weighed significantly more than those of the controls. Although this phenomenon is not uncommon for nematodes which increase root

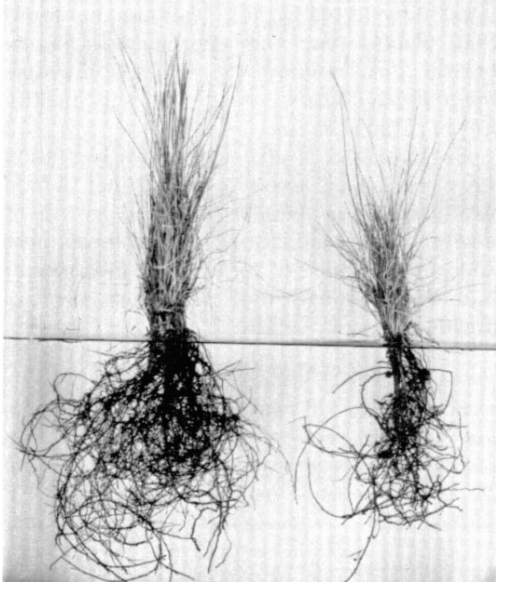

FIG. 2. Bundle of six 7-month-old longleaf pine seedlings from one pot inoculated with 10,000 *Meloidodera floridensis* larvae (right) and six uninoculated control seedlings (left).

weight by galling the roots (9), no galling was evident in seedlings parasitized by *S. brachyurum*. Therefore, these seedlings must have been stimulated to produce additional roots, since numerous actively growing lateral and short roots were observed when seedlings were harvested.

The mean stem diameter (0.68 cm) of seedlings inoculated with the high density of *H. galeatus* was significantly less than the controls (0.92 cm). All other species failed to cause any differences in stem diameters.

The final nematode assays expressed as mean numbers per pot for the low and high inoculum densities were as follows: *B. longicaudatus*, 240 and 590; *H. dihystera*, 1420 and 6600; *H. galeatus*, 950 and 5300; *T. claytoni*, 3750 and 1250; *M. floridensis*, 460 and 700; *P. brachyurus*, 10 and 15; and *S. brachyurum*, 960 and 1200. *H. galeatus, M. floridensis* and *P. brachyurus* were all recovered from roots placed in the mist chamber.

Final nematode densities were highly variable from species to species and apparently had little relation to the effect of nematodes on plant growth. Nematode mortality resulting from handling during inoculation or unfavorable conditions such as fluctuation in soil moisture and temperature may have accounted for some of this variability. Lack of appropriate methods for determining final nematode densities in plant tissue also added to this variability. The presence of mature females noted by direct examination and the recovery of larvae of *M. floridensis* from roots at the completion of this study indicated only that larvae in the inoculum had completed their life cycle. No nematodes were recovered from soil or roots taken from control pots.

This research shows that *H. galeatus, M. floridensis, P. brachyurus* and *T. claytoni* are pathogenic to longleaf pine. These data provide a basis for establishing control recommendations for nematode problems in longleaf pine plantations. Admittedly, the initial inoculum densities used in this study were higher than those found naturally in plantations. Although the potential of nematodes to cause yield losses to susceptible annual crops is generally related to initial population density (4), this relationship may be different with perennial crops. High initial populations account for stunting of recently transplanted seedlings, but lower initial populations may increase to significantly large numbers on a susceptible host during the first few growing seasons and thus stunt seedling growth during the second and third year. Individual species may be few in numbers, but, when several plant-parasitic species are involved, the aggregate in any one locality may constitute a high population. Also, nematode damage to perennials may tend to increase plant response to stresses such as extreme environmental conditions and facultative parasites.

Nematode-host relationships determined in greenhouse studies should logically be followed with studies to determine levels of plant tolerance in relation to economic loss on the basis of field data. Such studies would provide reliable information for making decisions concerning control.

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