Stunting of Southern Pine Seedlings by a Needle Nematode (*Longidorus* sp.)

Michelle M. Cram, Stephen W. Fraedrich, and Jeff Fields

Michelle M. Cram is a Plant Pathologist, USDA Forest Service, Forest Health Protection, 320 Green St, Athens, GA 30602-2044; telephone: 706.559.4233; email: mcram@fs.fed.us. Stephen W. Fraedrich is a Research Plant Pathologist, USDA Forest Service, Southern Research Station, 320 Green St, Athens, GA 30602-2044; telephone 706.559.4273; email: sfraedrich@fs.fed.us. Jeff Fields is a Nursery Coordinator, Georgia Forestry Commission, Flint River Nursery, Route 1 Box 40, Byromville, GA 31007; telephone 912.268.7308.

*Cram M.M., Fraedrich S.W., Fields J. 2003. Stunting of southern pine seedlings by a needle nematode (*Longidorus sp.*). In: Riley L.E., Dumroese R.K., Landis T.D., technical coordinators. National Proceedings: Forest and Conservation Nursery Associations—2002. Ogden, UT: USDA Forest Service, Rocky Mountain Research Station. Proceedings RMRS-P-28: 26–30. Available at: http://www.fcnanet.org/proceedings/ 2002/cram.pdf*

Abstract

An undescribed needle nematode (*Longidorus* sp.) was consistently associated with stunted loblolly pine seedlings at the Flint River Nursery in south Georgia. Seedlings in affected areas had root systems that were greatly reduced in size, and lacked lateral and fine roots. In a growth chamber experiment, the needle nematode significantly reduced the size of loblolly pine root systems in containers initially infested with 100 and 200 nematodes. Populations of the needle nematode increased in all containers during the course of the experiment.

Slash and longleaf pines have also been found to be hosts for the needle nematode. Cover crops typically used at the nursery, including sorghum, wheat and rye, were not found to be hosts. Yellow nutsedge was also not a host.

A survey conducted in pine seed orchards that border the nursery indicated that 37% of the soil samples were positive for the needle nematode. The nematode was not found in redcedar windrows, an oak seed orchard, and pine stands adjacent to the nursery.

Key Words

Pinus taeda, *Pinus elliottii*, *Pinus palustris*, fumigation, seedling stunting

---

Introduction

Areas of stunted pine seedlings have been periodically observed by personnel at the Flint River Nursery (Byromville, Georgia) for over a decade. In 1996 and 1997, we examined stunted loblolly seedlings in block I of field 7S, but the cause of the damage could not be determined. Blocks I and II were being used to test rotating pine with white oak crops (table 1). In 1998, blocks II and III were fumigated and sown with slash pine. According to nursery records, block I was too wet in 1998 to fumigate and was taken out of production. In 1999, stunting recurred in block II and was also found in block III. Seedlings were evaluated for pathogenic fungi and soil samples were forwarded to a nematode testing laboratory for evaluation. In spite of these efforts, the cause of the stunting remained undiagnosed. In the spring of 2000, blocks I-III were fumigated with MC33 (methyl bromide 33%/ chloropicrin 67%) and sown with loblolly pine in 2000 and 2001. Stunting was not observed in blocks I-III in 2000, but in 2001 stunting reappeared in all 3 blocks, with most of the damage in Block II.

A fumigation study was established in 1998 in blocks IV and V of field 7S (Cram and others 2002), which were adjacent to the areas affected by stunting. In the third year of the study (2000), patches of stunted loblolly pine seedlings appeared in nonfumigated plots. The damaged seedlings had poorly developed root systems that lacked lateral and feeder roots (fig. 1). In August 2000, soil samples were forwarded to a nematode laboratory where low levels of lesion (*Pratylenchus* sp.) and ring (*Criconemella* sp.) nematodes were found in both damaged and undamaged areas. Laboratory evaluations of roots for fungal pathogens were also inconclusive. However, during our examination of unwashed roots under the

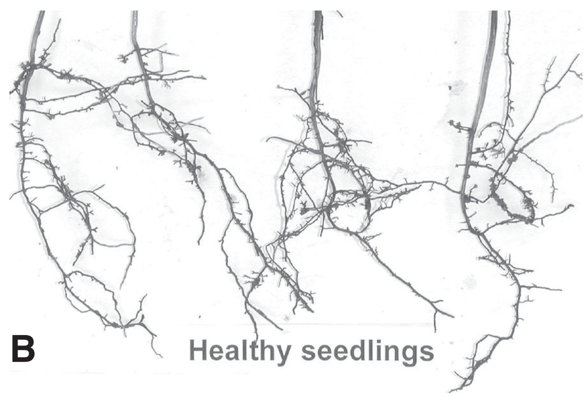
Table 1. Field history of fumigation and crop rotations of pine, hardwoods, and cover crops for blocks I-V in field 7S of Flint River Nursery, Byromville, Georgia.

Date	Block 1	Block 2	Block 3	Block 4	Block 5
1989	**MC33**[a]	**MC33**	**MC33**	**MC33**	**MC33**
1990	Lob [b]	Lob	Lob	Lob	Lob
1991	Lob	Lob	Slash	Slash	Slash
1992	Lob	Lob	Slash	*Basamid*Slash	*MC33*Slash
1993	Oak I Lob	Lob	Slash	Slash	Slash
1994	Oak	Oak	DogwoodISweetgum	Slash	Slash
1995	Oak	Oak	DogwoodISweetgum	Virginia	Virginia I Sand
1996	Lob I Oak	Oak	Lespedeza	Lespedeza	Wheat
1997	Lob	Lob	Sorghum	Sorghum	Sorghum
1998	Fallow	*MC33*Slash	*MC33*Slash	*Fumigation study*Lob	*Fumigation study*Lob
1999	Fallow	Lob	Lob	Lob	Lob
2000	*MC33*Lob	*MC33*Lob	*MC33*Lob	Lob	Lob
2001	Lob	Lob	Lob	Lob	Lob

[a]Methyl bromide 67% / chloropicrin 33%.
[b] Lob=loblolly pine.

dissecting microscope, we routinely observed a needle nematode (*Longidorus* sp.) associated with the stunted seedlings. This nematode had not been reported by the nematode testing laboratory. Samples of the needle nematode were sent to the USDA, ARS, Nematology Laboratory in Beltsville, Maryland, for identification and were determined to be an undescribed species (Handoo 2000). The nematode is large (7 to 8 mm long) and requires extraction techniques specific for large nematodes (Flegg 1967; Shurtleff and Averre 2000). This paper summarizes findings of our field and growth chamber studies to determine the cause of the seedling stunting, and to identify other possible hosts of the needle nematode.

A



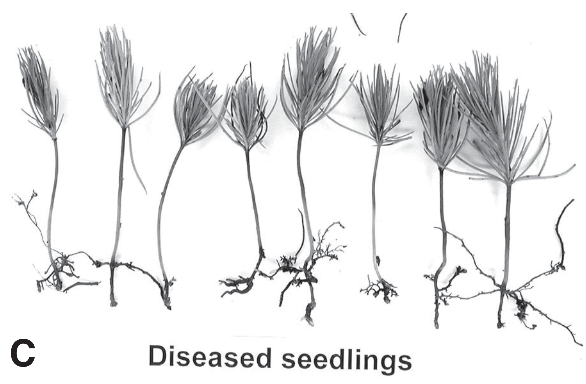


*Figure 1. Patches of stunted loblolly pine seedlings at Flint River Nursery **A**. Root systems of healthy **B**, and diseased **C**, 10-week old seedlings.*

Methods

Identifying the Causal Agent

Field Survey—Patches of stunted seedlings in the fumigation study ranged from 10 to 29.5 ft (3 to 9 m) long and were one seedbed wide. Soil and seedlings were sampled from 4 patches of stunted seedlings in August through October of 2000 (Fraedrich and Cram 2002). Samples were taken in August from the center of the patches, the margins, and 5 to 10 ft (1.5 and 3 m) from the margins. In October, samples were removed from the margins of the patches and 5 ft (1.5 m) from the margins. Nematodes were extracted using a modified Flegg (1967) technique with 90 µm aperture sieves (Fraedrich and Cram 2002).

Growth Chamber Study—The effect of the needle nematode on loblolly pine seedlings was tested by infesting microwaved field soil from Flint River Nursery with 0, 50, 100, and 200 nematodes, and planting 5 germinating loblolly pine seeds per container (Fraedrich and Cram 2002). There were 4 replications for each treatment. Seedlings were removed after 22 weeks and dry weights of the roots and shoots were measured. Population densities of needle nematodes in containers were also determined using the modified Flegg technique (Fraedrich and Cram 2002). Relationships between root dry weight, and initial and final needle nematode populations were determined by regression analysis (Draper and Smith 1981).

Host Range and Survey for the Needle Nematode

Host Range Studies—Initial host range tests were conducted on pine species normally grown at the nursery (Fraedrich and others 2002). The effect of the needle nematode on slash, longleaf and loblolly pine was evaluated by infesting microwaved nursery soil with 0 and 200 nematodes, and planting 5 germinated seedlings in each container. There were 4 replications for each pine species and treatment. Seedlings were removed after 26 weeks, and seedling root and shoot dry weights were measured. The population of needle nematodes was determined for each container.

A second study was conducted to evaluate the host suitability of wheat, rye, sorghum, and yellow nutsedge to the needle nematode. Loblolly pine and a fallow treatment were included for comparison. Containers of microwaved nursery soil were infested with 100 needle nematodes and planted with 5 germinating seeds or tubers (nutsedge) of each species. There were 4 replications for each treatment. Soil was removed from containers, and the needle nematodes were extracted and counted after 12 weeks.

Area Survey—Soils from various locations in and around the nursery were evaluated for needle nematodes in October 2001 and January 2002. Composite soil samples consisting of 8 to 10 cores at a 6 inch (15.2 cm) depth were taken along transects in 3 redcedar windrows, an oak seed orchard, and pine stands on lands adjacent to the nursery. Sixteen composite soil samples were also obtained from loblolly and slash pine seed orchards that border the nursery.

Results

Identifying the Causal Agent

Field Survey—Population densities of needle nematodes were greater in soil samples from the root zone of seedlings at the centers and margins of patches with stunted seedlings than in areas with healthy seedlings at distances of 1.5 and 3.0 m from the margins of patches. In August, 26.6 needle nematodes per 0.8 oz (25 g) soil were obtained from the center of patches, 13.1 at the margins, and 0.5 and 0.2 nematodes at 5 to 10 ft (1.5 and 3 m) from the margins, respectively. In October, evaluations of soil samples from the root zone of seedlings found an average of 25.8 needle nematodes per 0.8 oz of soil from the margins of patches, and 1.5 nematodes/0.8 oz soil at locations 5 ft outside the margin. Nematode extractions from bulk soil samples resulted in an average of 17.5 needle nematodes/6 in^3 (100 cc) in the margins of patches and 2.5 needle nematodes/6 in^3 at 5 ft from the margins. Seedling damage occurred in approximately 4% of the nonfumigated area within the fumigation study in 2000.

Growth Chamber Study—Loblolly pine seedlings in containers with 100 and 200 needle nematodes had damaged root systems similar to stunted seedlings in the field. Roots of affected seedlings lacked lateral and feeder roots. Root dry weight of seedlings decreased with respect to both the initial needle nematode dose (fig. 2A), and the final population per container (fig. 2B). The shoot dry weight of seedlings was not affected by initial or final populations of needle nematodes in containers.

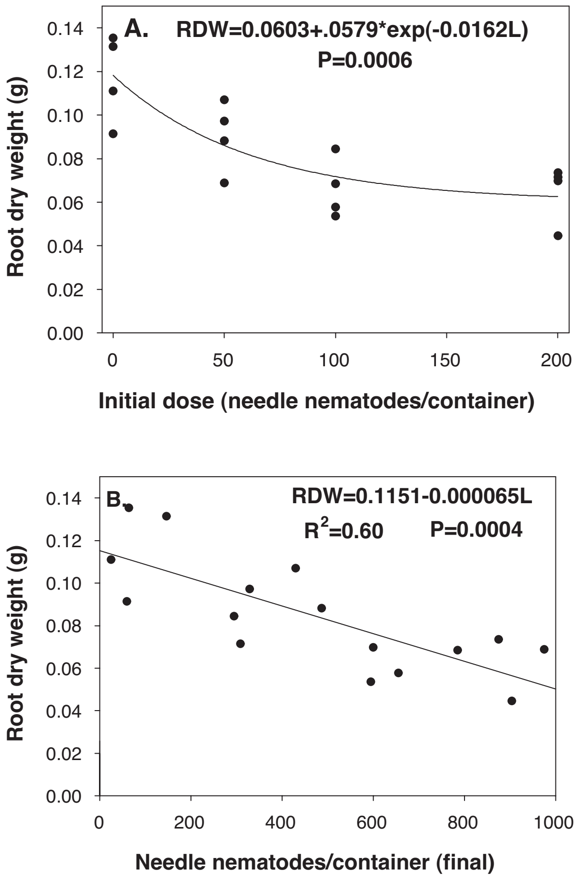


*Figure 2. Relationships between the initial dose of needle nematodes (L) and root dry weight (RDW) of loblolly pine seedlings **A**, and final population of needle nematodes per container and root dry weight **B**. Data was collected 22 weeks after infestation. Each data point represents the mean root dry weight of 5 seedlings per container.*

Host Range and Survey for the Needle Nematode

Host Range Studies—Population densities of the needle nematode increased on slash, loblolly and longleaf pine roots, and reduced the root dry weights of slash and loblolly but not longleaf pine (table 2). The needle nematode population densities decreased substantially in containers with wheat, rye, sorghum, and yellow nutsedge, as well as the fallow containers (table 3).

Area Survey—Surveys conducted in and around the nursery failed to recover the nematode in windrows composed of redcedar, an oak seed orchard, or in pine stands on lands adjacent to the nursery. However, the needle nematode was found in 37% of the soil samples from the loblolly and slash pine seed orchards that border the nursery.

Table 2. Final needle (*Longidorus*) nematode population densites and root dry weights of southern pine species 26 weeks after infestation.

Pine species	Initial needle nematode	Final needle nematode	Root dry weight
	- - - Number/container - - -		g
Loblolly pine	200	1257	0.159*
	0	0	0.295
Slash pine	200	1683	0.334*
	0	0	0.556
Longleaf pine	200	820	0.681
	0	3	0.825

* Means within species are significantly different ($P \leq 0.05$) according to t-test.

Discussion

Needle nematodes have been found in soils where southern pines are grown (Hopper 1958; Ruehle and Sasser 1962); however, there has been no report of these nematodes damaging loblolly pine or any other southern pine. The results of our survey indicate that the needle nematodes found were associated with the stunting of loblolly pine seedlings at Flint River Nursery. The ability of this needle nematode to feed and reproduce on roots of pine was confirmed in growth chamber studies. The nematode can also stunt the root systems of loblolly and slash pines. In growth chamber experiments, the lack of a difference in shoot weight is most likely due to the lack of stress from high summer temperatures that would occur under field conditions. Possible interactions of the

Table 3. Final needle (*Longidorus*) nematode population densities in containers with cover crops, yellow nutsedge, and loblolly pine 12 weeks after infestation with 100 needle nematodes.

Species*	Initial needle nematode	Final needle nematode
	- - - - - - Number/container - - - - - -	
Loblolly pine	100	1089
Sorghum	100	7
Wheat	100	3
Rye	100	1
Nutsedge (yellow)	100	3
Fallow	100	6

*Sorghum – *Sorghum bicolor*; Rye – *Secale cereale*; Wheat – *Triticum aestivum*; Loblolly pine – *Pinus taeda*; Yellow nutsedge –*Cyperus esculentus*.

needle nematode with other biotic and abiotic factors require additional study.

We suspect that the needle nematode was spread to our fumigation study in blocks IV and V by equipment during field preparations for fumigation of field 7S in 1998. The nematode populations appeared to have built up in blocks I and II that had been continually cropped with loblolly pine or white oak from 1990 to 1997 without fumigation. When fumigation was applied to affected blocks in 1998 and again in 2000, the stunting consistently reappeared within these fumigated areas during the second year of pine production. In 2001, soil samples from damaged areas of blocks II and III were processed with the modified Flegg technique (Fraedrich and Cram 2002), and the needle nematode was found consistently associated with stunted seedlings. Species of needle nematodes can occur at soil depths as great as 36 inches (91 cm) and in some soils routinely occur at depths of 24 to 36 inches (61 to 91 cm) (Shurtleff and Averre 2000). The fact that this undescribed needle nematode was able to cause stunting of pine seedlings in the second year following fumigation suggests that this species occurs at depths below the fumigation zone. This factor would make it difficult to manage this nematode with the conventional fumigation routinely used at most nurseries.

Loblolly, slash, and longleaf pines are hosts for the species of needle nematode found at the Flint River Nursery. The host range test indicates that the sorghum, wheat and rye evaluated are not suitable hosts, and therefore, can be used as cover crops in areas affected by this needle nematode. The use of these cover crops, or leaving the fields fallow, would be an important component of an integrated pest management program to manage this nematode. Additional studies are being conducted on the host range of the needle nematode, and survival of the nematode over time without a host.

References

Cram MM, Enebak SA, Fraedrich SW, Dwinell LD. 2002. Chloropicrin, EPTC, and plant growth-promoting rhizobacteria for managing soilborne pests in pine nurseries. In: Dumroese RK, Riley LE, Landis TD, technical coordinators. National Nursery Proceedings—1999, 2000, 2001. Ogden (UT): USDA Forest Service, Rocky Mountain Research Station. Proceedings RMRS-P-24. p 69-74.

Draper NR, Smith H. 1981. Applied Regression Analysis. 2nd ed. New York (NY): John Wiley & Sons Inc.

Flegg JJM. 1967. Extraction of *Xiphinema* and *Longidorus* species from soil by a modification of Cobb's decanting and sieving technique. Annals of Applied Biology 60:429-437.

Fraedrich SW, Cram MM. 2002. The association of a *Longidorus* species with stunting and root damage of loblolly pine (*Pinus taeda* L.) seedlings. Plant Disease 86:803-807.

Fraedrich SW, Cram MM, Handoo ZA. 2002. Host range and distribution of a Longidorus species associated with stunted loblolly pine seedlings. Phytopathology 92:S26.

Handoo ZA. 2000. Personal communication. Beltsville (MD): USDA ARS Nematode Laboratory.

Hopper BE. 1958. Plant-parasitic nematodes in soils of southern forest nurseries. Plant Disease Report 42:308-314.

Ruehle JL, Sasser JN. 1962. The role of plant-parasitic nematodes in stunting of pines in southern plantations. Phytopathology 52:56-68.

Shurtleff MC, Averre III CW. 2000. Diagnosing plant diseases caused by nematodes. St Paul (MN): The American Phytopathological Society.