Auburn University Southern Forest Nursery Management Cooperative

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ASSESSING STUNT NEMATODES ON PINE SEEDLING ROOTS

by
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INTRODUCTION

In the earliest extensive nematode survey, Hopper (1958) found stunt nematodes (*Tylenchorhynchus spp.*) at 8 of 35 southern forest nurseries. He concluded they contributed to stunting at three nurseries where "problem areas" contained ≥ 2,000 stunt nematodes per pint of soil (≥ 400 / 100 cc), but that fewer did not affect seedlings. The pathological severity of nematodes diminished greatly with the routine use of Methyl Bromide. Recently, however, there have been cases of seedling stunting that subsequent soil and seedling analysis indicated was related to the presence of nematodes. In addition, with the potential loss of Methyl Bromide as a nursery soil fumigant it is possible that nematode population dynamics in forest nurseries may change. For this reason we decided to investigate the effectiveness of a nematicide applied over-the-top of loblolly pine. While conducting this study, we also investigated the possibility of using a different laboratory technique for assessing nematode numbers. We have applied this new technique to "operational" samples sent to us from the field. The results of the nematicide study, the techniques investigation, and the operational evaluations are reported here.

METHODOLOGY

Loblolly seed were sown operationally in a Texas nursery on April 14, 1998. Single bed plots (120 by 5 feet) were randomly selected for treatment in areas where soil assays indicated there were abundant stunt nematodes. Oxamyl was applied over seedlings on May 21 at 0.95 lbs ai/acre. About 100 Seedlings were lifted from each plot on June 5 and shipped overnight express to Auburn and processed there without knowledge of treatment.

At Auburn, ten seedlings were randomly selected per sample, stems were removed and root systems placed in 80 mm diameter Petri plates with 25 ml of water. After five minutes, roots were removed, dried and weighed and nematodes in the water ("root-wash") were counted. The nematodes in 5% of each sample were counted using a dissecting microscope. The assay was modified slightly and redone once. The second time, the wash was passed through a 500 μ screen to remove some debris and then through a 43 μ screen to recollect the nematodes.

The technique was later used to count nematodes from a Louisiana and from a Georgia nursery with symptoms characteristic of stunt nematode damage. In August, both seedling and soil samples were obtained from beds in a Louisiana nursery containing both apparently normal and stunted seedlings. Nematodes were separated from the soil samples using the technique of Rodriguez-Kabana and Pope (1981). The Georgia nursery manager sent bulked seedling samples from normal and from stunted sections in each of two nursery blocks in early September.

For the nematicide study, the effects on seedling weights and on the number of nematodes per plant and per gram of root were analyzed using SAS GLM. The association between nematode abundance and root mass was analyzed using SAS regression.

RESULTS AND DISCUSSION

Technique Investigation

During the evaluation of the Texas nematicide study, we observed that stunt nematodes remained on bare-root seedlings after shipment and then separated from the roots when placed in water. This made it possible to assess nematodes per mass of root instead of per volume of soil. The technique for counting stunt nematodes should be of more general interest as a new means of investigation. Quantifying nematodes by root rather than soil volume better evaluates the host/parasite interaction. Unlike bulk-soil assays, numbers of nematodes are directly associated with host root masses so the correlation of their population with seedling size is more precisely evaluated

Counting nematodes washed per unit root avoids both averaging across blocks and on the smaller scale, differences in populations due to available root mass. This is better for stunt nematodes than the more standard bulking of soil samples. Bulking, even across patches of stunted seedlings, determines an average infestation that tends to be below expected thresholds for stunt nematodes (2,000 per pint or 400 / 100 cc). Nematode abundance, as measured by soil assays through the year, and the importance of environmental conditions to symptom development, complicate associating problems with these weakly pathogenic nematodes.

Subsamples from the nematicide study produced essentially the same estimates whether counted directly or screened first to remove debris before counting ($r = 0.97$ $p = 0.0001$). This indicates that nematodes were not lost during screening and that "cleaner" samples can be counted directly without loss of efficiency.

Nematicide Study

Soil assays over each of the last four years at the Texas nursery had not consistently indicated that stunt nematodes were associated with observed symptoms. Although present, they were least

abundant in early summer when symptoms first become obvious. The oxamyl study was initiated, in part, to see if it would improve seedling growth. Some researchers doubt that stunt nematodes stunt pine seedlings.

There was more root mass (α 0.05) in oxamyl treated plots, but a 66% reduction in nematodes per plant was not significant (Table 1) and seedling growth through the season was not satisfactorily improved (data not shown).

Table 1. The effects of oxamyl on individual seedling root dry weight and nematode numbers sampled on June 5.

Treatment	Roots	Number of Nematodes	
		Per Plant	Per gram of Root
Oxamyl	.33 gms	39	1.6
Control	.20	115	7.1
lsd	*.12*	*92*	*5.9*

Although nematodes per gram was better correlated with root weight ($r = -0.81$, $p = 0.002$) than was nematodes per seedling ($r = -0.65$, $p = 0.03$), drying and weighing require some additional time and preparation and are not required to confirm a nematode problem. This suggests that determining nematodes per plant was sufficient to associate nematodes with patches of localized stunting.

Operational Sampling

The analyses of Louisiana and Georgia nursery samples, though not replicated, represent typical efforts to assess the cause(s) of diseases in nurseries. Numbers of nematodes and comparative visual

Table 2. Seedling appearance, fresh weight and numbers of stunt nematodes washed from roots of loblolly seedlings from two nurseries.

Sample Source	Seedling		Nematodes per	
	Appearance	$\bar{x}$ Weight	mg root	100 cc soil
Louisiana				
	Normal	4.1 gms	0	0
	Small	2.7	270	540
	Stunted	0.7	300	600
Georgia				
Block A	Normal	9.4	0	-
	Stunted	1.1	160	-
Block B	Normal	4.5	9	-
	Stunted	0.8	1200	-

assessments and weights for sampled seedlings are presented in Table 2. The association ("significance") between size and stunt nematodes is apparent.

MANAGEMENT IMPLICATIONS

It seems probable that early inoculation is necessary to produce symptoms like those observed at Texas where we found nematodes to be very abundant (> 2,000 / 100 cc soil) at about the time seed were sown. Therefore, seedlings contact nematodes immediately after germination. Stunt nematodes feed on and restrict the development of emerging fine roots. Their impact is increased with soil moisture deficits.

Oxamyl appeared to be effective against nematodes in forest tree nurseries, but to be cost effective would have to be applied prior to damage being visual, i.e. during the first 2-3 weeks after germination.

The root-wash assay is a quick, simple alternative to standard bulked-soil assays for evaluating stunt nematodes on pine seedlings.

LITERATURE CITED

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