# ARTIFICIAL INOCULATION OF FUMIGATED NURSERY BEDS WITH ENDOMYCORRHIZAE

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Many nurserymen fumigate hardwood production areas to control weeds, root-rot fungi, damping-off fungi, and nematodes. Although growth is generally improved, some nurseries in North Carolina, South Carolina, and Georgia have reported problems with stunted sweetgum (Liquidambar styraciflua L.) seedlings following fumigation with methyl bromide. The stunted seedlings usually lacked endomycorrhizae, while healthy seedlings from the same bed were normally endomycorrhizal. Sweetgum has been shown to be highly dependent on the.endomycorrhizal association for optimum growth (1, 2), while sycamore (Platanus occidentalis L.) seems less dependent upon the endomycorrhizal association. Nonmycorrhizal sycamore seedlings can be grown in the nursery and survive when outplanted (4).

Bryan and Ruehle (1) stated "Fumigation would undoubtedly eradicate a significant quantity of indigenous inoculum of endomycorrhizal fungi. The resulting sweetgum seedlings would, therefore, be nonmycorrhizal or only mildly infected and could not grow at acceptable rates." Since natural reinfestation of the endomycorrhizae can be very slow or nonexistent (5), artificial introduction of endomycorrhizal spores may be beneficial. Results reported here indicate that artificial introduction of-nonfumigated soil may increase the diameter growth of sweetgum seedlings where methyl bromide fumigation had been used.

# Methods

At five nurseries throughout North and South Carolina, soil was fumigated with methyl bromide at a rate ranging from 300 to 450 pounds per acre. At a sixth nursery in Georgia, telone C was used instead of methyl bromide. Nursery plots were established us ing a split-split-plot design, with two species, sweetgum and sycamore, serving as the main plots. Six fertilizer treatments involving different rates and combinations of N, P, and K served as the subplots. The fertilizer plots were further split, with half the plots receiving an application of soil containing endomycorrhizal spores, root fragments, and litter from the upper 4 inches of the A layer of a hardwood stand. There were two replicates for each main plot. The size of each sub-subplot was 4 feet wide by 25 feet long. Sowing dates ranged from April 11, at the southernmost nursery, to May 1, 1974, at the northernmost nursery. Beds were watered daily from the time of sowing until after germination. Seedlings were thinned to a density of a 6/ft<sup>2</sup> for sycamore and to 10/ft<sup>2</sup> for sweetgum. Weeds were controlled by hand labor even though several nurseries had used mineral spirits on the sweetgum beds.

At the end of the growing season. seedlings were measured for height and separated into two groups by root-collar diameter. The larger group was greater than seven-sixteenths of an inch for sycamore and greater than five sixteenths of an inch for sweetgum. Eighteen co-dominant seedlings per sub-subplot were selected randomly and measured to the nearest inch for height growth.

At two nurseries, root systems from three seedlings per subsubplot were prepared according to the procedure of Phillips and Hayman (7) and examined microscopically for endomycorrhizal infection. Estimates of the percentage of linear root infection were made by counting the number of 0.2-inch segments with any signs of endomycorrhiza and dividing by the total number of segments (3).

# **Results and Discussion**

The addition of forest soil inoculum increased production of large-diameter sweetgum seedlings at three of the five nurseries that were fumigated with methyl bromide (table 1). No nursery showed a significant increase in sycamore seedlings with large diameters. This supports other studies indicating that sweetgurn

Adding soil with inoculum to fumigated beds increased production of large diameter sweetgum seedlings but had no such effect on sycamore.

<sup>&</sup>lt;sup>1</sup>Research for the paper was conducted while the author was a graduate student at North Carolina State University, Raleigh, N.C.

**Table 1.**—Effect of inoculating fumigated nursery soils with endomycorrizal fungi on height and diameter growth of sweetgum and sycamore seedlings

#### SWEETGUM Number of seedlings Rate of per 144 ft<sup>2</sup> with application diameter greater Average height Nursery Fumigant (1b ai/A) than 5/16 inch (inches) Not Not Inoculated inoculated Inoculated inoculated Methyl bromide А 450 $231^{1}$ 205 21.5 22.2 В Methyl bromide 300 184<sup>2</sup> 130 13.0 11.9 С Methyl bromide 400 22<sup>3</sup> 9 13.1<sup>2</sup> 11.3 D Methyl bromide 350 48 49 $21.5^{2}$ 25.0 Е Methyl bromide 300 43 38 10.3 9.7 F Telone C 424 199 225 24.3 25.7

## SYCAMORE

Nursery	Fumigant	Rate of application (1b ai/A)	Number of seedlings per 144 ft <sup>2</sup> with diameter greater than 7/16 inch		Average height (inches)		
			Inoculated	Not inoculated	Inoculated	Not inoculated	
А	Methyl bromide	450	105	98	24.2	22.3	
В	Methyl bromide	300	28	17	13.1 <sup>2</sup>	10.2	
С	Methyl bromide	400	65	71	21.7	20.0	
D	Methyl bromide	350	30	33	34.3	33.3	
Е	Methyl bromide	300	20	10	15.0	14.9	
F	Telone C	424	185	190	30.9	30.9	

<sup>1</sup> Analysis of variance indicates a significant difference from seedlings not inoculated at the 6 percent level.

<sup>2</sup> Analysis of variance indicates a significant difference from seedlings not inoculated at the 5 percent level.

<sup>3</sup> Analysis of variance indicates a significant difference from seedlings not inoculated at the 1 percent level.

is more dependent upon the endomycorrhizal association than is sycamore.

Since methyl bromide fumigation eliminates endomycorrhizal fungi in the top layers of soil, formation of endomycorrhizae may be delayed until roots reach viable spores in lower soil levels. The endomycorrhizal association may occur much earlier when soil containing viable endomycorrhizal spores is mixed into upper layers of the fumigated nursery bed; this could account for the increase in diameter growth of sweetgum. Microscopic examinations of root segments at the end of the growing season showed that inoculated seedlings and seedlings receiving no inoculation had the same amount of endomycorrhizal infection.

A disadvantage of using forest soil as an inoculum source is that it may contain weed seeds and other harmful soil organisms, which possibly could account for the decrease in height growth of sweetgum at Nursery D. A source of inoculum that might be relatively free of root pathogens and parasites is soil from covercropped areas. Endomycorrhizae have a wide host range, as indicated by the ability of one species to infect sycamore, sweetgum, yellow poplar, cottonwood, soft maple, citrus, peach, corn, soybeans, sorghum, cotton, and pepper (5): Because spore production may be higher at the end of the growing season (6), soil from cover-cropped areas should be collected after harvesting. Techniques are also being developed for large-stale artificial soil inoculation with specific endomycorrhizal fungi (4).

Effects of artificial inoculation on seedling growth may be reduced under high fertilization. This was demonstrated at nursery A by an interaction between inoculation and rate of phosphorus application (figure 1). With no addition of phosphorus, a 67-percent increase in sweetgum seedlings with large root collars resulted from inoculation; however, inoculation had no effect on diameter growth when 200 pounds per acre of phosphorus were applied.



**Figure 1.**—Interaction between inoculation of fumigated soil with endomycorrhizal fungi and application of phosphorus for sweetgum at nursery A.

The additional phosphorus had a dramatic effect on the growth of sweetgum by increasing the number of seedlings with large root collars by 111 percent. This supports earlier reports that a reduction in response to endomycorrhizae can occur when phosphorus is applied (*6*).

Soil fumigants other than methyl bromide have been evaluated for hardwood production (*8*). These are usually less expensive and not as destructive to the beneficial fungi. For example, methyl bromide fumigation was not used at nursery F, where growth of sweetgum and sycamore was equal to or better than that at any of the five nurseries where methyl bromide was used (table 1). At nursery F, inoculation with forest soil did not have a beneficial effect on the growth of sweetgum, presumably because fumigation with telone C did not reduce significantly the amount of indigenous endomycorrhizal fungi.

Results indicate that, following methyl bromide fumigation of areas to be used for growing hardwood species that are dependent on the endomycorrhizal association, the fumigated soil should be inoculated with soil containing viable endomycorrhizal fungi. Beneficial effects of introduced endomycorrhizal fungi may be masked by high fertilization, especially with phosphorus. When possible, soil fumigants that are not as destructive of endomycorrhical fungi as methyl bromide should be used.

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