QUALITY HARDWOOD SEEDLINGS REQUIRE EARLY MYCORRHIZAL DEVELOPMENT IN NURSERY BEDS

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Abstract.--Research during the past 3 years has revealed that many hardwood species require early endomycorrhizal infection in nursery beds to produce thrifty seedlings. If infection is delayed until late summer, as frequently occurs in production nurseries, seedlings of inferior grade are produced. In four different experiments with 18 half-sib progeny of sweetgum, root collar diameters of nonmycorrhizal seedlings averaged 0.17 cm and height averaged 5.0 cm, regardless of fertility regimes studied. Diameters and heights of mycorrhizal seedlings averaged 0.70 cm and 32.7 cm. These results indicate that seedling grade can be significantly improved by providing sufficient endomycorrhizal inoculum in nursery beds. The progeny from all our sweetgum selections have exhibited an obligate requirement for endomycorrhizal fungi, and all have produced good quality mycorrhizal seedlings. However, progeny from some mother trees have a greater percentage of larger-sized seedlings than progeny from others under a variety of experimental conditions. It may be feasible to develop a testing scheme to evaluate selected mother trees based on the morphological grade of their progeny in nursery beds under specified environmental conditions.

Additional keywords: Sweetgum, endomycorrhizal fungus, hardwood nursery practices.

Nurserymen find it hard to grow hardwood seedlings of consistently high quality for outplanting. This difficulty is a major obstacle to artificial establishment of many hardwoods. Research over the past 3 years, at the Forest Services' Mycorrhizal Institute in Athens, Georgia, suggests that a mycorrhizal deficiency in the nursery beds early in the growing season maybe a primary cause for the poor development of hardwood seedlings.

This conclusion is based on results from a large number of studies, many of which are still in progress. In this paper we make no attempt to describe our studies in detail or to prove our conclusions to some level of confidence. Our purpose here is to alert forest tree improvement workers to the possible importance of mycorrhizae for production of hardwood seedlings. Sweetgum is the primary example because our research is farthest advanced with that hardwood species. Despite their preliminary nature, we strongly emphasize findings of possible significance in tree selection and breeding.

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Infection delayed

Unlike pines and oaks which have ectomycorrhizal root symbionts, most hardwood species develop a symbiotic relationship with endomycorrhizal fungi. Spores of ectomycorrhizal fungi are readily disseminated by wind; those of endomycorrhizal fungi are disseminated primarily by soil water movement. Thus, while current nursery fumigation practices destroy the inoculum of both groups of fungi, ectomycorrhizal inoculum is restored to nursery beds much more quickly in the spring.

Reinfestation of nursery soil with endomycorrhizal fungi can occur in several ways. The root systems of hardwoods eventually grow below the zone of effective fumigation and come into contact with viable spores. Spores are moved into fumigated topsoil by earthworms and other microfauna. They are also moved in soil water movement during the growing season.

All these methods of bringing viable spores into contact with the developing root system have a major disadvantage: Infection does not occur until summer and there is insufficient time for seedlings to develop properly. The resulting seedlings are too large to plow under but too small to outplant.

The importance of endomycorrhizal fungi to hardwood seedling development was poorly understood until recently and research on it is still in the early stages. When seedlings were not developing well, no one thought of examining the roots under a microscope for the presence of endomycorrhizal symbionts. Nurserymen assumed that the problem was with moisture supply, fertility, or destructive root pathogens. The importance of endomycorrhizae for ion and water uptake by many hardwood species was not considered. Instead, fertility of nursery soils was increased--an approach that works with agronomic plants that lack their normal mycorrhizal associate. To obtain good growth of non-mycorrhizal crop plants up to 1000 ppm of phosphorus may have to be applied to the rooting media. Much work remains to be done before definite conclusions can be reached, but it appears that the same approach will not work with hardwood seedlings. Nursery beds may be overfertilized but few are under-fertilized. The seedlings apparently must have endomycorrhizal associates.

The importance of endomycorrhizal fungi to hardwood seedlings growing in nursery beds was first observed by us with sweetgum (*Liquidambar styraciflua* L.) in 1973. Progeny from 10 selected mother trees were planted in beds that were improperly fumigated due to a sudden and prolonged drop in temperature during fumigation. By early July all seedlings were badly discolored and even weekly applications of liquid fertilizers throughout June and July elicited no visual growth responses. In late July and early August individual clumps of seedlings turned green and began to elongate. Laboratory examination of the root systems from these elongated seedlings showed them all to be mycorrhizal. The root systems from the still discolored and nonelongating seedlings proved to be nonmycorrhizal.

FORMAL STUDIES

We have worked with 10 hardwood species during the past 3 years. Sweetgum has been studied for 3 consecutive years, and only the results with this species will be considered in any detail here. The bulk of the initial nursery work on the other 9 species was done during the 1976 growing season, and it is far too soon to be making conclusions about results.

Whenever possible, we planted half-sib progeny in our experiments. With sweetgum we recorded the location of the mother trees, thus some general conclusions about seed sources are possible.

Before seeding nursery beds we applied from 140 kg/ha to 1120 kg/ha of commercial 10-10-10 fertilizer, and we top dressed all seedlings including nonmycorrhizal controls with a total of 1680 kg/ha of NH_4NO_3 during the growing season. Calcium is important for hardwood root development and we have arbitrarily been standardizing this element in our microplots at 1120 kg/ha. All nursery soil is fumigated with MC-2 at the recommended rates to eliminate natural inoculum, destructive root pathogens and most weeds.

We maintain pure cultures of the endomycorrhizal fungi, *Glomus mosseae* and *Glomus fasciculatus* on sorghum roots in our laboratory. We also have a culture of mixed fungi obtained from a nursery bed on which sweetgum seedlings were growing in 1973. Several of the fungi in this mixture have not yet been identified. Infected sorghum roots are applied to treated plots. Soil leachates from the mycorrhizal culture are applied to control plots to standardize. other soil microorganisms. Endomycorrhizal fungal spores are removed from these leachates with appropriate filters.

EARLY RESULTS

Since our primary purpose has been to improve nursery production of commercial seedlings, most of the seedlings have been destructively sampled. Relatively few have been outplanted. On the basis of past experience, however, it is reasonable to assume that large, thrifty nursery seedlings will survive and grow better than smaller ones.

All hardwood species studied thus far have benefited tremendously from the incorporation of mycorrhizal inoculum into the nursery beds at the time of sowing. Furthermore, sweetgum seedlings have demonstrated an obligate requirement for these root symbionts when grown in natural soils. Root systems of hardwoods are mycorrhizal in almost all natural situations.

We have 2 years of data (1974 and 1975) on total biomass on nonmycorrhizal and mycorrhizal sweetgum seedlings. The mycorrhizal sweetgum seedlings have been producing 70 to 80 times as much total dry weight as nonmycorrhizal ones. Comparable differences are apparent for most of the other hardwood species tested.

Table 1 shows differences in root collar diameter and total height among half-sib progeny of sweetgum seedlings after 1 growing season. Results are from four different studies conducted over several years. The results from a large sweetgum study completed in 1975 have already been submitted for publication. In this initial work we studied the response of sweetgum seedlings from eight different mother trees to different fertility regimes (140, 280, 560, and 1120 kg/ha of 10-10-10) with and without the endomycorrhizal fungus, Glomus mosseae.

Table 1.--Root collar diameter (RCD) and total heights (HT) of mycorrhizal and nonmycorrhizal sweetgum seedlings obtained from three experiments during 1975 and 1976.-

	Control		Glomus mosseae		Glomus fasciculatus		Natural inoculum	
Mother tree selection	RCD cm	HT cm	RCD cm	HT cm	RCD cm	HT cm	RCD cm	HT cm
	1975 Experiment							
SG-74-2	.16	<5.0	.62	34.2				
SG-74-4	.16	<5.0	.57	34.1				
SG-74-5	.16	<5.0	.62	34.5				
SG-74-6	.16	<5.0	.73	40.5				
SG-74-7	.16	<5.0	.59	32.0				
SG-74-8	.16	<5.0	.70	35.8				
SG-74-9	.16	<5.0	.86	42.1				
SG-74-51	.16	<5.0	.66	32.5				
			1976 Experiment					
SG-SC-2	.19	3.8	.73	32.2	.80	36.5	.70	33.5
SG-SC-3	.17	3.6	.70	33.6	.93	41.2	.60	33.7
SG-SC-4	.18	5.2	.73	37.2	.92	43.9	.84	44.0
SG-74-7	.18	4.2	.75	30.4	.72	34.2	.88	34.8
SG-74-9	.22	5.7	.76	29.2	.85	30.2	.73	26.9
SG-75-2	.20	3.8	.60	25.8	.68	32.7	.48	22.9
SG-75-4	.16	3.5	.65	29.5	.70	29.4	.55	25.0
SG-75-6	.20	4.4	.66	31.1	.81	28.4	.69	29.7
SG-75-8	.17	4.1	.72	27.6	.71	29.0	.55	25.5

1/ Seedling data not tabulated by individual fertilizer treatments. One can not compare a specific mother trees' progeny between years because of almost 40 days growing season between years due to differences in sowing dates.

In this 1975 study, the seedlings with mycorrhizae grew as well at the lowest rate as at the highest rate of fertilizer application. We also found that more of the progeny from upland selections grew to large sizes. In 1974, the progeny from a single upland selection were substantially larger than those from a bottomland selection.

These two observations indicate that progeny of some upland selections may glean a greater benefit from mycorrhizae than progeny of some bottomland selections. However, the upland sources may simply represent superior genotypes, via natural selection, that have been subjected to greater environmental stress on less fertile sites with significantly poorer soil moisture relationships. Thus, we do not mean to infer that all upland selections are better than all bottomland selections. Quite the contrary; we have tested some bottomland individuals whose seedlings were very good and we would readily include them among the better trees we have tested.

Genetic predisposition to endomycorrhizal infection has not been positively demonstrated to our knowledge within any forest tree species, but then no effort has been made to determine if it exists. Without outplanting data no strong argument can be offered. The only information we have is from our nursery studies which suggest the possibility. Since we have studied at least 18 half-sib progeny of sweetgum, we will use this species to illustrate our point.

None of the progeny of sweetgum selections, regardless of fertility treatments, have developed much beyond the primary leaf stage without mycorrhizae. Progeny from all the parent trees have developed well regardless of fertility as long as they have mycorrhizae. This is dramatic proof of the obligate need of sweetgum for endomycorrhizae.

Some half-sib progeny have a higher percentage of large seedlings than other progeny. Thus, if an arbitrary size for superior seedlings is set 70 to 80 percent of the seedlings from one mother tree will meet the standard while only 30 to 40 percent of the seedlings from another mother tree may meet it. We realized that seedlings must be stratified by size and outplanted on many sites before the superiority of a given size class can be determined. However, based upon the distinctive results in our early tests, we feel it is possible and practical to develop nursery tests to identify mother trees which will produce seedlings of superior size with minimum fertility.

At this time, although the potential of our new mycorrhizal technology to the hardwood tree improvement programs in the Southeast is at best perhaps visionary, we feel its place in nursery management has been definitely established. Its implementation is imperative if we are to produce adequate numbers of plantable seedlings that are highly competitive in the field. Until recently there was little reason to presume that hardwood seedlings provided with adequate moisture and fertility in the nursery beds did not have a suitable environment in which to make optimum growth. Now, it has been demonstrated quite conclusively that hardwoods must have adequate levels of endomycorrhizal inoculum in the nursery beds early in the growing season it they are to attain the large size demanded by the forestry industries.

This new hardwood mycorrhizal technology will not need 10 or 20 years to be perfected. With a concerted effort and support by the forestry industries, this technology can be available in from 3 to 5 years. During this same time span, the potentials for incorporating mycorrhizal technology into the existing tree improvement programs can be easily explored. Foresters and the forestry industries are by nature conservative in accepting new practices. They should, however, carefully examine the potentials of this forestry related mycorrhizal technology instead of waiting for a decade to see what develops.