National Pisolithus tinctorius Ectomycorrhizae

Nursery Evaluation Results - 1978,

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Abstract.--In 1978, the national <u>Pisolithus tinctorius</u> (<u>P.t.</u>) ectomycorrhizae nursery evaluation was expanded to include 33 bare-root nurseries in 28 states, along with 18 container seedling studies in nine states (including Hawaii) and Canada. The results obtained to date are encouraging. The Ga. <u>P.t.</u> inoculum has been more effective and consistent than the Abbott <u>P.t.</u> inoculum in the production of <u>P.t.</u> ectomycorrhizae and stimulation of seedling growth and quality. The variable Abbott <u>P.t.</u> results were apparently caused by inoculum batch production problems that have since been identified and, hopefully, eliminated.

INTRODUCTION

During the past several years, the Institute for Mycorrhizal Research and Development, U.S.D.A. Forest Service, Athens, Georgia, has been conducting extensive research on mycorrhizae. This research has centered around one particular ectomycorrhizal fungus, <u>Pisolithus tinctorius</u> (P.t.) and its application to forest tree nurseries and field forestation (Cordell and Marx, 1977). Based on the successful results obtained from this pioneer research, cooperative <u>P.t.</u> ectomycorrhizae field evaluations have also been conducted by the Mycorrhizal Institute and Forest Insect and Disease Management, Southeastern Area -State and Private Forestry, U.S.D.A. Forest Service, Atlanta, Georgia, in both bareroot and container tree seedling nurseries. Results obtained from both the nursery and followup field outplanting evaluations have been highly successful on a wide variety of host seedling species, bare-root nursery soil and container media types, and forestation sites. Significant increases in ectomycorrhizal production along with seedling growth and quality (reduced culls) have been repeatedly

¹Paper presented by the senior author at the Intermountain Nurseryman's Association Meeting, Asven, Colorado, August 13-16, 1979.

²Respectively, National Coordinator, National Mycorrhizae Nursery Evaluation, Forest Insect and Disease Management, Southeastern Area, State and Private Forestry, USDA Forest Service, Asheville, North Carolina, and Director, Institute for Mycorrhizal Research and Development, Southeastern Forest Experiment Station, USDA Forest Service, Athens, Georgia. obtained in the nurseries (Marx and others, 1976; Ruehle and Marx, 1977). Significant increases in tree survival and growth have also been obtained on a variety of forestation sites (Marx and others, 1977: Berry and Marx, 1978).

In 1978, the national <u>Pisolithus tinctorius</u> (<u>P.t.</u>) ectomycorrhizae nursery evaluation was expanded to include 33 bare-root nurseries in 28 states, along with 18 container seedling studies in nine states (including Hawaii) and Canada. Seedling species involved in the bare-root nurseries included 11 species of pines, along with two varieties of Douglasfir and Fraser fir. Seedling species involved in the container nurseries included nine species of pines, along with Douglas-fir, western hemlock, black spruce, white spruce, bur oak, and eucalyptus.

OBJECTIVE

The objective of both the bare-root and container seedling evaluations was to compare the effectiveness of <u>P.t.</u> inoculum produced by the Mycorrhizal Institute - Athens, Georgia, (Ga. <u>P.t.</u>) and Abbott Laboratories - Chicago, Illinois, (Abbott P.t.) for ectomycorrhizal formation, seedling growth and quality in the nursery, and tree survival and growth in subsequent field outplantings. This is a cooperative project between the Institute for Mycorrhizal Research and Development, Athens, Georgia, Forest Insect and Private Forestry, Atlanta, Georgia, and various other U.S. Forest Service, State, industry, and university agencies.

METHODS

Bare-root Nurseries

As in previous nursery seedbed evaluations, randomly selected prefumigated seedbed plots were inoculated with either the Georgia or Abbott <u>P.t.</u> mycelium-vermiculite-peat inoculum immediately prior to planting. Prefumigation with soil fumigants equivalent to methyl bromide - 98% + chloropicrin -2% and methyl bromide - 67% + chloropicrin - 33% has been <u>mandatory</u> in obtaining successful P.t. nursery seedbed inoculations. The dried inoculum mixture (approximately 12 percent moisture content) was seedbed dosage rates - Georgia - 100ml/ft , Abbott - 200, 100, 50 ml/ft². The inoculum was then chopped into the upper 3-4 inch soil surface with a garden tool. Each of the five treatments - four P.t. treatments and one check - were replicated five times in a randomized 5-block design. Following the randomized 5-block design. Following the inoculation, conventional seeding, mulching and all other nursery cultural practices were maintained as usual. The nursery phase of this evaluation is scheduled for 1 to 3 years, depending on the rotation length of the seedling host species.

Container Nurseries

Each container evaluation was installed as a completely randomized design in a greenhouse at each location. The same dried P.t. mycelium-vermiculite-peat inoculum mixture was again used in these evaluations. Immediately prior to filling the containers, the inoculum was mixed in a clean soil mixer such as a small cement mixer cleaned with a chlorox-hot water solution prior to mixing. Precautions were also taken to avoid cross con-tamination of the Georgia and Abbott $\underline{P.t.}$ inoculum. Four P.t. inoculum-growth media volume to volume ratios were evaluated at each location--Ga. P.t.-1:15, Abbott P.t.-1:15, 1:7.5, and 1:30. Each of the four treatments along with a nontreated check treatment were replicated four times. The number of containers used per replicate per treatment varied at each location but averaged around 40-50. The growth media used consisted of either the standard peat-vermiculite 1:1 volume to volume Mycorrhizal Institute. All container seedling cultural practices including temperature control, watering, fertilization, seedling growth period, etc. were employed as deemed necessary for each seedling species and location. Each seed source was tandardized and the seed was stratified and treated with a fungicide such as Arasan[®] prior to seeding as dictated by local container seedling production requirements. However, fungicide drenches applied for damping off

However, fungicide drenches applied for control following seeding were omitted.

The duration of the container seedling evaluations ranged between 6 and 12 months, depending upon the geographic location.

Outplantings

Field outplantings are being established from all treatments represented in both the bare-root and container seedling nurseries with seedlings showing significant responses in <u>P.t.</u> ectomycorrhizae development. A randomized block design with five replicates per treatment and 49 seedlings per treatment replicate is being utilized seedings per treatment replicate is being utilized per site. Standard site preparation, spacing, etc. are being employed as deemed necessary for the host species, site condition, geographic location, etc. All trees are being hand planted. The outplantings are scheduled for a 10-year duration with annual tree survival, growth, and P.t. fruiting body development measurements the first 5 years and biannual measurements the remaining duration. Annual or biannual progress reports will also be prepared and submitted to all cooperators.

RESULTS

The results obtained to date from both the bare-root and container seedling evaluations are encouraging.

Bare-root Nurseries

The midseason or first-year sampling results from 33 nurseries showed an average of 19 percent ectomycorrhizae formation on seedling feeder P.t. roots by the Ca.-100 P.t. treatment and 5 percent by the Abbott-200 <u>P.t.</u> treatment and 5 percent Abbott <u>P.t.</u> inoculum batch effect is shown in Table 1. For example, Abbott <u>P.t.</u> batches 4 and 5 produced over 200 percent as much P.t. ectomycorrhizae as batch 2.

The first-year sampling results obtained from five bare-root nurseries in the midwestern United States treated with either Abbott <u>P.t.</u> batch 4 or S were also encouraging in most cases. However, the Ca.-100 P.t. treatment again produced the most effective and consistent results. This treatment produced an average of 14 percent P.t. ectomycorrhizae and 62 percent of the seedlings had some P.t. ectomycorrhizae. The Abbott-200 P.t. treatment produced an average of 10 percent P.t. ectomycorrhizae and 37 percent of the seedlings had some <u>P.t.</u> ectomycorrhizae. In addition, the Ga.-100 <u>P.t.</u> treatment plots showed a 250 perce percent increase in total ectomycorrhizae (P.t. and other) over the untreated check plots (20 percent vs. 8 percent). The Abbott-200 $\underline{P.t.}$ treatment plots showed over a 200 percent increase in ectomycorrhizae over the check plots (17 percent vs. 8 percent).

Table 1.--1978 National Ectomycorrhizae Nursery Evaluation--Summary of midseason root evaluations in 33 nurseries

	Ectomycorrhizae by						Seedlings		
Treatment	P.t. (%)			Total (%)			w/P.t. (%)		
GA 100	18.6 Abbott batch No.			26.5 Abbott batch No.			70.4 Abbott batch No.		
	Abbott 200	3.4	5.8	5.2	19.6	17.1	17.8	27.0	34.4
Abbott 100	2.2	5.8	4.9	19.6	17.1	17.7	24.5	31.6	30.0
Abbott 50	1.4	3.2	2.8	18.6	15.0	16.0	13.0	23.0	20.4
Control		0.1			13.8			1.4	

¹Poor quality Abbott <u>P.t</u>.inoculum; produced an average of 3 percent <u>P.t</u>. ectomycorrhizae on 4-month-old loblolly pine seedlings in inoculated field microplots at Athens, Ga., in 1978.

 2 Good to excellent quality Abbott <u>P.t.</u> inoculum; produced an average of 12 and 54 percent <u>P.t.</u> ectomycorrhizae, respectively, on 4-month-old loblolly pine seedlings in inoculated microplots in Athens, Ga., in 1978.

Table 2.--1978 National Ectomycorrhizae Nursery Evaluation--Summary of final root evaluations (1-0 seedlings) in 12 nurseries

Treatment	Ectomycorrhizae by P.t. (%)			Seedlings w/P.t. (%)			Cull seedlings (%)		
GA 100	27.8 Abbott batch No.			95.0 Abbott batch No.			18.4 Abbott batch No.		
	Abbott 200	3.2	10.5	5.7	17.8	61.0	32.2	29.1	13.8
Abbott 100	1.5	4.5	2.5	10.5	42.0	21.0	25.3	15.0	22.3
Abbott 50	2.4	5.5	3.4	12.5	33.0	19.3	28.2	17.4	24.6
Control		0.4			1.8			24.8	

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Final results obtained from 12, 1-0 southern and central United States bare-root nurseries were also quite variable--particularly with the Abbott <u>P.t.</u> batches (Table 2). Again, the Ga. <u>P.t.</u> provided the most favorable and consistent results. This inoculum produced 28 percent <u>P.t.</u> ectomycorrhizae on seedling feeder roots, increased seedling fresh weights 26 percent over controls, and decreased seedling cull percentages 26 percent less than controls. The Abbott <u>P.t.</u> batch effect is revealed again in Table 2. The nurseries treated with batches 4 and 5 had over a 300 percent increase in <u>P.t.</u> ectomycorrhizae formation and seedlings with <u>P.t.</u> ectomycorrhizae along with a significant reduction in cull seedlings as compared with the nurseries treated with batch 2.

Container Nurseries

Final results obtained from the container seedling evaluation with both the Ga. and Abbott $\underline{\text{P.t.}}$ inoculum were also highly encouraging. The Ga. P.t. 1:15 treatment produced

40 percent <u>P.t.</u> ectomycorrhizae on seedling feeder roots while the Abbott <u>P.t.</u> 1:15 treatment produced 20 percent <u>P.t.</u> ectomycorrhizae. This resulted in almost a 300 percent increase in total ectomycorrhizae development on the Ga. <u>P.t.</u> treated seedlings and over a 200 percent increase on the Abbott <u>P.t.</u> treated seedlings, as compared with the untreated check seedlings.

SUMMARY AND CONCLUSIONS

1. The ectomycorrhizal fungus <u>Pisolithus</u> <u>tinctorius</u> can be successfully artificially inoculated into prefumigated bare-root nursery seedbeds and greenhouse containers on a wide variety of conifer (pines, firs, spruces, Douglasfir, western hemlock and some hardwoods (oaks) species it a wide geographic range in the United States,

2. The Ga. <u>P.t.</u> inoculum was more effective and consistent than the Abbott <u>P.t.</u> inoculum in the production of <u>P.t.</u> ectomycorrhizae on seedling feeder roots along with significant increases in seedling growth and quality (cull reduction) in the bare-root nurseries.

3. The variable results obtained with the Abbott $\underline{P.t.}$ inoculum, primarily in the bare-root nurseries, were apparently caused by inoculum batch variations. Results obtained with Abbott P.t. inoculum batches 4 and 5 were comparable to those obtained with the Ga. $\underline{P.t.}$ inoculum in the bare-root nurseries. In addition, the Abbott P t. inoculum was very effective and comparable to the Ga. $\underline{P.t.}$ inoculum in the production of $\underline{P.t.}$ ectomycorrhizae on a wide variety of conifer and one hardwood (bur oak) species in the container seedling evaluation. Several contributing Abbott $\underline{P.t.}$ inoculum batch production problems have since been identified and, hopefully, corrected.

4. Consequently, there is presently a great deal of optimism concerning the practical application of commercially-produced P.t. inoculum to hare-root and container seedling nurseries

for the production of "tailored" seedling products. Abbott Laboratories plans to market a brand of P.t. "MycoRhizTM" in the southern United States in 1980,

5. Additional evaluations are being planned by the U.S. Forest Service in the southern U.S. concerning the practical inoculation of bareroot nursery seedbeds utilizing a modification of conventional seeding equipment.

6. The Mycorrhizal Institute and Abbott Laboratories are also continuing their cooperative efforts with various Federal, State, industry, and university agencies in widespread locations throughout the 'Tnited States and several foreign countries concerning the research, development, and application of other suitable ecto- and endomycorrhizal fungi in both bareroot and container nurseries.

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