

amounts of material from 168 named cultivars or species. This includes 37 apple, 6 apple rootstocks, 16 ornamental *Malus*, 4 pear, 9 tart cherry, 18 sweet cherry, 17 plum, 4 plum rootstocks, 6 apricot, 4 nectarine, 30 peach, 3 peach rootstocks and 15 ornamental *Prunus*. It is available for the asking and free of charge.

MYCORRHIZAL FUNGI IN RELATION TO SOME ASPECTS OF PLANT PROPAGATION

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The symbiotic association between a plant root and a mycorrhizal fungus is termed mycorrhiza. The specific types of mycorrhizal associations have been described in previous issues of the IPPS Proceedings (2,4). Mycorrhizal fungi are naturally occurring organisms in over 80% of the plant taxa. The vast majority of vascular plants have evolved to a dependency on mycorrhizae either for survival or to flourish. In many instances, mycorrhizal fungi facilitate increased growth and/or selective nutrient uptake and accumulation; tolerance to environmental stresses, such as drought, temperature extremes and soil acidity, and function in protecting roots from pathogenic infection. In addition, mycorrhizal fungi are also known to produce enzymes, vitamins and growth hormones that increase root size and longevity as well as rooting of cuttings.

Because the mycorrhizal state is a universal, natural association, its importance in nursery crop production may only become apparent when we disrupt the natural soil environment. Advances in the use of fertilizers, pesticides, steam sterilization or fumigation, soilless mixes, etc. to increase crop productivity have simultaneously diminished or eliminated the indigenous beneficial soil-borne mycorrhizal fungi. Consequently, severe stunting, special nutritional requirements, poor survival and/or growth, and increased disease susceptibility are often attributed to deleterious characteristics of a plant species or a failure of cultural practice, rather than absence of mycorrhizal fungi.

In order to benefit from mycorrhizal fungi in the nursery industry, we must be concerned with plant-fungus specificity, differences among fungal isolates producing specific effects

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under different cultural conditions, the economics of producing mycorrhizal inoculum and inoculating nursery crops.

In this study we would like to demonstrate some of the benefits one may obtain by incorporating mycorrhizal inoculum into seedbeds and container media during propagation of nursery crops.

MATERIALS AND METHODS

Isolate M3 of the ectomycorrhizal fungus (*Pisolithus tinctorius*) was obtained from sporocarp tissue collected in Laurel County, Kentucky. *P. tinctorius* isolate M1 (original no. 138) originally isolated in Georgia, was obtained from D.H. Marx, Institute for Mycorrhizal Research and Development, Athens, Georgia. Vegetative mycorrhizal inoculum of both isolates was produced by procedures of Marx and Bryan (5). After 3 months, the inoculum was washed and stored at 3°C for 24 hours before use. Inoculum of *Glomus fasciculatus* was prepared and added to the medium following procedures of Bryan and Kormanik (1).

All plants grown in the greenhouse were under an extended 18 hour photoperiod and prevailing greenhouse temperatures of 20 to 28°C day/20°C night (70° to 82°F/70°F night), and were watered with a complete minor element solution once a month (3).

Field Experiments with Ecto- and Endomycorrhizal Fungi. In the fall, 1977, seedbeds were prepared and fumigated with 67% methyl bromide — 33% chloropicrin at 397 kg/hectare and covered with 2 mil plastic. The following spring the land was rotovated and plots were boxed. Plot size was 0.84 m². Seed of each species was tested for viability and stratified. Planting density was 222 plants/m².

Pine species used were loblolly (*Pinus taeda*), Virginia (*P. virginiana*), pitch (*P. rigida*), shortleaf (*P. echinata*) and Scotch (*P. sylvestris*). Before seeding, plots were inoculated with *Pisolithus tinctorius* isolate M3 at a rate of 1.1 l/m². Redbud (*Cercis canadensis*) seed were planted as above except that plots were inoculated with inoculum of the endomycorrhizal fungus *G. fasciculatus* (6.2 l/m²).

Seeding was done on June 23, 1978. Each microplot was fertilized with 170 kg/hectare of a commercial 10-10-10 fertilizer. On July 16, 30 and August 13, 1978, 37.4 kg/hectare N was applied as NH₄NO₃.

Fertilizer/*Glomus fasciculatus* Interactions in Growth of Containerized Southern Magnolia (*Magnolia grandiflora*) Seedlings. Two month-old southern magnolia seedlings grow-

ing in flats containing sterilized peat:perlite (1:1 V/V) were transplanted on March 3, 1978, to 7.6 cm (3 inches) pots containing a sterilized mixture of composted hardwood bark and expanded shale (2:1 V/V) and 1.1 kg or 4.5 kg of 18-6-12 Osmocote/m³. Thirty-six seedlings were planted at each fertility rate and one half of each group were inoculated with *G. fasciculatus* inoculum incorporated at a rate of 1:8 (V/V) into the container medium. Plants were grown in the greenhouse.

Fertilizer/*Pisolithus tinctorius* Interactions in Growth of Containerized Oak Seedlings. In the fall, 1977, red oak (*Quercus rubra*), swamp chestnut (*Q. Prinus*, Syn.: *Q. michauxii*) and pin oak (*Q. palustris*) acorns were collected, screened for viability and stratified at 5°C until used. Quart milk cartons were filled with steamed, composted, hardwood bark and expanded shale (2:1 V/V), half containing inoculum of *P. tinctorius* isolate M1 (1:15 V/V). On February 3, 1978, red and swamp chestnut oak acorns were planted in the containers, grown for 2 months and fertilized with 18-6-12 Osmocote at 1.1 kg/m³. Pin oak seeds were also planted at the same time in Leach "Super Cell" containers. The potting medium was steamed peat:perlite (1:1 V/V), inoculated with *P. tinctorius* (isolates M1 or M3, 1:8 V/V), or not inoculated. The medium contained various rates of Osmocote 14-14-14. Plants were grown in the greenhouse.

On May 18, 1978, all oak seedlings were transplanted to 15 cm (1 gallon) pots containing composted hardwood bark and expanded shale (2:1 V/V). The mix was amended with 108.5/m³ of Peter's fritted trace elements and various rates of Sierrablen 19-6-10 + Fe. Plants were grown outdoors in a container nursery until fall, 1978.

RESULTS

Field Experiments with Ecto- and Endomycorrhizal Fungi. Inoculating fumigated seedbeds with *Pisolithus tinctorius* inoculum substantially increased stem diameters and height growth of all 5 conifer species (Table 1). Stem diameters of inoculated plants were 50 to 150% greater than non-inoculated plants while plant heights were 60 to 125% greater. The uninoculated plants were stunted and slightly chlorotic. The apical foliage of uninoculated plants of most species was purple in color, characteristic of a phosphorus deficiency.

The endomycorrhizal fungus *Glomus fasciculatus* increased growth of redbud seedlings in the field. After 3 and 5 months, redbud seedlings growing in soil inoculated with *G. fasciculatus* were 92 and 72%, respectively, taller than noninoculated plants (Table 2). Stem diameters were also 20% larger on plants growing in inoculated soil. No nutrient deficiency symptoms were

Table 1. Effect of ectomycorrhizal fungus *Pisolithus tinctorius* on growth of pine seedlings 5 months after planting.¹

Species	Stem Diameter		Height	
	mm		cm	
	Inoculated	Control	Inoculated	Control
Loblolly	2.4±.2	1.6±.03	14.9±1.2	7.7±.6
Virginia	1.9±.2	1.2±.1	7.5±0.5	4.7±.3
Shortleaf	2.3±.2	0.9±.1	7.2±.6	3.2±.2
Pitch	2.0±.2	1.2±.1	7.8±.7	3.7±.4
Scotch	1.8±.1	1.1±.1	4.1±.2	2.5±.1

¹ *P. tinctorius* isolate M3, Laurel Co., Ky. Means of 20 seedlings.

apparent on redbud seedlings in inoculated or uninoculated control plots.

Table 2. Effect of endomycorrhizal fungus *Glomus fasciculatus* on growth of redbud seedlings in field beds 3 and 5 months after planting.¹

	Height (cm)		Stem diameter (mm)
	3 mo.	5 mo.	5 mo.
	Control	11.5±0.8	26.1±2.8
<i>Glomus fasciculatus</i>	22.1±1.0	45.0±2.8	4.2±0.3

¹ Means of 20 to 41 seedlings.

Fertilizer/*Glomus fasciculatus* Interactions in Growth of Containerized Southern Magnolia Seedlings. Inoculation of container media with *G. fasciculatus* in combination with the recommended rate of 18-6-12 Osmocote (4.5 kg/m³) produced the largest magnolia seedlings (Table 3). Inoculated seedlings at this rate were twice as large as the noninoculated controls at the full fertilizer rate. Inoculated seedlings grown with one-fourth the recommended fertilizer were also nearly twice as large as noninoculated control seedlings after 7 mo.

Table 3. Endomycorrhizal *Glomus fasciculatus*/fertilizer interactions in height growth of containerized southern magnolia seedlings.¹

	Fertilizer rates ²			
	1.1 kg/m ³		4.5 kg/m ³	
	6 mo.	7 mo.	6 mo.	7 mo.
Control	4.0±0.2	4.2±0.3	5.5±0.4	5.8±0.4
<i>Glomus fasciculatus</i>	7.5±0.8	8.2±0.9	10.2±0.8	11.8±0.7

¹ Two-mo.-old plants transplanted into 7.6 cm pots. Means of 18 seedlings.

² 18-6-12 Osmocote.

Fertilizer/*Pisolithus tinctorius* Interactions in Growth of Containerized Oak Seedlings. Fertilizer increased height growth of pin oak seedlings grown in Leach 'Super Cells' (Table 4) Plants fertilized at the manufacturer's recommended rate (4.5 kg/m³) produced the best growth while plants fertilized with one-fourth the recommended rate (1.1 kg/m³) were intermediate in size between plants receiving no fertilizer and those fertilized at the recommended rate. At the recommended fertilizer

rate, isolate M3 of *P. tinctorius* produced significantly more growth than plants inoculated with isolate M1.

Table 4. *Pisolithus tinctorius*/fertilizer interactions in height growth of pin oak seedlings grown in Leach 'Super Cells' 18 weeks.

	Fertilizer ¹ rate		
	none	1.1 kg/m ³	4.5 kg/m ³
	Height (cm)		
Control	8.2a	10.2a	13.6b
<i>P. tinctorius</i> (M1)	8.5a	9.9a	14.2b
<i>P. tinctorius</i> (M3)	9.0a	10.6a	18.3c

¹ 14-14-14 Osmocote. Means followed by different letters significantly different (Duncan's multiple range test, 5% level).

In outdoor container nursery experiments, the largest red, pin, and swamp chestnut oak seedlings grown in 15 cm containers were those receiving recommended fertilization (4.5 kg/m³) and inoculated with *P. tinctorius* (Tables 5,6,7). After 9 mo. these plants were 20 to 60% larger than noninoculated plants fertilized at the same fertilization rate. Also the inoculated plants at the recommended fertilizer rate grew substantially more (84 to 115%) between the 5 and 9 mo. measurement dates than noninoculated controls (26 to 62%) at the same fertility level. At the low rate of fertilization, growth of all three oak species was poor during the 5 to 9 mo. interval. However, inoculated plants fertilized at the one fourth rate did grow slightly more (1 to 3 cm) than noninoculated control plants fertilized at the same rate.

Table 6. *Pisolithus tinctorius*/fertilizer interactions in growth of red oak seedlings grown in 15 cm pots in the outdoor container nursery.¹

	Height (cm)			
	1.1 kg fertilizer/m ³		4.5 kg fertilizer/m ³	
	5 mo.	9 mo.	5 mo.	9 mo.
Control	4.3±1.0	4.6±1.0	8.8±0.9	11.1±1.2
<i>P. tinctorius</i> (M1)	6.4±0.7	7.4±0.5	9.6±1.3	17.7±3.5

¹ 19-6-10 + Fe Sierrablen. Means of 10 plants. Seedlings were transplanted from quart milk cartons to 15 cm (1 gallon) containers.

Table 7. *Pisolithus tinctorius*/fertilizer interactions in height growth of swamp chestnut oak seedlings grown in 15 cm pots.¹

	Height (cm)	
	5 mo.	9 mo.
Control	15.9±2.7	22.1±3.0
<i>P. tinctorius</i> (M1)	14.6±2.1	31.4±4.6

¹ 19-6-10 + Fe Sierrablen, 4.5 kg/m³ Means of 12 plants.

Pin oak seedlings responded differently to the two isolates of *Pisolithus tinctorius* under differing experimental conditions. In the greenhouse — Leach tube experiment, isolate M3 pro-

Table 5. *Pisolithus tinctorius*/fertilizer interactions in growth of pin oak seedlings grown in 15-cm containers in the outdoor container nursery.¹

	Height (cm)						Stem diameter (mm)					
	1.1 kg fertilizer/m ³		4.5 kg fertilizer/m ³		1.1 kg fertilizer/m ³		4.5 kg fertilizer/m ³		1.1 kg fertilizer/m ³		4.5 kg fertilizer/m ³	
	5 mo.	9 mo.	5 mo.	9 mo.	5 mo.	9 mo.	5 mo.	9 mo.	5 mo.	9 mo.	5 mo.	9 mo.
Control	10.1±0.7	11.2±0.7	12.3±0.6	20.0±1.0	3.5±.1	3.7±.2	4.2±.1	4.2±.1	3.5±.1	3.7±.2	4.2±.1	6.3±.3
<i>P. tinctorius</i> (M1)	11.9±0.8	15.3±1.7	16.3±1.3	33.0±2.2	4.4±.2	5.6±.5	4.8±.2	4.8±.2	4.4±.2	5.6±.5	4.8±.2	8.9±.6
<i>P. tinctorius</i> (M3)	11.3±0.9	14.5±1.6	12.5±1.4	24.4±2.1	4.2±.2	5.3±.3	4.4±.2	4.4±.2	4.2±.2	5.3±.3	4.4±.2	6.9±.9

¹ 19-6-10 + Fe Sierrablen. Seedlings were transplanted from Leach 'Super Cells' when 4-mo-old into 15 cm containers. Means of 10 to 25 seedlings.

duced superior growth (Table 4) while in the outdoor container experiment, isolate M1 produced superior growth (Table 5).

DISCUSSION

These studies indicate mycorrhizal fungi may be beneficial to plant growers in a number of ways: maximizing plant growth and producing salable plants in a minimum time resulting in more efficient greenhouse utilization, reduced labor costs, and greater production turnover; improving plant appearance due to decreased nutrient deficiency symptoms; improving fertilizer utilization or decreasing fertilizer needed to produce plants the same size as nonmycorrhizal plants; and the opportunity to produce "super seedlings", plants specifically infected with a mycorrhizal fungus ecologically adapted to adverse conditions frequently encountered by consumers.

Invariably, we obtained superior growth of seedlings by inoculation with mycorrhizal fungi. Conifers and oaks grew better when inoculated with *Pisolithus tinctorius*, and redbud and magnolia grew better when inoculated with *Glomus fasciculatus*. Obtaining a growth response to mycorrhizal fungi depends upon at least two factors: the rate at which land or media become reinfected with natural mycorrhizal fungal inoculum, and the degree of dependency of a plant species on mycorrhizal fungi for growth. In Lexington, we apparently have little air-borne ectomycorrhizal inoculum so that land or media in greenhouse experiments seldom become infected; other geographical regions apparently have abundant air-borne ectomycorrhizal inoculum so that growth benefits of artificial inoculation may not be realized. Endomycorrhizal fungi are not air-borne; therefore, artificial media devoid of soil will always be deficient and the degree of endomycorrhizal deficiency observed in field production will depend on the efficiency of soil fumigation. Apparently, endomycorrhizal fungi are seldom eliminated but reduction of inoculum may produce an early lag in growth. At 3 months, inoculated redbud plants were 92% larger than uninoculated plants, while at 5 months, they were only 72% larger. The "catching up" of uninoculated plants late in the growing season may be due to an increase in indigenous endomycorrhizal inoculum or a lag in time of infection by indigenous propagules compared to the inoculated plots.

Conifers produced in mycorrhizal-deficient nurseries often have foliar discolorations typical of nutrient deficiencies, especially of phosphorus. Similar conditions may be found with endo mycorrhizal plants, such as sweet gum. While plants may be large enough to sell, they are not appealing to consumers. We noticed this particularly in Scotch pine, inoculated plants being a healthy green color while uninoculated plants were an

unthrifty chlorotic to purple color. Suitable mycorrhizal fungi yield plants with greater visual appeal than mycorrhizal-deficient plants.

Fertilizer is a significant cost item to nurserymen, especially those who use expensive slow-release types. These studies demonstrated that mycorrhizal fungi produced a better growth response when fertilizer was supplied at the recommended rate. Such a response was not obtained at one-fourth the recommended rate; however, in the event a grower was not interested in maximum size but rather desired maximum return on his fertilizer investment, a suitable rate between the two levels used here could probably be found. With a mycorrhizal plant, it should take less fertilizer to produce the same size non-mycorrhizal plant.

Ecologically-adapted mycorrhizal fungi show much potential for allowing superior growth of trees on such adverse sites as strip mine spoils. It appears probable that for any given set of soil conditions, mycorrhizal fungi adapted to those conditions can be found. Homeowners often must contend with adverse soil conditions, such as scalping and removal of top soil by builders, and alkaline soil conditions around building foundations due to mortar. Cost of plants is often not a factor to consumers if fail-safe plants can be obtained. The use of ecologically-adapted mycorrhizal fungi to meet such a demand has not been attempted, but probably mycorrhizal fungi more than any other factor offers potential for producing a fail-safe plant. It seems certain that there will soon be a market for specifically-infected seedlings.

In our studies, adequate fertilization was essential for obtaining growth responses to mycorrhizal fungi. Especially with endomycorrhizal fungi, high fertilization has been reported to inhibit infection and annual growth responses. We found no such effects with either ecto- or endomycorrhizal associations. Probably magnolia and redbud are species with high dependencies for endomycorrhizal fungi.

The two isolates of *P. tinctorius* we studied produced differing responses with pin oak under differing conditions. One isolate was superior with plants growing on peat-perlite in the greenhouse, the other being superior with plants growing on composted hardwood bark-shale in large containers outdoors. Definite conclusions cannot be made with regard to these differences; however, it seems probable that fungal isolates will have to be screened and selected for precisely defined production conditions in order to select isolates which produce the desired plant responses.

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BRUCE BRIGGS: We know that with high fertility it is hard to get a mycorrhizal infection. You appear not to have had this problem. Do you think it is related to your use of encapsulated fertilizer?

DALE MARONEK: You are right about high fertilizer levels inhibiting mycorrhizal establishment. We are presently examining the influence of encapsulated fertilizers on mycorrhizal infection. You may be right that the slow release types are not inhibiting.

MIKE DIRR: Can you compensate for the lack of growth in your pines, which looks like phosphorus deficiency, by adding additional phosphorus?

DALE MARONEK: In some cases, yes and in other cases, no.

MIKE DIRR: How host specific are the mycorrhizal fungi?

DALE MARONEK: Some are very broad and others are quite species specific.

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