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CULTURAL ENHANCEMENT OF MYCORRHIZAL FUNGI ON SOUTHERN PINES?

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Abstract -- The need for cultural enhancement of mycorrhizal fungi in the cultural of southern pines is discussed. It is suggested that any active program to improve seedling quality through enhancing mycorrhizal colonization can only be justified by demonstrating that more mycorrhizae are better, or different species of mycorrhizae improve seedling performance. Then the increase in performance must be greater in value than the costs incurred. With respect to these questions we have documented a case of mycorrhizal deficiencies frequently encountered in expanded bare root nurseries in the southeastern United States. Secondly we argue that claims of growth gains, after outplanting, due to inoculation with *Pisolithus tinctorius* may in fact be due to increased diameter in the nursery. We conclude by suggesting that insufficient evidence is found that would justify an active mycorrhizal enhancement program in the southeastern United States and discuss the data needed if such a program was to be generally practiced in operational regeneration programs regardless of region.

INTRODUCTION

The nature of forestry has changed in many parts of the world. In many places, forest management is no longer concerned with silvicultural manipulation to economically produce timber products with respect to sustainability (i.e., investment in regeneration) and maintaining environmental quality (i.e., Best Management Practices). In some cases forestry is practiced with respect to social goals of wilderness protection, maintenance of diversity, preservation of ecosystems. Others still are mined for timber without concern for the quality of the forests in the next rotation. Others still are managed for timber, but are located on public lands where less concern for profitable management exists. Regeneration strategies used in these systems may fundamentally differ from that when profitable timber management determines management practices.

The Southeastern region is rapidly becoming the woodbasket of the United States. Presently, the southeast accounts for nearly one-third of sawtimber, one-half of the plywood and two-thirds of the pulpwood produced in the United States. Timber and timber related enterprises are important to the economic structure of the region. In several southern states, timber is the most important sector of the manufacturing base and is the major crop. Due to a low percentage of federally owned land, timber production in the South is essentially a private exercise. Less than 5% of timberland is owned by the public, thus private landowners are supplying the raw materials that support this industry. In large part, these landowners manage their woods for multiple benefits; nevertheless, profitable timber management is consistently an important priority in their management. Few practices are included in the silvicultural systems unless they are demonstrably profitable.

Regeneration costs are perhaps the most intensely scrutinized. Costs of artificial regeneration in the southern U.S. can often reach $300 (US) per hectare. This cost is usually carried throughout a rotation of 25-35 years. Any additional cost in regeneration must produce an increase in value of wood produced that will achieve an acceptable return.

Several researchers have suggested that cultural practices be incorporated in seedling production so that enhanced mycorrhizal colonization is achieved. However, these practices often increase the cost of seedlings. For instance, inoculating seedlings with vegetative mycelium can increase costs by almost $8.00 per thousand seedlings. Before wide-spread inoculation programs of this sort are operationally practiced one or two simple questions must be answered. Is more mycorrhizae better, or is different mycorrhizae on root stems of pine advantageous?

Mycorrhizal deficiency

Some level of mycorrhizal colonization is necessary for adequate development of the southern pines. Without mycorrhizae, slash pine (*Pinus elliottii*) in Puerto Rico grew to a height of approximately 12 cm in five years (Vozzo, 1971). However, mycorrhizal deficiencies are not common in the southern pine region of the United States. Under most circumstances, pine seedlings are rapidly infected with ectomycorrhizal symbionts. Due to the ubiquitous nature of the air-borne spores, it is very difficult to grow non-mycorrhizal pine seedlings. In Georgia, an electronically air-filtered, plant-growth room is usually required to grow non-ectomycorrhizal seedlings (Marx and Bryan, 1969).

However, under certain conditions ectomycorrhizal deficiencies in tree nurseries have been reported. This deficiency occurs on newly expanded bare root pine nurseries on ``new ground." New ground can be defined as soil having no previous history of producing an ectomycorrhizal crop (South et al., 1988). Although eventually these seedlings become mycorrhizal, the delay in development retards seedling growth. The lack of early mycorrhizal development on seedlings growing on new ground can be observed by the stunted growth P deficient foliage on seedlings suffering from this condition (often showing a reddish-purple color at the needles' tips). In many cases interspersed throughout the seedbeds will be pockets of healthy seedlings in which viable inoculum was present and the seedlings developed mycorrhizae on their root systems early in their development. In Ohio, Indiana, and Missouri considerable difficulty of a similar nature was experienced in nurseries newly established on land previously used for agriculture (Auten, 1945).

The problem of mycorrhizal deficiencies in new nurseries has been known for some time. In the United Kingdom, a common practice was to inoculate new nurseries with mycorrhizal fungi from the transplants (Steven, 1932). Unfortunately, transplants are not common in the southern United States. Therefore, the problem can be mitigated by (1) artificial inoculation with vegetative mycelia (at a potential cost of $10 per thousand seedlings); (2) artificial inoculation with ectomycorrhizal spores (at a cost of $0.43 per thousand seedlings); or (3) early application of P fertilization (at a cost of $0.19 per thousand seedlings). Fertilization with a soluble form of P, phosphoric acid, as late as the end of July, resulted in only an 8% cull produced as compared to 62% culls when no P fertilization was done on ground suffering from ``New Ground Syndrome" (South et al., 1988). However, P fertilization may be less desirable since a greater level of ectomycorrhizal infection would result from artificial inoculation. This brings us to the question of whether if some is good, is more better?

Is more mycorrhizae better?

For southern pines, *Thelephora terrestris* (Tt) is the most common ectomycorrhizal symbiont. We are aware of no data to show growth benefits from increasing the percentage of short roots infected with this fungus. One study showed that container-grown pine seedlings with 26% Tt were no better than seedlings with 21% Tt (Ruehle et al., 1981). In fact, after two years in the field, seedlings planted with 16% Tt (and fertilized at a higher rate in the nursery) had 32 to 85% more volume per plot than seedlings with 26% Tt (Ruehle et al., 1981). Since the fertilized trees had larger root collar diameter (RCD) at planting (2.5 mm vs 1.8 mm) this suggests that a 0.7 mm difference in RCD at time of planting may be more important for growth than a 10 percentage point increase level of Tt infection. Unfortunately, there are no published studies that compare seedlings of equal morphology with different levels of naturally occurring Tt.

One study compared seedlings of equal morphology and varied the level of Tt by physically stripping ectomycorrhizae from root systems (Marx and Hatchell, 1986). They reported that stripping root systems by hand and reducing the level of Tt from 45% (on the controls) to 2%, reduced survival and growth. They concluded that reduced seedling performance resulted from removal of short roots infected with Tt. However, work by South and Stumpff (1988) showed that root stripping also reduces root growth potential. Further unpublished work has shown that root growth potential is also reduced when root stripping is done on non-mycorrhizal seedlings. Therefore, the observed reduction in survival and growth might be simply explained by a reduced ability to rapidly produce new roots.

Different is better?

Although Tt is a very common ectomycorrhizal symbiont, there are many other species that occur in the South. Benefits of artificial inoculation with *Pisolithus tinctorius* (Pt) has been shown for seedlings used in strip mine reclamation (Cordell et al., 1991). Of all the ectomycorrhizal inculcation programs tested to improve seedling performance on forest regeneration sites in the southern United States, Pt is the most widely tested fungus. Over the past decade the number of loblolly pine seedlings inoculated with Pt has continued to increase. However, most of the seedlings are used by the U.S. Forest Service and by coal companies. In 1990, only 0.5% of 1,500,000,000 loblolly pine seedlings were artificially inoculated with Pt. Although the technique has been available for years, the practice of inoculating southern pines for operational forest management has not been widely accepted, partly due to the experience that growth improvements attributable to Pt are less consistent and reduced in magnitude than those observed on rehabilitated coal spoils.

Why so few Pt seedlings?

As previously mentioned, costs incurred in regeneration must justify their expense. Thus for Pt seedlings to become a part of southern pine regeneration practices they must outperform their counter parts. In some reports, no benefits in growth due to inoculation have been reported (Leach and Gresham, 1983; Powers and Rowan, 1983). In some cases, inoculation with Pt has resulted in decreased field survival of container-grown stock (Echols et al., 1990). It appears that a consistent positive effect from artificial inoculation with Pt is difficult to obtain on average sites in the South.

In many cases, inoculation with Pt in the nursery increases seedling size in the nursery (Marx et al., 1984). In one case, inoculation of sand pine increased basal area of seedlings by 72% (Hatchell and Marx, 1987). As a result, volume per plot after 7 years was 35% greater for the seedlings with larger initial root collar diameters. In some cases, seedlings inoculated with Pt are larger at time of outplanting and survive and grow better than those with natural inoculated mycorrhiza. When initial seedling size and Pt infection levels are confounded, the results can be difficult to interpret. Therefore, Barnett (1982) suggests that in order to have a valid test of the hypothesis (that Pt inoculated seedlings perform better than non-inoculated seedlings), inoculated and control seedlings should have similar morphology at the time of outplanting. Although it is possible to design studies to test the effect of inoculation independent of initial differences in seedling size (e.g. Letho, 1990), few studies with southern pines have attempted to do so.

In one study, there was no significant different in initial morphology of loblolly pine seedling but large difference in Pt infection (Hatchell and Marx, 1987). Even though 55% of the short roots were infected with Pt at time of outplanting, the growth of loblolly pine after 7 years was no different than seedlings infected only with Tt. Despite the fact the seedlings were planted on a deep sandy soil, the presence of Pt did not improve seedling growth. At a cost of $10/liter for inoculum and a seedbed density of 302/m^2, the cost of inoculum for this study was $26.80/thousand seedlings. Although the cost of artificial inoculation doubled the seedling cost, there was no improvement in nursery growth and no improvement in field performance.

One study that attempted to minimize initial differences in seedling size was reported by Marx et al. (1988). Even though Pt inoculation in the nursery caused an above average increase in seedling size (a 40% increase in seedling basal area and a 39% increase in seedling weight), seedlings from the smallest Pt plot were selected so they more closely matched the control seedlings. These seedlings were outplanted and field performance was measured after 8 years. The authors suggest that Pt seedlings were able to withstand the stress due to drought after outplanting better than nursery run seedlings. They speculate that Pt inoculated seedlings were growing better during the 5th year after outplanting as a result of Pt remaining and expanding on the root system. However, closer scrutiny of the data may lead to other conclusions. If one accounts for the growth of different size seedlings by using seedling size as the X-axis and growth as the Y-axis it become apparent that Pt seedlings grew similarly to those that were not inoculated (see Figure 1). This suggests that, at least after the first year of growth, Pt seedlings had an advantage of size and this advantage was expanded upon after subsequent growth.

For species like longleaf pine, cost of experimental treatments involving Pt inoculation may cost more than $50 per thousand seedlings (Hatchell and Marx, 1987). If this treatment does not increase seedling size in the nursery, then the probability of an improvement in field performance may be minimal. However, increasing seedling size in the nursery can be inexpensively accomplished by sowing early, fertilizing more, lowering seedbed density, and proper irrigation. In some studies, naturally inoculated longleaf pine seedlings grown in the nursery at a density of 65/m^2 have out-performed seedlings inoculated with Pt and grown at 161/m^2 (Hatchell, 1986). For this study, cost of the Pt treatment was $142/thousand plantable seedlings ($10.80/$m^2$/76 plantable seedlings). Thus at this time, evidence is not persuasive to include inoculation of Pt in a regeneration strategy when profitable timber management is the objective.

Experiences from the southeastern United States may be relevant to forest management throughout the world, even in areas that do not necessarily manage for profitable timber production. Before wide-spread inoculation programs are started, the goals of management need to be clearly defined so that the benefits of the program can be judged within the context of the goals. Then rigorous criteria need to be defined so that the gains from various alternatives can be assessed. Lastly, through experimentation including outplanting performance, mycorrhizal inoculation can be judged with respect to the benefits, costs and alternatives that might meet the same criteria with less expense.

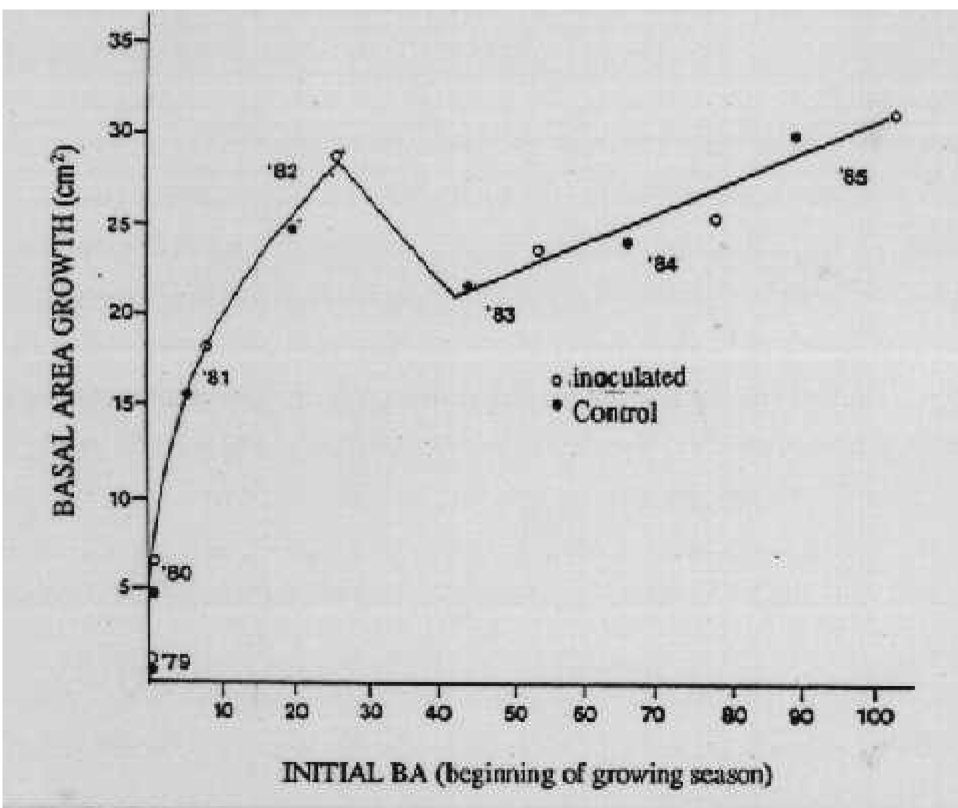

Figure 1. Effect of initial basal area, at the beginning of each growing season (x-axis) and inoculation with Pt on annual basal-area growth (y-axis) of loblolly pine (Mexal and South, 1991; adapted from Marx et al., 1988).

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