

MYCORRHIZAE AND ESTABLISHMENT OF TREES ON STRIP-MINED LAND^{1, 2}

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ABSTRACT

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This paper presents a brief introduction into ecto- and endomycorrhizal associations of plants and discusses their potential value in revegetation, especially reforestation, of strip-mined lands. The significance of ecologically adapted ectomycorrhizal fungi, such as *Pisolithus tinctorius*, to survival and growth of pines and other tree species has practical importance in these forestation efforts. Data from pilot studies on strip-mined coal spoils in Ohio, Virginia and Kentucky show that pines tailored with *Pisolithus* ectomycorrhizae prior to planting survive and grow significantly better than trees with other fungal symbionts. The performance of grasses and other herbaceous plants used in revegetation of strip-mined lands may also be improved by use of specific endomycorrhizal fungi. Manipulation of mycorrhizal fungi on roots of plants for revegetation purposes has great potential in the reclamation of drastically disturbed lands.

Microorganisms are present in great numbers near the feeder roots of plants and they play vital roles in numerous physiological processes. These dynamic microbial processes involve saprophytism, pathogenicity, and symbiosis. The most widespread symbiosis of plants is the mycorrhizal association which involves various root-inhabiting fungi and plant feeder roots. The prevalence of mycorrhizal associations on plants is so common under natural soil conditions that a non-

mycorrhizal plant is the exception rather than the rule. Only a few plants, such as sedges, crucifers and certain aquatics, do not form mycorrhizae. Other plants, and especially those of major economic importance to man, such as forest trees and agronomic crops, form abundant mycorrhizae on their roots.

There are three classes of mycorrhizae on plants. Ectomycorrhizae occur naturally on many of the important forest tree species of the world. All members of the gymnosperm family *Pinaceae*, e.g., pine, spruce, fir, larch, hemlock, etc., as well as certain angiosperms, e.g., willow, poplar, aspen, walnut, hickory, pecan, oak, beech, eucalypt, and others, are ectomycorrhizal. Several of these tree species can be either ectomycorrhizal or endomycorrhizal, depending on soil conditions.

ECTOMYCORRHIZAE

Ectomycorrhizal infection is initiated from spores or hyphae (propagules) of the fungal symbionts in the rhizosphere of feeder roots. The propagule is stimulated by root exudates and grows vegetatively over the feeder root surface, forming the fungus mantle. Following mantle formation, hyphae develop intercellularly in root cortex, forming the Hartig net which may completely replace the middle lamellae between the cortex cells. The Hartig net is the main distinguishing feature of ectomycorrhizae. Ectomycorrhizae may appear as simple unforked roots, bifurcate roots, multiforked (coralloid) roots, nodular-like roots, or in other configurations (fig. 1). Color of ectomycorrhizae is apparently determined by the color of the hyphae of the fungal symbionts. Ectomycorrhizae may be brown, black, white, red, yellow, or blends of these colors. Individual hypha, strands of hyphae, or rhizomorphs may radiate from fungus mantles into the

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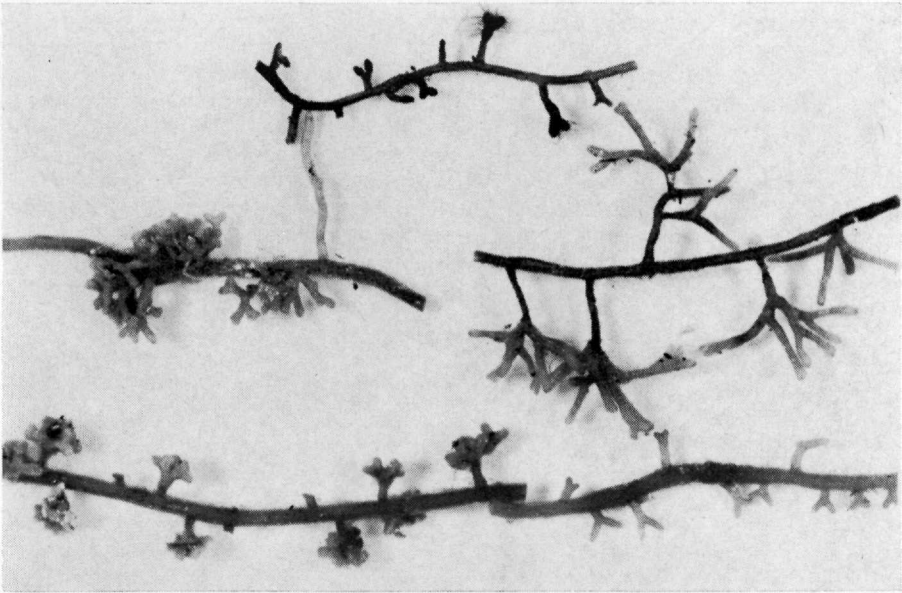


FIGURE 1. Five morphologically distinct forms of ectomycorrhizae of *Pinus taeda*, each formed by a different species of ectomycorrhizal fungus.

soil and to the base of fruit bodies of the fungi.

Most fungi which form ectomycorrhizae with forest trees are Basidiomycetes that produce mushrooms or puffballs. Certain Ascomycetes, such as truffles, also are symbiotic. Over 2,100 species of ectomycorrhizal fungi have been estimated to exist on trees in North America. These fungi produce millions of spores in their fruit bodies that are widely disseminated by wind and water. Most ectomycorrhizal fungi are dependent on their tree hosts for essential carbohydrates, amino acids, vitamins, etc., in order to complete their life cycles. Ectomycorrhizal development, therefore, is usually a prerequisite for fruit body production by these fungi, however, not all fungal species which form mushrooms and puffballs are ectomycorrhizal. Many of these fungi are saprophytes that play an important role in the decomposition of organic matter and the mineral cycle in forest soils.

Under normal forest soil conditions, many species of fungi are involved in the ectomycorrhizal associations of a forest, a single tree species, an individual tree,

or even a small segment of lateral root. As many as three species of fungi have been isolated from an individual ectomycorrhiza. Just as a single tree species can have numerous species of fungi capable of forming ectomycorrhizae on its roots, a single fungus species may enter into ectomycorrhizal association with numerous tree species. Some fungi, however, are apparently rather host specific, whereas others have broad host ranges and form ectomycorrhizae with numerous tree genera in diverse families.

ENDOMYCORRHIZAE

Most of the economically important agronomic and forage crops, as well as fruit and nut trees, such as peach, citrus, apple, plum, cherry, almond, etc., normally form endomycorrhizae. Important forest trees, such as maples, elms, gums, ash, sycamore, cottonwood, alder, and dogwood, and other trees not forming ectomycorrhizae normally form endomycorrhizae. As mentioned earlier, some of these trees can form both endo- and ectomycorrhizae. Endomycorrhizal fungi form a loose network of hyphae on feeder root surfaces, and do not develop the

dense fungus mantle found on ectomycorrhizae. Most often these fungi form large, conspicuous, thick-walled spores on the root surfaces, in the rhizosphere, and sometimes in feeder root tissues. Hyphae of the endomycorrhizal fungi penetrate the cell walls of the epidermis and progress into the cortical cells of the root. These infective hyphae may develop specialized absorbing or nutrient-exchanging structures (haustoria) called arbuscules in the cortex cells. Thin-walled, spherical-to-ovate vesicles may also be produced in the cortex cells by the fungus. The term "vesicular-arbuscular" mycorrhizae has been coined to denote this type of endomycorrhizae. As in ectomycorrhizae, endomycorrhizal fungus infection does not occur in meristematic or vascular tissues. Unlike ectomycorrhizae, however, endomycorrhizal infection does not cause major morphological changes in roots. Endomycorrhizae may have a yellowish color in comparison to noninfected roots.

The fungi which form endomycorrhizae with trees are mainly Phycomyces. They do not produce large, above-ground fruit bodies or wind-disseminated spores as do most ectomycorrhizal fungi, but some of them produce large zygospores and chlamydospores on or in roots. Some species also produce large sporocarps (5–10 mm diameter) containing many spores on roots. These fungi spread in soil by root contact, moving water, insects, or mammals. Most endomycorrhizal fungi of trees belong to the family *Endogonaceae*. Several genera and species have been identified; many more undoubtedly exist (Gerdemann and Trappe, 1974). They are so widespread that it is extremely difficult to find natural soils anywhere in the world that do not contain them. In the absence of a host, the spores of these fungi are able to survive for many years in the soil. Based on the limited amount of research done on endomycorrhizal fungi, most species appear to have very broad host ranges. For example, *Glomus mosseae* (= *Endogone mosseae*) will form endomycorrhizae with cotton, corn, pepper, soybeans, sorghum, and other agronomic crops, as well as sycamore, sweetgum, citrus, peach, and black locust.

ECTENDOMYCORRHIZAE

Another class of mycorrhizae are ectendomycorrhizae which are of lesser ecological importance than the other classes. They have the features of both ecto- and endomycorrhizae. Ectendomycorrhizae have a limited distribution in forest soils and are primarily found on roots of normally ectomycorrhizal forest trees. Very little is known about the species of fungi involved or their significance to tree growth, since only limited research has been attempted on them.

Ectomycorrhizal fungi are beneficial to the growth and development of trees. For some trees, such as *Pinus*, they are indispensable for growth under natural field conditions. Trees with abundant ectomycorrhizae have a much larger, physiologically active, root-fungus surface area for nutrient and water absorption than do trees with few mycorrhizae. This increase in surface area comes not only from the multi-branching habit of most ectomycorrhizae, but also from the extensive vegetative growth of hyphae of the fungus symbionts from the ectomycorrhizae into the soil. These extramatrical hyphae function as additional nutrient and water-absorbing entities. Ectomycorrhizae are able to absorb and accumulate in the fungus mantles nitrogen, phosphorus, potassium, and calcium more rapidly and for longer periods of time than nonmycorrhizal feeder roots. The fungi of ectomycorrhizae assist in the degradation of certain complex minerals and organic substances in soil and render essential nutrients from these materials available to the tree. Ectomycorrhizae also appear to increase tolerance of trees to drought, high soil temperatures, soil toxins (organic and inorganic), and extremes of soil pH caused by high levels of sulfur or aluminum. Ectomycorrhizae function as biological deterrents to infection of feeder roots by root pathogens, such as species of *Pythium* and *Phytophthora*. Induced hormone relationships in ectomycorrhizae by fungal symbionts extend the longevity (length of physiological activity) of these roots as compared to nonmycorrhizal roots.

Very little research has been accomplished on the value of endomycorrhizae to forest trees. Available research results

indicate that endomycorrhizae are more efficient nutrient-absorbing roots than are nonmycorrhizal roots. This applies especially to the absorption and utilization of phosphorus. The extensive network of hyphae of these fungi growing from endomycorrhizae exploit large volumes of soil that are rarely exploited by nonmycorrhizal roots. It is not known for certain if some trees are dependent on endomycorrhizae for growth as *Pinus* species are on ectomycorrhizae, however, recent evidence obtained from research at the Athens, Ga. laboratory indicates that sweetgum may need endomycorrhizae in order to become established after seed germination and to grow normally.

Many factors affect mycorrhizal development. In discussing them it is necessary to separate those which affect the tree from those which affect the fungal symbionts. The main factors influencing the susceptibility of tree roots to mycorrhizal infection appear to be photosynthetic potential and soil fertility. High light intensity and low to moderate soil fertility enhance the degree of mycorrhizal development, whereas the other extreme of these conditions, i.e., low light intensity (below 20% of full sunlight) and excessively high soil fertility reduce, or may even eliminate, mycorrhizal development. These factors appear to influence the biochemical status of feeder roots, such as controlling levels of reducing sugars or the synthesis of new feeder roots, which are prerequisites to symbiotic infection. Roots growing rapidly due to high soil fertility may actually outgrow their fungal symbionts. Generally, any soil or above-ground condition which influences root growth also influences mycorrhizal development to one degree or another. Factors which affect the fungal symbionts directly are those which regulate survival or growth of infective propagules or vegetative growth of the symbionts on roots. Extremes of soil temperatures, pH, moisture, etc., and presence of antagonistic soil microorganisms can affect the symbionts and thereby influence the mycorrhizal potential of the soil.

Several excellent texts have been published in recent years which compre-

hensively cover the different mycorrhizal associations (Harley, 1969; Marks and Kozlowski, 1973; Hacksaylo and Tompkins, 1973).

MYCORRHIZAE AND AFFORESTATION PRACTICES

Over 3,000 papers have been published on mycorrhizae of plants and most of these relate to forest trees (Hacksaylo and Tompkins, 1973). Much of this research has been very basic and has broadened our understanding of their complexity in forest soil ecosystems. Various studies have shown that a prerequisite to growth of many forest trees is the presence of ectomycorrhizae on their roots. This means that certain trees, such as *Pinus*, have an obligate requirement for ectomycorrhizae and cannot grow normally without them. This basic information shows that ectomycorrhizae formed by any fungus is better than no ectomycorrhizae at all on tree roots, especially pines. This point has practical significance to afforestation programs with normally ectomycorrhizal trees in areas of the world where their symbiotic fungi do not occur naturally. Mikola (1969) has demonstrated that there must be a parallel introduction of the essential ectomycorrhizal fungi if afforestation with these trees (*Pinus* in particular) is to succeed. There are many areas of the world where indigenous ectomycorrhizal trees and their symbiotic fungi do not occur naturally. In these areas, i.e., the high Andes of Peru (Marx, 1976a), Puerto Rico (Vozzo and Hacksaylo, 1971), Africa (Gibson, 1963), Australia (Bowen *et al.*, 1973), Asia (Oliveros, 1932), subalpine areas of Austria (Moser, 1963), former agricultural soils of Poland (Dominik, 1961), oak shelterbelts in the steppes of Russia (Imshenetskii, 1967), former treeless areas of the United States (Hatch, 1937), forestation attempts were either total or near failures until ectomycorrhizal infection occurred on tree roots. Symbiotic root infection was insured either by introductions of pure cultures of the fungi, soil containing ectomycorrhizal fungi, or manipulation of soil containing low levels of indigenous symbiotic fungi to encourage ectomycorrhizal development.

Natural soils of the world which have supported vegetation in the past have not been reported void of endomycorrhizal fungi. The broad host range of most species of endomycorrhizal fungi normally ensures endomycorrhizal development on desirable plants. Failure in afforestation of normally endomycorrhizal trees due to a deficiency of endomycorrhizal infection, therefore, has not been reported.

MYCORRHIZAE AND REVEGETATION OF STRIP-MINED LANDS

In observing the physical disruption of overburden strata during strip-mining operations, it becomes quite apparent that the resulting surface material does not physically, chemically, or biologically resemble any other landscape in the world. Physically, the surface material is often very rocky, steeply sloped, and depending on its mineral composition, has variable weathering rates. This material may be quite dark in color and absorb sufficient radiant energy to cause high surface temperatures. Chemically, the surface material may contain high levels of sulfur, aluminum, manganese, etc., which are limiting to plant growth. The material may contain only small quantities of the essential elements for normal plant growth. Biologically, any macro- or microbiological system which existed in the surface soil prior to stripping may now be buried dozens of feet under the stripped overburden. Some of the original topsoil may be mixed throughout the stripped profile, but even under these conditions the surface material is nearly a biological desert in comparison to the biological status of the original profile.

The status of the mycorrhizal potential of the new surface material is of concern to us. The literature is well documented with descriptions of the various physical and chemical properties of strip-mined lands (Hutnik and Davis, 1973). The questions I shall attempt to answer are—Can indigenous and recolonizing mycorrhizal fungi survive these "soil" properties? Are mycorrhizae essential to all plants which may naturally or artificially revegetate this surface material? Is there

significant potential for ecological selection among the vast number of species of mycorrhizal fungi to assure that adaptable ones will maintain themselves on roots of plants in these disturbed sites?

Ectomycorrhizal Associations.—There has been only limited research on mycorrhizal associations of plants on strip-mined lands and mining wastes. Schramm (1966) published a classic piece of work on plant colonization of anthracite wastes in Pennsylvania. He concluded that early ectomycorrhizal development was essential for seedling establishment of *Betula lenta*, *B. populifolia*, *Pinus rigida*, *P. virginiana*, *Populus tremuloides*, *Quercus rubra*, and *Q. velutina* on this waste material. The only generally successful original plant colonists of this bare and predominantly nitrogen-deficient waste were either nitrogen-fixing plants or certain ectomycorrhizal tree species. Schramm suggested that due to their year-round effectiveness, evergreen trees (pines) should receive special attention, and furnished strong evidence in support of his conclusions. Seedlings from either wind-blown or artificially planted seed of these tree species that did not have ectomycorrhizae were chlorotic and soon died. The majority of surviving seedlings, and especially those growing well, were heavily ectomycorrhizal. The main basidiomycetes observed by Schramm that developed near the surviving tree seedlings were *Inocybe lacera*, *Thelephora terrestris*, *Pisolithus tinctorius*, *Amanita rubescens*, and *Scleroderma aurantium* which form ectomycorrhizae on trees (Trappe, 1962). These observations by Schramm add additional evidence to that which has been already discussed on the need of these tree species for ectomycorrhizal associations.

Schramm (1966) traced the extensively developed mycelial strands formed by *P. tinctorius* from ectomycorrhizae of these various tree species through large waste volumes to the base of its basidiocarp. These mycelial strands are somewhat unique in that they are large and brilliant gold-yellow in color and were easily traced through the contrasting dark anthracite wastes. Some strands were traced through waste material as far as

15 ft from the seedlings to the basidiocarp. The ectomycorrhizae formed by *Pisolithus* were also yellow-gold in color and prolifically branched. He associated *P. tinctorius* ectomycorrhizae with the most vigorously growing seedlings. In most cases, it was the first symbiont on seedling roots. The other species of ectomycorrhizal fungi appeared on roots and produced basidiocarps primarily after litter had accumulated under the seedling canopy. These observations tentatively confirmed that *P. tinctorius* was the fungal symbiont forming the gold-yellow ectomycorrhizae on these tree species. Earlier, Bryan and Zak (1961) "synthesized" ectomycorrhizae with *P. tinctorius* on *Pinus echinata* seedlings in aseptic culture. This pine species, however, was not encountered by Schramm in Pennsylvania.

Schramm's work strongly suggests that only a few ectomycorrhizal fungi are capable of ecologically adapting to soil conditions on the anthracite wastes. High soil temperatures could be a limiting factor in specific mycorrhizal establishment. Marx and coworkers (1970) found that *P. tinctorius* formed more ectomycorrhizae on *P. taeda* seedlings at a constant soil temperature of 34°C than it formed at either 14, 19, 24, or 29°C. *Thelephora terrestris* did not form ectomycorrhizae at 34°C soil temperature and it formed more between 14 and 24°C than at 29°C. *T. terrestris* is one of the major ectomycorrhizal fungi on pine seedlings in nurseries throughout the United States, which probably accounts for its presence on planted coal spoils. It was on the seedling roots from the nursery prior to planting on the spoil. In a later study with these fungi, Marx and Bryan (1971) found *P. taeda* seedlings with *Pisolithus* ectomycorrhizae survived and grew as well at 40°C as they did at 24°C. Seedlings with *Thelephora* ectomycorrhizae or those without ectomycorrhizae did not survive well and did not exhibit growth at 40°C. These results may explain why *Pisolithus* is the primary symbiont on young volunteer seedlings on anthracite wastes. Schramm (1966) recorded soil temperatures between 35°C and 65°C in wastes at a depth of 2½ inches. Perhaps *Pisolithus* was dominant because high

soil temperatures restricted earlier establishment of the other fungi.

Prompted by Schramm's report and our findings on soil temperature, examinations were made in 1971 of various strip-mined wastes in the East. Our results were only observational, but we thought them to be significant. We found *Pisolithus* basidiocarps and its unique, gold-yellow ectomycorrhizae and mycelial strands to be the predominant, if not the only, ectomycorrhizal fungus on roots of *Pinus virginiana*, *P. taeda*, *P. resinosa*, and several *Betula* spp. on coal wastes in Indiana, Pennsylvania, Ohio, Virginia, West Virginia, Kentucky, Tennessee, and Alabama, as well as *P. echinata* and *P. taeda* on strip-mined kaolin wastes in Georgia. Some of these wastes had a soil reaction as low as pH 2.9, although most were between pH 3.5 and 5.5. *Pisolithus* has also been reported on coal wastes associated with *B. lenta*, *B. pendula*, *B. populifolia*, *Populus grandidentata*, *P. tremuloides*, and *Salix humilis* in West Germany (Meyer, 1968), *Pinus banksiana* in Missouri (Lampky and Peterson, 1963), and *Pinus* spp. in Indiana and Tennessee (Hile and Hennen, 1969).

On a coal waste near Fabius, Alabama, over 3,000 *Pisolithus* basidiocarps were observed under planted *P. taeda* in an area of less than ½ acre. From the mature basidiocarps in this area we collected over 1,300 g of dry basidiospores. There are approximately 1.1 billion spores per gram. These and other basidiospore collections have proved functional as inoculum for ectomycorrhizal development on pines (Marx, 1976b).

I previously discussed evidence which proved that any ectomycorrhizae on roots of trees such as *Pinus* are better than no ectomycorrhizae at all. It appears that one more premise can be made—certain species of ectomycorrhizal fungi are more beneficial to tree growth than others. Based on Schramm's observations and our reports, it appears that *Pisolithus* may be more beneficial to the establishment of *Pinus* and *Betula* species on strip-mined lands and wastes than are other species of ectomycorrhizal fungi. Making the assumption that *Pisolithus* ectomycorrhizae are instrumental in tree estab-

lishment and maintenance on strip-mined wastes, research was begun to develop techniques to "tailor" seedlings in the nursery with *Pisolithus* ectomycorrhizae. The working premises were simple—Why wait for natural means to establish *Pisolithus* on colonizing trees? Could tree survival and growth be improved by having *Pisolithus* ectomycorrhizae performed on the seedlings in the nursery prior to planting on the strip-mined lands? Recently, techniques were developed for this "tree tailoring" concept (Marx and Bryan, 1975) which were effective on several species of pines in conventional tree nurseries (Marx *et al.*, 1976c). Briefly, the techniques involved the production of pure culture, vegetative mycelial inoculum of *P. tinctorius* in a vermiculite-peat moss-nutrient substrate, or the use of basidiospores mixed with a physical carrier, such as moist vermiculite. Either inocula was used to infest artificially fumigated nursery soil and no further modifications of standard nursery practices were necessary. Effective soil fumigation (methyl bromide), done shortly before soil infestation, and the maintenance of reasonable levels of soil fertility through the growing season appear to be the two prerequisites for successful "tailoring" of pine seedlings with *Pisolithus*. These techniques also have been used successfully with other ectomycorrhizal fungi, such as *Thelephora terrestris* and *Cenococcum graniforme*.

The introduction of *Pisolithus* into nursery soils has significantly improved pine seedling quality in the nursery. Growth increases of between 100–150% after one growing season in the nursery have been encountered with seedlings of *P. taeda*, *P. strobus*, and *P. virginiana* (in the South) following successful soil infestation and ectomycorrhizal development by *Pisolithus*. After a few more years of research, ectomycorrhizal deficiencies in nurseries of many acres may be corrected by using pure cultures of highly beneficial symbionts. In the past these deficiencies were corrected by the addition of forest litter and humus to the nursery soil. This practice inadvertently introduced a considerable number of pests, such as weeds and disease-causing organ-

isms, into certain nurseries. Currently, *Pisolithus* is being tested in tree nurseries in Australia, Mexico, East Africa, Switzerland, and France, as well as in different parts of the United States. The practicability of introducing *Pisolithus* and other ectomycorrhizal fungi into the near-sterile root substrate of containerized seedlings is also being examined. Preliminary information suggests that growth in containers and field performance of these seedlings can be improved with specific ectomycorrhizae (Marx and Barnett, 1974). This application to containerized tree stock could be as relevant as it is to standard nursery-grown seedlings, since it is anticipated that in the near future container stock will account for as much as 20% of the seedlings used in reforestation in North America. Container-grown stock also has promise for reforesting strip-mined lands.

Since the techniques to "tailor" seedlings with *Pisolithus* ectomycorrhizae have only recently been developed, very little seedling performance information is available. In a greenhouse pot study, Nicholas (1971) found that *Pisolithus* ectomycorrhizae improved growth of *Betula pendula* and *Pinus resinosa* seedlings in variously amended bituminous coal spoils in comparison to seedlings without *Pisolithus*. Unfortunately, his test seedlings were not grown under the best of conditions during synthesis of ectomycorrhizae due to problems experienced in the mycorrhizal growth room located at the Athens, Ga. facility.

Since the winter of 1973, members of the Athens, Ga. Project have outplanted nearly 25,000 pine trees with specific ectomycorrhizae from Pennsylvania to Florida. These test plantings are located on strip-mined coal and kaolin spoils, eroded Copperhill, Tennessee, and routine reforestation sites. The field results look very promising, but due to the young age of the plantings, problems with field design, destruction of certain plots by cattle and trailbikes, and lack of winter hardiness of Georgia-grown seedling stock planted in the North, they can only be discussed in general terms. With few exceptions, seedlings of *P. taeda*, *P. virginiana*, *P. echinata*, *P. clausa*, *P. resinosa* and *P. elliotii* var. *elliotii* with

Pisolithus ectomycorrhizae survived and grew better than seedlings with ectomycorrhizae formed by other ectomycorrhizal fungi. The few exceptions can be explained on the basis of seedling condition and degree of ectomycorrhizal development at the time of planting. In certain tests, basidiospores of *Pisolithus* were placed on roots of standard nursery stock that had *Thelephora terrestris* ectomycorrhizae prior to planting on coal wastes. Some *Pisolithus* ectomycorrhizae developed from basidiospore inoculum and stimulated seedling height growth as much as 30 percent in comparison to seedlings without *Pisolithus*.

Last winter, pine seedlings grown in the Athens' nursery with either *Pisolithus* or *Thelephora* ectomycorrhizae were planted on strip-mined coal sites in Kentucky, Virginia and Ohio. The Kentucky site had been planted to pine five times previously on a yearly basis with nearly complete failure each year. In our planting on this site, survival of *P. virginiana* seedlings with *Thelephora* ectomycorrhizae was only 1.5 percent. Survival of seedlings with *Pisolithus* ectomycorrhizae was 45.5 percent. These seedlings grew significantly in one growing season and *Pisolithus* has completely colonized the roots and adjacent soil volumes. The Virginia site was not considered a major problem area and survival and growth of *P. taeda* seedlings were better than on the Kentucky site. *Pisolithus* ectomycorrhizae increased survival of these seedlings by 11%, and height and stem diameter by 21% and 26%, respectively, in comparison to seedlings with *Thelephora* ectomycorrhizae. In the Ohio planting near New Straitsville, *Pisolithus* ectomycorrhizae increased survival of loblolly pine seedlings by 34% and Virginia pine seedlings by 36% over similar seedlings ectomycorrhizal with *Thelephora*.

An inherent difficulty in research of this nature has become apparent to us and that is maintaining the integrity of the specific ectomycorrhizal association on these sites. *Pisolithus* recolonizes roots of test seedlings from natural sources so rapidly that valid growth comparisons between seedlings initially with and without *Pisolithus* is very erratic.

Quite often, over one-half of the seedlings without *Pisolithus* at planting will have variable quantities of *Pisolithus* ectomycorrhizae by the end of the first growing season on site.

Preliminary field results look promising, but are not in themselves conclusive. Much more research remains to be done. The results, however, point out the potential benefits of specific ectomycorrhizae on seedlings for specific sites. Many reforestation sites, including strip-mined lands, are characterized by soil factors which exert selective pressure on symbiotic fungi. Fungi which can tolerate these factors and are ecologically adapted to these sites, should be used to "tailor" seedlings prior to planting. Perhaps in this manner a persistent and physiologically active root system can be assured. This feature in itself may remedy some of the problems encountered in the past on the reclamation of these adverse sites. The same principle of symbiont selection and tailoring may also have application in the reforestation of more routine sites.

HOST RANGE AND GEOGRAPHIC DISTRIBUTION OF *PISOLITHUS TINCTORIUS*

There has been a great deal of interest generated throughout the world on the practical use of *Pisolithus* in re- and afforestation efforts on a variety of sites. Because of this interest and the obvious implications of plant quarantine in introducing *Pisolithus* to other areas, attempts have been made to obtain information from the literature and correspondence regarding its host range and geographic distribution. After a brief search, it became apparent that *Pisolithus* has a nearly worldwide distribution. It has been found in Europe, Asia, Africa, Central and South America, and in the Middle East. It has been reported in 22 states throughout the United States. Its tree host range is also quite impressive: Thirty-five species or varieties of pines, three species of *Betula*, two species of *Eucalyptus*, two species of *Populus*, and six species of *Quercus* have either been experimentally confirmed as hosts of *Pisolithus* or have been associated with this fungus consistently in the field. It

possible that "tailoring" these tree species with *Pisolithus* in the nursery or in containers may also improve their survival and growth in the field.

Endomycorrhizal Associations.—There has been only limited research on endomycorrhizae of plants on strip-mined wastes. Daft *et al.* (1974) found abundant endomycorrhizae on roots of a variety of herbaceous plants, including grasses, on anthracite and bituminous coal wastes in Pennsylvania and bituminous wastes in Scotland. They identified and collected spores of *Gigaspora gigantea* (= *Endogone gigantea*) from the Pennsylvania wastes and found that they would infect and stimulate growth of corn plants in spoil material. In addition to *G. gigantea*, other species of *Endogonaceae* were also found. They concluded that endomycorrhizae may be essential for the survival and growth of herbaceous plants on coal wastes.

Two of my associates (W. C. Bryan and W. J. Otrosina) have made observations on the incidence of endomycorrhizae on herbaceous plants on strip-mined land. On both artificially and naturally revegetated coal spoils in Kentucky and Virginia, as well as kaolin spoils in Georgia, they found a variety of wild and cultured grasses to be endomycorrhizal to one degree or another. They observed that not all seedlings of some grasses have endomycorrhizae on the same site. Planted and volunteer trees, such as sycamore, sweetgum, maples, and black alder, were also heavily endomycorrhizal. In all likelihood, the endomycorrhizal infection on the planted trees was on the roots prior to planting on the spoil, however, the significance is that these endomycorrhizae persisted.

Earl F. Aldon (USDA, Forest Service, Rocky Mountain Forest and Range Experiment Station, Albuquerque, New Mexico 87101, personal communication) found that endomycorrhizae significantly increased survival and growth of the four-wing saltbush (*Altriplex canescens*) on strip-mined coal spoils in New Mexico. Size index (height \times stem diameter) of the mycorrhizal seedlings was 231 and only 91 for the nonmycorrhizal seedlings after the second year on the spoil site.

Since some endomycorrhizal infection is apparent on wild grasses and volunteer trees on spoil material, the logical question to ask is, what was the source of the initial inoculum? As mentioned earlier, endomycorrhizal fungi do not have highly effective means of dissemination. Most geographic spread of these fungi is thought to be slow and mainly by moving water or, soil, insects, birds, and perhaps mammals (including man). Their presence on strip-mined lands may be accounted for through contamination of overburden material with the original topsoil. If some plants are dependent on, or at least stimulated by, endomycorrhizal infection, then perhaps by increasing the amount of available inoculum, either artificially or naturally, revegetation of spoil material with these plants may be enhanced.

It appears that research on the value of endomycorrhizae to survival and growth of plants, including trees, on strip-mined lands should be concentrated on testing a variety of fungi which persist on the spoil. In this manner, perhaps an ecologically adapted endomycorrhizal fungus can be found which may have a similar potential to that of the ectomycorrhizal fungus *Pisolithus tinctorius*. Apparently there are a multitude of species of endomycorrhizal symbionts of herbaceous plants from which the potential choice can be made (Nicolson, 1967). It would be extremely interesting to determine the value of endomycorrhizae to pioneer plants. Are they pioneer plants because they do not require endomycorrhizae for normal growth? If sufficient quality and quantity of endomycorrhizal inoculum were present in spoil material would the same succession of herbaceous species prevail? There is tremendous potential for practical research on this aspect of endomycorrhizal associations.

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