Loblolly pine mycorrhizae in East Tennessee

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A recent survey of six loblolly pine stands in East Tennessee yielded 53 species of fungi, six of which have previously been shown to be mycorrhizal with loblolly pine. In addition, four of the collected species formed by mycorrhizae with loblolly pine in synthetic culture.

Methods

Sporophores were collected from the soil surface of stand interiors in six stands of loblolly pixie *(Pinus taeda L.)* surveyed at intervals from October 1970 through December 1971. Identifications were confirmed from observation of fresh specimens.

Attempts were made to isolate pure cultures from all species collected. The procedure described by Hacskaylo (3) was used. Pure cultures of *Agaricus placomyces* Pk., *Amanita citrina* (Schaef.) S. F. Gray, *Collybia maculata* (Alb. & Schw.) Quel., *Leucopaxillus laterarius* (Peck) Singer & Smith, *Lactarius piperatus* (Fr) S.F. Gray, and *Russula fragilis (Fr.)* Fr. were obtained this way. Pure cultures of *Amanita rubescens (Fr.)* S.F. Gray, *Amanita virosa* Fr., *Amanita parcivolvata (Pk.)* Gilbert, *Boletus bicolor* Pk., *Clitocybe gibba (Fr.)* Kumner, *Paxillus atrotomentosus (Fr.)* Fr., were contributed by investigators throughout the United States and Canada.

The technique recommended by Hacskaylo (4) and modified by Marx and Zak (5) was used for synthetic culture of mycorrhizae. Half-gallon Mason jars were used as culture chambers. A substrate of 600 ml. of vermiculite mixed with 120 ml. of loosely packed peat moss was added to the jars and saturated with 450 ml. of the nutrient solution (adjusted to pH 4) prescribed for synthetic culture by Melin (6) and Norkrans (7). Jars were plugged with cotton balls reinforced with cheesecloth and immediately autoclaved for 30 minutes at 250 degrees and 15 lbs. pressure.

Sterile pine seedlings were obtained by using seed surface sterilized with 30 per cent hydrogen peroxide for 1 hour, planted on agar in petri dishes, and allowed to germinate. For each test, one sterile seedling was planted in the center of each of five culture jars. At the same time, jars were inoculated with blocks of agar cultured with suspected mycorrhizal fungi. Seedlings were then grown either on light tables or in a greenhouse waterbath. Two to 4 months following inoculation all seedlings were removed and examined under a binocular scope for the presence or absence of mycorrhizae.

Results

Fifty-two species of basidiomycetes were collected and identified from the six stands surveyed. Five of these, *Suillus* brevipes (Pk.) Kuntze, *Suillus luteus (Fr.)* S. F. Gray *Suillus granulatus (Fr.)* Kuntze, *Laccaria laccata (Fr.)* Berk. & Br., and *Pisolithius tinctorius* (Pers.) Coker & Couch, have previously been shown to form mycorrhizae with loblolly pine. In addition, mycorrhizae of the type formed by *Cenococcum graniforme* (Sow.) Ferd & Wings were observed in two stands.

Of the collected species for which pure cultures were available, *Agaricus placomyces, Amanita parcivolvata, Amanita* virosa, and *Boletus bicolor* formed mycorrhizae with loblolly pine in sterile culture.

Amanita citrina, Amanita parcivolvata, Paxillus atrotomentosus, and *Russula fragilis* produced mycorrhizaelike structures. All were branched dichotomously and hypertrophied, while root hairs were suppressed. The structures, however, lacked the fungal sheath characteristic of fully developed mycorrhizae.

Clitocybe gibba, Collybia maculate, Lactarius piperatus, and *Leucopaxillus laterarius* did not form mycorrhizae with lololly pine.

It has been stressed that the results of synthetic culture are not conclusive *(8, 9, 10).* The fact that a fungus enters the mycorrhizal relationship under artificial conditions of synthetic culture indicates only that the fungus has the ability to do so. Whether it does under field conditions is till open to question (8). Negative results in synthetic culture are especially questionable. Subsquent testing has shown several fungi to be mycorrhizal even though first reported as non-mycorrhizal (1, 2).