

Rejuvenation of Loblolly Pine Vegetative Buds  
by  
Shoot-tip Micrografting Techniques.

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Successful vegetative propagation of some mature conifer species depends on the ability to rejuvenate mature tissue and induce it to form juvenile tissue. One treatment often used for the induction of juvenility involves grafting of mature buds onto young rootstocks.

Micrografting techniques were tested as a means of rejuvenating meristematic tissue of 8-year-old loblolly pine (*Pinus taeda* L.) vegetative buds. In the main experiments, apical scions (the apical dome plus some sub-apex tissue, measuring 0.1-0.3 mm) were grafted onto 4-, 8-, 12-, and 16-week-old loblolly pine and slash pine (*P. elliottii* Engelm. var. *elliottii*) seedlings. They were grown singly in tubes under incandescent, fluorescent and "gro-sho" lights, creating individual greenhouse environments. Cleft, veneer and budding graft cuts were employed on foliated and defoliated seedlings. Both the epicotyl and hypocotyl regions served as graft sites. Seedlings were grafted in fall, winter, spring, and summer. In supplemental experiments, three chemicals (sodiumdiethyldithiocarbamate, polyvinylpyrrolidone, and benzoadeninepurine, at two concentrations each) and activated charcoal were applied to graft sites in attempt to improve grafting success and to induce juvenility. Rootstocks grafted in the fall were maintained in the light following grafting. Rootstocks grafted in subsequent experiments were maintained in the dark for five days following grafting, then placed in the light.

To assess anticipated successful rejuvenation, primary and mature loblolly pine needles were cultured *in vitro* on modified basal media (DCR). Primary needles showed abundant callusing, mature needles died. Additionally, apical scions from mature buds were grafted on primary needle callus to determine meristem tissue viability and activity. All scions responded by either callusing or by swelling, elongating and turning a bright green. The apical scions were therefore viable and capable of callusing and growing at the time of grafting.

There were no successful graft takes with 962 attempts. Although rootstocks developed healthy callus at the graft site, the scions did not. Scions became necrotic and died within 3-15 days, depending on treatment. Histological examination of the graft union tissue revealed a barrier of necrotic cells between the rootstock and scion, resin deposition, no vascular connections, nor cambial joining between the graft components.

It would appear that the micrografting methods employed in this study did not provide favorable circumstances for apical scions to fuse with rootstocks, or conversely. In summary, micrografting may not be a viable alternative in attempting rejuvenation and propagation of mature loblolly pine vegetative buds.