

A TISSUE CULTURE SYSTEM FOR STUDYING  
DISEASE RESISTANCE TO PHYTOPHTHORA CINNAMOMI

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Calli derived from embryos of Pinus taeda, P. echinata, P. taeda x P. echinata, and P. virginiana were screened for their resistance to Phytophthora cinnamomi Rands, the littleleaf disease pathogen. Approximately 50 clones of calli from each of at least two different seed sources of each pine species were selected, except for P. virginiana which only had one seed source. The optimum growth condition was established by testing the interactions of calli at different temperatures with growth regulators. A similar condition was determined for hyphae of P. cinnamomi. Results showed that a culture medium with  $10^{-5}$  M 2,4-D at 22°C gave best growth of calli and caused the least synergistic reaction between fungus and callus tissue. Three methods were used for evaluating the in vitro resistance reaction: (1) diameter growth of P. cinnamomi on the callus surface; (2) amount of intracellular hyphae and cytological change of infected callus cells; and (3) surface reaction of inoculated calli. Loblolly pine was demonstrated to be the most resistant species and shortleaf pine the most susceptible. However, susceptible clones were also found in loblolly pine and loblolly x shortleaf pine hybrids and resistant clones were found in shortleaf pine. Resistance reactions were always inversely correlated with a greater growth rate of P. cinnamomi on callus, a larger number of intracellular hyphae, and necrosis and accumulation of phenolic compounds in callus cells.