DEVELOPMENT OF IN VITRO AND IN VIVO SCREENING TECHNIQUES TO DETECT LITTLELEAF DISEASE RESISTANCE

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Two-month-old sand cultured pine seedlings were inoculated with zoospores and chlamydospores of Phytophthora cinnamomi Rands to determine in vivo susceptibility. Seedlings from sand culture were harvested and gently washed with tap water to remove sand particles. Root tips of the intact seedlings were then inoculated with differing dosages of zoospores or chlamydospores of P. cinnamomi in a moist chamber by using a micropipette inoculation technique. Two, five, and ten spores per microliter were used as inoculum dosage treatments.

Susceptibility increased with increasing dosage of both zoospores and chlamydospores for all the species tested. In order from most to least susceptible at the 5 spores/pl dosage were Pinus echinata, P. virginiana, P. taeda, and finally the P. taeda x P. echinata hybrid. Infection rates from chlamydospore inoculation were consistently higher than for zoospore inoculation in all species, indicating that the former have greater inoculum potential.

In vitro inoculation was done by using the same method as described above except that the plantlets were derived from embryonic cotyledons through organogenesis in a tissue culture system. Advantages of the latter include: 1) the elimination of interaction with other microorganisms; and 2) large quantities of genetically identical cloned plantlets were available for inovulation with different inocula and dosages. Preliminary results indicate that P. echinata is more susceptible than P. taeda and the infection rate of root tips was increased with increasing inoculum concentration for all the species tested.

In general, the in vitro inoculation results correlated well with the in vivo results, indicating that this screening method may be suitable for incorporation into a traditional breeding program for littleleaf disease resistance.