Agrobaterium Tumefaciens - Mediated Transformation And Protoplast Technologies For Loblolly Pine (*Pinus Taeda* L.)

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Genetic engineering is a useful method to compliment traditional breeding approaches to improve growth, wood and fiber properties in woody plant species. With genetic engineering, the transfer of novel genes can confer unique traits, such as herbicide tolerance, that are difficult or impossible to breed into woody plants. Genetic engineering is also important for scientists to dissect the functions of genes to significantly increase our understanding of forest tree genetics.

A genetic transformation procedure for loblolly pine (*Pinus taeda* L.) is being developed. Early stage loblolly pine somatic embryos are co-cultured for 2 days in the dark at 25°C with *Agrobaterium tumefaciens* containing the 2X35S promoter NPTII gene for selection. Transformed early stage embryos are selected on solid media containing geneticin for two months. Putatively transformed lines carrying the *nptII* gene were isolated and are being put through the maturation process.

We have also isolated protoplasts in high yields ($2.5.10^5$/g fw) from loblolly pine early stage somatic embryo cultures. Protoplast viability averaged 75.5%. The first divisions occurred after 7-9 days of culture, and we are currently following their growth. Electroporation readily transforms protoplasts from many plants. Electroporation conditions were optimized for the Douglas fir and loblolly pine protoplasts by monitoring the uptake of Calcein (3,3'-Bis[N,N'-bis(carboxymethyl)aminomethyl]fluorescein).