Transgenic Loblolly Pine Trees from Diverse Elite Families

Heather J. Gladfelter, J. Eric Gulledge, Sally Schwuchow, Ryan R. McCormack, Ron T. Kothera, Mike R. Becwar, Bob J. Kodrzycki, H. Dayton Wilde, and Marie B. Connett[1]

Abstract

Loblolly pine (*Pinus taeda* L.) has been the focus of genetic improvement for nearly 100 years because of the value of this species to the forestry industry. The application of gene transfer technology to loblolly pine improvement has been limited by the regeneration of transgenic tissue into plants. We have developed gene transfer systems that allow the regeneration of trees after the transformation of embryogenic cultures from a large number of genetically diverse families.

Genetic transformation was achieved by biolistic and *Agrobacterium*-mediated techniques. Biolistic transformation efficiency was increased by identifying the optimal target using secondary somatic embryogenesis and by determining the long-term effects of tissue culture manipulations. Improvements to selection and the tissue culture system facilitated the production of stable transformants from 72% of the cell lines attempted from 15 elite families, with an escape rate of less than 1%. Molecular analysis of transgenic trees produced from biolistic transformation found that 36% of the trees had three inserts or less. Transgenic trees produced by biolistics have exhibited normal morphology for up to five growing seasons, to date.

An *Agrobacterium*-mediated transformation system was developed for loblolly pine using tissue culture and selection procedures of the biolistic system. *Agrobacterium tumefaciens* has been used to produce transgenic trees of clones from elite loblolly families, as well as clones of *P. radiata* and *P. taeda* x *rigida*. Genomic blot analysis of *Agrobacterium*-transformed somatic embryos is ongoing. Field tests with *Agrobacterium*-transformed loblolly and the hybrid loblolly have been established each year since 2001. The efficiency of the *Agrobacterium* transformation system has made it possible for ArborGen to scale-up for high-throughput gene testing in a conifer. Transgenic trees have been produced with genes for lignin modification, accelerated growth, and flowering control.

---

[1] ArborGen, LLC, Summerville, SC 29484-8401.