

TRANSFORMATION OF *LIQUIDAMBER STYRACIFLUA* L. (SWEETGUM) USING
AGROBACTERIUM TUMEFACIENS

Z.-Z. Chen and A.-M. Stomp
Forestry Dept., North Carolina State University
Raleigh, NC

We have produced the first transgenic sweetgum plants using *Agrobacterium*-mediated DNA transfer. Transformation was accomplished by co-cultivation of leaf pieces or nodules with a disarmed binary *Agrobacterium tumefaciens* (pBI121 in C58z707). The C58 background was used because wild type C58 gave good gall formation on sweetgum seedlings. The mini-plasmid (pBI121) contains the selectable marker, neomycin phosphotransferase (NPT II) under the NOS promoter and the unselectable marker, beta-glucuronidase (GUS) under the CaMV35s promoter. Co-cultivated tissue was transferred to 500 mg/L, each, of carbenecillin and cefotaxime to decontaminate the tissue and 40 mg/L kanamycin sulfate to select transformed cells. Tissue was carried under selection for 6 months, subcultured every 2 weeks, in liquid medium to proliferate nodules and segregate a transgenic nodule population. Cultures were checked periodically for GUS expression during selection. After cultures grew freely without carbenecillin and cefotaxime, indicating decontamination of the tissue, shoot formation was induced on agar medium containing kanamycin following the procedures developed for nodule cultures of sweetgum (Chen & Stomp, in accompanying abstract). Individual shoots were excised from shoot producing nodule masses and rooted on medium containing kanamycin. Transformation has been verified using an ELISA test for the presence of NPT II, positive GUS staining and quantitative assay and analysis of DNA isolated from GUS positive plants by Southern hybridization. A number of transgenic nodular callus lines and transgenic plants have been produced using this method. These procedures provide a routine method for the production of transgenic sweetgum plants or cell cultures.