

ISOLATION AND CHARACTERIZATION OF TOBACCO CYTOCHROME P450 cDNAS INVOLVED IN HERBICIDE METABOLISM. Lily B. Gavilano and Balazs Siminszky, Post-Doctoral Research Associate and Assistant Professor, University of Kentucky, Agronomy Dept., Lexington, KY 40546.

Cytochrome P450 monooxygenases catalyze the metabolism of a large array of herbicides. While overexpression of P450 genes isolated from corn, arabidopsis and soybean have been shown to confer resistance to herbicides, very few P450 genes involved in herbicide metabolism have been isolated from tobacco. In addition to improving our understanding about the molecular biology of P450-mediated herbicide metabolism, the isolation and characterization of herbicide metabolizing P450s and their use for engineering herbicide-resistant tobacco may ultimately provide more weed management options in direct-seeded tobacco. The objective of this project is to isolate and characterize P450 genes that are involved in herbicide metabolism. We have identified 43 unique P450 sequences from a large tobacco EST database and used a modified 5' RACE PCR strategy for isolating the full-length version of 15 randomly selected P450 cDNAs. The full-length cDNAs were subcloned into a plant expression vector and the resulting construct was introduced into tobacco via *Agrobacterium*-mediated plant transformation. P450 transcript accumulation was measured by Northern blot analysis and seed were harvested from individuals exhibiting the highest level of mRNA expression. Experiments are underway to determine the degree of herbicide tolerance of each transgenic line against selected herbicides.