

Plant Gene Register

Nucleotide Sequence of the Tobacco (*Nicotiana tabacum*) Anionic Peroxidase Gene¹

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Peroxidases have been implicated in numerous physiological processes including lignification (Grisebach, 1981), wound-healing (Espelie et al., 1986), phenol oxidation (Lagrimini, 1991), pathogen defense (Ye et al., 1990), and the regulation of cell elongation through the formation of inter-chain covalent bonds between various cell wall polymers (Fry, 1986; Goldberg et al., 1986; Bradley et al., 1992). However, a complete description of peroxidase action in vivo is not available because of the vast number of potential substrates and the existence of multiple isoenzymes. The tobacco anionic peroxidase is one of the better-characterized isoenzymes. This enzyme has been shown to oxidize a number of significant plant secondary compounds in vitro including cinnamyl alcohols, phenolic acids, and indole-3-acetic acid (Mäder, 1980; Lagrimini, 1991). A cDNA encoding the enzyme has been obtained, and this enzyme was shown to be expressed at the highest levels in lignifying tissues (xylem and tracheary elements) and also in epidermal tissue (Lagrimini et al., 1987). It was shown at this time that there were four distinct copies of the anionic peroxidase gene in tobacco (*Nicotiana tabacum*). A tobacco genomic DNA library was constructed in the λ -phage EMBL3, from which two unique peroxidase genes were sequenced. One of these clones, λ POD1, was designated as a pseudogene when the exonic sequences were found to differ from the cDNA sequence by 1%, and several frame shifts in the coding sequences indicated a dysfunctional gene (our unpublished results). The other clone, λ POD3, described in this manuscript (Table I), was designated as the functional tobacco anionic peroxidase gene because of 100% homology with the cDNA. Significant structural elements include an AS-2 box indicated in shoot-specific expression (Lam and Chua, 1989), a TATA box, and two intervening sequences.

Table I. Characteristics of the tobacco anionic peroxidase gene

Organism:	<i>Nicotiana tabacum</i> L. (tobacco) cv Coker 176.
Location on Chromosome:	Nuclear genome, chromosome location not known.
Function:	Gene encodes an anionic peroxidase isoenzyme (donor: hydrogen-peroxide oxidoreductase, EC 1.11.1.7).
Clone Type:	Genomic.
Sources:	Genomic library in λ EMBL 3, constructed from DNA isolated from leaf tissue.
Techniques:	Various restriction fragments subcloned into Bluescript (Stratagene), complete supercoil dideoxy sequencing of both strands using synthetic oligonucleotide primers.
Method of Identification:	Nucleotide sequence comparison of exons with the cDNA clone for peroxidase (Lagrimini et al., 1987).
Expression Characteristics:	Anionic peroxidase is highly expressed in xylem-forming tissues, where it is involved in lignin synthesis (Lagrimini, 1991). Also, it is expressed to a lesser extent in other tissues, including epidermal tissue, where it has a possible role in cell elongation (Bradley et al., 1992).
Regulation:	Unknown.
Structural Features of Gene:	AS-2 box (Lam and Chua, 1989), TATA box, transcriptional start site, translational start site, translational stop codon, poly(A) addition site, and two intervening sequences.
Subcellular Localization of the Gene Product:	Cell wall.

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