

Plant Gene Register

Nucleotide Sequence of a *Populus tremuloides* Gene Encoding Bispecific Caffeic Acid/5-Hydroxyferulic Acid *O*-Methyltransferase¹

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Bi-specific OMT (EC 2.1.1.68) catalyzes the meta-specific methylation of caffeic acid and 5-hydroxyferulic acid to ferulic acid and sinapic acid, respectively (Higuchi, 1990). It is one of the key enzymes in angiosperm dicots mediating the formation of unique precursors for syringyl lignin, which is generally absent in gymnosperms (Higuchi, 1990).

OMT has been purified and its cDNA clones isolated from *Populus tremuloides* (Bugos et al., 1991, 1992), *Populus deltoides* X *Populus trichocarpa* (Dumas et al., 1992), and several other plants (Gowri et al., 1991; Callazo et al., 1992; Jaeck et al., 1992; Pellegrini et al., 1993). In this paper we report the nucleotide sequence encoding an OMT genomic clone, designated PTOMT, obtained from an angiosperm tree species. Two oligonucleotide primers corresponding to nucleotide positions 65 to 82 and to positions 1370 to 1388 of a full-length OMT cDNA (Ptomt1) from secondary developing xylem tissue of *P. tremuloides* (Bugos et al., 1991) were used for PCR using *P. tremuloides* total cellular DNA as a template. A 2.7-kb DNA fragment was amplified, subcloned, and sequenced in its entirety in both directions.

The *P. tremuloides* POMT gene contains four exons and three introns (Table I), downstream from the translation start codon. The amino acid sequence derived from this *P. tremuloides* genomic DNA is 99.7% homologous with that from Ptomt1 (Bugos et al., 1991).

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The GenBank accession number for the sequence reported in this article is U13171.

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Abbreviation: OMT, *O*-methyltransferase.

Table I. Characteristics of the OMT gene from *P. tremuloides*

Organism:

Populus tremuloides Michx.

Function, Pathway:

Encodes bi-specific OMT (EC 2.1.1.68), lignin biosynthesis.

Cloning Techniques:

Amplification of total nuclear DNA by PCR was carried out using *Taq* DNA polymerase and primers derived from the OMT cDNA (Ptomt1) sequence (Bugos et al., 1991). The primers used were 5'-GCTCTAGAGCATGGGTTCAACAGGTGAA (upstream primer) and 5'-GTTGGAAGCTTAAGCCAATAGG (downstream primer). These primers also include *Xba*I and *Hind*III restriction sites (underlined) at their 5' ends, respectively, to facilitate cloning into a plasmid vector. A 2.7-kb amplification product was cloned into pNoTA (5 Prime-3 Prime, Inc., West Chester, PA) and sequenced for both strands using the Sequenase system (United States Biochemical).

Sequence Identification:

DNA and deduced amino acid sequences were compared with nucleotide and deduced amino acid sequences of *P. tremuloides* cDNA (Bugos et al., 1991).

Features of Gene Structure:

Four exons (419, 311, 65, and 300 bp) and three introns (1102, 130, and 150 bp) are included in the amino acid coding region.

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