

Plant Gene Register

Nucleotide Sequence of a New cDNA for Peroxidase from *Arabidopsis thaliana*¹

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Peroxidases (EC.1.11.1.7) play an integral role in secondary cell wall biosynthesis by catalyzing the polymerization of cinnamyl alcohol into lignin and by forming rigid cross-links between cellulose, pectin, Hyp-rich glycoproteins, and lignin (Fry, 1986). They are very suitable as a defense marker because these enzymes oxidize several organic compounds (hydrogen donors) in the presence of H₂O₂ and are induced by wounding and fungal infection (Svalheim and Robertson, 1990; Graham and Graham, 1991). These various functions are consistent with the occurrence of multiple peroxidases, suggesting that different peroxidase isozymes are involved in different processes in specific parts of plants. Cloning of ARP gene could help in explaining the physiological role that each of the isozymes plays in plant cell wall biosynthesis and defense response. We have cloned two peroxidase isozyme genes of *Arabidopsis thaliana*, *prxCa* and *prxEa* (Intapruk et al., 1991, 1993). Here we present the nucleotide and deduced amino acid sequences of a new ARP cDNA, namely *prxCb*.

Using cDNA of *prxCa*, we screened a λ gt11 cDNA library of *A. thaliana*. Two positive clones containing partial cDNAs were isolated and sequenced. From DNA sequence analysis, the two clones differ by a 64-bp deletion at the 3' poly(A). Both clones have an open reading frame of 1059 bp encoding a polypeptide of 353 amino acid residues (*M*, 38,950) beginning with an ATG start codon at position 35 and ending with a TGA stop codon at position 1094. A putative poly(A) signal, AATAAA, was observed 34 bp upstream of the poly(A) sequence. The characteristics of the *prxCb* are summarized in Table I.

The amino acid sequences of nine peroxidases from Brassicaceae family, *A. thaliana* Ca, Ea (Intapruk et al., 1993); horseradish A2 (Welinder, 1992b), Cla, Clb (Fujiyama et al., 1988), C2, C3 (Fujiyama et al., 1990), E5 (Welinder, 1992a), and n (Bartonek-Roxa et al., 1991), were compared with the new ARP Cb, and the mature sequences show identities ranging from 51 to 93.5%. The alignment also shows that residues 34 to 56 of the ARP Cb, including the distal catalytic Arg³⁸ and His⁴², and residues 152 to 177 around proximal

Table I. Characteristics of a peroxidase cDNA from *A. thaliana*

Organism:	<i>Arabidopsis thaliana</i> (L.) Heynh Columbia ecotype.
Location of Gene:	Nuclear enclosed.
Gene Product:	Donor: H ₂ O ₂ oxidoreductase, a group of heme-containing enzymes.
Techniques:	Isolation of cDNA clones from λ gt11, subcloning into pBluescript SK-, and sequenced by dideoxy sequencing on both strands.
Clone Type:	cDNA full length, <i>prxCb</i> .
Method of Identification:	Sequence comparison to EMBL/Genbank data base; sequence identity to superfamily plant peroxidases (Welinder, 1992b).
Features of cDNA Structure:	1236 bp including 5' and 3' untranslated regions and poly(A) tail. Putative poly(A) signals are AATAAA.
Features of Protein:	Open reading frame of 1059 bp encoding a polypeptide of 353 amino acid residues of <i>M</i> , 38,950. Proximal and distal His residues bound to protoheme at 42 and 170 amino acid residues, respectively.
Regulation:	Unknown.
Codon Usage:	40.9% G + C and 24.8% A in the third position.
G/C Content:	45.0% in coding regions.
Antibodies:	Not available.

His¹⁷⁰ are highly conserved and characteristic for plant peroxidases (Urrutigoity et al., 1991). The predicted isoelectric point value of the mature ARP Cb polypeptide (excluding heme and calcium) is 8.4, a little more basic than that of ARP Ca, which is 8.1.

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The EMBL accession number for the sequence described in this article is X71794.

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Abbreviation: ARP, *Arabidopsis* peroxidase.

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