

Plant Gene Register

Isolation and Characterization of a cDNA for Spinach Cytosolic Ascorbate Peroxidase¹

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Reduced oxygen intermediates are produced in plant tissues from mitochondrial and chloroplastic electron transport pathways and from electron-saturated photosynthetic pigments (Bowler et al., 1992). For instance, the Mehler reaction is a primary source of reduced oxygen intermediates in chloroplasts in which dioxygen is reduced by Fd on the reducing side of PSI to produce $\cdot\text{O}_2^-$. The dismutation of $\cdot\text{O}_2^-$ to H_2O_2 occurs spontaneously or is catalyzed by superoxide dismutases (Neubauer and Yamamoto, 1992). Efficient scavenging of H_2O_2 is critical, since it can damage membranes and deactivate enzymes. In addition, H_2O_2 can react with $\cdot\text{O}_2^-$ to produce the highly reactive and dangerous hydroxyl radical ($\cdot\text{OH}$). The reduction of H_2O_2 by ascorbate is catalyzed by APX (EC 1.11.1.11). This enzyme is considered to be an indispensable part of the electron-scavenging pathway (Nakano and Asada, 1987). APX is a heme-containing homodimer found in all higher plants (Asada, 1992). Plant APXs include soluble cytosolic isoforms (Mittler and Zilinskas, 1991) and chloroplast-localized isoforms that may be stromal or thylakoid bound (Miyake et al., 1993).

We report here the isolation of a full-length cDNA from spinach (*Spinacia oleracea*) that encodes a cytosolic APX subunit (Table I). The cDNA is 1102 bp in length with a 750-bp open reading frame, along with 46 bp of 5' and 306 bp of 3' untranslated sequences. The nucleotide sequence of the spinach cytosolic APX open reading frame shows 74% identity with the open reading frame of a cytosolic APX cDNA from pea and 72% identity with one from Arabidopsis. The derived amino acid sequence identity among spinach, pea, and Arabidopsis is 83%.

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Abbreviation: APX, ascorbate peroxidase.

Table I. Characteristics of cDNA clone pSOAPX

Organism:	<i>Spinacia oleracea</i> immature leaves.
Techniques:	A full-length pea cDNA library was used to screen for homologous clones from a λ -ZAP spinach leaf library (Clontech, Palo Alto, CA). The longest cDNA was sequenced by dideoxy chain termination.
Features of the cDNA:	Contains an open reading frame with putative translation start codon at nucleotide 46 and stop codon at nucleotide 796. Putative polyadenylation signal at nucleotide 1045.
Features of the Derived Polypeptide:	250 amino acid residues, estimated subunit pI of 5.26, and estimated subunit molecular weight of 27,625.
Expression:	High levels of cytosolic APX were seen in immature leaves and shoots.

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The EMBL accession number for the sequence reported in this article is L20864.

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