

## Plant Gene Register

***lox1:Ps:2*, a *Pisum sativum* Seed Lipoxygenase Gene<sup>1</sup>**

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All plants contain LOXs (EC 1.13.11.12), which catalyze the hydroperoxidation of polyunsaturated fatty acids. In vegetative organs, plant LOX genes respond to drought, wounding, pathogen attack, and exposure to jasmonate (for refs. see Melan et al., 1994); metabolism of fatty acid hydroperoxides produced by LOXs can give rise to the plant growth regulators ABA and methyl jasmonate. In soybean seeds, short-chain carbonyl compounds, derived from LOX-produced fatty acid hydroperoxides, are responsible for off-flavors.

Plant LOX genes reported in the literature include those for vegetative LOX from *Arabidopsis thaliana* (Melan et al., 1994), *Phaseolus vulgaris* (Eiben and Slusarenko, 1994), and *Glycine max* (Kato et al., 1993) and for seed LOX (LOX-3, Yenofsky et al., 1988; SC514, Shibata et al., 1991) from *G. max*. We describe here a *Pisum sativum* LOX gene that corresponds to a seed mRNA that encodes a polypeptide similar in sequence to LOX-2 from soybean seed. We have named the gene *lox1:Ps:2*, in consultation with the International Society for Plant Molecular Biology Commission on Plant Gene Nomenclature.

The 5785 bp of *lox1:Ps:2* described in Table I include 1366 bp 5' to the translation initiation codon and 884 bp 3' to the stop codon. Apart from three nucleotide substitutions and one deletion of three nucleotides, the protein coding sequence is identical to that of the pea seed LOX-2 cDNA pPE320 (Ealing and Casey, 1989). Eight introns are in the same positions, relative to the coding sequence of *lox1:Ps:2*, as in the soybean LOX-3 gene (Yenofsky et al., 1988). The first intron is relatively large (314 bp, compared to 77–115 bp for the other seven); first introns in the *Arabidopsis LOX1*, *P. vulgaris Lox1*, and *G. max LOX-3*, L4, and SC514 genes are also large, comprising 1242, 1285, 448, 2542, and 624 bp, respectively. The protein sequence predicted by *lox1:Ps:2* contains the His, Asn, and Ile residues that are liganded to iron at the active site (see, e.g., Boyington et al., 1993). The *lox1:Ps:2* gene has extensive, overlapping tandem repeats between 600 bp and 1.3 kb upstream of the transcription start site (position 1340). There are also several motifs that correspond to known protein-binding sites and several sequences, including AACAAA, an "RY" repeat, and a perfect decanucleotide palindrome, that are thought to be important to the regulation of gene expression in developing seeds.

**Table I.** Characteristics of the *lox1:Ps:2* gene from pea

Organism:	Pea ( <i>Pisum sativum</i> L. cv Birte).
Function:	Lipoxygenase; fatty acid hydroperoxidation.
Techniques:	Clone $\lambda$ GEM12-CF-2 was isolated from a SauIII library of genomic DNA by plaque hybridization with the central EcoRI fragment from the insert of the pea seed LOX cDNA clone pPE923 described by Ealing and Casey (1989). The sequence of <i>lox1:Ps:2</i> within $\lambda$ GEM12-CF-2 (insert size approximately 12 kb) was determined by exonuclease III deletion/primer walking/dideoxy sequencing of EcoRI fragments cloned into Bluescript KS* using an automated (Pharmacia Automated Laser Fluorescent) DNA sequencer and by manual cycle sequencing of phage DNA using <sup>33</sup> P-labeled oligonucleotides and Taq polymerase. Both strands were sequenced at least twice.
Sequence Identification:	Comparison with cDNA sequence.
Expression and Regulation:	The mRNA corresponding to <i>lox1:Ps:2</i> is produced in a temporally regulated fashion during pea seed development (Domoney et al., 1990).
Chromosomal Location:	Pea seed LOX genes map to linkage group IV, close to <i>Np</i> and <i>le</i> (North et al., 1989; Domoney et al., 1991).
Features of the Predicted Amino Acid Sequence:	Differs from that predicted by cDNA pPE320 in three places: Leu <sup>333</sup> → Ile; Met <sup>561</sup> deleted; Leu <sup>578</sup> → Ile (numbers refer to the amino acid sequence predicted from pPE320).
Antibodies:	The anti-LOX antibody, "anti-B" (Domoney et al., 1990), recognizes the LOX-2-type polypeptide in pea seeds.

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The EMBL/GenBank accession number for the sequence reported in this article is X78580.

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Abbreviation: LOX, lipoxygenase.

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