

Update section

Sequence

Nucleotide sequence of a cDNA encoding a lipid transfer protein from wheat (*Triticum durum* Desf.)

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Lipid transfer proteins (LTPs) have been isolated from bacteria, yeasts, animals and plants and are characterized by their *in vitro* ability to catalyse the transfer of lipids between membranes (for a review on plant LTPs, see [1]). Three proteins which increased the *in vitro* phosphatidylcholine transfer between liposomes and mitochondria were identified in *Triticum durum* semolina [5]. One of the purified proteins has a molecular mass of 7 kDa, the molecular mass of the two others being 9 kDa.

A cDNA clone (pTd4.90) encoding a *T. durum* LTP was isolated, from a mid-maturation (22 days after flowering (DAF) seeds) cDNA library [3], using the *Sty I/Not I* fragment of the cDNA clone 9C2 encoding a maize 9 kDa LTP [9] as a probe. Figure 1 shows the nucleotide sequence of the 615 bp cDNA and the deduced amino acid sequence of the wheat LTP. The 615 bp sequence contains one open reading frame of 339 bp and a 274 bp 3' untranslated sequence. There are three putative sites of translation initiation, at position 12, 18 or 39, but neither of them are in agreement with the Joshi's rule [4] indicating that

probably this clone is not full-length. The coding sequence is particularly rich in G + C (64%). At position 460, there is one putative polyadenylation signal (AATAAG) 135 bp upstream from the polyadenylation site.

The open reading frame encodes a protein of 113 residues, of which residues 24–113 correspond to a wheat LTP. The N-terminus of the predicted mature protein is based on the N-terminal amino acid sequence (38 first residues) of the purified wheat 9 kDa LTP. The wheat mature LTP contains 90 amino acids and has a calculated molecular mass of 9608 Da. All the previously reported plant LTPs are synthesized as precursors. But, according to alignment with known plant LTP sequences, and especially with the barley sequence [6], the signal peptide deduced from clone pTd4.90 is presumably truncated of the first three amino acids.

The N-terminal sequence of the wheat purified 9 kDa LTP shows four different amino acids with residues 24–61 of the LTP deduced sequence (Fig. 1). This is in agreement with the presence of LTP isoforms as suggested by the purification of

The nucleotide sequence data reported will appear in the EMBL, GenBank and DDBJ Nucleotide Sequence Databases under the accession number X63669.

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GCGCTCAGGTAATGCTCATGGCCGTCGCCTTGGTGCTCATGCTCGCCGCGGTCCCGCGC 59
A Q V M L M A V A L V L M L A A V P R 19
GCTGCCGTGGCCATCGACTGCGGCCACGTTGACAGCTTGGTGAGACCC*TCCTGAGCTAC 119
A A V A I D C G H V D S L V R P C L S Y 39
GTTCAGGGCGGCCCCGGCCCGTCTGGGCAGTGCTGCGACGGCGTCAAGAACC*TCCTAAC 179
V Q G G P G P S G Q C C D G V K N L H N 59
CAGGCGCGATCCCAGAGCGATCGCCAAAGCGCTTGCAACTGCCTCAAGGGGATCGCTCGT 239
Q A R S Q S D R Q S A C N C L K G I A R 79
GGCATCCACAATCTCAACGAGGACAACGCCCGCAGCATCCCCCAAGTGCGGTGTCAAC 299
G I H N L N E D N A R S I P P K C G V N 99
CTCCCATACACCATCAGTCTCAACATCGACTGCAGCAGGGTGTAATGGGCGACGATCCGT 359
L P Y T I S L N I D C S R V • 113
CAAGCTGGTGCTCAGCTCATCCATCCACGTGGAGTTGAAGCGCGCAGCCTCTATCCCTAT 419
GTAGTATGGTCACTAGTTATGCGAGTTTATACTGAATATGAATAAGAACTCTCTCCAGCT 479
.....
GGCTTGCTGGTACTCCTCTGGAGGAGATCAGTATCTGTGTACCTGAGAGTTGAGAGTTTG 539
TACCATGGGCACTCCAGTGTTTATGGACTTTAATACATACAACCTCGTTCTGTTCAAAAA 599
AAAAAAAAAAAAAAAAA 615

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Fig. 1. Nucleotide sequence of the pTd4.90 cDNA and deduced amino acid sequence of a 9 kDa LTP. The signal peptide is underlined. Double underlined amino acids indicate differences with the N-terminal sequence of the purified 9 kDa LTP [5]: G and H are replaced with R and N respectively, D is missing and there is an insertion (I) denoted by $\uparrow$. Cysteine residues are marked with * and the putative polyadenylation signal is marked with dots.

two wheat 9 kDa proteins [5]. But, these differences could also be due to the fact that the *T. durum* cultivars used for the cDNA library construction and the LTP purification were different. On the contrary, there is a total conservation of the primary structure of the *T. durum* 9 kDa LTP deduced from clone pTd4.90 and a 9 kDa LTP isolated from *Triticum aestivum* seeds [2].

The wheat LTPs contain 8 cysteine residues including a doublet Cys-Cys and the sequence Cys-Asn-Cys which are highly conserved in plant LTPs. The sequence homologies between the *T. durum* 9 kDa LTP and other plant LTPs range from 28 to 70%. The lowest homologies (28–38%) are observed with the ricin LTPs [7, 8] and the highest homology (70%) with the barley LTP [6].

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