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THE SECRETORY PHOSPHOLIPASE A₂ (sPLA₂) GENE FAMILY IN DURUM WHEAT: IDENTIFICATION, CHARACTERIZATION AND EVIDENCE FOR A ROLE IN ADAPTATION TO DROUGHT STRESS

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To survive adverse stresses plants have developed complex signalling networks to perceive environmental stimuli and transduce the information over the plasma membrane into the cell where it activates specific signalling cascade. In this context, the production of lipid mediators triggered by phospholipases, throughout the generation of membrane phospholipid-derived second messengers, plays a pivotal role in plant response to environmental stresses. PLA₂ specifically hydrolyses phospholipids at the *sn*-2 position to yield free fatty acids (FFAs) and lysophospholipids, which are known to act as signalling molecules in a wide range of physiological and pathological processes. Currently, the PLA₂ superfamily consists of five different classes: the secretory PLA₂s (sPLA₂), the cytosolic Ca²⁺-dependent PLA₂s (cPLA₂), the cytosolic Ca²⁺independent PLA₂s (iPLA₂), the platelet activating factor acylhydrolases and the lysosomal PLA₂s. In plants, to the best of our knowledge, to this day, the only specific PLA₂s discovered belong to the class of secretory PLA₂s.

In the light of this, an investigation was carried out to identify and characterize the sPLA₂ gene family in durum wheat and to evaluate its involvement in durum wheat response to drought stress. On the basis of sequence homology with the four sPLA₂ genes of rice ($OssPLA_2I$: Os02g0831700, $OssPLA_2II$: Os03g0261100, $OssPLA_2III$: Os03g0708000, $OssPLA_2IV$: Os11g0546600) the full-length sequences corresponding to the four durum wheat sPLA₂s genes and to their corresponding transcripts were isolated and characterised. The genomic structure of the four sPLA₂ genes was determined by comparisons between the gene sequences and the corresponding expressed sequences. The isoforms I comprised 3 exons and 2 introns, while the other three isoforms comprised 4 exons and 3 introns. While the length of the exons was highly conserved among the sPLA₂ genes, the introns showed a high degree of variability. The four expressed sequences were identical to the coding sequences deduced from the corresponding sPLA₂ genes, thus demonstrating that they are all actively transcribed and potentially encode functional sPLA₂s isoforms as they contain all the domains typical of the plant sPLA₂s.

In leaves two genes encoding the isorform I and III were found to be up-regulated by water deficit, while the other two were found to be constitutively expressed. Consistently, as a consequence of the stress imposition, an increase was also observed in a Ca^{2+} -dependent PLA₂ activity whose biochemical characteristics resemble those of other well known plant sPLA₂s. In line with these findings, the analysis of the FFA pool of durum wheat leaves under water deficit revealed an increase in the amount of polyunsaturated molecules.

In the whole, the results obtained reveal the existence in durum wheat of a gene family encoding putative sPLA₂s and suggest a role of specific sPLA₂s isoforms in durum wheat response to water deficit.