STRUCTURAL CHARACTERISATION OF GENES ENCODING PROTEINS OF THE PDI FAMILY IN WHEAT

D’ALOISIO E., TANZARELLA O.A., PORCEDDU E., CIAFFI M.

Department of Agrobiology and Agrochemistry, University of Tuscia, Via S. C. De Lellis, 01100 Viterbo (Italy) - elisalauradal@hotmail.com

Protein Disulfide Isomerase (PDI) gene family, wheat, gene structure, expression analysis

The PDI (Protein Disulfide Isomerase) family includes several genes whose products are responsible for diversified metabolic functions, the proteins differ also for number and position of the active thioredoxin-like sites, for presence/absence of other domains and of the KDEL signal of retention in the endoplasmic reticulum. In plants the PDI family includes eight different phylogenetic classes. Isolation and characterization in wheat of the three homoeologous gene sequences encoding classical PDI (TaPDIL1-1) and of their promoter sequences have been reported previously. Some studies of molecular characterization, expression analysis and cell localisation in rice and maize have suggested the involvement of the classical PDI in the assemblage and deposition of storage proteins in these species. In wheat the likely involvement of the classical PDI, as well as the potential participation of PDI-like proteins, in the storage protein folding and in formation of high molecular weight protein aggregates makes their study particularly interesting. Our goal is the characterization of the complexity and diversity of the PDI gene family in wheat. A cross search using PDI-like sequences of rice in the wheat EST databases “TIGR wheat gene index” and “HarvEST Wheat” identified nine sequences coding for PDI-like proteins in wheat, whose full length cDNAs have been cloned. Phylogenetic analysis allowed the assignment of the ten PDI and PDI-like sequences of wheat to the eight phylogenetic groups identified in plants. Thus at least one gene has been cloned for each phylogenetic group. The search for conserved motives in the deduced amino acid sequences of the nine isolated genes, by comparison with sequences in different protein data bases, revealed a high level of structural similarity between the proteins encoded by genes belonging to the same phylogenetic group. The comparison of the genomic organisations of three wheat PDI-like genes (TaPDIL2-1, TaPDIL4-1 e TaPDIL5-1) with their orthologous of rice and Arabidopsis showed a high level of conservation of their structural features (exon/intron structure, exon length and position of the active sites) among members of the same phylogenetic group. Most likely such conservation reflects the essential functional role of their encoded proteins. The chromosome location of the genes encoding two wheat PDI-like proteins (TaPDIL4-1 and TaPDIL5-1) was determined through Southern analyses of DNA extracted from nulli-tetrasomic and ditelosomic lines of Chinese Spring. The three homoeologous gene sequences encoding TaPDIL4-1 were located in the short arm of the group 1 chromosomes, those encoding TaPDIL5-1 in the long arm of the group 5 chromosomes. Northern analysis of TaPDIL2-1, TaPDIL4-1, TaPDIL5-1 and TaPDL1-1 detected different expression patterns in the analysed tissues. Further studies will be necessary to complete the molecular characterisation of this multigenic family in wheat. The structural characterisation and detailed analysis of expression patterns will be extended to the remaining six genes encoding PDI-like proteins. An exhaustive knowledge of the structural features and regulation of the PDI family genes will be useful to design the most suitable strategies.
for their functional characterization, in particular for the silencing of single genes or of gene groups through the RNA interference (RNAi) technology. The effect on the characteristics of seed storage proteins produced by the progressive knock-out of PDI and PDI-like genes will allow the understanding of their role in the formation of protein aggregates and will highlight possible functional redundancies.