Title Computational annotation of genes differentially expressed during fruit ripening in olive
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Abstract

This study deals with the identification of large sets of differentially expressed genes in developing olive fruits and the subsequent computational annotation by means of different softwares. Total RNA from fruits of the cv. Leccino, sampled at three different developmental stages [i.e., 30 days after full bloom (DAF) (A), 90 DAF (pit hardening, B) and 130 DAF (veraison, C)], was used for the identification of differentially expressed genes putatively involved in main processes characterizing fruit growth, development and ripening. In order to isolate up- and down-regulated genes, four subtractive hybridization libraries were constructed: forward and reverse between A and B, forward and reverse between B and C. All sequences were preliminary analyzed through BlastX against non-redundant NCBI databases and about 60% of them showed similarity to known proteins. Library-specific cDNA repertories were annotated according to the three main vocabularies of the gene ontology (GO): cellular component, biological process and molecular function, respectively. BlastX analysis, GO terms mapping and annotation analysis were performed using the Blast2GO software, a research tool designed with the main purpose of enabling GO based data mining on sequence sets for which no GO annotation is yet available. Furthermore, the olive fruit-specific transcriptome dataset was used to query all known KEGG (Kyoto Encyclopaedia of Genes and Genomes) metabolic pathways for characterizing and positioning retrieved EST records within the drupe development. On the whole, our approach led to the identification of differentially expressed sequences that proved to be significantly differentiated in terms of GO category sequence amounts among the three developmental stages. Similarly, the integration of the olive sequence datasets within the MapMan platform for microarray analysis allowed us to display them into pathways along with metabolic and regulation diagrams useful for the definition of key functional categories in time course analyses for gene groups. Such characterization is a first step toward both, functional genomics and systems biology research in olive, especially for understanding gene regulatory networks and metabolic pathways in fruit growth, development and ripening.