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TRANSCRIPTOME ANALYSIS OF FOUR CHLOROPLAST DEVELOPMENTAL BARLEY MUTANTS

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We investigated four *albina* and *xantha* barley mutants representing successive steps in the chloroplast biogenesis and the corresponding wild type (WT) with the Affymetrix Barley1 GeneChip® (about 22.000 probe sets) to assess the variations of gene expression associated with chloroplast development. When mRNA isolated from leaves of mutant plants grown at 20°C were compared with mRNA extracted from green leaves of WT plants grown under the same conditions a number of probe sets were found more than 2 fold up- or down-regulated. Since the mutants analyzed represent successive steps in the chloroplast biogenesis, the number of probe sets up- or down-regulated decreased according to progress in chloroplast development. The number of genes up- or down-regulated during growth at 20°C was similar (19% of transcriptome) in the first three mutants (*alb-e*¹⁶, *alb-f*¹⁷ and *xan-s*⁴⁶), while for the fourth (*xan-b*¹²) the number of genes modified was reduced to 9% showing a clear normalization of the transcriptome as chloroplast development proceed. The genes up- and down-regulated in the mutants compared to WT were subdivided into classes to identify groups of genes whose expression is associated to a given stage of the chloroplast development. The up- and down- regulated gene lists were converted into the homologous sequences of Arabidopsis (cut off E-value = e^{-10}) and loaded onto the mapman software in order to gain information about the metabolic changes associated with the chloroplast development. Some keys metabolic pathways (e.g. chlorophyll or carotenoids biosynthesis) have been studied thoroughly: all intermediates of the pigment biosynthetic pathways were detected by HPLC analysis and all genes involved were verify by qRT-PCR. The cross-references between transcriptomic data and morphological or biochemical traits allowed to identify subregulons of genes regulated as pigments synthesis proceed. All together these data provide useful information for the understanding of the regulation of nuclear genes associated with chloroplast development.