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Comparative *De Novo* Transcriptomic Analysis of Photosynthetically Active and Non-Photosynthetically Active Tissues of Variegated *Pelargonium zonale* Leaves

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Abstract

Variegated *Pelargonium zonale* leaves have proven to be an excellent model system to examine source–sink interactions within the same organ providing the equal microenvironment conditions, unlike common shoot/root relation studies. Photosynthetically non-active (W) mesophyll cells contain smaller plastids lacking thylakoid membranes or starch granules, and exhibit no peroxisomes in comparison to photosynthetically active (G) cells. With the aim of gaining a deeper insight into molecular phenotype of W leaf tissue, particularly the one related to photosynthetic-dependent H_2O_2 metabolism, transcriptomes of these two metabolically contrasted tissues were compared.

High-quality total RNA from W and G leaf tissues was extracted according to our previously optimised protocol. Highly purified cDNA libraries were synthesized and sequenced on an Illumina platform. The ambiguous nucleotides, adapter sequences, and low-quality sequences were trimmed and the read quality was checked before and after the trimming. In total, 39763284 (with Q30=94.3%) and 42062153 (with Q30=94.0%) clean reads were obtained in G and W total RNA samples, respectively, and used to perform transcriptome assembly by Trinity software. After removing the redundancy, via Corset software, 139811 transcripts with 139575 unigenes were annotated through comparison with seven commonly used databases (NCBI non-redundant protein and nucleotide sequences; PFAM; Clusters of Orthologous Groups of proteins, Swiss-Prot, KEGG, GO).

Analysis of differentially expressed genes was performed using DESeq2 R package and revealed 4668 up-regulated genes and 6689 down-regulated genes in G tissue compared with W one. Among the up-regulated genes in G tissue, the majority was associated with cytoskeleton, photosynthetic processes, plastids, thylakoids and transport, while in W tissue up-regulated genes were mainly found to encode enzymes with ATPase activity, carbohydrate absorption and digestion, callose, pectin and linoleic acid metabolism. Moreover, a significant difference between these two tissues differing in H_2O_2 generation rate was observed in the expression level of genes involved in H_2O_2 scavenging. Enzymatic constituents of the ascorbate-glutathione cycle and glutathione-S-transferase were up-regulated in W tissue, while catalase, glutathione-peroxidases and three Class III peroxidases were all up-regulated in G tissue. The obtained transcriptome results were correlated with previously revealed morphological, biochemical, and molecular characteristics of these two tissues.

Keywords:

antioxidative metabolism, differential gene expression analysis, H_2O_2 scavenging, photosynthesis, source/sink metabolism, variegated plants.

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