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RNA-Seq analysis differentially expressed genes in purple pakchoi (*Brassica campestris* ssp. *chinensis* Makino.) under low temperature

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Low temperature is the main environmental factor that affect anthocyanin biosynthesis and accumulation in purple pakchoi, which is one of the most popular vegetables in China with high anthocyanin content. In the present study, the transcriptome profiles of the purple pakchoi cultivar "ziyi", which maintained at 5°C (low temperature, LT) and 20°C (normal temperature, NT) for 10 d, were analyzed using Illumina paired-endsequencing technology in order to reveal mechanisms associated with anthocyanin biosynthesis and accumulaiton at low temperatures. The anthocyanin content under 5°C treatment was gradually increased, and the purple color deepened as compared to that observed under 20°C. After *de novo* assembly and quantitative assessment of the obtained reads, 114,043 unigenes were obtained, and 76,369 sequences were annotated by aligning the sequences against five public databases. Of all the differentially expressed genes (DEGs), 4,172 and 3,918 up-regulated genes were identified in the 5°C and 20°C treatment groups, respectively. Twelve major anthocyanin accumulation- and biosynthesis-related genes, including transcription factors (TFs), were identified, and their expression levels were estimated. The structural genes *PAL, C4H, F3H, CHS*, and *CUT75G1* and *TFs HY5, MYB44*, and *MYB113* were induced by low temperature conditions. Overall, this is the first transcriptome sequencing analysis of this plant species under low temperature conditions. Studies of the DEGs involved in the anthocyanin accumulation and biosynthesis pathways provide insights into the regulatory mechanisms of anthocyanin synthesis and accumulation in purple vegetables at low temperature.

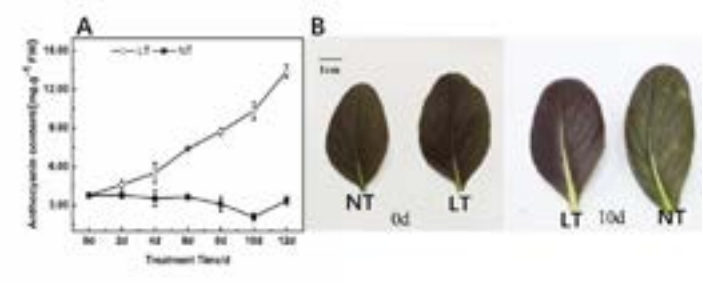

Fig. 1 Anthocyanin content and the color of purple pakchoi at 5°C and 20°C.Anthocyanin content changes in purple pakchoi treated with low temperatures from 0 d to 12 d (A); comparison of color in purple pakchoi under low temperature conditionsat 0 d and 10 d.

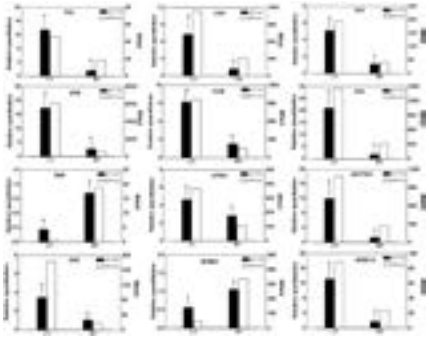

Fig. 2 Real-time PCR. Data represent the mean ± SE (n = 3 or 4). Single and double asterisks indicate statistical differences compared to the control (t-test, p<0.05). The relative gene expression of 12 randomly selected genes examined usingqRT-PCR (5 or 0.01, respectively)

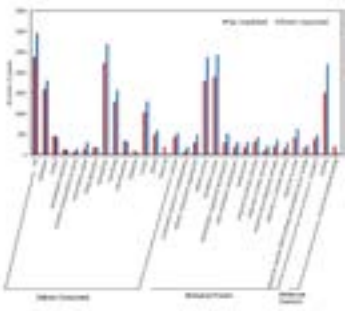

Fig. 3 Four major pathways likely implicated in the effects of low temperatures on purple pakchoi during IMC identified by a KEGG enrichment analysis. Thered line representsa p-value of 0.05

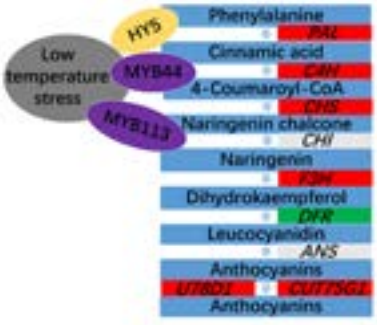

Fig. 4 Anthocyanin biosynthetic pathwaysin purple pakchoi under low temperature conditions. HY5, MYB44 and MYB113 are transcription factors.Genes involved in this pathway include PAL, C4H, CHS, F3H, DFR, ANS, U78D1and CUT75G1. Red representsthe up-regulation of the gene expression level, while green denotesdown-regulation.Gray indicates a non-DEG.

Recent Publications

1. N.U. Ahmed, J.I. Park, H.J. Jung, et al., Anthocyanin biosynthesis for cold and freezing stress tolerance and desirable color in Brassica rapa, Funct Integr Genomics. 2015 Jul;15(4):383-94.
2. C. Kang, O. Darwish, A. Geretz, et al., Genome-scale transcriptomic insights into early-stage fruit development in woodland strawberry Fragaria vesca., Plant Cell 25 (2013)1960-78.
3. Y. Qu, A. Zhou, X. Zhang, et al., De Novo Transcriptome Sequencing of Low Temperature-Treated Phlox subulata and Analysis of the Genes Involved in Cold Stress, Int J Mol Sci 16 (2015)9732.
4. X. Gu, Y. Chen, Z. Gao, et al., Transcription factors and anthocyanin genes related to low-temperature tolerance in rd29A:RdreB1BI transgenic strawberry, Plant Physiology & Biochemistry Ppb 89 (2015)31-43.
5. C. Zhang, H. Jia, W. Wu, et al., Functional conservation analysis and expression modes of grape anthocyanin synthesis genes responsive to low temperature stress, Gene 574 (2015)168-77.

Biography

Yuying Zhu has working on Shanghai Academy of Agricultural Sciences since 1984. Now he has his expertise in pakchoi cultivation and breeding. He has chosen 15 pakchoi varieties that have large area.

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