

EXPRESSION OF CALCIUM-DEPENDENT PROTEIN KINASE (CDPK) GENES IN VITIS AMURENSIS UNDER ABIOTIC STRESS CONDITIONS

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Abiotic stresses, such as extreme temperatures, soil salinity, or water deficit, are one of the major limiting factors of crop productivity worldwide. Examination of molecular and genetic mechanisms of abiotic stress tolerance in plants is of great interest to plant biologists. Calcium-dependent protein kinases (CDPKs), which are the most important Ca^{2+} sensors in plants, are known to play one of the key roles in plant adaptation to abiotic stress. CDPK is a multigene family of enzymes. Analysis of *CDPK* gene expression under various abiotic stress conditions would help identify those *CDPKs* that might play important roles in plant adaptation to abiotic stress. We focused on studying *CDPK* gene expression under osmotic, water deficit, and temperature stress conditions in a wild-growing grapevine *Vitis amurensis* Rupr., which is native to the Russian Far East and is known to possess high adaptive potential and high level of resistance against adverse environmental conditions. Healthy *V. amurensis* cuttings (excised young stems with one healthy leaf) were used for the treatments. For the non-stress treatment, we placed the cuttings in distilled water for 12 h at room temperature. For the water-deficit stress, detached cuttings were laid on a paper towel for 12 h at room temperature. For osmotic stress treatments, the cuttings were placed in 0.4 M NaCl and 0.4 M mannitol solutions for 12 h at room temperature. To examine temperature stress tolerance, the *V. amurensis* cuttings were placed in a growth chamber at +10°C and +37°C for 12 h. The total expression of *VaCDPK* genes was examined by semiquantitative RT-PCR with degenerate primers designed to the *CDPK* kinase domain. The total level of *CDPK* gene expression increased under salt and decreased under low temperature stress conditions. We sequenced 300 clones of the amplified part of different *CDPK* transcripts obtained from the analyzed cDNA probes. Analysis of the cDNA sequences identified 8 different *CDPK* genes (*VaCDPK1a, 1e, 1d, 2a, 3a, 3b, 3c, 3d*). We sequenced full cDNA sequences of the genes and analyzed their expression levels by real-time PCR and FAPP method, which has been recently developed by our research group. The prevalent *CDPK* transcript was *VaCDPK3a* under both non-stress and abiotic stress conditions. Under high-salt conditions, *VaCDPK1d, 1e, 3b, and 3d* transcripts were up-regulated. Under high mannitol conditions, expression of *VaCDPK1e* and *3b* was up-regulated, while expression of *VaCDPK1d, 3c, and 3d* was only slightly induced. Under water-deficit, expression of only *VaCDPK3b* and *3c* genes was induced. Cold stress induced expression of *VaCDPK2a* and *3d* genes; while hot stress induced expression of *VaCDPK1a, 1d, 1e, 2a, 3a, and 3c* genes. Taken together, the data show that the *VaCDPK* genes are transcriptionally regulated by osmotic, water-deficit, and temperature stresses. The differential expression of the *VaCDPK* genes during osmotic, water-deficit, and temperature stresses is suggestive of their involvement in the underlying signal transduction pathways.