Functional Characterization and Abiotic Stress-Responsive Expression of WRKY8/23/28-Dependent Genes MIOX2, AT2G45180, GATL6, and EXLA2 in Plants

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Abstract: MIOX2, AT2G45180, GATL6 and EXLA2 are Wrky8/23/28-dependent genes. 35S:WRKY8-SRDX down regulate 92 genes, 35S:WRKY23-SRDX down regulate 127 genes while 35S:WRKY28-SRDX down regulate 236 genes. MIOX2, AT2G45180, GATL6, and EXLA2 were included in those eight genes that were down regulated by all of three WRKY-SRDX complexes. MIOX2 has inositol oxygenase activity located in the cytoplasm involved in the L-ascorbic acid biosynthetic process, inositol catabolic process, oxidation-reduction process, and syncytium formation. AT2G45180 is located in the chloroplast thylakoid membrane, an extracellular region involved in lipid transport. GATL6 has polygalacturonate 4-alpha-galacturonosyltransferase activity and transferase activity, located in Golgi apparatus, an integral component of the membrane and involved in carbohydrate biosynthetic process, cell wall organization, pectin biosynthetic process, and polysaccharide biosynthetic process. EXLA2 is located in the extracellular region and is involved in plant-type cell wall loosening, plant-type cell wall organization, sexual reproduction, and unidimensional cell growth. In the present study, expression analysis of MIOX2, AT2G45180, GATL6, and EXLA2 genes was done. In the first section, bioinformatics analysis of MIOX2, AT2G45180, GATL6, and EXLA2 genes was presented, including gene structure, promoter cis-acting elements, and protein structure. The expression levels of different overexpression lines of MIOX2, AT2G45180, GATL6 and EXLA2 genes were significantly higher as compared to control. Sub cellular localization of MIOX2, AT2G45180, GATL6, and EXLA2 genes in Nicotiana benthamiana leaves revealed that MIOX2 expressed in the cytosol, AT2G45180 showed its expression in plastids, GATL6 revealed its expression in endoplasmic reticulum while EXLA2 localized in extracellular membrane. Expression analysis under abiotic stresses analyzed by gRT-PCR showed a dynamic response of MIOX2, AT2G45180, GATL6, and EXLA2 genes to abiotic stresses. Expression profiles of these genes revealed that MIOX2 and AT2G45180 genes were up regulated under cold stress and heat stress at 24-hour treatment while GATL6 and EXLA2 genes were down regulated under cold and heat stresses. Under salt and mannitol stresses MIOX2 were up regulated at 24 h treatment and AT2G45180 was up regulated at 24 h mannitol treatment while other genes were down regulated. MIOX2 and AT2G45180 genes were up regulated under dark stress, while GATL6 and EXLA2 were up regulated at 6 h dark treatment and down regulated at 12 h and 24 h dark treatment. Our study lays a foundation for further gene function and expression studies of MIOX2, AT2G45180, GATL6, and EXLA2 genes.

Keywords : WRKY, SRDX, MIOX2, AT2G45180, GATL6, EXLA2, stress, expression, GUS, down regulated, up regulated, treatment

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