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Biochemical Characterization of Transgenic Tobacco Plants That Express a Human Dehydroascorbate Reductase

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Dehydroascorbate reductase (DHAR; EC 1.8.5.1) is an enzyme that is critical for maintenance of an appropriate level of ascorbate in plant cells. Ascorbate (ascorbic acid, AsA) acts as an important antioxidant in both enzymatic and non-enzymatic reactions in plant cells. DHAR catalyzes the reduction of dehydroascorbate (DHA) to AsA by glutathione (GSH). To analyze the physiological role of DHAR in environmental stress adaptation, we developed transgenic tobacco (*Nicotiana tabacum* cv Xanthi) plants expressing human DHAR gene in chloroplasts and investigated their protection effect to oxidative stress induced by methyl viologen (MV). DHAR activity and AsA content in transgenic plants were 1.55 and 1.95 times higher than nontransgenic (NT) plants, respectively. In addition, oxidized glutathione (GSSG) content was approximately 2.9 times higher than NT plants. The ratios of AsA to DHA and GSSG to GSH were changed by the overexpression of DHAR as expected. When tobacco leaf discs were subjected to MV at 5 μ M, T₀ DHAR transgenic plants showed about 50% reduction in membrane damage relative to NT plants. These results indicate that DHAR may contribute to the protection against the oxidative stress. The characterization of T₁ DHAR transgenic plants is under investigation in terms of various stresses.

Keywords: ascorbate, dehydroascorbate, glutathione, methyl viologen (MV) stress