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The Role of Hydrogen Peroxide-Producing and Hydrogen Peroxide-Consuming Peroxidases in the Leaf Apoplast of Cowpea in Manganese Tolerance1[W]

Marion Maria Fecht-Christoffers, Hendrik Führs, Hans-Peter Braun, and Walter Johannes Horst*
Institute of Plant Nutrition (M.M.F.-C., H.F., W.J.H.) and Department of Applied Genetics (H.-P.B.),
University of Hannover, 30419 Hannover, Germany

The apoplast is considered the leaf compartment decisive for manganese (Mn) toxicity and tolerance in cowpea (Vigna unguiculata). Particularly apoplastic peroxidases (PODs) were proposed to be key enzymes in Mn toxicity-induced processes. The presented work focuses on the characterization of the role of hydrogen peroxide (H_2O_2)-producing (NADH peroxidase) and H_2O_2-consuming peroxidase (guaiacol POD) in the apoplastic washing fluid (AWF) of leaves for early stages of Mn toxicity and genotypic differences in Mn tolerance of cowpea. Leaf AWF of the Mn-sensitive cultivar (cv) TVu 91 but not of the Mn-tolerant cv 1987 showed an increase of guaiacol-POD and NADH-peroxidase activities at elevated AWF Mn concentrations. two-dimensional resolutions of AWF proteins revealed that cv TVu 91 expressed more and additional proteins at high Mn treatment, whereas Mn-tolerant cv TVu 1987 remained nearly unaffected. In both cultivars, NADH-peroxidase activity and accompanied H_2O_2 formation rate in vitro were significantly affected by Mn^{21}, p-coumaric acid, and metabolites occurring in the AWF. The total phenol concentration in the AWF was indicative of advanced stages of Mn toxicity but was rather unrelated to early stages of Mn toxicity and genotypic differences in Mn tolerance. The NADH oxidation by AWF PODs was significantly delayed or enhanced in the presence of the protein-free AWF from cv TVu 1987 or cv TVu 91, respectively. High-performance liquid chromatography analysis of AWF indicates the presence of phenols in cv TVu 1987 not observed in cv TVu 91. We conclude from our studies that the H_2O_2-producing NADH peroxidase and its modulation by stimulating or inhibiting phenolic compounds in the leaf apoplast play a major role for Mn toxicity and Mn tolerance in cowpea.