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# Root Proteome Analysis of Sorghum in Response to Cadmium using Label-free Proteomics Techniques

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### [Introduction]

The contamination of soil and groundwater with heavy metals poses significant environmental hazards, leading to considerable losses in plant productivity and human health problem. Cd is suggested to cause damage even at very low concentrations, and healthy plants may contain Cd levels that are toxic to mammals. Sorghum plants were able to accumulate large quantities of heavy metal in the shoots, as well as highly tolerant to metal pollution. In the present study, we focused on roots (the primary site of Cd exposure) to explore the Cd-response proteins and their possible functions in response to heavy metal stress. A high-throughput proteome technique was used to identify proteins that are highly regulated by Cd stress in sorghum roots and to deepen our understanding of the molecular basis of Cd responses in plants.

#### [Materials and Methods]

Seeds of *Sorghum bicolor* L. (BTX 623) were obtained from the National Germplasm Resources of USDA-ARS, plant stress and germplasm development unit, USA. Surface-sterilized seeds were germinated on moist filter paper in Petri dishes and grown in a growth chamber. After 5 days of planting, the seedlings were transplanted to a vessel containing Hoagland solution. After 10 days, the seedlings were exposed to different concentration of Cd (0, 100, 150  $\mu$ M CdCl<sub>2</sub>) as a Cd solution. After 5 days of Cd-treatment, the roots were harvested, and three replications were prepared for each treatment for morpho-physiological and proteomic analysis. 2-DE electrophoresis was performed to confirm the protein separation and MALDI-TOF/TOF MS was applied to identify the proteins.

## [Results and Discussions]

In whole plants, roots are the primary site through which heavy metals gain access. Plant molecular response to Cadmium (Cd) stress is characterized by the synthesis of stress-related proteins and signaling molecules. To better understand the Cd-responsive mechanisms of sorghum, the present study was conducted using the proteomic technique. An increase in Cd levels inhibited the uptake of interacting metal ions. Ten-day-old sorghum seedlings were exposed to different concentrations (0, 100, and 150  $\mu$ M) of CdCl<sub>2</sub>, and proteins were extracted from the roots of 15-day-old sorghum seedlings. The protein abundance involved in glycolysis and tricarboxylic acid cycle, including glyceraldehyde 3-phosphate dehydrogenase was changed in in sorghum roots after Cd treatment. Significant changes were absorbed in the levels of proteins known to be involved in carbohydrate metabolism, translation and stress responses. In addition, Cd stress had an inhibitory effect on carbon fixation, ATP production and the regulation of protein synthesis. As expected, most of the up-regulated proteins are involved in heavy metal detoxification and antioxidant processes. Enzyme activity analysis revealed that ascorbate peroxidase and glutathione S-transferase activity was stimulated by Cd treatment. Abundance changes of these proteins, together with their putative functions provide us a new insight that can lead to an integrated understanding of the molecular basis of Cd responses in plants.

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