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Leaf Proteome Responses of Azuki Bean at Early Vegetative Stage to Waterlogging Stress

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[Introduction]
Recently, Korea has been demanding measures to prevent the overproduction of rice by increasing the self-sufficiency rate of the crops through cultivating field crops in the rice field. Waterlogging is one of the major environmental stresses, and becoming increasingly frequent worldwide due to climate change, limiting plant growth, and thereby reducing the yield of crops. Azuki bean is typically considered high temperature and humid but highly sensitive to waterlogging during its life cycle, caused by poor drainage, flooding, and long periods of rainfall.

[Materials and Methods]
The seeds of Azuki bean were collected from the Rural Development Administration, Korea. Soil properties and growth characteristics of Azuki bean were measured during the second and fifth leaf stage. To confirm the expression pattern of the protein, the Azuki bean leaves (from two and five leaf stage) were collected and sampled to extract the protein. 2-DE electrophoresis was performed to confirm the protein separation and LTQ-FT-ICR MS was applied to identify the proteins.

[Results and Discussion]
Waterlogging caused a significant reduction in morphological and biochemical properties both in 2-leaf and 5-leaf stage. More than 400 protein spots were detected on 2-D gels from 2-leaf stage of which a total of 43 protein spots that exhibited more than a 1. 5-fold changes in intensity using image analysis software. However, a total of 21 differentially expressed proteins were successfully analyzed using LTQ-FT-ICR MS analysis. Among these proteins, a total of 14 proteins showed increased expression, and 7 proteins showed decreased expression in the treated samples compared to their levels in untreated seedlings. However, in the 5-leaf stage proteome, 28 protein spots were found to be expressed differentially ($p > 0.05$) upon waterlogging stress. A total of 16 proteins were increased significantly while 12 proteins were decreased abundances. In the waterlogged leaves from 2-leaf and 5-leaf stage, glyceraldehyde-3-phosphate dehydrogenase, glycine dehydrogenase, NADH dehydrogenase, malate dehydrogenase, and ATP synthase were specifically accumulated to manage energy consumption, and minimize oxidative damage. The abundance of the most identified protein species from the leaves that function in stress response and metabolism was significantly enhanced, while protein species involved in transcription, photosynthesis and regulation were severely reduced. qRT-PCR revealed that the expression levels of these genes correlated with their observed protein abundance. Taken together, these findings shed light on the complex mechanisms underlying waterlogging tolerance in Azuki bean.

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