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Proteomic Approach to Analyze Cold-Responsive Proteins in Rice Leaf Tissue

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Objectives

Cold stress is one of the major abiotic stresses in agriculture worldwide. To response to cold stress, plants must receive low temperature signals and transduce them into biochemical responses. After several primary transient response to cold, such as membrane depolarization and increase in cytosolic calcium concentration, there is an orchestration of subsequent events in plant physiology. To examine the response of rice to cold stress, change in protein expression was analyzed using a proteomic approach.

Material and Methods

1. Materials

Rice (*Oryza sativa* L. cv. Dongjinbyeo) was used in this study. Fourth- and fifth-leaf stage rice seedlings grown under natural light in a greenhouse (20°C/30°C) were used for stress treatment

2. Methods

For cold treatment, plants were transferred to growth chamber set at 4°C or 10°C and incubated for varying times. Total soluble proteins were extracted from leaf tissue. The soluble proteins were fractionated with 15% PEG. Protein profiles after cold treatment were analyzed by two-dimensional gel electrophoresis (2-DE). Protein spots were visualized by silver staining. Differentially expressed spots were identified by peptide mass fingerprinting using matrix assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS).

Results and Discussion

More than 1,000 protein spots were reproducibly detected on 2-DE gel including 43 that were up-regulated and 4 were down-regulated under 4°C treatment, 47 that were up-regulated and 7 were down-regulated under 10°C treatment, respectively. All of differentially expressed proteins spots were subjected to MALDI-TOF mass spectrometry followed by database searching which allowed the identification of 34 protein spots under 4°C treatment, 27 protein spots under 10°C treatment, respectively. These proteins are involved in redox regulation, reactive oxygen species scavenging, signal transduction, and nitrogen and animo acid metabolism-related proteins. Proteins involved in transcription factor or in plant defense mechanism were also identified and will be discussed.

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