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# Proteome Analysis of Nodulation in Hypernodulating Soybean Mutant

LIM, Chae Woo · HWANG, Cheol-Ho*

*School of Bioresources Sciences, Dankook University*

## Objectives

Proteome analysis was performed to investigate the proteins responsible for the difference shown in between hypernodulation mutant, SS2-2 and it's mother line, Sinpaldalkong2. Proteome expression map of root or nodule in either 0 or 21 days after an inoculation of *Bradyrhizobium japonicum*, respectively was analyzed to distinguish any difference between two soybeans. Based on these proteomic analyses, proteins involved in nodulation or more specifically hypernodulation will be examined collectively and their functions in the process of nodulation will be interpreted with information from the Legume genome project ongoing.

## Materials and Methods

1. Materials
   - Plant: Sinpladalkong2 (mother line) and SS2-2 (hypernodulation mutant)
     Bacteria: *Bradyrhizobium japonicum*, USDA110

2. Methods
   - Two soybeans were grown under controlled condition of 16 hrs light 26℃/23℃ (day/night).
     Nodule was induced by an inoculation of *Bradyrhizobium japonicum*, USDA110.
     Two dimensional electrophoresis was done following the protocol of BioRad withmodifications especially in sample preparation. The resulting gels were stained with Coomassie blue and the 2D images were analyzed using Melanie III.

## Results and Discussion

1. About 600 protein spots were seperated across the pI range of 4-7 in proteome expression map of roots or nodules(0 or 21 DAI) of both Sinpladalkong2 and SS2-2.

2. In comparison of proteome map of root and nodule from SS2-2 with Sinpaldalkopng2, it was found that five variable regions showed difference in spot patterns.

3. Newly induced, upregulated, and downregulated proteins are being characterized by MALDI-TOF/MS and identified in order to establish the database of nodule proteins.

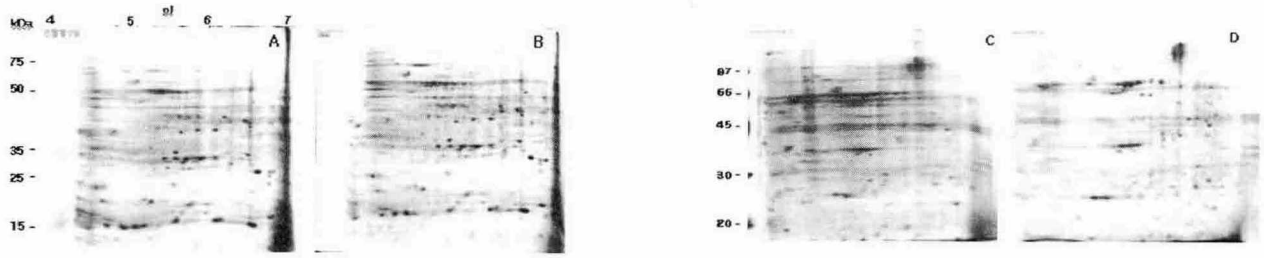


**Figure 1.** 2-D electrophoresis of proteins from Sinpaldalkong2 at 0 DAI root (A), 21 DAI nodule (D) and its Supernodulating mutant, SS2-2 at 0 DAI root (B), 21 DAI nodule (C). First dimension was focused using a 17cm IPG strip with a linear pH gradient 4-7 loaded with 2 mg of protein. For the second dimension, 12% SDS-PAGE gel were used. The separated proteins were visualized by Coomassie blue (R250) staining and analyzed using Melanie III.

*Corresponding author. Tel 042-860-4492 E-mail jeonjh@kribb.re.kr