P-214

Rhizobial genes involved in the symbiosis with host legumes

 \bigcirc Hien P. Nguyen¹, Omar M. Faruque¹, Hiroki Miwa¹, Takakazu Kaneko², Shusei Sato³, Shin Okazaki¹

¹Graduate School of Agriculture, Tokyo University of Agriculture and Technology, ²Faculty of Life Sciences, Kyoto Sangyo University, ³Graduate School of Life Sciences, Tohoku University

E-mail: nguyenphuochien92@gmail.com

Symbiosis specificity has been demonstrated in the interactions between rhizobia and leguminous plants. While the compatible symbiosis leads to formation of the nitrogen-fixing nodules, some cultivars exhibit incompatibility with the specific strains, resulting in ineffective nodulation. *Vigna radiata* cv. KPS1 and soybeans carrying *Rj4* gene are incapable of nodulation with *Bradyrhizobium elkanii* USDA61 due to presence of pathogen-like proteins. This work was performed to understand the molecular mechanisms underlying nodulation incompatibility of KPS1 and *Rj4* plants. By mutagenesis, we isolated six Tn5 mutants of USDA61, designated BE5, BE21, BE53, BE85, BE103 and BE168, which could nodulate KPS1 efficiently. The T3SS mutant BErhcJ could form nodules on both KPS1 and Rj4 soybean. Similarly, inoculation test on Rj4 soybean revealed that four mutants BE5, BE85, BE103 and BE168 were able to nodulate, suggesting the commonality of incompatible mechanism of KPS1 and Rj4 plants. Sequencing analysis of the Tn5-flanking sequence indicated that five genes encoding the cytosine deaminase, hypothetical proteins, manitol-binding protein and GTP pyrophosphokinase respectively were disrupted. The deduced gene products of these Tn5 mutants shared high similarity with the hypothetical proteins of pathogens. One tts box was found at 96-bp in upstream of *bel53* gene found in BE53, indicating that the gene product was involved in T3SS. To determine the infectious properties of USDA61 and its mutant, the infection process of BE21 was analysed using DsRed and gusA genes. Both USDA61 and BE21 infected KPS1 through infection threads (ITs) and formed bumps. Our results indicated that KPS1 halted both infection and nodule organogenesis of *B. elkanii* USDA61.