RECENT RESEARCH ACTIVITIES

Tissue-specific transcriptome analysis in nodules of *Lotus japonicus* identified a novel transporter, LjMATE1, that assists the Fe translocation to nodules by providing citrate

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Legume plants can establish symbiotic nitrogen fixation (SNF) in nodules as a plant-microbe collaboration, where the nutrients between host plant cells and their resident bacteria (rhizobium) are actively exchanged. As such major molecules nitrogen compounds and carbohydrates are known, while various minerals are also transported; however knowledge about the molecular basis of plant transporters mediating exchange of those metabolites is still very limited. To elucidate the metabolite dynamics relevant for SNF in nodules, three cell types were isolated from a nodule of a model legume, *Lotus japonicus*, using laser

microdissesction (LM) (Figure 1), and one-color microarray analysis was done. In our cell type-specific profiling, many genes were identified as being expressed in nodules in a spatial-specific manner. For example, a number of metabolic genes for a phenylpropanoid pathway were found as being highly expressed in the nodule cortex accompanied by those encoding putative transporters for organic metabolites.

Among them, a MATE-type transporter (LjMATE1) was identified as an infection zone-specific gene. Reporter gene experiments indicated that *LjMATE1* expression was restricted to the infection zone of nodules. To characterize the transport function of LjMATE1, we conducted a biochemical analysis using *Xenopus* oocyte as a heterologous system, and found that LjMATE1 is a citrate-specific transporter. The physiological role of LjMATE1 was analyzed with L. japonicus RNA interference (RNAi) lines, which revealed limited growth only under nitrogen deficient conditions with inoculation of rhizobia compared with the controls. It was noteworthy that Fe localization was clearly altered in nodule tissues of knock-down line and the Fe content in nodules was significantly lower than that in wild-type. These results strongly suggest that LiMATE1 is a nodule-specific transporter that assists the Fe translocation from the root to nodules by providing citrate.

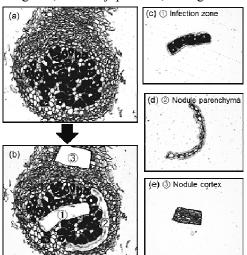


Figure 1. Isolation of three tissues from cross section of *L. japonicus* nodule using laser microdissection (LM). Cross section of a nodule; (a) before, and (b) after, LM. (c-e) Sectioned three cell-types; (c) infection zone, (d) nodule parenchyma; and (e) nodule inner cortex.

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References

[1] Takanashi K., Takahashi H., Sakurai N., Sugiyama A., Suzuki H., Shibata D., Nakazono M., Yazaki K., "Tissue-specific transcriptome analysis in nodules of *Lotus japonicus*", *Molecular Plant-Microbe Interaction*, vol. 25. pp. 869-876, 2012.

[2] Takanashi K., Yokosho K., Saeki K., Sugiyama A., Sato S., Tabata S., Ma J.F., Yazaki K., "LjMATE1 - a citrate transporter responsible for iron supply to nodule infection zone of *Lotus japonicus*", *Plant and Cell Physiology*, vol. 54. pp.585-594, 2013.