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学 位 論 文 題 名

Role of acid-tolerant arbuscular mycorrhizal fungi in plant acid tolerance and their
distribution along soil pH gradients

(耐酸性アーバスキュラー菌根菌の植物耐酸性に果たす役割と土壌 pH 勾配に
沿った分布)

This thesis consists of 14 figures, 9 tables, 105 references, General introduction, two Chapters, and General discussion in a total of 89 pages with five accompanying publications.

Plant productivity is constrained by soil acidity due to high-levels of aluminum that inhibits root elongation and thus nutrient uptake. *Miscanthus sinensis* is a common pioneer grass in early primary succession in acidic soil, but largely depends on the symbiotic associations with arbuscular mycorrhizal (AM) fungi during colonization of strongly acidic soil. AM fungi associate with most land plants and supply immobile nutrients, especially phosphorus (P), to their host plants. In the present study, the mechanism underlying the improved plant acid-tolerance by mycorrhizal formation was investigated with respect to acid-tolerance/sensitivity of AM fungi. Furthermore, the global distribution pattern of acid-tolerant/sensitive AM fungi along soil pH gradients was for the first time demonstrated by a large-scale field survey in conjunction with a model experiment.

1. Role of acid-tolerant arbuscular mycorrhizal fungi in plant acid tolerance

M. sinensis seedlings were grown at pH 3.2 and 5.2 in the presence of either P-fertilizer, an acid-sensitive fungus *Claroideoglossum etunicatum*, or an acid-tolerant fungus *Rhizophagus* sp. RF1. At pH 5.2, the growth and shoot P concentration of the plants were significantly increased both by P-fertilizer and the mycorrhizal fungi

irrespective of their sensitivity to soil acidity. Whereas, at pH 3.2, only the plants associated with the acid-tolerant fungus could grow sustainably. The plants grown at pH 3.2 showed more aluminum-deposition on the root tips, more damaged root tips, and shorter root length than those grown at pH 5.2, and no difference in the root damages among the treatments was observed. These observations suggested that the acid-tolerant AM fungus could confer the extra acid-tolerance on the host plants beyond the adaptability of the plant to acidic soil via providing an alternative pathway for nutrient uptake, and not via alleviating the root damages caused by aluminum.

2. Community structure of arbuscular mycorrhizal fungi along soil pH gradients

Six field sites that covered a soil pH range of 3.0–7.4 were chosen across Japan, and rhizosphere soils of the pioneer grass *M. sinensis* grown in the sites were collected. *M. sinensis* seedlings were grown on the soils in a greenhouse, and the community compositions of AM fungi in the roots were determined based on the sequences of fungal large subunit ribosomal RNA gene. Overall, about 2,700 clones were sequenced, and 52 AM fungal phylotypes (sequence types) were defined. AM fungal richness decreased significantly with increases in soil acidity. Ordination analysis showed that soil pH is a major driver of the communities, but geographic isolation and climatic factors were not. Nestedness analysis indicated that the communities in lower-pH soils were nested within those in higher-pH soils, implying that the fungi that occurred in acidic soil were not specific to acidic soil, but were commonly distributed from acidic to neutral soils. These observations were further confirmed by subsequent model experiment. The AM fungal communities collected from neutral and acidic soils were grown at three different pH levels in association with *M. sinensis* seedlings. In the community from the neutral soil, fungal richness decreased with increases in soil acidity, and the communities grown at lower pH were nested within that grown at highest pH. In contrast, the community from the acidic soil was unresponsive to the changes in pH. All these results suggest that soil acidity is a major constraint on AM fungal richness and acid-tolerant fungi are the generalists that occur over wide ranges of pH.

The present study provides new insight of the functional and ecological significance of arbuscular mycorrhizal fungi in acidic soil and is expected to contribute to restoration of acidic soil.

Therefore, we acknowledge that the author is qualified to be granted the Degree of Doctor of Philosophy in Agriculture from Hokkaido University.