Title
Difference in Acquisition of Soil Organic Nitrogen between Pak-Choi and Tomato

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Introduction

Effective utilization of organic matter is strongly desired to promote sustainable agriculture. Whereas it has been well known that growth responses to organic matter differ among plant species, the factors affecting these characteristics has not been elucidated. The most part of organic nitrogen (N) in organic matter is a proteaceous compound (Kumar et al., 2006). In soils, proteins are degraded by protease, and the released amino acids and peptides are mineralized to ammonium (NH_4) and nitrate (NO_3), which can be utilized by plant roots. Here, we propose two hypotheses accounting for the factors responsible for the differences in plant response to organic N. The first hypothesis is that higher protease activity in rhizosphere contributes to the ability to utilize organic N in soils. The rate-limiting step in degradation of organic nitrogen in soils is considered to be a protease-mediated process. Plant species with high ability to utilize organic N may make their rhizosphere environment more suitable for protease-producing microorganisms, or may secrete protease from roots, resulting in the enhancement of proteolysis in rhizosphere soils. The second hypothesis is that the differences in ability to absorb amino acids are responsible for the different characteristics of organic N utilization. Recently, it has been shown that many plant species can absorb amino acids as significant N-sources for their growth (Streeter et al., 2000). Plant species with high ability to absorb amino acids is considered to be superior characteristics when competing with rhizosphere microorganisms for N acquisition. In this study, plant utilization of organic N is comprehensively discussed by testing these hypotheses.

Materials and Methods

Pot experiment

Seedlings of pak-choi (*Brassica rapa* L. var. chinensis) and tomato (*Solanum lycopersicum* L.) were grown in pots in a greenhouse. Each pot contained 300 mL of vermiculite mixed with field soils, as sources of microbe inoculums (vermiculite : field soil = 4 : 1, v/v). Field soils were collected from Hokkaido Central Agricultural Experiment Station, Naganuma, Japan. For 10 years, 25000 kg of manure and 2000 kg of rice bran, and 1070 kg of fish meal ha^{-1} $year^{-1}$ have been applied to the field. Different sources of N (no N (–N), ammonium sulfate (AS), and cattle farmyard manure (CM)) were applied to each soil for the N treatment (70 mg N pot^{-1} in AS and CM treatments).

After 28-days treatment, seedlings and soils (rhizosphere and non-rhizosphere soils) were sampled. Plant N concentrations were determined. Soil protease activities were determined according to Ladd and Bulter (1972), using benzyloxycarbonyl phenylalanyl leucine (ZFL) as a substrate.

Determination of amino acid uptake rate

Excised roots of pak-choi and tomato were incubated in 10 mL of 0.2 mM $CaCl_2$ solution (pH5.2) containing different sources of nitrogen (glycine (Gly), glutamic acid (Glu), NO_3^- and NH_4^+) for 2 hours at 25 ^oC or 4 ^oC. N concentration in the solution before and after the incubation was determined and the decrease in the N concentration was calculated. Active uptake rate for each N source was estimated by subtracting the rate at 4 ^oC (passive N uptake) from that at 25 ^oC.

Results and Discussion

Plant growth was the best in the AS treatment in both species (Fig. 1). In tomato, no significant difference in growth was found between the –N and CM treatments. In contrast, the growth of pak-choi was significantly enhanced in the CM treatment in comparison with the –N treatment. N accumulation in the plants showed the same tendency with plant growth (data not shown). These results suggest that pak-choi can utilize organic N in soils more efficiently than tomato.

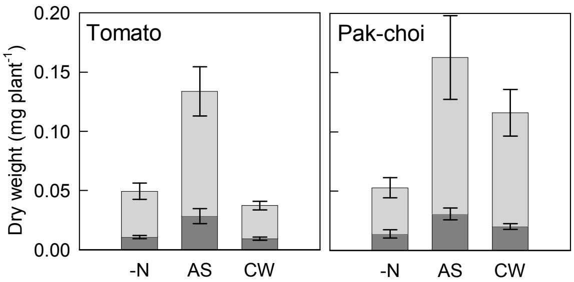

Fig. 1. Dry weight of tomato and pak-choi seedlings after the treatment. –N, no N application; AS, ammonium sulfate; CM, cattle farmyard manure.

As described in introduction section, protease activity in rhizosphere soils is one of the key factors that contribute to organic N acquisition in plant. In the rhizosphere soils, the highest activities were detected in the CM treatment irrespective of plant species (data not shown), which suggested that the organic matter application increased the activities. Although rhizosphere effect (Badalucco et al., 1996) was observed for the protease activities when comparing the activities between rhizosphere and bulk soils, there was no difference in this effect between two species (data not shown). Then, the correlation between rhizospheric protease

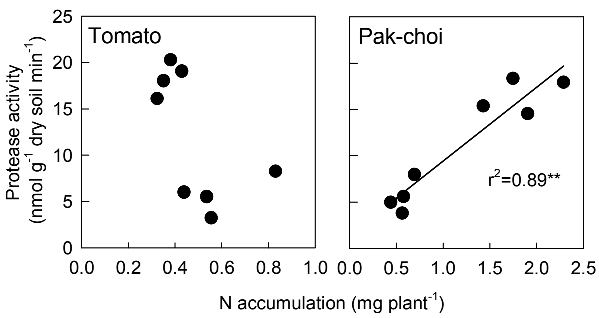

Fig. 2. Relationship between N accumulation and rhizospheric protease activity in tomato and pak-choi seedlings. **, significant at P<0.01

activity and plant total N accumulation was determined using individual replicates in both species grown with the –N and CM treatments (Fig. 2). A significant positive correlation was found only in pak-choi (r^2=0.89, P<0.01). These results suggest that the rhizospheric protease activity is a potential factor contributing to organic N acquisition in plant but other factors are likely to be involved in the mechanisms.

It is well known that plant roots compete with soil microbes for N acquisition in rhizosphere. It can be an advantage to take up amino acid N more efficiently to defeat rhizosphere microbes in N acquisition. Therefore, plant ability to absorb amino acid N may also be responsible for organic N acquisition in plant. As expected, Gly, which is one of the major free amino acid in soils (Streeter et al., 2000), was taken up at significantly higher rate in excised roots of pak-choi than of tomato (Table 1).

Table 1. Net uptake rate (μmol g^{-1} root FW h^{-1}) of NO_3^-, NH_4^+, Gly, and Glu in excised roots of tomato and pak-choi seedlings.

	NO_3^-	NH_4^+	Gly	Glu
Tomato	0.51 ±0.16	1.37 ±0.37	2.08 ±0.31	2.15 ±0.45
Pak-choi	0.12 ±0.07	0.70 ±0.42	5.55 ±1.76	1.95 ±1.02

In conclusion, this study strongly suggests that organic N acquisition in plant depends on at least the following two factors; protease activity in rhizosphere and ability to absorb amino acids. It would be interesting to characterize the microbiota in rhizosphere from the point of view of protease production and/or competition in amino acid acquisition.

References

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