



Title	Studies of properties of microorganisms in bulk and rhizosphere soils following the application of cover crops [an abstract of dissertation and a summary of dissertation review]
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## 学 位 論 文 内 容 の 要 旨

博士（環境科学）

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## 学 位 論 文 題 名

Studies of properties of microorganisms in bulk and rhizosphere soils following the application of cover crops

（緑肥施用土壌の根域および根圏における微生物特性に関する研究）

Cover crops, such as rye (*Secale cereale* L.) and hairy vetch (*Vicia villosa* Roth, HV), are commonly applied in crop production with multifunction. One of the functions is supplying inorganic nitrogen (N), the available N for subsequent plants. The supply is abundant during the decomposition phase of cover crop residues, within a month, but the utilization by the subsequent plants in the periods is limitedly performed. In bulk soil, bacteria and fungi play roles in the inorganic N supply from cover crop residues. However, the roles during the decomposition periods are barely known. In interaction with the subsequent plant, microorganisms inhabiting plant rhizosphere soils can assist N utilization (N uptake,  $N_{up}$ ) affecting plant yield. The plant–microbe interactions in the subsequent plant rhizosphere soil during cover crop decomposition is still unidentified yet. Therefore, to understand the effects of cover crops on horticultural systems, this study was carried out (1) to clarify N availability and its utilization by the subsequent plant, (2) to identify microorganisms contributing to the N availability in the bulk soil, and (3) to evaluate the plant–microbe interactions in the rhizosphere soil of subsequent plant cultivated during the decomposition periods of cover crop residues.

The two years (i.e., 2017 and 2018) of field and pot experiments were carried out. Treatments were soil without any cover crops (control) and soils with rye, HV, and mixed (rye+HV) cover crops. In the field, two ammonium sulfate application rates (i.e., 0 and 2.5 g N m<sup>-2</sup> in 2017 and 0 and 6 g N m<sup>-2</sup> in 2018) were applied to compare the effects of cover crops and synthetic N fertilizer. The effects of cover crops were clarified in the pot. Red leaf lettuce (*Lactuca sativa* L. var. *crispa* cv. red fire), the subsequent plant, was transplanted at 5 days after incorporation (DAI) of cover crops and harvested at a mature stage in the field and at the mid growth and mature stages in the pot to evaluate lettuce yield and  $N_{up}$ . Bulk and rhizosphere soils were collected

during the experiments (i.e., 5–38 DAI in 2017 and 3–31 DAI in 2018). The bulk soils were analyzed for concentration of soil inorganic N (i.e.,  $\text{NO}_3^-$ -N+ $\text{NH}_4^+$ -N), activity of the  $\beta$ -glucosidase enzyme (BG), and carbon-based soil microbial biomass (SMB). Bulk and rhizosphere soils from the pot were subjected to DNA-based molecular analysis to quantify and identify bacteria and fungi. Influenced microbial taxa, whose the relative abundance was affected by cover crops, were selected; and the relative abundance was correlated with values of BG activity and SMB or lettuce yield and  $N_{\text{up}}$ .

The N availability as concentration of soil inorganic N was (from high to low)  $\text{HV} > \text{rye}+\text{HV} > \text{rye} = \text{control}$ . This clarifies the common effects of the cover crops on N availability. In the field,  $\text{HV}-0\text{N}$  and  $\text{rye}+\text{HV}-0\text{N}$  promoted lettuce yield and  $N_{\text{up}}$  over  $\text{control}-0\text{N}$ . Conversely, lettuce performances in rye depended on 6N additional fertilizer. It indicates that HV and  $\text{rye}+\text{HV}$  could alternate application of synthetic N fertilizer. In the pot, however, HV suppressed plant yield and  $N_{\text{up}}$  in 2017 but promoted it in 2018 at the mid growth stage because HV effect was initial soil carbon dependent. Specific analysis of rhizosphere soil showed that plant–microbe interactions were interrupted as significant negative correlations between bacterial dominant taxa and yield and  $N_{\text{up}}$  in HV in 2017. On the contrary, vigorous lettuce growth in HV in 2018 showed that the roots recruited more beneficial microorganisms in the rhizosphere that had positively direct and indirect roles in yield and  $N_{\text{up}}$  promotions.

In bulk soil, bacterial and fungal DNA was relatively higher in cover crop-treated soils than in control within 5–10 DAI, concomitant with active N mineralization periods, up to 15 DAI, of cover crops. Further, microorganisms in bulk soil sequentially promoted BG activity and SMB during the decomposition periods, implying BG activity and SMB as the indicators of microbial roles in N availability. Specifically, family *Parachlamydiaceae* and unidentified bacteria of class SAR202 were positively in HV and  $\text{rye}+\text{HV}$  or negatively in rye correlated with BG activity and SMB. This indicates specific microbial roles in N availability from each cover crop.

This study demonstrated the technique to maximize the utilization of N supplied from cover crops during the residual decomposition periods. The technique can be adapted in the cover crop-horticulture rotation system. Furthermore, the microbial pathways of N supply and N utilization were molecularly revealed. In cover cropping system, thus, cover crops can be selected based on the N benefit and microbial feature.