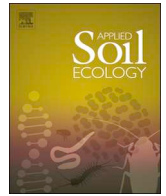




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Diazotroph abundance and community structure are reshaped by straw return and mineral fertilizer in rice-rice-green manure rotation

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ABSTRACT

The nitrogen (N)-fixing community in soil, a key functional player to replenish N pools from atmospheric N₂, can be radically changed by the addition of inorganic or organic substrate; however, the response of the diazotroph population to nutrient and residue management in green manure-based wetland rice rotations is poorly understood. Here, we investigated the diazotrophic community to inputs of a leguminous green manure Chinese milk vetch (*Mv*, *Astragalus sinicus*) using quantitative PCR and Illumina Miseq sequencing of the *nifH* gene. Five treatments were compared in a Milk vetch–early rice–late rice rotation: 1) Control, no fertilization and straw return (CK); 2) rice straw incorporation alone (Rs); 3) inorganic fertilizer application alone (F); 4) F plus Rs (FRs); 5) similar with FRs but high-stubble (~35–40 cm) of late rice straw was remained (FRh). The results showed that cultivation practice affected the magnitude of soil available nutrient pools (N, P and K), but not the soil organic matter and total N pools. The rice straw significantly repressed the *nifH* gene abundance compared to the control, and increased the number of diazotrophic bacteria species, Chao 1 values, Shannon and Simpson indexes more than other fertilization treatments. Multivariate regression tree analysis revealed that the community diversity and structure of diazotrophs were primarily shaped by soil nitrate and available P status, as well as C/N ratio. The most abundant genus *Bradyrhizobium* (21%–32%) tended to decrease in rice straw soil in comparison with the control but was significantly enhanced in the FRs treatment at seedling stage and in FRh treatment at flowering stage. Spearman correlation analysis showed that the dominant diazotrophic genera were positively related with soil available phosphorus, but responded differentially to soil total N, nitrate, and pH between the seedling and flowering stages. Overall, the planting and incorporation of vetch under different practices of residue and fertilizer management reshaped the diazotrophic community during the green manure season, highlighting the crucial roles of soil C, N, and P status or their ratios in shaping the population and diversity of diazotrophs.

1. Introduction

The excessive application of mineral fertilizer nitrogen (N) in agriculture has caused severe environmental issues (Le et al., 2010; Liu et al., 2013; Gao et al., 2018a). A major challenge for the sustainable production of rice is to maintain grain yield but with lower

environmental impacts (Chen et al., 2014). Substitution of the mineral fertilizer by organic substrates, such as straw and green manure is considered a promising way of reducing the chemical fertilizer input (Singh et al., 2005; Xie et al., 2016). Legumes are favored crops cultivated in the rice-green manure rotation systems because of their ability to fix N by biological fixation, and enhance N use efficiency and rice

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