

Diversity and activity of nitrogen fixing archaea and bacteria associated with micro-environments of wetland rice

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Wetland rice is one of the world's most important crop plants. The cultivation on waterlogged paddy soils is strongly limited by nitrogen (N), which is typically supplied by industrial fertilizers that are not only costly but also exhibit hazardous effects on the environment. It has been reported that "Biological Nitrogen Fixation" through N₂-fixing bacteria and archaea (diazotrophs) can alleviate the N-shortage in rice cultivation, thus carrying out an important ecosystem function. However, our understanding of the diversity and in situ N₂ fixation activity of diazotrophs in flooded rice fields is still rudimentary. Moreover, knowledge on the impact of biochemical gradients established by root activity (i.e. exudation, radial oxygen loss) on the functioning of N-fixing microorganisms in paddy soil ecosystems is limited.

We aimed at studying underlying processes on biologically relevant scales. Greenhouse studies were performed to identify key factors that control rice-diazotroph association and related N₂ fixation activities. Paddy soils of different geographical origin were cultivated with two commercially used genotypes of wetland rice. Samples were separated into bulk soil, rhizosphere soil, rhizoplane, and roots at flowering stage of rice plant development. These samples were subjected to functional assays and various molecular biological techniques in order to analyze the associated diazotroph communities.

Based on Illumina amplicon sequencing of *nifH* genes and transcripts, we show that the diversity and potential activity of diazotroph communities varies according to micro-environments. We will comparatively discuss the influence of (a) the soil microbial "seed bank" and (b) plant genotype in shaping the respective microbiomes and selecting for potentially active diazotrophs. Actual N₂ fixation activities of soil-genotype combinations and micro-environments will be shown on the basis of incubation assays using ¹⁵N₂-containing atmospheres. Areas of potential N-transfer between diazotrophs and rice roots will be presented via the detection and visualization of spatial colonization patterns of selected diazotrophic groups on rice rhizoplanes. Our approach will help to increase the understanding of the contribution of Biological Nitrogen Fixation to rice cultivation in paddy soil ecosystems.