Geophysical Research Abstracts
Vol. 17, EGU2015-15251, 2015
EGU General Assembly 2015

Tracking microbial colonization patterns associated with micro-environments of rice

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The interface between soil and roots (i.e. the rhizosphere) represents a highly dynamic micro-environment for microbial populations. Root-derived compounds are released into the rhizosphere and may attract, stimulate, or inhibit native soil microorganisms. Microbes associated with the rhizosphere, in turn, may have deleterious, neutral, or promoting effects on the plant. Such influences of microbial populations on the plant and vice versa are likely to be greatest in close vicinity to the root surface. It is therefore essential to detect and visualize preferential micro-sites of microbial root colonization to identify potential areas of microbe-plant interaction.

We present a single-cell based approach allowing for the localization, quantification, and visualization of native microbial populations in the rhizosphere and on the rhizoplane of soil-grown roots in situ. Catalyzed reporter deposition fluorescence in situ hybridization (CARD-FISH) in combination with confocal laser scanning microscopy was applied to observe colonization densities and patterns of microbial populations associated with wetland rice. Hybridizations with domain- and phylum-specific oligonucleotide probes showed that the growth stage of the rice plant as well as the distance to the root surface had a strong influence on microbial colonization patterns. Three-dimensional visualizations of root-associated microbes revealed micro-sites of preferential colonization. Highest cell numbers of archaea and bacteria were found at flowering stage of rice plant development. Irregular distribution patterns of microbiota observed at early growth stages shifted towards more uniform colonization with plant age. Accordingly, the highest colonization densities shifted from the tip to more mature regions of rice roots. Methanogenic archaea and methanotrophic bacteria were found to be co-localized at basal regions of lateral roots. Beneficial effects of a close association with root surfaces were indicated by proportionally higher numbers of methane-oxidizing bacteria on the rhizoplane compared to the rhizosphere. Such spatial effects could not be observed for methanogenic archaea.

As a consequence, the detection and visualization of microbial colonization patterns on a micro-scale via CARD-FISH represents an instrumental approach in revealing potential sites of interaction between microbes and plants in soil micro-environments.