

## **Endo-hyphal microbiome: a core of bacteria associated with the nitrogen cycle**

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**Project Goals: The interactions that occur in the rhizosphere between fungi and bacteria are fundamental for the ecological network of the soil. Over the past few years, scientist have described the diversity of microbial communities in a variety of soils associated with plants, yet little is known about the specific ecological significance and how these interactions are established. Bridging this knowledge gap could address multiple DOE priorities, as the effect of these interactions can have positive repercussions on the reduction of chemical fertilizers and increment of plant resistance towards abiotic and biotic stresses. Here we present preliminary results on the composition and possible function that these fungal-bacterial interactions could have on the soil ecosystem. Based on the isolation and identification of possible fungal-bacterial association from the soil, and combining bioinformatics and molecular tools, we are on the verge of understanding the importance of these associations.**

Over the past several years, the scientific community has described the diversity of microbial communities in a variety of soils associated with plants, but at present, little is known about the specific diversity of the soil fungal microbiome, involving bacteria colonizing the surface of fungi (i.e., exo-bacteria) or existing within fungal hypha (i.e., endobacteria). This study aimed to collect, identify, and characterize several fungi and their associated (endo- and exo-) microbiome collected from the rhizosphere of several plants. Microcosm devices called fungal highway columns, containing one of four plant-based media as attractants, were placed in the rhizosphere of six different plants. The isolated fungi and their associated endo- and exo- bacteria were identified by sequencing of the ITS (fungi) or 16S (bacteria) rRNA regions, followed by Scanning Electron Microscope (SEM) and fluorescence in situ hybridization (FISH) imaging. Most of the fungi recovered are known plant pathogens, such as *Fusarium*, *Pleosporales*, and *Cladosporium* together with species associated with the soil, e.g. *Kalmusia*. The bacteria recovered were previously described as plant promoters, such as *Bacillus*, *Rhizobium*, *Acinetobacter* or *Ensifer*. The interactions between fungi and exo-bacteria recovered from any single medium were further investigated via confrontation assays. From the reconstruction of the potential co-occurring bacterial-fungal association in the rhizosphere, we discovered that the most promiscuous exo-

bacterium group (associated with diverse fungi) was *Bacillus*, which presented either neutral or negative interactions with different fungi. Furthermore, some endobacteria and exo-bacteria identified seemed to be host-specific. These findings suggest a complex interaction between fungi and bacteria in the rhizosphere.

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