

## Context Dependent Mycorrhizal Resource Exchange in Bioenergy Cropping Systems

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**Project Goals: The LLNL Bioenergy SFA seeks to support sustainable and predictable bioenergy crop production through a community systems biology understanding of microbial consortia that are closely associated with bioenergy-relevant crops. We focus on host-microbial interactions in algal ponds and perennial grasses, with the goal of understanding and predicting the system-scale consequences of these interactions for biomass productivity and robustness, the balance of resources, and the functionality of surrounding microbial communities. Our approach integrates ‘omics measurements with quantitative isotope tracing, characterization of metabolites and biophysical factors, genome-enabled metabolic modeling, and trait-based representations of complex multi-trophic biological communities, to characterize the microscale impacts of single cells on system scale processes.**

Mutualistic associations between plants and mycorrhizal fungi can enhance plant productivity, resilience to stress, and carbon (C) allocation belowground. A better understanding of mycorrhizal relationships can inform more sustainable management of cellulosic bioenergy crops, such as switchgrass (*Panicum virgatum* L.), a C4 perennial grass championed for its high biomass yields and tolerance to a broad spectrum of climatic conditions and soils unsuitable for intensive agriculture. We are investigating context-dependent resource exchange between *Panicum hallii*—a model species closely related to switchgrass—and two mycorrhizal fungi: the arbuscular mycorrhizal (AM) fungus *Rhizophagus irregularis* and the sebacinoid mycorrhizal fungus *Serendipita bescii*. Both fungi have been found growing in association with a wide range of plant species, including switchgrass and several other bioenergy crops. Due to differences in their genomic repertoires, we hypothesize that each fungus confers plant benefits and ecosystem services through unique mechanisms.

We grew *P. hallii* with and without *R. irregularis* and *S. bescii* in microcosms containing ‘live’ soil harvested from a marginal Oklahoma pasture. We restricted the soil moisture in half of the microcosms in order to assess plant and mycorrhizal response to water limitation. Additionally, half of the microcosms were grown in a <sup>12</sup>CO<sub>2</sub> atmosphere and half in a <sup>13</sup>CO<sub>2</sub> atmosphere. The microcosms were harvested destructively at 5, 8, and 12 weeks after the onset of <sup>13</sup>CO<sub>2</sub> labeling.

This approach allows us to track plant- and mycorrhizal-derived  $^{13}\text{C}$  into other microbial taxa, soil C pools, and C fluxes ( $\text{CO}_2$ , volatiles, dissolved organic C). We used root and hyphal exclusion chambers to examine biogeochemical fluxes and multipartite microbial interactions in spatially distinct ecological niches.

Although AM and sebacinoid fungi were identified in the native soil microbial community, qPCR analyses show that the *R. irregularis* and *S. bescii* strains used as inoculants were more abundant in roots and soils harvested from inoculated microcosms. Overall, total plant biomass was slightly higher in microcosms inoculated with *R. irregularis* than in uninoculated microcosms and those inoculated with *S. bescii*. The relationship between mycorrhizal inoculation and total plant biomass diminished over time under water-limited conditions, but not under water-replete conditions. Additionally, the ratio of shoot:root biomass was higher in plants grown under water-replete conditions—particularly those inoculated with *S. bescii*. Isotopic analyses and NanoSIMS imaging show that plants allocated a substantial quantity of C to their mycorrhizal partners and other C pools. After 12 weeks of growth in a  $^{13}\text{CO}_2$  atmosphere, recently photosynthesized  $^{13}\text{C}$  accounted for up to 10% of rhizosphere soil C ( $0.4 \text{ mg } ^{13}\text{C excess g}^{-1} \text{ soil}$ ) and 0.7% of hyphosphere soil C. Although much of this plant- and mycorrhizal-derived C was retained below ground, some returned to the atmosphere as  $\text{CO}_2$ . We are using quantitative stable isotope probing (qSIP) to assess taxon-specific microbial response to mycorrhizal presence and soil moisture availability. Our results shed light upon the complex and dynamic nature of plant-microbe interactions and their potential role in sustainable bioenergy crop production.

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