Understanding Variation in the Switchgrass Microbiome Across Scales: Evidence for Both Host Filtering and Environmental Control

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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.

Switchgrass (*Panicum virgatum*) is a promising biofuel species that can be cultivated across a large geographic range including marginal lands with low inputs. Recent work demonstrates that components of the switchgrass microbiome can substantially modify plant physiology, growth, and stress tolerance, which can impact both plant production and ecosystem function. However, a key remaining gap in understanding the role of the switchgrass microbiome in plant success is how the microbiome varies across sites, and what controls that variation. To address this, we characterized fungal communities in switchgrass soils, roots, and leaves at 14 sites ranging from the coast to the mountains of North Carolina. The sites varied in climate, soil properties, and management (monoculture, mixed-grass, or tree-intercropping). The samples were sequenced (ITS rRNA) on Illumina MiSeq. We analyzed how plant size, site characteristics, and climate factors affected fungal community structure.

There was significant site-level variation in switchgrass-associated soil, root, and leaf fungi, which was largely explained by local conditions. Fungal community composition in all samples was primarily affected by soil texture, which ranged from very sandy (86% sand) to very clayey (37% clay) across sites. In soils, fungi also varied with pH, P, Mg, and MAT. In contrast, root and leaf fungi were most strongly affected by plant size and soil DOC. Despite shared environmental drivers, there was little overlap in fungal identity between roots and leaves, which shared only 17% of fungal taxa. Soils shared <10% of fungal taxa with roots and <1% with leaves, suggesting that the plant is filtering some taxa from the available pool as well as being colonized by taxa from other sources. Only roots and soil fungi were affected by site management, which mainly reflected the 5x greater SOM in sites with intercropped trees.

Our findings highlight that soil and plant characteristics are associated with divergence in switchgrass microbiomes across landscapes, and that switchgrass-associated fungi are not a

simple reflection of the taxa found in soils. Because we also isolated ~2000 fungi representing ~400 taxa from roots and leaves at these sites, this work also facilitates the next step to relating function to composition across the landscape.