

Plant-microbe and microbe-microbe interactions mediate switchgrass sustainability: following rhizosphere microbial communities during switchgrass establishment

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Project Goals: Switchgrass (SG; *Panicum virgatum* L.) is a perennial grass native to the tallgrass prairie and one of the most promising bioenergy crops in the U.S., with potential to provide high-yield biomass on marginal soils unsuitable for traditional agricultural crops. A persistent concern for bioenergy cultivation of SG with low-input management, is improving seedling establishment and resistance to abiotic and biotic stresses. We hypothesize that successful establishment and sustainable cultivation of SG in marginal soils is in part enabled by beneficial plant-microbial interactions. We are investigating the succession of rhizosphere microbial communities associated with high- and low-performing SG plants grown in nutrient-limited soils at two Oklahoma field sites. The outcome of this research will provide a better genomic basis for SG cultivation in marginal soils, and will expand our knowledge of the interactions between soil microbiomes, plants and ecosystems.

In the soils surrounding roots (rhizosphere), biotic, chemical and physical drivers enrich for specific bacterial and fungal communities. These organisms play multiple roles, and some may benefit plant productivity via help with nutrient acquisition, water uptake and pathogen suppression. We are investigating the composition, function and succession of rhizosphere microbial communities during plant cultivation in an effort to better understand the plant-microbiome interactions that enable plant survival and adaptation under stressed conditions, such as drought and nutrient deficiency.

To study the establishment phase plant-soil microbiome characteristics of SG growing in 'marginal' nutrient or water-limited soils, we selected two Noble Foundation research farms, both remnants of the Dust Bowl Era in Oklahoma. One, Red River Farm has sandy loam soil low in NO₃-N and organic matter; the other, Third Street Farm, has a silt loam soil with relatively low phosphorus availability. Five hundred Alamo AP13 non-clonal seedlings were planted into each field in May-June of 2016. Other than hand weeding during the summer, no management or water/ nutrients was supplied to the fields. Thirty plants were randomly selected for multi- time point monitoring of rhizosphere and bulk soil samples to study community succession during SG establishment. These same plants were non-destructively sampled over the first growing season at 5 time points: early and late vegetative growth, reproductive growth, maximal growth, and senescence. To test the association between rhizosphere communities and SG growth, plant growth parameters such as plant height, tiller numbers and flowering time were measured; biomass yield was assessed at the end of growth season. Within each plot plant growth was

highly variable, with between 20-143 tillers per plant. Overall, plants at the Red River site had better growth and higher biomass yield than at Third Street.

To better understand factors influencing the establishment and functional properties of rhizosphere microbial communities, we conducted amplicon sequencing of marker genes specific to bacteria, fungi and soil eukaryotes, and monitored physio-chemical characteristics of soil beneath targeted plants, including gravimetric moisture, pH, and NO_3/NH_4 . We are structuring our analyses to assess differences in environmental drivers, community assembly, and succession patterns in both high- and low- performing plants on our two soils of different nutrient status. A subset of samples of particular interest will be characterized using shotgun metagenomics and GeoChip hybridization to further characterize the microbial functional properties.

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