Dissecting the role(s) of host genotype and phytobiome composition and function in the successful establishment of switchgrass (*Panicum virgatum* L) on marginal soils.

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Project Goals: (1) Identify high- and low-performing switchgrass (SG) genotypes in marginal soils and determine the functional succession of SG-associated microbial communities during successful SG establishment of each. (2) Characterize plant–microbe and microbe–microbe interactions within and between SG and its microbiome, particularly when challenged by water or nutrient stress. (3) Determine how low-input SG production in marginal soils may enhance ecosystem sustainability metrics such as: C storage, nutrient availability, and soil food webs. (4) Integrate and synthesize experimental data to reveal plant-microbe interactions and the underlying mechanisms critical to SG effects on ecosystem sustainability.

To jump start research aimed at achieving the ambitious goals listed above, *PI Saha* has identified high and low biomass genotypes from fifteen families of a switchgrass nested association mapping (NAM) population (see figure 1). The NAM population was derived by crossing fifteen diverse parents, selected from a diversity panel pre-screened for certain useful agronomic traits like early vigor and yield, to a recurrent parent AP13 (draft genome, microarray and other genomic resources available). Subsequently, 10 F_1s from each of 15 families were chain crossed to develop a NAM population of 2,000 genotypes (see figure 2). The population has been evaluated in field experiments at two locations for two years. Soil quality for these trials had adequate levels of nitrogen, phosphorus and organic matter. Based on overall performance across environments, 80 high yielding and 20 low yielding genotypes were identified. Clonal ramets are being generated for 4-6 high-yielding genotypes in the greenhouse for initial testing on soils poor in nitrogen, phosphorus, or both. Based on these results, a single high-yielding genotype (on these marginal soils) will be chosen for subsequent analyses.

To dissect the role of the microbiome in facilitating switchgrass growth on marginal soils, seedlings will be planted at 2-3 Oklahoma sites with soils characterized as nutritionally depleted of organic matter. *PI Craven* will collect bulk soil for baseline microbial metagenomic analysis, and subsequently follow the succession of switchgrass rhizosphere microbial and microfaunal communities associated with establishment-phase plants that are high and low performers. Rhizospheric soil and root tissues will also be collected from each for the isolation of bacterial and fungal endophytes. We have established high-throughput screens for many potential useful microbial traits, including N-fixation, ACC deaminase activity, and solubilization of inorganic phosphorus (CaPO4). Bacterial or fungal strains found to contain one or more will be candidates for SIP tracer studies and simplified community modeling and analysis.

To elucidate differences in nutrient use efficiency (NUE), flux and utilization, *PIs Scheible* and *Udvardi* will carry out physiological, transcriptomic, metabolomics and fluxomic studies on plants grown under optimal or limiting nutrient conditions. Switchgrass growth experiments using plants grown from Alamo seed have been initiated using sand to define nutritional

parameters for subsequent studies. Transcriptomic and metabolomic studies will reveal genes and processes that are involved in acclimation to nutrient limitation, and which are conserved or not between switchgrass genotypes or plant species more broadly. This will help to clarify how comparable switchgrass is to other species, and will also help to identify molecular and genetic markers for subsequent analyses. A qPCR platform will be developed to determine expression of SG homologs to known P and N-signaling, -metabolism and -status indicator genes, including microRNAs. Transcriptomic and metabolomic studies of plants devoid of microbes will serve as a base-line for comparison of plants associated with microbes, with the objective of identifying nutritional and other services provided by microbes to plants, and vice versa.

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