

## **Title: Integration of Experimental and Modeling Approaches to Understand, Predict, and Modulate Rhizosphere Processes for Improved Bioenergy Crop Productivity**

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**Project Goals: Short statement of goals.** This project couples novel lab and field studies to develop the first predictive model of grass-microbiomes based on new mechanistic insights into dynamic plant-microbe interactions in the grasses *Sorghum bicolor* and *Brachypodium distachyon*. The results will be used to predict plant mutants and microbial amendments, which improve low-input biomass production and validate these predictions in lab and field studies. We will determine the mechanistic basis of dynamic exudate exchange in the grass rhizosphere. A specific focus will be on the identification of plant transporters and proteins that regulate root exudate composition and how specific exudates select for beneficial microbes that increase plant biomass and nitrogen use efficiency (NUE). To do this we will use stable isotope dilution mass spectrometry methods to quantify nitrogen (N) flow between plants and microbes and use plant mutants to identify plant genes that influence exudation. Furthermore, we will develop two predictive genome-scale rhizosphere models (*S. bicolor* and *B. distachyon*) that are grounded in lab and field data. These models will be used to predict plant growth and exudate metabolism to maximize N flux to the plant as a function of light and N availability over a diurnal cycle. Finally, we will use these models to design genetic and microbial interventions that are tailored to increase plant biomass production under N limited conditions by optimizing plant-microbe interactions. We will also design and test the effects of amendments added at different times of the day and night to increase plant productivity by altering the microbiome to alleviate N limitation. The most favorable and robust approaches will be tested under marginal field conditions.

**Abstract text:** To avoid competition with food crops and maximize their economic and environmental benefits, bioenergy crops should be grown on marginal soils with minimal inputs, especially inputs of energy intensive synthetic N fertilizer. Thus, bioenergy crops are more reliant on microbial-driven organic N for nutrition than food crops. Root exudates are thought to play a critical role in recruiting and maintaining beneficial microbes including those that make N available to the plant. Up to 25% of all photosynthetically fixed carbon is released into the rhizosphere through root exudates. These exudates provide a critical source of nutrition for diverse microorganisms and are composed of many compounds including organic acids, amino acids, and mono- and polysaccharides. However, it is unclear if this exchange of exudates for plant benefits holds during the night, when the plants are not photosynthesizing. Our preliminary data suggest that exudate composition shows strong circadian rhythms. Unraveling the nature of plant and microbial drivers behind these dynamic interactions is crucial for understanding the dynamics of N cycling and uptake in the rhizosphere. Predictive models are necessary to understand the complexity of these processes and optimize sustainable bioenergy systems. We hypothesize that exudate and bacterial community engineering will enable structuring of rhizosphere communities that enhance N supply and/or reduce microbial competition for organic N thereby increasing plant NUE.

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