Title: Leaf Carbon and Nitrogen Isotope Composition in Diverse Sorghum Lines Under Differential Water and Nitrogen Treatments

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Project Goals:

The *overall project goal* is to establish a foundational, systems-level understanding of plant, microbial, and environmental interactions that will lead to strategies for enhancing growth and sustainability of sorghum through genetic and microbial adaptations to water and nitrogen limited environments.

The ***specific objectives*** of the research presented here are to:

1. Conduct phenotypic characterizations of a diverse panel of sorghum genotypes to define photosynthetic and isotope response under drought and low nitrogen conditions.
2. Test sorghum genotype by environment interactions in both controlled environment and field growth conditions.
3. Determine if measurements of leaf carbon and nitrogen isotope composition can screen for differences in water and nitrogen use efficiency in diverse sorghum genotypes.

Towards achieving our project goals we have screened 30 diverse sorghum lines under controlled environment growth conditions in the Bellweather Phenotyping System at the Danforth Center. This population included 18 energy, 2 grain and 10 sweet sorghum lines. Under the phenotyping system two separate experiments were conducted using a random block design to study the growth, photosynthetic and stable isotope response of this diverse sorghum panel to changes in nitrogen and water availability. Additionally, whole plant nitrogen and water use efficiency were estimated from the phenotyping data. In the first experiment each line was provided with two levels of nitrogen (n=3 to 4 plants per line per treatment) over a three week time period. In the second experiment, each line received a constant nutrient supply under three different watering treatments over a three-week growth period (n=3 to 4 per line per treatment). Individual plants were imaged daily at 4 different angular rotations to calculate plant growth and approximate biomass accumulation. Additionally, weighing and watering of plants was automated to maintain the required soil volumetric water content and the predetermined nutrient supply. Towards the end of both experiments the upper most fully expanded leaf from individual plants was used for gas exchange measurements with a LI-6400XT open gas exchange system (Li-COR Biosciences, Inc. Lincoln, NE). A portion of the same leaf was sampled for nitrogen and carbon isotope composition ($\delta^{15}N$ and $\delta^{13}C$, respectively). Leaf samples for $\delta^{15}N$ and $\delta^{13}C$ analysis were also collected from two field locations in Nebraska

in both 2016 and 2017. The western Nebraska field site was used to study response to drought conditions and the eastern location was used to characterize response to low nitrogen.

Initial characterization of these data sets suggests significant variation between the sorghum genotypes in their responses to nitrogen and water availability. For the controlled environment phenotyping experiment data will be presented on differences in nitrogen and water use efficiency, rates of CO_2 assimilation, stomatal conductance, intrinsic transpiration efficiency (TE_i) and photosynthetic nitrogen use efficiency (PNUE). Measurements of total leaf C/N content and leaf $\delta^{13}C$ and $\delta^{15}N$ will be presented from both the controlled environment and field experiments.

Future directions

Leaf level and whole plant traits will be assessed across genotypes in response to both changes in nitrogen and water availability. This information will be analyzed in comparison to field grown material to help identify and select for genomic traits and potentially elite lines for enhanced nitrogen and water use efficiency in sorghum.

Funding statement

This project is funded by the DOE BER Sustainable Bioenergy Research Program, Award DE-SC0014395, and was also supported by DOE JGI Community Science Program.