

Investigation of Carbonic Anhydrase to Improve Sorghum C₄ Photosynthetic Efficiency

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Project Goal: Leverage *Setaria viridis* as a model system to develop novel technologies and methodologies to redesign the bioenergy feedstock *Sorghum bicolor* to enhance water use and photosynthetic efficiencies.

Abstract:

One objective of this project is to optimize the expression of carbonic anhydrase (CA) to increase the residence time of carbon dioxide in the leaf to improve C₄ photosynthesis and drought resilience. Phylogenetic and gene expression analysis of CA across the grasses demonstrates that there are several α , β , γ - CA genes present in all grass genomes that exhibit a conserved expression in leaf tissue regardless of photosynthetic type. We are working to understand the role of alternative splicing of the most abundant leaf expressed β -CA, with the working hypothesis that two transcript isoforms facilitate CA being localized to different sub-cellular locations. On-going experiments include investigation of CA protein targeting and biochemical analysis of the alternatively spliced protein products. Results suggest that translation of the most abundant C₄ CA transcript does not begin in exon 1 as previously annotated. These results provide insights for designing constructs to optimize CA expression in Sorghum. Constructs that are currently being made for Sorghum include a β -CA overexpression construct and a transcriptional activator, dCas9-TV, for all β -CA isoform promoters with 1-2 sgRNAs per isoform. Further research on CA promoter elements that drive elevated and cell-specific gene expression, will help refine strategies for editing CA promoters to improve photosynthesis in Sorghum.

Funding Statement: This project is funded by grant DE-SC0018277 from The DOE Department of Biological and Environmental Research