

## Improving Photosynthesis in C<sub>4</sub> Bioenergy Crops

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### Project Goals

**The Renewable Oil Generated with Ultra-productive Energycane (ROGUE) project aims to engineer the two most productive American biofuel crops, energycane and *Miscanthus*, to produce a sustainable supply of biodiesel, biojet fuel and bioproducts. The three main objectives of this work are:**

- 1) To improve the conversion of sunlight into plant biomass/metabolites through photosynthesis without the need for increased quantities of either water, or fertilizer.**
- 2) To transfer ROGUE technologies from the lab bench to crops through an efficient pipeline.**
- 3) To test technologies through replicated field trials.**

### Abstract

Many important C<sub>4</sub> bioenergy crops, such as maize, sorghum and sugarcane, are tropical, warm-season forage grasses with limited productivity under chilling conditions, as compared to temperate C<sub>4</sub> grasses, e.g. *Miscanthus × giganteus*, *Panicum virgatum* and *Spartina pectinata* (1). This is due to the limited amount of pyruvate orthophosphate dikinase (PPDK) and rubisco which restrain phosphoenolpyruvate (PEP) regeneration (1–3). In order to expand potential growing seasons and increase the range of latitudes where biofuel crops can grow, two main hypotheses are formed to increase photosynthetic ability of these crops under different conditions. We hypothesize that the upregulation PPDK in C<sub>4</sub> crops by engineering PPDK native gene from closely-related species, e.g. sorghum, elevates photosynthesis and biomass. This will be tested by introducing a synthesized native Sorghum bicolor PPDK gene into *Miscanthus* and energycane to determine the level of photosynthetic improvement in the greenhouse and by field trials. We also plan to modulate the level of PPDK regulatory protein (PDRP) which controls posttranslational modifications of the rate-limiting PPDK under different light conditions (4–6) increases PPDK activity in plants. We will first perform the in silico modelling to understand the potential impact of modulating PDRP activity, then assemble the transgenic construct followed by transient expression of the gene in protoplasts of bioenergy crops, and finally perform physiological assessments in resulting transgenic crops.

## References

1. Long SP, Spence AK (2013) Toward Cool C<sub>4</sub> Crops. *Annu Rev Plant Biol* 64(1):701–722.
2. Wang D, Naidu SL, Portis AR, Moose SP, Long SP (2008) Can the cold tolerance of C<sub>4</sub> photosynthesis in *Miscanthus x giganteus* relative to *Zea mays* be explained by differences in activities and thermal properties of Rubisco? *J Exp Bot* 59(7):1779–1787.
3. Naidu SL, Moose SP, Al-Shoaibi AK, Raines CA, Long SP (2003) Cold Tolerance of C<sub>4</sub> photosynthesis in *Miscanthus x giganteus*: Adaptation in Amounts and Sequence of C<sub>4</sub> Photosynthetic Enzymes. *Plant Physiol* 132(July):1688–1697.
4. Ohta S, Ishida Y, Usami S (2006) High-level expression of cold-tolerant pyruvate, orthophosphate dikinase from a genomic clone with site-directed mutations in transgenic maize. *Mol Breed* 18(1):29–38.
5. Chastain CJ, et al. (2011) Functional evolution of C<sub>4</sub> pyruvate, orthophosphate dikinase. *J Exp Bot* 62(9):3083–3091.
6. Burnell JN, Chastain CJ (2006) Cloning and expression of maize-leaf pyruvate, Pi dikinase regulatory protein gene. *Biochem Biophys Res Commun* 345(2):675–680.

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