

A Systems Approach to Enhancing Seedling Establishment for Increased Yields in the Oilseed Crop, *Camelina sativa*

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Project Goals:

Our research plan aims to establish the non-food oilseed crop plant, *Camelina sativa*, as a commercially viable, dedicated biofuel and bioproduct feedstock. We are focused on improving seed and oil yields by employing an integrated genetic and metabolic systems approach to increase the rates of photosynthetic CO₂ capture and conversion to triacylglycerols (TAGs). *Camelina* has the advantages of low agronomic inputs and natural resistance to biotic and abiotic stresses relative to other oilseed crops, and *Camelina* oil-based blends have been tested and approved as liquid transportation fuels. *Camelina* also benefits as a synthetic biology platform from a fully sequenced genome, well-established molecular genetic tools, and numerous resources available from its close relative, *Arabidopsis thaliana*. Despite these advantages, the major limitation in widespread adoption of *Camelina* as an industrial oilseed crop is its modest oil yield. Our project will address yield directly by employing a tissue-specific and whole-plant systems approach to identify the major regulatory mechanisms that limit 1) **carbon fixation in photosynthetically active source tissues (leaves)**, 2) **the transport of fixed carbon from source to sink tissues (seeds)**, and 3) **the allocation of fixed carbon to TAG production**. The limiting factors identified in our analyses will be individually validated and combined using multi-gene stacking and genome editing technologies to engineer *Camelina*.

Abstract:

Varying environmental conditions, including changes in temperature, light, water and nutrient availability, result in energy and metabolic imbalances that significantly impact crop productivity and agricultural production. Metabolic interactions between chloroplasts and mitochondria, including photorespiratory metabolism, are known to be essential in mitigating these imbalances, and strategies to enhance the plasticity of these networks have the potential to enhance crop resilience to environmental stress. We previously described improved carbon assimilation, increased water and nitrogen use efficiency, and higher seed yields in *Camelina* by expressing LIP36, a mitochondrial carrier that promotes oxaloacetate/malate exchange. Metabolic systems analysis demonstrated that LIP36 transport activity (1) enhances the non-cyclic phase of the mitochondrial tricarboxylic acid (TCA) pathway in photosynthetic tissues, and (2) promotes redox exchange between the mitochondria and cytosol. In doing so, LIP36 contributes to cellular redox balance and optimal photorespiratory metabolism, while favoring anabolic over respiratory metabolism. This results in increased stress resilience and improved seed yields under limiting environmental conditions.

In the current study, we took advantage of LIP36 tissue-specific expression to examine the role of mitochondrial metabolic networks in optimizing respiratory, anabolic, and redox metabolism in early seedling development, root growth/architecture, and seed development. We will present combined metabolic, physiological and transcript profiling data that demonstrate a positive impact of LIP36 expression on early seedling establishment. This includes increased shoot growth and root elongation within the first 10 days following germination. Metabolic and transcript analysis demonstrate that the primary impact is on cellular redox balance. Our data suggest that the ability to increase the plasticity of redox exchange between the cytoplasm and mitochondria, may decrease ROS generation and alter the expression of important stress resilience pathways that increase robust seedling establishment. Enhancing seedling establishment is a key agronomic factor for improving the stand and the ultimate yields of crops grown on marginal lands. We anticipate that this trait could contribute to increased adoption of Camelina as an industrial oilseed crop.

This work is funded by grant #DE-SC0018269 from the Department of Energy BER program.