

Genomics and Phenomics to Identify Yield and Drought Tolerance Alleles for Improvement of Camelina as a Biofuel Crop

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Project Goals: Plant oils represent renewable sources of energy-dense hydrocarbons that can be used for biofuel, but a major challenge is to produce these oils in high-yielding non-food crops that can grow under marginal conditions. Our goal is to improve the suitability of camelina as a bioenergy crop. The major objectives are to: 1) Develop and apply automated, non-destructive high-throughput phenotyping (HTP) protocols to evaluate the phenotypic diversity of a camelina panel consisting of 250 accessions, grown under well-watered and water-limited conditions; 2) Discover alleles/genes controlling morphological, physiological, seed, and oil yield traits using genome-wide association studies; and 3) Identify, test, and validate useful germplasm under diverse environments and marginal production areas. These studies will enable the discovery of new genes associated with crop yield and stress tolerance and identify high-yielding cultivars that are suitable for certain geographical regions.

In recent years, *Camelina sativa* has received considerable attention as a potential non-food biofuel crop, but significant challenges remain to develop stable, high-yielding, geographically adapted germplasm suitable for biofuels production. Here, we will use advanced high-throughput phenotyping and genomics-based approaches to discover useful gene/alleles controlling seed yield and oil content and quality in camelina under water-limited conditions, and identify high-yielding cultivars suitable for production in different geographical regions. Our experimental design includes complimentary field- and greenhouse-based HTP experiments conducted on a panel of 250 camelina accessions grown under well-watered and water-limited conditions. In year 1 of the study, the entire panel was analyzed using the greenhouse-based LemnaTec system available at the Donald Danforth Plant Science Center. The greenhouse-based system was used to monitor the growth of two replicates of each accession under well-watered and water-limited conditions from 7 to 35 days after planting. Phenotype data for each plant at each time point was derived from the approximately 200,000 images produced during the experiment using the PlantCV software package. Seeds were collected from each plant for further analysis.

In preparation for the field-based phenotyping experiments to be conducted in Maricopa, AZ and Sidney, NE, the entire population was planted in Maricopa during year 1 for multiplication of seed. Pilot studies were also conducted at each location with 10 selected varieties that showed genotypic and phenotypic diversity. In Nebraska, the ten selected varieties were planted in 5 replicates. The control was irrigated with approximately 1" per week and the treatment was not irrigated after stand establishment. The decreases in yield due to the treatment ranged from 26% to 1%, demonstrating good treatment effects and genotypic differences in the camelina lines

tested. A new field phenotyping cart for camelina plots with multiple sensing modules was constructed. Phenotyping was conducted at two time points. Initial analysis of the canopy temperature data showed distinct differences in the canopy temperatures between treatments towards the end of the growing season. In-depth analysis of 2017 phenomics/yield data for the ten selected varieties is ongoing. These preliminary trials helped us to establish the methods that will be used in 2018 and 2019 field trials to screen the 250 accessions of camelina for tolerance to drought based on yield reductions and phenomics data.

In Maricopa, the ten camelina varieties were planted in 4-row plots, three replicates each, under well-irrigated and reduced irrigation conditions. A high-clearance tractor was equipped with proximal sensors and imagers to collect field-based HTP data including crop height, canopy temperature and canopy multi-spectral reflectance. The HTP data were collected on a weekly basis throughout the growing season. The reflectance data were used to construct vegetation indexes. In addition to field-based HTP measurements, traditional morphological and physiological traits were collected and analyzed. Raw data were processed to remove outliers then the cleaned data sets were analyzed using analysis of variances and correlation analyses. The results showed that there were temporal associations between vegetation indexes and camelina growth stages. The indexes reached the highest values around flowering time then decreased in value at later growth stages when chlorophyll was degraded due to leaf senescence and desiccation. There were significant genotypic variations among tested varieties for measured indexes. Significant associations between HTP-measured traits and traditional morphological and physiological traits were observed and indicated the feasibility of using field-based HTP related traits to predict and measure traits controlling camelina plant structure, growth, and response to environmental conditions.

Collectively, these studies will serve as a foundation for future experiments that aim to characterize the phenotypes of the entire camelina panel planted under well-watered and water-limited conditions in the field. Genome-wide association studies will then be employed to identify genes and genetic markers associated with agronomically important traits (Goal 2). Comparison of results between field- and greenhouse-grown plants will also help determine how well the greenhouse-based studies translate to findings in the field. The results will also help identify a subset of plant lines that will be analyzed in an advanced yield trials at four different locations, including Maricopa, AZ, High Plains, NE, Quincy, FL, and Morris, MN (Goal 3).

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