Developing Non-food Grade *Brassica* Biofuel Feedstock Cultivars with High Yield, Oil Content, and Oil Quality that are Suitable for Low Input Production Dryland Systems.

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**Project Goals:** (1) Develop superior non-food grade oilseed cultivars suitable for a range of PNW and other US environments with blackleg and abiotic stress resistance suitable for high-quality biofuel feedstocks; (2) Dissect genetic architecture of industrial *Brassica* oilseed germplasm for yield, quality, and resistance to blackleg fungus through a genome-wide association study with high-density genetic markers; (3) Develop and apply marker-assisted genomic selection techniques for blackleg resistance that will accelerate development of disease resistant oilseed cultivars; and (4) Use bioinformatics to identify putative PRR resistance genes that confer durable resistance to blackleg and use transgenic approaches to introgress PRR genes into adapted cultivar background.

**Abstract:** This project utilizes rapeseed (*Brassica napus*) and Indian mustard (*B. juncea*) oilseed germplasm and the long-term industrial oilseed breeding program at the University of Idaho, and the agronomy and molecular biology expertise from the University of Idaho and Washington State University. We have recently completed the first full growing season of the project.

Two hundred and forty spring *B. napus* genotypes were planted under field condition in spring of 2017. All these accessions had previously been genotyped, and were planted in replicated trials into ground with a proven history of blackleg. Plots were evaluated throughout the growing season for blackleg disease lesions on leaves. Few diseases symptoms were observed due to drier than usual weather post-planting. In addition, 20 spring *B. napus* lines were planted at two locations in replicated field trials where blackleg was controlled (via two foliar fungicide applications) and without fungicide. Blackleg lesions were recorded throughout the growing season. At maturity, seed was harvested and stem canker recorded, although few incidences were noted.

In fall of 2017, 280 winter *B. napus* genotypes were planted under field condition at two locations in northern Idaho (Grangeville, 4 replicates with smaller plots, and Genesee, two replicates with larger plots to allow combine harvest at maturity). All these accessions had previously been genotyped, and were planted in replicated trials into ground with a proven history of blackleg disease. To date, we have recorded blackleg leaf lesions in fall of 2017 and we will repeat lesion counts on two occasions in spring of 2018. Blackleg has always appeared a greater problem in winter *B. napus* than in spring plantings and significantly greater lesions counts were observed and markedly different resistance levels were found within the genotypes screened. At harvest, we will record yield from the Genesee trial which has larger plot size compared to Grangeville. All of these genotypes were included in an earlier project were we recorded seed yield. However, in that study we had severe winterkill in two of the four years and so yield data on the winter types was limiting. The 2017-2018 plots look good at the time of reporting, and the additional yield data will be invaluable to add to our data set and allow greater genotype by association studies.

In addition, 20 winter *B. napus* lines were planted at Grangeville ID, in replicated field trials where blackleg was controlled (via two foliar fungicide applications) and without fungicide. Blackleg lesions were recorded in fall of 2018 and we will repeat lesion counts on two occasions in spring of 2018. At maturity, seed will be harvested and stem canker recorded, although few
incidences were noted. This trial will help us understand the potential yield loss caused by blackleg disease in Pacific Northwest winter B. napus cultivars without fungicide control or genetic plant resistance to blackleg.

A genetically diverse genotypic collection of B. juncea had been collected at the University of Idaho. This diversity panel included Plant Introduction accessions (268 lines) from USDA, along with germplasm from within the University of Idaho B. juncea breeding program (232 lines). To ensure, continuity between genotyping and phenotyping, seed from each of the 500 genotypes was planted in the glasshouse in fall of 2016. A single plant was harvested from each accession and that seed thereafter used to plant the first field evaluation trials in spring of 2017. While the selected plants were at the seedling stage in the glasshouse a leaf sample was taken from each genotype and freeze dried for genotyping. In spring of 2017 the glasshouse seed increase produced sufficient seed to allow planting of a replicated yield trial at one location (Genesee) and a seed increase plot at another location (Moscow). A wide range of plant morphological data was recorded on plots from the replicated yield trial including, crop establishment, flower date, plant height, lodging, pod length, seeds pod−1, and maturity. Any blackleg lesions observed were counted, and other disease and insect damage recorded. At crop maturity, seed was harvested and yield recorded. A sample of seed was taken from each plot and is currently being evaluated for oil content, oil quality, and seed mean quality.

Pattern Recognition Receptor (PRR) mediated resistance is dependent on perception of pathogen-associated molecular pattern (PAMP) by pattern recognition receptor (PRR) in plants. In addition, PRR-mediated resistance is potentially durable because PAMPs, as structural elements of molecules, are highly conserved and usually essential for liability or lifestyle of pathogens; and because pathogens are less likely to evade host resistance through mutation or deletion of PAMPs, compared with virulence effectors. We have been successful in identifying several putative PRR genes and these are being further investigated, before we begin transferring these PRR genes into the regionally adapted B. napus and B. juncea oilseed cultivars being selected in the breeding program. In addition we plan to pyramid multiple PRR genes into a single winter B. napus line by multiple crossing among transgenic plants expressing individual PRR genes.

Progress results from all research to date will be discussed in the context of the work that is to follow. The genetically superior Brassica non-food grade oilseed cultivars developed will drastically increase domestic industrial oilseed production. In addition, we will identify new and novel plant resistance genes for blackleg resistance and molecular marker assisted selection tools to accelerate plant breeding procedures. Increased domestic production of biofuel feedstock oil will reduce dependence and importation of fossil fuels, increase fuel security, and ensure US agricultural competitiveness with foreign countries.

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