

## Genetics and Genomics of Pathogen Resistance in Switchgrass

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**Project Goals: This project wants to establish switchgrass as a key component of the bioenergy industry in the US by developing cultivars with the right suite of genes for high biomass, ethanol yield, and with good quality profiles and durable resistance to diseases.**

**The specific objectives are:**

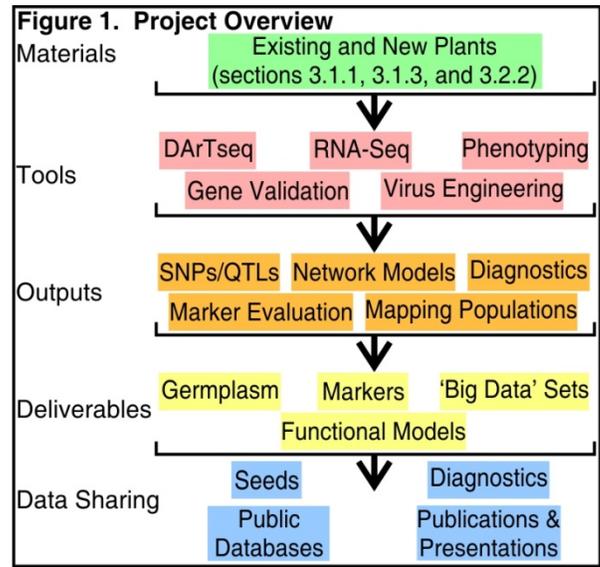
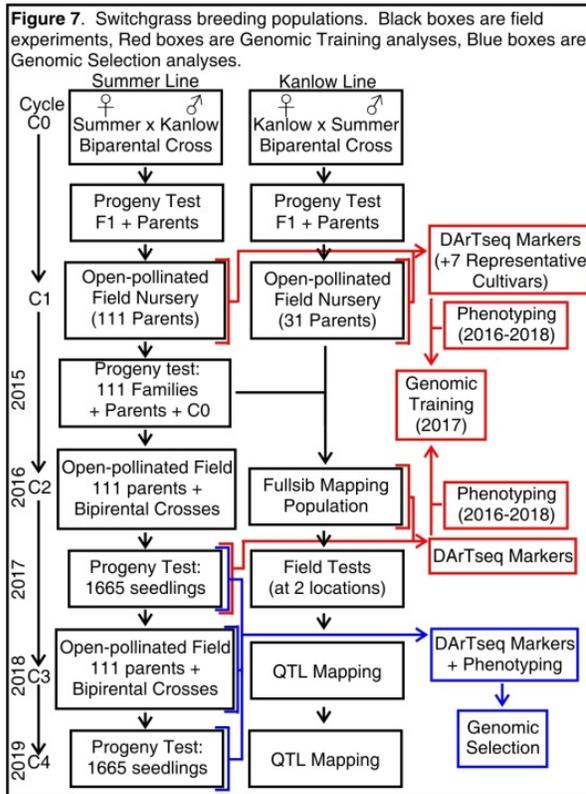
- (1) Understand the genetic and genomic bases of pathogen response in regionally-adapted upland and lowland switchgrass breeding populations with contrasting disease symptoms under field conditions,**
- (2) Dissect the molecular underpinnings of the broad resistance or tolerance to pathogens in ‘Kanlow’ vs ‘Summer’, and**
- (3) Discover the molecular differences that permit systemic viral infections in some switchgrass plants, but not in other genetically-related plants.**

Switchgrass (*Panicum virgatum*) is susceptible to a number of fungal (*Puccinia emaculata* and *Uromyces graminicola*) and viral (*Panicum mosaic virus*) pathogens, making yield stability and biofuel output under biotic stress important selection criteria for biofuel feedstock improvement. This project leverages the differential reactions to challenges from the rust and viral pathogens of a composite tetraploid switchgrass population derived from crossing Kanlow (lowland) with Summer (upland) ecotypes, a population already under improvement for bioenergy-related traits. Three successive generations (parents and offspring) will be measured phenotypically for quantitative traits (biomass yield, rust, and cell wall components) and genomically with molecular SNP (single-nucleotide polymorphisms) markers derived from DArT-seq (see Figure). Prediction models will be developed from these assessments based on quantitative (BLUP, animal model, breeding values, and index selection) and molecular genetics (QTL mapping, genomic selection) methodologies and combined to deploy the best aggregate “phenotype-genotype”.

The USDA-ARS and UNL labs in Lincoln, NE, are highly experienced with breeding perennial grasses like switchgrass since the 1930s and have developed and published robust NIRS (near-infrared spectroscopy) calibration equations for quality (SCW) traits. Breeding methodologies are being continually refined and optimized to maximize gains from selection. Selection is practiced in two stages, first with selection of the best 25-30% of tested families from which the top 10% of individuals are chosen as parents of the next generation. Recombinations are carried out by open-pollination which will be augmented with poly- and biparental crosses to capitalize on all types of gene actions (ie additive and dominance, more specifically) and maximize genetic gains. A pedigree will be established to follow the flow of genes from parents to progeny across generations and to increase the efficiency of prediction models by including the pedigree and molecular relationship matrices. The two rust pathogens are prominent in field trials established at the UNL Agricultural Research and Development Center (located near Mead, NE) and intense

natural disease pressures prevail yearly to allow discrimination of tolerant or resistant from susceptible genotypes and segregation of phenotypes/genotypes with all combinations of high/low yield and high/low lignin. Ratings will be carried out on a 1-5 scale and taken twice during the crop cycle. This discrimination will facilitate QTL mapping in a derived population by crossing susceptible with tolerant parents in order to define the regions governing rust, biomass yield, and SCW quality traits. QTL mapping combined with genomic selection will ensure identification of genes with major and minor effects and establish the foundation for better refinement of those genes.

The functional transcriptional networks, underpinning the broad rust and viral disease resistance or tolerance in the Kanlow population, will be studied. RNAseq data will be collected to interrogate the transcriptional changes underlying defense responses in switchgrass, based on the overall and specific differences in gene expression in Kanlow and Summer. These studies will be augmented by engineering viral genomes with fluorescent markers to visualize viral movement and pathogenesis in plants, developing antibodies to coat proteins to detect virus in field-grown samples, and by using recombinantly-expressed viral proteins to identify interacting plant proteins that are crucial for the spread of infection.



*This research project is supported by the DOE Office of Science, Office of Biological and Environmental Research (BER), grant no. DE-SC0016108.*