Project FY22-DU-004: Genetic Characterization and Introgression of FHB Resistance in Durum Wheat

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1. What are the major goals and objectives of the research project?

The overall goal of this project is to genetically characterize FHB resistance and transfer the resistance into durum wheat. The specific objectives of the research are:

1. Finely map the 2A QTL for FHB resistance derived from the Divide X PI 254188 cross.
2. Develop user-friendly DNA markers for the 2A QTL.
3. Introgress the 2A QTL into durum wheat varieties.
4. Screen EMS mutants derived from ND Riverland and Kronos for FHB resistance.
5. Identify genes related to FHB susceptibility in durum wheat.

2. What was accomplished under these goals or objectives? *(For each major goal/objective, address these three items below.)*

What were the major activities?

71 EMS mutant lines showing less susceptibility to FHB compared to the wildtype Kronos were re-evaluated twice in the greenhouse and we confirmed that 11 out 71 consistently showed a high level of FHB resistance with an average disease severity below 33%.

We identified the mutated genes in those 11 FHB resistant mutant lines derived from Kronos. Those genes that were mutated in four or more FHB resistant mutant lines but not changed in susceptible mutant lines are considered to be related to FHB susceptibility and selected as targets for FHB resistance improvement in durum wheat using the wheat x maize-mediated gene editing technology established in our lab.

We re-evaluate the most susceptible and most resistant individual plants generated by EMS method from the durum wheat line (ND Riveland-Fhb1) carrying the Fhb1 gene. The disease data will be used to select the most resistant and susceptible mutants which will be sequenced in the future to identify the candidate gene suppressing the Fhb1 resistance gene in the Durum genetic background.

We have successfully developed the genome editing system using the wide hybridization between wheat varieties and maize plants expressing Cas9 enzyme and single guide RNA (sgRNA) and five wheat genes related to disease susceptibility or resistance have been edited in various wheat genotypes. We have generated double haploid seeds from most of the mutants. Some of the mutants have been phenotyped for the certain disease.

We also test the established genome editing system on the Durum wheat and we successfully generated the targeted mutation of *TaHRC1* gene in the durum wheat variety Divide.

What were the significant results?

11 EMS mutants developed from Kronos were identified to consistently showed a high level of FHB resistance.

10 protentional candidate genes mutated in resistant mutants but not in susceptible mutants have been identified and are considered to be related to FHB susceptibility. Selected genes will be performed for gene expression analysis in durum wheat.

List key outcomes or other achievements.

A quick and effective genome editing approach without wheat transformation was developed for both spring wheat and durum wheat.

We could significantly increase the disease resistance to certain pathogen by editing genes related to disease susceptibility.

3. What opportunities for training and professional development has the project provided?

Three Ph.D. students are involved in this project and have obtained training in FHB screening, genotyping, EMS mutant generation, and genome editing through wide crosses between wheat and maize.

4. How have the results been disseminated to communities of interest?

Some of the research results were presented in conference presentations and peer-reviewed articles published in scientific journals.

5. What do you plan to do during the next reporting period to accomplish the goals and objectives?

Perform gene expression analysis of candidate genes selected for gene editing using real time RT-PCR.

Construct binary vectors expressing Cas9 and sgRNA for editing candidate genes for FHB susceptibility in durum wheat.

Transform vectors into corn to generate transgenic corn plants for wide hybridization and generate haploid plants from wide hybridization between durum wheat varieties.

Sequence some of the EMS mutants from ND Riverland-Fhb1 to identify the candidate genes suppressing the Fhb1 resistance in durum wheat.