PI: Cai, Xiwen | Agreement #:

Project FY22-DU-002: Introgression and Understanding of Hexaploid-derived FHB Resistance Genes in Durum

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

The major goals of this research project are to develop FHB-resistant durum wheat germplasm by alien introgression and provide a better understanding of the effects of the durum genetic background on FHB resistance. The specific objectives of this project are to: 1) Transfer the whole chromosome containing major FHB resistance QTL from hexaploid to adapted durum varieties by alien chromosome-mediated substitution; 2) Characterize inheritance and epistasis of the hexaploid-derived FHB resistance genes in the near-isogenic adapted durum backgrounds by targeted chromosome substitution and recombination; and 3) Select the targeted chromosome recombinants with FHB resistance in the adapted durum backgrounds for germplasm/variety development.

2. What was accomplished under these goals or objectives?

What were the major activities?

- 1) Made crosses to introduce common wheat chromosome 3D, 4D, and 6D into adapted durum cultivars 'Divide' and 'ND Riveland' by their substitutions for durum chromosomes 3B, 4B, and 6A using chromosome-specific markers. This will allow for introgression of chromosomes 3B, 4B, and 6A, which contain major FHB resistance QTL, from Sumai 3 and PI 277012 into Divide and ND Riveland.
- 2) Made crosses to transfer Sumai 3 chromosomes 3B and 6B and Wangshubai chromosome 4B into LDN durum using LDN DS 3D(3B), DS 6D(6B), and DS 4D(4B) and chromosome-specific markers.
- 3) Developed molecular markers specifically targeting the short and long arms of the chromosomes under wheat homoeologous group 3, 4, 5, and 6 for D-genome chromosome introgression into durum.
- 4) Performed marker-assisted introgression of the novel FHB resistance gene *Fhb7*^{The2} into Divide, ND Riveland, MTD18348, Miwok, and winter durum breeding lines.
- 5) Phenotyped and genotyped the Divide x PI 277012 RIL population (n=203) and performed QTL and epistatic analysis.
- 6) Developed and verified new Fhb7-specific STS and KASP markers for MAS in durum genotypes.

What were the significant results?

- 1) Developed bridging materials for the introduction of common wheat chromosome 3D, 4D, and 6D into adapted durum cultivars 'Divide' and 'ND Riveland'. This will allow for introgression of chromosomes 3B, 4B, and 6A, which contain major FHB resistance QTL, from Sumai 3 and PI 277012 into Divide and ND Riveland.
- 2) Generated progenies for the introgression of Sumai 3 chromosomes 3B and 6B and Wangshubai chromosome 4B into LDN durum using LDN DS 3D(3B), DS 6D(6B), and DS 4D(4B).
- 3) Developed 14 PCR allelic competitive extension (PACE) markers specifically targeting the short and long arms of the chromosomes under wheat homoeologous group 3, 4, 5, and 6. They have been used in the D-genome chromosome introgression into durum.

PI: Cai, Xiwen | Agreement #:

- 4) Incorporated the novel FHB resistance gene *Fhb7*^{The2} into Divide, ND Riveland, MTD18348, Miwok, and winter durum breeding lines through a marker-assisted backcrossing breeding pipeline.
- 5) Completed FHB phenotyping and genotyping of the Divide x PI 277012 RIL population (n=203) and performed QTL and epistatic analysis. Linkage maps of all 14 durum chromosomes were constructed in this RIL population. Seven FHB resistance QTL were identified on chromosome 5A, 4A, 2B, and 5B. Epistatic effects for FHB resistance were detected at the loci between A and B subgenomes in this population. FHB phenotyping of another RIL population involving PI 277012 and another durum line D00095 is in progress. Comparative analysis of these two RIL populations sharing the same resistance source in the different backgrounds will provide insight into the epistatic effect of this resistance source in durum.
- 6) Newly developed *Fhb7*^{The2}-specific STS and KASP markers for MAS in durum genotypes.

List key outcomes or other achievements.

- 1) Developed elite FHB-resistant durum breeding lines containing *Fhb7*^{The2}. They exhibited significant resistance in the FHB nurseries (Lincoln, NE and Fargo, ND) and greenhouse screenings. Undesired linkage drag has not been observed with *Fhb7*^{The2} in these introgression lines. They will be released to the FHB research and durum breeding community for variety development. This work will lead to an impactful breakthrough in durum FHB breeding and enable durum wheat to gain significant resistance to FHB.
- 2) Revealed the additive effect of *Fhb7*^{The2} and other FHB resistance genes.
- 3) Obtained additional phenotyping and genotyping data for further FHB resistance QTL and epistatic analysis in durum.
- 3. What opportunities for training and professional development has the project provided? One postdoc has been hired to work on this research project. This research project has offered him a great opportunity to learn the procedure and principles of FISH/GISH and chromosome-specific marker development from reference genomes and cDNA sequences. In addition, the postdoc has received training in genetic analysis, chromosome engineering, genomics, and bioinformatics. These learning and research experience have facilitated their career development in plant genetics and breeding.
- **4.** How have the results been disseminated to communities of interest?

 Research results from this project were presented in the 2024 FHB Forum and will be published in the peer-reviewed journal.

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

- Conducting backcrosses and marker/chromosome analyses to continue chromosome substitution-mediated FHB resistance gene introgression from hexaploid into durum for epistatic analysis and germplasm development.
- 2) Advancing generation of the two populations derived from the durum x common wheat crosses by single seed decent.
- 3) Making additional backcrosses and performing MAS for *Fhb7*^{The2} introgression into adapted spring and winter durum genotypes.
- 4) Performing FHB evaluation of the $Fhb7^{The2}$ introgression lines in the FHB nurseries in Lincoln, NE and Fargo, ND.

FY24 USDA-ARS/USWBSI Performance Progress Report

PI: Cai, Xiwen | Agreement #:

- 5) Evaluating another durum RIL population for FHB resistance with shared parent with the previous tested RIL population in the greenhouse.
- 6) Performing epistatic analysis in the RIL population(s) with phenotyping and genotyping data.
- 7) Increasing seed of FHB-resistant RILs to validate their resistance in the field conditions (Lincoln, NE and Fargo, ND).