Project 1: Identify and Map Novel QTL for FHB Resistance in Durum Wheat

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1. **What are the major goals and objectives of the research project?**
   The major goal of this project was to identify, map, and deploy QTLs for FHB resistance in the emmer wheat (*Triticum turgidum* L. subsp. *dicoccum*) line PI 254188. Therefore, the specific objectives are:

   1) Develop a mapping population with recombinant inbred lines (RILs) derived from the cross between Divide and PI 254188;
   2) Phenotype FHB resistance and morphological traits of the mapping population from the Divide/PI 254188 cross in greenhouse and field;
   3) Construct a genetic linkage map of the population using 90k-SNP chips;
   4) Identify DNA markers linked to QTL for FHB resistance in PI 254188;
   5) Transfer and pyramid the FHB resistance QTL into adapted durum wheat cultivars.

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

   a) What were the major activities?
   (1) Generated additional DNA markers for the major QTL on chromosome 2A identified in the mapping population derived from the cross between Divide and PI 254188.
   (2) Evaluated a mapping population consisting of 200 RILs (F2:7) derived from the cross between Joppa and one FHB resistant RIL (DPI-RIL-10), which was selected from the cross between Divide and PI 254188.
   (3) Developed near-isogenic lines with Fhb1 in the durum wheat cultivar ND Riveland.

   b) What were the significant results?

   (1) 22 STARP markers were developed in the QTL region of chromosome 2A identified in the mapping population derived from the cross between Divide and PI 254188.
   (2) In a greenhouse evaluation, the FHB severity ranged from 14.3% to 87.44% for the mapping population derived from the cross between Joppa and DPI-RIL-10. In the field inoculation experiment, the FHB severity ranged from 8.3% to 80.0% for the same mapping population.
   (3) A progeny derived from the cross between a durum wheat line and the spring wheat line ND2710 carrying Fhb1 was used as the donor of Fhb1 to cross with the durum wheat cultivar ND Riveland. Then, ND Rivaland was used as recurrent parent in multiple cycles of backcrosses and marker-assisted selection, and near-isogenic lines with Fhb1 were developed in the genetic background of ND Rivaland.

   c) List key outcomes or other achievements.

   (1) STARP markers for the 2A QTL were developed, which will be used for marker-assisted selection of the QTL during introgression of the FHB resistance into other durum wheat cultivars.

(2) FHB phenotyping data were collected from both greenhouse and field inoculation experiments for the mapping population derived from the cross between Joppa and DPI-RIL-10. These data will be used for fine mapping of the QTL for FHB resistance in the population.

(3) Near-isogenic lines of durum wheat cultivar ND Rivaland with Fhb1 were generated. These lines will be used directly in durum wheat breeding programs.

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

The project provided trainings to one research associate and two Ph.D. students on FHB phenotyping, QTL mapping, marker development and near-isogenic line creation. The PI and the participants of this project have attended two conferences.

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

Nothing to report

Project 2: Expand FHB Screening Capacity for Durum Wheat

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

The major goal and objective of this research project is to expand the FHB nursery at Fargo location and establish a new FHB nursery at Langdon location to accommodate and screen more advanced breeding lines, mapping populations, and introgression germplasm from durum wheat researchers working in the DUR-CP.

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

a) What were the major activities?

In the 2021 summer season, we planted 2,960 hill plots of durum wheat materials in the FHB nursery at the Fargo location for FHB evaluation. The durum wheat materials included advanced breeding lines, mapping populations, and introgression germplasm from four PIs (Steven Xu, Xuehui Li, Shaobin Zhong, and Elias Elias) in the durum wheat CP. One mapping population consisting of 660 hill plots were inoculated by the point inoculation method and the remaining 2,300 hill plots were inoculated by the Fusarium-infested corn inoculum spread on the field in mid-June. Overhead misting systems were used for disease development.

A new FHB nursery was established at Langdon for FHB screening in collaboration with Dr. Venkata Chapara at the NDSU Langdon Research Station, and 1,850 hill plots from two PIs (Xu and Zhong) were planted in the 2021 summer season. At this location, the Fusarium-infested corn inoculum and overhead misting systems were used for disease infection and development.

b) What were the significant results?

High quality disease data were collected from the mapping population which was point-inoculated at the Fargo nursery. These data are useful for QTL mapping of the FHB resistance in the mapping population. Due to extreme dry weather conditions, the disease level was low on the durum wheat materials inoculated by the Fusarium-infested corn inoculum.

c) List key outcomes or other achievements.

The FHB nursery at the Fargo location provided larger capacity for screening of durum wheat materials from PIs involved in the USWBSI program. The FHB nursery at Langdon provided another location for FHB evaluation.

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

The FHB nurseries provided trainings for four Ph.D. students in FHB phenotyping.

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

Nothing to report

Project 3: Genetic Characterization and Integrated Deployment of FHB Resistance in Spring Wheat

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

Our overall goal is to clone and characterize a major QTL for FHB resistance in PI 277012 and quickly deploy it along with another major FHB resistance QTL Fhb1 in the four spring wheat breeding programs of the Spring Wheat Coordinated Project (SPR-CP). The specific objectives of this project are:

1) Clone and characterize the major FHB resistance QTL Qfhb.rwg-5A.2 in the wheat line PI 277012.
2) Quickly introgress Qfhb.rwg-5A.2 and Fhb1 into elite spring wheat cultivars and breeding lines by backcrossing, marker-assisted selection and speed breeding approach.
3) Determine the expression of Qfhb.rwg-5A.2 and its interaction with Fhb1 in different genetic backgrounds.

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

a) What were the major activities?

(1) Conducted sequence analysis and gene annotation of the PI 277012 genome.
(2) Developed additional DNA markers for both QTL (*Qfhb.rwg5A.1* and *Qfhb.rwg5A.2*) identified in PI 277012.
(3) Made crosses between selected RILs with QTL for FHB resistance derived from PI 277012 and susceptible wheat genotypes (Dayn, WA8283, and Grandin) to generate recombinants.
(4) Screened additional EMS mutants generated from PI 277012 in the greenhouse.
(5) Continued to peform backcrossing, marker-assisted selection and speed breeding for introgression of Qfhb.rwg-5A.2 and Fhb1 into 11 elite spring wheat cultivars or breeding lines developed by spring wheat breeders at NDSU, University of Minnesota, SDSU, and WSU, respectively.

b) What were the significant results?

(1) Based on genotyping and phenotyping of selected recombinant lines, one scaffold (utg001995l, 9.8 Mb) was identified from genome assembly of PI 277012 sequenced with the PacBio HiFi sequencing technology.
(2) Two flanking makers and one peak marker were developed for the *Qfhb.rwg5A.1* region. A total of 88 STARP markers were developed from utg001995l for the *Qfhb.rwg5A.2* region. These markers will be used to identify the recombinants containing either or both of the QTL.

(3) 12 FHB susceptible EMS mutants were identified from screening approximately 510 M2 individuals derived from 217 and 57 EMS M1 plants of GP112 and GP226, two FHB resistant RILs derived from the cross between Grandin and PI 277012). These FHB susceptible EMS mutants will be used for validation of the candidate genes for *Qfhb.rwg5A.2*.

(4) Five RILs (GP288, GP512, GP646, GP693 and GP737) containing targeted region of *Qfhb.rwg5A.2* were selected for crossing with Dayn and F2 seeds were generated for identification of recombinants. Recombinants were identified from two crosses, GP112 X Dayn and GP112 X WA8283. Three recombinants carrying the *Qfhb.rwg5A.2* region only (no *Qfhb.rwg5A.1*) were selected from the cross between Dayn and GP112 for further backcrossing with Dayn. Two of them are less susceptible (disease severity was 41.5% and 51.67%, respectively) and one is susceptible (disease severity was 80.00%).

(5) By backcrossing, marker-assisted selection and speed breeding, Qfhb.rwg-5A.2 and Fhb1 were integressed into nine spring wheat cultivars. The backcross generations for each FHB resistance QTL are shown as follows:

Recipient cultivar or line	× GP112	× Alsen
Glenn	F1BC6	F1BC7
Alsen (with *Fhb1*)	F1BC6	-
ND VitPro	F1BC6	F1BC7
Linkert	F1BC6	F1BC7
Lang-MN (with *Fhb1*)	F1BC6	-
MN10201 (with *Fhb1*)	F1BC6	-
Surpass	F1BC6	F1BC7
WA8283	F1BC6	F1BC7
Dayn	F1BC6	F1BC7

c) List key outcomes or other achievements.

(1) High quality whole genome assembly of PI 277012 was generated with the HiFi sequencing technology and one 9.8 Mb scaffold covering the target QTL was identified. These sequence resources will facilitate the isolation of the candidate genes for *Qfhb.rwg5A.2*.

(2) New PCR-based DNA markers were developed for both *Qfhb.rwg5A.1* and *Qfhb.rwg5A.2.* These markers will be useful for transferring the two QTL into other wheat varieties by marker-assisted selection.

(3) Additional FHB susceptible EMS mutants were identified from RILs with *Qfhb.rwg5A.2* derived from PI 277012. These EMS mutants will be useful for validation of the candidate gene(s) to be cloned.

(4) The two major FHB resistance QTL (Qfhb.rwg-5A.2 and Fhb1) were introduced into various spring wheat cultivars and breeding lines by backcrossing, marker-assisted selection and speed breeding approach, and near-isogenic lines at F1BC6 or F1BC7 generations have been developed.

3. **What opportunities for training and professional development has the project provided?**
   The project provided one research associate and two Ph.D. students with training on genome analysis, QTL mapping and marker development as well as map-based gene cloning. The PI and the participants of this project attended two conferences.

4. **How have the results been disseminated to communities of interest?**
   The FHB resistant wheat lines developed and DNA markers associated with the FHB resistance have been provided to and used by other wheat researchers and breeders.

Project 4: Increase Capacity of Two Coordinated FHB Nurseries for Spring Wheat

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1. **What are the major goals and objectives of the research project?**
   The major goal and objective are to expand the FHB nursery at Fargo location and establish a new nursery at Langdon location to accommodate and screen more advanced breeding lines, mapping populations, and introgression germplasm from NDSU spring wheat breeding program and other researchers who are working in the spring wheat CP.

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

   a) **What were the major activities?**
      We planted spring wheat materials at both Fargo and Langdon locations for FHB screening in the 2021 summer season.
      At the Fargo location, we planted 3,992 hill plots of spring wheat materials, including advanced breeding lines, mapping populations, and introgression germplasm from three PIs (1,100 hill plots from Andrew Green, 1,200 hill plots from Steven Xu, and 1,692 hill plots from Shaobin Zhong), who are working in the spring wheat CP. Among the 3,992 hill plots, 1,692 hill plots were inoculated by the point inoculation method and 2,300 hill plots were inoculated by application of the Fusarium-infested corn inoculum in mid-June. Overhead misting systems were used for both inoculation methods for disease development.

      At the Langdon location, we planted 1,550 hill plots from two PIs (850 hill plots from Zhong and 700 hill plots from Xu) for FHB screening in collaboration with Dr. Venkata Chapara at the NDSU Langdon Research Station. The Fusarium-infested corn inoculum was applied at this location and overhead misting systems were used for FHB development.

   b) **What were the significant results?**
      Quality disease data were collected from the point-inoculated wheat materials at the Fargo nursery in 2021 summer season. These data are useful for FHB resistance identification, QTL mapping, and selection of advanced breeding lines for spring wheat cultivar development. Due to extreme dry weather conditions, the disease level was low on the spring wheat materials inoculated by the Fusarium-infested corn inoculum.

c) List key outcomes or other achievements.

The FHB nursery at the Fargo location provided larger capacity for screening of spring wheat materials from PIs involved in the USWBSI program. The FHB nursery at Langdon provided another location for FHB evaluation.

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

The FHB nurseries provided trainings for one research associate and four Ph.D. students in FHB phenotyping.

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

Nothing to report

Publications, Conference Papers, and Presentations

Please include a listing of all your publications/presentations about your FHB work that were a result of funding from your FY21 grant award. Only citations for publications published (submitted or accepted) or presentations presented during the **award period** should be included.

Did you publish/submit or present anything during this award period?

☒ Yes, I've included the citation reference in listing(s) below.
☐ No, I have nothing to report.

Journal publications as a result of FY21 grant award

List peer-reviewed articles or papers appearing in scientific, technical, or professional journals. Include any peer-reviewed publication in the periodically published proceedings of a scientific society, a conference, or the like.

Identify for each publication: Author(s); title; journal; volume: year; page numbers; status of publication (published [include DOI#]; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).

Poudel, B., Mullins, J., Puri, K.D., Leng, Y., Karmacharya A., Liu, Y., Hegstad, J., Li, X., and Zhong, S. 2022. Molecular mapping of quantitative trait loci for Fusarium head blight resistance in the Brazilian spring wheat cultivar 'Surpresa'. Front. Plant Sci. 12:778472. https://doi.org/10.3389/fpls.2021.778472.
Status of publication: published.
Acknowledgment of federal support: yes.

Books or other non-periodical, one-time publications as a result of FY21 grant award

Report any book, monograph, dissertation, abstract, or the like published as or in a separate publication, rather than a periodical or series. Include any significant publication in the proceedings of a one-time conference or in the report of a one-time study, commission, or the like.

Identify for each one-time publication: Author(s); title; editor; title of collection, if applicable; bibliographic information; year; type of publication (book, thesis or dissertation, other); status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).

None

Other publications, conference papers and presentations as a result of FY21 grant award

Identify any other publications, conference papers and/or presentations not reported above. Specify the status of the publication.

Wang, R., Axtman, J., Salsman, E., Hegstad, J., Fiedler, J., Xu, S., Zhong, S., Elias, E., and Li, X. 2021. Recurrent Selection to Develop Fusarium Head Blight Resistance Germplasm for Durum Wheat. In: Proceedings of the 2021 National Fusarium Head Blight Forum complied and edited by the USWBSI Networking & Facilitation Office (NFO), Virtual, December 6-7, 2021. Retrieved from: https://scabusa.org/forum/2021/2021NFHBForumProceedings.pdf

Zhang, W., Danilova, T., Zhang, M., Ren, S., Zhu, X., Zhang, Q., Zhong, S., Dykes, L., Fiedler, J., Xu, S., Boehm, J., Cai, X. 2021. A Diploid Tall Wheatgrass-Derived Fhb7 Allele Integrated into Wheat B Genome Conditions FHB Resistance in Wheat. In: *Proceedings of the 2021 National Fusarium Head Blight Forum complied and edited by the USWBSI Networking & Facilitation Office (NFO), Virtual, December 6-7, 2021. Retrieved from: https://scabusa.org/forum/2021/2021NFHBForumProceedings.pdf*

Zhong, S., Shrestha, S., Poudel, B., Karmacharya, A., Zhao, M., Leng, Y., Mullins, J., Chu, C., and Xu, S. 2021. Identification of QTL for Type I Resistance to Fusarium Head Blight in Two Spring Wheat Mapping Populations. In: In: Proceedings of the 2021 National Fusarium Head Blight Forum complied and edited by the USWBSI Networking & Facilitation Office (NFO), Virtual, December 6-7, 2021. Retrieved from: https://scabusa.org/forum/2021/2021NFHBForumProceedings.pdf