USDA-ARS | U.S. Wheat and Barley Scab Initiative

FY21 Performance Progress Report

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Cover Page

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USDA-ARS Agreement ID:	59-0206-0-160
USDA-ARS Agreement Title:	Transfer of FHB Resistance to NDSU Hard Red Winter Wheat Breeding Material
FY20 USDA-ARS Award Amount:	$58,437
Recipient Organization:	North Dakota State University Department of Plant Sciences Dept. 7670, PO Box 6050 Fargo, ND 58108-6050
DUNS Number:	80-388-2299
EIN:	45-6002439
Recipient Identifying Number or Account Number, if any:	FAR0031913
Project/Grant Period:	7/10/21 - 7/9/23
Reporting Period End Date:	7/9/2022

USWBSI Individual Project(s)

USWBSI Research Category*	Project Title	ARS Award Amount
HWW-CP	Transfer of FHB Resistance to NDSU Hard Red Winter Wheat Breeding Material	$58,437
	FY21 Total ARS Award Amount	$58,437

I am submitting this report as an: ☐ Annual Report ☒ Final Report

I certify to the best of my knowledge and belief that this report is correct and complete for performance of activities for the purposes set forth in the award documents.

GF Marais _____ 6/16/2022 _____

Principal Investigator Signature Date Report Submitted

* BAR-CP – Barley Coordinated Project
DUR-CP – Durum Coordinated Project
EC-HQ – Executive Committee-Headquarters
FST-R – Food Safety & Toxicology (Research)
FST-S – Food Safety & Toxicology (Service)
GDER – Gene Discovery & Engineering Resistance
HWW-CP – Hard Winter Wheat Coordinated Project
MGMT – FHB Management
MGMT-IM – FHB Management – Integrated Management Coordinated Project
PBG – Pathogen Biology & Genetics
TSCI – Transformational Science
VDHR – Variety Development & Uniform Nurseries
NWW –Northern Soft Winter Wheat Region
SPR – Spring Wheat Region
SWW – Southern Soft Red Winter Wheat Region

Project 1: Transfer of FHB Resistance to NDSU Hard Red Winter Wheat Breeding Material

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

1. Increase (annually) the frequencies of four FHB resistance genes within the NDSU breeding population through careful planning and execution of new convergent crosses coupled with marker screening and agronomic evaluation of the segregating progenies.
2. Hasten the selection of high yielding, FHB resistant inbred lines in each of three 2-year selection studies. Specific, well-chosen crosses will be employed and large numbers of progeny will be extensively evaluated.
3. Initiate (annually, in a greenhouse) the development of 500-600 new (near-random) single seed descent (SSD) inbred lines from select crosses that each segregate for one or more FHB resistance QTL (plus resistance to the wheat rusts). From the second year, the F_4 populations will be grown in the field for resistance selection and pure line development.
4. Conduct an annual Winter Wheat x Fungicide performance trial (field) to evaluate the response of advanced breeding lines and controls to fungicide application for the reduction of DON content.

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?

Objective 1: F_2 from 663 crosses (made in 2021) were planted in September 2021 for field selection in the summer of 2022. 521 new crosses were made among 55 parents in February 2022. The parents were primarily identified from among field planted routine NDSU breeding program material. FHB phenotyping was not effective due to its laborious nature, limited personnel, irregular natural epidemics and low heritability (in single plant assessments) in segregating populations. However, marker analyses that were performed at the ARS Genotyping Center in Fargo greatly aided the selection of FHB resistant parents. In addition, marker and phenotyping data from the Northern FHB Screening Nursery, NRPN, RGON and ARS Genotyping laboratory at KSU were used to identify resistant parents. Of the 521 new crosses, 325 involved one or more FHB resistant parent (primarily *Fhb1*, but also *Qfhs.ifa-5A*, *Qfhb.rwg-5A.1*, *Qfhb.rwg-5A.2* and the Everest 2DL_QTL). Use of a highly specific *Fhb1* marker allowed rapid progress in establishing the gene; however, lack of universal markers for the remaining genes seriously hampers their introgression.

Objective 2. Study (i): The transfer of resistance QTL *Qfhb.rwg-5A.1* and *Qfhb.rwg-5A.2* from PI277012 (in GP80) to HRWW was completed. During the report period, a winter wheat B_3F_1 population (GP80/Novus-4//Monument/3/2*ND Noreen) of 132 plants was evaluated for SNP haplotypes (Infinium wheat 90K SNP platform), two SSR markers and FHB Type II resistance (greenhouse). Nine lines were derived and their resistance confirmed in a further greenhouse FHB trial. The results suggested that eight lines had resistance comparable to GP80 with the *Qfhb.rwg-5A.2* markers occurring in all eight and the

Qfhb.rwg-5A.1 markers occurring in four lines. The eight selections constitute a valuable HWW resistance breeding resource and were already used in the 2022 crossing block. The selections will be included and evaluated in field trials from 2023 and will continue to be used extensively in future crossing blocks. Study (ii): Pre-breeding was done aiming to develop semi-dwarf inbred lines that are FHB resistant, winter-hardy, high-yielding, and also leaf, stem, and stripe rust resistant. Eight cross combinations were produced utilizing eight winter wheat genotypes with one or more of the desired traits. Greenhouse-based single seed descent (SSD) inbreeding with phenotypic selection steps was used for line development. F_3-derived F_4 populations were evaluated for grain yield in an un-replicated field trial. Four single plants (spikes) were selected (phenotype) from each of the nine highest yielding families and threshed individually. Five seeds from each selected spike were used for marker-based selection. Study (iii): A double cross (16M10) was subjected early generation yield selection in an attempt to derive high yielding lines with *Fhb1*. Following modified SSD inbreeding from a large F_1 population, near-random $F_{3:4}$ inbred lines were established and planted in an un-replicated yield trial. The 40 best $F_{3:5}$ lines were again planted in a replicated yield trial. The highest yielding selections were then analyzed with markers to establish inbred lines.

Objective 3: Annually, SSD inbreeding is initiated from 30-40 promising (high yield with resistance to FHB and other diseases) cross combinations to achieve generation acceleration and hasten the development of FHB resistant inbred lines. SSD is used in place of doubled haploid (DH) development as it is cheaper and in line with the project's available resources. Modified SSD with selection for simply inherited traits is also less wasteful compared to DH development. Crosses are chosen that have at least one parent that is believed to be FHB resistant. F_2 seedlings (± 96 plants/cross) are screened with mixed leaf and stem rust inoculum and during all stages of inbreeding the plants are selected (greenhouse) for height and fertility. The $F_{2:3}$ is planted in the field in the fall and subjected to single plant selection in the ensuing summer. Three sets of SSD selections have been handled in this manner during the report period.

Objective 4: A variety X fungicide evaluation trial with 22 entries was planted at Casselton. The trial followed a split plot layout (three replicates) with half of the plots treated with Prosaro at 8.2 fl oz/acre at flowering (applied on 6/4, 6/7, 6/9, and 6/11 to allow for variation in flowering dates among the varieties). Corn inoculum (FHB) was applied on 5/26/21 and 5/28/21 (the earliest varieties started to head around 6/1/21).

b) What were the significant results?

Objective 1: The presence (marker detected) of resistance QTL *Fhb1* (and to a lesser extent *Qfhs.ifa-5A*) increased among advanced inbred lines. Currently, 10 of the 23 State-wide Elite Trial entries have *Fhb1* (three of these are also included in the NRPN and seven in the RGON). Of the 180 advanced (Senior) yield trial entries, 68 have *Fhb1*. With regard to the 554 new Junior Trial inbred lines, 357 have *Fhb1*. Thus, *Fhb1* has now been firmly established in each program generation. *Fhb6*, *Qfhb.rwg-5A.1*, *Qfhb.rwg-5A.2* and the Everest 2DL_QTL were introduced for the first time in the F_2 to F_3 generations. The

segregating populations and inbred lines were furthermore evaluated for multiple adaptive (ND) traits. Phenotyping and marker selection (where appropriate markers were available) were done to also raise the presence of resistance to the cereal rust diseases and increase the possibility to develop inbred lines with broad disease resistance.

Objective 2. Study (i): The *Qfhb.rwg-5A.1* and *Qfhb.rwg-5A.2* QTL have small individual effects and were difficult to transfer in the absence of reliable markers; however, we could identify and apply SNP haplotypes of the donor line (GP80) to aid transfer. Eight homozygotes for one or both resistance genes were obtained following three backcrosses to winter wheat (approximately 93% winter wheat background recovered) and were confirmed in a replicated greenhouse trial. The selections had resistance similar to the spring wheat donor and were calculated to have 75% to 82% of ND Noreen genetic background. ND Noreen is believed to have background (native) FHB resistance that appears to bolster the PI277012-derived resistance. Derivatives with the genes are now being evaluated further and will be applied in a wide range of new crosses. F_4 intermediates from the transfer that segregate for the resistance markers will be planted in the field in September 2022 for continued selection in 2023. Markers *Xbarc186* (*Qfhb.rwg-5A.1*), *Xgwm2136* and KASP marker 5AL-8.0K (*Qfhb.rwg-5A.2*) could be useful for predicting the presence of the respective genes; however, both *Xbarc186* and KASP-5AL-8.0K give cross parent-specific polymorphisms that limit their usefulness. *Xgwm2136* appears to produce a unique polymorphism associated with the resistance, yet the marker may be less tightly linked to the resistance. Study (ii): Two of the nine originally selected (yield) F_4-derived families completely lacked *Fhb1,* whereas the remaining seven families were homozygous for *Fhb1*. Following the marker screens, 140 F_5-derived inbred lines with *Fhb1* and additional favorable (marker-predicted) resistance gene combinations were established from the seven families for continued yield testing. In addition to *Fhb1*, the presence of rust resistance genes *Lr34* (58%), *Lr46* (46%), *Lr68* (52%), *Yr17* (71%) and the 1BL.1RS translocation (15%) could also be determined. Few reliable and specific disease resistance markers are available in wheat, and it is highly likely that additional rust, bacterial leaf streak and tan spot resistance genes that were present among the parents will also occur among the progeny. The selections will be planted in the field as single rows in September 2022. Study (iii): Based on the marker results and phenotypes in observation/increase rows, four cross 16M10 lines with the *Fhb1* and rust resistance gene markers were singled out for further testing and included in the 2022 Preliminary (Junior) yield trials.

Objective 3: In 2022, three sets of SSD material were handled, including: (i) 501 F_4 rows derived from 40 crosses made in 2020 (planted at Casselton in September 2021). Selections from these rows will progress to F_5 head rows (planted in September 2022). (ii) 656 F_3 rows that derive from 44 crosses made in 2021 (planted at Casselton in September 2021). Following selection, F_4 rows will be planted in September 2022. (iii) F_1 of new 31 cross combinations (made in 2022) were greenhouse planted in March 2022. Following greenhouse selection, the $F_{2:3}$ will be field planted (September 2022) together with the other earlier derived SSD derivatives

Objective 4: Very dry growing conditions prevailed throughout the state and no significant incidences of foliar diseases occurred at Casselton in 2021. In the fungicide trial, no significant FHB infection occurred, and no DON analyses were done. There was also no significant effect of the fungicide treatment on any of the traits measured and no differential responses of varieties to fungicide application either. A new (2022) Winter Wheat Variety trial with 22 entries and the same statistical design was planted at Casselton in the fall of 2021.

c) List key outcomes or other achievements.

Based on marker results, it is belied that *Fhb1* has now been successfully incorporated into a significant proportion of the NDSU winter wheat breeding population and in all phases of the selection scheme.

Indications are that the very promising resistance gene combination *Ofhb.rwg-5A.1* and *Qfhb.rwg-5A.2* present in the spring wheat PI277012 has been successfully incorporated into our winter germplasm. The genes are expected to contribute significantly to the improvement of FHB resistance in winter wheat.

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

Sagar Adhikari MS thesis title: Pre-breeding to combine disease resistance and agrotype genes in hard winter wheat. Graduated in February 2022.
Bipin Neupane MS thesis subject: A diallel study - Evaluation of winter wheat lines for FHB genetic background resistance. Joined the project in June 2021, expected to graduate in the spring of 2023.
Bhanu Dangi MS thesis subject: Evaluation of winter wheat for FHB genetic background resistance. Joined the project in January 2022; expected to graduate in the spring of 2024.

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

Promising, advanced breeding material was entered in regional nurseries such as NRPN (5 lines), RGON (25 lines), Northern Scab Nursery (15 lines); USDA stem rust (100 lines), stripe rust nurseries (USDA, KS 160 lines; USDA WSU 100 lines) and statewide variety trials (ND - 2 lines; SD - 1 line; MN - 1 line; MT - 1 line). Data on the submitted material get listed in on-line reports of the respective nurseries and are accessible to researchers in the hard winter wheat producing states. The results of the Variety X Fungicide trial of 2020/21 were incorporated in the NDSU annual publication “North Dakota Hard Red Winter Wheat Trial Results and Selection Guide” (available at https://www.ndsu.edu/agriculture/ag-hub/publications/north-dakota-hard-red-winter-wheat-variety-trial-results-2021-and-selection)

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).

None

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).

Adhikari, S. (2022). Pre-breeding to combine disease resistance and agrotype genes in hard winter wheat. MS Thesis (accepted February 2022, North Dakota State University); acknowledgment of federal support - yes.

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.

Clair Keene, Joel Ransom, Francois Marais, Senay Simsek and Andrew Friskop (NDSU Main Station); Eric Eriksmoen (North Central Research Extension Center, Minot); John Rickertsen (Hettinger Research Extension Center); Glenn Martin (Dickinson Research Extension Center). North Dakota Hard Winter Wheat Variety Trial Results for 2021 and Selection Guide (A1196-21, September 2021). Available online at: North Dakota Hard Red Winter Wheat Variety Trial Results for 2021 and Selection Guide | NDSU Agriculture and Extension