

Project FY22-HW-003: Transfer of FHB Resistance to NDSU Hard Red Winter Wheat Breeding Material

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

Objective 1. Systematically raise the frequencies of target FHB resistance QTL in the HRWW breeding program through convergent crosses, marker use and agronomic evaluation.

Objective 2. Select for genetic background (native) FHB resistance among advanced lines and utilize it in the breeding program.

Objective 3. Develop F₄ single seed descent (SSD) inbred lines from crosses that segregate for FHB resistance QTL (plus resistance to the wheat rusts) in each project year.

Objective 4. Conduct an annual Winter Wheat x Fungicide field trial to evaluate genotype response 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.)

Objective 1: Major activities.

- In the 2025 crossing block 48 parents were crossed in 456 single cross combinations. Parents had one or more of the traits: FHB resistance (*Fhb1*, *Fhb7*, or “native” = 12 lines), leaf rust resistance (18), stem rust resistance (12), stripe rust resistance (9), grain yield (19), semi-dwarf height (47). Thirty-four per cent of crosses involved at least one FHB resistant parent.
- Introgression of *Fhb7* from XWC14-255-3-1: The first aim was to replace the agronomically poor “Chinese Spring” genetic background with well-adapted HRWW genetic backgrounds using marker-assisted, modified backcross breeding. Three backcrosses were made to different winter wheat parents relying on a dominant, *Fhb7*-diagnostic marker. Five diverse HRWW parents were used for the final backcross to recover 93.3% of HRWW genetic background. A second aim was to also recover genotypes in which *Fhb7* occur together with one or more additional larger-effect FHB resistance QTL or background resistance QTL. The final backcross parents were therefore diverse with respect to *Fhb1*, *Qfhb.rwg-5A.1*, *Qfhb.rwg-5A.2* and un-identified background resistance QTL.

Objective 1: Significant results.

- Selection for cold-tolerance, high yield and broad disease resistance. (a) 500 new F₅ inbred lines (2024) advanced to 2025 Junior yield trials. Marker analyses (USDA Wheat Genotyping Laboratory, Fargo) to predict *Fhb1*, *Qfhb.rwg-5A.2*, and key rust resistance genes showed that 193 (39%) of the new lines had *Fhb1* whereas 14 (3%) had *Qfhb.rwg-5A.2*. (b) 180 F₆ lines (2025 Senior yield trials) included 98 (54%) with *Fhb1*. These will be selected for cold survival, plant phenotype, broad disease resistance, grain yield and processing quality. (c) 24 lines (of which 6 have *Fhb1*) were included in the 2025 Statewide Elite Yield Trial.
- *Fhb7*: 322 B₃F₂ from six modified backcross lineages were tested (markers) for *Fhb7*, *Qfhb.rwg-5A.1*, *Qfhb.rwg-5A.2*, *Fhb1* and additional disease resistance genes *Lr34*, *Lr46*, *Lr68* (leaf rust), *Sr24* (stem rust) and *tsn1* (tan spot). The same 322 plants were then evaluated for FHB Type II resistance and overall phenotype in a greenhouse. The combined data were considered to identify two sub-groups of plants for continuation of the project.

Objective 1: Key outcomes.

- Promising advanced lines have been included in 2025 disease evaluation nurseries (FHB, leaf rust, stem rust, stripe rust), the Regional Germplasm Observation Nursery (RGON) and Northern Regional Performance Nursery (NRPN). Apart from disease and phenotypic data,

the USDA Genotyping Laboratory (KSU) runs extensive marker tests on regional nurseries, facilitating selection of the better lines which will then again be involved in annual crosses to achieve steady, ongoing pyramiding of the diverse resistances.

- ***Fhb7***: (a) Twenty-two B₃F₂ plants either had *Fhb7* singly (4 plants) or were pyramided genotypes with two to four of *Fhb7*, *Qfhb.rwg-5A.1*, *Qfhb.rwg-5A.2* and *Fhb1*. Most QTL pyramids were only partially homozygous for the targeted QTL and the plants had acceptable height, spike characteristics and fertility. The B₃F_{2:3} was therefore replanted for a second cycle of marker screening to recover additional, homozygous pyramided genotypes. (b) Ten B₃F₂ plants with good agrotypes and strong type II FHB resistance scores were homozygous for resistance QTL *Fhb7* (7 plants); *Fhb7* & *Qfhb.rwg-5A.2* (1 plant); *Fhb7* & *Qfhb.rwg-5A.1* (1 plant); and *Qfhb.rwg-5A.1* & *Qfhb.rwg-5A.2* (1 plant). The ten selections are being retested in a second (replicated) greenhouse FHB resistance trial to confirm the resistance and to select the agronomically best, resistant B₃F_{2:3} segregates within each family. (c) Finally, B₃F_{2:3} families that lacked *Fhb7* but nonetheless showed strong FHB resistance (segregated for alternative and/or background FHB resistance QTL) were planted for field evaluation in 2025. (d) Four B₃F_{2:3} and five (different) B₃F_{2:4} families, homozygous for *Fhb7*, were used for making new crosses in 2024 and 2025, respectively.

Objective 2: Major activities.

- Following a diallel study (2022) and a first testcross analysis (2023), a second testcross analysis (2024) was done in which 20 well-adapted genotypes with unknown FHB resistance were crossed to three heterogeneous testers. Tester #1 was a F_{2:3} line, homozygous for FHB resistance QTL *Qfhb.rwg-5A.1* and -5A.2 (markers) and it carried FHB background resistance from ND Noreen. Testers #2 and #3 were F₁ hybrids of ND Noreen with 18Nord-107 and 19Nord-129, respectively (each parent has background resistance that was detected in the earlier diallel trial). The trial attempted to: (a) identify additional sources of background resistance; (b) pyramid unknown and uncharacterized, background FHB resistance QTL with known, larger effect FHB resistance QTL. Since FHB resistance QTL appear to be mostly dominant and complementing (first two trials), the best F₁ plants within the most resistant cross combinations were likely to involve several, complementing resistance QTL. **Objective 2: Significant results.**

- The second (2024) testcross trial showed that 21Nord-156, 21Nord-153 and 20SSD-19K505-8-2 have promising levels of background resistance.
- Highly significant differences in FHB resistance of lines, testers, and their interactions were found. Many crosses exhibited superior resistance, with 19 F₁ hybrids having (average) IS < 24%. Presence of stronger resistance appeared to result from accumulated, complementing FHB resistance alleles. It also appeared as though “smaller effect” background resistance QTL can have equal or better effect on overall resistance compared to the presence of *Fhb1*.

Objective 2: Key outcomes.

- To date, nine different HRWW genotypes were found to have good FHB background resistance. These have, and are being used as cross parents to improve FHB resistance.
- The study underscored the utility of line x tester crosses to select and accumulate known, “larger-effect” and under-utilized, “small-effect” resistance QTL in FHB resistance breeding.
- The most resistant progeny will be selected in the field to develop and test pure lines.

Objective 3: Major activities.

- SSD inbreeding was initiated (annually) from promising crosses (high yield with resistance to FHB and other diseases) to achieve generation acceleration. ± 96 F₂ seedlings/cross were

screened with mixed leaf and stem rust inoculum. During inbreeding the plants were also selected for height and fertility. The F₄ was field planted in the fall of year 2 and single plants were selected in year 3. F₅ pure lines were established in year 4.

Objective 3: Significant results.

- (a) SSD inbreeding resulted in 250 pure lines from 44 crosses made in 2021. These were included in un-replicated yield trials in 2025. (b) In 2022, SSD inbreeding was initiated from 31 crosses of which 30 had resistance QTL from one or more of the following sources: background resistance (12), *Fhb1* (20), *Qfhb.rwg-5A.1* (4), *Qfhb.rwg-5A.2* (4). From these, 945 F₅ inbred lines were derived for field evaluation in 2025 (c) In 2023, SSD inbreeding was started with F₂ of 30 crosses. FHB resistance QTL were present in 21 crosses as follows: background resistance (13), *Fhb1* (3), *Qfhb.rwg-5A.1* (8), *Qfhb.rwg-5A.2* (8). The F₄ (322 rows) were planted for making head selections in 2025. (d) In 2024, 30 crosses were used to initiate a new set of SSD; 17 crosses had *Fhb1* present and 4 had background resistance.

Objective 3: Key outcomes.

- (a) In 2025, 250 new SSD inbred lines from 44 crosses (2021) included 82 lines with *Fhb1* and another 12 lines with *Q fhb.rwg-5A.1*. (b) In 2025, marker screens will again be done on the 2022-initiated SSD to identify those with *Fhb1* and *Q fhb.rwg-5A.1*. (c), d) Marker assessments pertaining to the 2023 and 2024 crosses will be done in 2026, 2027.

Objective 4: Major activities.

- An annual variety X fungicide evaluation trial (Casselton) with 22 entries followed a split plot layout (3 replicates) with half of the plots treated with Prosaro at 8.2 fl oz/acre at flowering (applied on three different dates to allow for variation in flowering dates). Corn inoculum (FHB) was applied on two dates, the first with the onset of heading in the earliest varieties.

Objective 4: Significant results.

In 2024 wet, humid conditions were encountered during the growing season. However, due to late disease onset, FHB infections were sporadic and non-uniform.

Objective 4: Key outcomes.

- Fungicide treatment did not cause significant differential yield or DON responses.

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

- Kripa Rijal is expected to graduate in the spring of 2025 with MS thesis title: "Improvement of Fusarium Head Blight resistance in winter wheat."

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

Advanced lines were entered in regional nurseries [NRPN (5); RGON (30); Northern Scab (15); USDA stem rust (100); USDA KS Stripe rust (152); USDA WSU stripe rust (120)]; statewide variety trials [ND (3); UMN (2)], and WQC Northern Grow-outs (3). Data on the submitted material get listed in on-line reports of the respective nurseries. Variety X Fungicide trial results were reported on the NDSU website.

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

"Larger"-effect resistance QTL *Fhb1*, *Fhb7*, *Qfhb.rwg-5A.1* & *-5A.2* and nine sources of significant, "smaller"-effect background FHB resistance QTL are being pursued. Marker-aided, convergent crosses and Pedigree breeding will be employed to widely disperse the variability within the breeding population and to combine it with winter survival, yield, broad adaptation, good processing quality and wide spectrum disease resistance.