

Project 2: A Double Haploid Initiative to Speed Development of FHB Resistant Soft Winter Wheat

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

This proposal expands the regional Double Haploid (DH) initiative to quickly develop and release high-yielding varieties that contain effective FHB resistance pyramids. Objectives are: 1) Develop DH lines that combine multiple effective FHB resistance genes/QTL in a high-yielding background; 2) Utilize molecular markers to track FHB genes/QTL and enrich F1 DH populations; and 3) Share new DHs with all VDHR-SWW breeders after the initial culling such that each breeder evaluates about 475 new DHs each year.

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

What were the major activities?

The collaborative DH project has been very successful at rapidly developing pure lines with good FHB resistance. Three new varieties derived from DHs funded through this initiative with good FHB resistance were released between 2022 and 2023 by Virginia Tech, 15VDH-FHB-MAS-38-02 (2022, private license), 15VDH-FHB-MAS33-13 (2022, private license), 15VDH-FHB-MAS25-15 (2023, public released as 'VT Pitman'). All three contain *Fhb1*.

In 2022, a single cross (18VDH-FHB-MAS07-173-03 / 16VDH-SRW03-018) segregating for 7 FHB QTL was chosen to produce a larger number of DH lines (350) for selection of lines with many QTL combinations. Fifty eight F1 seeds from this cross were sent to Heartland in the summer of 2022 to produce 258 DH lines. These parents were chosen because they completely complementary FHB QTL (line 18VDH-FHB-MAS07-173-03 contains *Fhb1* (3BS), FHB3B.Bess, FHB1A.Neusse, and FHB6A.Neusse, while line 16VDH-SRW03-018 contains FHB1B.Jtwn, FHB2B.Bess, FHB.3BL) yet have the same vernalization alleles, same photoperiod sensitivity alleles, and the same *Rht* alleles as determined by inhouse genotyping with SSR and KASP markers. Both parents were developed from this long running project, and have demonstrated strong agronomic performance, with 16VDH-SRW03-018 ranking first in the overall mean across the Uniform Southern Nursery in 2021, and 18VDH-FHB-MAS07-173-03 ranking first in the Mason Dixon trial at Warsaw, VA in 2022. Thirteen lines from this cross were visually selected for inclusion into the primary breeding pipeline, while all 258, were harvested to create a mapping population to investigate epistatic interactions between FHB QTL. All lines will be planted in both yield plots as well as in the scab nursery for 2025. In our proposal (FY22), we demonstrated that FHB QTL do not appear to act in an additive fashion, and it is unclear if all have non-zero effects in all genetic backgrounds. We hope that by standardizing the genes for photoperiod sensitivity, vernalization and plant height, we can remove these confounding factors to dissect potential interactions at previously detected FHB QTL. This effort is in contrast to the selection of DH crosses based on GEBVs of DON, FDK and other agronomic traits in Project 2. No crosses will be sent for DH production in 2024 due to a lack of funding.

Table 1: Crosses sent for DH production in 2023 through genome-enabled selection or parents with complementary QTL.

Pedigree	Year	No. DH	Fhb1 ^a	FDK ^b	DON ^c	Yield ^d	Selection Index ^e
18VTK10-23 // 17VTK4-29 / 15VDH-FHB-MAS38-01	2023	60	seg	-1.28	-1.19	1.48	3.69
VA22FHB-111 / 15VDH-FHB-MAS33-13	2023	40	seg	-2.00	-1.58	0.17	3.18
VA22W-207 / 19VT2-44-20	2023	50	seg	-2.37	-2.27	-0.78	2.75
VA20FHB-18 // VA20W-135 / 17VTK4-29	2023	60	seg	0.19	-0.57	1.29	2.54
18VDH-FHB-MAS07-173-03 // 15VDH-FHB-MAS33-13 / 15VDH-FHB-MAS38-01	2023	50	fixed	-2.12	-0.36	-0.91	2.32
18VTK10-23 // 15VDH-FHB-MAS33-13 / VA17W-75	2023	50	seg	-1.17	-1.60	0.75	3.26
15VDH-FHB-MAS33-13 / DH20SRW02-073	2023	60	seg	-1.24	-1.55	0.87	3.24
18VTK10-23 // 15VDH-FHB-MAS33-13 / 15VDH-FHB-MAS38-01	2023	50	seg	-1.38	-1.48	0.79	3.35
17VTK4-29 / 15VDH-FHB-MAS33-13	2023	60	fixed	-1.67	-0.84	1.99	4.00

^a status of *Fhb1* in the cross. “Fixed” means both parents are homozygous for the *Fhb1* allele, wheels seg indicates that only one parent contains the allele, and a dash indicates neither parent has *Fhb1*. Crosses were not designed with *Fhb1* explicitly, but were clearly favored due to low FDK and DON.

^b Genomic prediction of FDK in standard units from the population mean (i.e. z-scores of GBLUP).

^c Genomic prediction of DON in standard units from the population mean (i.e. z-scores of GBLUP).

^d Genomic prediction of Grain Yield in standard units from the population mean (i.e. z-scores of GBLUP).

^e Selection index included genomic predictions for FDK, DON, grain yield (shown in b-d), test weight, heading date, powdery mildew and leaf rust (not shown).

In fall 2023, 730 DH lines sent for DH production in 2022, were planted in single rows for 2024 evaluation. Of these, 134 lines were selected for advancement to year-1 trials in the 2025 SRW Observation nursery. Five grams of each line are being partitioned for all collaborators on the ust 2024.

What were the significant results?

Very poor DH seed was received back from Heartland Plant Innovations in the fall of 2022 from the FY21 project. This drastically limited the number of DH lines available for selection. Plant materials (67 F1 plants) were shipped to Heartland Plant Innovations Inc in the fall of 2021, but were damaged enroute. Heartland salvaged what they could but were only able to develop 38 DH lines instead of the expected 550. An additional 81 lines were sown in headrows from DH lines that were received the previous year (2021) with too few seeds (< 6), using seed grown in the greenhouse. For F1 lines submitted in 2022 and for the foreseeable future, only F1 seed will be sent. It is unclear to what degree the lack of DH lines produced and the poor quality of that seed had to do with damage during shipping. Including lines with insufficient seed for field planting from the previous year, 48 lines were advanced from headrows to the 2024 SRW Observation nursery. These lines were not shipped to collaborators due to limited and poor seed quality. Of these, only about 6 lines appear to have agronomic performance acceptable enough to advance to year-2 yield testing.

List key outcomes or other achievements.

- 1) A new public variety, 'VT Pitman', was released in the spring of 2023, tested under the name 15VDH-FHB-MAS25-15. This variety is a sister line to 15VDH-FHB-MAS33-13 released in 2022, and has similar FHB resistance, and carries the *Fhb1* gene. In contrast to 15VDH-FHB-MAS33-13, 15VDH-FHB-MAS25-15 has demonstrated better powdery mildew resistance. Both lines have the *H13* allele for hessian fly resistance.
- 2) A mapping population of 258 DH lines was created from the cross 18VDH-FHB-MAS07-173-03 / 16VDH-SRW03-018, and will be planted in

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

PhD student Sunilda Frias is working on the sparse testing approach in the SRW Observation nursery, providing opportunities to learn how genomic prediction can be used outside traditional GS methods.

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

Five grams of each selected DH line will be shipped to collaborators in the fall (August) of 2024.

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

Funding for the DH project was cut to zero for FY24. Thus, no crosses were sent for DH in 2024, as it takes one year for development and we will have to pay for the DH sent in 2023. I intend to continue to accompany the Barley Improvement Committee and the Wheat Improvement committee to meet with congress to stress the importance of this research and help to restore funding to the 15 million authorized in the farm bill.