

Project FY22-SW-005: Genome-enabled Selection for FHB Resistance in Winter Wheat

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

1. Evaluate available adapted wheat germplasm for FHB resistance and determine the predictive ability of FHB traits, including visual scores, FDK and DON within the Virginia germplasm. Develop new varieties on soft winter wheat with improved resistance to fusarium head blight and excellent agronomic performance.
2. Determine the linear (and potentially non-linear) genetic relationships (i.e. genetic correlation) between morphology (e.g. height), phenology (e.g. heading date, flowering time), and FHB traits (FHB index, FDK, DON), and their impact on agronomic performance
3. Use genomic information to estimate genetic and breeding values for advancement and mating decisions, respectively, to improve FHB resistance and agronomic performance. Redesign mate-pair decision making to include multiple traits and a selection index to maximize response to selection for both FHB traits and other agronomic and disease traits.
4. Evaluate response to selection and any unexpected consequences thereof.

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

What were the major activities?

Most progress so far has been made on objectives 1 and 3. In spring 2023, a new public soft red winter (SRW) wheat variety, developed primarily from support from the USWBSI VDHR, was released under the name 'VT Pitman'. 'VT Pitman' performed well in state official variety trials, in 2024, ranking 20 out of 150 entries for grain yield across locations in Virginia, and placing 2nd across the state in North Carolina. Four SRW lines were released in the spring of 2024, 15VTK-1-101, 17VTK4-29, VA19FHB-36, and 16VDH-SRW02-023. Lines 17VTK4-29 and VA19FHB-36 were specifically developed for FHB resistance, with the first containing *Fhb1*, and the latter containing FHB QTL *Fhb1B-Jamestown*, and *Fhb1A-Neuse*.

Several high yielding lines with excellent FHB resistance are currently under evaluation in late stage regional trials. Genomic information is used to estimate genetic and breeding values at all breeding stages, such that selection on predicted FHB traits is now conducted at every generation. Genomic prediction of expected mate pair performance is being used to guide all crossing decisions.

Genomic prediction of crosses using predicted family merit

A genomic prediction model was trained using data from the VT breeding program across five years, 2019-2023. This dataset included 41,568 phenotypic records (plots), representing 164,774 phenotypic data points for 4,840 breeding lines measured for 14 traits (unbalanced).

Included in the dataset were phenotypic records for 4,645 scab plots with 2,998 FHB scores, 4,181 FDK, and 3,525 DON measurements after filtering for heritability greater than 0.3 from 1,916 lines evaluated in the misted inoculated nurseries in Virginia from 2019 through 2023. Data from 2021 was excluded due to very low FHB pressure. All lines were genotyped with genotyping by sequencing markers, totaling 9,286 after variant calling, filtering and imputation. Genomic BLUPs were fit and marker effects estimated through backsolving.

Elite parents (51 inbred lines) were selected based on superior performance in advanced yield trials. Early generation parents (44 inbred lines) were selected based on predicted genomic merit of lines evaluated in first year yield trials (2023) in one plot in 1 or 2 locations using a sparse testing design. F1 seed (168 F1s) from the previous year of crossing (2022) was also selected based on the previous year's genomic estimated family merit. Inbred lines were planted in 9 to 12 pots, while F1s were limited to 3-4 pots each so that remaining seed could be planted earlier that fall. All selected parents were Virginia lines, or F1s between Virginia Lines to ensure high genetic connectivity for prediction.

Expected family genotypes were calculated for all possible mate-pair combinations (32,896 predicted families) using 9,286 GBS markers and 47 loci of interest that were screened in house for inbred parents, and predicted for F1s from parental genotypes. Expected family merit for each of the 14 traits was calculated by the product of the expected GBS genotype values for each family and the predicted marker effects from the trained genomic selection model. A selection index was created by weighting the traits based on perceived importance within the program, with Yield, FDK, DON and test weight given the most weight. The initial intention for this project was to assign economic weights, but these are not trivial to estimate. Economic weights are highly influenced by the market (e.g. price of grain, dockage for DON, test weight, etc.) and regional importance of traits (e.g. disease pressure is not uniform across our target population of environments). Crosses with a high coefficient of coancestry were generally avoided to reduce population level inbreeding. Crosses sent for doubled haploid generation in 2023 were chosen based on both the selection index and prior knowledge of the parents.

Evaluation of across season genomic predictive ability

Multivariate prediction of FHB traits is predictive across years, with previous years data able to predict unobserved lines in the following year, and is generally marginally better than single trait prediction across years (Figure 1). Loss of prediction accuracy in 2022 is likely due to the use of new races, suggesting there may be race specific tolerance in wheat. Investigation into the degree of race specificity and tolerance / susceptibility needs further work to determine to what degree this is a problem. Year to year variability in response to infection may be due to, among timing of infection, which races are most prevalent at a given site. If so, understanding what leads to race specific tolerance / susceptibility would allow breeders to develop lines more stable to year to year (i.e. race to race) variation.

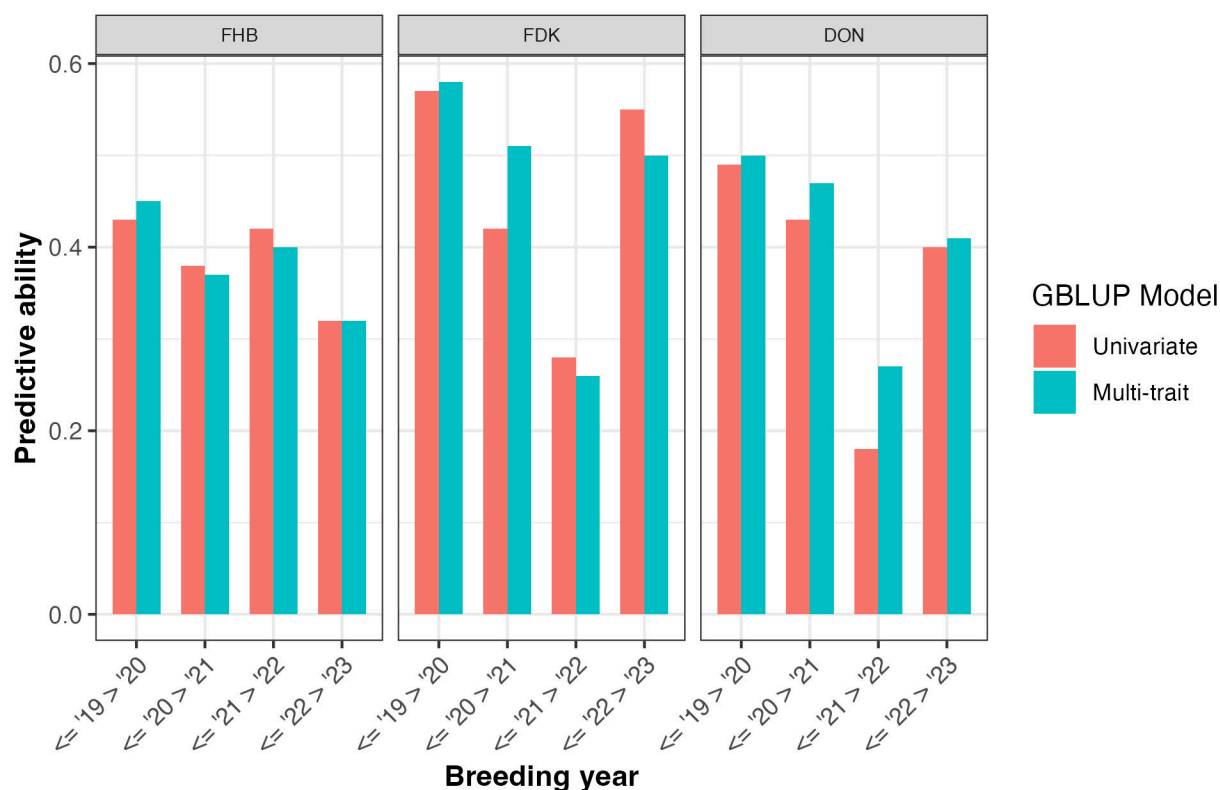


Figure 1: Predictive ability of univariate and multivariate (FHB scores, FDK and DON together) using previous years data to predict the performance of lines in a given year. Axis labels indicate using previous years to predict performance in the following year, for example “<= '22 > '23”, the years 2019-2022 used to predict 2023.

Evaluation of line performance in Misted Inoculated nursery

Regional trials and early generation trials (first and second year) were evaluated in a misted inoculated nursery. The scab nursery was intentionally planted last (i.e. late) to increase the likelihood of warmer temperatures during flowering, but was significantly earlier than previous years due to good weather and an expected due date for the PI’s son in early December (Nov 18, 2021, Dec 2, 2022; Nov 9 2023). This strategy was started for the harvest year 2022, and was continued for the 2023 harvest due to excellent infection and high disease pressure. Disease pressure appeared visually less in 2024 compared to previous years, and FDK values were observed lower than previous years.

A total of 1,860 soft winter wheat plots were evaluated in the misted inoculated nursery for the 2024 harvest season, representing 925 breeding lines and check varieties. Regional nurseries included the Northern Uniform Scab (47 entries), Preliminary Northern Uniform Scab (40 entries), Southern Uniform Scab (59 entries), Uniform Eastern (28 entries), Uniform Southern (40 entries), Gulf Atlantic (60 entries), and Mason Dixon (79 entries) nurseries. Local nurseries evaluated included the second year SRW Preliminary (168 entries) and a portion of the first year SRW Observation (245 entries out of 857). While the other scab trials have a complete entry list, the SRW Observation was subset to reduce the total number of lines under FHB evaluation as part of the sparse testing design. All families were sampled to maximize the prediction accuracy

of FHB traits in unobserved lines. Entries from all regional trials are being prepped for planting in Fall 2024.

Crosses

A total of 302 single crosses and 333 top-crosses (F1 by elite) were made, as well as 13 double crosses (F1 by F1) of specific interest in spring of 2023. Each day, a list of females and males that were ready for crossing was used to subset the list of potential crosses, and used to make high ranking crosses based on the selection index. A training dataset consisting of data from 2019 to 2023 was used to train the genomic prediction model. A set of 158 crosses were also made to introgress a dominant male sterility gene from New York germplasm into a Virginia germplasm background for the second cycle of backcrossing to elite Virginia materials. The dominant male sterile gene is intended to facilitate crossing in a future rapid cycling breeding scheme.

Scab nursery

All trials were given a visual score for FHB approximately 2 weeks after flowering, after scab infection was allowed to progress and was very apparent, but before dry down. The Uniform Scab nurseries, Northern, Preliminary Northern and Southern, as well as the Mason Dixon and Gulf Atlantic were scored for FDK after harvest, and were processed for DON accumulation (Table 2). Other nurseries are in progress for FDK and DON sample processing (Table 3).

Key outcomes

- 1) A pipeline for genomic prediction of mate pairs for multiple traits and single genes, including FHB traits, has been fully implemented
- 2) All regional nurseries we participate in (7) were screened in our misted inoculated nursery for FHB scores. FDK and DON traits are currently being collected.
- 3) Lines with improved resistance and agronomic traits have been advanced to regional nurseries. One DH line with improved FHB resistance (including *Fhb1*), 15VDH-FHB-MAS25-15 was released as a public variety in the spring of 2023, under the name 'VT Pitman'.
- 4) Four SRW lines were released in the spring of 2024, 15VTK-1-101, 17VTK4-29, VA19FHB-36, and 16VDH-SRW02-023. Lines 17VTK4-29 and VA19FHB-36 were specifically developed for FHB resistance, with the first containing *Fhb1*, and the latter containing FHB QTL *Fhb1B-Jamestown*, and *Fhb1A-Neuse*.

Table 2: Results from Uniform Scab nurseries in 2023. Means, broad-sense heritability, and error standard deviation for FHB scores (0-9), FDK (%) and DON (ppm) for the 2023 scab nursery are shown.

Year: 2023	NUWWSN			PNUWWSN			SUWWSN		
Trait	FHB (0-9)	FDK %	DON ppm	FHB (0-9)	FDK %	DON ppm	FHB (0-9)	FDK %	DON ppm
Mean	3.0	12.7	7.7	3.9	26.9	5.0	4.6	27.0	11.95
Broad-sense heritability	0.63	0.64	0.91	0.4	0.57	0.59	0.45	0.73	0.83
Error standard deviation	0.79	5.9	1.7	1.02	12.7	2.0	1.02	11.5	4.48

Table 3: Results from collaborative nurseries in 2023. Means, broad-sense heritability, and error standard deviation for DON (ppm) and FDK (%) for the 2023 scab nursery are shown. Statistics on FHB scores are omitted for brevity.

Year: 2023	Gulf Atlantic		Mason Dixon		Uniform Eastern		Uniform Southern	
Trait	DON ppm	FDK %	DON ppm	FDK %	DON ppm	FDK %	DON ppm	FDK %
Mean	10.5	44	3.6	48	3.7	60	4.0	TBD
Broad-sense heritability	0.58	0.63	0.36	0.53	0.64	0.51	0.22	TBD
Error standard deviation	5.0	13	0.96	15	0.8	13	1.05	TBD

Table 4: Results from Uniform Scab nurseries in 2024. Means, broad-sense heritability, and error standard deviation for FHB scores (0-9), FDK (%) and DON (ppm) for the 2023 scab nursery are shown.

Year: 2024	NUWWSN			PNUWWSN			SUWWSN		
Trait	FHB (0-9)	FDK %	DON ppm	FHB (0-9)	FDK %	DON ppm	FHB (0-9)	FDK %	DON ppm
Mean	4.3	11.9	TBD	6.0	9.5	TBD	4.8	20.9	TBD
Broad-sense heritability	0.29	0.66	TBD	0.3	0.71	TBD	0.02*	0.80	TBD
Error standard deviation	1.5	6.1	TBD	1.0	3.36	TBD	1.73	6.7	TBD

* Reasoning for low heritability for FHB scores in the Southern Uniform is unknown.

Table 5: Preliminary results from collaborative nurseries in 2024. Means, broad-sense heritability, and error standard deviation for FHB scores (0-9) and FDK (%) for the 2023 scab nursery are shown.

Year: 2024	Gulf Atlantic		Mason Dixon		Uniform Eastern		Uniform Southern	
Trait	FHB (0-9)	FDK %	FHB (0-9)	FDK %	FHB (0-9)	FDK %	FHB (0-9)	FDK %
Mean	5.4	23.9	4.0	15.8	5.7	TBD	4.2	TBD
Broad-sense heritability	0.41	0.63	0.51	0.68	0.46	TBD	0.42	TBD
Error standard deviation	1.2	8.4	1.3	8.03	1.34	TBD	1.56	TBD

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

Postdoctoral associate, Dr. Moshood Bakare, learned to score FHB traits in the field, as well as FDK and DON sample preparation. Dr. Bakare worked with postdoctoral associate Felipe Sabadin to develop the prediction model for mate-pair decisions. He is currently working on a manuscript genetic relationships between FHB traits and other agronomic, phenologic and morphologic traits to determine trade-offs and unintended consequences of selection. Graduate students Lexi Perry and Sunilda Frias assisted Moshood Bakare in scoring FHB traits in the field, and thus gained experience recognizing and quantifying FHB.

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

Results from the regional nurseries have been supplied to the lead PI's on those efforts, and we will continue to provide FDK and DON data as it becomes available. All 2024 harvest data will be uploaded to T3 as data becomes available for access by other breeders and interested parties.

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

We are currently working on a manuscript assessing the genetic relationships between FHB traits and other agronomic, phenologic and morphologic traits to determine trade-offs and unintended consequences of selection. Additionally we will evaluate the efficacy of genomic prediction through validation of families in the 2026 scab nursery.

Approximately 2,000 wheat plots will be evaluated in our misted inoculated FHB nursery, including Y1, Y2 and elite lines from our program and lines from other programs entered into regional nurseries.