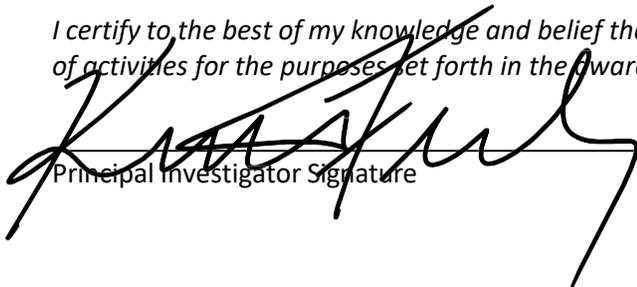**Cover Page**

USDA-ARS Agreement ID:	59-0206-2-126
USDA-ARS Agreement Title:	Breeding Fusarium Head Blight (FHB) Resistant Winter Wheat and Barley
Principle Investigator (PI):	Katherine Frels
Institution:	University of Nebraska
Institution UEI:	HTQ6K6NJFHA6
Fiscal Year:	2022
FY22 USDA-ARS Award Amount:	$140,655
PI Mailing Address:	University of Nebraska, Department of Agronomy and Horticulture 377H Plant Sciences Building, 1875 N 38th St Lincoln, NE 68583-0915
PI E-mail:	kfrels2@unl.edu
PI Phone:	402-472-8709
Period of Performance:	May 1, 2022 – April 30, 2026
Reporting Period End Date:	April 30, 2023

USWBSI Individual Project(s)

USWBSI Research Category[*]	Project Title	ARS Award Amount
BAR-CP	Developing Scab Resistant and Low DON Winter Barley Varieties for the Great Plains	$31,159
HWW-CP	Breeding for Scab Resistance in NE HWW by Optimizing Introgression and Selection	$109,496
	FY22 Total ARS Award Amount	$140,655

I am submitting this report as an: ☒ Annual 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.

_____   7/25/23 _____
Principal Investigator Signature   Date Report Submitted

Project 1: Developing Scab Resistant and Low DON Winter Barley Varieties for the Great Plains

1. **What are the major goals and objectives of the research project?**
   Objectives:
   1- *Increase phenotypic screening of UNL barley germplasm for FHB resistance and DON accumulation.*
   2- *Develop new FHB resistant barley germplasm and develop mapping populations for the University of Nebraska barley program.*
   3- *Improve selection of germplasm with increased FHB resistance and reduced DON accumulation*

   Due to limited acres of barley production and hot, dry weather in the production regions, FHB resistance was not historically a consideration for the UNL barley program. However, we are seeing both expanded interest in growing winter barley in Nebraska and increased risk of FHB due to rainfall at flowering in some areas of Nebraska. Expanding our knowledge of what, if any, native (minor gene, quantitative) tolerance to FHB is present in our unique Great Plains adapted barley germplasm is key to maintaining safe, low-DON feed barley production in Nebraska. We will utilize phenotypic and genotypic analysis to improve our breeding and selection methods for barley FHB resistance, and we will support the development of collaborative FHB screening nurseries for the winter barley community.

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?**
   Obj. 1: Increase phenotypic screening of UNL barley germplasm for FHB resistance and DON accumulation. Our key accomplishment for FY22 funding was completing our second year of testing UNL barley and the Winter Malting Barley Trial in our inoculated mist nursery. We also completed the first year of evaluating the Winter NABSEN. We had good incidence in our nursery with average incidence reaching 48% for the winter NABSEN and 51% for all barley evaluated. Severity was low for most genotypes evaluated likely due to the hot, dry weather that started in June 2022 through June 2023. In Fall 2022, we again planted the UNL barley trials, WMBT, and Winter NABSEN in our inoculated mist nursery, however, this trial was subjected to severe-exceptional drought for the entire growing season. Results from 2023 will be reported when available.

   Obj. 2: Develop new FHB resistant barley germplasm and develop mapping populations for the University of Nebraska barley program. With additional phenotypic data, we have been able to increase the number of crosses made to improve FHB tolerance or maintain tolerance in the UNL barley program. In the March 2023 crossing block, we made 16 successful crosses specifically for improving FHB tolerance. We made 28 additional crosses with a parent demonstrating moderate tolerance in the 2022 Lincoln, NE inoculated nursery. This is a 50% increase in crosses made with a FHB tolerant parent compared to our March 2022 crossing block.

   Obj. 3: Improve selection of germplasm with increased FHB resistance and reduced DON accumulation. We submitted 342 unique barley breeding lines for genotyping with the 3k chip developed by Dr. Fiedler. Data was returned to us in June 2023 and will be processed for use in genomic selection and association mapping after the 2023 harvest season.

b) What were the significant results?

Focusing on the results of the 2022 UNL Elite Barley Trial (EBT), we saw relatively low disease severity due to the hot, dry weather, but very good incidence in the disease nursery. Of the 40 lines in the YET, % were at or below the trial average for severity (Figure 1) and 47% were at or below the trial average (Figure 2) for incidence. DON values averaged 0.64 ppm in the entire trial, but the highest DON content line in this trial had 3.9 ppm. This line, NB21214, is our leading malting-type barley line. However, these DON results combined with the high incidence (60%) observed likely means this line will not be released as a cultivar and we will need to prioritize crossing it with FHB tolerant parents.
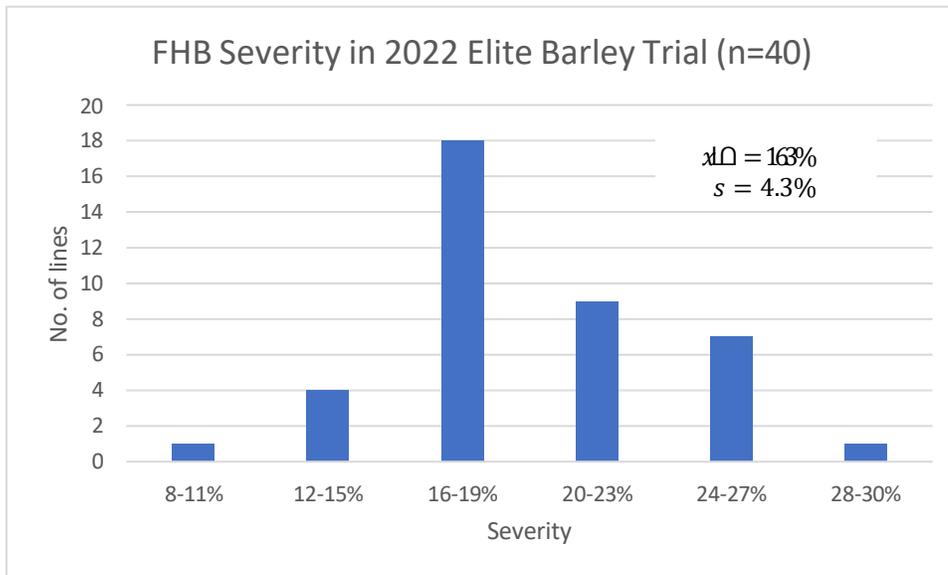

Figure 1. FHB severity in the 2022 Elite Barley Trial. These are the top performing barley breeding lines in the program. They are primarily 6-row feed barley at this time.
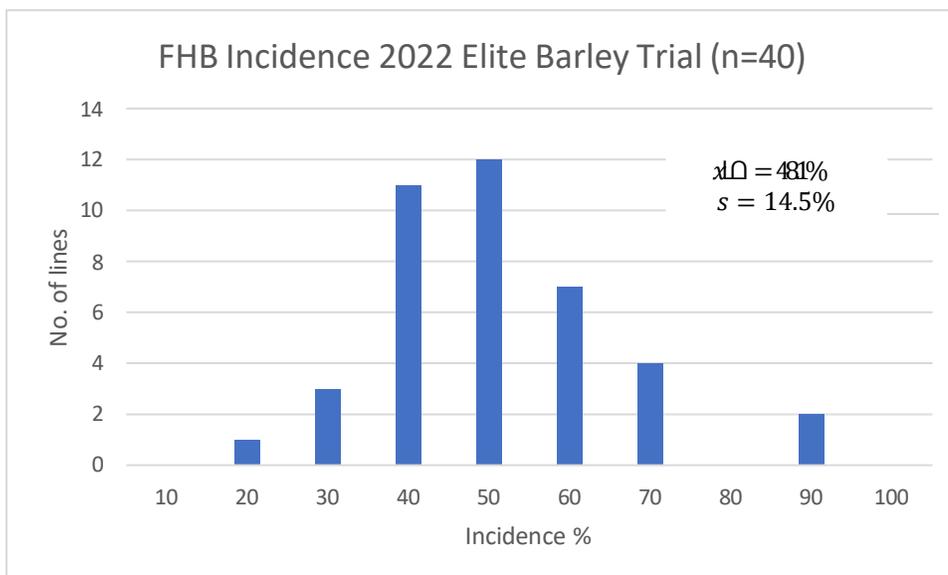

Figure 2. FHB incidence in the 2022 Elite Barley Trial.

Results for Objective 2 and 3 are pending additional inbreeding generations and analysis.

c) **List key outcomes or other achievements.**
We received two barley DH populations (from 2021 cycle) and submitted one new cross with FHB tolerant parents for DH production in spring 2023.

3. **What opportunities for training and professional development has the project provided?**
Ph.D. students Sheryl Sierra and Sydney Graham are leading the UNL barley breeding efforts. They are gaining hand-on experience in designing and improving a breeding pipeline by integrating disease screening data such as FHB while maintaining germplasm necessary for meeting our goals of releasing 6-row feed barley cultivars and eventually 2-row malting cultivars adapted to the southern and central Great Plains. During FY22-23, Sheryl and Sydney implemented designed crossing schemes and initiated genotyping efforts for the UNL barley program.

4. **How have the results been disseminated to communities of interest?**
We continue to use Twitter and university outreach publications to reach our growers. In addition, our variety development and release committee is evaluating how to improve marketing of UNL barley varieties. Barley is discussed at UNL field days and seed days. Dr. Frels presented an update on the UNL barley breeding efforts at the 2022 USWBSI National Fusarium Head Blight Forum last December.

Project 2: Breeding for Scab Resistance in NE HWW by Optimizing Introgression and Selection

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1. **What are the major goals and objectives of the research project?**
Objectives:
   1- Increase the proportion of University of Nebraska wheat breeding crosses targeted towards FHB resistance and DON accumulation.
   2- Improve evaluation and selection of germplasm with increased FHB resistance and reduced DON accumulation.
   3- Evaluate the effect of genotype x fungicide treatments for FHB in our conventional vs intensive management breeding trials.

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?**
Obj. 1: In 2022, we successfully made 20 planned crossed targeting the introgression of major genes such as *Fhb1* and 30 additional crosses with potential for FHB resistance through native (minor gene) resistance. This amounted to 6.5% of the wheat crosses made in 2022. In 2023, we made 26 crosses to introgress *Fhb1* and 7 crosses involving *Fhb6*. 101 other crosses were made using parents that have good phenotypic tolerance to FHB or reduced DON levels. In total, 23% of the crosses we made in 2023 involve some form of tolerance to FHB. We are also working with Drs. Fang Wang and Xiwen Cai to introgress the FHB7 QTL described in Zhang et al. (2022) into Nebraska wheat germplasm that is either 1) top agronomic performing or 2) has either *Fhb1* or *Fhb6* to develop FHB-tolerance gene stacks.

Zhang, W., Danilova, T., Zhang, M., Ren, S., Zhu, X., Zhang, Q., Zhong, S., Dykes, L., Fiedler, J., Xu, S. and Frels, K. Cytogenetic and genomic characterization of a novel tall wheatgrass-derived Fhb7 allele integrated into wheat B genome. 2022. Theoretical and Applied Genetics. https://doi.org/10.1007/s00122-022-04228-3

Obj. 2: We continue to evaluate the FHB regional nurseries for public and private breeding programs in the hard winter wheat region. In 2022, we had good incidence in our inoculated mist nursery, but severity was low for most genotypes evaluated likely due to the hot, dry weather that started in June and lasted through harvest in Nebraska. Some highlights from nurseries that we participate in include:

- NUWWSN- a collaborative trial with winter soft and hard wheat programs evaluated 49 lines in 2022. The average severity for lines was 15 and average incidence was 25 in this trial.
- We evaluated the UNL Wheat State Variety Trial entries for the south-central NE trial. This is the region that is most likely to be affected by FHB. In 2022, the trial contained 27 commercial or precommercial lines. The UNL lines tended to have better FHB resistance than private industry lines entered in this trial, demonstrating the success of our USWBSI project in developing improved resistance in our program through both major and minor gene activity.
- We tested BC_3F_3 *Fhb1* introgression families from Dr. Fang Wang's dissertation research-improved severity compared to the rest of the UNL breeding program and best index values (% of symptomatic spikelets on all heads sampled) in the program.

2022 was the last year we evaluated the wheat preliminary yield trial (PYT) in the inoculated mist nursery. Due to the large number of entries and space limitations in the nursery, we cannot replicate the entries and the heritability of our FHB tolerance scores was low. Starting in 2023, we will utilize multi-

year phenotypic BLUPs from the genotypes in the advanced and elite trials to generate genome-estimated breeding values via genomic prediction for the entries in the PYT.

Obj. 3: Evaluate the effect of genotype x fungicide treatments for FHB in our conventional vs intensive management breeding trials. Due to the hot and dry conditions in Nebraska in 2022, we did not observe FHB symptoms in our conventional management trial. Therefore, we were not able to observe Genotype x Management effects when comparing with the results of the fungicide treated trial. We planted this trial again in 2023, but the location was subjected to severe to exceptional drought throughout the growing season.

b) What were the significant results?

We increased the number of crosses made with parents having either major or minor gene tolerance to FHB by 168% by planning crosses to maximize the use of the parents with major genes or native resistance.

We have identified multiple Nebraska breeding lines with good native tolerance via our phenotypic screening. These results were described in greater detail in the FY21 project final report. Our primary need now is for improved genomic prediction methods for FHB tolerance in our PYT genotypes. Dr. Fang Wang tested different genomic prediction methods for new genotypes. The first predicted a new set of entries with data from prior years (Table 1), while the second predicted the new entries with prior years' data plus a subset of the new entries tested in the current environment (Table 2). The latter scenario appears to be the most accurate. For example, using phenotypic data from 40% of the new entries in 2017 (r=0.16) improved the accuracy of the genomic predictions for FHB severity over leaving out all 2017 data from the model (r=0.025). However, all models had lower prediction accuracy than recommended for use in a breeding program. We will need to test methods for optimizing the training population for improved accuracy prior to deploying these models for selection.

Cross-validation strategy	Size of training dataset	Size of testing dataset	Prediction Accuracy of Severity	Prediction Accuracy of Incidence
NA2015	1112	67	0.17	-0.22
NA2016	974	205	0.13	0.006
NA2017	975	204	0.025	0.10
NA2018	979	200	0.04	0.17
NA2019	913	266	0.22	-0.073

Figure 1. Evaluating the prediction accuracy of predicting new entries with other years data. NA2015 signifies predicting the 2015 entries with all other years of data, NA2016 signifies predicting the 2016 entries with all other years of data, and so on.

Cross-validation strategy	Size of training dataset	Size of testing dataset	Prediction Accuracy of Severity	Prediction Accuracy of Incidence
NA2017(-20%)	1139	40	0.20*	0.22^{ns}
NA2017(-40%)	1099	80	0.16***	0.21^{ns}
NA2017(-60%)	1059	120	0.13***	0.19^{ns}
NA2017(-80%)	1019	160	0.07	0.17
NA2019(-20%)	1125	54	0.32^{ns}	0.04***
NA2019(-40%)	1073	106	0.30*	0.005*
NA2019(-60%)	1019	160	0.28***	-0.02***
NA2019(-80%)	965	214	0.25	-0.05

Figure 2: Two examples of utilizing prediction models generated with previous years' data plus a subset of the experimental year's entries. NA2017 indicates predicting the 2017 new entries with data from 2015, 2016, 2018, and 2019 and subsets of 20-80% of the new experimental entries.

c) **List key outcomes or other achievements.**
We have added a partial FTE postdoc position to optimize genomic prediction methods for selecting FHB tolerant breeding lines in Nebraska germplasm.

3. **What opportunities for training and professional development has the project provided?**
This project has provided partial funding to train several interns (2) and one post-doc in plant production, disease inoculation and data collection and analysis. We encourage graduate students and post docs to attend the Scab Forum, but their schedules did not allow attendance in 2022.

4. **How have the results been disseminated to communities of interest?**
We continue to use Twitter and university outreach publications to reach our growers. FHB and management methods for wheat are discussed at UNL field days and seed days. Dr. Frels presented an update on the effort to select FHB tolerant cultivars for Nebraska producers at the 2022 USWBSI Scab Forum last December.

Publications, Conference Papers, and Presentations

Please include a listing of all your publications/presentations about your <u>FHB work</u> that were a result of funding from your FY22 grant award. Only citations for publications <u>published</u> (submitted or accepted) or presentations <u>presented</u> during the **award period** should be included.

Did you publish/submit or present anything during this award period May 1, 2022 – April 30, 2023?
☒ Yes, I've included the citation reference in listing(s) below.
☐ No, I have nothing to report.

Journal publications as a result of FY22 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.

  Nothing to Report

Books or other non-periodical, one-time publications as a result of FY22 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.

  Nothing to Report

Other publications, conference papers and presentations as a result of FY22 award
Identify any other publications, conference papers and/or presentations not reported above. Specify the status of the publication.

Frels,K., F. Wang, S. Wegulo, X. Cai, V. Belamkar, and P.S. Baenziger. (2022). Selecting for Fusarium resistance in the Great Plains. Proceedings of the 2022 National Fusarium Head Blight Forum; Tampa, FL. December 4-6. Retrieved from: https://scabusa.org/forum/2022/2022NFHBForumProceedings.pdf.
Status: Abstract Published and Presentation made
Acknowledgement of Federal Support: YES

Asif, M., S. Wegulo, J. Stevens, K. Frels, H. Hallen-Adams, and K. Eskridge. Effects of Fungicides and Cultivar Resistance on Fusarium Head Blight of Wheat. Proceedings of the 2022 National Fusarium Head Blight Forum; Tampa, FL. December 4-6. Retrieved from: https://scabusa.org/forum/2022/2022NFHBForumProceedings.pdf.
Status: Abstract Published and Poster Presented
Acknowledgement of Federal Support: YES