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

FY21 Performance Progress Report

Due date: July 26, 2022

Cover Page

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Fiscal Year:	2021
USDA-ARS Agreement ID:	59-0206-0-187
USDA-ARS Agreement Title:	Microbial Interactions with Wheat and Barley Pathogens.
FY20 USDA-ARS Award Amount:	$36,251
Recipient Organization:	Virginia Tech. School of Plant and Environmental Sciences 213 Latham Hall (0390), 220 Ag Quad Ln Blacksburg, VA 24061
DUNS Number:	003137015
EIN:	54-6001805
Recipient Identifying Number or Account Number, if any:	423546
Project/Grant Period:	5/15/21 - 5/14/23
Reporting Period End Date:	5/14/2022

USWBSI Individual Project(s)

USWBSI Research Category*	Project Title	ARS Award Amount
PBG	Managing the Phytomicrobiome for Increased Disease Resistance	$36,251
	FY21 Total ARS Award Amount	$36,251

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.

David C Haak

Principal Investigator Signature

21-JUL-2022

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: Managing the Phytomicrobiome for Increased Disease Resistance

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

Our goal in this two-year USWBSI project is to characterize the host genotype x microbiome x pathogen interactions among wheat varieties with varying resistance levels in a systems framework. The specific objectives are:

1. Describe the metagenomic network landscape for the interactions of host-microbiome-pathogen in susceptible, moderate resistant, and resistant wheat varieties across two locations.
2. Identify microbe and microbial community nodes associated with *Fusarium* load.
3. Identify microbial metabolism genes and correlate their abundance with *Fusarium load.*

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. Describe the metagenomic network landscape for the interactions of host-microbiome-pathogen in susceptible, moderate resistant, and resistant wheat varieties.
This objective was designed to generate information about the interactions between host resistance genotype and the microbial community under pathogen infection. We have generated substantial amplicon data describing these interactions across 18 genotypes and two growth stages and 3 locations. We have also generated preliminary long read metagenomic information for a subset of samples representing infected and uninfected samples. We have overcome numerous challenges and generated tissues from samples collected this growing season. Currently we are extracting DNA from these samples and preparing libraries for sequencing.

Objective 2. Identify microbe and microbial community nodes associated with Fusarium load.
This objective analyses data produced from sequencing samples in objective 1 with an aim to connect microbial taxa with the presence of *Fusarium*. Analysis of these data is underway. Already, we have identified patterns of community shifts associated with the presence of *Fusarium*. We are generating random forest classification models with the aim to connect microbial community changes with *Fusarium* load.

Objective 3. Identify microbial metabolism genes and correlate their abundance with Fusarium load.
Similar with Objective 2, this objective uses data generated in objective 1 to and assign function to microbial reads to identify functions and genes that change in abundance among community members in the presence of fusarium. While still in the early analytical stages we have generated metabolic pathway models from the preliminary metagenomic sequencing.

b) What were the significant results?

Under objective 1, preliminary analyses of these data have revealed some interesting findings. First, we have found that bacteria in this system comprise 85% of the most abundant reads. Among the fungal taxa present *Fusarium spp.* and *Cercospora spp.* are among the most abundant. Predictably, among the *Fusarium spp.* present *F. graminearum* shifts in abundance from 54% to > 90% corresponding with visual FHB ratings. Surprisingly, however, in fields with little to no visual FHB, *F. graminearum* is 1 of 8 *Fusarium spp.* present.

Exploring the communities present under objective 2 we find that these communities are relatively simple, with just 289 bacteria identified (thus far) across samples. Of these notable shifts in these communities occurs among host resistance genotype (R, MR, S), even grown in the same field, with taxa like *Sphingomonas* exhibiting substantially reduced relative abundance in MR and R lines in the presence of *Fusarium.* Conversely members of the genus *Methylobacterium* do not show a change in relative abundance across R, MR, or S hosts. Finally, we find shifts in particular members of these genera, which have previously been identified with suppression of FHB, across host genotype.

While still early investigating shifts in functional roles across these data under objective 3 we have identified over 2,000 unique functional categories. Within these categories functional modules include metabolic pathways, environmental metabolism, cofactor biosynthesis, and importantly, secondary metabolism. Additional analyses are underway.

c) List key outcomes or other achievements.

This work has already uncovered some important interactions among the microbial members in the wheat grain head microbiome. In addition, preliminary results from this work have been used as preliminary data in a large federal proposal. The bioinformatic workflows that are being built for these data have been used in graduate training and education as part of a course “Introduction to Microbial Community Analysis”.

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

This project has supported the training of an undergraduate and graduate student, as well as one postdoctoral research associate. Graduate student training has included cross-training in field, lab, and bioinformatic techniques and approaches. The results from work completed to date have been used to enhance training in a graduate level course in bioinformatics. Finally, results from an undergraduate project investigating endophytes in mature kernels, is being prepared for publication.

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

These results are being prepared for a presentation at the National Fusarium Head Blight Forum 2022. In addition, we have connected with growers about this project through collaborations with the small-grains breeding program at Virginia Tech.

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).

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).

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.