USDA-ARS/
U.S. Wheat and Barley Scab Initiative
FY17 Final Performance Report
Due date: July 31, 2018

Cover Page

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Fiscal Year:	2017
USDA-ARS Agreement ID:	N/A
USDA-ARS Agreement Title:	Enhancing FHB Resistance by Epigenetic Modification of Durum Cultivars.
FY17 USDA-ARS Award Amount:	$ 45,599

USWBSI Individual Project(s)

USWBSI Research Category*	Project Title	ARS Award Amount
DUR-CP	Enhancing FHB Resistance by Epigenetic Modification of Durum Cultivars.	$ 45,599
	FY17 Total ARS Award Amount	**$ 45,599**

SHAHRYAR KIANIAN

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DN: c=US, o=U.S. Government, ou=Department of Agriculture, cn=SHAHRYAR KIANIAN, 0.9.2342.19200300.100.1.1=12001002925979
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Principal Investigator Date

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* MGMT – FHB Management
FST – Food Safety & Toxicology
GDER – Gene Discovery & Engineering Resistance
PBG – Pathogen Biology & Genetics
EC-HQ – Executive Committee-Headquarters
BAR-CP – Barley Coordinated Project
DUR-CP – Durum Coordinated Project
HWW-CP – Hard Winter Wheat Coordinated Project
VDHR – Variety Development & Uniform Nurseries – Sub categories are below:
    SPR – Spring Wheat Region
    NWW – Northern Soft Winter Wheat Region
    SWW – Southern Soft Red Winter Wheat Region

Project 1: *Enhancing FHB Resistance by Epigenetic Modification of Durum Cultivars.*

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

The immediate objectives were to:

1. characterize the epigenetic changes of FHB resistant durum cultivars produced by altering the DNA methylation pattern, and
2. characterize durum cultivars missing portions of chromosome 2A region that may contain a FHB suppressor locus.

The ultimate goal of this project is to enhance the resistance in durum cultivars by removal of a persistent suppression mechanism.

2. What was accomplished under these goals? *Address items 1-4) below for each goal or objective.*

1) major activities and 2) specific objectives

- Transcriptome analysis of tissue from most resistance and susceptible parents (Obje. 1)
- Crossing and advancement of most resistant epigenetic mutants for characterization of the stability of resistance (Obje. 1)
- Advancemnt of irradiated lines by selfing for marker analysis and selection of homozygous deletions for phenotypic characterization (obje. 2)

3) significant results

- Due to cost associated with library construction and sequencing for transcriptome analysis was reduced to 27 samples (initial plan was for 144 samples). Two most resistant lines (E.25.10 & E.25.30), along with a susceptible check (Ben) were inoculated under controlled conditions and tissues were collected at 3 different time points (mock inoculation, 12h and 48h) and from three biological replicates, resulting into 27 samples, and used for RNAseq analysis. Transcriptome analysis (with a mean library size of ≈ 200 bp, quality scores of ≥Q30 and approximate coverage of 32 - 50 million reads per samples) of the selected lines (treated and susceptible check) is continuing to discover the candidate gene(s). Preliminary analysis revealed differential expression (elevated and suppressed ≥5 folds; log2 value) of many (~100) novel candidate genes (uncharacterized proteins) in E.25.10 and E.25.30 as compared to Ben for response to Fusarium infection. In addition, several known genes such as, 3-ketoacyl-CoA synthase, lipid transfer protein (LTP) PR61, argininosuccinate lyase, thaumatin-like protein, ATP synthase also were differentially expressed ≥5 folds in the modified lines (E.25.10 and E.25.30) as compared to the checks. We will verify the expression levels of the candidate genes using real-time quantitative PCR. The uncharacterized candidate genes will be characterized using bioinformatics analysis and reverse genetics approach utilizing the existing durum TILLING population.

- The two most resistant epigenetically modified lines were backcrossed to durum cultivars and advanced for three generations without selection or screening for resistance. The BC_1F_3 plants were evaluated for the first time in the field and greenhouse. Visual scoring for the FHB infection (~10% infection) and DON analysis (1 ppm as compared to 10 ppm in control) revealed stable inheritance of the resistance in the backcross-derived lines. We are performing additional phenotypic evaluations to confirm our results.

- The radiation hybrid population missing portions of chromosome 2A has been advanced to homozygousity. We will marker screening in the coming year to identify overlapping deleted segments of the chromosome and begin the phenotypic screen.

4) key outcomes or other achievements

- Identification of FHB resistant durum lines by epigenetic modification
- Identification of a number of genes possibly underlying the FHB resistance in these modified lines by initial transcriptome analysis
- Initial analysis of backcross lines indicating stability of FHB resistance
- Development of homozygous durum populations missing portions of chromosome 2A

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

Dr. Jitendra Kumar is the postdoctoral scientist on this project. Drs. Dill-Macky and Kianian have been actively advising/mentoring Dr. Kumar as he advances through his career. He has actively participated at various on-campus meeting (e.g., Department of Plant Pathology Seminar series). He has made several oral presentations to various groups (e.g., departmental, Cereal Disease Laboratory, and lab groups) and has been active in preparing publications from his research.

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

Through presentations and publication of outcomes.

Training of Next Generation Scientists

Instructions: Please answer the following questions as it pertains to the FY17 award period. The term “support” below includes any level of benefit to the student, ranging from full stipend plus tuition to the situation where the student’s stipend was paid from other funds, but who learned how to rate scab in a misted nursery paid for by the USWBSI, and anything in between.

1. **Did any graduate students in your research program supported by funding from your USWBSI grant earn their MS degree during the FY17 award period?** NO. There is no support for a graduate student.

   If yes, how many?

2. **Did any graduate students in your research program supported by funding from your USWBSI grant earn their Ph.D. degree during the FY17 award period?** NO. There is no support for a graduate student.

   If yes, how many?

3. **Have any post docs who worked for you during the FY17 award period and were supported by funding from your USWBSI grant taken faculty positions with universities?** No

   If yes, how many?

4. **Have any post docs who worked for you during the FY17 award period and were supported by funding from your USWBSI grant gone on to take positions with private ag-related companies or federal agencies?** No

   If yes, how many?

Release of Germplasm/Cultivars

Instructions: In the table below, list all germplasm and/or cultivars released with full or partial support through the USWBSI during the FY17 award period. All columns must be completed for each listed germplasm/cultivar. Use the key below the table for Grain Class abbreviations. *Leave blank if you have nothing to report or if your grant did NOT include any VDHR-related projects.*

Name of Germplasm/Cultivar	**Grain Class**	**FHB Resistance** (S, MS, MR, R, where R represents your most resistant check)	**FHB Rating** (0-9)	**Year Released**

Add rows if needed.

NOTE: List the associated release notice or publication under the appropriate sub-section in the 'Publications' section of the FPR.

Abbreviations for Grain Classes

Barley - BAR
Durum - DUR
Hard Red Winter - HRW
Hard White Winter - HWW
Hard Red Spring - HRS
Soft Red Winter - SRW
Soft White Winter - SWW

Publications, Conference Papers, and Presentations

Instructions: Refer to the FY17-FPR_Instructions for detailed instructions for listing publications/presentations about your work that resulted from all of the projects included in the FY17 grant. Only include citations for publications submitted or presentations given during your award period. If you did not have any publications or presentations, state 'Nothing to Report' directly above the Journal publications section.

NOTE: Directly below each reference/citation, you must indicate the Status (i.e. published, submitted, etc.) and whether acknowledgement of Federal support was indicated in publication/ presentation. See example below for a poster presented at the FHB Forum:

> Conley, E.J., and J.A. Anderson. 2016. Accuracy of Genome-Wide Prediction for Fusarium Head Blight Associated Traits in a Spring Wheat Breeding Program. In: Proceedings of the XXIV International Plant & Animal Genome Conference, San Diego, CA.
> Status: Abstract Published and Poster Presented
> Acknowledgement of Federal Support: YES (poster), NO (abstract)

Journal publications.

Books or other non-periodical, one-time publications.

Other publications, conference papers and presentations.

Kumar, J., F. S. Xu, E.M. Elias, R. Dill-Macky, and S.F. Kianian. FHB resistance in durum wheat by means of epigenetic modification. Proceeding of the National Fusarium Head Blight Forum. Dec. 2017 poster #24

> Status: Abstract Published and Poster Presented
> Acknowledgement of Federal Support: YES (poster), YES (abstract)