USDA-ARS/
U.S. Wheat and Barley Scab Initiative
FY13 Final Performance Report
July 15, 2014

Cover Page

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Fiscal Year:	FY13
USDA-ARS Agreement ID:	59-0206-9-084
USDA-ARS Agreement Title:	Evaluation, Breeding, and Genomics of FHB Resistance in Wheat and Barley.
FY13 USDA-ARS Award Amount:	$ 148,749

USWBSI Individual Project(s)

USWBSI Research Category[1*]	Project Title	ARS Award Amount
BAR-CP	Development and Characterization of Winter Barley for Resistance to FHB and DON.	$ 29,211
VDHR-SWW	Improving FHB Resistance in SRW Wheat via MAS and Mapping in Native Sources.	$ 112,951
VDHR-SWW	Developing Double Haploids to Expedite Mapping and Enhance FHB Resistance in SRWW.	$ 6,587
	FY13 Total ARS Award Amount	**$ 148,749**

Carl A. Griffey — July 5, 2014

Principal Investigator — Date

[*] MGMT – FHB Management
FSTU – Food Safety, Toxicology, & Utilization of Mycotoxin-contaminated Grain
GDER – Gene Discovery & Engineering Resistance
PBG – Pathogen Biology & Genetics
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: *Development and Characterization of Winter Barley for Resistance to FHB and DON.*

1. What major problem or issue is being resolved relevant to Fusarium head blight (scab) and how are you resolving it?

Scab is a perpetual problem for barley growers and end users in Virginia, and epidemics such as that occurring in 2013 and prior years not only reduce yields and quality, but also reduce barley's marketability due to low test weights and DON toxin. In the past several years, we have developed and advanced populations and pure lines derived from crosses between superior winter barley breeding lines and cultivars from our program with FHB resistant spring barley lines. However, the FHB resistant spring barley lines are not adapted to our environment and lack resistance to other prevalent diseases. Thus, the program has more recently initiated research to characterize and validate QTL and to identify diagnostic markers for FHB resistance in our native barley sources. Current diagnostic markers for FHB resistance (three SSR markers each for QTL on chromosomes 2H and 6H) from spring barley along with markers for other diseases (two SNP markers for leaf rust, three SNP markers for powdery mildew, one SNP marker for net blotch), yield (one SNP marker) and quality (one SNP marker) are being used for MAS in the Virginia Tech barley program. Mapping populations to characterize FHB resistant in the winter barley cultivars Nomini and Eve are being developed in our program. About 300 RIL each for the populations Eve/Doyce and Eve/VA07H-35WS were developed via single seed descent in the greenhouse and seed of the RILs is currently being increased in a grow-out nursery in Aberdeen, Idaho. The RIL mapping populations will be phenotyped for FHB in VA, KY, NC, and possibly in the scab nursery in Nanjing, China in 2015 and 2016. A Thoroughbred/Nomini mapping population will be used to characterizing FHB resistance in hulled barley cultivar Nomini. It is currently in the F_3 generation, and is being advanced to develop RILs in the greenhouse at Virginia Tech. A doubled haploid population also will be developed from a cross of Secretariat/Nomini in 2015 and subsequently will be used in this study. The Thoroughbred/Nomini and Secretariat/Nomini populations will be evaluated for FHB in the field (KY, NC, VA, and possibly in Nanjing, China) in 2016 and/or 2017. The genotyping will be done at Virginia Tech in collaboration with USDA-ARS, Fargo, ND.

Breeding populations derived from crosses made with FHB resistance sources (AC Alberte, Atahulpa, MN Brite, and Fredrickson) are in advanced generations. This season (2013-14), three elite FHB resistant barley lines were evaluated in Virginia's State Variety Trial, 19 advanced FHB resistant lines were evaluated in a preliminary yield test, 21 FHB resistant lines were tested in an observation yield trial, and 75 pure lines and 80 populations were evaluated for FHB resistance in our scab nursery and advanced in the program.

2. List the most important accomplishments and their impact (i.e. how are they being used) to minimize the threat of Fusarium Head Blight or to reduce mycotoxins. Complete both sections; repeat sections for each major accomplishment:

Accomplishment:

We have continued to make progress improving resistance to FHB. Marker analysis of Nomini, VA06H-48, and Eve showed that the FHB marker haplotypes for these genotypes are different than the haplotypes of the FHB resistant genotypes Chevron and Fredrickson. Recombinant inbred lines are being developed from crosses between Thoroughbred/Nomini, Eve/Doyce and Eve/VA 07H-35WS. Double haploid lines will be developed from Secretariat/Nomini. The recombinant inbred lines will be used to characterize FHB resistance in Nomini and Eve. In addition, pure lines from populations derived from crosses between known FHB resistant spring barley lines and adapted winter barely lines are being evaluated for FHB resistance and agronomic performance.

Impact:

Screening resistant genotypes in our program with molecular markers on chromosomes 2H and 6H for FHB and DON revealed QTL regions which may confer unique resistance in the Virginia Tech germplasm. Use of diagnostic markers for FHB and also to select for resistance to other diseases, yield, and quality will enhance variety development in the Virginia Tech barley program. Breeding populations and pure lines derived from crosses to incorporate FHB resistance from Chevron, Frederickson, Atahulpa, Tibetan, Island, ND 20448, AC Alberte, MN-Brite, Quest (M1222), and other elite genotypes are being advanced and tested in the program and will provide for superior cultivars and/or germplasm.

Project 2: *Improving FHB Resistance in SRW Wheat via MAS and Mapping in Native Sources.*

1. What major problem or issue is being resolved relevant to Fusarium head blight (scab) and how are you resolving it?

Development of competitive wheat cultivars having FHB resistance derived from exotic sources, such as Fhb1 derived from Sumai 3, has been hindered by linkage drag. In addition, progress has been hindered by the lack of adequate characterization and validation of FHB resistance in adapted native sources and unavailability of diagnostic markers needed to implement marker assisted incorporation and pyramiding of diverse QTL for FHB resistance. FHB resistance in the SRW wheat cultivar Massey was mapped and resistance in Ernie was validated and fine mapped. Mapping of FHB resistance in the SRW wheat cultivars Roane and Jamestown was conducted in a northern and southern set of RILs and phenotyped by cooperators in AR, GA, KY, LA, MD, MO, NC, and VA for two years. Double haploid lines of a Pioneer26R46/Tribute mapping population were evaluated for FHB phenotypes in AR, KY, NC and VA for a second year during 2014. Marker assisted selection (MAS) is being used to both enhance the level of scab resistance and to accelerate the development of superior scab resistant cultivars. Markers linked to scab resistance genes located on wheat chromosomes 3BS (*Fhb1*) and 5AS of Ning 7840 (Sumai 3 derivative), 1B of Jamestown, 2B, 3BSc, 4B and 5A of Ernie and 3BSc of Massey are being used to screen, characterize and select parents and their progeny for scab resistance genes. Twelve top cross

populations developed between 2008 and 2010 with either Ernie or Ning 7840 (or other Sumai3 derivatives) in their pedigrees were screened via MAS to enrich FHB resistance in these breeding populations. In 2013, FHB breeding materials evaluated in scab nursery and/or field tests included: 199 populations, 1500 headrows, and more than 800 pure lines.

2. List the most important accomplishments and their impact (i.e. how are they being used) to minimize the threat of Fusarium Head Blight or to reduce mycotoxins. Complete both sections; repeat sections for each major accomplishment:

Accomplishment:

In a Pioneer 25R47/ Jamestown population, three putative QTL were associated with FHB on chromosomes 1B, 3BS, and 5A. A QTL on 1B in Jamestown was associated with resistance to FHB, whereas alleles in Jamestown for a QTL on chromosome 5A were associated with FHB susceptibility. Markers associated with these QTL are Xwmc500 (chromosome 1B), Xgwm285 (chromosome 3BS), and Xgwm304 (chromosome 5A). The QTL on chromosome 1B from Jamestown reduces FHB severity by 10% and DON content by 2 ppm. Genotypes having Jamestown alleles at the 5A QTL had 5% higher FHB severity, while Jamestown alleles at the 3BS QTL had a small to negligible effect on fusarium damaged kernel (FDK). Since resistance to scab is quantitative and additive in nature, combining the QTL on chromosome 1B from Jamestown with other validated FHB resistance QTL, such as *Fhb1*, will facilitate pyramiding and further enhancement of FHB resistance. The results showed that selection of Jamestown alleles for the QTL on chromosome 1B and negative selection for alleles of Jamestown for the QTL on 3BS and 5A will reduce FHB severity, FDK, and DON content.

A subset of 42 extreme lines (21 resistant and 21 susceptible RILs) in the Pioneer 25R47/ Jamestown population was genotyped with 9,000 SNP markers at Monsanto Company. Selective genotypic analysis identified two additional putative QTL for FHB on chromosomes 2B and 6D in addition to those identified on 1B, 3BS, and 5A. The remaining 142 RILs are currently being genotyped using 90,000 SNP markers in collaboration with USDA-ARS genotyping laboratory at Raleigh, NC and Fargo, ND. Further genotyping with these SNP markers and analysis of markers associated with QTL for FHB resistance in other populations (FG95195/Jamestown, 170 RILs and Jamestown/LA97UC113-124, 77 RILs) already assessed for FHB resistance will facilitate validation of these QTL and help identify additional QTL and diagnostic markers associated with these QTL. The diagnostic markers Xwmc500 (chromosome 1B) and Xgwm304 (chromosome 5A) identified in the study are being used for MAS in the Virginia Tech wheat breeding program.

During 2012-2013, phenotypic data was collected in a Pioneer26R46/Tribute double haploid (DH) population in AR (Milus), KY (Van Sanford), NC (Murphy), MD (Costa), and VA (Griffey). Twenty two polymorphic SSR markers were used to genotype the population. Single marker analysis indicated that SSR markers Xbarc261 (chromosome 2D), Xgwm285 (3BSc), Xwmc471 (3BSc), Xwmc418 (3BSc), and Xwmc443 (5D) were significantly associated with FHB incidence at multiple locations, and markers Xgwm47 (2A), Xbarc95

(2D), Xbarc261 (2D), Xgwm285 (3BSc), Xmmc471 (3BSc), Xwmc418 (3BSc), Xwmc805 (5A, 5D), and Xwmc443 (5D) were significantly associated with FHB severity across multiple locations. Similarly, the SSR marker Xgwm47 (2A) was significantly associated with DON content across several locations. Although the SSR markers Xbarc95 and Xbarc261 on chromosome 2D were significantly associated with FHB incidence and FHB severity in single marker analysis, the markers were not linked to the QTL in composite interval mapping. Since the markers are linked to dwarfing gene *Rht8* and the photoperiod sensitivity gene *Ppd-D1*, more markers are needed to confirm a pleiotropic (joint) effect or whether an independent QTL for FHB resistance is present in this region. Based on single marker analysis, the putative QTL for FHB resistance on chromosomes 2A, 5A, and 5D might be unique to Tribute. The population is being genotyped with 90,000 SNP markers in collaboration with USDA-ARS genotyping laboratory at Raleigh, NC and Fargo, ND. During 2013-14, a second year of phenotypic data is being collected by collaborators in AR (Milus), KY (Van Sanford), NC (Murphy), and VA (Griffey). The results from the first year will be validated and diagnostic marker(s) will be identified for use in MAS breeding in the Virginia Tech and other wheat breeding programs.

Impact:

Progress in introgression and pyramiding of FHB resistance in SRW wheat has been hindered by lack of knowledge of the identity and diversity of QTL governing resistance, lack of QTL validation, and lack of reliable diagnostic markers to deploy in MAS. Identification and validation of consistent QTL in native sources such as Ernie (on chromosomes 2B, 3BSc, 4B and 5A), Massey (3BSc), and Jamestown (1B) has potential to enhance both breeding effectiveness and level of FHB resistance in SRW wheat. Once these QTL and those postulated in Tribute are validated and diagnostic markers identified, they can be used for MAS to enhance scab resistance in wheat breeding programs.

Project 3: *Developing Double Haploids to Expedite Mapping and Enhance FHB Resistance in SRWW.*

1. What major problem or issue is being resolved relevant to Fusarium head blight (scab) and how are you resolving it?

Research is focused on shortening breeding cycles through the development of doubled haploid populations and enhancing FHB resistance via MAS breeding efforts in selection of parents, designing crosses, gene introgression and pyramiding, population enrichment, and selection of pure lines. Marker haplotypes of parents for validated FHB resistance QTL and other traits of importance such as dwarfing genes, disease and insect resistance, rye translocations, and quality are being assessed and utilized to enhance breeding efficiency. Markers linked to scab resistance genes located on wheat chromosomes 1B of Jamestown, 2DL, 3BS (*Fhb1*) and 5AS of Ning 7840 (Sumai 3 derivative), 2B, 3BSc, 4B and 5A of Ernie are being used to screen, characterize and select parents and their progeny for scab resistance genes. Seven crosses were made to pyramid *Fhb1*, and QTL on chromosomes

5AS, 2DL, and 3BSc (Ernie) in spring 2014. One or more of these crosses will used to develop doubled haploid populations at Heartland Plant Innovations. Phenotyping of the populations will be conducted in AR, GA, KY, LA, NC, and VA.

2. List the most important accomplishments and their impact (i.e. how are they being used) to minimize the threat of Fusarium Head Blight or to reduce mycotoxins. Complete both sections; repeat sections for each major accomplishment:

Accomplishment:

A top cross MD03W61-09-7 (*Fhb1*) / Jamestown (QTL on 1B) // GA04570-10E46 was made in spring 2013. The doubled haploid (DH) population consisting of 250 lines was developed at Heartland Plant Innovations in Manhattan, KS in 2013. The DH lines were genotyped for the 1B QTL of Jamestown, *Fhb1*, *Lr9*, *Sbm1*, and 1B.1R in our lab and evaluated in headrows at Warsaw in 2014. Selected DH lines will be grown to increase seed stocks, and then will be shared with other cooperators. A single cross MDC07027-12-24 / VA11W-108 was made in spring 2013, and up to 200 DH lines containing gene *Fhb1* and characterized for QTL on chromosomes 2DL and 5AS are currently being developed at Heartland Plant Inovations in Manhattan, KS. The DH lines will be evaluated in headrows in 2015 and selected lines will be genotyped and subsequently shared with cooperators.

Impact:

Use of a combination of DH and MAS technologies will shorten the breeding cycle and enhance development of FHB resistance germplasm and varieties. The DH lines selected on the basis of marker genotypes and desirable agronomic traits will be distributed to cooperating breeders for use in their programs. Select DH lines will be advanced and tested in routine yield nurseries and superior lines will be released as varieties.

Include below a list of all germplasm or cultivars released with full or partial support of the USWBSI during the FY13 award period. List the release notice or publication. Briefly describe the level of FHB resistance.

None

Include below a list of the publications, presentations, peer-reviewed articles, and non-peer reviewed articles written about your work that resulted from all of the projects included in the FY13 grant. Please reference each item using an accepted journal format. If you need more space, continue the list on the next page.

Berger, G., A. Green, P. Khatibi, W. Brooks, L. Rosso, S. Liu, S. Chao, C. Griffey, and D. Schmale, III. 2014. Characterization of Fusarium Head Blight (FHB) Resistance and Deoxynivalenol Accumulation in Hulled and Hulless Winter Barley. Plant Dis. 98(5):599-606.

Berger, G., S. Liu, M. Hall, W. Brooks, S. Chao, G. Muehlbauer, B.-K. Baik, B. Steffenson and C. Griffey. 2013. Marker-trait associations in Virginia Tech winter barley identified using genome-wide mapping. Theor. Appl. Genet. 126:693-710.

Khatibi, P., G. Berger, J. Wilson, W. Brooks, N. McMaster, C. Griffey, K. Hicks, N. Nghiem, and D. Schmale, III. 2014. A comparison of two milling strategies to reduce the mycotoxin deoxynivalenol in barley. Jour. of Agri. and Food Chem. DOI:10.1021/jf501208x

Liu, S., C.A. Griffey, M.D. Hall, A.L. McKendry, J. Chen, W.S. Brooks, G. Brown-Guedira, D. Van Sanford, and D.G. Schmale. 2013. Molecular characterization of field resistance to Fusarium head blight in two U.S. soft red winter wheat cultivars. TAG. DOI 10.1007/s00122-013-2149-y

Presentations

Malla, S., C. Griffey, G. Milus, J.P. Murphy, A. Clark, D. Van Sanford, J. Costa, N. McMaster, and D. Schmale III. 2013. Mapping FHB resistance in native SRW wheat cultivar Tribute. Pp. 25. In: S. Canty, A. Clark, Y. Salat, and D. Van Sanford (Eds.), Proceedings of the 2013 National Fusarium Head Blight Forum; Dec. 3-5; Milwaukee, WI.

Wright, E., C. Griffey, S. Malla, D. Van Sanford, S. Harrison, J.P. Murphy, J. Costa, G. Milus, J. Johnson, A. McKendry, D. Schmale III, A. Clark, and N. McMaster. 2013. Characterization of FHB resistance in SRW wheat Roane and Jamestown NAM populations. In: S. Canty, A. Clark, Y. Salat, and D. Van Sanford (Eds.), Proceedings of the 2013 National Fusarium Head Blight Forum; Dec. 3-5; Milwaukee, WI.