

**USDA-ARS/
U.S. Wheat and Barley Scab Initiative
FY09 Final Performance Report
July 15, 2010**

Cover Page

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Fiscal Year:	2009
USDA-ARS Agreement ID:	59-0790-7-078
USDA-ARS Agreement Title:	Contribution of Local Inoculum Sources to Regional Atmospheric Populations of <i>G. zeae</i> .
FY09- USDA-ARS Award Amount:	\$ 99,044

USWBSI Individual Project(s)

USWBSI Research Category*	Project Title	ARS Adjusted Award Amount
FSTU	Diagnostic Testing Services for Deoxynivalenol in the Eastern United States.	\$ 64,980
PBG	Mycotoxin Potential and Aggressiveness of FHB Pathogens used for USWBSI Research.	\$ 21,854
MGMT	Within-Field Inoculum from Corn Debris and the Management of FHB/DON.	\$ 12,210
	Total Award Amount	\$ 99,044

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 Winter Wheat Region
 SWW – Southern Sinter Wheat Region

Project 1: *Diagnostic Testing Services for Deoxynivalenol in the Eastern United States.*

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

There is a growing need to develop and expand USWBSI diagnostic testing services for mycotoxins throughout the United States. DON testing services are vital to the development of new varieties of wheat and barley with reduced mycotoxin potential and are necessary to identify and/or exclude appropriate strategies for managing FHB. FY09 DON testing services at Virginia Tech provided analytical services necessary to develop new cultivars of wheat and barley with reduced potential for DON contamination and to improve chemical and cultural practices necessary to reduce DON contamination in wheat and barley.

2. List the most important accomplishment and its impact (i.e. how is it 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 FY09, DON was quantified from over 6,000 samples (runs, internal checks (controls), and re-runs) of wheat and barley from four USWBSI investigators (Bergstrom, Cowger, Griffey, and Grybauskas) in four states (New York, North Carolina, Virginia, and Maryland). DON was also quantified from over 400 samples for industry (Osage BioEnergy in Hopewell, Virginia), and Virginia Tech researchers (Rideout and Stromberg). Most of the samples received for testing in FY09 were 100g kernel lots from FHB field trials, but some were ground 5-25g samples from greenhouse experiments. Extraction, clean-up, and quantification of DON were conducted following standard protocols using a GC/MS. DON testing services were managed by two talented scientists (Patricia Gundrum and Diane Reaver) and four dedicated undergraduates.

Impact:

The goals of this work were to provide analytical services necessary to develop new cultivars of wheat and barley with reduced potential for DON contamination and to facilitate DON testing that will improve chemical and cultural practices necessary to reduce DON contamination in wheat and barley. This work directly addresses Goal #1 of the Action Plan to 'Provide analytical support for DON/trichothecene quantitation for Initiative's stakeholders'. We are providing DON testing services for approximately 15,000 wheat and barley samples from at least four USWBSI investigators in four states. Schmale routinely interacts with stakeholders in VA to discuss new diagnostic technologies for DON and related management strategies for FHB, an effort aligned with Goal #2 of the Action Plan to 'Provide requisite information on DON/ trichothecene safety issues to producers, millers, researchers, risk assessors and regulators'.

Project 2: *Mycotoxin Potential and Aggressiveness of FHB Pathogens used for USWBSI Research.*

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

USWBSI investigators often rely on a ‘favorite’ set of strains of FHB pathogens for their field and greenhouse trials, yet little is known about the mycotoxin potential and aggressiveness of these strains. Knowledge of the aggressiveness and mycotoxin potential of FHB pathogens used in USWBSI research may assist in selecting appropriate strains for field and greenhouse experiments and may help explain observed variations in FHB and/or DON among field and greenhouse trials. We are screening strains that have been used (or are currently being used) in inoculations of field and/or greenhouse trials to support USWBSI VHDR research. The aims of the project are to: (1) measure the aggressiveness of individual strains on a series of susceptible and moderately-resistant cultivars of wheat and barley, (2) determine the relative concentrations of trichothecene mycotoxins produced by these strains following a series of controlled inoculations, and (3) identify individual strains to the level of species, based on both biological and phylogenetic species recognition.

2. List the most important accomplishment and its impact (i.e. how is it 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 received 44 strains of FHB pathogens from 7 USWBSI investigators (P. Murphy, S. Zong, P. Paul, T. Friesen, G. Bergstrom, C. Griffey, and G. Milus. Receipt, handling, and transport of the strains were conducted under USDA-APHIS permit P526P-09-02605. Trichothecene mycotoxin genotyping of the strains has shown that all three genotypes (3ADON/DON, 15ADON/DON, and NIV) are present in the collection of 44 strains. Portions of three genes have been amplified for nearly all of these strains, and sequencing and phylogenetic analysis are underway. Inoculations of greenhouse-grown wheat plants and resulting mycotoxin analyses are scheduled for FY10.

Impact:

The project directly addresses Goal #1 of the Action Plan to ‘Characterize genetic variation in the pathogen population with regard to aggressiveness toward plants and mycotoxin potential’. We are screening strains representing nearly a decade of FHB research in the U.S. Knowledge of the aggressiveness and mycotoxin potential of FHB pathogens used in USWBSI research may assist in selecting appropriate strains for field and greenhouse experiments and may help explain observed variations in FHB and/or DON among field and greenhouse trials.

Project 3: *Within-Field Inoculum from Corn Debris and the Management of FHB/DON.*

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

Knowledge of the relative contribution of within-field inoculum sources of *G. zeae* to infection of local wheat and barley is important for developing and/or excluding strategies for managing FHB. Our experimental objective is to quantify the relative contribution of within-field corn debris as an inoculum source of *G. zeae* for FHB and DON contamination in 20 variable wheat or barley environments over two years, all in regions where corn is the predominant crop in the agricultural landscape and corn debris is left on the land surface over large areas. In 2009, research was conducted in a total of ten commercial wheat fields in five states (Illinois, Missouri, Nebraska, New York, and Virginia), each following a non-susceptible crop. Over these environments we encountered six severe epidemics (in Illinois, Missouri, and Virginia), two moderate epidemics (in New York), and two mild epidemics (in Nebraska). Each field contained small experimental plots with local corn debris, infested residue, or no residue (control). Wheat heads above each microplot were rated at approximately soft dough stage for FHB incidence, severity, and index. At grain maturity, at least 100 heads from each experimental plot were harvested, dried and shipped to Cornell where grain was threshed from a subsample of heads and sent to Virginia Tech for DON analysis.

2. List the most important accomplishment and its impact (i.e. how is it being used) to minimize the threat of Fusarium head blight or to reduce mycotoxins. Complete both sections (repeat sections for each major accomplishment):

Accomplishment:

DON levels did not differ significantly between small experimental plots with or without corn debris in any of the ten commercial wheat fields (five states, two locations per state). FHB was observed in four fields, and DON was detected in every field. FHB symptoms at soft dough were a poor predictor of DON ppm, *G. zeae* was recovered from a large percentage of mature spikes at every location except Lincoln and Mead, NE, suggesting late infections in some fields with no or few symptoms at soft dough. Local corn debris resulted in significant increases over no debris in FHB only in one field (Riner, VA), and did not result in a significant increase in DON in any location.

Impact:

FHB and DON contamination may not be significantly impacted by a reduction (e.g., tillage) of corn debris from single wheat fields in major corn-producing regions. Our research addresses Goal #3, to develop a full understanding of specific factors influencing infection and toxin accumulation that can be used to develop the next generation of scab and DON risk assessment measures. Ultimately, our efforts will aid in developing and/or excluding strategies for managing FHB and will help refine forecasting/risk assessment models for FHB.

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 grant. Please reference each item using an accepted journal format. If you need more space, continue the list on the next page.

Bergstrom, G.C., Waxman, K.D., Schmale, D.G., Bradley, C.A., Sweets, L.E., Wegulo, S.N., and Keller, M.D. 2009. Effects of within-field corn debris in microplots on FHB and DON in ten U.S. wheat environments in 2009. Page 22 in Proc. 2009 National Head Blight Forum, Orlando, FL. Date of Meeting.

Keller, M.D., Waxman, K.D., Bergstrom, G.C., and Schmale, D.G. 2010. Local distance of wheat spike infection by released clones of *Gibberella zeae* disseminated from infested corn residue. Plant Disease. *In press*.

Keller, M.D., Thomason, W.E., and Schmale, D.G. 2009. Influence of crop residues and disease resistance on FHB in Virginia wheat. Page 59 in Proc. 2009 National Head Blight Forum, Orlando, FL.

Khatibi, P., Griffey, C.A., and Schmale, D.G. 2009. Mitigating deoxynivalenol contamination in hulless barley and fuel ethanol co-products. Phytopathology 99: S204.

Liu, S., Brooks, W., Chao, S., Griffey, C., Hall, M., Gundrum, P., Berger, G., Khatibi, P., and Schmale, D.G. 2009. Association analyses of SNP markers with scab resistance in winter feed barley. Page 133 in Proc. 2009 National Head Blight Forum, Orlando, FL.

Schmale, D.G. and Munkvold, G.P. 2009. Mycotoxins in crops: A threat to human and domestic animal health. The Plant Health Instructor DOI: 10.1094/PHI-I-2009-0715-01.

Schmale, D.G. 2009. Linking field and atmospheric populations of toxigenic fusaria. Page 181 in Proc. 2009 National Head Blight Forum, Orlando, FL.

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Project: Diagnostic Testing Services for Deoxynivalenol in the Eastern United States.

**FY09 FPR – USWBSI ADDENDUM
DON Service Labs – Quality Control Data**

Insert below Quality Control Data/Results from the FY09 Award Period (May 09-May 10):

Quality control data is collected at Virginia Tech via the blind testing of samples with unknown DON levels (coordinated by J. Gillespie , NDSU). We also test subsamples of grain lots in each GC/MS run with low, medium, and high DON (to test for consistency among GC/MS runs) and run known standards at the beginning and end of the GC/MS run (to test for consistency within a GC/MS run).

We participated in the blind testing of the following samples during the funding period. Comparative data was provided by J. Gillespie via email (James.Gillespie@ndsu.edu). Lab ID ‘S’ is the Virginia Tech lab (highlighted in grey).

a. Barley 84 and Malt 84

Lab ID	Method	Sample	Results (ppm DON)
A	GC/MS	Barley 84	0.58
		Malt 84	0.00
D	Diagnostix EZ-TOX	Barley 84	0.80
		Malt 84	<0.10
E	GC/ECD	Barley 84	0.60
		Malt 84	<0.20
F	GC/MS #1	Barley 84	0.57
		Malt 84	0.14
F	GC/MS #2	Barley 84	0.57
		Malt 84	0.13
K	Neogen 5/5	Barley 84	0.20
		Malt 84	0.00
M	Diagnostix EZ-TOX	Barley 84	0.55
		Malt 84	0.05
N	EZ-TOX	Barley 84	0.64
		Malt 84	0.10
O	EZ-TOX	Barley 84	0.50
		Malt 84	0.00
P	EZ-TOX	Barley 84	0.49
		Malt 84	0.02
Q	Biotek EZ-TOX	Barley 84	0.62
		Malt 84	0.10
S	GC/MS	Barley 84	0.41
		Malt 84	0.12

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b. Barley 85 and Malt 85

Lab ID	Method	Sample	Results (ppm DON)
A	GC/MS	Barley 85	0.00
		Malt 85	0.00
D	Diagnostix EZ-TOX	Barley 85	<0.10
		Malt 85	<0.10
E	GC/ECD	Barley 85	<0.20
		Malt 85	<0.20
F	GC/MS #1	Barley 85	0.11
		Malt 85	N.D.
F	GC/MS #2	Barley 85	0.10
		Malt 85	N.D.
K	Neogen 5/5	Barley 85	0.00
		Malt 85	0.00
M	Diagnostix EZ-TOX	Barley 85	0.01
		Malt 85	0.01
N	EZ-TOX	Barley 85	0.10
		Malt 85	0.10
O	EZ-TOX	Barley 85	0.00
		Malt 85	0.00
P	Neogen 5/5	Barley 85	0.04
		Malt 85	0.01
Q	Biotek EZ-TOX	Barley 85	0.10
		Malt 85	0.10
S	GC/MS	Barley 85	0.12
		Malt 85	0.07