

**USDA-ARS/  
U.S. Wheat and Barley Scab Initiative  
FY14 Final Performance Report  
July 15, 2015**

**Cover Page**

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<b>Fiscal Year:</b>	FY14
<b>USDA-ARS Agreement ID:</b>	59-0206-2-082
<b>USDA-ARS Agreement Title:</b>	Diagnostic Testing Services for Deoxynivalenol in the Eastern U.S.
<b>FY14 USDA-ARS Award Amount:</b>	\$ 105,877

**USWBSI Individual Project(s)**

<b>USWBSI Research Category*</b>	<b>Project Title</b>	<b>ARS Award Amount</b>
FSTU	Diagnostic Testing Services for Deoxynivalenol in the Eastern U.S.	\$ 66,550
PBG	Enzymatic Detoxification of Deoxynivalenol.	\$ 39,327
	<b>FY14 Total ARS Award Amount</b>	<b>\$ 105,877</b>

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Principal Investigator

\_\_\_\_\_  
Date

\* MGMT – FHB Management

FSTU – Food Safety, Toxicology, & Utilization of Mycotoxin-contaminated Grain

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

WES-CP – Western 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:** *Diagnostic Testing Services for Deoxynivalenol in the Eastern U.S.*

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

Demand continues for USWBSI diagnostic testing services for mycotoxins across 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. FY14 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 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 FY14, DON data was delivered for 4,306 wheat and barley samples from six USWBSI investigators (Laskar, Glover, Griffey, Grybauskas, Rideout, and Schmale). The testing number does not include controls, checks, and re-runs. Most of the samples tested in FY14 were 100g kernel lots from FHB field trials, but some were smaller lots (~5g samples) from laboratory experiments. We also processed samples associated with DON during ethanol production and DON detoxification studies. Extraction, clean-up, and quantification of DON were conducted following standard protocols using a GC/MS. DON testing services were coordinated, supported, and managed by a talented research associate (Niki McMaster). The Schmale Lab at Virginia Tech continues to be committed to the long-term management of a successful and productive mycotoxin testing lab for the USWBSI.

**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 wheat and barley samples from USWBSI investigators. Schmale routinely interacts with stakeholders in the mid-Atlantic 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:** *Enzymatic Detoxification of Deoxynivalenol.*

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

The ultimate goal of our project is to reduce mycotoxin contamination in wheat and barley used for feed and food. This research aims to provide an efficient control strategy for reducing mycotoxins in wheat and barley throughout the United States. We are testing enzymes from unique sources for their ability to detoxify DON, and working to identify novel enzymes (and microbes that produce them) to destroy DON.

**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 new Ph.D. student, Ms. Nina Wilson, started on the project and has made excellent progress. First, in an effort to develop a tool to screen potential candidate DON detoxification enzymes, we have engineered a number of yeast strains to be sensitive to DON. One of these strains is sensitive to DON at 100 ppm, but not sensitive to de-epoxy DON (the detoxified product) at the same concentration. This tool is being used to screen enzymes for DON detoxification. Second, we have identified a new promising candidate epoxide hydrolase. This candidate is a styrene oxide isomerase that destroys epoxides. Third, we have been working with DON detoxifying microorganisms that were isolated from the field and cultured in the presence of high concentrations of DON. Three mixed cultures of microbes, and one pure culture, were able to consistently detoxify nearly all of the DON (100 ppm) in laboratory experiments. These microbes are currently being characterized, and exposed to DON and related derivatives in follow-up lab experiments.

**Impact:**

This research will offer new strategies for detoxifying DON in wheat and barley products. We have initiated collaborations with Vireol Bio Energy and AgBiome to explore opportunities to transfer this technology to the commercial sector in the future. The proposed work directly addresses the PBG goal to ‘Develop new strategies for reducing impact of FHB disease and mycotoxin contamination in barley and wheat’, and the PBG research need to ‘identify enzymes to detoxify DON’.

### **Training of Next Generation Scientists**

**Instructions:** Please answer the following questions as it pertains to the FY14 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 FY14 award period?**

No

**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 FY14 award period?**

No, but a new Ph.D. student (Nina Wilson) started on the FY14 project supported by the USWBSI.

**If yes, how many?**

- 3. Have any post docs who worked for you during the FY14 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 FY14 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?**

**Include below a list of all germplasm or cultivars released with full or partial support of the USWBSI during the FY14 award period. List the release notice or publication. Briefly describe the level of FHB resistance. *If not applicable because your grant did NOT include any VDHR-related projects, enter N/A below.***

N/A

**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 FY14 grant. Please reference each item using an accepted journal format. If you need more space, continue the list on the next page**

Brooks, W.S., M. E. Vaughn, G. L. Berger, C. A. Griffey, W. E. Thomason, R. M. Pitman, S. Malla, J. E. Seago, D. W. Dunaway, E. G. Hokanson, H. D. Behl, B. R. Beahm, D. G. Schmale, N. McMaster, T. Hardiman, J. T. Custis, D. E. Starner, S. A. Gulick, S. R. Ashburn, E. H. Jones, Jr., D. S. Marshall, M. O. Fountain, T. D. Tuong, M. J. Kurantz, R. A. Moreau, and K. B. Hicks. 2014. Registration of 'Atlantic' Winter Barley. *Journal of Plant Registrations* 8: 236-241. DOI:10.3198/jpr2014.04.0019crc

Prussin, A.J., Marr, L.C., Schmale, D.G., and Ross, S.D. 2015. Experimental validation of a long-distance transport model for plant pathogens: Application to *Fusarium graminearum*. *Agricultural and Forest Meteorology* 203:118-130.

Khatibi, P.A., McMaster, N., Musser, R., and Schmale, D.G. Survey of Mycotoxins in Corn Distillers' Dried Grains with Solubles from Seventy-Eight Ethanol Plants in Twelve States in the U.S. in 2011. Participated in ISM Workshop -Training Course titled "Detection techniques for mycotoxins in the food/feed chain" in Bari, Italy, October 6-10, 2014. Audience: Academia, Government. Scope: International. Reason: course participant. Number of Times Delivered: 1. Audience Size: 40. Hours of Instruction: 0.25

Wilson, Nina, 2014. Modification of the Mycotoxin Deoxynivalenol with Enzymes and Microorganisms. Poster and presentation for the International Society for Mycotoxicology (ISM) Workshop: Detection Techniques for Mycotoxins in the food/feed chain in Bari, Italy, October 6-10, 2014 Audience: Academia and Industry. Scope: International. Reason: Learning Experience and Informational. Number of Times Delivered: 1. Audience: ~35.

Wilson, Nina, 2014. Modification of the Mycotoxin Deoxynivalenol with Enzymes and Microorganisms. Poster and presentation for the USWBSI in St. Louis, Missouri, December 7-9, 2014. Audience: Academia, Industry, and Farmers. Scope: National. Reason: Learning Experience and Informational. Number of Times Delivered: 1. Audience: ~175.

PI: Schmale, David

Project: Diagnostic Testing Services for Deoxynivalenol in the Eastern U.S.

### FY14 FPR – USWBSI ADDENDUM DON Service Labs – Quality Control Data

Insert below Quality Control Data/Results from the FY14 Award Period (approx. May 2014-May 2015):

Quality control data were collected at Virginia Tech through (a) the blind testing of samples with unknown DON levels (coordinated by the USWBSI through Trilogy Analytical Laboratories), and (b) the testing of subsamples of grain lots in each GC/MS run (to test for consistency among GC/MS runs). Known standards are run throughout the the GC/MS run to establish our standard curves.

- a. QC data for blind testing of samples from Trilogy Labs (coordinated by Trilogy Labs, and communicated through Sue Canty; scabusa@scabusa.org). Lab ID ‘Lab3’ is the Virginia Tech lab (highlighted in grey). Lab IDs 1-4 are other USWBSI labs. Data are in ppm.

	Trilogy Sample	Trilogy Quant	Lab1	Lab2	Lab3	Lab4-1	Lab4-2
Aug 2014	Low	0.70	0.60	0.70	0.59	0.56	0.65
	High	8.70	6.50	7.90	6.87	6.37	6.63
	Med	2.10	2.42	2.10	1.71	1.62	1.87
Sept 2014	Low	0.50	0.59	0.50	0.56	0.44	0.46
	High	7.90	7.13	6.20	6.85	5.71	6.24
	Med	2.90	2.46	2.20	2.68	1.78	1.96
Oct 2014	Low	0.50	0.59	0.50	0.47	0.49	0.47
	High	6.40	5.77	5.20	5.85	5.40	5.42
	Med	4.50	4.12	3.50	4.28	3.82	3.81
Nov 2014	Low	1.00	0.92	1.00	0.89	0.75	0.84
	High	7.00	5.77	6.10	5.88	5.1	5.58
	Med	4.90	4.38	4.40	4.52	3.72	4.29
Dec 2014	Low	0.50	0.47	0.50	0.48	0.45	0.46
	High	8.90	7.44	7.00	7.61	5.77	6.11
	Med	2.60	2.25	2.20	2.41	1.75	1.95
Jan 2015	Low	0.5	0.49	0.50	0.53	0.49	0.49
	High	7.9	7.75	7.00	7.19	6.43	6.20
	Med	2.4	2.01	1.90	2.05	1.59	1.64
Feb 2015	Low	1.60	1.40	1.30	1.41	1.12	1.17
	High	7.00	5.94	5.70	5.82	5.40	5.21
	Med	2.60	2.35	2.00	2.58	1.95	1.97

**PI:** Schmale, David

**Project:** Diagnostic Testing Services for Deoxynivalenol in the Eastern U.S.

	Trilogy Sample	Trilogy Quant	Lab1	Lab2	Lab3	Lab4-1	Lab4-2
Mar 2015	Low	1.00	0.98	0.90	0.81	0.85	0.86
	High	8.90	7.58	6.10	6.14	6.89	7.16
	Med	3.50	3.03	3.00	2.35	2.77	3.06
Apr 2015	Low	1.50	1.66	1.40	1.43	1.29	1.34
	High	7.90	9.03	6.80	6.54	6.50	6.57
	Med	4.90	5.50	3.90	4.18	4.17	4.32

b. QC data from internal checks of subsamples of grain lots from Trilogy (14-Apr-03) in each GC/MS run (to test for consistency among GC/MS runs). Trilogy sample 14-Apr-03 was measured 149 times and determined to have an average DON concentration of 6.99 ppm with a standard error of 0.57.

