### USDA-ARS/ U.S. Wheat and Barley Scab Initiative FY08 Final Performance Report (approx. May 08 – April 09) July 15, 2009

## **Cover Page**

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Fiscal Year:	2008
USDA-ARS Agreement ID:	59-0790-6-066
USDA-ARS Agreement	Structural and Functional Studies of Trichothecene Biosynthetic
Title:	Enzymes.
FY08 USDA-ARS Award	\$ 20,720
Amount:	$\phi$ $37,120$

#### **USWBSI Individual Project(s)**

USWBSI		<b>ARS</b> Adjusted
Research		Award
Category <sup>*</sup>	Project Title	Amount
PBG	Development of improved Enzymes for the Inactivation of	\$39,720
	Trichothecene Toxins.	
	Total Award Amount	\$ 39,720

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

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:** Development of improved Enzymes for the Inactivation of Trichothecene Toxins.

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

The major goal for the past fiscal year was to develop an improved enzyme for inactivating fungal mycotoxins. It was planned to utilize the three-dimensional structure of trichothecene 3-*O*-acetylase from *F. sporotrichioides* and *F. graminearum* (Tri101) as a framework to develop a modified enzyme with improved efficacy towards the inactivation of DON and nivalenol. The work utilizes a combination of biomolecular structural engineering coupled with kinetic and thermodynamic measurements.

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 three sections (repeat sections for each major accomplishment):

#### Accomplishment:

Over the past year a modified version of Tri101 from *F. graminearum* has been created that has a greater thermal stability than the wild type enzyme. This is being transferred into barley in collaboration with Lynn Dahleen to test its efficacy as a method for combating scab in the field.

In addition the structure of Tri3 a 15-O-trichothecene acetyltransferase isolated from F. sporotrichioides has been determined. A kinetic analysis shows that TRI3 is an efficient enzyme with the native substrate, 15-decalonectrin, but is inactive with either DON or nivalenol. The structure of TRI3 complexed with 15-decalonectrin provides an explanation for this specificity and shows that Tri3 and Tri101 (3-O-trichothecene acetyltransferase) are evolutionarily related. The active site residues are conserved across all sequences for TRI3 orthologs, suggesting that differences in acetylation at C15 are not due to differences in Tri3. The tri3 deletion mutant shows that acetylation at C15 is required for DON biosynthesis even though DON lacks a C15 acetyl group.

### Impact:

Although the research is at an early stage, the preliminary results suggest that it should be possible to create a better enzyme for inactivating the Fusarium toxins. The biological impact of these findings will not be known until transgenic barley has been created.

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.

Garvey GS, McCormick SP, Alexander NJ, Rayment I. Structural and functional characterization of TRI3 trichothecene 15-O-acetyltransferase from Fusarium sporotrichioides. Protein Sci. 2009, **18**(4):747-61.

If your FY08 USDA-ARS Grant contained a VDHR-related project, include below a list all germplasm or cultivars released with full or partial support of the USWBSI. List the release notice or publication. Briefly describe the level of FHB resistance. If this is not applicable (i.e. no VDHR-related project) to your FY08 grant, please insert 'Not Applicable' below.

Not applicable.