

**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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| <b>Fiscal Year:</b>                | 2008   |
| <b>USDA-ARS Agreement ID:</b>      | NA   |
| <b>USDA-ARS Agreement Title:</b>   | Uniform Testing of Biocontrol Agent <i>Cryptococcus flavescens</i> OH 182.9. |
| <b>FY08 USDA-ARS Award Amount:</b> | \$ 19,860  |

**USWBSI Individual Project(s)**

| <b>USWBSI Research Category*</b> | <b>Project Title</b>   | <b>ARS Adjusted Award Amount</b> |
|----------------------------------|--|----------------------------------|
| MGMT                             | Uniform Testing of Biocontrol Agent <i>Cryptococcus flavescens</i> OH 182.9. | \$19,860                         |
|                                  | <b>Total Award Amount</b>  | <b>\$ 19,860</b>                 |

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Principal Investigator

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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:** *Uniform Testing of Biocontrol Agent *Cryptococcus flavescens* OH 182.9.*

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

Results from multiple researchers supported in part by the USWBSI demonstrate that no single control measure from among pesticides, biological control, cultural control, disease forecasting and the use of resistant varieties is likely to reduce FHB to economically acceptable levels. Using biological control measures as part of an integrated management strategy against Fusarium head blight (FHB) is understudied yet has considerable potential for significantly contributing to the reduction of FHB and deoxynivalenol (DON) in wheat.

In previous work, we discovered microbial strains that reduced FHB in the greenhouse and field and demonstrated enhanced reduction of FHB via mixing fungicide-tolerant variants of our biocontrol agents with fungicides (Schisler et al., 2002). In more recent work, we discovered that the application of cold-shock temperatures during fermentation of OH 182.9 enhanced the survival and efficacy of the cells produced (Zhang et al., 2005; Dunlap et al., 2007).

However, impediments to including biological control products in the integrated management of FHB and DON in wheat include the lack of cost effective media for mass producing biomass of active biocontrol strain *Cryptococcus flavescens* OH 182.9 and the need for larger scale, proof-of-concept field tests of the original and stress tolerant variants of OH 182.9.

In work conducted in part with FY08 funding from the USWBSI, three cost effective, commercially feasible liquid culture medium for producing *C. flavescens* OH 182.9 were developed that utilized inexpensive, industrial grade sources of carbon and nitrogen. These media were then compared with a laboratory grade medium for equivalence in producing high quantities of efficacious inoculum of strain OH 182.9. In a separate course of study, biomass of cold-shocked OH 182.9 and a stress tolerant variant of the wild type strain were tested in greenhouse studies followed by field studies conducted in Michigan, Ohio, Illinois and Missouri.

**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 all three sections (repeat sections for each major accomplishment):**

**Accomplishment:**

An economically feasible, industrial-grade liquid medium was developed that supports the production of high quantities of efficacious biomass of FHB antagonist *Cryptococcus flavescens* OH 182.9. In our research, several liquid media were prepared using affordable, readily available sources of carbon and nitrogen. A medium that featured an industrial source of casein digest as well as carbon loading and C:N ratios that mimicked those used in our semi-defined laboratory medium produced biomass of FHB antagonist OH 182.9 that was similar in quantity to the laboratory medium. Additionally, biomass produced in the

casein-digest-based medium was identical to that produced in the laboratory medium in its efficacy in reducing FHB symptoms in both greenhouse and field studies.

**Impact:**

The development of liquid culture media that are economical to produce and support the production of high quantities of efficacious biomass of biocontrol agents is crucial to the production of affordable biocontrol products for the agricultural marketplace. The improved economics that result from producing inoculum of FHB antagonist OH 182.9 in our newly devised, commercially feasible liquid medium enhances the likelihood of bringing an OH 182.9-based, FHB biocontrol product to the marketplace.

**Accomplishment:**

Prothioconazole-tolerant (PTCT) variants of FHB antagonist OH 182.9 were shown to be more effective in reducing symptoms of FHB in greenhouse, and to a lesser extent, in field trials than the progenitor wild-type strain. PTCT variants reduced FHB disease severity by as much as 83% in greenhouse studies compared to 36% for the wild-type strain. Field studies confirmed the trend of PTCT variants reducing FHB symptoms to a greater extent than the wild-type OH 182.9 strain.

**Impact:**

The discovery that several PTCT variants of FHB antagonist OH 182.9 possess enhanced efficacy in reducing FHB is a key step in the process of developing successful FHB control strategies that combine fungicide and OH 182.9 treatments. Because wheat heads can not be treated with fungicides after flowering, combinations of fungicides with selected PTCT variants of FHB antagonist OH 182.9 are likely to reduce pathogen infection at both flowering and, importantly, after flowering when antagonist colonization of the wheat head would provide more lasting protection against new infections.

**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.**

Schisler, D.A., Slininger, P.J. and Boehm, M.J. Development of co-cultivated mixtures of antagonists active against Fusarium head blight of wheat. *Phytopathology* 97:S104. 2007.

Schisler, D.A., Boehm, M.J., Paul, P., and Slininger, P.J. Evaluation of prototype commercial media for the production of Fusarium head blight antagonist *Cryptococcus flavescens* OH 182.9. In: Proceedings of the National Fusarium Head Blight Forum; 2008 December 2-4, Indianapolis, IN. Lexington, KY: University of Kentucky. pp. 60-63. 2008.

Schisler, D.A., Dunlap, C.A., Slininger, P.J., Zhang, S., Boehm, M.J. and Jackson, M.A. Fermentation: a key determinant of successful biocontrol product development. *J. of Plant Pathology* 90 (2, Supplement):S2.126.

**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.