USDA-ARS/
U.S. Wheat and Barley Scab Initiative
FY10 Final Performance Report
July 15, 2011

Cover Page

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Fiscal Year:	FY10
USDA-ARS Agreement ID:	NA
USDA-ARS Agreement Title:	Efficacy and Characterization of Fungicide and *Cryptococcus flavescens* Mixtures.
FY10 USDA-ARS Award Amount:	$ 20,000

USWBSI Individual Project(s)

USWBSI Research Category*	Project Title	ARS Award Amount
MGMT	Efficacy and Characterization of Fungicide and *Cryptococcus flavescens* Mixtures.	$ 20,000
	Total ARS Award Amount	**$ 20,000**

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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
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: *Efficacy and Characterization of Fungicide and Cryptococcus flavescens Mixtures.*

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

Despite considerable research effort and progress, the significant and consistent reduction of Fusarium head blight (FHB) and the mycotoxin deoxynivalenol (DON) it produces in wheat and barley remains elusive. Research results clearly indicate, however, that utilizing an integrated pest management approach against FHB achieves the greatest level of disease/toxin control. The use of yeast biological control agent *Cyptococcus flavescens* OH 182.9 (NRRL Y-30216) as part of an integrated management strategy against FHB has considerable potential to contribute to the reduction of FHB and DON. Application of the fungicide-tolerant yeast strain variant OH 182.9 3C after wheat flowering when fungicides are not approved for use could reduce post-flowering infections by the FHB pathogen. Alternatively, a tank mix of fungicide and yeast biocontrol agent OH 182.9 3C applied at flowering could provide immediate and lasting protection against FHB and DON due to yeast OH 182.9 survival on wheat head infection courts after protection from the fungicide component has diminished. The yeast component could be especially useful in limiting the total DON content in harvested grain by combating new, DON producing infections by *F. graminearum* that can occur during early to late grain development. In studies conducted in the last year, field studies were conducted on quantifying the colonization dynamics of strain OH 182.9 under differing integrated application protocols to confirm results obtained in first year field studies. Additionally, for a second year we determined the level of FHB disease reduction associated with the colonization levels observed in order to direct research on producing and formulating the biocontrol agent to enhance the ability of cells to colonize pathogen infection courts, thereby improving biocontrol effectiveness. Studies were also conducted for a second year to confirm first year data generated to elucidate the chemical and physical properties of wheat head surfaces from the time of wheat head emergence from "boot" until the latter stages of kernel development. Such knowledge contributes to understanding how and when to apply fungicides and/or biocontrol agents to improve spray coverage and, concomitantly, treatment effectiveness. With this information on wheat head surfaces, formulations of fungicides and/or cells of biocontrol agents could also be better tailored to overcome surface characteristics that impede ideal spray coverage of infection courts on wheat heads.

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: *Demonstrated, for a second consecutive year, yeast biological control agent Cryptococcus flavescens OH 182.9 extensively colonized wheat head tissues and contributed to reducing FHB and DON when combined with a fungicide registered for use against Fusarium head blight (FHB) disease.*
Combining prothioconazole (PTC) tolerant FHB biocontrol agent *C. flavescens* OH 182.9 3C with a triazole fungicide has great potential for limiting FHB disease and pathogen formation

of the mycotoxin deoxynivalenol (DON) in grain since new pathogen infection can take place after fungicides can no longer be applied but when populations of the biocontrol agent could still be active on wheat head tissues. Field experiments were conducted on wheat in two locations in Illinois to determine how successfully yeast OH 182.9 3C colonized wheat head tissues when the biocontrol agent was applied alone or in combination with a commercial fungicide containing prothioconazole and tebuconazole either at or seven days after wheat flowering. Populations of PTC tolerant variant OH 182.9 3C were not affected by the presence of fungicide. After rain events, the yeast made up > 50% of the total microbial population recovered from specific wheat head tissues from 8 to 12 days after flowering, demonstrating for a second year the propensity of OH 182.9 3C to aggressively compete in colonizing infection court tissues on wheat heads. Disease reduction associated with the various treatments again supported the observation that the population of fungicide-tolerant variant of OH 182.9 3C on infection court tissues was not inhibited by the presence of fungicide and that treatments that contained both the variant and fungicide provided the greatest arithmetic reduction in FHB symptoms and DON. In other studies, fungicide treatment at flowering followed by treatment with cocultured biocontrol yeasts produced at the ARS lab in Peoria, IL were the most effective in reducing the mycotoxin deoxynivalenol in field trials conducted in multiple states with the treatment of spraying fungicide at flowering and the dual cultured yeast product five days later reducing DON by an average of 37%.

Impact: The feasibility of using fungicide tolerant biocontrol strain OH 182.9 3C as part of an integrated disease management program to reduce FHB and DON in wheat was demonstrated for a second year. Specifically, these studies showed that a yeast biocontrol agent/fungicide tank mix, or yeast sprayed after fungicide application results in the yeast colonizing wheat heads to high levels and contributes to overall FHB disease reduction, providing further support for the soundness of commercially producing this product to provide an additional FHB disease control tool for wheat producers and processors.

Accomplishment: *Confirmed that the hydrophobicity of wheat head tissues is lower at the time of wheat flowering than earlier in wheat head development.*

A waxy cuticle covering the surfaces of lemma and glume tissues on wheat heads mediates interactions between these tissues and the biotic environment. The chemical and physical properties of the surfaces of these plant tissues contribute to the nature of these interactions. For a second year, contact angle measurements demonstrated that the surface chemistry and ultrastructure of lemma and glume tissues changed around the time of anthesis for both a susceptible and moderately resistant wheat cultivar, with the tissue surfaces reaching a minimum in hydrophobicity immediately after flowering.

Impact: The discovery that the surface chemistry and ultrastructure of wheat head tissues changed during the course of wheat head development provides evidence that formulations of fungicides and/or biocontrol agents may need to be optimized for the specific stage of wheat head development in order to account for changing surface roughness and hydrophobicity levels on wheat heads. Improved delivery, coverage and retention of biocontrol agents and/or fungicides should ultimately result from spray formulations that are tailored for application to a specific stage of wheat head development.

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., Janisiewicz, W. J., Boekhout, T., and Kurtzman, C. P. 2011. Agriculturally important yeasts: biological control of field and postharvest diseases using yeast antagonists, and yeasts as pathogens of plants. Pages 45-52 in: The Yeasts (Fifth Edition) C. P. Kurtzman, J. W. Fell and T. Boekhout, eds. Elsevier B.V., Maryland Heights, Missouri.

Schisler, D.A., Khan, N, Boehm, M.J. 2011. Bacteria and yeasts for reducing Fusarium head blight in cereals and selection thereof. Canadian Patent No. 2,323,019; issued March 8, 2011.

Schisler, D.A., Paul, P., Boehm, M.J., Bradley, C.A., Dunlap, C.A. 2010. Colonization of wheat heads by antagonist *Cryptococcus flavescens* OH 182.9 when applied alone or in combination with different concentrations of Prosaro® and the effect on Fusarium head blight development in field-grown wheat. Pages 98-102 in: The 2010 National Fusarium Head Blight Forum, A. C. S. Canty, A. Anderson-Scully, D. Ellis, D. Van Sanford, ed. University of Kentucky, Milwaukee, WI.

Schisler, D.A. 2010. Good guys vs. bad guys, wheat is a battlefield. Interbusiness Issues, May 2010. http://www.peoriamagazines.com/ibi/2010/may/good-guys-vs-bad-guys.

Yuen, G. Y., Jochum C.C., Halley S.A., Sweets L.E., Kirk, W., Schisler, D.A. 2010. 2010 Uniform biological control trials-preliminary results. Pages 112-115 in: The 2010 National Fusarium Head Blight Forum, A. C. S. Canty, A. Anderson-Scully, D. Ellis, D. Van Sanford, ed. University of Kentucky, Milwaukee, WI.