USDA-ARS/ U.S. Wheat and Barley Scab Initiative FY05 Final Performance Report (approx. May 05 – April 06) July 14, 2006

Cover Page

PI:	Frances Trail
Institution:	Michigan State University
Address:	Department of Botany & Plant Pathology
	342 Plant Biology Lab
	East Lansing, MI 48824
E-mail:	trail@msu.edu
Phone:	517-432-2939
Fax:	517-353-1926
Fiscal Year:	2005
FY05 ARS Agreement ID:	58-3640-2-138
Agreement Title:	Studies of Inoculum Formation and Genomics in Gibberella
	zeae.
FY05 ARS Award Amount:	\$ 58,500

USWBSI Individual Project(s)

USWBSI Research		ARS Adjusted
Area [*]	Project Title	Award Amount
EDM	Genomics of Gibberella zeae, the Head Scab Fungus.	\$ 58,500
	Total Award Amount	\$ 58,500

Principal Investigator

Date

- CBC Chemical & Biological Control
- EDM Epidemiology & Disease Management
- FSTU Food Safety, Toxicology, & Utilization
- $GIE-Germ plasm\ Introduction\ \&\ Enhancement$

^{*} BIO – Biotechnology

VDUN - Variety Development & Uniform Nurseries

Project 1: Genomics of Gibberella zeae, the Head Scab Fungus.

1. What major problem or issue is being resolved and how are you resolving it?

Our lab continues to pursue strategies for limiting epidemics of *Gibberella zeae*. Our approach has been to study the life cycle to develop biology-based approaches to control. Perithecia, and the ascospores they discharge, are an important component of epidemics, as they are the source of primary inoculum in the field. The focus of this project was to continue to identify and study genes that disrupt ascospore discharge and perithecium development. We have completed an expression analysis using the Affymetrix Fusarium Genechips® that included 5 stages of perithecium development. Previously, we found that accumulation of lipids during the final stages of colonization of the plant is important for subsequent perithecium development (Guenther and Trail, 2005). This year, examination of the microarray data identified genes important to the metabolism of lipids throughout development. Interestingly, genes for lipid accumulation are expressed in culture just after perithecium induction; whereas genes for lipid degradation are expressed during the development of the perithecium (Figure 1). Because this gene expression is so striking, we chose to disrupt one of the genes that is likely to regulate lipid usage. The gene FG01571, encoding a hormone sensitive lipase (HSL), is a "starvation enzyme" in humans, where it signals for fats to be broken down for energy when cells are beginning to starve. In G. zeae, HSL is up-regulated significantly in culture as perithecia form. Mutants disrupted in *hsl* were analyzed for lipid content. In the *hsl* mutant, in culture, lipids continued to accumulate during perithecium development, in contrast to the wild-type, where lipid amounts declined (Figure 2). In culture, the medium is rich, and the perithecia develop normally. We are in the process of testing perithecium development in culture on a less-rich medium and in planta.

We have also been working on the disruption of 2 aquaporins (water channels), a P-type ATPase, and 2 chloride channels. Each of these genes is believed to have a role in perithecium function. The disruptants of the chloride channels did not have an identifiable phenotype. The former 3 genes have disruptions in progress, but should be completed by the end of the summer.

2. List the most important accomplishment and its impact (how is it being used?). Complete all three sections (repeat sections for each major accomplishment):

Accomplishment: The HSL gene (FG01571) was disrupted and the disrupted mutant accumulated lipids continuously up to 22% by 72 hrs after induction. This is in contrast to the discrete storage vs. usage cycle of the wild-type PH-1 cultures which accumulated a maximum of 10% lipid/dry weight just before perithecium formation. This indicates a defect in lipid usage in this mutant and we are examining the effects *in planta*. Our manuscript on lipid usage during perithecium development and pathogenicity is submitted.

Impact: Building a complete understanding of how fungi develop *in planta* and survive in the crop debris is the goal of our research. We have begun to unravel the nutritional relationship between host and fungus, and have shown how the fungus requires accumulates and used in order to develop perithecia. We are now investigating strategies to disrupt lipid storage, which is required for overwintering and perithecium production in the spring. As a result of that accomplishment, what does your particular clientele, the scientific community, and agriculture as a whole have now that they didn't have before?:

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We will shortly publish the information on how lipids are accumulated and used during sexual development and pathogenicity. This information will provide the research community with a better understanding of nutritional interaction between host and pathogen.



Figure 1: Average normalized gene expression for genes involved in triacylglycerol (TAG) storage and breakdown in culture. At 0H, perithecia are induced. At 144H, perithecia are mature.



Figure 2: Lipid accumulation during perithecium development between the *hsl*- mutant and the wild-type PH-1. TAG: triacylglycerol.

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.

Funded entirely by USWBSI:

Qi, W., Kwon, C. and F. Trail. 2006. Microarray analysis of transcript accumulation during perithecium development in *Gibberella zeae* (anamorph *Fusarium graminearum*). Molecular Genetics and Genomics 276:87-100.

Guenther, J. and Trail, F. 2005. The development and differentiation of *Gibberella zeae* (anamorph: *Fusarium graminearum*) during colonization of wheat. Mycologia 97 (1): 232-240. COVER.

Guenther, J., Hallen, H.E. and Trail, F. 200-. Accumulation of triglycerides in sexual development by *Gibberella zeae* (anamorph *Fusarium graminearum*). Submitted.

Recent publications relevant but funded at least in part by other resources:

Goswami, R., Xu, J.R., Trail, F. Hilburn, K. and H. C. Kistler. 2006. Genomic analysis of hostpathogen interaction between *Fusarium graminearum* and wheat during early stages of disease development. Microbiology *in press*.

Gueldener, U., K.-Y. Seong, J. Boddu, S. Cho, F. Trail, J.-R. Xu, G. Adam, H.-W. Mewes, G.J. Muehlbauer, and H. C. Kistler. 2006. Development of a *Fusarium graminearum* Affymetrix GeneChip For profiling fungal gene expression *in vitro* and *in planta*. Fungal Genetics and Biology, 43 (5): 316-325.

Gaffoor, I., and F. Trail. 2006. Characterization of two polyketide synthase genes involved in zearalenone biosynthesis in *Gibberella zeae*. Appl. and Environ. Microbiol. 72:1793-1799.

Gaffoor, I., Brown, D. W., Plattner, R., Proctor, R. H., Qi, W., and Trail, F. 2005. Functional analysis of the polyketide synthase genes in the filamentous fungus *Gibberella zeae* (anamorph *Fusarium graminearum*). Euk Cell. 4:1926-1933.

Trail, F., Gaffoor, I., and Vogel, S. 2005. Ejection mechanics and trajectory of the ascospores of *Gibberella zeae* (anamorph *Fusarium graminearum*). Fungal Genetics and Biology: 42:528-533