USDA-ARS/

U.S. Wheat and Barley Scab Initiative FY06 Final Performance Report (approx. May 06 – April 07) July 16, 2007

Cover Page

PI:	Gary Muehlbauer
Institution:	University of Minnesota
Address:	Department of Agronomy and Plant Genetics
	411 Borlaug Hall
	1991 Upper Buford Circle
	St. Paul, MN 55108
E-mail:	muehl003@tc.umn.edu
Phone:	612-625-6228
Fax:	612-625-1268
Fiscal Year:	2006
USDA-ARS Agreement ID:	59-0790-4-116
USDA-ARS Agreement	Molecular Genetic Approaches to Develop Scab Resistance.
Title:	
FY06 ARS Award Amount:	\$ 109,392

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Award Amount
GET	Developing and Characterizing Transgenic Wheat for Scab Resistance.	\$ 60,763
HGG	Mechanisms and Essential Genes for Resistance to Fusarium Head Blight.	\$ 48,629
	Total Award Amount	\$ 109,392

Principal Investigator	Date

HGR – Host Genetics Resources

 $HGG-Host\ Genetics\ \&\ Genomics$

PGG – Pathogen Genetics & Genomics

VDUN – Variety Development & Uniform Nurseries

(Form - FPR06)

^{*} CBCC – Chemical, Biological & Cultural Control

EEDF - Etiology, Epidemiology & Disease Forecasting

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

GET – Genetic Engineering & Transformation

PI: Muehlbauer, Gary

USDA-ARS Agreement #: 59-0790-4-116

Project 1: Developing and Characterizing Transgenic Wheat for Scab Resistance.

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

Fusarium head blight (FHB or scab) is a disease that can devastate wheat. To enhance FHB resistance in wheat, we are developing transgenic wheat lines with potential resistance genes and testing these lines for scab resistance.

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

Accomplishments:

- 1. We screened 17 transgenic wheat lines in the 2006 summer field trial and 24 lines in the 2007 summer field trial (collaboration with R. Dill-Macky). Due to dry and hot conditions we did not obtain usable results in the summer 2006 trial.
- 2. We screened 33, 33, and 42 transgenic wheat lines in the fall 2006, in the winter 2006, and in the spring 2007 greenhouse, respectively. Not all transgenic plants were screened in each trial. In two trials, we identified one line carrying a lipid transfer protein, two lines carrying a glutathione-S-transferase and two lines carrying a jasmonic acid-inducible Myb gene that exhibited significant (P<0.05) reduction in FHB severity compared to the nontransformed Bobwhite control. In one trial, we also identified three lines carrying a lipid transfer protein, three lines carrying a glutathione-S-transferase, two lines carrying a jasmonic acid-inducible Myb gene, three lines carrying carrying a barley germin-like gene, and one line carrying a pathogenesis-related 1 gene that exhibited significant (P<0.05) reduction in FHB severity compared to the nontransformed Bobwhite control. The three lines carrying the germin-like gene have only been screened once.
- 3. We developed five lines in the cv. Alsen genetic background that carried either the α-purothionin, thaumatin-like proteinl or β-1,3-glucanase transgene. Each of these transgenes showed a reduction in FHB severity under field conditions. Each of these lines—also carried the 3BS FHB resistance QTL. We conducted three greenhouse trials with these lines. In general they exhibited low FHB severity equivalent to Alsen. However, we could not detect a significant reduction in disease severity in the transgenic plants compared to Alsen.
- 4. Nine wheat lines carrying a rice pathogenesis-related 1 (PR-1) transgene, 15 wheat lines carrying a rice jasmonic acid-inducible Myb transgene, and seven lines wheat lines carrying a barley germin- like transgene were developed.

<u>Impact:</u> (Accomplishment 1) We will have the results from the field test in August 2007. (Accomplishment 2) We identified a set of transgenic wheat lines that performed well in the greenhouse. In particular the lines carrying the germin-like gene are encouraging. These lines will be further tested in the greenhouse and field.

(Accomplishment 3) We added proven transgenes to the Alsen background. These lines exhibited low levels of FHB severity. However, the variation in scab severity in the screens did not allow us to determine if the transgene increased the level of resistance in Alsen.

(Accomplishment 4) We developed additional transgenic wheat lines for FHB testing.

PI: Muehlbauer, Gary

ARS Agreement #: 59-0790-4-116

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?

The scientific community now has access to transgenic wheat lines that exhibit reduced FHB severity. These lines provide another source of variation for FHB resistance. In addition, the scientific community now has new information on the functional impact of a variety of genes on FHB resistance.

PI: Muehlbauer, Gary

USDA-ARS Agreement #: 59-0790-4-116

Project 2: Mechanisms and Essential Genes for Resistance to Fusarium Head Blight.

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

Fusarium head blight (FHB or scab) is a disease that can devastate the small grains wheat and barley. The wheat and barley transformation efforts have a limited number of genes that have the potential to reduce FHB. In addition, there is a limited amount of information pertaining to the molecular genetic interaction between the small grains and *Fusarium graminearum*. Our goal is to identify the mechanisms and essential genes for wheat and barley scab resistance.

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

Accomplishments:

3.

- 1. Described the impact of trichothecene mycotoxins on the expression patterns of barley genes during the barley-*F. graminearum* interaction. Identified 58 barley genes that respond to trichothecene accumulation. These genes encode proteins involved in trichothecene detoxification and transporter function, cell death and ubiquitination processes. A paper describing this work is in press at Molecular Plant-Microbe Interactions. All data are available at PLEXdb (http://www.plexdb.org/).
- 2. We are describing the gene expression patterns of barley genes in three near-isogenic line pairs carrying resistant and susceptible alleles for two QTL on chromosome 2H and a QTL on chromosome 3H for Fusarium head blight resistance.
- 3. We are describing the gene expression patterns of wheat genes in a near-isogenic line pair carrying resistant and susceptible alleles for the chromosome 3BS FHB resistant QTL. A manuscript is in preparation describing this work.
- 4. A graduate student from my laboratory visited Dr. Steve Scofield's laboratory (Purdue University, USDA-ARS, West Lafayette, IN) to learn the viral induced gene silencing (VIGS) system for functionally testing genes for resistance to FHB.

Impact:

(Accomplishment 1) Identified barley genes that respond to trichothecene accumulation. These genes potentially control a variety of cellular processes including: trichothecene detoxification and transport, cell death and ubiquitination processes. Data was released on the web.

(Accomplishments 2 and 3) We are identifying genes that differentiate the resistant from susceptible interactions, and may be involved in providing resistance, for important QTL in barley and wheat. All data will soon be released on the web.

(Accomplishment 4) A graduate student in my laboratory received VIGS training from S. Scofield's laboratory.

PI: Muehlbauer, Gary

USDA-ARS Agreement #: 59-0790-4-116

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?

The scientific community now has access to an unprecedented amount of expression data that defines the barley and wheat-*F. graminearum* interactions. In particular, the gene expression in response to trichothecene accumulation is known. These gene expression data will be used to test the functional significance of particular genes in transgenic plants and in the viral induced gene silencing system (collaboration with S. Scofield), and to develop molecular markers for breeding applications.

PI: Muehlbauer, Gary

USDA-ARS Agreement #: 59-0790-4-116

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.

Publications:

- Mackintosh, C.A., J. Lewis, L.E. Radmer, S. Shin, S.J. Heinen, L.A. Smith, M.N. Wyckoff, R. Dill-Macky, C.K. Evans, S. Kravchenko, G.D. Baldridge, R.J. Zeyen and G.J. Muehlbauer. 2007. Overexpression of defense response genes enhances the resistance of wheat to Fusarium Head Blight. *Plant Cell Reports* 26:479-488.
- Boddu, J., S. Cho and G.J. Muehlbauer. Transcriptome analysis of trichothecene-induced gene expression in barley. *Molecular Plant-Microbe Interactions* (In press).
- Nduulu, L.M., A. Mesfin, G.J. Muehlbauer and K.P. Smith. 2007. Genetic analysis of Fusarium head blight resistance and heading date in barley using near-isogenic lines with contrasting alleles for a QTL region on chromosome 2(2H). *Theor. Appl. Genet.* (In press).
- Cuomo, C.A., U. Güldener, J-R. Xu, F. Trail, B.G. Turgeon, A. Di Pietro, J.D. Walton, L-J. Ma, S.E. Baker, M. Rep, G. Adam, J. Antoniw, T. Baldwin, S. Calvo, Y-L. Chang, D. DeCaprio, L.R. Gale, S. Gnerre, R.S. Goswami, K. Hammond-Kosack, L.J. Harris, K. Hilburn, J.C. Kennell, S. Kroken, J.K. Magnuson, G. Mannhaupt, E. Mauceli, H.-W. Mewes, R. Mitterbauer, G. Muehlbauer, M. Münsterkötter, D. Nelson, K. O'Donnell, T. Ouellet, W. Qi, H. Quesneville, M.I.G. Roncero, K-Y. Seong, I.V. Tetko, M. Urban, C. Waalwijk, T.J. Ward, J. Yao, B.W. Birren, and H.C. Kistler. The *Fusarium graminearum* genome reveals a link between localized polymorphism and pathogen specialization. *Science* (In review).
- Chang, Y-L., S. Cho, H.C. Kistler, H-C. Sheng and G.J. Muehlbauer. Bacterial artificial chromosome-based physical map of *Gibberella zeae* (*Fusarium graminearum*). *Genome* (Accepted pending revisions 6-07).
- Jia, H., S. Cho and G.J. Muehlbauer. Transcriptome analysis of the wheat 3BS Fusarium head blight resistance QTL during *Fusarium graminearum* infection. In preparation for submission to *Molecular Plant-Microbe Interactions*.

Abstracts:

- Shin S.H., J.M. Lewis, C.A. Mackintosh, A. Elakkad, K. Wennberg, S.J. Heinen, R. Dill-Macky and G. J. Muehlbauer. 2006. Transgenic wheat with enhanced resistance to Fusarium Head Blight. Natl. Scab Forum Abstracts.
- Boddu, J., H. Bilgic, H. Jia, S. Cho, G.J. Muehlbauer. 2006. Transcriptome analysis of barley and wheat infected with *Fusarium graminearum*. Natl. Scab Forum Abstracts.
- Jia, H., S. Cho, K.P. Smith and G.J. Muehlbauer. 2007. Transcriptome analysis of wheat and barley near-isogenic lines during *Fusarium graminearum* infection. Plant and Animal Genome Abstracts, P739.

PI: Muehlbauer, Gary

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Presentation:

"Exploiting genomics for understanding the barley-*Fusarium graminearum* interaction" at the University of Nebraska, Lincoln, NE