

**USDA-ARS/
U.S. Wheat and Barley Scab Initiative
FY07 Final Performance Report (approx. May 07 – April 08)
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Cover Page

PI:	Tilahun Abebe
Institution:	University of Northern Iowa
Address:	Department of Biology 144 McCollum Science Hall Cedar Falls, IA 50614
E-mail:	Tilahun.Abebe@uni.edu
Phone:	319-273-7151
Fax:	319-273-7125
Fiscal Year:	2007
USDA-ARS Agreement ID:	59-0790-6-057
USDA-ARS Agreement Title:	Engineering Barley with Antifungal Gene Gastrodianin to Enhance Resistance to Scab Disease.
FY07 ARS Award Amount:	\$ 32,664

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
GET	Engineering Barley with Anti-Fungal Gene Gastrodianin for Resistance to Scab Disease.	\$32,664
	Total Award Amount	\$ 32,664

Principal Investigator

Date

* CBCC – Chemical, Biological & Cultural Control
EEDF – Etiology, Epidemiology & Disease Forecasting
FSTU – Food Safety, Toxicology, & Utilization of Mycotoxin-contaminated Grain
GET – Genetic Engineering & Transformation
HGR – Host Genetics Resources
HGG – Host Genetics & Genomics
IIR – Integrated/Interdisciplinary Research
PGG – Pathogen Genetics & Genomics
VDUN – Variety Development & Uniform Nurseries

Project 1: *Engineering Barley with Anti-Fungal Gene Gastrodianin for Resistance to Scab Disease.*

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

There is very little genetic resistance to *Fusarium* head blight (FHB) in barley. Genetic engineering has a great potential in the development of FHB resistant genotypes. So far, however, engineering resistant barley has been unsuccessful because of lack of genes that effectively inhibit *Fusarium graminearum*. Transformation with the common pathogenesis-related proteins such as, chitinases, glucanases, and thaumatin-like proteins (TLPs), did not significantly improve resistance. There is a need to study efficacy of other fungal resistance genes against *F. graminearum*. We are working with an antifungal gene *gastrodianin* isolated from a Chinese medicinal plant *Gastrodia elata*. *Gastrodianin* is known to inhibit growth of *F. graminearum* and other saprophytic fungi. The major routes of *F. graminearum* infection of barley kernels are through the husk (lemma and palea) and the apex of florets. To successfully block infection, expression of *gastrodianin* in transgenic Golden Promise barley was targeted to the husk and epidermis of the kernel using a spike-specific *Lem2* promoter that was isolated from Morex barley.

**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: Golden Promise barley was co-transformed with plasmids pLem2VGM2 (containing *GFP* and *gastrodianin*) and pAHC25 (containing a selectable *bar* gene). Both *GFP* and *gastrodianin* are driven by the spike-specific *Lem2* promoter. Fifteen T₀ transformants were recovered. Of these, eleven were fertile, two plants were sterile and two plants were lost to contamination. Integration of *gastrodianin* into the genome of Golden Promise was verified by PCR. Accumulation of *gastrodianin* transcript was confirmed by RT-PCR. Western analysis using a polyclonal antibody raised against *gastrodianin* protein also detected a band at 12 kDa, which corresponds to the size of the mature protein. We planned to test resistance of transgenic plants to *F. graminearum* in fall 2007 and spring of 2008. Unfortunately, the transgenic plants grow very slow and we were unable to collect seeds for planting T1 and T2 generation. Instead, greenhouse testing is scheduled for fall 2008.

Impact: Use of the tissue-specific *Lem2* promoter will direct accumulation of *gastrodianin* protein in the spike of transgenic plants. This is advantageous because the protein will only be expressed in the tissues *F. graminearum* must cross to infect the kernel. In addition, constitutive expression of proteins is an energy-demanding process and can interfere with metabolic processes resulting in dwarf plants. Restricting expression of *gastrodianin* to the spike could reduce possible side effects of the protein on growth.

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?:

Transgenic Golden Promise plants expressing *gastrodianin* can be crossed to elite barley lines to develop varieties that are resistant to *F. graminearum*. Ultimately, resistant varieties can increase yield and quality of grain in areas where scab disease is a major problem.

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.

None