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

Cover Page

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Fiscal Year:	2005
FY05 ARS Agreement ID:	59-0790-3-081
Agreement Title:	RNAi Control of Deoxynivalenol Contamination of Barley.
FY05 ARS Award Amount:	\$ 39,128

USWBSI Individual Project(s)

USWBSI		
Research		ARS Adjusted
Area [*]	Project Title	Award Amount
BIO	RNAi Control of Deoxynivalenol Contamination of Barley.	\$ 39,182
	Total Award Amount	\$ 39,128

Principal Investigator

Date

- CBC Chemical & Biological Control
- EDM Epidemiology & Disease Management
- FSTU Food Safety, Toxicology, & Utilization
- GIE Germplasm Introduction & Enhancement

^{*} BIO – Biotechnology

VDUN - Variety Development & Uniform Nurseries

Project 1: RNAi Control of Deoxynivalenol Contamination of Barley.

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

One of the most severe mycotoxin problems in the U.S. is trichothecene contamination of small grains by *Gibberella zeae* (anamorph *Fusarium graminearum*) in a disease called scab or Fusarium head blight (FHB). Here we propose a novel method to control trichothecene contamination in barley using RNA interference (RNAi) technology to block mycotoxin production. RNAi is a conserved eukaryotic gene regulatory mechanism often referred to as gene silencing. We propose to silence expression of a key transcription factor gene (*tri6*) for the control of tricothecene production in *Fusarium* by transforming barley and wheat with an inverted repeat sequence of *tri6* (we propose two crops as one may work better than the other). It is hypothesized that the inverted repeat transcript will be fragmented into small RNA species known as siRNAs as a part of a conserved eukaryotic silencing mechanism. These siRNAs will then be taken up by hyphae and trigger a silencing mechanism in *Fusarium*. This research is designed to quickly control mycotoxin contamination of barley and wheat.

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: Our most important accomplishment was to show that RNAi silencing can reduce trichothecene production and decrease virulence in *Fusarium graminearum*. Also, the progress from this SCAB grant enabled us to obtain enough data that led to a successful bid for a NRI USDA research grant to continue the topic of this research.

Whereas we still do not know if RNAi technology can control trichothecene production in barley and/or wheat, we have made progress in this regard. Currently we have several lines of wheat callus tissue harboring RNAi vectors geared to suppress 2 genes (*tri6* and GFP) in *Fusarium graminearum*. We have shown that GFP strains of *Fusarium graminearum* fluoresce green on wheat callus and we are at the step to see if the GFP containing wheat callus can suppress this florescence.

Impact: This is a new technology for *F. graminearum* and allows researchers worldwide to apply this technology for knock down experiments when disruption of a gene is lethal and/or the researcher is interested in speed and qualitative regulation of gene expression in this fungus. Several labs world wide have asked for our RNAi silencing vector.

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 availability of a new method to control gene regulation in *F. graminearum* represents an important advance in *F. graminearum* research allowing for alternative ways to examine function of genes in the fungus.

FY05 (approx. May 05 – April 06) PI: Keller, Nancy ARS Agreement #: 59-0790-3-081

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.

Hammond TM, Keller NP (In preparation) RNAi mechanisms in fungi. Invited Chapter for book edited by S Osmani and G. Goldman entitled *Aspergillus* genomics

Hammond TM, Keller NP (2005) RNA silencing in *Aspergillus nidulans* is independent of RNA dependent RNA polymerases. Genetics 169: 607-617.

McDonald T, Brown D, Keller N P, Hammond T (2005) Inverted repeat transgenes silence mycotoxin production in *Aspergillus* and *Fusarium* species. Mol Plant Microbe Interactions 18:539-545.