

USDA-ARS / USWBSI
FY03 Final Performance Report (approx. May 03 – April 04)
July 15, 2004

Cover Page

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Year:	FY2003 (approx. May 03 – April 04)
FY03 ARS Agreement ID:	NA
FY03 ARS Agreement Title:	Control of FHB in barley through transformation.
FY03 ARS Award Amount:	\$ 60,788

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
BIO	Development of Fusarium head blight resistant transgenic plants in barley.	\$ 60,788
	Total Amount Recommended	\$ 60,788

Principal Investigator

Date

* BIO – Biotechnology
CBC – Chemical & Biological Control
EDM – Epidemiology & Disease Management
FSTU – Food Safety, Toxicology, & Utilization
GIE – Germplasm Introduction & Enhancement
VDUN – Variety Development & Uniform Nurseries

Project 1: Development of *Fusarium head blight resistant transgenic plants in barley.*

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

Resistance to FHB in barley and durum is limited, with only a few sources of partial resistance available for cultivar development. Transgenic barley and durum expressing antifungal and antitoxin genes may provide breeders with additional sources of resistance to incorporate into breeding programs. Single gene insertions have provided some protection, comparable to resistant checks, but disease and DON levels are still too high to provide complete protection. Our objective is to insert multiple antifungal and antitoxin genes into barley and durum, develop homozygous lines expressing the genes, and test their effects on FHB and DON. These were: modified Trichothecene acetyl transferase (*mTri101*) from *Fusarium sporotrichoides*; ATP-binding cassette transporter (*Tri12*) from *F. sporotrichoides*; thaumatin-like protein (*tlp*) from *Oryza sativa*; chitinase (*chi*) from *O. sativa*; glucanase (*glu*) from *F. venenatum*; endochitinase (*endo*) from *F. venenatum*; and exochitinase (*exo*) from *F. venenatum*.

2. What were the most significant accomplishments?

Eighteen out of 44 durum transgenic lines (T₁'s) have been analyzed by PCR and Southern, and the other 26 lines are in the greenhouse for further analysis. Northern analyses as well as western and IEF gel analyses are being conducted to characterize protein differential expressions in spike and leaf tissues of transgenic durum wheat and barley lines Tri101 and *tlp* (T₃'s). New putative transgenic plants that were regenerated in the 2003 grant cycle, and the minimum number of transformation events include:

Antifungal or Antitoxin Gene/s	No. of plants	No. of events
Glucanase + bar	33	4
Endochitinase + bar	2	2
<i>tlp</i> + <i>chi</i>	16	10
<i>tlp</i> + Tri12	10	5
<i>tlp</i> + mTri101	6	5
<i>chi</i> + Tri12	2	2
<i>chi</i> + <i>exo</i>	1	1

T₀'s of putative *glu*+ transgenic barley lines were all positive for *bar* and glucanase following PCR analysis. All other putative transgenic plants (T₀'s) that contained the *bar* gene and the gene/s of interest were selected for T₁ progeny analysis.

Conlon and Lacey backcross-derived lines expressing Tri101 and PDR5 lines were selected in the field in Aberdeen, ID (collab. Phil Bregitzer) and advanced in the greenhouse to identify homozygous lines for field testing in 2004. Backcrossing of lines expressing *chi* and *tlp* was started in early 2004.

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 you grant. Please reference each item using an accepted journal format. If you need more space, continue the list on the next page.

Tobias, D.J., A.K. Jha and L.S. Dahleen (2004) Transformation of Barley with Antifungal and Antitoxin Genes. In Vitro Biology Abstract Issue Vol. 40, p. 53-A (P-2041) Presented in 2004 World Congress on In Vitro Biology, May 2004, San Francisco, CA.

Dahleen, L.S. and M. Manoharan. 2003. Transformation of barley with two antifungal genes. National Fusarium Head Blight Proceedings, p. 12.

Manoharan, M. and L.S. Dahleen. 2003. Regeneration and genetic transformation of durum wheat. National Fusarium Head Blight Proceedings, p. 29.

Dahleen, L.S. 2003. Transformation to provide new genes for FHB resistance: a summary of current US public research. 3rd Canadian Workshop on Fusarium Head Blight, proceedings p. 89.