

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

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<b>Fiscal Year:</b>	2007
<b>USDA-ARS Agreement ID:</b>	59-0790-6-069
<b>USDA-ARS Agreement Title:</b>	Modification of the Ribosomal Target to Enhance Resistance to Trichothecene Mycotoxins.
<b>FY07 ARS Award Amount:</b>	\$ 43,190

**USWBSI Individual Project(s)**

<b>USWBSI Research Area *</b>	<b>Project Title</b>	<b>ARS Adjusted Award Amount</b>
GET	Modification of the Ribosomal Target to Enhance Resistance to Trichothecene Mycotoxins.	\$43,190
	<b>Total Award Amount</b>	<b>\$ 43,190</b>

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Principal Investigator

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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: Modification of the Ribosomal Target to Enhance Resistance to Trichothecene Mycotoxins.**

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

Our main goals in the project funded during the FY07 period was to develop DON resistant forms of the wheat L3 genes and to generate transgenic wheat plants with these genes to determine if overexpression of the mutant forms of the wheat L3 genes will confer resistance to FHB in 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:**

We have successfully demonstrated that expression of a truncated form of the yeast L3 gene (L3 $\Delta$ ), which corresponds to the first 99 amino acids of the yeast ribosomal protein L3, in transgenic wheat plants leads to improved resistance to Fusarium Head Blight (FHB) and a reduction in DON levels. During FY07 we have isolated genes encoding the wheat ribosomal L3 proteins. We have engineered the same mutations into the wheat L3 genes that previously conferred resistance to FHB in the yeast L3 genes. We prepared constructs with the DON-resistant point-mutant forms of the wheat *RPL3A1* gene and a truncated wheat *RPL3A1* gene (L3 $\Delta$ ) downstream of the maize *Ubiquitin1* promoter and first intron. Each coding region included V5 and 6xHis epitope tags so that the new proteins can be distinguished from native wheat L3. These constructs were provided to Dr. Ann Blechl for transformation into wheat. Dr. Blechl has produced several new transgenic wheat lines with the wheat L3 $\Delta$  and the full-length wheat *RPL3A1* mutants, W258K and W258C. We plan to identify homozygous lines and characterize expression of the wheat L3 genes. These lines will be evaluated in the greenhouse for resistance to FHB during FY08 and DON levels will be analyzed.

**Impact:**

We have demonstrated FHB resistance in transgenic wheat plants for the first time by expressing a modified form of the yeast ribosomal protein L3 gene. Our results also provided the first evidence that resistance to DON correlates with resistance to FHB and results in reduced accumulation of DON in transgenic wheat plants. We have now developed constructs containing the mutated forms of the wheat L3 genes and produced transgenic wheat plants with these genes. If transgenic wheat lines containing the modified wheat L3 genes show resistance to FHB and DON accumulation, these transgene loci will provide sources of novel resistance to FHB. These genes can be transformed into other wheat cultivars, such as the Hard Winter Wheat and Durum, as well as into barley plants. We anticipate that the transgenic wheat lines overexpressing the DON resistant forms of the wheat L3 genes may be more acceptable to the public.

**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?:**

Developing effective FHB resistance through transgenics is one of the strategies that can be used to reduce the impact of FHB in wheat. Our work has demonstrated the feasibility of this approach. We have produced transgenic wheat lines that showed resistance to FHB and reduced DON accumulation in the greenhouse tests. During FY08, these lines were grown in a transgenic nursery established by the University of Minnesota and were included in the tours of this nursery that were open to the public. The transgenic lines are being evaluated in a FY08 field test at the University of Minnesota by Dr. Ruth Dill-Macky. If these lines show resistance in the field, they could be used as breeding material for introduction of the FHB resistance into elite wheat cultivars or breeding lines and can be combined with other sources of resistance to provide growers new wheat cultivars with effective FHB resistance.

**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**

J. E. McLaughlin, A. Bin ‘Umer, J. Schifano, A. Tortora, S. McCormick, N. E. Tumer 2008. Genome wide analysis of trichothecene mycotoxin mechanisms of action in *Saccharomyces cerevisiae*. In preparation.

Di, R., and N. E. Tumer. The N-terminal 99 amino acids of yeast ribosomal protein L3 reduce the toxicity of pokeweed antiviral protein in *Saccharomyces cerevisiae*. In preparation.

Di, R., A. Blechl, R. Dill-Macky, A. Tortora, and N. E. Tumer. 2007. Expression of the N-terminal 99 amino acids of yeast ribosomal protein L3 in transgenic wheat confers resistance to *Fusarium* head blight. *Mol. Plant Breeding* 5:283.

### **Oral presentations in conferences**

N. E. Tumer. 2007 Resistance to FHB by expression of modified L3 genes in wheat. National Fusarium Head Blight Forum Dec. 2-4 2007, Kansas City, MO

Di, R., A. Blechl, R. Dill-Macky, A. Tortora, and N. E. Tumer. 2007. Expression of the N-terminal 99 amino acids of yeast ribosomal protein L3 in transgenic wheat confers resistance to *Fusarium* head blight. The 2<sup>nd</sup> International Conference of Plant Molecular Breeding, Hainan, China.