USDA-ARS / USWBSI FY04 Final Performance Report July 15, 2005

Cover Page

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Year:	FY2004 (approx. May 04 – April 05)
FY04 ARS Agreement ID:	58-5325-2-758
FY04 ARS Agreement Title:	Engineering Wheat's Resistance to <i>Fusarium</i> Infection and
	Trichothecenes.
FY04 ARS Award Amount:	\$ 48,780

USWBSI Individual Project(s)

USWBSI Research Area [*]	Project Title	ARS Adjusted Award Amount
BIO	Modification of the Ribosomal Target to Enhance Resistance to Trichothecene Mycotoxins.	\$ 48,780
	Total ARS Award Amount	\$ 48,780

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: 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 major goal in this project was to develop transgenic wheat plants that are resistant to the Fusarium mycotoxin, deoxynivalenol (DON). Trichothecenes inhibit protein synthesis by targeting ribosomal protein L3. Pokeweed antiviral protein (PAP), a ribosome inactivating protein (RIP) binds to L3 to depurinate the -sarcin/ricin loop of the large rRNA. Plants transformed with the wild type PAP show lesions and express very low levels of PAP because PAP autoregulates its expression by destabilizing its own mRNA. We showed that transgenic tobacco plants expressing both the wild type PAP and a truncated form of yeast L3 (L3 Δ) are phenotypically normal. PAP mRNA and protein accumulation is increased in these plants, indicating that L3A suppresses the autoregulation of PAP mRNA stability. Ribosomes are not depurinated in the transgenic plants expressing PAP and $L3\Delta$, even though PAP is associated with ribosomes. The expression of the endogenous tobacco ribosomal protein L3 is upregulated in these plants and they are resistant to the Fusarium mycotoxin, DON. These results demonstrate that expression of an N-terminal fragment of yeast L3 leads to trans-dominant resistance to PAP and the trichothecene mycotoxin, DON, providing evidence that both toxins target L3 by a common mechanism.

2. What were the most significant accomplishments?

<u>Accomplishment:</u> The contamination of important agricultural products, such as wheat, barley or maize with the trichothecene mycotoxin, deoxynivalenol (DON) due to infection with *Fusarium* is a worldwide problem because DON accumulates in grain to levels posing a threat to human and animal health. Favorable weather conditions led to a dramatic re-emergence of the *Fusarium* Head Blight (FHB) in the USA in the last decade, resulting in significant economic losses. Our work demonstrated for the first time that introduction of a modified form of ribosomal protein L3 into transgenic plants led to *trans*-dominant resistance to DON, providing evidence that DON targets ribosomal protein L3 to inhibit protein synthesis.

<u>Impact:</u> This is the first time that high level of resistance to DON has been obtained by modifying its ribosomal target. The resistance is durable and is transmitted to the homozygous progeny in tobacco. It is the first step in developing wheat plants that are resistant to *Fusarium*. It will provide wheat researchers a tool for studying the effects of DON on the peptidyltransferase center of wheat ribosomes and a tool for breeding wheat cultivars resistant to *Fusarium* Head Blight.

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.

Refereed Publications:

R. Di and <u>N. E. Tumer</u> (2005). Expression of a truncated from of ribosomal protein L3 confers resistance to pokeweed antiviral protein and the *Fusarium* mycotoxin deoxynivalenol. *Molecular Plant Microbe Interact.* 18, 762-770.

S. C. Popescu and <u>N. E. Tumer</u> (2004). Silencing of ribosomal protein L3 genes in *N. tabacum* reveals coordinate expression and significant alterations in plant growth, development and ribosome biogenesis. *Plant J.* 39, 29-44.

Presentations:

R. Di and N. E. Tumer (2005). Expression of a truncated form of ribosomal protein L3 confers resistance to pokeweed antiviral protein. American Society for Virology 24th Annual Meeting, The Pennsylvania State University, University Park, Pennsylvania. June 18-22. Abstract No. 29-1.

R. Di and N. E. Tumer (2005). Expression of a truncated form of ribosomal protein L3 confers resistance to pokeweed antiviral protein and the Fusarium mycotoxin deoxynivalenol. The 5th Symposium on Post-transcriptional Regulation of Plant Gene Expression. June 8-12, 2005. The University of Texas at Austin. Abstract No. 39.