

**U.S. Wheat and Barley Scab Initiative
Annual Progress Report
September 18, 2000**

Cover Page

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Grant Number:	59-0790-9-026
Grant Title:	Fusarium Head Blight Research
Amount Granted:	\$83,000.00

Project

Program Area	Objective	Requested Amount
Biotechnology	Enhance scab resistance in winter wheat germplasm by plant transformation.	\$70,000.00
Variety Development & Uniform Nurseries	To enhance variety development of scab resistant varieties.	\$70,000.00
	Requested Total	\$140,000.00¹

Principal Investigator

Date

¹ Note: The Requested Total and the Amount Granted are not equal.

Project 1: Enhance scab resistance in winter wheat germplasm by plant transformation.

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

Traditional plant breeding has limited genetic resources for Fusarium head blight (FHB). Many of these sources appear to be multigenic and are difficult to manipulate in conventional or even with molecular marker assisted plant breeding. In this research our key goal was to increase the sources of resistance to Fusarium head blight (FHB) through the use of novel, highly biologically active genes from nontraditional sources. We approached this goal by collecting a set of inhibitor of programmed cell death (PCD) or antiapoptotic genes (lead candidate genes: IAP and ced9) and antifungal genes (lead gene: lactoferrin). Our objective was to transform Bobwhite with these genes using microprojectile or *Agrobacterium tumefaciens* mediated transformation, grow the progeny and screen them for resistance or tolerance to FHB. As these genes may not be effective, a second objective was to continue collecting genes that may be effective in reducing the devastating effects of FHB.

2. Please provide a comparison of the actual accomplishments with the objectives established.

Using microprojectile and *Agrobacterium tumefaciens* mediated transformation; we have made 24 events with IAP, 3 with ced9 (there was a problem with the ced9 construct and we had to redo the construct and transformations), and 24 events with lactoferrin. The strategy in using antiapoptotic genes is to affect the infection process and requires testing at the whole plant level. The strategy in using lactoferrin is to express a known antifungal protein, which should affect FHB. Extracts from transgenic tobacco plants expressing lactoferrin (when compared to transgenic tobacco plants not expressing lactoferrin) were shown to inhibit FHB growth in petri dishes. The progeny from the IAP, ced9, and lactoferrin transgenic lines have been screened in the growth chamber. Preliminary results from our screen for FHB for IAP identified some families expressing a level of tolerance to FHB that merits further testing. The preliminary results for lactoferrin were also promising. Plants containing lactoferrin (57.6 ± 8.8 , mean \pm standard error) had a lower level of FHB than plants, which did not contain lactoferrin (91.2 ± 10.1). For both genes, transgenic families having the highest level of tolerance and appropriate controls (we are including the best conventionally developed FHB susceptible and resistance lines, and nontransgenic Bobwhite) are being retested to confirm these preliminary results. Additional genes (e.g. the antiapoptotic gene, Bcl-x1, and some derivatives) have been collected for and are being inserted into wheat.

3. What were the reasons established objectives were not met? If applicable.

Our objectives were met for IAP, lactoferrin, and collecting additional genes which may be highly useful in controlling FHB. We had problems with ced9's construct and had to redo the construct and the transformations. Seed from T₀ plants for ced9 are being harvested.

4. What were the most significant accomplishments this past year?

The most significant accomplishment would be the identification of a promising gene (lactoferrin) for alleviating the devastating effects of FHB.

Project 2: To enhance variety development of scab resistant varieties.

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

Our main objective was to develop germplasm that is tolerant to Fusarium head blight (FHB), which will be the future base for cultivar development for the high rainfall and irrigated acreage in the central Great Plains. To meet this goal, we are collecting adapted and exotic germplasm from throughout the world and crossing them into adapted hard red and white winter wheat cultivars. Once our transgenic FHB tolerance is verified, we will rapidly incorporate and pyramid those transgenes into common wheat. Traditionally about one third of Nebraska's wheat acreage is in the FHB risk area (between 600 to 700,000 acres) and the University of Nebraska wheat breeding program has developed cultivars grown on 80% of Nebraska, as well as, being widely grown in the FHB risk areas in adjacent states in South Dakota and Kansas. In addition to collecting germplasm, a key need has been to develop effective screens to allow selection for FHB tolerance.

2. Please provide a comparison of the actual accomplishments with the objectives established.

We have collected elite germplasm from the northern Great Plains and eastern United States, as well as, from China and have evaluated for agronomic performance, the germplasm of Central Europe. The crossing continues as would be expected in any traditional breeding effort. We are developing an effective greenhouse screen, which will be used mainly for better parent identification and have the key skilled personnel in place. This screen has been used to evaluate our transgenic materials. We are in the process of building a field-screening nursery based on mist irrigation with appropriate controls to screen 1000 lines. The irrigation pipe and controls have been purchased and received. The irrigation well has been dug and the line feeds put in place. We would like to acknowledge the advice of our colleagues at South Dakota State University (particularly Drs. Yue Jin and Jackie Rudd) in helping us develop our greenhouse and field screening protocols.

3. What were the reasons established objectives were not met? If applicable.

Basically, all of our objectives have been met. Traditional plant breeding, by nature, is a time consuming effort. We have nearly completed a scheduled phase-in of needed facilities and appropriate disease screens which allow the FHB tolerant material to be fully incorporated in the plant breeding program and properly screened for FHB tolerance, agronomic performance, adaptation, and end use quality.

4. What were the most significant accomplishments this past year?

The most significant accomplishments for last year are the continued incorporation and generation advance of FHB tolerant germplasm, the development of a FHB tolerance greenhouse screen, and purchase of necessary equipment for a FHB tolerance field screen. NE94654 has been recommended for release in 2000-2001. It appears to have low level of FHB tolerance which may explain its below average FHB incidence scores in the data reported so far in the FHB screening nursery. NE94654 is a line that seems well adapted to FHB risk areas of Nebraska.

Year: 2000
PI: Stephen Baenziger
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Progress Report

Publications:

Dickman, M. B., Y.K. Park, T. Clemente, and T. Oltersdorf, and R. French. Modulation of plant-pathogen interactions by animal anti-apoptotic genes. *Nature Biotechnology*: in review.