U.S. Wheat and Barley Scab Initiative FY02 Final Performance Report (approx. May 02 – April 03) July 15, 2003

Cover Page

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Grant Number:	59-0790-0-061
Grant Title:	Fusarium Head Blight Research
FY02 ARS Award Amount:	\$ 6,829

Project

Program Area	Project Title	USWBSI Recommended Amount
CBC	Management of Fusarium Head Blight With Biological Control Agents.	\$7,000
	Total Amount Recommended	\$7,000

Principal Investigator

Date

Project 1: Management of Fusarium Head Blight With Biological Control Agents.

- 1. What major problem or issue is being resolved and how are you resolving it?
- a) We became aware that the chemical composition of the growth medium for growing the bacterial biocontrol agents (BCAs) could have a significant effect on the efficacy of the BCAs in controlling FHB. The potato dextrose broth (PDB) we have used to culture the BCAs contains dextrose (glucose) as carbon source, which we now know is not the best carbon source for production of antibiotics such as iturin that might be a major part of the antagonistic effect against FHB. (Also, studies by our lab and others suggest that PDB sprayed onto wheat might stimulate *Fusarium graminearum* and lead to increased FHB symptoms on wheat. It would be more desirable to use a growth medium for the BCAs that could be sprayed onto wheat which would not lead to as much stimulation of *F*. *graminearum*). A defined growth medium (called original defined medium) for some *Bacillus* species that was previously described in the literature, with mannitol as the carbon source and glutamic acid as carbon/nitrogen source, allowed very good growth of the four BCAs we have studied in either broth or agar-solidified form
- b) We were told by Gary Yuen at the University of Nebraska-Lincoln (who used plating methods to enumerate cell numbers of one of our BCAs grown in the original defined broth medium applied to field plots in Nebraska) that cell numbers for our BCA in this broth were lower by an order of magnitude or more than for the BCAs used by other investigators, grown in various broth media. After I conferred with David Schisler at USDA-CRS in Peoria who has had extensive experience in growth medium formulation, I modified the defined medium recipe so it contained 2.5 times more C than originally and 2.1 times more N than originally, in hopes that cell numbers and accordingly antibiotic production would increase in the modified medium. After 10 days in the modified defined broth medium at 30° C, plate counts for strains 1BA and 1BC averaged 6.7 X 10⁵ CFU/ml. After 72 hours hours in the PDB at 30° C, plate counts were 1.5 X 10⁴ CFU/ml. Visible turbidity in both the PDB and defined medium broth formulations suggested that cell numbers should be more like 10⁷ CFU/ml. We will investigate whether the plating efficiency of our BCAs is low, and how to make the plating efficiency higher.

c) Further trials will be conducted in the greenhouse and field plots with the *Bacillus* strains we have isolated and characterized in the laboratory, to evaluate their efficacy in controlling FHB.

2. What were the most significant accomplishments?

a) We verified that a defined growth medium allowed growth of our BCAs. Cell-free acid precipitates from 10-day old defined broth cultures of our four BCAs were extracted with methanol/water and then spotted onto sterile paper disks which were then applied to plates inoculated with *F. graminearum*. This type of extraction method should harvest iturin-like compounds from the broth medium. Zones of inhibition developed around disks from all four isolates, but the greatest inhibition was noted for strains 1BA and 1BC.

b) We verified that modifying the defined medium by raising C and N led to an apparent increase in cell numbers of our BCAs, compared to the original defined broth formulation.c) We continued to work in cooperation with D. Schisler, G. Yuen, M. Draper and G. Bergstrom on the biocontrol of FHB. We worked with Schisler to raise the C and N of our defined medium; and worked with Yuen to help devise a plating medium for counting BCAs in field experiments.

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.

Baye, N., and B.H. Bleakley. 2002. Multiple approaches in attempted identification of bacterial strains used in the biological control of Fusarium Head Blight. Abstract H-128, p. 244. *In* Abstracts of the 102nd General Meeting of the American Society for Microbiology. American Society for Microbiology, Washington, D.C.

Baye, N., B.H. Bleakley, M.A. Draper, and K.R. Ruden. 2002. Effect of bacterial growth medium composition on antifungal activity of *Bacillus sp.* strains used in biological control of Fusarium Head Blight. Abstract, p. 54. *In* 2002 National Fusarium Head Blight Forum, December 7-9, 2002. Cincinnati, OH.

Baye, N., and B.H. Bleakley. 2002. Taxonomic affiliation of bacterial strains used in the biological control of Fusarium Head Blight suggests possible role of lipopeptide antibiotic in fungal antagonism. Abstract, p.55. *In* 2002 National Fusarium Head Blight Forum, December 7-9, 2002. Cincinnati, OH.

Draper, M.A., B.H. Bleakley, K.R. Ruden, N. Baye, A.L. LeBouc, and S.M. Schilling. 2002. Uniform trials for biological control agent performance in the suppression of Fusarium Head Blight in South Dakota-2002. Abstract, pp. 65-66. *In* 2002 National Fusarium Head Blight Forum, December 7-9, 2002. Cincinnati, OH.

Bleakley, B.H. 2003. Biological control of foliar and head diseases of wheat. AD-421 Progress Report (CRIS Report).