

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

Cover Page

PI:	Bruce Bleakley
Institution:	South Dakota State University
Address:	Department of Biology & Microbiology NPBL-Box 2140D, Room 252 Brookings, SD 57007
E-mail:	Bruce_Bleakley@sdstate.edu
Phone:	605-688-5498
Fax:	605-688-5624
Year:	FY2004 (approx. May 04 – April 05)
FY04 ARS Agreement ID:	59-0790-0-061
FY04 ARS Agreement Title:	Management of Fusarium Head Blight with Biological Control Agents.
FY04 ARS Award Amount:	\$ 8,780

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
CBC	Management of Fusarium Head Blight with Biological Control Agents.	\$ 8,780
	Total ARS Award Amount	\$ 8,780

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: 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 conducted studies with *Bacillus* strain 1BA to see if it produces iturin and/or surfactin, cyclic lipopeptide antibiotics that have antifungal properties. We initially thought that known iturin and surfactin standards had significantly different retention times than products produced by this *Bacillus* strain, but closer examination of the HPLC traces showed that 1BA produced peaks that were similar if not identical to those of the known standard compounds. We have a HPLC method that will work to separate iturin and surfactin from *Bacillus* broth culture supernatants. However, the analysis time to run one sample is quite long (over one hour), which limits the number of samples that can be run in a reasonable amount of time. We plan to examine other solvent systems for use with HPLC, to see if retention times for iturin and surfactin can be reduced.
- b) We attempted to enumerate the population level of *Bacillus* 1BA, and other bacterial strains applied to wheat and barley at anthesis in field plot experiments to control FHB, to see if these biocontrol agents survive and persist or grow in the field after they are sprayed onto plant material. We were able to get good counts of Gary Yuen's *Lysobacter* strain, because it has known resistance to antibiotics that inhibit most natural microflora that are present on wheat and barley. However, we were unable to obtain good population estimates of the other biocontrol bacteria used in field plot studies, because it was not known what antibiotics and/or other chemical and physical factors could be tolerated by these strains that inhibit the natural microflora resident on wheat and barley heads. We plan to examine chemical and physical factors that can be tolerated by *Bacillus* 1BA which would inhibit the resident normal microflora found on wheat and barley heads at anthesis, so that population estimates of this bacterium can be made on inoculated plant material in field plots. If possible we will use the wild type parental strain of *Bacillus* 1BA in these studies, since creating a mutant strain that is resistant to one or more antibiotics would necessitate that we test the mutant to verify that it had the same characteristics and antagonistic effect against FHB as the parent wild type bacterium.

2. What were the most significant accomplishments?

- a) We verified that both iturin and surfactin are produced in defined broth media by *Bacillus* 1BA; and that the composition of the media and the time of incubation both have an influence on the amounts of these lipopeptides produced. We determined a broth medium composition and incubation time that optimized the amounts of iturin and surfactin produced by 1BA. Hypothesizing that these two lipopeptides are important in the antagonism of FHB, we will use the broth composition and incubation time that provide highest levels of production of these lipopeptides by 1BA when growing it for field plot application.
- b) We verified that application of our biocontrol bacteria resulted in reduction of DON levels in South Dakota field plots in some situations. We continued to work with D. Schisler, G. Yuen, M. Draper, and G. Bergstrom in a collaborative multi-state effort to optimize biocontrol technology for FHB.

Impact: Collaborating scientists benefited from this new scientific knowledge (optimal growth medium and incubation time) by being able to grow 1BA for field plot application under conditions that helped optimize bacterial lipopeptide production. New scientific information included patterns of lipopeptide production by biocontrol strain *Bacillus* 1BA in a defined broth medium. Information will hopefully facilitate development of one or more bacterial biocontrol agents to reduce FHB and/or DON in field situations.

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

Bleakley, B.H., and N.L. Baye. 2004. Use of HPLC in examining culture supernatants of bacteria used in biological control of FHB for the presence of iturin. *In* Canty, S.M., Boring, T., Versdahl, K., Wardwell, J., and Ward, R.W. (Eds.), Proceedings of the 2nd International Symposium on Fusarium Head Blight; incorporating the 8th European Fusarium Seminar; 2004, 11-15 December; Orlando, FL, USA. Michigan State University, East Lansing, MI. p. 292.

Draper, M.A., B. Bleakley, K.R. Ruden, K.D. Glover, S.M. Schilling, D.S. Wittmeier, and G. Lammers. 2004. 2004 Uniform trials for the performance of biological control agents in the suppression of Fusarium Head Blight in South Dakota. *In* Canty, S.M., Boring, T., Versdahl, K., Wardwell, J., and Ward, R.W. (Eds.), Proceedings of the 2nd International Symposium on Fusarium Head Blight; incorporating the 8th European Fusarium Seminar; 2004, 11-15 December; Orlando, FL, USA. Michigan State University, East Lansing, MI. p. 297.

Yuen, G.Y., B.H. Bleakley, M.A. Draper, C.C. Jochum, E.A. Milus, K.R. Ruden, and L.E. Sweets. 2004. Results from the 2004 standardized evaluation of biological agents for the control of Fusarium Head Blight. *In* Canty, S.M., Boring, T., Versdahl, K., Wardwell, J., and Ward, R.W. (Eds.), Proceedings of the 2nd International Symposium on Fusarium Head Blight; incorporating the 8th European Fusarium Seminar; 2004, 11-15 December; Orlando, FL, USA. Michigan State University, East Lansing, MI. pp. 380-382.

Bleakley, B.H. 2005. Biological control of foliar and head diseases of wheat. AD-421 Progress Report (CRIS Report).