

**USDA-ARS / USWBSI
FY04 Preliminary Final Performance Report
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Cover Page

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Year:	FY2004 (June 04 – June 05)
FY04 ARS Agreement ID:	59-0790-4-134
Agreement Title:	Validating a Rapid Immunological Test for <i>Fusarium graminearum</i>.
FY04 ARS Award Amount:	\$ 14,634

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
FSTU	Quantifying <i>Fusarium</i> in Seeds and Grain Products for Laboratory and Industrial Use.	\$ 14,634
	Total ARS Award Amount	\$ 14,634

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: *Quantifying Fusarium in Seeds and Grain Products for Laboratory and Industrial Use.*

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

FHB resistance in small grains is portended to be additive gene action by minor genes. Attempts have been made to find molecular markers for using genetic maps using mapping populations that express variation for FHB resistance (using ocular scores) or the mycotoxin DON in barley. Barley mapping populations generally contain 75 to 85 individual lines necessitating field experimentation because greenhouse space is insufficient to accommodate replicated plantings. Disease expression and DON production are highly variable in the field resulting in high experimental errors (CV's) associated with FHB scores and DON analysis. Consequently, mapping studies have failed to find alleles associated with disease resistance and have only found identifiable genes responsible for morphological (2-row vs. 6-row, plant height, and heading date) features associated with disease avoidance, not resistance. This suggests better methods (field and/or laboratory) are needed to better assess the disease to better identify the true resistance/susceptibility phenotypes and genotypes of mapping and breeding populations. Previously, we developed a monoclonal antibody-based test to quantify *Fusarium graminearum* in barley. The objectives of this project were to 1) compare antibody quantification of *F. graminearum* with that of FHB scores, ergosterol, ELISA quantification of Fusarium, and RT-PCR with DON in commercially harvested seed; 2) compare errors associated with antibody quantification of *F. graminearum*, FHB scores, and DON analysis in North American barley scab evaluation nurseries (NABSEN); and 3) test whether environmental conditions affect DON production and antigen abundance in isolates of *F. graminearum* grown in vitro.

2. What were the most significant accomplishments?

The results from these studies indicate ELISA quantification gave better estimates of *Fusarium* disease infestation than RT-PCR, ocular estimates of incidence or FHB severity because of higher correlations with DON in commercial grain samples. In addition, the ELISA analyses gave lower experimental than FHB scoring and DON analysis. The CV's from these experiments ranged between 21 and 26 for ELISA, 30 to 66 for FHB scores, and 50 to 62.4 for DON. Isolates of *Fusarium graminearum* varied in DON production due to media in which they were grown, osmotic potential of media, and temperature, despite mass of mycelia in cultures not differing (with exception of temperature). The antigen to which the ELISA antibodies are specific did not differ per unit mass of mycelium regardless of environmental variables imposed upon the *Fusarium* cultures. Collectively, these data demonstrate that ELISA quantification of *Fusarium* is a better analytical tool than visual ratings, DON analysis, ergotsterol, or RT-PCR because it provides more accurate and precise estimates of the disease. Improved diagnostics through ELISA quantification of *Fusarium* should yield greater genetic improvement to Fusarium resistance through traditional screening and plant breeding methods as well as enhancing the ability to find molecular markers associated with resistance genes. ELISA can also be used to differentiate potentially toxic from non-toxic commercial grain samples.

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.

Hill, N.S., P. Schwarz, R. Horsley, S. Neate, B. Steffenson, L. Dahleen, B. Cooper, B. Kittle, and A. Jones. 2004. Simple and rapid immunoquantification of *Fusarium* in barley and its relationship with DON and FHB scores. In S.M. Canty, T. Boring, J. Wardwell, and R.W. Ward (eds). Proceedings of the 2nd International Symposium on Fusarium Head Blight; incorporating the 8th European Fusarium Seminar; 11-15 December, 2004; Orlando, FL. East Lansing: Michigan State Univ. pp. 403.

S.M. Neate, P.B. Schwarz, N.S. Hill and R.D. Horsley. 2004. The relationships between *Fusarium* head blight visual symptoms, *Fusarium* biomass and deoxynivalenol levels in barley. In S.M. Canty, T. Boring, J. Wardwell, and R.W. Ward (eds). Proceedings of the 2nd International Symposium on Fusarium Head Blight; incorporating the 8th European Fusarium Seminar; 11-15 December, 2004; Orlando, FL. East Lansing: Michigan State Univ. pp. 415.

N. Hill, A. Jones, S. Neate, P. Schwarz, K. Smith, R. Dill-Mackey, L. Dahleen, R. Horsley, B. Cooper. 2005. Simple and rapid immunoquantification of *Fusarium* in Barley. American Brewing Chemist's Society. 11-15 June, Savannah, Georgia.