

USDA-ARS / USWBSI
FY03 Final Performance Report (approx. May 03 – April 04)
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Cover Page

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Year:	FY2003 (approx. May 03 – April 04)
FY03 ARS Agreement ID:	NA
FY03 ARS Agreement Title:	Global molecular surveillance of FHB species and their mycotoxin potential.
FY03 ARS Award Amount:	\$ 37,073

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
EDM	Global molecular surveillance of FHB species and their mycotoxin potential.	\$ 37,073
	Total Amount Recommended	\$ 37,073

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: Global molecular surveillance of FHB species and their mycotoxin potential.

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

Although the primary etiological agent of FHB, *Fusarium graminearum*, has been regarded as a single, panmictic species worldwide, phylogenetic analyses of DNA sequences from 11 nuclear genes totaling 13.6 kb have shown that this morphospecies actually consists of 11 phylogenetically distinct and biogeographically structured species (hereafter referred to as the *Fg* clade)]. In addition to the unexpectedly high level of species diversity within the *Fg* clade, the virulence-associated trichothecene mycotoxin genes are under a novel form of balancing selection, which may have important consequences for the fitness and aggressiveness of FHB pathogens on particular hosts or in particular environments. PCR-based trichothecene toxin assays have also shown that 6 of the 11 species within the *Fg* clade are still segregating for 15ADON, 3ADON or NIV chemotype. Taken together, these studies suggest that the combined species and mycotoxin diversity of the *Fg* clade is remarkably high. However, it appears that only a fraction of this diversity is currently represented within North America. Therefore, the inadvertent introduction of novel *Fg* clade pathogens or chemotypes into the U. S. via global trade has the potential to exacerbate the FHB problem. To address this issue, we have developed protocols for the PCR multiplex amplification of two sets of chemotype-specific primers designed from TRI3 and TRI12 within the trichothecene toxin gene cluster to examine chemotype diversity in a collection of FHB isolates from the U. S., China and Brazil. Development of robust molecular tools for *Fg* clade species identification and chemotype determination have significantly improved disease surveillance and global monitoring efforts, and will make available for the first time detailed information on the geographic and host distributions of FHB pathogens and their trichothecene chemotypes, enhancing current knowledge of the ecology, epidemiology and population dynamics of these mycotoxigenic cereal pathogens.

2. What were the most significant accomplishments?

Two multiplex PCR tests for chemotype identification of B-trichothecene FHB pathogens have been developed and validated. Using these tests, chemotype diversity has been assessed in a collection of 900 isolates from the U.S., 200 isolates from China, 75 isolates from Brazil, and 19 isolates from Italy. Within the U.S., the vast majority of isolates (89.5%) have a 15ADON chemotype. The 3ADON chemotype was found in 10% of isolates, but was geographically limited, occurring only in North Dakota and Minnesota. The NIV chemotype was found at low frequency (0.5%). Chemotype frequencies were more balanced within Brazil and China, although the 15ADON chemotype was completely absent from the three Chinese populations surveyed to date. Work on screening 1814 FHB strains for species identification from the countries listed above and from a broader global collection, based on DNA sequence data from two loci totaling 2.48 megabase pairs, has been completed. Multilocus DNA sequence data has identified two additional species of B-trichothecene producing fusaria, bringing the total number of phylogenetically distinct species in this group to 14. DNA sequencing of the A-trichothecene FHB portion of the project had been completed, with portions of seven loci sequenced totaling 7 kb of DNA sequence. Preliminary analysis of these data indicates that 25 phylogenetically distinct species are represented in this collection of A-trichothecene FHB pathogens. This DNA sequence database will serve as the basis for the development of high thru put molecular methods for the accurate identification of FHB pathogens, host/geographic distributions and their mycotoxin potential.

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.

Peer-reviewed articles:

O'Donnell, K., Ward, T.J., Geiser, D.M., Kistler, H.C. and Aoki, T. 2004. Genealogical concordance between the mating type locus and seven other nuclear genes supports formal recognition of nine phylogenetically distinct species within the *Fusarium graminearum* clade. *Fungal Genetics and Biology* 41:600-623.

Geiser, D.M., del Mar Jiménez-Gasco, M., Kang, S., Makalowska, I., Veeraraghavan, N., Ward, T.J., Zhang, N., Kuldau, G.A., O'Donnell, K. 2004. FUSARIUM-ID v.1.0: A DNA sequence database for identifying *Fusarium*. *European Journal of Plant Pathology* 110:473-479.

Presentations:

Poster/Published abstract:

O'Donnell, K., Ward, T.J., Geiser, D.M., Kistler, H.C., Gale, L.R., and Aoki, T. Global genetic diversity of *Fusarium graminearum* clade species and their mycotoxin potential. National Fusarium Head Blight Forum Proceedings. p. 149. 2003.

Gale, L.R., Ward, T.J., Balmas, V., Kistler, H.C. Detection of distinct subpopulations of *Fusarium graminearum*, lineage 7. National Fusarium Head Blight Forum Proceedings. p.131. 2003.

Gale, L.R., Ward, T.J., Balmas, V., Kistler, H.C. Population subdivision in *Fusarium graminearum* lineage 7 in the United States is correlated with toxin chemotype. 22nd Fungal Genetics Conference. v.50 (Suppl.) p. 145. 2003.

Kistler, H.C., Birren, B., Ma, L.-J., Calvo, S., Galagan, J., Gale, L.R., O'Donnell, K., Trail, F., Ward, T., and Xu, J.-R. The whole genome sequence of the wheat and barley pathogen *Fusarium graminearum*. Abstracts of the 7th European Conference on Fungal Genetics. p.210. 2004.

Kuldau, G.A., Ward, T.J., O'Donnell, K., Archibald, D.D., Jimenez-Gasco, M., Zitomer, N., Geiser D.M. Reassessment of type-A trichothecene-producing *Fusarium* using molecular phylogenetics and HPLC-MS toxin profiles. Chemical and Biological Terrorism Defense Gordon Conference. Buelton, CA, 2004.

Zitomer, N.C., Geiser, N.C., Ward, T.J., Jiminez-Gasco, M., Archibald, D.D., O'Donnell, K., Kulda, G.A. The use of HPLC-MS to characterize toxin-production from fusaria based on new phylogenetic concepts within the genus. American Phytopathological Society Meeting. Anaheim, CA, 2004.

Invited Lectures:

O'Donnell, K. Discordant evolution of trichothecene toxins and species within the *Fusarium graminearum* clade: Evidence from multigene genealogies. Seminar presented at the Department of Plant Pathology, Pennsylvania State University, 2003.

O'Donnell, K. Discordant evolution of trichothecene toxins and species within the *Fusarium graminearum* clade: Evidence from multigene genealogies. Seminar presented at the Department of Plant Pathology, Michigan State University, 2003.

Ward, T.J. An evolutionary framework for the comparative and functional genomics of the *Fusarium graminearum* species complex. 2nd International *Fusarium* Genomics Workshop held in conjunction with the 7th European Conference on Fungal Genetics, Copenhagen, Denmark, 2004.

Ward, T.J. Unusual patterns of evolution in the trichothecene mycotoxin gene cluster of plant-pathogenic *Fusarium*, Department of Biological Sciences, Illinois State University, Normal, Illinois, 2003.