

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:	Genomics, population genetics and development of Gibberella zeae.
FY03 ARS Award Amount:	\$ 76,098

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
EDM	Diversity of Gibberella zeae populations from the U.S., China and Italy.	\$ 48,781
EDM	Genomics of Gibberella zeae, the head scab fungus.	\$ 27,317
	Total Amount Recommended	\$ 76,098

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: *Diversity of Gibberella zae populations from the U.S., China and Italy.***1. What major problem or issue is being resolved and how are you resolving it?**

The re-emergence of FHB in past decade is likely due to a combination of factors including unfavorable climatic conditions, changes in agronomic practices and the lack of high levels of genetic resistance in currently planted wheat and barley. Another unknown but potentially important factor for the disease is the level of genetic variation in the pathogen. In order to assist effective plant breeding and disease management programs, it is essential to understand the sources and extent of genetic variation in the head blight pathogen both in the U.S. and worldwide.

Genetic diversity of populations from China, the U.S. and Italy have been inferred based on allele differences at polymorphic, single copy loci defined by RFLPs and multilocus haplotypes have been constructed for each strain. Genetic data on strains have been arranged into geographic populations corresponding to the country of origin or defined regions within each country (e.g. state, county, field etc.) and analyzed according to geographic source. In order to determine the degree of outcrossing in the fungus, the extent of linkage disequilibrium between pairs of loci were calculated.

2. What were the most significant accomplishments?

In our analysis of 702 *F. graminearum* strains collected in 1999 – 2000 from 89 fields in 53 counties in nine Midwestern states, only 31 strains were determined to be 3 acetyl deoxynivalenol (3ADON) producers, while the overwhelming majority of isolates were 15 acetyl deoxynivalenol (15ADON) producers. Only two nivalenol producing strains were identified, one from Missouri and one from Minnesota. 3ADON strains were limited in distribution to ND and northwestern MN. Limited frequency (4.4% of total strains and 7% of MN and ND strains) and distribution may indicate recent introduction of 3ADON strains and/or reduced fitness of these strains.

In 2003 we made collections of the Fusarium head blight pathogen to learn more about the population genetic dynamics of the 3ADON population. Sample sites included counties in MN and ND where 3ADON strains had been previously obtained, as well as additional counties in the two states, from which 3ADON strains either had not been recovered previously and/or from which no *F. graminearum* samples have been examined yet. Tests are underway to determine the distribution and the frequency of the 3ADON chemotype in ND and MN, and whether the 3 ADON strains are being assimilated by recombination into the resident North American 15 ADON population.

Project 2: *Genomics of Gibberella zeae, the head scab fungus.***1. What major problem or issue is being resolved and how are you resolving it?**

There is a lack of knowledge concerning the way in which the head blight pathogen, *Fusarium graminearum* causes disease in plants. This basic knowledge will be required to develop novel strategies for the control of the disease and the mycotoxins produced by the fungus. Genomics technology makes it possible to study the expression of potentially all of the genes in an organism. Agricultural scientists have begun using this technology to improve crops and study pathogenicity. A genome project for the scab fungus provides a unique opportunity to harness this technology for the study of the disease cycle of this important fungus. One direct method to access a large number of expressed genes is to partially sequence individual clones from a cDNA library, called Expressed Sequence Tags (ESTs). Our goal was to sequence ESTs from libraries created from *Fusarium graminearum*-infected wheat heads subtracted with wheat heads infected with a low virulence strain of the fungus or mock inoculated plants.

2. What were the most significant accomplishments?

Fungal genes expressed during the initial stages of infection on wheat were identified using suppressive subtractive hybridization. A subset of up-regulated genes have been targeted for gene deletion to ascertain their role in pathogenicity. Two such genes, encoding a putative histidine kinase and ABC transporter have been confirmed to be up-regulated in infected plants and deletion mutants of the genes have been created. Tests are underway to determine if strains containing deletion mutations are altered in pathogenicity.

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.

Gale, L.R., Ward, T.J., Balmas, V., and Kistler, H.C. Detection of distinct subpopulations of *Fusarium graminearum* lineage 7 in the U.S. 2003 National Fusarium Head Blight Forum Proceedings. p. 139. 2003.

Goswami, R., Trail, F., Xu, J.R. and Kistler, H.C. Analysis of gene expression in *Fusarium graminearum* during infection on wheat. 2003 National Fusarium Head Blight Forum Proceedings. p. 140. 2003.

Kistler, H.C., Birren, B., Calvo, S., Galagan, J., Gale, L.R., Ma, L.-J., Trail, F., and Xu, J.R. The whole genome sequence of *Fusarium graminearum* lineage 7. 2003 National Fusarium Head Blight Forum Proceedings. p. 141. 2003.

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. 2003 National Fusarium Head Blight Forum Proceedings. p. 149. 2003.

Seong, K., Zhao, X., Tracy, M., Trail, F., Kistler, H.C. and Xu, J.R. Identifying virulence factors in *Fusarium graminearum* using forward and reverse genetic approaches. 2003 National Fusarium Head Blight Forum Proceedings. p. 176. 2003.

Seong, K., Tracy, M., Kistler, H.C. and Xu, J.R. REMI mutagenesis *Fusarium graminearum*. 2003 National Fusarium Head Blight Forum Proceedings. p. 177. 2003.

Kistler, H.C., Gale, L.R., Xu, J.-R., Trail, F., Birren, B. Genomics of *Fusarium graminearum*: Priorities and plans for genome annotation, functional analysis and integration of the genetic map with the draft genome sequence assembly. Abstracts for the 3rd Conference on Fungal Genetics and Molecular Biology, The Japan Society for Fungal Genetics and Molecular Biology, p. 10-11. 2003.

Kistler, H.C., Birren, B., Calvo, S., Galagan, J., Gale, L.R., Ma, L.-J., Trail, F., Xu, J.-R. The whole genome sequence of the wheat and barley scab fungus, *Fusarium graminearum*. Abstracts for the Plant and Animal Genome XII conference. p.89. San Diego, CA, 2004.

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. Copenhagen, Denmark. 2004.

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.

Suga, H., Gale, L.R. and Kistler, H.C. 2004. Development of VNTR markers for two *Fusarium graminearum* clade species. *Molecular Ecology Notes* 4, in press.