USDA-ARS/
U.S. Wheat and Barley Scab Initiative
FY07 Final Performance Report (approx. May 07 – April 08)
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Cover Page

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Fiscal Year:	2007
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USDA-ARS Agreement Title:	Genetic Diversity and Genetic Mapping of *Gibberella zeae*.
FY07 ARS Award Amount:	$ 32,629

USWBSI Individual Project(s)

USWBSI Research Area*	Project Title	ARS Adjusted Award Amount
PGG	Genetic Diversity in and Genetic Mapping of *Gibberella zeae*.	$32,629
	Total Award Amount	**$ 32,629**

15 July 2008
Principal Investigator Date

* CBCC – Chemical, Biological & Cultural Control
EEDF – Etiology, Epidemiology & Disease Forecasting
FSTU – Food Safety, Toxicology, & Utilization of Mycotoxin-contaminated Grain
GET – Genetic Engineering & Transformation
HGR – Host Genetics Resources
HGG – Host Genetics & Genomics
IIR – Integrated/Interdisciplinary Research
PGG – Pathogen Genetics & Genomics
VDUN – Variety Development & Uniform Nurseries

Project 1: *Genetic Diversity in and Genetic Mapping of Gibberella zeae.*

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

A number of phylogenetic lineages (now 11) are known with *G. zeae* and have been formally described as phylogenetic species. Considerable genetic diversity in gene sequence exists between the lineages, but the lineages are not reproductively isolated under laboratory conditions suggesting that the lineages may represent (formerly) isolated populations and not distinct species. Published sequences for lineage-associated genes are from a relatively few strains. We sequenced four genes in *G. zeae* strains from the United States to determine whether the published sequences were representative of those present in the field. These same genes also have been sequenced from 472 isolates representing multiple lineages from South America, which should enable a comparison of these populations as well. From these sequences we will be able to determine the relatedness of these populations and determine if there are unique sequences present in the US populations of this fungus.

2. List the most important accomplishment and its impact (how is it being used?). Complete all three sections (repeat sections for each major accomplishment):

Accomplishment:

Previously, Amplified Fragment Length Polymorphisms (AFLPs) were used to evaluate the genetic diversity of 523 isolates from North America and assign them to one of 503 haplotypes (presumptive clones) belonging to *F. graminearum* lineage 7 (*F. graminearum sensu stricto*). We sequenced four genes [Mating Type (*MAT*), *TRI-101*, Reductase (*RED*), and β-tubulin (*TUB2*)] from strains representing ~350 of these haplotypes. In all cases, multiple alleles were found (some not previously reported): *RED* – 19 (15) alleles, *TUB2* – 3 (1) alleles, *MAT* – 7 (4) alleles, and *TRI101* – 10 (7) alleles. All alleles present at a frequency > 1% were recovered from multiple locations, suggesting that these populations are well-mixed. The new alleles represent 35% of the total *RED* sequences, < 1% of the *TUB2* sequences, 2.2% of the *MAT* sequences and 9.4% of the *TRI101* sequences.

Genetic relatedness networks for these four genes (using the available lineage tester strains to represent nine of the eleven described lineages) are not homologous with regards to the association of the lineages. This lack of concordance between gene genealogies is consistent with a hypothesis that these lineages are distinct but that all form a part of a single species and should not yet be accorded species status. The polytomies that characterize these genealogies also are consistent with all nine lineages being members of the same species rather than members of a number of different species. The data from the North American strains blurs the differences between lineage 7 and the other lineages by identifying additional alleles that are intermediate between the described alleles and those in lineage 7. These data also provide evidence for intragenic recombination at some loci, which when combined with the relatively large number of alleles suggests that these populations are relatively old and probably stably established.

Impact:

The taxonomic status of *G. zeae*/*F. graminearum* is of critical importance for plant quarantine and trade measures. If there are a number of species then each must be treated separate-

ly and the presence/absence of a particular species can be used as a non-tariff trade barrier. Our results strongly suggest that while isolated populations of *F. graminearum* exist, these populations are not reproductively isolated and should be recognized as portions of a single, large, diverse species rather than as nine discrete entities. Sequencing of additional DNA variants is consistent with this conclusion as alleles that are "missing" from the initial alignments are found and begin to blur the distinctness of the various groups. Recognition of a single species does not materially impact the plant quarantine regulations currently in place, nor alter the application/implementation of current trade practices.

As a result of that accomplishment, what does your particular clientele, the scientific community, and agriculture as a whole have now that they didn't have before?

The controversy over species or lineage as the proper designation for the groups observed within *F. graminearum* has been ongoing. These data suggest that a single species is most appropriate since the differences between lineages are now less distinct than they were in the past. They also suggest that lineage/speciation techniques that rely on one or a few single nucleotide polymorphisms for their diagnostic capability have an error rate that has yet to be quantified, and that these techniques should not be used beyond the research level until the error rates are determined.

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.

Books:

1. **Leslie, J. F.**, R. Bandyopadhyay & A. Visconti, eds. 2008. *Mycotoxins: Detection Methods, Management, Public Health and Agricultural Trade.* CABI, Kew, UK. 476 pp.

Refereed journal articles:

1. Hornok, L., C. Waalwijk & **J. F. Leslie**. 2007. Genetic factors affecting sexual reproduction in toxigenic *Fusarium* species. *International Journal of Food Microbiology* **119:** 54-58.
2. **Leslie, J. F.**, L. L. Anderson, R. L. Bowden & Y.-W. Lee. 2007. Inter- and intra-specific genetic variation in *Fusarium*. *International Journal of Food Microbiology* **119:** 25-32.
3. Ramirez, M. L., M. M. Reynoso, M. C. Farnochi, **J. F. Leslie** & S. N. Chulze. 2007. Population genetic structure of *Gibberella zeae* from wheat in Argentina. *Food Additives and Contaminants* **24:** 1115-1120.
4. Bentley, A. R., **J. F. Leslie**, E. C. Y. Liew, L. W. Burgess & B. A. Summerell. 2008. Genetic structure of *Fusarium pseudograminearum* populations from the Australian grain belt. *Phytopathology* **98:** 250-255.
5. Lee, J., **J. F. Leslie** & R. L. Bowden. 2008. Expression and function of sex phermones and receptors in the homothallic ascomycete *Gibberella zeae*. *Eukaryotic Cell* **7:** 1211-1221.
6. Lee, J., J. E. Jurgenson, **J. F. Leslie** & R. L. Bowden. 2008. Alignment of genetic and physical maps of *Gibberella zeae*. *Applied and Environmental Microbiology* **74:** 2349-2359.
7. Bentley, A. R., M. G. Milgroom, **J. F. Leslie**, B. A. Summerell & L. W. Burgess. 2008. Spatial aggregation in *Fusarium pseudograminearum* populations from the Australian grain belt. *Plant Pathology* (in press).
8. Bowden, R. L., I. Fuentes-Bueno, **J. F. Leslie**, J. Lee & Y.-W. Lee. 2008. Methods for detecting chromosomal rearrangements in *Gibberella zeae*. *Cereal Research Communications* (in press).
9. **Leslie, J. F.**, and R. L. Bowden. 2008. *Fusarium graminearum*: When species concepts collide. *Cereal Research Communications* (in press).

Book chapters:

1. Bandyopadhyay, R., R. A. Frederiksen & **J. F. Leslie**. 2008. Priorities for mycotoxin research in Africa identified by using the nominal group technique. In: *Mycotoxins: Detection Methods, Management, Public Health and Agricultural Trade* (J. F. Leslie, R. Bandyopadhyay & A. Visconti, eds.), pp. 19-26. CABI, Kew, UK. 476 pp.
2. Shelton, B. G. & **J. F. Leslie.** 2008. Comparative risks of airborne and foodborne molds and mycotoxins. In: *Mycotoxins: Detection Methods, Management, Public Health and Agricultural Trade* (J. F. Leslie, R. Bandyopadhyay & A. Visconti, eds.), pp. 317-324. CABI, Kew, UK. 476 pp.

Abstracts and meeting presentations:

1. Anderson, L. L., Y.-W. Lee, R. L. Bowden & **J. F. Leslie**. 2007. Relationships between alleles at lineage diagnostic loci in *Fusarium graminearum*. *Fungal Genetics Newsletter* **54(Suppl.):** 67.
2. Lee, J., R. L. Bowden & **J. F. Leslie**. 2007. Pheromone functions in *Gibberella zeae*. *Fungal Genetics Newsletter* **54(Suppl.):** 134.
3. Lee, J., **J. F. Leslie** & R. L. Bowden. 2007. Functions of the sex pheromones of *Gibberella zeae*. *Proceedings of the 2007 National Fusarium Head Blight Forum (Kansas City, Missouri)*: 30.
4. Reynoso, M. M., M. L. Ramirez, **J. F. Leslie** & S. N. Chulze. 2007. Trichothecene chemotypes of isolates of *Gibberella zeae* recovered from wheat in Argentina. *Proceedings of the 2007 National Fusarium Head Blight Forum (Kansas City, Missouri)*: 34.

Dates and locations of invited presentations by Dr. Leslie that contained information from this project but for which there is no published abstract:

1. Bioforsk, Ås, Norway – 03/07.
2. College of Life Sciences, Dalian Nationalities University, Dalian, China – 04/07.
3. Shenyang Agricultural University, Shenyang, China – 04/07.
4. Faculty of Agricultural & Life Sciences, Seoul National University, Seoul, Korea – 05/07.
5. FABI, University of Pretoria, Pretoria, South Africa – 11/07.
6. Norwegian National Veterinary Institute, Oslo, Norway – 04/08.
7. Faculty of Agricultural & Life Sciences, Seoul National University, Seoul, Korea – 05/08.