

Sustainable Plant Protection Systems



Grain Molds, Mycotoxins and Stalk Rots of Sorghum and Millet

Project KSU 101
John F. Leslie
Kansas State University

Principal Investigator

John F. Leslie, Dept. of Plant Pathology, Kansas State University, Manhattan, Kansas 66506-5502

Collaborating Scientists

Dr. Ranajit Bandyopadhyay, International Institute for Tropical Agriculture, Ibadan, Nigeria
Dr. R. L. Bowden, USDA-ARS Plant Science & Entomology Research Unit, Manhattan, Kansas
Dr. A. E. Desjardins, NCAUR, USDA-ARS, Peoria, Illinois
Dr. Mamourou Diourté, IER, Bamako, Mali
Dr. J. Peter Esele, Sererre Agric. & Animal Production Res. Institute, NARO, Soroti, Uganda
Dr. Laszlo Hornok, Agricultural Biotechnology Center, Godollo, Hungary
Drs. D. J. Jardine, C. R. Little, J. S. Smith & C. Toomajian, Kansas State Univ., Manhattan, KS
Dr. Chagemu J. Kedera, Kenyan Plant Health Inspection Service, Nairobi, Kenya
Drs. W. F. O. Marasas, G. S. Shepard & H. F. Vismer, Med. Res. Council, Tygerberg, So. Africa
Dr. Neal McLaren, University of the Free State, Bloemfontein, South Africa
Drs. Michael Wingfield and Brenda Wingfield, University of Pretoria, Pretoria, South Africa

Introduction and Justification

Sorghum and millet are plagued by numerous diseases, most of which have a fungal etiological agent. Stalk rot and grain mold, the most important diseases on a worldwide basis for which there is no effective management regime can be caused by several species of *Fusarium*, although at least 25 additional fungal genera may be present as secondary invaders or members of a disease complex. Separating and identifying the roles and risks associated with the various members of this complex fungal community is necessary to estimate the risks posed by different members of the community and to provide breeders with the correct targets for resistance breeding. Fungi that cause grain mold also are linked with stand establishment problems as the seeds that are produced may germinate poorly or the germinated seedlings may be killed by fungi that accompanied the seed.

Fusarium spp. and the secondarily invading *Aspergillus* spp. may produce mycotoxins such as aflatoxins, fumonisins, ochratoxin, deoxynivalenol and zearalenone. These toxins may reduce the quality of the grain as a food/feed source as well as the value of the grain in a cash market scenario. These toxins are associated with a variety of human and animal health problems including acute toxicity and death, increased incidence of cancer, inhibition of normal growth and development, immune suppression and increased disease susceptibility, increased risks of birth defects, and reduced nutritional and economic value of the resulting grain. In most host-country settings these risks are inadequately quantified due to limited medical data reporting systems.

Fusarium and related species and the diseases they cause offer the most attractive targets for improved management that could be of importance in a global context. Isolates of *Fusarium* recovered

from sorghum and millet have long been a taxonomist's nightmare. Many species lack morphological characters that can be used to clearly and cleanly differentiate them from other related species, and many cultures are misidentified, if identified at all. Many of these cultures also have been identified as *Fusarium moniliforme*, a name that has now been abandoned due to the numerous species that it has been associated with. As all strains with the *F. moniliforme* name often were assumed (incorrectly) to be equivalent in terms of pathogenicity, breeding materials often were challenged with an improper strain with correspondingly inconsistent results. For example, *F. verticillioides* is a common pathogen of maize that once was termed *F. moniliforme*, as was *F. thapsinum*, a major cause of sorghum stalk rot. Challenging sorghum plants with *F. verticillioides* when screening for stalk rot resistance results in unpredictable results, as the only plants that become diseased are those infected by *F. thapsinum* due to natural causes. A similar challenge with *F. thapsinum*, however, can effectively flatten an experiment planted with a sensitive variety. Results from previous studies sponsored by INTSORMIL have indicated that the dominant *Fusarium* species varies by location, e.g., *Fusarium andyazi*, in southern Africa, *F. thapsinum* in West Africa, *F. proliferatum* in Egypt, and an as yet unnamed new species that is common from West Africa through Egypt and East Africa (Kenya and Uganda). Within region variation suggests that as many as 20 additional species remain to be described. Until they have been effectively separated it is difficult to determine which species are common in one area and less common in others. Such studies also are needed to enable breeders to effectively challenge the materials in their programs. The *Fusarium* species associated with pearl millet and finger millet also have been examined in a somewhat cursory manner. *Fusarium pseudonygamai* is the dominant species on pearl millet, while finger millet is host to an amazingly diverse group of *Fusarium* spp. (between 40 and 60 from samples taken in Uganda

in 2000). The *Fusarium* species on these crops are not known to be associated with production problems, but may produce mycotoxins that could contaminate grain. Identifying the toxins produced, if any, and their levels is particularly important for strains found on finger millet as this grain often is used to produce a weaning food for children. These children would be particularly susceptible to the reductions in mental and physical development that can result from sub-acute exposure to these toxins.

Objectives and Implementation Sites

- Identification of *Fusarium* species associated with pearl and finger millet and with grain mold and stalk rot of sorghum. Kansas, South Africa, Mali and Uganda.
- Mycotoxins in sorghum and millets. Kansas, South Africa and Nigeria.
- Strengthen host-country research capacity. Kansas, South Africa (Malaysia & South Korea).

Contribution to INTSORMIL Objectives

Collectively, the planned work impacts INTSORMIL objectives 2, 4, 5 and 7. Fewer mycotoxins in the grain improve food and nutritional quality of sorghum and pearl millet. Reduced disease pressure increases the yield and yield stability. Information on biotic stresses is being disseminated through the existing workshops and co-authored scientific publications and the training of graduate students and visiting scientists. Assisting INTSORMIL breeders with the development of germplasm resistant to various pathogens increases yield and yield stability.

Research Methodology and Strategy

Species Identification

After field collection, strains are subcultured to a selective medium to purify cultures from bacterial and most other fungal contaminants. These cleaned cultures are genetically purified by sub-culturing individual macro- or microconidia (of uninucleate origin) that have been separated from the remainder of the colony by micromanipulation. Three different species concepts are used in *Fusarium* – morphological, biological and phylogenetic. Most species from sorghum and millet are very similar to one another morphologically, which means that the morphological characters are insufficient to differentiate the species, thus either biological or phylogenetic concepts and strategies are usually employed after an initial morphological observation confirms that the strains have the morphological characters common to most sorghum/millet *Fusarium* species. At this point cultures are grown for three days and DNA is isolated from all strains. DNA from strains is run through an Amplified Fragment Length Polymorphism (AFLP) protocol. At the end of the first run, strains with visibly similar patterns are grouped together and rerun to confirm their similarity. Genes with species specific sequences, usually one encoding β -tubulin (tub-2) and/or another encoding translocation elongation factor 1- α (tef-1) are amplified by PCR and sequenced. If there is less than 1% difference between the sequences obtained and those available for standard strains, then the group is considered to have been successfully identified. If there are tester strains available for

sexual crosses for a known species, then the identity of the remaining strains in the group are confirmed by crosses.

In many cases for strains from sorghum and millets in Africa, the species is one that has not been described. In such cases, additional strains are sequenced to confirm that the first set of sequence data typifies the group. At this time, a search for the sexual stage begins. Crosses are made in all possible pairwise combinations of all strains, with each strain serving as both the male and as a female parent in a cross (this results in the number of crosses made being the square of the number of strains in the group, e.g., 50 strains => 2500 crosses that must all be re-peated at least twice => 5000 crosses total), with the goal of finding strains that are fertile as the female parent. The number of crosses can be reduced by up to 1/2 if the mating type of the strains can be determined molecularly before the crossing process begins. Once fertile strains are identified, female fertility usually must be improved through crosses with other female fertile strains, which may be a very time-consuming process. Once the sexual stage has been successfully identified then photographs of critical morphological features are made, strains are deposited in appropriate international culture collections and herbaria and the new species can be written up for publication. No more than 2-3 new species can be processed at any single time.

Most of this work is done at KSU with samples collected from numerous African countries including Egypt, Ethiopia, Mali, Nigeria and South Africa with the help of colleagues based there.

Mycotoxin Production

In vitro assessment of mycotoxin production requires collaboration with other scientists who are equipped with the necessary apparatus for chemical analyses. The presence of the fumonisin, beauvericin and fusaproliferin mycotoxins can be evaluated after growth on rice in laboratory culture for up to 30 days. The contents are extracted in acetonitrile:water, run through a clean-up column to remove contaminants, derivatized, if necessary, to enable detection, and finally quantified by using an HPLC protocol.

Converted rice (usually the Uncle Ben's brand) commonly is used for these studies and the toxin levels produced can be high. Three mycotoxins were tested: fumonisin, fusaproliferin and beauvericin. Fumonisin is important due to their association with esophageal cancer and neural tube defects in humans, their ability to cause a number of diseases in domesticated animals, and their effects as trade barriers. Fusaproliferin and beauvericin are not associated with a disease syndrome of humans or domesticated animals but are toxic to cells maintained in cell culture. Beauvericin is insecticidal and yeasts that synthesize this compound have been used as bio-control agents. Beauvericin also is very effective in permeabilizing cell membranes and thereby facilitating the entry of other mycotoxigenic molecules.

Strengthening Research Capacity

Present workshops on Scientific Writing and Scientific Research Ethics as requested. Organize annual *Fusarium* Laboratory workshop.

Research Results

Species Identification – Mali

A large pre-existing fungal population isolated from sorghum stored on farm in rural Mali is being analyzed for DNA polymorphisms and species identification. All cultures have been cleaned and purified by the micromanipulation of single spores to yield pure cultures. DNA has been extracted from most of the nearly 1200 strains, some AFLP comparisons have been run, enabling preliminary working groups of strains to be identified. These AFLP comparisons are being rerun with different primer pairs to confirm the preliminary results. Most of the observed patterns are significantly different from those of known species. Molecular work on this group has been limited as we worked to develop highly fertile mating type testers needed for description and for practical identification purposes of the new species. We have recently (after a series of some 15 backcrosses and sib crosses) tentatively identified suitable strains for use as testers for both mating types in this fungus. These testers are now being tested for their utility in identifying the species through crosses with field-collected isolates.

Species Identification – Uganda

A group of 400 strains from finger millet collected in Uganda are in a more advanced state of evaluation. Approximately 20% of the group appeared morphologically to belong to *Fusarium verticillioides*, a species known to produce fumonisins. Following AFLP tests, this set could be subdivided into two groups. The members of one group (32 strains) crossed readily with the tester strains of *F. verticillioides* and produced fumonisin B1 and B2 mycotoxins (Table 1). These strains were polymorphic at tub-2 (28 strains sequenced) with 21 strains with the usually dominant allele and seven strains with an allele that is usually rare. In *tcf-1*, there were five alleles present, an unusually high number, with three of them being new for this species, another unusually high number. Five nucleotide positions were altered in one or more of the alleles, with one change at a silent (no amino acid change) location in the coding sequence and the other four in one of the introns. All but one strain was capable of serving as both a male and a female parent in a cross, suggesting that sexual recombination is both frequent and important in the *F. verticillioides* life cycle in this environment. The high levels of female fertility are unusual as other populations of this fungus often have 50%, and sometimes

Table 1. Toxin and fertility status of strains of *F. verticillioides* collected from finger millet in Uganda. N.D. – No Data.

Strain Number	Location	MAT Allele	♂/♀ Fertility	Fumonisin (µg/g)	
				B ₁	B ₂
9374	Apac Kole, Akolo	A-2	♂ + ♀	N.D.	N.D.
9397		A-2	♂ + ♀	30	60
9401		A-2	♂ + ♀	980	990
9646	Tororo, Tororo, Mukuja	A-1	♂ + ♀	N.D.	N.D.
9669		A-2	♂ + ♀	790	440
9689		A-2	♂ + ♀	96	38
9705		A-2	♂ + ♀	1,030	430
10069	Bugiri, Bukawli, Kapyanga	A-1	♂ + ♀	N.D.	N.D.
10072		A-2	♂ + ♀	N.D.	N.D.
10076		A-1	♂ + ♀	5,700	4,200
10077		A-1	♂ + ♀	N.D.	N.D.
10082		A-2	♂ + ♀	8	4
10084		A-1	♂	12,400	6,200
10086		A-1	♂ + ♀	740	560
10087		A-1	♂ + ♀	160	110
10089		A-2	♂ + ♀	290	140
10090		A-2	♂ + ♀	N.D.	N.D.
10093		A-1	♂ + ♀	450	340
10099		A-1	♂ + ♀	750	620
10108		A-2	♂ + ♀	460	490
10112		A-1	♂ + ♀	12	8
10115	A-1	♂ + ♀	1300	450	
10116	A-1	♂ + ♀	250	190	
10120	A-2	♂ + ♀	74	54	
10125	A-2	♂ + ♀	2	1	
10133	A-2	♂ + ♀	56	34	
10134	A-1	♂ + ♀	540	175	
10136	A-2	♂ + ♀	230	130	
10137	A-2	♂ + ♀	60	490	
10139	A-2	♂ + ♀	130	60	
10142	A-2	♂ + ♀	N.D.	N.D.	
10143	A-2	♂ + ♀	1100	480	

Table 2. Toxins produced by 55 strains of *Fusarium sp. nov.* isolated from finger millet in Uganda. All strains are male fertile, with female fertility still being tested. N.D. – No Data; T – trace (0.1-0.99 µg/g). None of the strains produced detectable levels of fusaproliferin or enniatins.

Strain Number	Location	MAT Allele	Total Fumonisin (µg/g)	Beauvericin (µg/g)
9440	Apac Kole, Akolo	K-2	39	1410
10314	Serota, Serrere	N.D.	3.1	< 1.0
10316		K-2	1.0	< 1.0
10317		K-2	1.7	2,700
10319		K-1	2.5	3,500
10320		K-1	1.5	740
10323		K-1	< 0.1	< 1.0
10324		K-2	N.D.	N.D.
10325		K-1	T	< 1.0
10329		K-1	< 0.1	1,400
10330		K-2	< 0.1	< 1.0
10336		K-2	N.D.	N.D.
10339		K-1	N.D.	N.D.
10340		K-2	T	4,200
10341		K-1	< 0.1	< 1.0
10342		K-1	< 0.1	N.D.
10344		K-2	N.D.	1,700
10347		K-1	T	560
10349		K-1	< 0.1	< 1.0
10350		K-2	< 0.1	< 1.0
10351		K-2	< 0.1	830
10352		K-2	< 0.1	< 1.0
10353		K-1	N.D.	N.D.
10354		N.D.	< 0.1	< 1.0
10355		K-1	< 0.1	160
10356		K-1	T	930
10357		K-1	< 0.1	4,500
10359		N.D.	N.D.	N.D.
10361	K-1	< 0.1	610	
10362	K-1	N.D.	N.D.	
10363	K-1	< 0.1	6,400	
10364	K-1	< 0.1	2,000	
10365	K-1	< 0.1	1,600	
10366	K-1	< 0.1	860	
10367	K-1	T	47	
10368	N.D.	< 0.1	5,500	
10369	K-1	T	590	
10370	K-1	< 0.1	1,300	
10371	Serota, Serrere	K-1	< 0.1	600
10372		K-1	T	1,000
10373		K-1	< 0.1	3,700
10376		K-1	< 0.1	1,200
10377		K-1	N.D.	N.D.
10378		K-2	n.d.	830
10379		K-2	T	940
10380		K-1	T	990
10381		K-1	N.D.	N.D.
10382		K-1	T	2,100
10385		K-1	< 0.1	930
10386		K-1	T	380
10387		K-1	< 0.1	130
10388		K-1	< 0.1	2,000
10389		K-1	< 0.1	770
10390		K-1	< 0.1	810
10392		K-1	< 0.1	2,200

fewer, of the strains that can function as female parents. The high levels of polymorphism and sexual recombination in this strain set are typical of those found in an area near the center of diversity for a species. The levels of toxin these strains can produce suggests that they are of potential concern as contaminants of finger millet grain when it is processed into human food.

The second group of strains (Table 2) is a new biological species that remains to be named and formally described. This species is a part of the *Liseola* section of the genus, i.e., the section to which most of the other *Fusarium* strains isolated from sorghum and millet belong. Although it is morphologically indistinguishable from *F. verticillioides*, the new species can be differentiated on the basis of toxin production (produces beauvericin but little or no fumonisins), AFLP banding patterns, DNA sequence data for the *tef-1* and *tub-2* loci, and cross fertility amongst members of the species but not with members of *F. verticillioides*. This biological species also appears to be quite fertile. In preliminary tests that are now being verified, most of the isolates can serve as both the male and the female parent of a cross. Thus, sexual recombination is frequent in this species. Unlike the new species from Mali, identification of good female fertile tester strains was easily accomplished following a single intercross of two field strains. A few of the strains in the new Ugandan species produce low to trace levels of fumonisins, but most produce significant quantities of beauvericin. Beauvericin production is important because of its insecticidal properties. If the species is one that is merely present on sorghum and millet rather than a pathogen of sorghum and millet then it could possibly be used as a biocontrol agent through the displacement of more pathogenic or toxigenic fungi and as a means for insect control against larvae of various stalk borers.

Strengthening Research Capacity

Workshops held and number of attendees included in non-degree training report.

Networking Activities

Editorial and Committee Service (2008)

- Editor, Food Additives and Contaminants (2006-2009)
- International Society for Plant Pathology, *Fusarium* Committee (2000-2013)
- MycoRed Steering Committee (2007-2013)

Research Investigator Exchanges (2008)

- Hungary – September 2-6
- Italy – February 13-24; August 29 – September 2; September 26 – October 4
- Kenya – September 20-25
- Malaysia – June 14 – July 3
- Mozambique – October 25-31
- Norway – April 15-25
- South Africa – November 1-20
- South Korea – May 18-25
- Turkey – August 3-10

Other Collaborating Scientists (Host Country)

- Dr. Sofia Chulze, Department of Microbiology, National University of Rio Cuarto, Rio Cuarto, Argentina.
- Dr. Sandra Lamprecht, Plant Protection Institute, Agricultural Research Council, Stellenbosch, South Africa.
- Drs. Yin-Won Lee & Jungkwan Lee, Dept. of Plant Pathology, Seoul National University, Seoul, South Korea.
- Drs. Antonio Logrieco, Antonio Moretti & Giuseppe Mulé, Inst. Sci. of Food Production, CNR, Bari, Italy.
- Dr. Baharuddin Salleh, School of Biological Sciences, Universiti Sains Malaysia, Penang, Malaysia.
- Dr. Brett Summerell, Royal Botanic Gardens, Sydney, Australia.

Other Collaborating Scientists (U.S.)

- Drs. Charles W. Bacon and Tony Glenn, USDA Russell Research Center, Athens, Georgia
- Dr. Gary N. Odvody, Texas Agricultural Experiment Station, Corpus Christi, Texas

Recipients of Fusarium Cultures in 2008 (Other than Collaborators)

- Ridao Azucena, University of Buenos Aires, Buenos Aires, Argentina.
- Peter Cotty, USDA-ARS, University of Arizona, Tucson, Arizona.
- David Geiser, Pennsylvania State University, University Park, Pennsylvania.
- Fungal Genetics Stock Center, University of Missouri-Kansas City, Kansas City, Missouri.
- Carla Klittich, Dow Agrosiences, Indianapolis, Indiana.
- Ralf Kristensen, Institute of Veterinary Medicine, Oslo, Norway.
- Robert H. Proctor, Mycotoxin Research Unit, NCAUR, USDA-ARS, Peoria, Illinois.
- David Schmale, Dept. Plant Pathol. & Weed Science, Virginia Tech. Univ., Blacksburg, VA.
- Keith Seifert, Agriculture and Agri-Foods Canada, Ottawa, Ontario, Canada.
- Amir Sharon, Department of Plant Sciences, University of Tel Aviv, Tel Aviv, Israel.
- Bettina Tudzynski, Westfaelische Wilhelms University, Muenster, Germany.
- Cees Waalwijk, DLO Institute for Plant Protection, Wageningen, The Netherlands.

Publications and Presentations (2008)

Seminar, Workshop & Invited Meeting Presentations (International Locations Only)

Norwegian National Veterinary Institute, Oslo, Norway – April, 2008.
Faculty of Agricultural & Life Sciences, Seoul National Univ., Seoul, Korea – May, 2008.

Science University of Malaysia, Penang, Malaysia – June, 2008.
University of Malaysia Terengganu, Terengganu, Malaysia – July, 2008.
IUMS International Mycology Congress, Istanbul, Turkey – September, 2008.
Pan-African Environmental Mutagenesis Conf., Cape Town, So. Africa – November, 2008.
FABI, University of Pretoria, Pretoria, South Africa – November, 2008.

Books (2008)

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

Journal Articles (2008)

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.

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* 36 (suppl. B): 603-608.

Carter, L. L. A., J. F. Leslie & R. K. Webster. 2008. Population structure of *Fusarium fujikuroi* from rice and water grass in California. *Phytopathology* 98: 992-998.

Kabbage, M., J. F. Leslie, K. A. Zeller, S. H. Hulbert & W. W. Bockus. 2008. Genetic diversity of *Mycosphaerella graminicola*, the causal agent of Septoria leaf blotch, in Kansas winter wheat. *Journal of Agricultural Food and Environmental Sciences* vol. 2 <http://www.sci-entificjournals.org/journals2008/articles/1377.pdf>.

Lee, J., J. F. Leslie & R. L. Bowden. 2008. Expression and function of sex pheromones and receptors in the homothallic ascomycete *Gibberella zeae*. *Eukaryotic Cell* 7: 1211-1221.

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.

Leslie, J. F., and R. L. Bowden. 2008. *Fusarium graminearum*: When species concepts collide. *Cereal Research Communications* 36 (suppl. B): 609-615.

Book Chapters (2008)

Bandyopadhyay, R., R. A. Frederiksen & J. F. Leslie. 2008. Priorities for mycotoxin re-search 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.

Coulbaly, O., K. Hell, R. Bandyopadhyay, S. Hounkponou & J. F. Leslie. 2008. Economic impact of aflatoxin contamination in Sub-Saharan Africa. In: *Mycotoxins: Detection Methods, Management, Public Health and Agricultural Trade* (J. F. Leslie, R. Bandyopadhyay & A. Visconti, eds.), pp. 67-76. CABI, Kew, UK. 476 pp.

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 (2008)

Lee, J., J. F. Leslie & R. L. Bowden. 2008. Functions of the sex pheromones in *Gibberella zeae*. *Rivista di Patologia Vegetale* 90: S3.26.

Leslie, J. F., J. Lee, J. E. Jurgenson & R. L. Bowden. 2008. An update of the genetic map of *Gibberella zeae*. *Rivista di Patologia Vegetale* 90: S3.26.

Lima, C. S., S. S. Costa, M. A. Campos, J. F. Leslie & L. H. Pfening. 2008. Etiology of mango malformation and PCR detection of its causal agent in Brazil. *Rivista di Patologia Vegetale* 90: S3.65.

Minnaar-Ontong, A., L. Herselman, W. M. Kriel & J. F. Leslie. 2008. Population dynamics of *Fusarium Head Blight* in South Africa. *Rivista di Patologia Vegetale* 90: S3.66.

Ecologically-Based Management of Sorghum and Pearl Millet Insect Pests in Africa and the United States

Project WTAMU 101
Bonnie B. Pendleton
West Texas A&M University

Principal Investigator

Dr. Bonnie B. Pendleton, Assistant Professor of IPM, Entomology, Div of Agriculture, Box 60998, West Texas A&M University, Canyon, TX

Collaborating Scientists

Mr. Hamé Abdou Kadi Kadi – Entomologist, INRAN, B.P. 60, Kollo, Niger
Dr. Niamoye Yaro Diarisso – Entomologist/Scientific Coordinator, IER, B.P. 258, Bamako, Mali
Mr. Fernando Chitio – Entomologist/District Director, IIAM, Box 36, Nampula, Mozambique
Dr. D. C. Munthali – Entomologist, Private Bag 0027, Botswana College Agriculture, Gaborone
Dr. Gary C. Peterson – Sorghum Breeder, Texas AgriLife Research, Lubbock, TX 79401
Dr. Gerald J. Michels, Jr. – Entomologist, Texas AgriLife Research, Amarillo, TX 79106
Dr. Michael W. Pendleton – Electron Microscopist, Microscopy and Imaging Center, Texas A&M University, College Station, TX 77843-2257

Introduction and Justification

Entomologists, breeders, pathologists, economists, and extension agents in Mali, Niger, Mozambique, Botswana, and the US are educating students and farmers in IPM and developing, evaluating, and transferring pest management technologies for insects of sorghum and millet. Development and adoption of ecologically-based technologies will decrease loss by insects in the field and storage, reduce pesticide use, conserve soil and water without contamination by pesticides, and increase yield of food and feed for domestic use and income from marketing. Sorghum and millet are damaged by such biotic stresses as greenbug, *Schizaphis graminum*, and yellow sugarcane aphid, *Sipha flava*, in the US and sugarcane aphid, *Melanaphis sacchari*, in Africa that suck juice from leaves and vector viruses. Larvae of sorghum midge, *Stenodiplosis sorghicola*, feed on the ovary and can cause 100% loss of grain. Larvae of millet head miner, *Heliocheilus albipunctella*, tunnel in spikes. Southwestern corn borer, *Diatraea grandiosella*, in the US and maize stalk borer, *Busseola fusca*; and spotted stem borer, *Chilo partellus* in Africa tunnel in stalks, causing susceptibility to disease and lodging. Grain storage can take advantage of greater market price but result in more damage by insects that annually destroy 35% of grain worldwide. Pests of stored grain include the maize weevil, *Sitophilus zeamais*.

Objectives and Implementation Sites

This project is contributing to INTSORMIL objectives to facilitate markets by managing insects that damage yield and quality of sorghum and millet; improve food and nutritional quality to enhance marketability and consumer health by grain not contaminated by pests or pesticides; increase stability and yield through crop and natural resources management by IPM strategies not dependent on pesticides; develop and disseminate information on biotic stresses

to increase yield and quality by integrated management strategies against insects; enhance stability and yield through genetic technologies by determining differences among strains of insects and speeding development of resistant cultivars with yield and quality; and develop partnerships with agencies improving sorghum and millet and betterment of people through collaboration among scientists at West Texas A&M University, Texas AgriLife Research, and Texas A&M University in the US and Institut D'Economie Rurale in Mali, Institut National de la Recherche Agronomique du Niger, Instituto de Investigacao Agraria de Mocambique, Botswana College of Agriculture, private industries, volunteer organizations, and other agencies.

Specific objectives were to: 1) support entomology and IPM research and education of scientists in African countries; 2) collaborate with scientists in Africa and the US to develop and deliver IPM strategies against insects that damage sorghum and millet in the field and storage by improved understanding of biology, ecology, and population dynamics of insect pests and damage they cause; evaluation of potential arthropod pests; agronomic practices to prevent damage by insects and reduce pesticides; cultivars with greater yield and resistance to biotic and abiotic stresses; 3) provide education for students; and 4) develop partnerships with ICRISAT and PVOs engaged in improvement of sorghum and millet production and betterment of people. By presentations and publications, extension and other agencies will be assisted with transferring pest management information to farmers, scientists, and others in Africa and the US.

Research Methodology and Strategy

Evaluating potential pests and understanding the life histories of insect pests and natural enemies. Undergraduate Jody Gilcrest evaluated the effect of photoperiod on greenbug biotypes on sor-

ghum. M.S. student Zachary Eder determined the effect of yellow sugarcane aphids on biomass of sorghum. A M.S. student used pheromones to monitor seasonal abundance of southwestern corn borer moths in Texas. Using agronomic practices to manage pests. Intercropping grasses to draw stalk borers from sorghum or millet was evaluated by Dr. Yaro in Mali and Mr. Chitio in Mozambique. Developing germplasm resistant to biotic constraints. The PI and African entomologists collaborated with breeding projects in Mali, Mozambique, Niger, and Texas, and Milo Genetics and Monsanto for evaluating sorghum and millet for resistance to millet head miner, sorghum midge, greenbug, sugarcane aphid, stalk borers, and storage beetles. Studying pests of stored grain. Dr. Yaro and the PI prepared a color brochure and posters for hundreds of farmers to manage storage pests in Mali. A M.S. student determined what grain stage maize weevil infests sorghum in the field and evaluated resistance of stored sorghum grain. Electron microscopy and energy dispersive spectroscopy were used by Dr. Michael Pendleton to relate the depth of starch concentration in sorghum grain to resistance to maize weevil. Transferring insect pest management technologies. Mr. Chitio and Mr. Abdou Kadi Kadi assisted in transferring two sorghums in Mozambique and two sorghums and three millets to hundreds of farmers in Niger. Field demonstrations, workshops, brochures, posters, and training manuals were used or being prepared to teach farmers, extension, and others to recognize pest problems and evaluate, adapt, and implement IPM options. Undergraduate and graduate university students in the US, Botswana, and Niger assisted with research and were educated in entomology and IPM.

Research Results

Undergraduate Jody Gilchrest evaluated 14:10 and 10:14 light:dark hour photoperiods on biotype E and I greenbugs on susceptible ATx399 x RTx430 sorghum at constant daily dark and light temperatures of 10 and 23°C. Photoperiod but not biotype significantly affected greenbugs and should be considered when evaluating for resistance. Only 55 and 60% of biotype E and I greenbugs at 10:14 light:dark hours survived to produce offspring, while all produced nymphs at 14:10 light:dark hours. Biotype E and I greenbugs began producing nymphs in 14.0 days at 10:14

light:dark hours but only 9.5 and 9.6 days at 14:10. Fecundities of biotype E and I greenbugs were 2.3 and 2.7 times greater at 14:10 than 10:14 light:dark hours. Longevities of biotype E and I greenbugs were 1.3 and 1.4 times longer at 14:10 than 10:14 light:dark hours. (Table 1)

Master's student Zachary Eder evaluated the effect of yellow sugarcane aphids on biomass of sorghum ATx399 x RTx430. Plants at the third true-leaf stage were infested with 0, 10, 25, or 40 aphids per plant for 14 days. Plants infested with 40 aphids weighed less than 1/5 as much and were 1/2 as tall as check plants with no yellow sugarcane aphids. (Table 2)

A M.S. student from Colombia monitored southwestern corn borer moths in pheromone traps from May to October in Texas. Numbers of moths varied among locations and differed with weather. Moths of the 1st generation were trapped from late-June until mid-July, with a peak on 1 July. Second-generation moths were trapped from the 1st week of August through 1st week of September, with more than 50% captured before the middle of August.

Dr. Yaro assisted five farmers from each of Finkolo and Zandradowou villages in different agroecological zones with Sudan climate in the Sikasso region of Mali who evaluated *Andropogon gayanus* in three border rows 50 cm apart with 30 cm between plants to attract stalk borers away from 15 x 10-m plots of millet. *Andropogon* was transplanted on 7 and 8 July and millet planted on 18 and 17 July 2008 at Finkolo and Zandradowou. Ten millet plants were sampled from diagonals of the plot and *Andropogon* plants were sampled from each compass direction. Numbers of pests and natural enemies, percentages of deadhearts, and numbers of larvae and pupae of other borer species were counted on *Andropogon* and millet at the vegetative stage 30 days after emergence, tillering (70-80 DAE), and harvest (100-110 DAE). Damage was more at Finkolo than Zandradowou. Millet surrounded by millet (check) was more damaged (5.3) than by *Andropogon* (1.7), while 3.6% of *Andropogon* plants had deadhearts from borers. (Table 3)

Table 1.

Photoperiod (light:dark hours)	Greenbug biotype	Pre-reproductive period(days)	Reproductive period (days)	Total fecundity (nymphs)	Longevity (days)
10:14 (winter)	E	14.0 a A	15.7 a A	25.7 a A	37.9 a A
	I	14.0 a A	17.2 a A	23.5 a A	39.5 a A
14:10 (summer)	E	9.5 a B	23.5 a B	59.6 a B	50.7 a B
	I	9.6 a B	23.7 a A	63.5 a B	54.0 a B

Means followed by the same lower-case letter for a photoperiod or by the same upper-case letter for the same biotype in a column do not differ significantly.

Table 2.

Yellow sugarcane aphids per plant	Weight (g)	Height (cm)
0 (check)	0.93	65.1
10	0.32	46.5
25	0.17	34.0
40	0.18	32.6

Table 3.

Village	Farmer	% deadhearts of vegetative millet infested by stalk borers		
		Millet surrounded by millet	Millet surrounded by <i>Andropogon gayanus</i>	<i>Andropogon gayanus</i>
Finkolo	Diakalia BALLO	10.3	1.5	2.0
	Issouf BALLO	4.3	1.5	3.0
	Abdoulaye KONE	5.2	2.8	5.5
	Seybou KONE	6.6	2.3	7.1
	Oumar TRAORE	2.5	0.5	4.6
	Nouhoum DJOURTHE	0.0	3.7	1.2
	Siaka DJOURTHE	12.0	0.0	3.0
Zanradougou	Tidiani SANOGO	1.5	1.5	2.4
Mean		5.3	1.7	3.6

Table 4.

Village	% plants infested by stalk borers		
	Millet without <i>A. gayanus</i>	Millet with <i>A. gayanus</i>	<i>Andropogon gayanus</i>
Finkolo	9.1	7.8	4.4
Zanradougou	10.2	7.9	5.0

Dr. Yaro assisted farmers using *Andropogon gayanus* to attract stalk borers to 5 rows 20 m long and 10 m apart intercropped with local millet planted 6-9 July 2009 at Finkolo and Zanradougou. A randomized complete block with 10 farms was used at each village. Millet was intercropped with millet (check) or *Andropogon*. Natural enemies, and deadhearts and empty spikelets caused by stalk borers were sampled on 10 millet and 10 grass plants randomly selected on 5-8 September from diagonals of the plots. Millet was 19% less damaged surrounded by *Andropogon* than millet. (Table 4)

Mr. Chitio used napier grass as a border of three rows and Desmodium grass between rows of sorghum to trap stalk borers at Nampula Research Station in Mozambique. Rainfall stopped early, and the grass did not grow adequately to obtain results.

Mr. Abdou Kadi Kadi and millet breeder Mr. Issaka Ahmadou assessed at the Regional Agricultural Research Center in Kollo, Niger, damage by millet head miner and yield of HKB, H80-10GR, Taram, SOSAT-C, Mangarana, HKP-GMS, ICMV IS89305, Zati-b, Mangarana x ICMV IS89305, SOSAT-C x HKB, SOSAT-C x ZATB, and Tchoumo pearl millets developed at INRAN. The design was a completely randomized block with three replications. Each sub-plot was 12 m² with 4 rows 3 m long and 1 m between rows and hills. Five spikes of each genotype per replication were randomly selected and tagged. Five days later and until maturity, spikes were checked for larvae, pre-pupae, and pupae of millet head miner. At maturity, spikes were cut and damage assessed using a 1-9 scale where: 1 = <10, 2 = 11-20, 3 = 21-30, 4 = 31-40, 5 = 41-50, 6 = 51-60, 7 = 61-70, 8 = 71-80, and 9 = >81%. The percentage of infested spikes ranged from 24.2 to 62.4% for Tchoumo and SOSAT-C88 and was correlated with damage that ranged from 1.9 for Tchoumo to 4.3 for HK_GMS (20-50% mined spikes). The percentage of infested spikes was 38.4 and 40.9% on HKB and Taram (local varieties from Gaya zone in Dosso); damage was 1.9 and 2.1 (10-30% mined spikes). H80-10GR, SOSAT-C88, and HKP GMS millets were transferred to farms. (Table 5)

At Bazaga (Birni N’Konni), sorghum midge-resistant SSD-35 and its early maturing female parent Mota Maradi were less damaged (1.5 = 10-20% and 3.0 = 20-31% damaged spikelets) by sorghum midge and yielded more (862.5 and 737.5 kg ha⁻¹) than local El Mota (4.3 and 587.5 kg ha⁻¹). At the 1st planting at Doguérawa, damage by sorghum midge was 1.0 (<10% damaged spikelets) for SSD-35, 3.9 (31-40% damaged spikelets) for Mota Maradi, and 2.9 (21-30% damaged spikelets) for El Mota. At the 2nd planting, damage to SSD-35 was 1.3 (<10% damaged spikelets), 3.9 (31-40%) for Mota Maradi, and 2.0 (11-20% damaged spikelets) for El Mota. SSD-35 yielded 687.5 and 700.0 kg ha⁻¹ from the 1st and 2nd plantings. Yield of Mota Maradi did not differ between the 1st (937.5 kg ha⁻¹) and 2nd plantings (1,000.0 kg ha⁻¹). Yield of El Mota was greater in the 1st (743.8 kg ha⁻¹) than 2nd planting (562.5 kg ha⁻¹). (Table 6)

In 2009, SSD-35 and Mota Maradi were introduced on farms in seven villages of Birni N’Konni and Madaoua, Niger. Nine extension agents, 28 farmers, and “Taymako” organization of 80 farmers at Doguéraoua multiplied seed of SSD-35 on 3.5 hectares. SSD-35 was evaluated by 48 farmers and produced by 184 farmers on 67 hectares in two areas of Tahoua, Niger. In six villages, 104 and 67 farmers adopted SSD-35 and Mota Maradi planted on 24 and 16 hectares, respectively. Seed of SSD-35 was multiplied on 43 hectares by 84 farmers from five villages, and Mota Maradi was multiplied on 12 hectares by eight farmers from three villages. In 2008, the two varieties were introduced at farms in five villages of two regions by four extension agents, 20 farmers, and “Taymako” who did four tests with two planting dates at a site. Bagged grain of SSD-35 is sold by the private company “Semences Améliorées ALHERI” at Douchi.

Twenty-five sorghums from the Mozambique breeding program and 17 varieties from the U.S. differed significantly in damage (1-5 scale) by stalk borers at Namialo Agriculture Research Center. Twelve varieties differed significantly in resistance to sugarcane aphid. ICSB654 was resistant to stalk borers. Sima was

Table 5.

Millet evaluated at Kollo, Niger	Damage (1-9 scale)	% spikes infested by millet head miner
Tchoumo	1.9 ± 1.0 a	24.2 ± 1.6 a
Mangarana	2.9 ± 2.0 b	28.4 ± 2.2 b
HKB	1.9 ± 1.1 a	38.4 ± 2.1 c
Taram	2.1 ± 1.2 b	40.9 ± 1.5 c
ICMV IS89305	2.2 ± 1.2 b	44.2 ± 2.6 ab
Zatib	2.8 ± 1.9 b	44.9 ± 2.3 ab
Mangarana x ICMV IS89305	2.5 ± 1.4 b	52.8 ± 3.1 bc
SOSAT-C x HKB	2.7 ± 1.3 b	55.6 ± 2.9 bc
H80-10 GR	2.8 ± 1.4 b	57.3 ± 1.3 abc
HKP GMS	4.3 ± 2.7 c	58.1 ± 1.7 abc
SOSAT-C 88 x Zatib	3.1 ± 1.5 ab	59.7 ± 3.3 abc
SOSAT-C88	3.8 ± 2.2 ab	62.4 ± 3.8 cd
CV (%)	2.8	47.2
LSD	2.5	52.3

Table 6.

Sorghum	Damage by sorghum midge (1-9)			Yield (kg ha ⁻¹)		
	Bazaga	1st planting, Doguérawa,	2nd planting, Doguérawa,	Bazaga	1st planting, Doguérawa,	2nd planting, Doguérawa,
SSD-35	1.5 ± 0.3c	1.0 ± 0.1c	1.3 ± 0.3a	862.5 ± 13.6a	687.5 ± 9.7a	700.0 ± 12.3b
Mota Maradi	3.0 ± 0.4b	3.9 ± 0.4c	3.6 ± 0.3b	737.5 ± 14.7b	937.5 ± 17.3c	1,000.0 ± 7.9c
El Mota (check)	4.3 ± 0.7a	2.9 ± 0.7b	2.0 ± 0.6c	587.5 ± 10.9c	743.8 ± 12.4b	562.5 ± 10.5a
C.V (%)	35.5	43.2	36.8	48.6	39.7	45.4
L.S.D	1.5	1.9	0.3	139.0	193.7	137.5

Table 7.

25 sorghums	Stalk borer damage		Stalk borer damage		Sugarcane aphid damage
	17 sorghums	12 sorghums	12 sorghums	12 sorghums	
104GRD	1.25c	04CS-452-4-1	1.083b	SDS-3047/722E-8	1.01c
ICSB654	1.25c	04CS-573-3-1	1.3ab	Sima	1.15bc
Ent#64DTN	1.27bc	02CS-30932	1.31ab	GV SIMS710E-2	1.16bc
SPV1411	1.30abc	02CS-30445	1.37ab	SDS-1958-1-3-2/724E-5	1.16bc
ICSB324	1.63abc	03CM-15012-BK	1.62a	(SDS-5006*USV-187)E-4	1.30ab
E36-1	1.68ab	04CS-798-7-1	1.62a	ZSV-15-4/723E-3	1.30ab
ICSV700	1.73ab	03CM-1104-BK	1.65a	Macia	1.37ab
ICSR93034	1.75a			ICSV-93010-1/708E-9	1.45a
CV%=17.56		CV%=11.53		CV%=10.91	

little damaged by stalk borers, aphids, and sorghum midge. (Table 7)

The PI evaluated 54 sorghum hybrids developed by Monsanto for resistance to greenbug biotypes E and I, with most more resistant than the check. The PI evaluated 441 sorghum lines Milo Genetics developed for resistance to greenbug biotype I, with most more resistant than the check.

M.S. student Suhas Vyavhare put three female and two male newly emerged maize weevils into each of 10 vials with 5.0 g of kernels from sorghum plants at different growth stages in a field. Each grain in the 10 vials was evaluated for damage on a scale of 1-5, numbers of live and dead weevil adults were counted, grain in each vial was weighed, and moisture was determined. After

1 week, 23.1% of kernels at the hard-dough stage but only 6.1 and 12.0% of kernels at the soft-dough and physiological maturity were damaged. (Table 8)

Suhas Vyavhare evaluated 26 genotypes of stored sorghum for resistance to maize weevils. Five newly emerged weevils were put with 5.0 g of grain in each of 10 vials. Each grain in the 10 vials of one kind of sorghum was evaluated for damage on a scale of 1-5, numbers of live and dead weevils were counted, and grain in each vial was weighed once every 3 weeks. Eighteen new weevils were produced per gram of B.HF8 by 63 days after infestation. At 84 days after infestation, weight loss ranged from 55.6% for B.HF8 to 12.7%. (Table 9)

Dr. Michael Pendleton developed a technique to relate the lo-

Table 8.

Sorghum stage	Grain moisture (%)	% damaged kernels 1 week after infestation
Soft dough	50	6.1
	34	15.4
Hard dough	32	23.1
	25	16.7
Physiological maturity	16	12.0

Table 9.

Sorghum Genotype	Cumulative new maize weevils/g of grain by 63 DAI	% weight loss at 84 DAI
(5BRON151/(7EO366*GR107B-90M16)*Tegemeo)-HG7-CC2-CABK-CABK	3.1	12.7
(SV1*Sima/TS23250)-LG15-CG1-BG2-(03) BGBK-LBK-PRBK	2.9	13.8
Sureño	4.2	14.5
(B35*B9501)-HD9	5.9	18.9
(Dorado*Tegemeo)-HW15-CA1-CC2-LG1-CABK	5.0	20.4
(Dorado*Tegemeo)-HW13-CA1-CC2-LGBK-CABK	4.8	21.3
(Macia*TAM428)-LL9	4.4	22.3
Tegemeo	4.6	22.8
(Kuyuma*5BRON155)-CA5-CC1-CABK-CABK	5.3	23.4
(VG153*(TAM428*SBIII)-23-BE2-EE2-BE1	7.5	25.1
(9MLT176/(MR112B-92M2*TX2880)*A964)-CA3-CABK-CCBK-CABK-CABK	7.4	26.5
Macia	6.8	27.1
(Dorado*Tegemeo)-HW14-CA1-CC2-CABK-CABK	6.2	27.4
(Macia*TAM428)-LL2	6.1	27.7
(5BRON151/(7EO366*GR107B-90M16)*Tegemeo) -HG1-LGBK-CABK-CABK	7.0	30.2
(M84-7*VG153)-LBK-PR7-L4-L2-	8.4	30.8
(6BRON161/(7EO366*Tx2783)*CE151) -LG5-CG2- (03)-BG1-BG2-LBK-PR	6.0	31.4
(850G4300-5*Tx2782)-SM5-CM2-SM2-SM1-CABK-CMBK-CMBK	11.0	32.4
CE151	9.9	35.8
B409	11.5	36.5
(Segaolane*WM#322) LG2-LG2-(03)-BG1-LG1-LGK-PRBK	7.8	39.9
(*Tx2864*PI550607))))-PR3-SM6	12.0	36.9
(9MLT176/(MR112B-92M2*TX2880)*A964)-LG8-CABK-LGBK-LGBK-CABK	11.9	40.8
(Tx2864*PI550607))))-PR3-SM6	14.2	44.5
(A964*P850029)-HW6-CA1-CC1-LGBK-CABK	14.6	51.9
B.HF8	17.7	55.6

cation of starch in sorghum grain to resistance to maize weevil. Cross sections of grains were subjected to iodine vapor that produced a stable complex with starch. The iodine molecules were detected using a scanning electron microscope equipped with an energy dispersive spectroscopy (EDS) detector. Information from the EDS detector was related to the image of the sorghum grain so areas with concentrations of starch/iodine complex were determined. The greater the depth of the concentration of starch granules from the surface, the more resistant was the grain to maize

weevil. Starch was concentrated 60 micrometers from the surface of the grain of the most resistant genotype but only 20 micrometers from the surface of less resistant sorghum.

Ten and 9 hectares of Macia and Sima were planted at IIAM research stations at Namapa, Namialo, Nampula, Manetil, Mapupulo, Mutuali, and Sussundenga in Mozambique. A total of 5.5 hectares each of Macia and Sima was planted on farms at Liomagarue, Nametil, Malema, and Murrupula. Totals of 185,500 and

153,000 kg of Macia and Sima were produced at the seven research stations, and 26,125 and 36,300 kg were produced at the four farms to distribute to farmers or sell to NGOs to give to farmers.

To enhance germplasm, 12 local sorghum varieties were collected in Montepuez, Namuno, Ancuabe, Balama, Chiure, Ribaue, and Malema districts in Nampula and Cabo Delgado provinces in Mozambique. The sorghums resist storage pests because the grain is flint but matures late in 6 months. Farmers wanted varieties not damaged by birds or pests in the field or storage and were willing to use intermediate sorghum to overcome periods without cereal.

Networking Activities

Workshops and Meetings. The PI and students attended and presented research at the Sorghum Improvement Conference of North America and Great Plains Sorghum Conference, Amarillo, TX, 10-12 August 2009; 6th International Integrated Pest Management Symposium, Portland, OR, 24-26 March 2009; 57th Meeting of the Southwestern Branch of the Entomological Society of America, Stillwater, OK, 23-26 February 2009; and the 56th Annual Meeting of the Entomological Society of America, Reno, NV, 16-19 November 2008.

Research Investigator Exchanges

From 16-27 October 2008, the PI discussed and reviewed research with scientists from INRAN in Niger and IER in Mali. From 5-16 March 2009, the PI traveled with Drs. John Sanders, Ouendeba Botorou, Jeremy Foltz, and Niamoye Yaro to Finkoloni, Kaniko, Magnabougou, N'Garasso, Piza, Tingoni, and Wallo in Mali to meet with 199 farmers including 71 women, give them a color brochure written in French, and tell them how to manage insect pests in stored grain. From 16-28 April 2009, the PI discussed and reviewed research and education with scientists and students from IIAM in Mozambique and Botswana College of Agriculture. The PI met in Mali with the coordinators of the West Africa regional program to discuss and prepare workplans and budgets from 26-30 August 2009.

Research Information Exchange

The PI advised extension, National Sorghum Producers, and seed companies on management of sorghum insects. Four hundred ninety-five sorghums developed for resistance to biotype E and I greenbugs were evaluated for Milo Genetics and Monsanto. Supplies and funding were provided to Mr. Chitio in Mozambique, Dr. Yaro in Mali, Mr. Abdou Kadi Kadi in Niger, and Dr. Munthali in Botswana. The PI, Dr. Yaro in Mali, Mr. Abdou Kadi Kadi in Niger, and Dr. Alain Ratnadass, Entomologist, CIRAD/ICRISAT Niger, discussed collaborative entomology research for the "Cereals for the Drylands" proposal to the Bill and Melinda Gates Foundation. The PI and Dr. Yaro prepared a color brochure entitled "Les Insectes Nuisibles du Sorgho Stocke et La Gestion Integree des Insectes Nuisibles des Stocks" and traveled in March with Drs. John Sanders, Ouendeba Botorou, and Jeremy Foltz to tell hundreds of farmers how to manage storage pests. During release of SSD-35, Mr. Abdou Kadi Kadi worked with eight extension agents, 28 farmers, "Taymako" association of 80 farmers from Doguéraoua, and a student intern from Faculté d'Agronomie,

Université Abdou Moumouni de Niamey, Niger. Forty farmers in Niger were informed about sorghum midge through field training on identification, biology, damage assessment, and control. Eight extension agents were informed how SSD-35 was developed and trained how to identify and control insect pests of millet and sorghum in Niger. While collaborating with Dr. Kadri Aboubacar, Faculté d'Agronomie, Université Abdou Moumouni de Niamey, Mr. Abdou Kadi Kadi was involved with field practical training, supervising writing of reports, and the committees of five student interns in 2008. In 2009, he supervised two interns involved with testing and adoption of SSD-35 at Doguéraoua and surveying farmer knowledge of sorghum insect pests at Madaoua, Niger.

Germplasm Conservation and Distribution

H80-10GR, SOSAT-C88, and HKP GMS millets were transferred to farms in Niger. Sorghum midge-resistant SSD-35 and Mota Maradi were introduced on farms in seven villages of Birni N'Konni and Madaoua. Nine extension agents, 28 farmers, and "Taymako" organization of 80 farmers at Doguéraoua multiplied SSD-35 on 3.5 hectares. SSD-35 was evaluated by 48 farmers and produced by 184 farmers on 67 hectares in two areas of Tahoua. In six villages, 104 and 67 farmers adopted SSD-35 and Mota Maradi planted on 24 and 16 hectares, respectively. Seed of SSD-35 was multiplied on 43 hectares by 84 farmers from five villages, and Mota Maradi was multiplied on 12 hectares by eight farmers from three villages. In 2008, the two sorghum varieties were introduced at farms in five villages. Bagged grain of SSD-35 is sold by the private seed company "Semences Améliorées AL-HERI" at Doutchi.

Ten and 9 hectares of Macia and Sima were planted at seven IIAM research stations and 5.5 hectares each on four farms in Mozambique. Totals of 211,625 and 189,300 kg of Macia and Sima were produced to distribute to farmers or sell to NGOs to give to farmers. To enhance existing germplasm 12 local sorghum varieties were collected in Montepuez, Namuno, Ancuabe, Balama, Chiure, Ribaue, and Malema districts in Nampula and Cabo Delgado provinces.

Publications and Presentations

Journal Articles

- Ayyanath, M.M., B.B. Pendleton, G.J. Michels, Jr., and R.A. Bowling. 2008. Effect of greenbug (Hemiptera: Aphididae) from resistant sorghum on developmental rates of convergent lady beetle (Coleoptera: Coccinellidae). *Southwestern Entomologist* 33: 191-197.
- Pendleton, B.B., M.W. Pendleton, and E.A. Ellis. 2008. Using energy dispersive spectrometry to compare the efficiency of metal coating techniques for scanning electron microscopy of insects. *Texas Journal of Microscopy* 39: 14.
- Pendleton, M.W., E.A. Ellis, B.B. Pendleton, and A. Holzenburg. 2008. Two methods of conductive coating for scanning electron microscopy of maize weevils *Sitophilus zeamais* compared by energy dispersive spectroscopy. *Microscopy and Microanalysis* 14 (Supplement 2): 704CD. Cambridge University Press, New York, NY.
- Damte, T., B.B. Pendleton, and L.K. Almas. 2009. Cost-ben-

efit analysis of sorghum midge *Stenodiplosis sorghicola* (Coquillett)-resistant sorghum hybrid research and development in Texas. *Southwestern Entomologist* 34: 390-400.

Pendleton, B.B., and S. Veerabomma. 2008. Effects of soil water and nitrogen on fitness of greenbug (Hemiptera: Aphididae) on sorghum. *Southwestern Entomologist* 33: 281-287.

Pendleton, B.B., A.L. Palousek Copeland, and G.J. Michels, Jr. 2009. Effect of biotype and temperature on fitness of greenbug (Hemiptera: Aphididae) on sorghum. *Journal of Economic Entomology* 102: 1624-1627.

Peterson, G.C., K. Schaefer, and B.B. Pendleton. 2009. Registration of 16 sorghum germplasm lines. *Journal of Plant Registrations* 3: 203-205.

Proceedings

Garzon, C.A., B.B. Pendleton, J. Michels, and R. Fegley. 2009. Monitoring to establish a model of southwestern corn borer (Lepidoptera: Crambidae) dynamics for the Texas High Plains. In *Proceedings of the 57th Annual Meeting of the Southwestern Branch of the Entomological Society of America and the Annual Meeting of the Society of Southwestern Entomologists*, 23-26 February 2009, Stillwater, OK. P. 1.

Miscellaneous

Yaro Diarisso, N., and B.B. Pendleton. 2009. Les insectes nuisibles du sorgho stocke. La gestion integree des insectes nuisibles des stocks.

Presentations

Eder, Z., and B.B. Pendleton, Yellow sugarcane aphid effects on sorghum yield; Gilchrest, J.R., and B.B. Pendleton, Effect of photoperiod on fitness of greenbug biotypes E and I on sorghum; and Pendleton, M., E.A. Ellis, F.M. Chitio, and B.B. Pendleton, Using scanning electron microscopy with energy dispersive spectroscopy to identify starch in sorghum grain resistant to maize weevil. Sorghum Improvement Conference of North America and Great Plains Sorghum Conference, 10-12 August 2009, Amarillo, TX.

Pendleton, B.B. IPM for insect pests of sorghum and millet. *Global Food Shortages: Role of IPM*. 6th International Integrated Pest Management Symposium, 24-26 March 2009, Portland, OR.

Garzon, C.A., B.B. Pendleton, J. Michels, and R. Fegley. Monitoring to establish a model of southwestern corn borer (Lepidoptera: Crambidae) dynamics for the Texas High Plains. 57th Annual Meeting of the Southwestern Branch of the Entomological Society of America, 23-26 February 2009, Stillwater, OK.

Bowling, R., B.B. Pendleton, and G.J. Michels, IPM Program insect monitoring program in the northwest Texas Panhandle; Garzon, C.A., B.B. Pendleton, R. Bowling, and G.J. Michels, Monitoring to establish a model of southwestern corn borer (Lepidoptera: Crambidae) dynamics for the Texas High Plains; and Pendleton, M.W., B.B. Pendleton, E.A. Ellis, and A. Holzenberg, Two methods of conductive coating for scanning electron microscopy of maize weevils, *Sitophilus zeamais*, compared by energy dispersive spectroscopy. 56th Annual Meeting of the Entomological Society of America, 16-19 November 2008, Reno, NV.

