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# 2006 Progress Report

GMPRC Annual Report

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Grain Marketing and Production Research Center  
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# WELCOME TO GMPRC

Since its establishment in 1970, the Agricultural Research Service's Grain Marketing and Production Research Center (GMPRC) has been the U.S. Department of Agriculture's main location for conducting research on measuring and controlling the quality of cereal grains throughout the grain industry.

Our MISSION is to **“Conduct innovative research and develop new technologies to solve problems relating to natural resources conservation and the production, harvesting, storage, marketing, and utilization of grain to ensure a safe, abundant, and high quality grain supply.”**

Located in Manhattan, Kansas, GMPRC is situated in the heart of the Great Plains, which includes thirteen states that produce more than 2/3 of all U.S. wheat, corn, sorghum, and soybeans. Operating from a 60,000 square foot facility and the nation's only 50,000 bushel (700 metric ton) capacity research grain elevator, the Center is composed of five research units:

- BIOLOGICAL
- ENGINEERING
- GRAIN QUALITY AND STRUCTURE
- PLANT SCIENCE AND ENTOMOLOGY
- WIND EROSION

Our VISION is to be **“The Customer's Choice for solving problems in natural resources conservation and the production, harvesting, storage, marketing, and utilization of grain”** and we welcome the opportunity to serve all segments of the food industry from producers to consumers.

Dr. James E. Throne, Research Leader  
Biological Research Unit

Dr. Floyd E. Dowell, Research Leader  
Engineering Research Unit

Dr. Scott Bean, Acting Research Leader  
Grain Quality & Structure Research Unit

Dr. Robert L. Bowden, Research Leader  
Plant Science & Entomology Research Unit

Dr. Edward L. Skidmore, Research Leader  
Wind Erosion Research Unit

Dr. Thomas G. Shanower  
Center Director

# **INTERACTIONS WITH KANSAS STATE UNIVERSITY**

Kansas State University (KSU) has an exceptionally strong program in agricultural research and education. GMPRC maintains a close working relationship with KSU and the Kansas Agricultural Experiment Station and Cooperative Extension Service. One of the GMPRC research units, Plant Science and Entomology, is housed on the KSU campus. GMPRC scientists enjoy a close working relationship with scientists at KSU. A majority of the GMPRC scientists have adjunct faculty positions and, as a result, approximately 40 undergraduate and graduate students conduct research at GMPRC each year.

# Progress Report Designed for Customers

This Progress Report is designed to provide specific information about each of the active research projects at GMPRC in terms that are easily understood. The report is being provided as a pdf document on our website. Please feel free to print or download the document.

Each major research project in the Agricultural Research Service, including those at GMPRC, is identified by a number from the Current Research Information System (CRIS). In addition, we frequently develop Specific Cooperative Agreements (SCAs) or Cooperative Research and Development Agreements (CRADAs) with other groups such as universities, other federal agencies, or private companies. All of the results from these various agreements are incorporated into the report for each of the specific CRIS projects that they are associated with.

The information in this Progress Report is organized by Research Unit and a complete Table of Contents is provided on the following pages. For each project described, we have provided a statement of the problem that we are trying to solve, the goals and objectives for this particular research activity, the results obtained during 2006, and our future goals for this research activity for the next three years. We also have included a list of publications along with a contact person if the reader has additional questions or needs more detailed information on a project.

We certainly appreciate your comments and suggestions concerning ways that we can improve this report. We encourage you to continue to send your comments via mail, telephone, FAX, or email to:

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# **BIOLOGICAL RESEARCH UNIT**

The mission of the Biological Research Unit is to develop new and improved methods, approaches, and strategies for the management of insect pests that attack grain and stored products. In order to decrease the levels of chemical pesticides used to protect our food supply, safer more effective alternatives must be developed. Specific research projects for this Unit include:

- |                           |                                                                                                        |
|---------------------------|--------------------------------------------------------------------------------------------------------|
| CRIS - 5430-43000-025-00D | Integrated Management of Insect Pests in Stored Grain and in Processed Grain Products                  |
| CRIS - 5430-43000-026-00D | Genomics and Proteomics of Stored-Product Insects for Development of New Biopesticides                 |
| CRIS - 5430-43000-027-00D | Ecology, Sampling, and Modeling of Insect Pests of Stored Grain, Processing Facilities, and Warehouses |

**CRIS 5430-43000-025-00D**

## **Integrated Management of Insect Pests in Stored Grain and in Processed Grain Products**

**Project Leader:** F. Arthur

**Investigators:** F. Arthur and J. Lord

**Full-Time Scientist Equivalents (SYs):** 2.0

**Start Date:** 04/06/05

**Termination Date:** 04/05/10

**Problem:** Stored-product insect pests reduce the quality of stored grain and grain-related products in the U.S. and in the world. Over 12 billion bushels of corn and wheat are grown in the U.S. each year, with a value of over 25 billion dollars. In addition, over a billion bushels of barley, oats, rice, rye, and sorghum are grown in the U.S. each year, with a value of over 3 billion dollars. It is estimated that postharvest losses to these grains due to insects are 5 to 10%, or about 1.4 to 2.8 billion dollars. Losses to processed commodities, which are difficult to quantify, may greatly exceed dollar losses to raw commodities because of their greater economic value.

There is a need to improve integrated pest management (IPM) of stored-product insects by identifying new control agents that are safer and more environmentally friendly, and the factors that affect susceptibility to these agents; developing effective control strategies using combinations of these agents; testing the application and integration of these control strategies in actual industrial environments; and determining the vulnerabilities of economically important insect pests to various control agents. Insect pathogens are potential additions to integrated pest management programs for stored products, but they may not give effective control by themselves; however, control could be enhanced by using them in combinations or along with disruption

of insect immune systems.

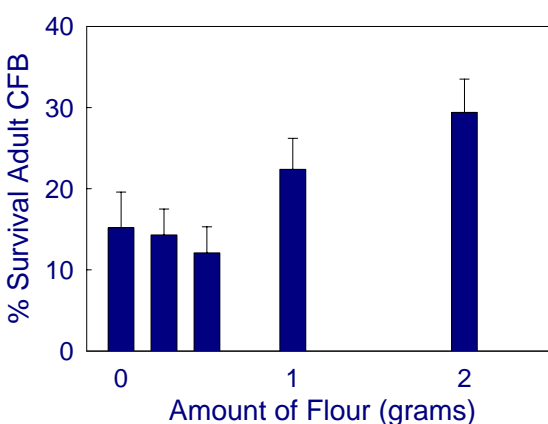
The loss of methyl bromide used as a fumigant to treat an entire facility presents us with a unique opportunity to develop smaller-scale insect control strategies that specifically target locations within a food production or storage facility that are vulnerable to insect pest infestation. Our goal is to improve IPM through selective application of safer control agents and non-chemical control methods and to identify the vulnerabilities in insect immune systems that can be exploited to make these control agents more effective. This will benefit food manufacturers and handlers by decreasing the levels of chemicals currently needed to protect our food supply from insect pests and it will lead to a wholesome, more dependable food supply for consumers.

**Objectives:** This research has three major objectives: 1) Identify and refine safe, environmentally friendly control methods and physical controls to manage stored-product insect pests; 2) Evaluate selective targeted controls and application strategies to manage insect pests in actual industrial environments; and 3) Identify vulnerabilities in insect physiological and biochemical stress responses that could be exploited to improve the effectiveness of these control methods.

## Results and Impact:

### 1. Food Material Negatively Affects Control from Aerosol Insecticides.

Aerosol insecticides are used to control insects in food processing facilities, but we need more information to optimize the effectiveness of these aerosols. GMPRC scientists conducted a field test whereby adult confused flour beetles were exposed with 0 to 2,000 mg of flour to a pyrethrin aerosol inside an open warehouse.



Survival of beetles increased with the presence of flour, indicating that the food material compromised effectiveness of the aerosol. There were also problems with uneven distribution of the aerosol inside the test warehouse. Although aerosols show potential for replacing whole-plant fumigations, if proper sanitation is not practiced the aerosol will be less effective for insect control, particularly with adult beetles. Also, the aerosol systems must deliver the product throughout the entire facility that is being treated.

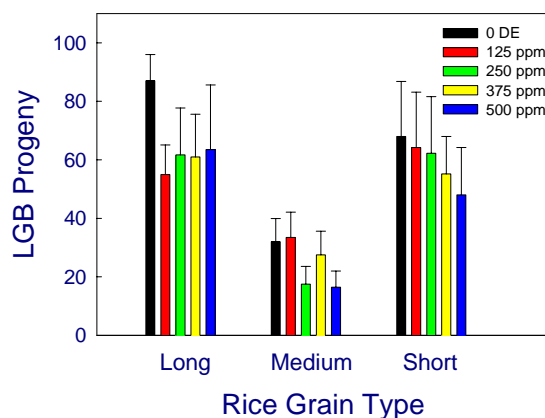
### 2. Diatomaceous Earth Is Not Equally Effective On All Grains.

Diatomaceous earth (DE) is used for insect control in stored grain, but more information is needed to optimize efficacy. GMPRC scientists treated rough rice with the labeled application rates of two

commercial DE products and exposed adult lesser grain borers, a major pest of stored grains, for different time periods and temperatures. Adult mortality increased with increasing time of exposure up to a maximum of 60 to 70% for both commercial formulations, and there was extensive progeny production in the treated rice. Results show that although these two commercial DE formulations had previously given good control of the lesser grain borer on stored wheat, they were not as effective on rough rice. Therefore, different application rates of DE may be necessary to give complete control of the lesser grain borer on different grain types.

### 3. The Insect Growth Regulator Methoprene Eliminates Progeny Production of the Lesser Grain Borer.

GMPRC scientists combined the natural inert dust diatomaceous earth (DE) with the insect growth regulator (IGR) methoprene and applied the combination at different rates to long-grain, medium-grain, and short-grain rough rice.

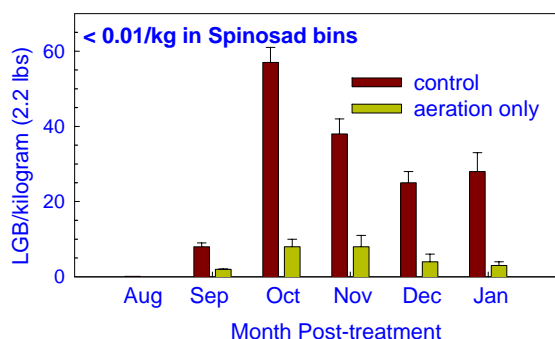


Mortality of adults was only 60% at the label rate of 500 ppm DE, and there were differences among the rice types. However, when methoprene was included in the insecticide treatment, no offspring were produced. Results show that methoprene by

itself will eliminate progeny production of the lesser grain borer, but the addition of DE will give some control of adults and could limit damage from adult feeding.

#### 4. Biological Pesticide Spinosad Controls the Lesser Grain Borer in Stored Wheat.

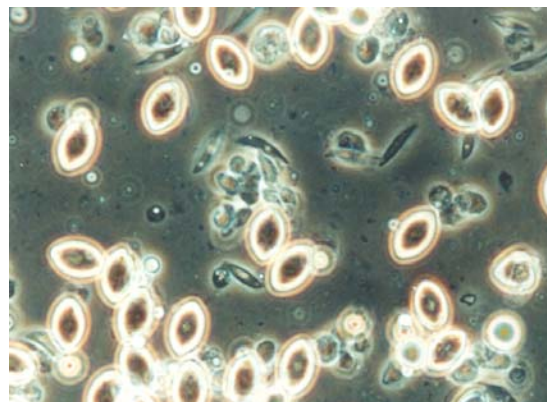
New insecticides are needed to control insects in stored grain. GMPRC scientists tested the newly approved and environmentally safe insecticide spinosad on Kansas farms and in laboratory studies to examine efficacy and persistence under typical field conditions.



In the field tests, no live lesser grain borers were found in any grain treated with spinosad, the residues killed adults during a 6-9 month storage period, there were few progeny, and residues did not degrade over time. Data were used to support registration, which was granted by the US-EPA.

#### 5. Test Assay Developed to Detect Insect Pathogens in Stored-Product Insect Pests.

Naturally occurring pathogens of insects can potentially be exploited for insect control, but a simpler way to determine their presence in insects is required. GMPRC scientists developed a test assay to detect *Mattesia oryzaephili*, a pathogen of grain beetles through tests with laboratory colonies from different field sources.



This assay will enable researchers to assess the impact of *M. oryzaephili* on pest populations and to determine the potential to exploit the pathogen by conservation or introduction where it does not occur, and will also allow scientists to maintain health and vigor of laboratory colonies by monitoring these cultures for insect pathogens.

#### Goals for 2007, 2008, and 2009:

##### Specific tasks in 2007 will be to:

1. Continue evaluations for new applications or uses of the IGRs hydroprene and methoprene on different surface substrates.
2. Analyze data from completed studies on new applications or uses of the IGRs hydroprene and methoprene on different surface substrates.
3. Correlate stress response in insects with susceptibility to pathogens.
4. Evaluate combinations of IGRs and DEs as surface treatments for control of insects in stored grain.
5. Continue with heat treatment studies of rice kernels for control of insects (test different species of insects).

6. Conduct second year of study on summer aeration of stored wheat for control of insects.
7. Analyze data for lab and field studies with targeted treatments for control of insects in grain bins.
8. Analyze data and write manuscript from study with surface compared to whole-bin treatments of insecticides for control of insects in stored wheat.
9. Continue study to collect insect population and *Mattesia* prevalence data in stored grain.
10. Continue study with *Mattesia* and parasitic wasps for control of insects in stored wheat.
11. Complete studies to determine interaction between DE dusts and lipids in different species of insects, and analyze data from initial studies.
12. Continue micro-array studies for genetic tests with insect immune responses to stress.
13. Quantify genetic responses of insects to pathogens, heat treatments, and other physical controls.

**Specific tasks in 2008 will be to:**

1. Continue evaluations for new applications or uses of IGRs on different surface substrates.
2. Write manuscripts on completed studies on new applications or uses of the IGRs hydroprene and methoprene on different surface substrates.
3. Analyze data and write manuscripts on combination studies of IGRs and DEs as surface treatments for control of insects in

stored grain.

4. Analyze data for heat treatment studies of rice kernels for control of insects (test different species of insects).
5. Conduct third year of study on summer aeration of stored wheat for control of insects.
6. Complete analyses and write manuscripts for lab and field studies with targeted treatments for control of insects in grain bins.
7. Model effectiveness of *Mattesia* to suppress specific stored-product insect species.
8. Complete data analysis and write manuscripts on studies to determine interaction between DE dusts and lipids in different species of insects, and analyze data from initial studies.
9. Develop experimental protocols to measure insect immune responses.
10. Document susceptibility of insects with silenced immune response genes to pathogens.
11. Continue to quantify genetic responses of insects to pathogens, heat treatments, and other physical controls.

**Specific tasks in 2009 will be to:**

1. Continue evaluations for new applications or uses of IGRs on different surface substrates.
2. Complete analysis of data and write manuscripts for heat treatment studies of rice kernels for control of insects.
3. Analyze data for study on summer aeration of stored wheat for control of insects.

4. Write manuscripts on studies with *Mattesia* and parasitic wasps for control of insects in stored wheat.

5. Complete data analysis and write manuscripts on studies to determine interaction between DE dusts and lipids in different species of insects, and analyze data from initial studies.

6. Determine gene regulation by insects in response to stress induced by control agents.

7. Begin analysis of results on susceptibility of insects with silenced immune response genes to pathogens.

8. Confirm gene systems involved in regulation of production of heat shock proteins by insects in response to physical controls and begin analysis of results.

#### **Specific Cooperative Agreements for This Project Included:**

None

#### **Summary of 2006 Publications/Patents:**

1. Arthur, F.H., Siebenmorgen, T.J. 2005. Historical weather data and predicted aeration cooling periods for stored rice in Arkansas. *Applied Engineering in Agriculture* 21: 1017-1020.

2. Arthur, F.H., Casada, M. 2005. Feasibility of summer aeration to control insects in stored wheat. *Applied Engineering in Agriculture* 21: 1027-1038.

3. Arthur, F.H. 2005. Initial and delayed mortality of late-instar larvae, pupae, and adults of *Tribolium castaneum* and *Tribolium confusum* (Coleoptera: Tenebrionidae) exposed at variable temperatures and time

intervals. *Journal of Stored Products Research* 42: 1-7.

4. Arthur, F.H., Hagstrum, D.W., Flinn, P.W., Reed, C.R., Phillips, T.W. 2005. Insect populations in grain residues associated with commercial Kansas grain elevators. *Journal of Stored Products Research* 42: 226-239.

5. Lord, J.C. 2006. Interaction of *Mattesia oryzaephili* with *Cephalonomia* spp. (Hymenoptera: Bethyridae) and their hosts *Cryptolestes ferrugineus* (Coleoptera: Laemophloeidae) and *Oryzaephilus surinamensis* (Coleoptera: Silvanidae). *Biological Control* 37: 167-172.

6. Mohandass, S.M., Arthur, F.H., Zhu, K.Y., Throne, J.E. 2006. Hydroprene: mode of action, current status in stored-product pest management, insect resistance, and future prospects. *Crop Protection* 25: 902-909.

7. Mohandass, S., Arthur, F.H., Zhu, K., Throne, J.E. 2006. Hydroprene prolongs development time and increases mortality of eggs of Indianmeal moth (Lepidoptera: Pyralidae). *Journal of Economic Entomology* 99: 1007-1016.

8. Vardeman, E.A., Arthur, F.H., Nechols, J.R., Campbell, J.F. 2006. Effect of temperature, exposure internal and depth of diatomaceous earth on distribution, mortality, and reproduction of the lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) in stored wheat. *Journal of Economic Entomology* 99: 1017-1024.

9. Toews, M.D., Pearson, T.C., Campbell, J.F. 2006. Imaging and automated detection of *Sitophilus oryzae* L. (Coleoptera: Curculionidae) pupae in hard red winter wheat. *Journal of Economic Entomology* 99: 583-592.



10. Arthur, F.H. 2005. Impact of aeration on insect pest management in stored rice [abstract]. National Entomological Society of America Annual Meeting, Ft. Lauderdale, FL, November 6-9, 2005.

11. Arthur, F.H. 2006. Insect growth regulators (IGRs) in pest management programs [abstract]. 5th National IPM Conference, St. Louis, MO, April 3-6, 2006.

12. Lord, J.C. 2005. Perspectives for biological control of stored-product pests using entomopathogens, alone and in combination with beneficial insects [abstract]. National Entomological Society of America Annual Meeting, Ft. Lauderdale, FL, November 6-9, 2005.

13. Campbell, J.F., Toews, M.D. 2005. Monitoring Indianmeal moth inside and outside. AIB Quarterly, Quality Assurance and Food Safety, Fall, 2005.

**For More Information on This Project  
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**CRIS 5430-43000-026-00D**

## **Genomics and Proteomics of Stored-Product Insects for Development of New Biopesticides**

**Project Leader:** R. Beeman

**Investigators:** R. Beeman and B. Oppert

**Full-Time Scientist Equivalents (SYs):** 2.0

**Start Date:** 04/06/05

**Termination Date:** 04/05/10

**Problem:** Insects reduce the quality of stored grain and other stored products in the U.S. and in the world. Over 12 billion bushels of corn and wheat are grown in the U.S. each year, with a value of over 25 billion dollars. In addition, over a billion bushels of barley, oats, rice, rye, and sorghum are grown in the U.S. each year, with a value of over 3 billion dollars. It is estimated that postharvest losses to these grains due to insects are 5 to 10%, or about 1.4 to 2.8 billion dollars. Losses to processed commodities are difficult to quantify, but probably greatly exceed the losses to raw commodities. Traditional chemical controls used by the cereal foods industry are being lost due to insect resistance, reduced public acceptance, and changes in regulatory oversight. There are major gaps in our basic knowledge of the mechanisms insects use to perceive and respond to their environment, and in the systems for insect growth, development, digestion, metabolism, and survival. These knowledge gaps are hampering the development of new integrated pest management (IPM) strategies for stored-product insect pests. The need for novel systems of insect control based on knowledge of basic biology coincides with unprecedented advances in genomic analysis of pest insects, which can fuel the growth of such knowledge.

**Objectives:** The project has three major objectives: 1) to develop new gene and protein manipulation technologies for discovery of novel biological targets in pest insects; 2) to characterize vulnerable genetic and physiological pathways such as those involved in digestion, tissue fluid regulation, and immunity; and 3) to develop DNA fingerprinting technologies for identifying populations and infestation sources, and for incorporation into IPM systems for stored-product insects.

The proposed research will lead to deeper knowledge of gene families and protein pathways that regulate vital physiological processes in insect tissues. Attaining these objectives will reveal novel protein targets and could lead to safer, more selective inhibitors for insect pest control. Knowledge of *Tribolium* genetic content will lead to rapid advances in biological knowledge and control strategies for other similar pest species. Genetic analyses will foster improvements in molecular fingerprinting of insects, leading to a better understanding of the structure and dynamics of insect populations.

Expected products include improved biocontrol agents that block molting or reproduction or that inhibit digestion, as well as novel gene and protein targets that can be inhibited by feeding on transgenic crops.

Anticipated benefits include new lines of cereals that express insect control proteins to confer pest resistance. New DNA fingerprinting methods will improve the accuracy of species and biotype identification, and also facilitate monitoring of population movements and more accurate identification of sources of infestation and pest resurgence.

This research will benefit farmers, producers, and academic and industrial partners involved in storage and processing of grain and grain products. Scientists will benefit from basic knowledge of insect genome organization and comparative genomic sequence, and from knowledge of gene pathways that regulate embryonic and larval growth and development, and digestive physiology. Finally, the research will benefit consumers demanding ecologically sound pest control systems founded on basic knowledge of insect biology. SCA partners are involved in whole-genome sequence analysis and annotation of *Tribolium*, in the evaluation of wheat protease inhibitors as biopesticides, in the search for osmoregulatory, digestive or neuroendocrine components for new biopesticide targeting, and in the RNAi-based screening of solute transporters for discovery and validation of genes involved in pesticide transport or excretion.

## Results and Impact:

**1. Manual Annotation Teams Organized and Implemented for the *Tribolium* Genome Project.** Without accurate annotation and functional analysis of an insect genome sequence, the value of that sequence to science and industry is reduced. Our laboratory was selected as the official clearing house for manual annotation of all "pest biology" genes in *Tribolium*, and analysis has been completed on >200 such genes,

including P450 (cyp) genes, cuticle protein genes, trans-membrane transporter genes, and others. The starting point for this manual effort is the putative gene list derived from the automated annotation of the *Tribolium* genome sequence, which was completed in May 2006 and includes 16,400 gene models. This accomplishment resulted from the joint efforts of the ARS Biological Research Unit in Manhattan, KS, Kansas State University, the Baylor College of Medicine's Human Genome Sequencing Center, the National Center for Biotechnology Information, and participants at the International *Tribolium* Genomics Meeting in Gottingen, Germany, in 2005. The functional analysis of genes characterized in the manual annotation will have far-reaching impact on broad knowledge of insect genome evolution, physiological adaptations in pest and beneficial beetle species, and the identification of novel targets for pest control exploitation.



We identified more than 100 genes that encode structural proteins just in the exoskeleton (cuticle). This figure shows increasing expression of one of the adult-specific "hard cuticle" proteins in the maturing pupa, revealed by insertion of a fluorescent reporter gene into the cuticle protein gene.

## **2. Genes Identified that Regulate Chitinous Structures in Insect Cuticle and Midgut.**

Insect-specific physiological structures and processes such as chitin and molting can be targeted for design of insect-specific biopesticides. We found approximately 20 new cuticle/chitin genes in the *Tribolium* genome sequence, isolated many of the corresponding cDNAs, and described the deleterious effects on development when the functions of some of these genes are inhibited, using the technique of RNA interference. The new genes include chitinases, chitin deacetylases, syntaxin, obstructors, cuticle proteins, and peritrophins, and have functions that include cuticle synthesis, assembly, modification, and degradation. This work was done in cooperation with Kansas State University. Examination of the formation, regulation and reutilization of the insect exoskeleton and midgut peritrophic membrane is revealing a great variety of candidate genes for biopesticide-mediated disruption.

## **3. Biochemical Analysis of Beetle Digestion Yields Valuable Insights.**

A comprehensive biochemical and genetic study of digestive proteases in *Tenebrio molitor* has yielded valuable insights into the complexity of digestion in this beetle storage pest. Multiple forms of cysteine- and serine-type proteases were compartmentalized, presumably for maximal activity and ensuring an efficient as well as tightly-regulated control of food digestion. These enzymes operate in a concerted fashion to digest oat proteins. Enzymes that initiate digestion were speculated to be primary targets for inhibitors, such as plant protease inhibitors, that can be developed as new control products for beetle storage pests. This information also will be used to study the regulation of digestive

enzymes in the genome of the closely related beetle, *Tribolium castaneum*.

### **Goals for 2007, 2008, and 2009:**

#### **Specific tasks in 2007 will be to:**

1. Continue manual adjustment of genome annotation and gene discovery.
2. Complete testing of promoters and transactivators and incorporation into vectors for gene tagging, enhancer trapping and misexpression. Enter data for insertional mutagenesis library into BeetleBase.
3. Conduct Northern and qPCR analysis of proteinase gene expression. Initiate comparison of Bt toxin binding proteins in coleopteran pests. Initiate proteinase studies with Bt toxins.
4. Initiate microarray experiments.
5. Maintain data-stream into BeetleBase.
6. Initiate statistical analysis of data from DNA fingerprinting study.

#### **Specific tasks in 2008 will be to:**

1. Continue manual adjustment of genome annotation and gene discovery.
2. Complete screening of insertional mutant libraries for physiological defects, enhancer patterns, or other traits.
3. Design new biopesticides based on proteinase inhibitor and toxin binding studies.
4. Conduct microarray analyses using oligo-based hybridization.

5. Develop theories of population structure of stored-product beetles.

**Specific tasks in 2009 will be to:**

1. Complete manual adjustment of genome annotation and gene discovery.
2. Maintain data-stream into BeetleBase.
3. Estimate the degree of sequence conservation within the Coleoptera.
4. Complete statistical analysis of the data from DNA fingerprinting study.

**Specific Cooperative Agreements for This Project Included:**

Human Genome Sequencing Center, Baylor College of Medicine, Houston, Texas

Departments of Biochemistry and Entomology, Kansas State University, Manhattan, Kansas

**Summary of 2006 Publications/Patents:**

1. Oppert, B.S., Walters, P., Zuercher, M.C. 2006. Digestive proteinases of the larger black flour beetle, *Cynaues angustus* (Leconte) (Coleoptera: Tenebrionidae). *Bulletin of Entomological Research* 96: 167-172.
2. Lewis, S.M., Kobel, A., Fedina, T., Beeman, R.W. 2005. Sperm stratification and paternity success in red flour beetles. *Physiological Entomology* 30: 303-307.
3. Arakane, Y., Muthukrishnan, S., Kramer, K.J., Specht, C.A., Tomoyasu, Y., Lorenzen, M.D., Kanost, M., Beeman, R.W. 2005. The *Tribolium* chitin synthase genes TcCHS1 and TcCHS2 are specialized for synthesis of

epidermal cuticle and midgut peritrophic matrix. *Insect Molecular Biology* 14: 453-463.

4. Oppert, B.S. 2006. Two-dimensional analysis of proteinase activity. *Journal of Biochemical and Biophysical Methods* 67: 173-179.

5. Bolognesi, R., Arakane, Y., Muthukrishnan, S., Kramer, K.J., Terra, W.R., Ferreira, C., Beeman, R.W. 2005. Sequences of cDNAs and expression of genes encoding chitin synthase and chitinase in the midgut of *Spodoptera frugiperda*. *Insect Biochemistry and Molecular Biology* 35: 1249-1259.

6. Elpidina, E.N., Tsybina, T.A., Dunaevsky, Y.E., Belozersky, M.A., Zhuzhikov, D.P., Oppert, B.S. 2005. Purification and characterization of a chymotrypsin-like proteinase from the midgut of *Tenebrio molitor* larvae. *Biochemie* 87: 771-779.

7. Li, H., Oppert, B.S., Higgins, R.A., Huang, F., Buschman, L.L., Gao, J., Zhu, K.Y. 2005. Characterization of cDNAs encoding three trypsin-like proteinases and quantitative analysis of mRNA in Bt-resistant and -susceptible strains of *Ostrinia nubilalis*. *Insect Biochemistry and Molecular Biology* 35: 847-860.

8. Petrek, J., Vitecek, J., Vlasinova, H., Kizek, R., Kramer, K.J., Adam, V., Klejduš, B., Havel, L., Beeman, R.W. 2005. Application of computer imaging, stripping voltammetry and mass spectrometry to study the effect of lead (Pb-EDTA) on the growth and viability of early somatic embryos of Norway spruce (*Picea abies*/L.Karst.). *Analytical Bioanalytical Chemistry* 383: 576-586.

9. Matsumiya, M., Arakane, Y., Haga, A., Muthukrishnan, S., Kramer, K.J., Beeman, R.W. 2006. Substrate specificity of chitinases from two species of fish, greenling, *Hexagrammos otakii*, and common mackerel, *Scomber japonicus*, and the insect, tobacco hornworm, *Manduca sexta*. *Insect Biochemistry and Molecular Biology* 70: 971-979.
10. Suderman, R.J., Dittmer, N.T., Kanost, M.R., Kramer, K.J., Beeman, R.W. 2006. Model reactions for insect cuticle sclerotization: cross-linking of recombinant cuticular proteins upon their laccase-catalyzed oxidative conjugation with catechols. *Insect Biochemistry and Molecular Biology* 36: 353-365.
11. Tamez-Guerra, P., Damas, G., Iracheta, M.M., Oppert, B.S., Gomez-Flores, R., Rodriguez-Padilla, C. Differences in susceptibility and physiological fitness of Mexican field *Trichoplusia ni* (Hubner) strains exposed to *Bacillus thuringiensis*. *Journal of Economic Entomology* 99: 937-945.
12. Hogenkamp, D.G., Arakane, Y., Zimoch, L., Merzendorfer, H., Kramer, K.J., Beeman, R.W., Kanost, M.R., Specht, C.A., Muthukrishnan, S. 2005. Chitin synthase genes in *Manduca sexta*: characterization of a gut-specific transcript and differential tissue expression of alternately spliced mRNAs during development. *Journal of Insect Biochemistry and Molecular Biology* 35: 529-540.
13. Beeman, R.W., Lorenzen, M.D., Brown, S.J., Stuart, J.J. 2005. The selfish gene Medea in *Tribolium castaneum*: an update [Abstract]. *Tribolium Transgenics Meeting and International Tribolium Genetics Meeting*, Gottingen, Germany, August 1-5, 2005.
14. Beeman, R.W., Lorenzen, M.D., Brown, S.J., Gibbs, R., Weinstock, G., Richards, S., Liu, Y. 2005. Integration of the recombination and physical maps with the genome sequence of *Tribolium castaneum* [Abstract]. *Tribolium Transgenic Meeting and International Tribolium Genetics Meeting*, Gottingen, Germany, August 1-5, 2005.
15. Lorenzen, M.D., Wang, L., Brown, S.J., Beeman, R.W. 2005. Annotation of the *Tribolium castaneum* genome [Abstract]. Presentation at the *International Tribolium Genetics Conference*, Gottingen, Germany, August 1-5, 2005.
16. Beeman, R.W., Lorenzen, M.D., Brown, S.J. 2005. Unveiling the first beetle genome [Abstract]. *National Entomological Society of America Annual Meeting*, Fort Lauderdale, FL, November 6-9, 2005.
17. Lorenzen, M.D., Beeman, R.W. 2006. The *Tribolium* genome project and its impact on pest management strategies [Abstract]. Presentation, 2006 *International Plant Resistance to Insects Workshop*, West Lafayette, IN, April 9-12, 2006.

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**CRIS 5430-43000-027-00D**

## **Ecology, Sampling, and Modeling of Insect Pests of Stored Grain, Processing Facilities, and Warehouses**

**Project Leader:** P. Flinn

**Investigators:** P. Flinn, J. Campbell, and J. Throne

**Full-Time Scientist Equivalents (SYs):** 3.0

**Start Date:** 04/28/05

**Termination Date:** 04/27/10

**Problem:** Insects reduce the quality of stored grain and other stored products in the U.S. and in the world. Over 12 billion bushels of corn and wheat are grown in the U.S. each year, with a value of over 25 billion dollars. In addition, over a billion bushels of barley, oats, rice, rye, and sorghum are grown in the U.S. each year, with a value of over 3 billion dollars. It is estimated that postharvest losses to raw grains due to insects are 5 to 10%, or about 1.4 to 2.8 billion dollars. Losses to processed commodities are difficult to quantify, but probably greatly exceed the losses to raw commodities. Many of the insecticides used by the cereal foods industry are being lost due to insecticide resistance or regulatory changes. Thus, alternative, economically viable methods for controlling these insects and reducing losses to raw and processed commodities are required. Our goal is to improve integrated pest management (IPM) of stored-product insects through development of better monitoring methods and use of computer models to optimize control strategies.

**Objectives:** This project has nine main goals: 1) Improve understanding of the abiotic factors that govern stored-product insect population dynamics; 2) Determine pest status of insect species, particularly emerging pests; 3) Determine how nutritional factors and

commodity quality can affect stored-product pest population dynamics and pest management decisions; 4) Develop improved sampling methods for insects in static and moving bulk commodities; 5) Improve and optimize pheromone/food traps for monitoring stored-product pest insect populations; 6) Improve methods for interpreting trap catches to aid in making stored-product pest management decisions; 7) Determine sampling-based action thresholds for stored-product insect pests; 8) Determine interaction of control technologies and optimize their combined use; and 9) Develop user-friendly tools to aid storage managers in making pest control decisions.

Attaining these objectives will provide benefits to managers of grain storage and grain processing facilities, food warehouses, and retail stores and to consumers by optimizing monitoring and management strategies for stored-product insect pests. Pest resurgence after control treatments is a major problem for the grain processing industries, and this research will improve our understanding of that problem. The research will determine the importance of emerging pests, such as psocids which are small, soft-bodied insects, sometimes called booklice. Anticipated products are improved insect monitoring and sampling technologies,

improved interpretation methods for sampling and monitoring systems, new computer simulation models that can be used to optimize monitoring and management strategies in grain storage and processing facilities, strategies for avoiding pest resurgence after control treatments are applied, and new information regarding the prevalence and environmental conditions that cause outbreaks of emerging pests, such as psocids.

Potential users of the knowledge developed during this project will be farmers; managers of grain storage and grain processing facilities, warehouses, and retail stores; pest control operators; manufacturers of storage-monitoring equipment; consumers; extension agents; and scientists. Input from these customers will be made through participation in a variety of technical and non-technical meetings. Unit members meet regularly with the Food Protection Committee of the International Association of Operative Millers to transfer research results and determine research needs; this committee consists of about 25 members who are responsible for pest and quality control for most of the major U.S. milling companies.

## **Results and Impact:**

### **1. Long-Term Population Trends of Stored-Product Insects at Flour Mills.**

There is limited data available on the long-term population trends of stored-product insects in food processing facilities or the impact of pest management tactics on pests in the field, but this information is critical for the development of integrated pest management programs. Scientists in the Biological Research Unit monitored several commercial sites, including flour mills, warehouses, and feed mills to evaluate seasonal trends in pest

populations inside and outside facilities, pest species diversity, and the impact of treatments such as fumigation with methyl bromide or sulfuryl fluoride, aerosol fogging, or chemical pesticide sprays on population dynamics. This information will be used to help in the development of population models of pests in the field and to evaluate the efficacy of different management tools and ultimately to develop integrated pest management programs for the food industry.

### **2. Dispersal Ability of the Lesser Grain Borer.**

The lesser grain borer, is one of the most damaging insects pests of stored wheat in the U.S. High immigration rates of this pest in farm bins in Kansas suggests that this movement is important in establishing pest populations in bins. The dispersal ability of these beetles and the potential sources are unknown. Scientists in the Biological Research Unit in collaboration with scientists at Kansas State University studied the dispersal ability of this species in the field using mark-release-recapture. Beetles dispersed an average distance of 380 meters, with a range from 50 meters to 3.6 kilometers, the distance of the furthest traps. Understanding the dispersal distances of a pest can help in the identification of important sources of these important pests and open new targets for pest management.

### **3. Myosin ELISA Test May Underestimate Levels of Insect Infestation in Grain.**

The insect fragment count has been used as a standard procedure in food analysis for many years; however, it requires technical training and is time consuming and relatively expensive. An alternative to the insect fragment count is a commercial immunoassay (ELISA) method that detects the insect muscle protein, myosin; however, there are concerns that myosin may break down over time.



Scientists in the Biological Research Unit conducted studies using the ELISA test. Hard red winter wheat was infested with larvae of the lesser grain borer and then fumigated to kill the larvae inside the kernels. Infected kernels were then "aged" at 90°F for 0, 14, 28, or 56 days after fumigation.



X-ray showing insects inside wheat kernels.

Myosin degradation was most rapid in the first two weeks after the larvae were killed, decreasing 58%. There were no significant differences in myosin levels between samples that were 14, 28, and 56 days old. These results indicate that the myosin ELISA test may underestimate the amount of insect contamination in grain that has been previously fumigated.

**4. Beneficial Insects Control Indianmeal Moth.** The Indianmeal moth is a serious pest of raw and finished stored products and attacks both packaged and bulk commodities as well as spillage. Scientists in the Biological Research Unit in collaboration with scientists at Kansas State University showed that a harmless parasitoid wasp, *Trichogramma deion*, was very effective in finding and

killing Indianmeal moth eggs on shelves in a simulated retail store environment.



Dying Indianmeal moth larvae parasitized by *Habrobracon hebetor*.

*Trichogramma deion* killed more Indianmeal moth eggs on open-type shelving than on gondola-type shelving. The presence of packages on the shelves did not interfere with the wasp locating and killing the Indianmeal moth eggs. *Trichogramma* goes after the moth eggs before they can develop into adult moths. This could provide a new tool for the retail organic food industry to manage insect pests.

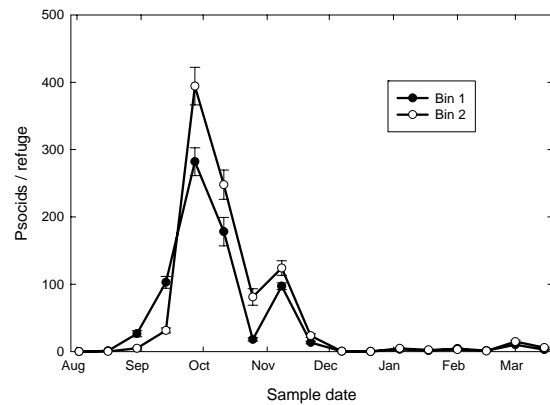
#### **5. Combination of Two Parasitoid Insects Works Best to Suppress Indianmeal Moth.**

The Indianmeal moth is a serious pest of raw and finished stored products and attacks both packaged and bulk commodities as well as spillage. Scientists in the Biological Research Unit in collaboration with scientists at Kansas State University showed that a combination of packaging and two species of parasitic wasps, *Trichogramma deion* and *Habrobracon hebetor*, provided good moth suppression. *Trichogramma* goes after the moth eggs before they can develop into damaging larvae, and *Habrobracon* finishes the job by killing any larvae that develop from eggs that *Trichogramma* may have missed. This could provide a new tool for the retail organic food industry to manage insect pests.

**6. Seasonal Distribution of Psocids in Stored Wheat.** Psocids are an emerging problem in grain stored in the U.S. and in grain processing facilities. Scientists in the Biological Research Unit conducted preliminary studies to determine which species of psocids were present in a feed mill, a grain elevator, and in wheat stored in steel bins. We then conducted a more extensive study in steel bins containing wheat to determine temporal and spatial distribution of psocids in the wheat. We also compared several sampling methods for the psocids – cardboard refuges, grain trier samples, and automated sampling using the StorMax Insector system. The predominant psocid species found in all locations was *Liposcelis entomophila*.

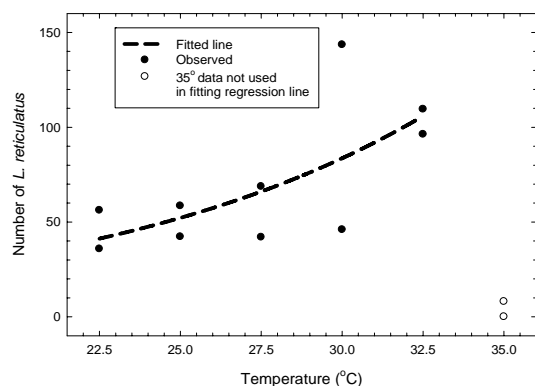


In the study on temperospatial distribution, infestation levels before the bins were filled with wheat were low, but some psocids were present in the empty bins. Number of psocids in cardboard refuges on the wheat surface were low immediately after bins were filled in July, peaked in late September, dropped to almost zero in December as temperatures dropped during winter, and then remained at low levels until the study was ended in April.



Number of psocids in cardboard refuges was indicative of number of psocids in grain samples. The results indicate that cardboard refuges may provide an efficient method for sampling psocids in bins of wheat, and that psocid populations can increase quickly to high levels during storage even though they are low early in the storage period.

**7. Effects of Environmental Conditions on Development of the Psocid *Lepinotus reticulatus*.** Psocids are an emerging problem in grain stored in the U.S. and in grain processing facilities, and environmental conditions play a major role in determining how quickly their populations develop. Scientists in the Biological Research Unit showed that *L. reticulatus* could not live and reproduce when relative humidity was at or below 55%. At 75% relative humidity, population growth was greatest at 30 degrees C., and dropped to almost zero at 35 degrees C.



These results indicate that it should be possible to effectively manage *L. reticulatus* populations using heat disinfestations with temperatures above 35°C.

#### Goals for 2007, 2008, and 2009:

##### Specific tasks in 2007 will be to:

1. Write paper on results of digital X-ray detection of internal-feeding insects in grain samples.
2. Analyze red flour beetle behavior in spillage data and write paper on results.
3. Conduct warehouse beetle tracking experiments.
4. Analyze red flour beetle response to traps in experimental boxes.
5. Complete DNA extraction, PCR with microsatellite primers, and analyze data for population genetics study.
6. Continue monitoring studies at food facilities and initiate analysis of data to determine how spatial distributions of insect pests change before, during, and after control treatments.

7. Conduct mark-recapture studies to assess dispersal of insect pests.

8. Continue monitoring pest population levels and environmental conditions in food facilities to determine how long-term population dynamics of stored product insects influences pest resurgence following treatment.

9. Continue measuring emigration and spatial variation in treatment efficacy in food facilities to evaluate the potential for insect pests to survive in food residues and to avoid treated areas.

10. Conduct experiments on the influence of perimeter treatments on red flour beetle immigration and emigration.

11. Develop simulation model for warehouse beetle.

12. Refine how the models simulate the movement of insects within and outside of flour mills, and the effects of control treatments on movement and mortality.

13. Conduct simulation studies with the models to determine optimal management strategies.

14. Continue to sample farm bins and processing facilities in Kansas and Oklahoma for psocids to investigate the ecology and potential impact of emerging pest species.

15. Investigate the relationship between psocid density in grain and subsequent fragment counts in flour.

**Specific tasks in 2008 will be to:**

1. Write paper on prediction of insect fragments in flour using digital X-ray detection.
2. Analyze warehouse beetle tracking data and prepare paper.
3. Study warehouse beetle response to traps in experimental boxes.
4. Prepare paper on red flour beetle pheromone trapping and interpretation.
5. Prepare papers on results of population genetic analysis of red flour beetle.
6. Conduct source identification experiments with red flour beetle.
7. Conclude monitoring studies at food facilities and analyze field efficacy and rate of rebound.
8. Conclude mark-recapture studies to assess insect dispersal.
9. Complete DNA extraction and PCR with microsatellite primers and analyze data for population genetics study.
10. Analyze data and prepare manuscript on monitoring pest population levels and environmental conditions in food facilities to determine how long-term population dynamics of stored-product insects influences pest resurgence following treatment.
11. Continue measuring emigration and spatial variation in treatment efficacy in food facilities to evaluate the potential for pests to survive in food residues and avoid treated areas.

12. Analyze data and write paper on the influence of perimeter treatments on red flour beetle immigration and emigration experiments to determine how long-term population dynamics of stored-product insects influences pest resurgence following treatment.

13. Validate and collect data to refine the warehouse beetle simulation model.

14. Refine the red flour beetle, Indianmeal moth, and warehouse beetle models to include other factors.

15. Conduct simulation studies with the models.

16. Analyze data and prepare manuscript on the prevalence of psocids in stored wheat and processing facilities to investigate the ecology and potential impact of emerging pest species.

**Specific tasks in 2009 will be to:**

1. Analyze data for warehouse beetle response to traps in experimental boxes.

2. Analyze data and prepare paper on warehouse beetle pheromone trapping and interpretation.

3. Analyze data and prepare paper on red flour beetle pheromone trapping and interpretation.

4. Analyze data from monitoring studies in food facilities to determine how spatial distributions of insect pests change before, during, and after control treatments.

5. Analyzed data and write paper on DNA extraction and PCR with microsatellite

primers and analyze data for population genetics study.

6. Analyze data and prepare paper on the emigration of insects and spatial variability of treatment efficacy in food processing facilities.

7. Continue to refine the red flour beetle, Indianmeal moth, and warehouse beetle models.

8. Write papers on simulation models and treatment strategies for the red flour beetle, Indianmeal moth, and warehouse beetle.

9. Analyze data and prepare paper on the economic impact and pest ecology of emerging pests in stored grain and warehouses, such as psocids.

#### **Specific Cooperative Agreements for This Project Included:**

Department of Electrical and Computer Engineering, Kansas State University, Manhattan, Kansas

Department of Entomology, Oklahoma State University, Stillwater, Oklahoma

Department of Agricultural & Biochemical Engineering, Purdue University, West Lafayette, Indiana

Department of Entomology, Kansas State University, Manhattan, Kansas

#### **Summary of 2006 Publications/Patents:**

1. Campbell, J.F. 2005. Fitness consequences of multiple mating on female *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae). *Environmental Entomology* 34: 833-843.

2. Campbell, J.F. Stored product insect behavior. In: J.W. Heaps, Editor, *Insect Management for Food Storage and Processing*. St. Paul, MN. AACC International. p. 39-51.

3. Flinn, P.W., Kramer, K.J., Throne, J.E., Morgan, T.D. 2005. Protection of stored maize from insect pests using a two-component biological control method consisting of a hymenopteran parasitoid, *Theocolax elegans*, and transgenic avidin maize powder. *Journal of Stored Products Research* 42: 218-225.

4. Perez-Mendoza, J., Throne, J.E., Maghirang, E.B., Dowell, F.E., Baker, J.E. 2005. Insect fragments in flour: relationship to lesser grain borer (Coleoptera: Bostrichidae) infestation level in wheat and rapid detection using near-infrared spectroscopy. *Journal of Economic Entomology* 98: 2282-2291.

5. Qureshi, J.A., Buschman, L.L., Throne, J.E., Ramaswamy, S.B. 2006. Dispersal of adult *Diatraea grandiosella* (Lepidoptera: Crambidae) and its implications for corn borer resistance management in *Bacillus thuringiensis* maize. *Annals of the Entomological Society of America* 99: 279-291.

6. Ramos-Rodriguez, O., Campbell, J.F., Ramaswamy, S. 2006. Pathogenicity of three entomopathogenic nematode species to some major stored product insect pests. *Journal of Stored Products Research* 42: 241-252.

7. Scholler, M., Flinn, P.W., Grieshop, M., Zdarkova, E. 2006. Biological Control of Stored Product Pests. In: J.W. Heaps, Editor, *Insect Management for Food Storage and*

Processing. St. Paul, MN. AACC International. p. 67-87.

8. Campbell, J.F. 2006. Assessing the fitness consequences of parasite infection decisions [abstract]. Society of Nematologists Proceedings, Kauai, Hawaii, June 18-21, 2006.

9. Flinn, P.W. 2006. Areawide IPM for insects in commercial grain elevators [abstract]. 5th International IPM Symposium, St. Louis, MO, April 4-6, 2006.

10. Lewis, E., Campbell, J.F., Griffin, C., Kaya, H., Peters, A. 2006. Behavioral ecology of entomopathogenic nematodes. *Biological Control* 38: 66-79.

11. Campbell, J.F., Ramos-Rodriguez, O., Ramaswamy, S. 2005. Do entomopathogenic nematodes have potential as biological control agents of stored product insects? [abstract]. Society for Invertebrate Pathology Meeting, Anchorage, Alaska, August 7-11, 2005.

12. Flinn, P.W. 2005. Recent technological advances for managing insect pests of stored-grain. Meeting Proceedings of the V Brazilian Congress of Agroinformatics, Londrina, Brazil [CD-ROM].

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# ENGINEERING RESEARCH UNIT

The mission of the Engineering Research Unit is to: (1) develop technologies to improve grain quality assessment, handling, and storage through innovative engineering research; (2) transfer knowledge and technology that meet the needs of consumers and the grain industry; (3) provide engineering expertise through cooperative research with other laboratories, agencies, universities, and industry groups; and (4) maintain an environment that fosters teamwork, innovation, and personal growth. Specific projects for this Unit include:

- |                           |                                                                                            |
|---------------------------|--------------------------------------------------------------------------------------------|
| CRIS - 5430-43440-005-00D | Improved Handling and Storage Systems for Grain Quality Maintenance and Measurement        |
| CRIS - 5430-44000-015-00D | Objective Grading and End-Use Property Assessment of Single Kernels and Bulk Grain Samples |

## Improved Handling and Storage Systems for Grain Quality Maintenance and Measurement

**Project Leader:** M. Casada

**Investigators:** M. Casada, F. Dowell, and P. Armstrong

**Full-Time Scientist Equivalents (SYs):** 1.7

**Start Date:** 08/15/04

**Termination Date:** 08/14/09

**Problem:** Worldwide grain markets are changing and customers are now demanding grain that meets their specific needs for end use properties and that is purer, safer, and more wholesome. They are demanding grain with fewer insects, diseases, and pesticides and either no genetically modified (GM) grain or strictly controlled levels of GM grain.

(AAWCO) has asked ARS to evaluate their existing packing factor data.

The introduction of transgenic crops into the U.S. grain handling system has shown that the infrastructure is largely unable to preserve the identity of specialty grains to the desired level of purity. Fundamental data are needed on commingling during handling, along with methods to minimize or eliminate this problem to effectively separate grains with special desirable characteristics, which adds value compared to commodity grains. A survey of our industry focus group showed a strong support for this study of identity preservation of grain.

Aeration is an underused tool for controlling insects and other risks in stored grain without the use of chemical pesticides, particularly in small grains in warm climates, a significant omission leading to continued storage losses of 5 to 10% in stored grain in some U.S. climates. Appropriate improved control strategies and improved monitoring systems are needed so that aeration will be more widely used to reduce pesticide use in stored grain. Even when aeration is used to reduce or eliminate pesticide use in stored grain, pesticides are still used to sanitize bins before storage; thus, a chemical-free method of pre-filling sanitation of grain storage bins is also needed. Our industry focus group survey showed strong support for studies of stored grain insect control, and especially strong support for these entirely chemical-free methods. With typical grain losses of 5 to 10% to stored grain insects, total losses in the U.S. exceed \$1 billion.

Existing packing factor data are of unknown reliability and are widely mistrusted in the industry. Accurate data are required for government-mandated inventory control and are a crucial component of new quality management systems being developed to enable source verification in the grain handling industry. The Farm Service Agency (FSA) is not able to do a thorough review and, as a result, along with the Association of American Warehouse Control Officials

Grain dust has adverse health effects and can lead to explosion disasters such as the 121 reported agricultural dust explosions in the last ten years (1993 - 2002) that killed 16 people, injured 149, and caused over \$100 million of facility damage. Improved system design procedures are needed for grain dust collection systems and data are needed on



dust emission, composition, and particle size distribution during grain unloading as affected by type of grain, moisture content, grain quality, grain flow rate, and drop height.

**Objectives:** The specific objectives of this research are: 1) Measure grain commingling levels during elevator handling for use in developing procedures, decision support systems, and instrumentation to facilitate value-added and identity-preserved grain segregation operations; 2) Develop new stored grain packing factors with known accuracy as needed for common grains in trade over a range of field conditions; 3) Develop improved aeration, monitoring, and sanitation systems and best management practices for quality maintenance and insect control in stored grain; and 4) Measure dust emission, particle-size distribution, and air entrainment during grain unloading to facilitate reducing dust emissions from grain handling operations and equipment.

The potential impact of this research is that improved grain drying, storing, aerating, and handling while maintaining quality and energy efficiencies will increase food wholesomeness, safety, and market competitiveness. Several specific problems relating to quality maintenance, grain purity, energy, and safety during grain handling, aeration, and storage are being resolved by this project. This research is relevant to producers, elevator and dryer operators, equipment and facility design engineers, marketers and handlers of grain, Extension Service specialists, and government regulatory agencies.

## **Results and Impact:**

**1. Summer Aeration of Stored Wheat in the Hard Red Winter Wheat Belt.** It is difficult to aerate wheat immediately after

harvest as part of an integrated pest management (IPM) strategy in much of the hard red winter (HRW) wheat belt due to high ambient temperature as well as high nighttime relative humidity. The high ambient temperatures are well known, but the complicating effect of high humidity has not been well documented. Using 50 years of historical weather data, we developed maps showing contours of average hours when summer aeration is effective after accounting for the complicating effect of high humidity. The actual hours available for effective grain cooling were reduced by an average of 68% for 12% moisture content wheat and by 88% for 10% moisture content wheat compared to the hours suggested by ambient temperatures when neglecting the humidity effect. These results indicate that previously published design data suggested fan sizes less than one-half that actually needed for effective summer aeration. These maps allow engineers to properly design aeration systems with correct fan sizes for summer aeration and clarify where the climate makes summer aeration impractical.

**2. Heat Treating Grain Bins to Control Insects.** Our previous research has shown that heat treatments are effective as a non-chemical method for disinfestation of empty grain storage bins. We developed an empirical economic risk model to compare variable costs for five tested heating systems for disinfestation of empty, 5000 bu grain storage bins with fitted drying floors. The high-output, 29 kW, propane heating system had the lowest cost and risk level of all heating systems and achieved the target temperature of 50°C within 2 hours at all test locations. Lower power systems requiring complex heat distribution or recirculation were not cost effective and exhibited higher risk levels of insect survival. These results indicate that properly-sized portable propane heat

treatment systems are equal to chemical applications for low-cost, low-risk disinfestation of empty bins, but without the concerns that arise with using chemicals.

### **3. Monitoring Moisture Content of Stored Grain.**

Grain temperature and moisture content (MC) are fundamentally important for safe grain storage. Temperature monitoring of grain is relatively easy, but there are no commercially available sensors for directly monitoring MC. Relative humidity (RH), however, can be an indirect way to measure MC by measuring the RH of the air surrounding the grain. The air RH comes into equilibrium with the grain depending on the grain MC and temperature (T). MC can thus be predicted from equations that are experimentally derived and use RH and T. Unfortunately these equations are not perfect and, thus, have inherent equation error in addition to the error of the RH and T sensing. This research found that RH and T sensor error did not influence grain MC prediction as greatly as the equation error. With development of better equations, accuracy could be increased for grain moisture content monitoring in bins.

#### **Goals for 2007, 2008, and 2009:**

##### **Specific tasks in 2007 will be to:**

1. Finalize basic procedures and recommendations for IP handling operations; obtain additional data.
2. Complete initial wheat data and model calibration for wheat; seek additional funds based on these wheat results.
3. (a) Third year of field trials; complete simple physical model; (b) complete data analysis & economic model, evaluate additional research needs; (c) data collection

on performance and reliability of wireless sensors, develop model and use guidelines; (d) examine signal analysis of insect monitor and implement improvements.

4. Complete dust emission data and continue model development.

##### **Specific tasks in 2008 will be to:**

1. Begin revision of expert system; obtain additional data; evaluate specific equipment for design improvements.
2. Begin data collection for 2nd grain type; continue seeking funds, or begin large project if funds in place.
3. (a) Begin complete physical model; evaluate further field trials; (b) none in this FY; (c) design practical sensors for RH/Temp monitoring, test large scale storage applications; (d) determine improvements in monitoring based on storage tests.
4. Complete model refinement, evaluate need for more data and dust control methodologies.

##### **Specific tasks in 2009 will be to:**

1. Obtain additional data; finalize revised expert system. Begin recommendations for design improvements.
2. Continue data collection for second grain type on a small scale, or large scale if funding is in place.
3. (a) Continue work on complete model; possibly, more field trials; (b) none in this FY; (c) Examine adaptation of CO<sub>2</sub> sensors; (d) Use guidelines determined from tested effectiveness of improved methods.

**Specific Cooperative Agreements for This Project Included:**

Department of Biological and Agricultural Engineering, Kansas State University, Manhattan, Kansas

**Summary of 2006 Publications/Patents:**

1. Akdogan, H.P., Casada, M. 2006. Climatic humidity effects on controlled summer aeration in the hard red winter wheat belt. Transactions of the ASABE. 49(4): 1077-1087.
2. Armstrong, P.R., Uddin, S., Zhang, N. 2006. Accuracy of grain moisture content prediction using temperature and relative humidity sensors. Applied Engineering in Agriculture. 22(2): 267-273.

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**CRIS 5430-44000-015-00D**

## **Objective Grading and End-Use Property Assessment of Single Kernels and Bulk Grain Samples**

**Project Leader:** T. Pearson

**Investigators:** T. Pearson, F. Dowell, and P. Armstrong

**Full-Time Scientist Equivalents (SYs):** 2.3

**Start Date:** 09/25/04

**Termination Date:** 09/24/09

**Problem:** The U.S. is a major consumer and exporter of cereal grains. However, for the U.S. grain industry to remain competitive internationally and to meet domestic consumer demands for quality, it must continually improve the quality of grain and grain products. We propose to address three important problems that the U.S. grain industry faces in order to improve end-use quality of grain: 1) Grain properties that have the most influence on final or end-use product quality are not well known. For example, we do not know how most wheat kernel properties affect bread quality. This makes selection of grain difficult for buyers, and it is difficult for breeders to know what traits should be propagated; 2) Many instruments to detect quality of whole grains suffer from poor accuracy, high cost, being too slow, requiring toxic chemicals, or do not directly measure what customers need to know: end-use qualities; 3) In many cases, economically viable instruments do not exist to measure and/or sort whole grain defects that occur in small fractions of grain but can have a deleterious effect on quality, such as insect-damaged wheat kernels or mold damaged grain.

**Objectives:** This technology will enable grain handlers to detect high-quality specialty grains, GMO's and food safety concerns such

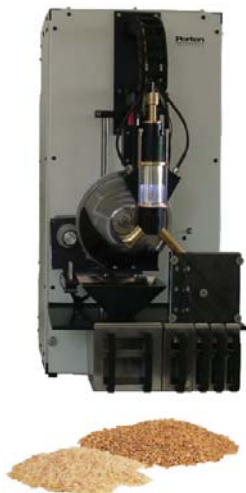
as toxins, biosecurity issues, quarantine issues, etc. for subsequent segregation. The technology will also help handlers improve quality by sorting individual kernels to improve quality and food safety of the grain. Millers and bakers will gain insight into properties of the kernels that correlate to higher quality products, thus enabling them to better select grain for their specific needs. In some cases, technologies will enable sorting of grain to improve low-quality grain to a higher quality, such as by removing fungal-damaged or low-protein kernels from mixed lots. Given knowledge of grain properties that produce premium end-use qualities and non-destructive methods to measure these properties, grain customers will be able to purchase grain that more consistently meets their quality needs and producers will be able to segregate grain lots with higher quality. In addition, breeders will be able to use this technology to identify single kernels with traits that would be desirable to propagate.

New technology and information developed through this research will be of use throughout the entire grain industry where quality and/or safety are of a concern. This includes producers, breeders, growers, grain handlers, marketers, millers, bakers, and government agencies such as the Extension

Service, FGIS/GIPSA, FSIS, APHIS, and OSHA.

## Results and Impact:

**1. Applications Developed for Automated NIR Sorting Technology Commercialized through a CRADA with a Company in Stockholm, Sweden.** The system was demonstrated at several international conferences and is being publicly marketed. The system automatically scans individual wheat kernels, and then sorts kernels based on specific attributes such as protein content, hardness, amylose content, etc. The system is now being used by breeders to select specific traits from early generation breeder samples. This will significantly reduce the time and expense required to develop cultivars with specific end-use traits. The system is also being evaluated for use in detecting food safety attributes such as vomitoxin during routine grading. Although it was developed for wheat, it is also finding applications in sorghum and millet.



SKNIR system developed by ERU and commercialized by Perten Instruments.

**2. Fast Single-Kernel NIR Measurement of Grain and Oil-Seed Attributes.** Fast single kernel analysis using near-infrared spectroscopy (SKNIR) can significantly improve many areas where single kernel analysis is presently used, such as in grain and oil-seed breeding programs. Current rates of SKNIR systems are around 1 kernel/s. For this work, instrumentation was designed and tested on corn and soybeans at rate of 10 kernels/s. The design allows measurement of kernels in near free-fall conditions to realize the faster rate. Results show the instrument worked well for predicting corn and soybean moisture and soybean protein. Future work will focus on the mechanical feeding and sorting of kernels and methods to bring corn measurement accuracy to the level of soybean measurements.

**3. Rapid Methods for Predicting Grain, Flour, and End-Use Quality.** Eight quality variables that can be measured rapidly were chosen for predicting various breadmaking quality parameters (bake water absorption, bake mix time, proof height, loaf weight, crumb score, and loaf volume, and loaf volume regression) of hard red winter (HRW) and hard red spring (HRS) wheat samples. The variables used in the predictions were: test weight; average hardness, weight, length, and diameter as measured by the SKCS 4100; and protein content, mixograph water absorption, and average total wet gluten as measured by near-infrared spectroscopy. Based on the eight-variable model obtained separately for HRW and HRS wheat, loaf volume and loaf specific volume can be predicted with a model  $R^2$  greater than 0.80; bake water absorption, bake mix time, and proof height with  $R^2$  ranging from 0.58 to 0.78; loaf weight, crumb score and loaf volume regression can be predicted only with  $R^2$  ranging from 0.27 to 0.39. For loaf volume

and loaf specific volume, which has the highest potential of being predicted using rapid measurement techniques, the single highest partial R2 that accounted for 98% of the model R2 of both HRW and HRS wheat was flour protein content. Additionally, flour protein content was a significant variable for predicting bake mix time, loaf weight, and loaf volume regression for HRW wheat and bake water absorption and bake mix time for HRS wheat.

#### **4. Detecting Durum Wheat Quality.**

Durum wheat production accounts for approximately 8% of the wheat production worldwide, and is mainly used to make semolina for macaroni, spaghetti, and other pasta products. The best durum wheat for pasta products should appear hard, glassy and translucent, and have excellent amber color, good cooking quality, and high protein content. Nonvitreous (starchy) kernels are opaque and softer, and result in decreased yield of coarse semolina. Thus, vitreousness of durum wheat has been used as one of the major quality attributes in grading. Traditionally, grain grading has been primarily done by visual inspection by trained personnel. This method is subjective and tedious. It also produces great variations in inspection results between inspectors. We used digital imaging technology for determining durum vitreousness. Results showed that 100% of non-vitreous kernels and 92.6% of mottled kernels, which is one of the hardest defect categories to consistently detect visually, could be correctly classed.

**5. Improving the Quality of White Wheat through Rapid Sorting.** White wheat is gaining acceptance throughout the Midwest as a class that can improve our competitiveness in export markets. All breeding programs in the Midwest are developing white wheat

cultivars. We are able to improve the quality of white wheat cultivars being used in breeding programs by removing wheat of other classes, such as red wheat, from samples using high speed sorting procedures developed through an agreement with Satake, Inc. There is no other technology available to remove these contaminating kernels. Almost all white wheat being developed in the Midwest and Pacific Northwest is now shipped to our research unit for purification through our sorter. Our sorting has reduced the development time for these new cultivars by several years, has saved the breeders hundreds of hours, and has salvaged some cultivars that would have been terminated if our technology was not available.



**High speed sorter used to improve grain quality.**

**6. Reducing Mycotoxins in Corn.** A high-speed single-kernel sorter was used to remove mycotoxins from white corn. It was found that using spectral absorbance at 500nm and 1200nm could distinguish kernels with aflatoxin-contamination. When these two spectral bands were applied to sorting corn at high speeds, reductions in fumonisin averaged 82% for corn samples with an initial level of

aflatoxin over 10 ppb. Most of the fumonisin is removed by rejecting approximately 5% of the grain. This technology will help insure the safety of the US food and feed supply.

### **7. Detecting Insect Fragments in Flour.**

Primary pests of stored cereals that develop and feed inside grain kernels are the main source of insect fragments in wheat flour. The Food and Drug Administration (FDA) has set a defect action level of 75 or more insect fragments per 50 gram of flour. The current standard flotation method for detecting insect fragments in flour is very labor intensive and expensive. We investigated the potential of near-infrared spectroscopy (NIRS) to detect insect fragments in wheat flour at the FDA defect action level. Fragments counts with both the NIRS and the standard flotation methods correlated well with the actual number of fragments present in flour samples. However, the flotation method was more sensitive below the FDA defect action level than the NIRS method. Although the flotation method is very sensitive at the FDA action level, this technique is time consuming (almost 2 h/sample) and expensive. Although NIRS currently lacks the sensitivity of the flotation method, it is rapid, does not require sample preparation, and could be easily automated for a more sophisticated sampling protocol for large flour bulks. Therefore, this method should be reexamined in the future because NIRS technology is rapidly improving.

**8. Properties of Corn Kernels Infected by Fungi.** Near infrared spectra, x-ray images, color images, near infrared images, and physical properties of single corn kernels were studied to determine if combinations of these measurements could distinguish fungal infected kernels from non-infested kernels. Kernels used in this study were inoculated in

the field with eight different fungi: *Acremonium zeae*, *Aspergillus flavus*, *Aspergillus niger*, *Diplodia maydis*, *Fusarium graminearum*, *Fusarium verticillioides*, *Penicillium* spp.

### **9. Detecting Fungal Damaged Corn.**

Results indicate that kernels infected with *Acremonium zeae* and *Penicillium* were difficult to distinguish from non-infested kernels while all of the other severely infected kernels could be distinguished with greater than 91% accuracy. A neural network was also trained to identify infecting mold species with good accuracy, based on the near infrared spectra. These results indicate that this technology can potentially be used to separate fungal infected corn using high speed sorter; and, automatically and rapidly identify the fungal species of infested corn kernels. This will be of assistance to breeders developing fungal resistant hybrids as well as mycologists studying fungal infected corn.

**10. Relation of Single Wheat Kernel Particle Size Distribution to Perten SKCS 4100 Hardness Index.** Material from single kernels crushed on the SKCS 4100 was collected and milled in a fabricated mill, which simulates the last two rolls of a Quadramat Jr. The PSD of each single kernel was then measured using a laser particle counter. It was found that the difference between the maximum and minimum slope of the PSD below 55 micrometers could distinguish most of the hard and soft kernels. These slopes correspond to a peak in the PSD between 20 to 30 micrometers. Particle size distributions from soft kernels normally have a peak in this particle size range while hard kernels have a small, or no, peak. SKCS low level data, as well as the raw crush profile, were analyzed to find a correlation with this slope. After stepwise selection, HI, and three

normalized crush profile values were used to predict the PSD slope. The predicted slope correctly classified 95% of the hard and soft kernels. These results indicate that a calibration for the SKCS based on single kernel particle size is possible and this may give a better indication of end use quality of a wheat sample.

**11. Low-cost Bi-chromatic Image Sorting Device for Grains.** A low-cost linescan imaging system was developed to inspect and sort grains and other products at high speeds (40 kernels/s). The device captures bi-chromatic images from opposite sides of each kernel and processes the images in real time using high speed microcontrollers. Detection of scab-damaged wheat kernels was used in this study to establish system feasibility and limits. Simple image statistics and intensity histograms were used as features and were able to distinguish good kernels from scab-damaged kernels with 95% accuracy. For each kernel, image acquisition required approximately 15 ms, while 5 ms were required for image processing and classification. The controller can output a signal to divert (sort) kernels or save the images on a compact flash card for transfer to a personal computer for off-line analysis. All parts for the system cost less than \$2000.

**12. Camera Attachment for Automatic Measurement of Single-wheat Kernel Size on a Perten SKCS 4100.** Wheat kernel size any shape is an important quality factor and characteristic for adjusting milling processes. Measuring kernel size is tedious and time consuming so it cannot be done as often as some wheat millers would like. Automated machines for measuring kernel size suffer from inaccuracies and/or high cost. The Perten Single Kernel Characterization System (SKCS 4100) is an automated instrument

which measures several single kernel quality characteristics such as weight, moisture content, hardness, and diameter. Of all of these measurements, the diameter measurement is the least accurate. A low cost color camera was attached to an SKCS 4100 to enable more accurate kernel size determinations. Using image data combined with SKCS data, errors in estimating kernel length and diameter were reduced by 56% and 66%, respectively.

**13. Detection of Damaged Wheat Kernels by Impact-acoustic Emissions.** A system was built that is able to distinguish good wheat kernels from a variety of damaged kernels by dropping kernels, one at a time, onto a steel plate and digitally analyzing the resulting sounds from the impact. The types of damage studied were insect damaged kernels with exit holes, hidden insect damaged kernels without exit holes, sprout damage, and scab damage. It was found that 98% of the good kernels and 87% of the insect damaged kernels with exit tunnels can be distinguished from each other. Accuracy for scab and sprout damaged kernels was 70% and 45% for hidden insect damaged kernels. The device should be capable of inspection rates exceeding 40 kernels/s, or ~70g/min. It is non-destructive and can be made to sort kernels into one of three different groups. This technology should help grain inspectors and millers better ascertain the quality of a wheat load under consideration.

**14. Applying NIR Sorting Technology to Other Disciplines.** The NIR spectroscopy procedures developed for determining single kernel attributes were found to apply to determining characteristics of single insects and other commodities. Thus, we applied NIR spectroscopy to detecting insect parasitoids, insect species, insect age grading,



and fig quality in cooperation with the Biological Research Unit, ARS USDA, Manhattan, KS; the Dept. Entomology at KSU, Manhattan, KS; the CDC, Atlanta, GA; and the Horticultural Crops Research Laboratory, Fresno, CA. Results showed we could detect parasitized weevils and flies, fly and mosquito age, stored grain insect species, and fig quality using NIR spectroscopy. This information can be used to develop control strategies for various pest insects and to automate fig grading.



Using ERU NIR technology to scan mosquitoes for the International Atomic Energy Agency.

**Goals for 2007, 2008, and 2009:**

**Specific tasks in 2007 will be to:**

1. Detect wheat kernel defects using single kernel acoustics from impact emissions. Test prototype with field samples, including mold damaged, broken kernels, germinated and kernels damaged by insects that are external feeders.
2. Detect characteristics of single corn kernels using NIR spectroscopy. Develop calibrations.
3. Detect and remove kernels with mycotoxin-producing molds. Verify sorter

performance with more samples.

4. Detect mutants for corn breeders. Measure chemical constituents in groups of mutants and normal kernels.
5. Detect single-kernel oat milling parameters. Develop SKCS groat size, oat-fill rate measurement.
6. Detect insect fragments in flour. Develop a prototype instrument for research/ industrial lab.
7. Predict end-use quality. Verify models with field data. Begin studying other classes.

**Specific tasks in 2008 will be to:**

1. Detect wheat kernel defects using single kernel acoustics from impact emissions. Test prototype with field samples, including mold damaged, broken kernels, germinated and kernels damaged by insects that are external feeders.
2. Detect characteristics of single corn kernels using NIR spectroscopy. Begin development of commercial prototypes.
3. Detect and remove kernels with mycotoxin-producing molds. Transfer technology to industry.
4. Detect mutants for corn breeders. Modify models as needed.
5. Detect single-kernel oat milling parameters. Incorporate measurement criteria into a commercial SKCS.
6. Detect insect fragments in flour. Compare imaging and human inspection. Redesign as needed.

7. Predict end-use quality. Modify models as needed.

**Specific tasks in 2009 will be to:**

1. Detect wheat kernel defects using single kernel acoustics from impact emissions. Transfer design and software to industry.
2. Detect characteristics of single corn kernels using NIR spectroscopy. Finalize testing of the system.
3. Detect mutants for corn breeders. Transfer technology; isolate mutant genes for propagation.
4. Detect single-kernel oat milling parameters. Field trials with oat milling/breeding industry.
5. Detect insect fragments in flour. Compare imaging and human inspection. Redesign as needed.
6. Predict end-use quality. Transfer models to FGIS and others.

**Specific Cooperative Agreements for This Project Included:**

JAG Services, Inc., Manhattan, Kansas

Department of Biological and Agricultural Engineering, Kansas State University, Manhattan, Kansas

**Summary of 2006 Publications/Patents:**

1. Armstrong, P.R., Maghirang, E.B., Xie, F., Dowell, F.E. 2006. Comparison of Dispersive and Fourier-Transform NIR Instruments for Measuring Grain and Flour Attributes.

Applied Engineering in Agriculture. 22(3): 453-457.

2. Baye, T.M., Pearson, T.C., Settles, M.A. 2006. Calibration development to predict maize seed composition using single kernel near infrared spectroscopy. Journal of Cereal Science. 43(2): 236-243.

3. Delwiche, S.R., Graybosch, R.A., Hansen, L.E., Souza, E., Dowell, F. 2006. Single kernel near-infrared analysis of tetraploid (durum) wheat for classification of the waxy condition. Cereal Chemistry. 83(3):287-292.

4. Delwiche, S.R., Pearson, T.C., Brabec,, D.L. 2005. High-speed optical sorting of soft wheat for reduction of deoxynivalenol. Plant Disease Journal. 89(11):1214-1219.

5. Haff, R.P., Jackson, E.S., Pearson, T.C. 2005. Non-destructive detection of pits and pit fragments in dried plums. Applied Engineering in Agriculture.21(6):1021-1026.

6. Perez-Mendoza, J., Throne, J.E., Maghirang, E.B., Dowell, F.E., Baker, J.E. 2005. Insect fragments in flour: relationship to lesser grain borer (Coleoptera: Bostrichidae) infestation level in wheat and rapid detection using near-infrared spectroscopy. Journal of Economic Entomology 98: 2282-2291.

7. Toews, M.D., Pearson, T.C., Campbell, J.F. 2006. Imaging and automated detection of *Sitophilus oryzae* L. (Coleoptera: Curculionidae) pupae in hard red winter wheat. Journal of Economic Entomology 99: 583-592.

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# GRAIN QUALITY AND STRUCTURE RESEARCH UNIT

The mission of the Grain Quality and Structure Research Unit, which includes the Hard Winter Wheat Quality Laboratory, is to ensure a high quality and safe U.S. grain supply for our customers by: (1) conducting basic and applied research to identify the physical characteristics and structural/biochemical components that govern quality; (2) developing rapid, precise, and accurate predictive technologies for quality assessment; and (3) evaluating the end-use quality of breeding lines. Specific projects for this Unit include:

- |                           |                                                                                                 |
|---------------------------|-------------------------------------------------------------------------------------------------|
| CRIS - 5430-44000-016-00D | Characterization of Grain Biochemical Components Responsible for End-Use Quality                |
| CRIS - 5430-44000-017-00D | Enhanced End-Use Quality and Utilization of Sorghum Grain                                       |
| CRIS - 5430-44000-018-00D | Enhancement of Hard Winter Wheat Quality and Its Utility (Hard Winter Wheat Quality Laboratory) |

**CRIS 5430-44000-016-00D**

## **Characterization of Grain Biochemical Components Responsible for End-Use Quality**

**Project Leader:** M. Tilley

**Investigators:** M. Tilley, J. Wilson, and  
vacant position

**Full-Time Scientist Equivalents (SYs):** 2.5

**Start Date:** 10/01/04

**Termination Date:** 09/30/09

**Problem:** The baking industry encounters tremendous loss in the areas of quality assurance to customers, consistency of product, processing parameters and product waste. It is estimated that as much as 10% of a company's product is relegated to waste due to lack of flour consistency. This variability in composition and quality of wheat flour is dependent upon genetic, environmental and supply chain factors. For example, not only will the protein content vary from lot to lot, but within a given protein content range (specified by end user requirements), the quality of the protein will vary considerably. Even when basic product specifications, such as ash, moisture, and protein content are met, the dough forming and baking characteristics of a given lot of flour are highly unpredictable.

Similarly, ingredient and processing variability translates into large economic losses in the baking industry. The U.S. baking industry annually uses about 42 billion pounds of flour, valued at about \$4.2 billion, to produce an estimated \$33 billion of baked goods. Variability in flour quality means that there are opportunities for large savings in the areas of raw material selection, processing, and distribution. Further, improved methodologies to better analyze the root causes of flour variability will open the door to the production of higher value products.

The baking industry has limited tools at its disposal to deal effectively with the variability of wheat flour, its major raw material. All currently utilized industry tools for assaying and/or testing flour are highly empirical, offering little insight into the biochemical basis of (a) what constitutes a "quality" flour, (b) how a particular lot of flour can be characterized for its suitability for a given application, or (c) how a particular lot of flour should be processed to maximize product quality.

**Objectives:** The objectives of this project are to: (1) Determine the roles and interactions of the major biochemical components of cereal grains (starch, storage proteins and enzymes) as they relate to food quality and functionality; (2) Define the role of the environment on functional properties of biochemical components that affect end-use properties; (3) Apply information generated in previous objectives towards development and refinement of methods to rapidly predict grain quality. Towards this end, investigation to determine role(s) of wheat starch granule size distributions on variations in functionality during bread-making will be performed. These approaches will utilize differential scanning calorimetry to provide thermal data on individual starch granule populations and starch fractions (pure A- and B- type granules) will be used for reconstitution

experiments to determine how size classes affect bread-making quality. The mechanism(s) of action of oxidative enzyme addition to flour on storage proteins and non-storage proteins will be determined by addition of enzymes followed by functional analysis and examination of protein fractions and polymers using SDS-PAGE, size exclusion HPLC, and multi-angle laser light scattering. Environmental effects on starch and storage proteins will be determined using the above approaches with samples that have been exposed to controlled temperature and irrigation regimes during development. The potential of novel microfluidic devices will be thoroughly examined as a means to develop extremely rapid and highly reproducible separation of proteins for grain cultivar identification and quality prediction.

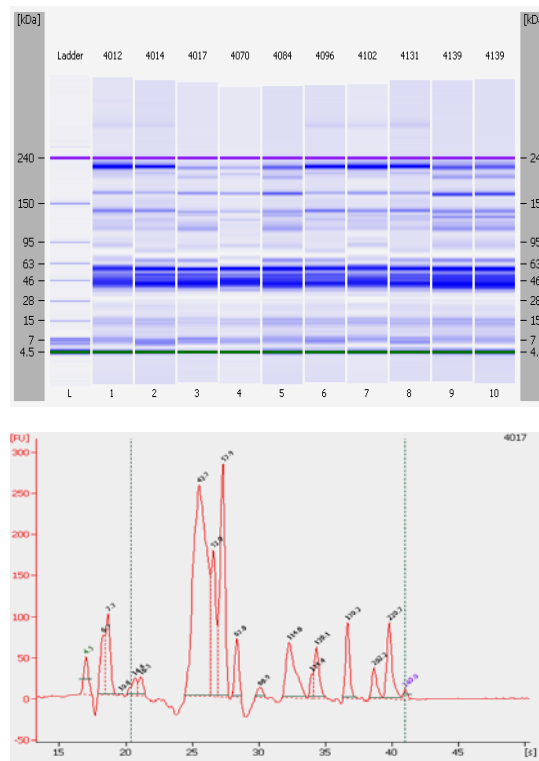
Advances in the knowledge of grain biochemical characteristics that determine the physical and functional properties critical to processing and end-product quality, and development of rapid, accurate methods to measure these quality determinants are essential to maintain a competitive position for U.S. grain in global markets. This project will provide the cereal food industry with the tools needed to define the end-use performance of cereal grains.

## Results and Impact:

### 1. Isolation of Large Quantities of Starch.

We have developed a method to isolate large amounts of starch separated into size fractions for baking and chemical testing. The sonication procedure for starch isolation has been scaled up and tested allowing us to isolate starch from up to 40 grams of starch at a time. This procedure has decreased the time it takes to isolate starch from flour from ~1 hour per sample to ~10 minutes. The correction model is currently being applied to

~ 100 HRW and 98 HRS wheat obtained from FGIS. This data is being correlated to bake quality data to better understand the importance of starch size distribution.



### 2. Microfluidics Library Development.

Knowledge of cereal proteins is important both for predicting end-use performance and for identification. The development of new or improvement of existing methods for deriving models for predicting quality traits and/or quickly identifying grain cultivars will assist in determining their roles in relation to quality. Microfluidic systems (lab-on-a-chip) produce fast separations using very small liquid volumes. We have purchased a microfluidics instrument and begun to identify quality-related protein patterns and unique proteins associated with specialty wheats, particularly waxy wheats. The effect of environment on proteins and quality might be available through analysis of HMW-GS isoforms or through quantification of

albumins and globulins. In addition, it is envisioned that the gliadin patterns, amounts of albumins and globulins, and HMW-GS isoforms of specific wheat varieties with known quality traits will be stored in a database and when one of those patterns (or combinations) is matched, it will tag the sample as a good quality sample.

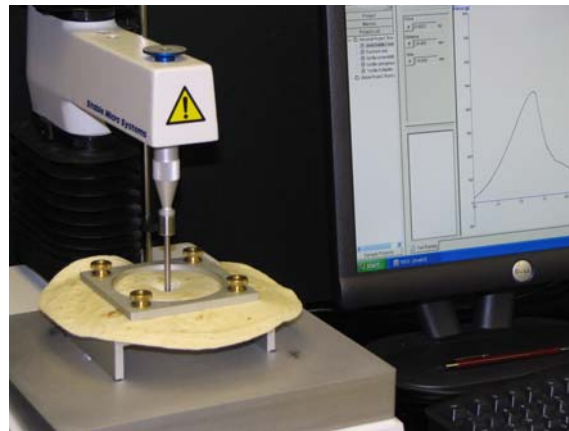
### 3. LDS Correction Model Detects Environmental Differences in Starch Ratios.

The environmental impact on the starch size distribution during grain filling of hard red winter wheat has been collected and collated. These samples were all grown at the Kansas State University Agronomy Study plots over a 6 year period of time. Statistical analysis of this data is nearing completion and correlations to some weather conditions have been noted. The larger starch size distributions (everything  $> 20 \mu\text{m}$ ) are positively correlated to increased cumulative precipitation, while the 5-20  $\mu\text{m}$  starch size distributions are negatively correlated to decreased cumulative precipitation and decreased cumulative and average evapotranspiration rates. Decreasing soil and air temperatures also appear to have a negative correlation to the 5-20  $\mu\text{m}$  starch populations, while increasing soil and air temperatures has a positive correlation to the 20-30  $\mu\text{m}$  starch populations.



### 4. Quality of Spelt Wheat and Its Starch.

Flours from 5 spelt cultivars grown over 3 years were evaluated as to their bread baking quality and isolated starch properties. The starch properties included amylose contents, gelatinization temperatures (differential scanning calorimetry), granule size distributions and pasting properties. Milled flour showed highly variable protein content and was higher than hard winter wheat, with short dough-mix times indicating weak gluten. High protein cultivars gave good crumb scores, some of which surpassed the hard red winter wheat (HRW) baking control. Loaf volume was correlated to protein and all spelt varieties were at least 10-15% lower than the HRW control. Isolated starch properties revealed an increase in amylose in the spelt starches of between 4 - 7 % over the HRW control. Negative correlations were observed for the large A-type granules to bread crumb score, amylose level, and final pasting viscosity for cultivars grown in year 1999 and to pasting temperature for samples grown in 1998. Positive correlations were found for the small B- and C-type granules relative to crumb score, loaf volume, amylose, and RVA final pasting viscosity for cultivars grown in year 1999, and to RVA pasting temperature in 1998. The environmental impact on spelt properties seemed to have a greater effect than genetic control.



## **5. Effect of Emulsifiers on Textural Properties of Whole Wheat Tortillas During Storage.**

Tortillas have become the most prevalent ethnic bread in the U.S., often replacing white pan bread. As a result, tortillas are the fastest growing segment of the U.S. baking industry with annual sales over US\$6 billion and growth exceeding 10% per year. Sustaining this tremendous demand requires sufficient definition and determination of fundamental quality characteristics of wheat flour tortillas. Three emulsifiers (SSL, GMS, de-oiled lecithin) impacted the textural quality of 100% WW tortillas during storage. However, the amount of emulsifier incorporated into the formulation was crucial. SSL was more effective at its lowest usage level (0.125%), unlike the de-oiled lecithin, which was most effective at its highest usage level (2%). The diameters of tortillas with mid (0.25%) and high (0.50%) levels of added SSL were significantly smaller than the rest of the tortillas. Rollability scores and rupture force (Fr) of tortillas were improved with emulsifier addition. Control tortillas consistently resulted in higher Fr values as well as lowest rollability scores at the end of full storage. None of the emulsifiers studied enhanced the stretchability of tortillas, as it abruptly declined during the first two days of storage. Type and level of emulsifier addition to tortillas should be determined carefully as it influences textural properties besides shelf life.

### **Goals for 2007, 2008, and 2009:**

#### **Specific tasks in 2007 will be to:**

1. Isolation of large quantities of starch, separated into size fractions for baking and chemical testing. Continued testing the correction model for starch size distributions by laser diffraction sizing (LDS).

2. Determine if particular starch granule ratios are markers for quality traits. Continue isolating the starch fractions from different environments for chemical analysis. Compare starch size distributions and chemical analysis to different environments. Use LDS and our correction model to detect environmental differences in starch ratios. Continue testing amylose/amylopectin ratios, pasting profiles, DSC temperatures and lipids of A, B, and C type starch fractions.

3. Determination of the contribution of individual glutenin subunits on enzyme mediated crosslinking in gluten functionality. Correlate information obtained on HMW-GS polymer formation with data on bread or tortilla quality characteristics.

4. Continue characterization of enzyme effects on protein interactions between glutenin and albumins. Begin investigation of effect of HMW-GS contribution of wheat on enzyme mediated crosslinking.

5. Continue investigating lab-on-a-chip system to extract, separate and identify wheat varieties. Test unknown and difficult to classify samples against the library developed. Improve extraction methodologies and separation techniques incorporating novel methods available at that time.

#### **Specific tasks in 2008 will be to:**

1. Isolation of large quantities of starch, separated into size fractions for baking and chemical testing. Continued testing the correction model for starch size distributions by laser diffraction sizing (LDS).

2. Bake studies using reconstituted gluten and starch fractions. Begin testing, amylose /amylopectin ratios, pasting profiles, differential scanning calorimetry (DSC)



temperatures and lipids of A, B, and C-type starch fractions. Chemical analysis of the starch fractions. Correlation analysis comparing bake data, starch size distributions and chemical analysis.

3. Continue characterization of effect of HMW-GS contribution of wheat on enzyme mediated crosslinking. Determine the sizes of polymeric fractions and the MW distribution of polymeric proteins from various near-isogenic lines.

4. Compare starch size distributions and chemical analysis to different environments. Proceed with analysis of starch from wheat grown in different environmental conditions to determine correlations of environmental effects on starch size distribution.

5. Relate polymer sizes and molecular weight distributions to quality characteristics, provided by the HWWQL.

6. Lab-on-a-chip will be developed to identify quality-related proteins and unique proteins associated with specialty wheats. The effect of environment on proteins and quality will be analyzed.

**Specific tasks in 2009 will be to:**

1. Complete bake studies and chemical analysis of the starch fractions. Determine correlations of starch size distribution to baking and chemical parameters.

2. Complete characterization of effect of HMW-GS contribution of wheat on enzyme mediated crosslinking.

3. Determine correlations of environmental effects on starch size distribution.

4. Determine if particular proteins are markers for quality traits and the feasibility of lab-on-a-chip system for on the spot analysis.

**Specific Cooperative Agreements for This Project Included:**

None

**Summary of 2006 Publications/Patents:**

1. Akdogan, H., Tilley, M., and Chung, O.K. 2006. Effect of emulsifiers on textural properties of whole wheat tortillas. *Cereal Chemistry*. 83(6):632-635.

2. Akdogan, H., Tilley, M., Bean, S.R., and Graybosch, R.A.. Differentiation of allelic variations of the HMW glutenin subunits of wheat flours by use of mixing parameters and polymeric protein content. *Proceedings of the 2006 Gluten Workshop* (in press)

3. Schober, T.J., Bean, S., Kuhn, M. 2006. Gluten proteins from spelt (*Triticum aestivum* ssp. *spelta*) cultivars: A rheological and size-exclusion high-performance liquid chromatography study. *J. Cereal Sci.* 44(2): 161-173.

4. Tilley, M., Akdogan, H., and Chung, O.K. Effect of ingredients on tortilla quality. *Proceedings of the First International Symposium on Cereal Science*, October 18 - 21 2006, Wuxi China. (in press)

5. Tilley, M., Pierrucci, V.R.M., Tilley K.A., Chung, O.K. 2006. Effects of Processing on Wheat Tortilla Quality: Benefits of Hard White Wheat. *Chinese J. Food Science* 11:152-158.

6. Tilley, M., Bean, S.R., and Tilley, K.A. 2006. Capillary electrophoresis for monitoring dityrosine and 3-bromotyrosine synthesis. *J. Chromatography A*. 1103:368-371.

7. Tilley, M., F.E. Dowell, O.K Chung, S.H. Park, E.B. Maghirang, B.W. Seabourn, T.C. Pearson, F. Xie, H. Akdogan, M.E. Casada, J.D. Wilson, S.R. Bean, T.J. Schober, P.R. Armstrong, M.S. Caley, D.L. Brabec, S.Z. Xiao, L.M. Seitz, R.K. Lyne, J.E. Throne, F.H. Arthur, D.B. Bechtel, G.L. Lookhart, and M.S. Ram. 2006. Wheat research in the U.S. Grain Marketing and Production Research Center. Annual Wheat Newsletter 52:158-169.

8. Wilson, J.D., Kaufman, R.C., Park, S. 2006. The environmental impact on starch size distribution in developing hard red winter wheat [abstract]. AACC International Meeting. Poster Paper No. 267.

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## Enhanced End-Use Quality and Utilization of Sorghum Grain

**Project Leader:** S. Bean

**Investigators:** S. Bean, M. Tilley, J. Wilson,  
and vacant position

**Full-Time Scientist Equivalents (SYs):** 2.5

**Start Date:** 10/01/04

**Termination Date:** 09/30/09



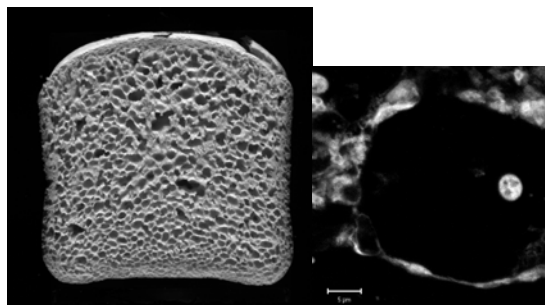
**Problem:** In recent years, sorghum production has declined in the U.S. Sorghum is a low-input, drought-tolerant crop grown in several parts of the U. S. and around the world. Sorghum is used primarily as animal feed in the U.S. (second only to maize), although 30 to 40% of worldwide production is used as human food. In 1998, the U.S. produced ~20% of the worldwide sorghum supply. Annually, 30 to 50% of the U.S. sorghum crop is exported. Therefore, new uses for sorghum could represent new markets for U.S. agriculture. In addition, the drought-tolerance of sorghum makes it attractive for future growth in areas of low water availability. Increased utilization of sorghum could serve as a tool for rural renewal in areas where sorghum is a major crop and where water is limited for production of other crops such as maize and soybeans.

Sorghum has potential for several uses including a source of renewable bio-industrial products such as ethanol, lactic acid, and biodegradable films and packaging. Sorghum

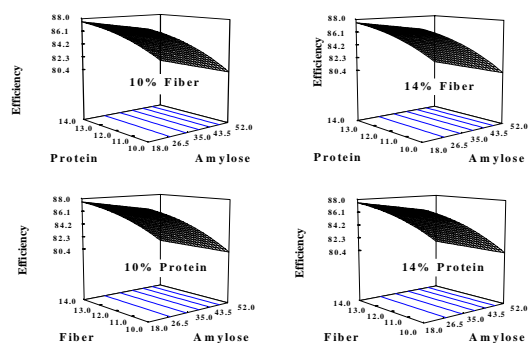
also represents a safe food for the 2-3 million people who cannot eat wheat due to celiac disease. However, several obstacles must be overcome in order to increase the utilization of sorghum. While some research directed at using sorghum in food products and industrial products (such as biodegradable films) has been carried out, comparatively little research has been conducted on the relationship between sorghum biochemistry and end-use quality and utilization.

**Objectives:** This project will focus on the relationships between sorghum biomolecules and end-use quality and utilization of sorghum with the goal of improving their functionality. Understanding these relationships will identify the components of sorghum that are responsible for end-use quality. Knowledge of these relationships will also allow for new uses of sorghum to be developed.

### Results and Impact:

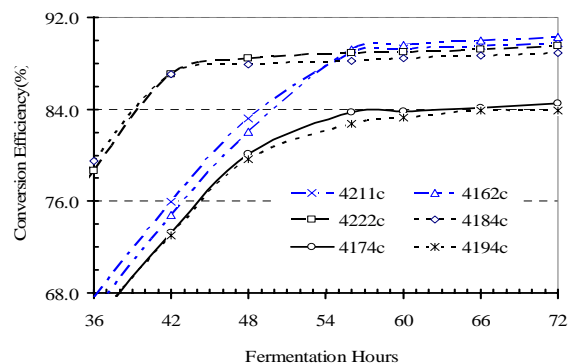


**1. Improved Sorghum Bread Quality.** It is currently estimated that there are between 2-3 million people in the U.S. alone that have celiac disease. Celiac disease requires a life long avoidance of wheat, barley, rye, and possibly oats, which severely limits food choices. Sorghum is a safe food for celiacs, however, production of high-quality bread from it is difficult. Recent research has shown that using a sourdough process significantly improves sorghum bread quality, especially crumb structure and loaf height. Sourdough fermentation results in degradation of soluble proteins. Upon baking, this prevents interference of aggregated denatured proteins with the starch gel. This study also allowed us to determine the factors necessary to produce high quality starch based wheat-free breads.



**2. Impact of Starch Composition on Ethanol Production.** The effects of amylose: amylopectin ratio, and protein and fiber contents on ethanol yields were evaluated by using artificially formulated media made from commercial corn starches with different contents of amylose, corn protein, and corn fiber, as well as different cereal sources, including corn, sorghum, and wheat with differing amylose contents. Second-order response-surface regression models were used to study the effects and interactions of amylose: amylopectin ratio, and protein and fiber contents on ethanol yield and conversion efficiency. The results showed that the amylose content of starches had a significant

( $P < 0.001$ ) effect on ethanol conversion efficiency. Neither the effect of protein and fiber contents, nor the interactions among amylose, protein, and fiber were significant at  $P = 0.05$  level. Conversion efficiencies increased as the amylose content decreased, especially when the amylose content was less than 35%. The reduced quadratic model fits the conversion efficiency data better than the full quadratic model does. Fermentation tests on mashes made from corn, sorghum, and wheat samples with different amylose contents confirmed the adverse effect of amylose content on fermentation efficiency. High-temperature cooking with agitation significantly increased the conversion efficiencies on mashes made from high-amylose (35-70%) ground corn and starches. A cooking temperature of 160 °C or higher was needed on high-amylose corn and starches to obtain a conversion efficiency equal to that of normal corn and starch.



**3. Factors Governing Ethanol Production Determined.** The goal of this research is to understand the key factors impacting ethanol production from grain sorghum. Seventy genotypes and elite hybrids, with a range of chemical compositions and physical properties selected from approximately 1200 sorghum lines, were evaluated for ethanol production, and were used to study the relationships among composition, grain structure, and

physical features that affect ethanol yield and fermentation efficiency. Variations of 22% in ethanol yield and 9% in fermentation efficiency were observed among the 70 sorghum samples. Genotypes with high and low conversion efficiencies were associated with attributes that may be manipulated to improve fermentation efficiency. Major characteristics of the elite sorghum genotypes for ethanol production by the dry-grind method include high starch content, rapid liquefaction, low viscosity during liquefaction, high fermentation speed, and high fermentation efficiency. Major factors adversely affecting the bioconversion process are tannin content, low protein digestibility, high mash viscosity, and an elevated concentration of amylose-lipid complex in the mash.



**4. Use of Ultrasound to Isolate Starch From Sorghum.** Sorghum starch plays an important role in both the production of sorghum food products and fermentation of sorghum to produce products like fuel ethanol. To improve the utilization of sorghum, it is necessary to understand the physical and chemical properties of its starch. Research on starch usually requires the use of purified starch. There are no rapid, simple methods for isolating sorghum starch available. Therefore, we investigated the use of ultrasound (sonication) as a method for sorghum starch purification. We found that under optimum conditions a high purity starch could be isolated with only 2 min of sonication. Starch could be isolated from either sorghum flour or sorghum whole meal. Other cereal starches from whole wheat meal, wheat flour, corn,

rice, and barley were also rapidly obtained using this method. This method can be used to rapidly purify starch for analytical tests and could possibly be scaled up for use as an industrial starch isolation method.

**5. Large Scale Extraction of Sorghum Proteins.** Sorghum proteins have the potential to be used as a bio-industrial renewable resource for applications such as biodegradable films and packaging. This project was designed to evaluate the effect of interactions between sorghum protein extraction and precipitation conditions on the yield, purity, and composition of sorghum protein fractions. Proteins were extracted with 70% ethanol under non-reducing conditions, with ultrasound, or under reducing conditions using either sodium metabisulfite or glutathione as the reducing agent. Several conditions were used to isolate the extracted proteins via precipitation, including lowering ethanol concentrations alone or in combination with lowering pH to 2.5, or by adding 1M NaCl to the extract. Combinations of these conditions were also tested. All precipitation conditions effectively precipitated proteins and both lowering the pH and the addition was 1M NaCl to the extracts enhanced precipitation in some cases. However, the conditions that precipitated the maximum amount of protein or highest purity of protein varied according to how the proteins were initially extracted. Precipitated proteins were characterized by RP-HPLC, SEC, HPCE, and SDS-PAGE to compare the protein fractions composition. Non-reduced and sonicated samples were found to have a much wider  $M_w$  distribution than reduced extracts. Thus, extraction and precipitation conditions influenced the isolated proteins yield, purity, and composition. Since the extraction and purification processes influenced the composition, purity, biochemical properties, it may be possible to

prepare protein fractions with unique functionalities for specific end-uses.

**Goals for 2007, 2008, and 2009:**

**Specific tasks for 2007 will be to:**

1. The effect of protein cross-linking on ethanol production from sorghum will be investigated.
2. A rapid method for measuring ergosterol content in sorghum will be developed to assist sorghum breeders in improving sorghum grain quality.
3. Visco-elastic dough formation in artificial sorghum protein-starch dough systems will be investigated and changes to sorghum proteins during mixing will be elucidated and compared to wheat proteins during mixing.

**Specific tasks for 2008 will be to:**

1. The cross-linking of sorghum proteins during kernel development will be investigated to understand how such cross-linking occurs and how the environment impacts the extent of protein cross-linking.
2. The extent of protein-protein interaction and protein-starch interaction in artificial sorghum dough systems will be determined. This work will continue to examine the factors necessary to produce visco-elastic dough from sorghum.

**Specific tasks for 2009 will be to:**

1. Methods for the use of reduction-oxidation systems to form visco-elastic dough directly from sorghum flour will be developed.
2. Sorghum proteins and starch will be modified to improve their functionality.

**Specific Cooperative Agreements for This Project Included:**

Department of Biological and Agricultural Engineering, Kansas State University, Manhattan, Kansas

Department of Biological Systems Engineering, University of Nebraska, Lincoln, Nebraska

Department of Soil and Crop Sciences, Texas A&M University, College Station, Texas

Department of Animal Sciences, University of Wisconsin, Madison, Wisconsin

Department of Pharmaceutical and Biomedical Sciences, University of Georgia, Athens, Georgia

Department of Food Science and Nutrition, Texas A&M University, College Station, Texas

**Summary of 2006 Publications/Patents:**

1. Bean, S., Chung, O.K., Tuinstra, M.R., Pedersen, J.F., Erpelding, J.E. 2006. Evaluation of the single kernel characterization system (SKCS) measurements of sorghum grain attributes. *Cereal Chem.* 83:108-113.
2. Bean, S., Ioerger, B.P., Park, S.H., Singh, H. 2006. Interaction between sorghum protein extraction and precipitation conditions on the yield, purity, and composition of purified protein fractions. *Cereal Chem.* 83:99-107.
3. Park, S. H., Bean, S. R., Wilson, J. D., and Schober, T. J. 2006. Rapid isolation of sorghum and other cereal starches using sonication. *Cereal Chem.* 83:611-616.

4. Corredor, D.Y., Bean, S.R., Schober, T.J., and Wang, D. 2005. Effect of decorticating sorghum on ethanol production and composition of DDGS. *Cereal Chem.* 83(1): 17-21.

5. Lee, K.-M., Bean, S. R., Alavi, S., Herrman, T. J., and Waniska, R. D. 2006. Physical and biochemical properties of maize hardness and extrudates of selected hybrids. *J. Agric. Food Chem.* 54: 4260-4269.

6. Schober, T. J., Bean, S. R. Kuhn, M. 2006. Combination of fundamental rheology and size-exclusion high-performance liquid chromatography in the study of gluten proteins from spelt wheat (*Triticum aestivum* ssp. *Spelta*) cultivars. *Journal of Cereal Science* 44: 161-173.

7. Seitz, L.M. 2005. 3-deoxyanthocyanidins and other phenolic compounds in grain from sorghum sister lines with white, red, and yellow pericarp. Abstract No. 270 in: 2005 AACC Annual Meeting Program Book. p.153. Meeting Abstract.

8. Wang, D., Wu, X., Bean, S., Wilson, J.P. 2006. Ethanol production from pearl millet by using *Saccharomyces cerevisiae*. *Cereal Chemistry* 83:127-131.

9. Zhan, X., Wang, D., Bean, S., Mo, X., Sun, X.S., Boyle, D. 2006. Ethanol production from extrusion-cooked sorghum flour. *Industrial Crops and Products.* 23:304-310.

10. Taylor, J., Schober, T., Bean, S. R. 2006. Non-traditional uses of sorghum and pearl millet. *J. Cereal Sci. Journal of Cereal Science* 44: 252-271.

11. Wu, X., Zhao, R., Wang, D., Bean, S. R., Seib, P. A., Tuinstra, M. R., and Campbell, M. 2006. Effects of amylose, corn protein, and

corn fiber contents on production of ethanol from starch-rich media. *Cereal Chem.* 83:569-575.

12. Park, S. H., Bean, S. R., Chung, O. K., and Seib, P. A. 2006. Levels of protein and protein composition in hard winter wheat flours and the relationships to breadmaking. *Cereal Chem.* 83:418-423.

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CRIS 5430-44000-018-00D

## Enhancement of Hard Winter Wheat Quality and Its Utility (Hard Winter Wheat Quality Laboratory)

**Project Leader:** B. Seabourn

**Investigators:** B. Seabourn, Y. Chen, and  
vacant position

**Full-Time Scientist Equivalents (SYs):** 3.0

**Start Date:** 10/01/04

**Termination Date:** 09/30/09



**Problem:** Wheat is the world's most important crop, in terms of both total acreage planted and human consumption. A new wheat cultivar requires 13-15 years to develop and is released at a cost of nearly \$1 M. Without intrinsic quality evaluation, long range breeding programs in the U.S. will face the risk of releasing a wheat cultivar of unknown quality with serious economic impact on the milling and baking industry. Quality evaluation of wheat breeding lines during the entire breeding cycle is the first step in keeping U.S. wheat competitive in world markets.

Wheat breeders urgently need information on the quality of experimental varieties that have been grown under different environments. The HWWQL is the critical link in the development of new hard winter wheat varieties in the U.S. However, this has traditionally required the breeder to spend a

| ID        | Milling Score | Rating    | Baking Score | Rating    | 1RS | Trait Deficiencies |
|-----------|---------------|-----------|--------------|-----------|-----|--------------------|
| Khakof    | 48.6          | Average   | 55.8         | Good      |     | 6.                 |
| Roughider | 46.9          | Poor      | 51.3         | Average   |     | 9.                 |
| Abilene   | 43.7          | Poor      | 62.1         | Very Good |     | 2.                 |
| Tandem    | 66.2          | Very Good | 64.5         | Very Good |     |                    |
| Crimson   | 62.0          | Very Good | 55.6         | Average   |     |                    |
| SD89180   | 59.6          | Very Good | 61.3         | Good      |     |                    |
| SD89188   | 43.5          | Poor      | 64.2         | Very Good |     |                    |
| SD89205   | 48.8          | Average   | 59.0         | Good      |     | 3.5.               |
| ND8974    | 45.6          | Poor      | 38.5         | Very Poor |     |                    |
| ND9043    | 54.9          | Good      | 50.4         | Average   |     | 10.                |
| ND9094    | 55.9          | Good      | 45.6         | Poor      |     |                    |
| ND9257    | 52.9          | Average   | 61.4         | Very Good |     |                    |
| ND9272    | 33.1          | Very Poor | 50.1         | Average   |     | 3.                 |
| ND9274    | 32.7          | Very Poor | 54.1         | Average   |     | 2.4.               |
| NE91631   | 34.1          | Very Poor | 44.2         | Poor      |     | 11.                |
| NE91648   | 50.6          | Average   | 40.9         | Very Poor |     |                    |
| NE90479   | 60.4          | Very Good | 56.3         | Good      |     | 5.6.               |
| NE92522   | 49.7          | Average   | 41.1         | Very Poor |     |                    |
| NE92578   | 56.3          | Good      | 46.7         | Poor      |     |                    |

large amount of time evaluating raw quality data. Therefore, the HWWQL developed a simple, user-friendly relational database system to quickly summarize and interpret end-use quality data. The database offers tremendous flexibility in that it allows the breeder to change the algorithms by which the database evaluates and interprets end-use quality. Thus, the breeder can “define” quality according to his/her end-use product or targeted customer preference. Food manufacturers and other customers can also use the information in this database, which is updated on a yearly basis, to determine which varieties possess the quality traits needed to make each of the products that they produce.

**Objectives:** Recent changes in the international marketing of wheat require



simple, inexpensive, fast, and accurate tests to enable buyers to identify wheat samples with specific end-use properties at the first point of sale. The current wheat marketing system is primarily based on physical rather than intrinsic end-use quality. In order to survive stiff competition in the global wheat market, it is imperative for U.S. producers to deliver a crop with end-use properties desired by buyers (both domestic and international), and for producers to be rewarded for the quality of their crop in addition to yield. Therefore, developing rapid, accurate and objective end-use quality predictors for the breeding community and the grain industry is one of our main objectives. Quality prediction systems are being developed using near-infrared (NIR) reflectance/transmittance spectral data combined with milling/dough/bread quality data. In addition, in order to expand the usage of hard winter wheat into non-bread products, such as Asian noodles and tortillas, we are developing small-scale testing methods for noodles and tortillas to evaluate early generation breeding lines with the ultimate goal of predicting non-bread product quality. Development of new cultivars that contain desirable intrinsic end-use qualities is a long and costly process. The development of rapid, accurate, and objective end-use quality predictors for the grain industry was identified in the 1990 Food Security Act as essential to maintaining U.S. competitiveness in world grain markets. Domestic commercial bakeries could save considerable time and expense if an on-line quality monitoring system could be incorporated into the dough formulation and mixing process. Therefore, one goal of this project is to extend our knowledge of the interaction of the basic components (protein, starch, lipid, and water) in a flour water system, and to thus provide a more rapid, objective, and reliable means for determining the end use performance of a given flour sample. This new information will ultimately

facilitate the increased utilization of U.S. wheat flour for new and unique commercial products, including Asian noodles and/or other products such as tortillas or pizza crust.



Wheat bread is a major commodity in the world. Millions of loaves of bread per day are produced in automated bakeries. Since dough is made in 2,000 pound batches, if the dough cannot be used due to variations in its properties, such as stickiness or shorter mixing time requirement, the bakery would have to throw out that batch and clean the entire system at a cost of about \$10,000. If the problem is not found until the bread comes out of the oven, then the cost rises to \$132,000 per batch due to the number of batches that are in the system when the problem is found. These costs include costs associated with utilities, cleanup, and lost product. The mixing of dough, as it is practiced commercially or in the quality control laboratory, has been largely one of visual and tactile observations of the dough mass as it changes in color and texture, consistency, and stickiness, or its response to work input via the mixograph, farinograph, or some other imitative device. Thus, current practices are largely subjective in nature. A more objective method for measuring the rheology of dough systems, preferably one based on physical-chemical aspects of dough component interactions, have long been desired by cereal chemists. Therefore, rapid methods to predict wheat flour quality, without

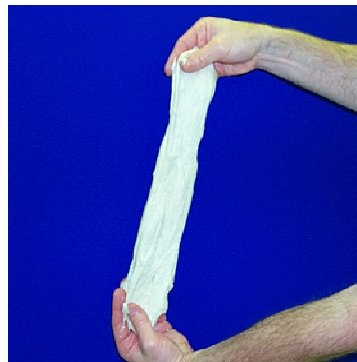
expensive and time-consuming tests such as test baking are urgently needed by the baking industry. Furthermore, rapid prediction methods for breadmaking quality would enhance the U.S. export of hard winter wheats because international buyers of U.S. wheat want to know what quality they are receiving prior to purchase.

One of the most important quality parameters to the baking industry is bake water absorption requirement for bread formulation. Research to determine the environmental effects on and rapid methods to accurately evaluate the water absorption potential of flour is needed so industry can optimize the formulation of their product.

## **Results and Impact:**

**1. Program Component.** Work at the HWWQL includes the evaluation of intrinsic end-use quality of hard winter wheat progenies; the development of rapid methods for quality differences in bread, noodles, and tortillas; and development of prediction models. The development of accurate end use quality prediction techniques is vital as we move from a marketing system based on U.S. government price supports to one where end use quality traits establish the value of crops in a global market. This project addresses several problems regarding the identification and determination of quality parameters that are key to ensuring the value of U.S. grain.

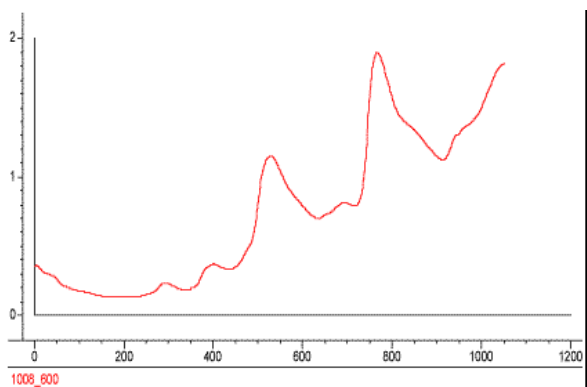
**2. Dough Functionality (Rheology).** (1) The mixing of dough, as it is practiced commercially or in the quality control laboratory, has been largely one of visual and tactile observations of the dough mass as it changes in color and texture, consistency, and



stickiness, or its response to work input via the mixograph, farinograph, or some other imitative device. Thus, current practices are largely subjective in nature. A more objective method for measuring the rheology of dough systems, preferably one based on physical-chemical aspects of dough component interactions, have long been desired by cereal chemists. Fifty-five hard red winter wheat flours with varying protein contents and mixing times were scanned with an infrared spectrometer after being mixed with a mixograph for 1 minute. End-use milling and baking properties for each sample were evaluated. Changes in protein structure during 1 minute of mixing were highly correlated to mixograph mix time, dough tolerance, bake mix time, and loaf volume potential. Statistical analysis showed that 73%, 81%, and 70% of the total variance in mixograph mix time, mixing tolerance, and bake mix time could be explained by the relationship between protein structure and these parameters, respectively. These results demonstrate the potential of infrared spectroscopy to make rapid and predictive determinations of dough functionality, which would be very useful in the wheat breeding lab as a screening tool, or in an online bakery process.



**3. Program Component.** Work at the HWWQL includes the evaluation of intrinsic end-use quality of hard winter wheat progenies; the development of rapid methods for quality differences in bread, noodles, and tortillas; and development of prediction models. The development of accurate end use quality prediction techniques is vital as we move from a marketing system based on U.S. government price supports to one where end use quality traits establish the value of crops in a global market. This project addresses several problems regarding the identification and determination of quality parameters that are critical to ensuring the value of U.S. grain.



**4. NIR Calibration.** Calibrations for Asian noodle color prediction were revised to include the '05 crop year data from the HWWQL. The results of this work yielded

slightly improved calibrations for noodle color prediction at 24 hours. It is expected that these calibrations will continue to improve with the continued accumulation sample data; we only have a few crop years of data for this new test within the HWWQL. We also continued development of NIR calibrations for flour ash, which has been difficult to do the narrow range of ash values for the advanced experimental lines typically seen in the HWWQL. This work also yielded improved calibrations. Continued work with these equations will involve incorporating data from various millstreams to add more variation and range to the calibration set. These improved results demonstrate the potential of infrared spectroscopy to make rapid and predictive determinations of flour quality, which would be very useful in the wheat breeding lab as a screening tool, or in a bakery quality control lab to fully identify raw material quality characteristics prior to processing.

**5. Frozen Dough Quality.** The use of frozen dough has seen tremendous growth in the last 30 years due to the benefit of producing a fresh baked product while saving time, equipment, and labor costs. However, it has been observed that frozen dough does not have the same (poorer) functionality that they had before freezing. Therefore, the confirmed results from the study above were used to investigate the resultant changes in end-use functionality wheat flour dough of varying quality as a result of freezing and thawing. Infrared measurements were made on eight optimally mixed flour-water doughs using hard red winter wheat flours of low, medium, and high protein content and short, medium, and long optimum mixing time before and after freezing. Our results indicated that protein structures associated with optimum flour quality decreased as result of freezing. With a method to measure these subtle changes in dough quality, bakers can make

intelligent decisions toward mitigating detrimental changes frozen dough quality.

**Goals for 2007, 2008, and 2009:**

**Specific tasks in 2007 will be to:**

1. Study both genetic and environmental effects on protein and lipid contents and composition with breeders samples grown at various locations.
2. Determine by FTIR and Raman spectroscopy the influence of covalent bonding on dough strength, specifically disulfide bonds within and between polypeptide chains as well as bonds between amino acid side-chains in the gluten polymer (e.g. tyr-tyr bonds).
3. Screen breeder samples for PPO, noodle color, and noodle-making quality; establish texture analyzing processes for raw and cooked noodles.
4. Begin to develop small scale rheological methods and analyze the molecular size distribution of tortilla dough proteins to predict end-use properties of tortillas.
5. Continue the mandated activity of the HWWQL with evaluation of end use quality of wheat breeding lines. Standard tests will be conducted to evaluate wheat quality and end use performance such as physical grain tests, milling, mixing, and baking tests, in addition to characterization of biochemical quality determinants. We will continue to play a major role in Wheat Quality Council activities and collaborate with the U.S. Wheat Associates, various state wheat commissions, Kansas State University, and the American Institute of Baking in evaluating the quality of U.S. hard winter wheat for both domestic and export customers. Upon the availability of

increased resources, we will expand quality testing for non bread products as requested by our customers. The fundamental wavelengths responsible for end use performance will be incorporated into existing NIR instrumentation for use in the commercial industry. Work on understanding the changes that occur in proteins, starches, and lipids during dough formation will continue.

6. Continually add graphic images of the mixograph curve for each wheat line in the HWWQL database for the next release of the database, as well as an interface for user-selected statistical analyses; a new, online real-time version of the database will be launched for even easier access to the HWWQL data via the world-wide web; we will study both genetic and environmental effects on end-use quality and biochemical components (composition) with breeders samples grown at various locations; continue with the instrumental analyses of the interaction of the various chemical components in flour during dough formation. The prediction models of wheat quality factors will be fine-tuned using several cereal testing instruments linked to the biochemical data from the MU's sister CRIS.

**Specific tasks in 2008 will be to:**

1. Investigate the effect of variation in kernel hardness and weight in a wheat sample on milling and baking properties and continue to study the relationships between wheat physical characteristics and end-use properties, which may be used to segregate wheat based on quality.
2. Study protein-starch, protein-lipid, and starch-lipid interactions in a model system during mixing, fermentation, and baking stages.

3. Determine the influence of various standard ingredients on dough rheology by FTIR; screen breeder samples for PPO, noodle color, and eating quality, based on a texture analysis of raw and cooked noodles.

4. Continue to provide early generation information to breeders that predict the tortilla making properties of wheat.

**Specific tasks in 2009 will be to:**

1. Complete prediction models for quality attributes desired by breeders and industry; apply mid-IR information on dough rheology to the development of near-IR measurement of important dough rheology parameters during mixing; develop an online "real-time" dough monitoring system to be used by the HWWQL and baking industry.

2. Develop NIR calibration models for rapid measurement of PPO and noodle-making quality in breeder and commercial samples.

3. Complete prediction models for tortilla rheology and quality attributes desired by breeders and industry, and incorporate these rapid measurements into our routine measurements of breeding program samples.

4. After prediction models are successfully tested and introduced into breeding programs, and if project length has not run its course, controlled blending of varieties to determine the effect(s) of blended wheat on the correlation with wet chemistry methods vs. NIR will be investigated in order to simulate samples typically encountered in commercial production and marketing.

**Specific Cooperative Agreements for This Project Included:**

Department of Grain Science and Industry,  
Kansas State University, Manhattan, Kansas

**Summary of 2006 Publications/Patents:**

1. Akdogan, H.P. and Chung, O.K. 2005. Effects of different emulsifiers on the textural properties and shelf-stability of 100% whole wheat flour tortillas. Abstract No. 309 Page 163 in: Program Book of the 90th Annual Meeting of the AACC. [Abstract]

2 Carver, B., Hunger, R., Klatt, A., Edwards, J., Porter, D.R., Verchot-Lubicz, J., Martin, B., Seabourn, B.W., Rayas-Duarte, P. 2006. Registration of 'Deliver' wheat. Crop Sci. p.1819-1820.

3 Carver, B.F., Smith, E.L., Hunger, R.M., Klatt, A.R., Edward, J.T., Porter, D.R., Verchot-Lubicz, J., Rayas-Durate, P., Martin, B.C., Krenzer, E.G. 2006. Registration of 'Ednurance' wheat. Crop Science. 46:1816-1817. Available: <http://crop.scijournals.org/content/vol46/issue>.

4 Chung, O.K., Park, S., Bean, S., Xiao, Z.S. 2005. Relationships between cooked alkaline noodle texture and solvent retention capacity (SRS), SDS-sedimentation, mixograph, and protein composition. Abstract No. 227 in: 2005 AACC Annual Meeting Program Book. p.141. Meeting Abstract.

5. Seibel, W., Chung, O.K., Weipert, D., Park, S. 2006. Cereals and cereal products. Encyclopedia of Agriculture.

6. Park, S., Seabourn, B.W., Xie, F., Chung, O.K. 2005. Prediction of alkaline noodle color and polyphenol oxidase activity using near-infrared reflectance (NIR) spectroscopy of wheat grain, meal and flour. Abstract No. 224 in: 2005 AACC Annual Meeting Program Book. p.140. Meeting Abstract.

7. Haley, S.D., Quick, J.S., Johnson, J.J., Peairs, F.B., Stromberger, J.A., Clayshulte, S.R., Clifford, B.L., Rudolf, J.B., Seabourn, B.W., Chung, O.K., Jin, Y., Kolmer, J.A. 2005. Registration of 'hatcher' wheat. Crop Science. Vol. 45: 2654-2655.

8. Xie, F., Seabourn, B.W., Chung, O.K., Seib, P.A. 2005. Study of wheat gluten secondary structure conformational changes in frozen dough using FT-HATR Mid-Infrared Spectroscopy.. Abstract No. 245 in: 2005 AACC Annual Meeting Program Book. p.146. Meeting Abstract.

**For More Information on This Project Contact:**

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# PLANT SCIENCE AND ENTOMOLOGY RESEARCH UNIT

The mission of the Plant Science and Entomology Research Unit is to support the U.S. wheat industry by developing and providing genetic solutions to economically important biotic and abiotic stress problems in hard winter wheat. We will focus on resistance or tolerance to several recalcitrant or emerging problems that impact grain production and/or quality. In addition, the Unit hosts a Cereal Crop Genotyping Laboratory that will provide marker-assisted selection research and services to public and private wheat breeders. The specific research project for this Unit is:

CRIS - 5430-21000-005-00D

Genetic Enhancement for Resistance to Biotic and Abiotic Stresses in Hard Winter Wheat

**CRIS 5430-21000-005-00D**

## **Genetic Enhancement for Resistance to Biotic and Abiotic Stresses in Hard Winter Wheat**

**Project Leader:** R. Bowden

**Investigators:** G. Bai, R. Bowden, M. Chen, J. Fellers, Z. Ristic, and vacant position

**Full-Time Scientist Equivalents (SYs):** 6.0

**Start Date:** 07/19/03

**Termination Date:** 07/18/08

**Problem:** In the USA, winter wheat (both hard and soft types) is planted on approximately 35 million acres annually and the farm gate value is approximately 4 billion dollars. In the central and southern Great Plains region, winter wheat is the most important crop and many rural communities are highly dependent upon it. Approximately 50% of wheat produced in this region is shipped internationally and supports a large wheat export industry. In addition to its value as a grain crop, wheat is also used as an important cool season pasture crop for the livestock industry.

Yields of wheat in the central and southern Great Plains are typically less than half of those achieved in more favorable environments, such as the Pacific Northwest. The difference between actual and attainable yields is due to multiple biotic and abiotic stress factors. In addition to yield losses, these stresses also cause losses in grain quality. Host plant resistance or tolerance to these biotic and abiotic stresses is the most economical and environmentally sound method of stabilizing and increasing wheat yields and protecting grain quality in this challenging environment.

**Objectives:** The goal of this project is to develop and provide genetic solutions to several economically important biotic and

abiotic stress problems in hard winter wheat. The focus is on several recalcitrant and/or emerging problems. The specific objectives of this project are: 1) Enhance resistance of wheat to rusts; 2) Enhance resistance of wheat to Hessian fly; 3) Enhance resistance of wheat to Karnal bunt, 4) Enhance resistance of wheat to Fusarium head blight, 5) Enhance wheat post-anthesis heat tolerance; and 6) Develop improved methods of marker-assisted selection of wheat.

### **Results and Impact:**

**1. Wheat May Have Both Resistance and Susceptibility Genes for Fusarium Head Blight.** We identified several quantitative trait loci (QTLs) for Fusarium head blight (FHB) resistance and the molecular markers closely linked to the QTLs in an experimental wheat line with chromosome 7A from resistant cultivar Sumai 3 substituted into the variety Chinese Spring. A total of 10 QTLs were identified with significant main effects on FHB resistance. These QTLs appeared to condition both resistance and susceptibility to FHB. We propose that adding FHB enhancing QTLs or removal of susceptibility QTLs both may significantly enhance the degree of wheat resistance to FHB in a wheat cultivar.



**2. Gene Expression Dramatically Different in Plants Resistant or Susceptible to Hessian Fly.** We identified genome-wide alterations in gene expression in wheat plants during compatible and incompatible interactions with Hessian fly. One thousand, one hundred and nineteen genes were found to be consistently either up- or down-regulated during compatible and/or incompatible interactions. Functional analysis of the genes with known functions revealed that the genes encoding proteins involved in direct antibiotic defense and the genes encoding enzymes involved in the phenylpropanoid, cell wall, and lipid metabolism were the major targets for differential regulation during compatible and incompatible interactions. Our results indicated that a combination of the enhancement of antibiotic defense, the evasion of nutrient metabolism induction, and the fortification and expansion of the cell wall, are likely the collective mechanism for host plant resistance observed during incompatible interactions. In contrast, induced susceptibility including the suppression of antibiotic defense, the induction of nutrient metabolism, the weakening of cell wall, and the inhibition of plant growth appeared to be the necessary conditions for Hessian fly virulence during compatible interactions.

**3. Molecular Markers for New Hessian Fly Resistance Gene.** H22, a major Hessian fly resistance gene in wheat, has been mapped to the distal region of wheat chromosome 1DS. Tightly linked markers have been identified. These markers can be used for map-based cloning of the H22 gene and can also be used for marker-assisted selection.

**4. Novel Hessian Fly Gut Proteins.** A group of genes that encode small secreted proteins from the gut of Hessian fly have been isolated and characterized. However, the exact functions of these proteins are not yet known

since no sequence similarity could be identified between these proteins and any known sequences in public databases. Their abundance suggests that these proteins perform important functions in the gut of this insect and therefore might be a useful target for developing new control measure in the future.

**5. New Method of Gene Enrichment Found.** A new method for genome sequencing of gene rich regions of DNA was developed. By using methyl sensitive restriction enzymes, DNA clones were enriched for low copy DNA, which contains most of the gene coding regions of the genome. This method may help accelerate genome sequencing efforts for several organisms, including wheat.

**6. Heat Tolerance in Wheat Correlates with Chaperone Protein.** We obtained evidence that suggests that chloroplast protein synthesis elongation factor, EF-Tu, may play a role in heat tolerance in wheat. EF-Tu has been shown to function as a “chaperone” protein that protects other proteins from degradation during heat stress. Twelve cultivars of winter wheat were assessed for heat tolerance and relative endogenous levels of EF-Tu under heat stress conditions. The results showed that heat stress induces accumulation of EF-Tu in mature plants (plants at flowering stage) of all 12 cultivars. However, the cultivars that show greater accumulation of EF-Tu under heat stress, display better heat tolerance.

**7. Possible New Type of Aluminum Tolerance.** On acid soils, aluminum toxicity can be a limiting factor in wheat yields. We screened 600 wheat lines from Asia and the USA and identified 70 cultivars with at least moderate tolerance to aluminum toxicity. Half of them show the susceptible allele of the aluminum-activated malate release (*ALMT1*)

gene. Among them one Chinese landrace, FSW, showed no expression of *ALMT1* and no malate release into culture solution. This finding suggested that aluminum tolerance in FSW may not be due to malate release, a well-recognized mechanism of aluminum tolerance, and other mechanisms may be involved in tolerance in FSW.

**8. Progress Made Toward Cloning Durable Rust Resistance Gene *Lr46*.** The gene *Lr46* has provided partial resistance to wheat leaf rust and has been durable for many years in spring wheat cultivars. We located *Lr46* on a small segment of wheat chromosome 1B. Molecular markers were found closely flanking *Lr46*. These markers will be useful for continued progress in cloning this interesting gene. They may also be useful for marker assisted selection.

**Goals for 2007, 2008, and 2009:**

**Specific tasks in 2007 will be to:**

1. Continue to evaluate introgression breeding populations with major gene resistance to wheat leaf rust in the field.
2. Continue to evaluate introgression breeding populations with minor gene adult plant resistance to leaf rust and stripe rust in the field and greenhouse. Minor gene adult plant resistance is often more durable.
3. Continue to develop molecular markers for resistance to rust.
4. Complete studies and submit manuscripts on gene expression studies of durable rust resistance genes.
5. Continue work on avirulence genes in leaf rust. Develop a system to test the avirulence function of rust haustorial secreted proteins.

6. Initiate introgression breeding program for new sources of resistance to wheat stem rust into hard red and hard white winter wheat. These new sources provide resistance against exotic races of stem rust.

7. Determine global expression patterns of genes encoding Hessian fly salivary gland proteins using microarray and real-time-PCR technologies.

8. Screen wheat breeding lines from cooperating breeding programs for resistance to Hessian fly.

9. Continue to evaluate introgression breeding populations with resistance to Karnal bunt.

10. Screen wheat breeding lines from cooperating breeding programs for resistance to Karnal bunt.

11. Continue marker-assisted backcross breeding for resistance to Fusarium head blight with regional breeding programs.

12. Determine heat tolerance of selected wheat lines from ICARDA and begin characterization of key proteins related to heat tolerance.

13. Continue to collect genotyping data in collaboration with Wheat CAP (Coordinated Agricultural Project).

14. Develop a mapping population for a new source of aluminum tolerance in wheat. Aluminum toxicity is a major limiting factor in acid soils.

15. Complete QTL analysis of two populations for pre-harvest sprouting tolerance. Sprouting tolerance is crucial for the introduction of hard white winter wheat varieties.

**Specific tasks in 2008 will be to:**

1. Release 1 or more germplasms with major gene resistance to leaf rust. These will be used by wheat breeders to increase resistance to leaf rusts.
2. Release 1 or more germplasms with minor gene adult plant resistance to rust. These can be used by wheat breeders to increase resistance to leaf rusts.
3. Continue work on avirulence genes in leaf rust. Test the avirulence function of rust haustorial secreted proteins.
4. Continue introgression breeding program for new sources of resistance to wheat stem rust into hard red and hard white winter wheat.
5. Genotype new mapping populations for stem rust resistance genes from hard winter wheat.
6. Determine the differences in the expression profile of the genes encoding Hessian fly salivary gland proteins during compatible and incompatible interactions.
7. Screen wheat breeding lines from cooperating breeding programs for resistance to Hessian fly.
8. Release 1 or more germplasms with resistance to Karnal bunt.
9. Screen wheat breeding lines from cooperating breeding programs for resistance to Karnal bunt.
10. Complete phenotyping and genotyping of the new mapping populations with resistance to Fusarium head blight.

11. Continue marker-assisted backcross breeding for resistance to Fusarium head blight with regional breeding programs.

12. Continue characterization of key proteins related to heat tolerance in selected wheat lines from ICARDA.

13. Continue to collect genotyping data in collaboration with Wheat CAP (Coordinated Agricultural Project).

14. Submit manuscripts on pre-harvest sprouting tolerance mapping.

**Specific tasks in 2009 will be to:**

1. Submit manuscript on the avirulence function of leaf rust haustorial secreted proteins.

2. Release germplasm lines with several new sources of resistance to stem rust in hard red and hard white winter wheat backgrounds.

3. Identify molecular markers associated with new stem rust resistance genes.

4. Submit manuscript on the molecular basis for Hessian fly resistance or susceptibility during compatible and incompatible interactions.

5. Screen wheat breeding lines from cooperating breeding programs for resistance to Hessian fly.

6. Generate new crosses with wild wheat relatives that contain Hessian fly resistance.

7. Release 1 or more germplasms or cultivars with resistance to Karnal bunt.

8. Screen wheat breeding lines from cooperating breeding programs for resistance to Karnal bunt.
9. Complete analysis on the new mapping populations for resistance to Fusarium head blight and write manuscript.
10. Complete analysis of heat tolerance in selected lines from ICARDA.
11. Phenotype and genotype RIL population of FSW/Wheaton for aluminum tolerance.

**Specific Cooperative Agreements for This Project Included:**

Department of Entomology, Kansas State University, Manhattan, Kansas

Department of Plant Pathology, Kansas State University, Manhattan, Kansas

Department of Agronomy, Kansas State University, Manhattan, Kansas

Department of Agricultural Economics, Kansas State University, Manhattan, Kansas

Department of Biology, Kansas State University, Manhattan, Kansas

Experiment Station, Texas A&M University, College Station, Texas

Department of Soil and Crop Sciences, Texas A&M University, College Station, Texas

Department of Plant & Soil Sciences, Oklahoma State University, Stillwater, Oklahoma

Center for Maize and Wheat Improvement (CIMMYT), Mexico D.F., Mexico

**Summary of 2006 Publications/Patents:**

1. Bernardo, A., Bai, G., Guo, P., Xiao, K., Guenzi, A., Ayoubi, P. *Fusarium graminearum* - induced differential gene expression between Fusarium head blight-resistant and susceptible wheat cultivars. *Functional and Integrative Genomics*.
2. Jia, G., Chen, P., Qui, G., Bai, G., Wang, X., Wang, S., Zhou, B., Zhang, S., Liu, D. 2005. QTLs for Fusarium head blight response in a wheat DH population of Wangshuibai/Alondra's'. *Euphytica* 146:183-191.
3. Lamoureux, D., Patterson, D.G., Li, W., Gill, B.S., Fellers, J.P. 2005. The efficacy of cot-based gene enrichment in wheat (*Triticum aestivum*). *Genome*. 48:1120-1126.
4. Ma, H., Bai, G., Zhou, L., Carver, B. 2005. Molecular mapping of a quantitative trait locus for aluminum tolerance in wheat cultivar Atlas 66. *Journal of Theoretical and Applied Genetics*. 112:51-57.
5. Ma, H., Bai, G., Zhang, X., Lu, W. 2006. Main effects, epistasis and environmental interactions of QTLs for Fusarium head blight resistance in a recombinant inbred population. *Phytopathology* 96:534-541.
6. Maddur, A.A., Liu, X., Zhu, Y., Fellers, J.P., Oppert, B.S., Park, Y., Bai, J., Wilde, G.E., Chen, M. Cloning and characterization of protease inhibitor-like cDNAs from the Hessian fly [*Mayetiola destructor* (Say)]. *Insect Molecular Biology* 15: 485-496.
7. Marza, F., Bai, G., Carver, B. 2005. QTLs for yield and related traits in the wheat population, Ning 7840 x Clark. *Theoretical and Applied Genetics*.

8. Webb, C., Szabo, L.J., Bakkeren, G., Garry, C.E., Staples, R.C., Fellers, J.P. 2006. Transient expression and insertional mutagenesis of *Puccinia triticina* using biolistics. *Functional and Integrative Genomics*.
9. Webb, C.A., Fellers, J.P. 2006. Cereal rust genomics and the pursuit of virulence and avirulence factors. *FEMS Immunology and Medical Microbiology*.
10. Wu, Y., Taliaferro, C., Bai, G., Martin, D., Anderson, J., Anderson, M., Edwards, R. 2006. Genetic analyses of Chinese *Cynodon* accessions by flow cytometry and AFLP markers. *Crop Science* 46:917-926.
11. Xu, X., Bai, G., Carver, B., Shaner, G., Hunger, R. 2006. Molecular characterization of a powdery mildew resistance gene in wheat cultivar Suwon 92. *Phytopathology*. 96:496-500.
12. Xu, X., Bai, G., Carver, B., Shaner, G.E. 2005. A QTL for early heading date in wheat cultivar Suwon 92. *Euphytica*.
13. Yang, J., Bai, G., Shaner, G. Novel QTLs for *Fusarium* head blight resistance in wheat cultivar Chokwang. *Theoretical and Applied Genetics*. 111:1571-1579.
14. Yu, J., Bai, G., Cai, S., Ban, T. 2006. Marker-assisted characterization of Asian wheat lines for resistance to *Fusarium* head blight. *Theoretical and Applied Genetics*. DOI 10.1007/500122-006-0297-2.
15. Brooks, S.A., Huang, L., Herbel, M.N., Gill, B.S., Brown Guedira, G.L., Fellers, J.P. 2006. Structural variation and evolution of a resistance gene island in natural populations of wild wheat. *Theoretical and Applied Genetics*.
16. Liu, X.M., Brown Guedira, G.L., Hatchett, J.H., Owuoche, J.O., Chen, M. 2005. Genetic characterization and molecular mapping of a Hessian fly resistance gene transferred from *T. turgidum ssp. dicoccum* to common wheat. *Journal of Theoretical and Applied Genetics*.
17. Liu, X., Fellers, J.P., Zhu, Y., Mutti, N.S., El-Bouhssini, M., Chen, M. 2006. Cloning and characterization of cDNAs encoding carboxypeptidase-like proteins from the gut of Hessian fly [*Mayetiola destructor* (Say)] larvae. *Insect Biochemistry and Molecular Biology*. 36:665-673.
18. Lobo, N.F., Behura, S.K., Aggarwal, R., Chen, M., Collins, F.H., Stuart, J.J. 2006. Genomic analysis of a 1 mb region near the telomere of Hessian fly chromosome x2 and avirulence gene *vh13*. *Biomed Central (BMC) Genomics*.
19. Ma, H., Bai, G., Lu, W.Z. 2006. Quantitative trait loci for aluminum resistance in wheat cultivar Chinese Spring. *Plant and Soil Journal* 283:239-249.
20. Ma, H., Zhang, K., Gao, L., Bai, G., Chen, H., Cai, Z., Lu, W. 2006. QTL for the resistance to wheat *Fusarium* head blight and deoxynivalenol accumulation in Wangshuibai under field conditions. *Plant Pathology*. DOI:10.1111/J.1365-3059.2006.01447.x.
21. Mateos-Hernandez, M., Singh, R.P., Hulbert, S.H., Bowden, R.L., Huerta-Espino, J., Gill, B.S., Brown Guedira, G.L. 2006. Targeted mapping of ESTs linked to the adult plant resistance gene *Lr46* in wheat using synteny with rice. *Functional and Integrative Genomics*. 2006 March 6(2):122-131.

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# **WIND EROSION RESEARCH UNIT**

The mission of the Wind Erosion Research Unit is to increase understanding of wind erosion processes; develop reliable predictive tools; develop control practices; and disseminate information and technology for sustaining agriculture, protecting the environment, and conserving natural resources.

**Research Activities:**

**CRIS 5430-11120-007-00D**

**Particulate Emissions From Wind Erosion:  
Processes, Assessment, and Control**

**Project Leader:** E. Skidmore

**Investigators:** E. Skidmore, L. Hagen, L. Wagner, and J. Tatarko

**Full-Time Scientist Equivalents (SYs):** 4.0

**Start Date:** 09/01/02

**Termination Date:** 08/31/07

**Problem:** Particulate emissions from wind erosion continue to threaten the sustainability of agriculture and the maintenance of environmental quality. Total suspended particles (TSP) in the air is commonly used as a measure of dust emissions. Annual fine particle emissions from wind erosion are estimated at 5,400 tons. Dust emissions from eroding fields enter suspension and become part of the atmospheric dust load. These emissions obscure visibility, pollute the air, create traffic hazards, foul machinery, and imperil animal and human health. Deposition of wind-blown sediments in drainage pathways and on water bodies significantly deteriorates water quality. The EPA has set National Air Quality Standards for fine particle concentrations in the air.

Accomplishment of the above stated objectives will provide a more reliable scientific-basis for improving prediction of particulate emissions into the atmosphere, developing soil-, climate-, and crop-specific control strategies, and assessing erosion damage and environmental impact. Beneficiaries from this technology will include: State and Federal regulatory agencies, those making national resource inventories, conservation planners, those formulating policies relating to conservation reserve programs and global change, land managers, and in-effect all global inhabitants.

**Results and Impact:**

**Objectives:** This threat is being resolved by accomplishing objectives to: 1) Improve Wind Erosion Prediction System (WEPS) for cropland and extend it to range, forest, and disturbed lands. 2) Increase understanding of particulate emissions from wind erosion processes. 3) Couple WEPS with appropriate databases to inventory dust emissions including PM-10 over large areas. 4) Develop new and evaluate viable practices for reducing dust emissions from wind erosion and incorporate findings into WEPS.

**1. Progress on the Wind Erosion Prediction System (WEPS) 1.0.** The latest and most cutting-edge model for forecasting wind erosion damage is a step closer to reaching growers and landowners in the wind-prone regions of the country. WEPS 1.0 can simulate weather, soil and crop conditions, and wind erosion on a daily basis. It can also project the emission of the tiny dust particles referred to as PM-10 that may pose risks to human health and the environment. Using WEPS 1.0, individual farmers will be able to formulate specific wind erosion control practices. The software can guide growers to



the right approach—whether it's establishing a soil-stabilizing crop cover, establishing wind breaks and barriers or reducing soil's erodibility by appropriate tillage.

**2. Action Plan for Implementing Recommendations of an Expert Review.** A panel of experts assessed the Units programs in regard to: relevance, scientific merit, capacity, and national/international leadership. After review of the scientific accomplishments, publications and current research program of WERU, the panel recommended several things that would increase the scientific merit and capacity of the Unit in harmony with agency mission. The Unit has responded with the development of an Action Plan for implementing recommendations. These are summarized in an Action Plan Summary table. Some of the recommendations extend beyond the Unit to ARS and NRCS agencies.

**Goals for 2007, 2008 and 2009:**

**Specific tasks in 2007 will be to:**

1. Develop new and evaluate viable practices for reducing dust emissions from wind erosion and incorporate findings into WEPS.
2. Determine best management practices (BMPs) to reduce the rate of roughness degradation.
3. Develop BMPs to maintain the crop residue's ability to reduce wind erosion.
4. Evaluate products to reduce dust emissions.
5. Couple WEPS with appropriate databases to inventory dust emissions including PM-10 over large areas. Collaborate with WSU scientists who are trying to incorporate WEPS

into their regional AIRPACT model.

6. Compare Zobeck soil database and Fryrear sieving data to soil and Management Submodel predictions.
  7. Publish WEPS 1.0 Technical documentation as an official USDA handbook.
  8. Continue to update WEPS 1.0 science and interface technical documentation as needed based on model changes.
  9. Certify WEPS 1.0 works for the thirty high priority core crops.
  10. Schedule training and testing of WEPS outside the Great Plains.
  11. Determine surface friction velocity as influenced by vertical and horizontal distribution of biomass.
  12. Concentrate on objectives and tasks on Project Plan 5430-11120-007-00D (termination date 08/31/2007) that are not yet fully accomplished, develop next OSQR project plan and start accomplishing tasks identified on next plan.
  13. Evaluate field data to determine the effectiveness of applied bio-solids on soil wind erodibility.
- Specific tasks in 2008 will be to:**
1. Conduct field-scale evaluation of quantitative process-level modules to evaluate management alternatives for semi-arid cropping systems.
  2. Modify WEPS so that it can be used as a Dust Event Prediction Warning System (DEPAWS).

3. Combine wind erosion prediction and water erosion prediction models.
4. Determine what fractions of dust are PM 10 and PM 2.5 from different soil types.
5. Identify minimum data necessary to run WEPS.
6. Develop fine-scale wind parameters maps to incorporate into WEPS..

**Specific tasks in 2009 will be to:**

1. Determine soil aggregate status as influenced by weather, intrinsic soil properties, land use, vehicular and animal traffic.
2. Determine threshold friction velocities as a function of soil temporal properties, and determine size distribution (including PM10 and PM2.5) of eroding aggregates created by erosion processes (entrainment of loose soil, clod/crust abrasion, and breakage of saltation-size aggregates) on bare soils as a function of temporal and intrinsic soil properties.
3. Items not accomplished in 2007 and 2008 that are still relevant are identified and resources obligated as appropriate.
4. High priority items identified by customers that are relevant to Unit's mission.

**Specific Cooperative Agreements for This Project Include:**

Department of Agronomy, Kansas State University, Manhattan, Kansas

**Summary of 2006 Publications/Patents:**

None

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