

Western Extension/Education Research Activity - WERA 066

Integrated Management of Russian Wheat Aphid and other Cereal Arthropod Pests

Report Information

Annual Meet Date: 2/25/2009

Period Covered: 2/2008 to 2/2009

Next Meeting: September 2010, Ft. Collins, CO

Meeting Participants

Colorado State University:	Tom Holtzer
Kansas State University:	Phil Sloderbeck
New Mexico State University:	Bryan Fontes
North Dakota State University:	Kirk Anderson
Oklahoma State University:	Tom Royer
Texas Agrilife Extension:	Ed Bynum
	Roxanne Fegley
USDA-ARS Stillwater, Oklahoma:	Cheryl Baker
	Barbara Driskel
	Do Mornihnweg
	Gary Puterka
	Kevin Surfran
	Jim Webster
USDA-ARS West Lafayette, Indiana:	Christie Williams
ARC-SGI Bethlehem, South Africa	Vicki Tolmay

Minutes of Meeting February 25, 2009

WERA 066 was held in conjunction with the joint meeting of the South West Branch of the Entomological Society of America and the Society of south West Entomologists, which ran from February 23-26 in Stillwater, OK. The session took place in the Wes Watkins Center, room 101, on the campus of Oklahoma State University.

1:30 pm – Business Meeting

Chair Tom Royer called the meeting to order and began with a round of introductions by those in attendance. Tom then read the minutes of the 2008 meeting that was held in Ft. Collins, CO. Phil Sloderbeck moved that the minutes be approved and Do Mornhinweg seconded the motion, which then carried.

Tom Holtzer, the WERA 066 Administrative Advisor, informed the group that a mid-term review is due this year. The report will focus on outcomes and impacts of this group. This report will be prepared by Chair Tom Royer and Secretary Christie Williams.

1:55 pm – Discussion and Oral Summaries of State Reports

North Dakota - Kirk Anderson

Several surveys were conducted during the past two years. The 2007 IPM Survey focused on grain aphids, grasshoppers, cereal leaf beetle, wheat stem maggot and barley thrips. The use of insecticide was a key factor in preserving the high quality of barley so that farmers could sell the crop for malting rather than as lower quality feed barley.

In addition, 2008 surveys were done for orange wheat blossom midge, wheat stem saw fly, plus other insects and diseases. The 2008 Hessian fly Survey characterized the utility of a five-component blend of sex-pheromones to determine geographic distribution, seasonal abundance and number of generations per year. Results indicated that the Hessian fly is distributed state-wide. Populations peaked in midsummer and again in September, indicating a lack of aestivation. The fall emergence occurred after the suggested wheat planting date (fly-free date).

A general discussion about Hessian fly followed. Researchers from several states reported using or having interest in using the Hessian fly pheromones to characterize populations. Phil Sloderbeck said that, similar to North Dakota, in Kansas Hessian fly flights were later in the fall than expected, after the fly-free date for planting wheat. Some years, such as 2008, wheat must be planted late due to weather conditions. At the Colorado-Kansas border Hessian fly populations seemed high, probably due to two factors: higher than normal rainfall in August causing sprouting of volunteer wheat that serves as insect refuge and continuous no-till wheat, without rotation or fallow, that allows survival. Hessian fly hadn't been seen in that region for over 20 years. Pyrethroids were effective in controlling Hessian fly. A new flyer on Hessian fly is on Phil's web site <http://www.oznet.ksu.edu/library/entml2/MF2866.pdf>.

Kirk Anderson mentioned that in North Dakota some durum wheat cultivars that were never intentionally bred for Hessian fly-resistance are quite resistant. Similar observations were made in Oklahoma. Christie Williams notes that durum wheats have been a strong source of resistance genes that have been introgressed into common wheat in the Purdue breeding program (added to the minutes).

It was mentioned that the fly-free dates that were first promoted in the 1920s may not be accurate today. Hessian flies have been observed to fly even in cold weather. It was mentioned that a fall infestation of Hessian fly may not necessarily lead to high spring infestations or crop damage. This is believed to be because Hessian flies damage the tiller on which they reside and tillers that form later are normal unless a secondary infestation occurs. A short discussion on Hessian fly population movement followed. It was reported that an edge effect can be observed in a field if adjacent to a no-till field with high populations. But Kirk Anderson and Christie Williams mentioned that Hessian flies are not strong flyers so don't migrate far, but could be dispersed by wind. Marion Harris has a paper published on this topic.

Oklahoma – Do Mornhinweg, Cheryl Baker and Gary Puterka
Fifty Russian Wheat Aphid-resistant barley lines have been released to date. In collaborative research QTL analysis identified two major and one minor locus

contributing to resistance, and mapping of resistance genes is being pursued. Differentials for biotype 1 and 2 are being pursued. Studies are ongoing to monitor RWA biotype diversity throughout the US and identify zones where RWA may become holocyclic. Parasitoids of greenbug have been identified.

A general discussion followed, concerning whether different RWA biotypes were distinct introductions or whether the same biotypes spontaneously evolved in multiple locations.

Texas – Roxanne Fegley and Ed Bynum

Greenbug infestations are now at levels that warrant treatment. Also, RWA and Hessian fly have been found in Texas. A study of perennial grasses offers an opportunity to monitor movement and virus vectoring of wheat curl mite.

Indiana – Christie Williams

Several molecular studies are underway. The Shukle lab is characterizing genes encoding Hessian fly secreted salivary gland proteins, believed to be effectors that induce plant susceptibility as well as avirulence proteins that induce resistance. The sequence diversity of these genes is greater in Israeli populations than in the US. These Israeli genes may be ancestral types.

The Shukle and Williams labs have done a transmission electron microscopy study showing that midgut microvilli are disrupted in avirulent Hessian fly larvae feeding for just three hours on resistant wheat plants. After six hours of feeding the microvilli are absent. The microvilli do not deteriorate in virulent larvae and larvae starved on filter paper. Thus, the resistant wheat plant quickly responds and inflicts damage to avirulent larvae.

The Williams lab has identified several genes encoding lectins that may disrupt the Hessian fly larval midgut. These lectins target high-mannose glycoproteins that are believed to be common in insect midgut. Virulent Hessian fly larvae alter the physiology of their host plants, causing them to produce amino acids and polyamines that are beneficial to the insects. In addition their data supports findings by the Harris lab that has shown that epidermal cells of susceptible plants degrade when under attack. Virulent larvae are able to cause loss of cutin in the cuticle of wheat leaves at feeding sites and suppress production of mRNA encoding lipid transfer genes. These changes are thought to lead to loss of cell integrity and general increase in permeability that delivers nutrients to the larvae.

The Stuart lab is using a map-based approach to locate Hessian fly genes that allow them to be virulent on wheat containing resistance genes *H6*, *H9* and *H13*. These genes have been mapped to 80- to 120-kb segments on chromosomes X1 and X2. Associative mapping with Hessian fly populations from five southeastern states identified diagnostic markers, visualized on agarose gels, for flies virulent to these genes and supports the gene-for-gene basis of interaction with wheat.

4:40 pm - Identification of Secretary and Location for Next Meeting

Gary Puterka was nominated by Do Mornhinweg he and accepted. The current secretary, Christie Williams will become the next chair. Kevin Surfran moved that Ft. Collins Co be the location of the 2010 WERA 066 meeting and Phil Sloderbeck seconded the motion, which then carried.

4: 48 pm - Adjournment

Phil Sloderbeck moved the meeting be adjourned and Do Mornhinweg seconded the motion, which then carried.

**Western Extension/Education Research Activity WERA 066
Integrated Management of Russian Wheat Aphid
and Other Cereal Arthropod Pests
State Reports - February 2009**

**Department of Bioagricultural Sciences and Pest Management
Colorado State University, Fort Collins, Colorado**

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WERA-066 Objectives Addressed

Develop of integrated management strategies for cereal aphid and other arthropod pests in small grains to improve economic viability of small-grain cropping systems while maintaining environmental quality.

Facilitate research into improved integrated pest management approaches at the field and landscape level to manage cereal arthropod pests in the western U.S.

Coordination of biological control, host plant resistance and cropping system research including evaluation of natural enemy performance and resistant cultivars alone and in combination in order to identify complementary management systems.

Coordinate research in genetics, genomics, physiology, taxonomy, and ecology of arthropod pests and their natural enemies that aid in implementing integrated management strategies in diverse agricultural systems.

Enhance efficient development of resistant varieties by coordinating the identification, monitoring, and characterization of Russian wheat aphid and Hessian fly biotypes.

Current Accomplishments

Biological control

Pitfall traps were established at three cropping systems sites. Spiders are being identified. A manuscript on carabids was published. (Objectives A, B2)

Uniform aphid natural enemy observations are taken at all three locations (Objectives A, B2)

Plant Resistance

Russian wheat aphid biotype 1-resistant wheat cultivars are now planted on more than 25% of Colorado's wheat acreage. The level of use has remained constant even though RWA-1 seems to have been largely replaced by RWA-2. (Objectives A, B3)

Winter wheat lines with the 2414-11 resistance source continue to be advanced. Lines with other sources are in earlier stages of development. (Objectives A, B1)

Resistant feed barley varieties were tested on-farm from 2006 to 2008. 'Stoneham' and 'Sidney' are resistant to known RWA biotypes and generally have performed better than 'Otis', their recurrent parent, under very dry conditions and in the presence of Russian wheat aphid. (Objectives A, B1)

Surveys were conducted to determine the presence of Dn4-virulent Russian wheat aphids. Russian wheat aphid remained scarce in 2008, but the percentage of samples containing Dn-4 virulent

aphids has continued to increase since the initial survey in 2004 (98% in 2008). No virulence to 2414-11 was detected. (Objectives A, B3)

Biology and Management

Dryland cropping systems studies are ongoing at three locations in eastern Colorado. Stoneham, a RWA-resistant feed barley, has been added to some rotations. Generally, rotations have been modified to incorporate more forages, and sunflower has been eliminated. (Objectives A, B1, and B2)

Aphid flights were monitored at four locations by means of suction traps. Trap catches were extremely low but higher than 2007 (Objectives A, B2)

High elevation noncultivated grass hosts were surveyed for a fifth season. Overwintering Russian wheat aphids appear to be widespread in noncultivated grasses at higher elevations, but their relationship to wheat production is unknown. (Objectives A, B2, and B3)

Several new insecticide treatments were tested against Russian wheat aphid in wheat and barley. Cobalt (Dow Ag Sciences) was equal or superior to the standard Lorsban treatment for the second year in a row. A foliar neonicotinoid product (Actara, Syngenta) and a ketoenol (Ultor, Bayer) did not perform well but might have a place in the malt barley market. (Objective A)

A foliar neonicotinoid product (Actara, Syngenta) and a ketoenol (Ultor, Bayer) were tested against Russian wheat aphid in barley. Aphid abundance was too low to assess the efficacy of these products. (Objective A)

Publications

Haley, S.D., J.J. Johnson, F.B. Peairs, J.S. Quick, J.A. Stromberger, J.D. Butler, H.R. Miller, E.E. Heaton, J.B. Rudolph, B.W. Seabourn, G. Bai, Y. Jin, J.A. Kolmer and X. Chen. 2008. Registration of 'Bill Brown' Wheat. *J. Plant Registrations* 2: 218-223.

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Randolph, T. L., S. C. Merrill, and F. B. Peairs. 2008. Reproductive rates of Russian wheat aphid (Hemiptera: Aphididae) Biotypes 1 and 2 on a susceptible and a resistant wheat at three temperature regimes. *J. Econ. Entomol.* 101: 955 - 958.

Miller, H. R. and F. B. Peairs. 2008. Ground beetles (Coleoptera: Carabidae) in Colorado dryland cropping systems. *Southwestern Entomologist* 33: 31 - 42.

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Giles, K., G. L. Hein, and F. B. Peairs. 2008. Areawide pest management of cereal aphids in dryland wheat systems of the Great Plains, USA. Pp. 441 - 466 in: Koul, O., G. Cuperus, and N. Elliott, eds., *Areawide Pest Management: Theory and Implementation*. CAB International. Cambridge, MA.

Merrill, S. C., F. B. Peairs, H. R. Miller, T. L. Randolph, J. B. Rudolph, and E. E. Talmich. 2008. Reproduction and development of Russian wheat aphid Biotype 2 on crested wheatgrass, intermediate wheatgrass, and susceptible and resistant wheat. *J. Econ. Entomol.* 101: 541-545.

Weiland, A. A., F. B. Peairs, T. L. Randolph, J. B. Rudolph, S. D. Haley, and G. J. Puterka. 2008. Biotypic diversity in Colorado Russian wheat aphid (Hemiptera: Aphididae) populations. *J. Econ. Entomol.* 101: 569-574.

Peairs, F. B., J. B. Rudolph, T. L. Randolph, and S. Merrill. 2008. 2007 Colorado field crop insect management research and demonstration trials. Colorado State Univ. Agric. Exp. Sta. Tech. Rep. TR08-06, 34 pp.

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Analyzing the diversity of secreted salivary gland transcripts in Hessian fly populations from Israel and the United States

Alisha J. Johnson (Ph.D. student, Purdue University), Ming-Shun Chen (USDA-ARS Research Entomologist, Manhattan, KS), Richard H. Shukle (USDA-ARS Research Entomologist, West Lafayette, IN)

The salivary glands and midgut of the larval Hessian fly are the primary interfaces with wheat. Little is known about the roles of these two organs in the interactions between larval Hessian fly and wheat. However, secreted salivary gland proteins (SSGPs) in the larval Hessian fly are hypothesized to be the effectors reprogramming host-plant tissues in compatible interactions with susceptible wheat and to be the avirulence gene products eliciting resistance in incompatible interactions with resistant wheat. Initial comparison of the transcripts encoding SSGPs in populations of Hessian fly from Israel has revealed greater diversity than is present in populations from the United States. Some of these divergent SSGPs in Israeli populations fall basal in phylogenetic analyses to clades containing families of SSGPs, suggesting these divergent SSGPs may represent ancestral types. Through comparative analyses, bioinformatics, and functional analyses we seek to gain insight into the possible evolution of these SSGP effectors and their roles in the interactions between larval Hessian fly and wheat.

Ultrastructural changes in the midgut of Hessian fly larvae feeding on resistant wheat

Richard H. Shukle (USDA-ARS Research Entomologist, West Lafayette, IN), Subhashree Subramanyam (Postdoctoral Associate, Purdue University), Christie E. Williams (USDA-ARS Research Molecular Biologist, West Lafayette, IN)

The Hessian fly is present in all the wheat producing regions of the United States and is the most important insect pest in the southeastern soft-winter-wheat region. Genotypes of the pest that can overcome formerly resistant wheat continue to appear and pose a threat to wheat production. There is a need to better understand the mechanisms by which resistant plants are able to prevail over larval attack. The midgut is one of the major interfaces between the larval Hessian fly and its host plant. The goal of the present study was to determine if ultrastructural changes occur in the midguts of larvae feeding on resistant wheat compared to larvae feeding on susceptible wheat and larvae experiencing starvation while removed from the plant. Results have revealed that within three hours of initiating feeding on resistant wheat midgut microvilli were disrupted and after six hours microvilli were absent. These microvilli disruptions observed in Hessian fly larvae feeding on resistant wheat were similar to those occurring in midgut microvilli of *Drosophila* larvae fed on a diet containing 1% wheat germ agglutinin. These results suggest the midgut is a major target of toxic plant compounds such as lectins that may play a pivotal role in resistance.

Characterization of wheat plant processes manipulated by Hessian fly larvae

Subhashree Subramanyam (Postdoctoral Researcher, Purdue University, West Lafayette, IN), Kurt Saltzmann (USDA-ARS Postdoctoral Researcher, West Lafayette, IN), Jill Nemacheck (USDA-ARS Biological Research Technician, West Lafayette, IN), Ming-Shun Chen (USDA-ARS, K-State Univ.), James Clemmens (Purdue University), Christie Williams (USDA-ARS Research Molecular Biologist, West Lafayette, IN)

Interactions of first-instar Hessian fly larvae with resistant wheat plants result in induction of plant defenses. One component of this defense response is the production of lectins that can function as feeding deterrents or antinutritional proteins when ingested by larvae. Wheat lectin genes that are responsive to avirulent Hessian fly larvae were cloned into expression vectors the proteins were harvested from *E. coli*. One of the lectins has been used in glycan binding arrays demonstrating its high affinity for high-mannose glycoproteins. This class of glycoproteins is common to larval midguts, suggesting that the lectin functions to disrupt the integrity of digestive membranes. Immunodetection indicated that the lectin increases in abundance once resistant plants are challenged with larvae and that larvae ingest the lectin. Messages encoding class III peroxidases also increased in abundance in resistant plants suggesting a role in resistance for genes involved in production of reactive oxygen species.

Interactions of first-instar Hessian fly larvae with susceptible wheat plants result in alteration of plant processes that enhance the host environment for the insect. Increased abundance of amino acids essential to insect development and production of polyamines are two examples of processes manipulated by virulent Hessian fly larvae. In addition,

the leaf cuticle rapidly loses cutin, which serves as a structural component and protective surface. This decrease in cutin occurs at the same time, and possibly due to suppressed transcription of lipid transfer protein genes. Consequently, regions of the leaf hosting virulent Hessian fly larvae increase in permeability and nutrients are delivered to the larvae.

Hessian fly-resistance genes

Steven S. Xu (USDA-ARS, North Dakota State Univ.), Marion O. Harris (North Dakota State Univ.), Christie Williams (USDA-ARS Research Molecular Biologist, West Lafayette, IN)

Previously published papers indicated that Hessian fly-resistance genes *H26* and *H32* reside in similar locations within the wheat genome. An integrated mapping approach demonstrated that *H26* and *H32* are either alternative alleles or tightly linked resistance genes that are effective against slightly different larval genotypes. Molecular markers were developed that will be useful in introgressing these genes into cultivars.

Publications since February 2007

1. Shukle, R. H., Mittapalli, O., Morton, P. K., Chen, M. S. 2009. Characterization and expression analysis of a gene encoding a secreted lipase-like protein expressed in the salivary glands of the larval Hessian fly, *Mayetiola destructor* (Say). *Journal of Insect Physiology*, 55; 104-111.
2. Shukle, R. H., Yoshiyama, M., Morton, P.K., Johnson, A. J., and Schemerhorn, B.J. 2008. Tissue and developmental expression of a gene from Hessian fly encoding an ABC-active-transporter protein: Implications for Malpighian tubule function during interactions with wheat. *Journal of Insect Physiology*, 54(1); 146-154.
3. Mittapalli, O. and Shukle, R. H. 2008. Molecular characterization and responsive expression of a defender against apoptotic cell death homologue from Hessian fly, *Mayetiola destructor*. *Comparative Biochemistry and Physiology Part B*, 149; 517-523.
4. Mittapalli, O., Neal, J. J., and Shukle, R. H. 2007. Antioxidant defense response in a galling insect. *Proceedings of the National Academy of Sciences (USA)*; 104(6); 1889-1894.
5. Mittapalli, O., Sardesai, N., and Shukle, R. H. 2007. cDNA cloning and transcriptional expression of a peritrophin-like gene in the Hessian fly, *Mayetiola destructor* (Say). *Archives of Insect Biochemistry and Physiology*, 64; 19-29.
6. Mittapalli, O., Neal, J. J., and Shukle, R. H. 2007. Tissue and life stage specificity of glutathione S-transferase expression in the Hessian fly, *Mayetiola destructor*: Implications for resistance to host allelochemicals. *Journal of Insect Science*, 7; article 20.
7. Tarver, M. R., Shade, R. E., Shukle, R. H., Moar, Muir, W. M., Murdock, L. M., and Pittendrigh, B. P. 2007. Pyramiding of insecticidal compounds for control of the cowpea bruchid (*Callosobruchus Maculatus* F.). *Pest Management Science*, 63(5); 440-446.

8. Giovanini MP, Saltzmann KD, Puthoff DP, Gonzalo M, Ohm HW Williams CE. 2007. A novel wheat gene encoding a putative chitin-binding lectin is associated with resistance against Hessian fly. *Mol. Plant Path.* 8:69-82. 2007.
9. Saltzmann KD, Giovanini MP, Zheng, C, Williams CE. 2008. Virulent Hessian fly larvae manipulate the free amino acid content of host wheat plants. *J. Chem. Ecol.* 34:1401-1410.
10. Subramanyam S, Smith DF, Clemens JC, Webb MA, Sardesai N, Williams CE. 2008. Functional characterization of HFR-1, a high-mannose N-glycan-specific wheat lectin induced by Hessian fly larvae. *Plant Physiology* 147:1412-1426.

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Progress toward understanding Hessian fly biotypes.

It has long been hypothesized that single *Avirulence* (*Avr*) genes in the Hessian fly determine the virulence and avirulence of Hessian fly biotypes to specific Hessian fly resistance (*R*) genes in wheat. Taking a map-based approach to test this hypothesis, we have attempted to position the *Avr* mutations that condition virulence to *R* genes *H6*, *H9*, and *H13* in wheat. Using backcross populations and recombinant inbred lines, we positioned *Avr* gene *vH9*, which corresponds to *R* gene *H9*, to a 120-kb segment of the genome near the telomere of the short arm of Hessian fly chromosome X1. Using the same approach, we positioned *Avr* gene *vH13*, which corresponds to *R* gene *H13*, to an 80-kb segment of the genome near the telomere of the short arm of Hessian fly chromosome X2. We then performed association mapping using Hessian fly populations collected in Georgia, Alabama, South Carolina, North Carolina, and Florida to determine if the positions of the genes could be further resolved. Results suggest that the mutations that caused virulence to both *H9* and *H13* can be easily observed on agarose gels and may serve as diagnostic markers for Hessian flies that are virulent to these genes.

Results clearly indicate that Hessian fly virulence and avirulence to specific resistance genes in wheat results from mutations in single genes. Thus, the wheat-Hessian fly interaction clearly appears to have a gene-for-gene basis.

The Hessian fly physical map is available as a Hessian fly Web FPC:
<http://genome.purdue.edu/WebAGCoL/Hfly/WebFPC/>.

Publications

- Andersson, MN, J Haftmann, JJ Stuart, S Cambron, MO Harris, SP Foster, S Franke, W Francke, Y Hilbur. 2008. Identification of sex pheromone components of the Hessian fly, *Mayetiola destructor*. J Chem. Ecol. DOI 10.1007/s10886-008-9569-1.
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Kansas

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PROJECTS:

Greenbug Resistance Studies

J. C. Reese, L. R. Campbell, Ken Kofoid, G. R. Reeck and Harold Trick.

Twelve sorghum germplasm lines tolerant to greenbug feeding damage were released in late 2007. However, KAES never announced this release. The manuscript describing these materials has been submitted to J. Plant Registrations, but is not accepted yet. We also have plans to test transgenic wheats in the coming months.

Inheritance and Categories of Resistance in Wheat to Russian Wheat Aphid (Hemiptera: Aphidiade) Biotype 1 and Biotype 2.

Shah A. Khan, Miramuthu Murugan, Sharon Starkey, Aurora Manley, and C. Michael Smith.

Emergence of Russian wheat aphid, *Diruaphis noxia* (Kudjumov), biotype 2 (RWA2) in Colorado has made all known *Dn* genes, except the *Dn7* gene from rye, *Secale cereale*, vulnerable and has warranted exploration for sources of resistance to RWA1 and RWA2. Antibiosis resistance to RWA1 and RWA2 was identified in the wheat breeding line KS94H871. Additional experiments indicated that tolerance and antixenosis are not operating in KS99H871. Segregation studies involving F₂-derived F₃ families indicated that KS99H871 resistance to RWA1 is controlled by one dominant gene and one recessive gene, whereas resistance to RWA2 is controlled by only one dominant gene. This new genetic resource may serve as a good source of resistance in future breeding programs with proper understanding of the genetics of resistance.

Feeding Behavior of Russian Wheat Aphid (Hemiptera: Aphididae) Biotype 2 in Response to Wheat Genotypes Exhibiting Antibiosis and Tolerance Resistance.

Sonia Lazzari, Sharon Starkey, John Reese, Andrea Ray-Chandler, Raymond McCubrey, and C. Michael Smith.

Kansas

Wheat genotypes containing the *Dnx*, *Dn7*, *Dn6*, and *Dn4* genes for resistance to the Russian wheat aphid, *Diuraphis noxia* (Kurdjumov), along with *Dn0*, a susceptible control, were assessed to determine the categories of *D. noxia* biotype 2 (RWA2) resistance in each genotype and RWA2 feeding behaviors on *Dnx* and *Dn0* plants by using the electronic penetration graph technique. At 14 d post-infestation, *Dn0* plants exhibited intense chlorosis and leaf rolling, and all test genotypes expressed some degree of chlorosis and leaf rolling, except *Dn7*, which was not damaged. Both *Dn7* and *Dnx* expressed antibiosis effects, significantly reducing the numbers of aphids on plants and the intrinsic rate of aphid increase. *Dn6* plants appeared to contain tolerance, exhibiting tolerance index measurements for leaf and root dry weight and plant height that were significantly lower than those of the susceptible *Dn0* plants. Principal component analyses indicated that antibiosis and leaf rolling data explained 80% of the variance among genotypes. Electronic penetration graph analysis demonstrated contrasting results between RWA1 and RWA2 phloem sieve element phase feeding events, but results indicated that *Dnx* resistance factors are present in the sieve element cells or phloem sap. Plants containing *Dnx* exhibit antibiosis resistance to *D. noxia* RWA2 similar to that in plants containing the rye-based *Dn7* gene without the negative baking quality traits associated with *Dn7*.

Global Phylogenetics of an Invasive Aphid Species: Evidence for Multiple Invasions into North America.

Xiang Liu, Jeremy L. Marshall, Petr Stry, Owain Edwards, Gary Puterka, L. Dolatti, Mustapha El Bouhssini, Joyce Malinga, Jacob Lage, and C. Michael Smith.

Critical to the study of an invasive species is understanding the number of invasions that have occurred, as well as the rate or potential of post-invasion adaptation and geographic range expansion. One virulent, invasive insect species that has caused much damage in the United States is the Russian wheat aphid, *Diuraphis noxia* (Kurdjumov). Past research on *D. noxia* has suggested that up to eight biotypes, defined based on their ability to damage different wheat and barley genotypes, have diverged and radiated across the western United States from a single, common ancestral invasion in 1986. The goal of our study was to address the basic question of “are all biotypes of *D. noxia* the by-product of a single invasion or multiple invasions into North America?”. We utilized the genome-wide technique of amplified fragment length polymorphisms (AFLPs), in combination with collections of *D. noxia* from around the World, to assess this question, as well as patterns of genetic divergence. We found that there were at least two invasions into North America, each resulting in subsequent post-invasion diversification that has since yielded multiple biotypes.

Categories of Resistance in Barley Against Russian Wheat Aphid, *Diuraphis noxia* Biotypes 1 and 2.

Miriamuthu Murugan, Shah A. Khan, Sharon Starkey and C. Michael Smith.

The emergence of Russian wheat aphid, *Diruaphis noxia* (Kudjumov), biotype 2 (RWA2) in Colorado has made all known *Dn* genes vulnerable except the *Dn7* gene from rye, *Secale cereale*, and has warranted exploration for sources of resistance to RWA1 and

Kansas

RWA2. The mechanism of resistance with the resistant donor plants is considered important in terms of their influence to exert selection pressure over the aphid population for selection into new virulent population. We report tolerance and antibiosis categories of resistance to RWA1 and RWA2, in the barley cultivar Stoneham. The rate and degree of expression of resistance by Stoneham against RWA1 and RWA2 though not similar, is greater than Sidney, which showed partial resistance. Antixenosis was not apparent in either Sidney or Stoneham against either RWA1 or RWA2. Tolerance in Stoneham, expressed as reduced tissue dry weight loss and reduced tolerance index values, indicated that cultivation of Stoneham will delay the chances of RWA biotype selection. The reactions of Stoneham to RWA2 indicate that it is a good source of donor for future resistance breeding strategies against RWA.

Interactions Among Biological Control, Cultural Control and Barley Resistance to the Russian Wheat Aphid in Colorado, Kansas and Nebraska.

Paola Sotelo, C. Michael Smith, Frank B. Peairs, Terri Randolph, and Gary L. Hein.

This research is being conducted to determine the interaction among biological control, cultural control and barley resistance to the Russian wheat aphid resistance in fields located in Colorado, Kansas, and Nebraska. The experimental design is a split-plot design with two main plot treatments (early, and delayed planting dates). Within each main treatment plot, four split-plot treatments (varieties) are randomized. In 2007, these treatments included two new *D. noxia* biotype 1 and 2 resistant barley cultivars, Stoneham and Sydney, and the susceptible cultivar Otis under triamethoxam-protected and unprotected regimes. Aphid, natural enemy and incidence sampling was conducted at four dates from late May through early July in 2007. Differences in the mean number of aphids were detected by location and variety during the second sampling, when unprotected Otis plants contained the highest numbers of aphids in Colorado and Kansas. On the third sampling date, unprotected Otis plants in Nebraska contained the highest numbers of aphids. There were complex interactions between the natural enemies observed in the three fields and these are being subjected to further analyses.

Affect of Carbon/nitrogen metabolism and allocation on plant-parasite interactions. Zhu, L., Liu, X.M., Liu, X., Jeannotte, R., Reese, J.C., Harris, M., Stuart, J.J., and Chen, M.S.

Carbon/nitrogen (C/N) metabolism and allocation within the plant have important implications for plant-parasite interactions. Many plant parasites manipulate the host by inducing C/N changes that benefit their own survival and growth. Plant resistance can prevent this parasite manipulation. We used the wheat-Hessian fly (*Mayetiola destructor*) system to analyze C/N changes in plants during compatible and incompatible interactions. The Hessian fly is an insect but shares many features with plant pathogens, being sessile during feeding stages and having avirulence (*Avr*) genes that match plant resistance genes in gene-for-gene relationships. Many wheat genes involved in C/N metabolism were differentially regulated in plants during compatible and incompatible interactions. In plants during compatible interactions, the content of free carbon-containing compounds (C-compounds) decreased 36%, whereas the content of free nitrogen-containing compounds increased 46%. This C/N shift was likely achieved

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through a coordinated regulation of genes in a number of central metabolic pathways including glycolysis, the tricarboxylic acid cycle, and amino acid synthesis. Our data on plants during compatible interactions support recent findings that Hessian fly larvae create nutritive cells at feeding (attack) sites and manipulate host plants to enhance their own survival and growth. In plants during incompatible interactions, most of the metabolic genes examined were not affected or down-regulated.

Analysis of Transcripts and Proteins Expressed in the Salivary Glands of Hessian Fly

Chen, M.S., Zhao, H.X., Zhu, Y.C., Scheffler, B., Liu, X.M., Liu, X., Hulbert, S., and Stuart, J.J.

Hessian fly (HF) (*Mayetiola destructor*) larvae are thought to manipulate host growth and metabolism through salivary secretions. However, the transcriptome and proteome of HF salivary glands have not been systematically analyzed. In this research, we analyzed Expressed-Sequence-Tags (EST) representing 6,106 cDNA clones randomly selected from four libraries made from dissected salivary glands. We also analyzed the protein composition of dissected salivary glands using one- and two-dimensional gel electrophoresis as well as LC-MS/MS analysis. Transcriptomic analysis revealed that approximately 60% of the total cDNA clones and 40% of assembled clusters encoded secretory proteins (SP). The SP-encoding cDNAs were grouped into superfamilies and families according to sequence similarities. In addition to the high percentage of SP-encoding transcripts, there was also a high percentage of transcripts encoding proteins that were either involved directly in protein synthesis or in house-keeping functions that provide conditions necessary for protein synthesis. Proteomic analysis also revealed a high percentage of proteins involved in protein synthesis either directly or indirectly. The high percentage of SP-encoding transcripts and high percentage of proteins related to protein synthesis suggested that the salivary glands of HF larvae are indeed specialized tissues for synthesis of proteins for host injection. However, LC-MS/MS analysis of 64 proteins did not identify any SPs corresponding to the cDNA sequences. The lack of accumulation of SPs in the salivary glands indicated the SPs were likely secreted as soon as they were synthesized.

Differential Responses of Wheat Inhibitor-Like Genes to Hessian Fly Attacks during Compatible and Incompatible Interactions.

Junxiang Wu, Xuming Liu, Shize Zhang, Yu-Cheng Zhu, R. Jeff Whitworth, Ming-Shun Chen

Four groups of inhibitor-like genes encoding proteins with diverse structures were identified from wheat. The majority of these genes were upregulated by avirulent Hessian fly, *Mayetiola destructor* (Diptera: Cecidomyiidae), larvae during incompatible interactions, and were downregulated by virulent larvae during compatible interactions. The upregulation during incompatible interactions and downregulation during compatible interactions resulted in 4- to 30-fold differences between the expression levels in resistant plants and those in susceptible plants. The increased expression of inhibitor-like genes during incompatible interactions suggested that these genes are part of defense mechanisms in wheat against Hessian fly attacks, whereas the downregulation of these genes during compatible interactions suggested that virulent larvae can suppress plant

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defenses. Both the upregulation of the inhibitor-like genes during incompatible interactions by avirulent larvae and the downregulation during compatible interactions by virulent larvae were through mechanisms that were independent of the wound response pathway.

Testing Hessian Fly Populations for Virulence to Known Resistance Genes.

Chen M.S., Echegaray E., Whitworth, R.J., Wang H., Sloderbeck, P.E., Knutson, A., Giles K.L. Peairs, F. B. and Walker T.

In recent years, the number of wheat fields heavily infested by Hessian fly has increased in the Great Plains of the U.S. Historically, resistance genes in wheat have been the most efficient means of controlling this insect pest. To determine which resistance genes are still effective in this area, virulence of six Hessian fly populations from Texas, Oklahoma, and Kansas was determined, using the resistance genes *H3*, *H4*, *H5*, *H6*, *H7H8*, *H9*, *H10*, *H11*, *H12*, *H13*, *H16*, *H17*, *H18*, *H21*, *H22*, *H23*, *H24*, *H25*, *H26*, *H31*, and *Hdic*. Five of the tested genes, *H13*, *H21*, *H25*, *H26*, and *Hdic*, conferred high levels of resistance (>80% of plants scored resistant) to all tested populations. Resistance levels for other genes varied depending on which Hessian fly population they were tested against. Biotype composition analysis of insects collected directly from wheat fields in Grayson county, Texas, revealed that the proportion of individuals within this population virulent to the major resistance genes was highly variable (89% for *H6*, 58% for *H9*, 28% for *H5*, 22% for *H26*, 15% for *H3*, 9% for *H18*, 4% for *H21*, and 0% for *H13*). Results also revealed that the percentages of biotypes virulent to specific resistance genes in a given population are highly correlated ($r^2 = 0.97$) with the percentages of susceptible plants in a virulence test. This suggests that virulence assays, which require less time and effort, can be used to approximate biotype composition.

We are currently looking to survey several locations in Kansas and have recently obtained a sample from Eastern Colorado.

Integrated Management of the Hessian fly in Kansas.

R. Jeff Whitworth, Phillip E. Sloderbeck and Brian McCornack

We recently submitted a grant to study the usefulness of pheromone traps to monitor Hessian fly activity and predict the proper wheat planting dates. For years Kansas has use the “fly-free date “ as a way to reduce Hessian fly injury. This date was originally established more than 60 years ago using a set of planting date studies from the 1930’s. Recent Hessian fly outbreaks have called this date into question. We hope to use pheromone trapping data to refine the fly-free date and decide if it is still a useful way to avoid Hessian fly infestations.

K-State Wheat Insect Extension Activities

P. E Sloderbeck, J. P. Michaud and R. J. Whitworth

We continue to add to our wheat insect website at:
<http://www.entomology.ksu.edu/DesktopDefault.aspx?tabindex=195&tabid=405>
and to revise our wheat insect management guide annually
(<http://www.oznet.ksu.edu/library/ENTML2/MF745.PDF>).

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In addition we cooperate with the Plant Pathology Department to revise the Wheat Variety Disease and Insect Ratings each year (<http://www.oznet.ksu.edu/library/plant2/mf991.pdf>).

Plus we make presentations at numerous County Extension meetings and crop tours as requested.

We recently revised our Extension publication on the Hessian fly (<http://www.oznet.ksu.edu/library/entml2/MF2866.pdf>) and added new publications on the bird cherry-oat aphid (<http://www.oznet.ksu.edu/library/entml2/MF2823.pdf>) and flea beetle (<http://www.oznet.ksu.edu/library/entml2/MF2832.pdf>).

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Publications:

Book Chapter

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North Dakota

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Highlights of the 2008 Small Grains Season

The 2008 growing season (April through September) could be described as cooler and drier than normal. The western third of the state was in a moderate to severe drought during much of the growing season. The drought had a major impact on wheat yields in the western part of the state with some areas experiencing a total crop loss. However, in central North Dakota and in the Red River valley of eastern North Dakota, timely rains occurred that provided ideal growing conditions for the wheat crop. Wheat harvest began at the beginning of August and progressed at a good pace due to optimal harvesting weather. Most growers were done harvesting by the beginning of September. Quality and yields were highly variable. The average yield for hard red spring wheat was estimated at 38.5 bushels/acre, an increase of 2.5 bushels/acre when compared to 2007. The average yield for durum wheat was estimated at 25.0 bushels/acre, a decrease of 4.5 bushels/acre when compared to 2007.

2007 IPM Survey

Maps from the 2007 IPM survey in North Dakota were uploaded onto the NDSU IPM website at the following address: (<http://www.ag.ndsu.nodak.edu/aginfo/ndipm/>)

A state-wide survey of small grain diseases and insects continued during the 2008 growing season. Types of insects monitored include: aphids (species not distinguished), grasshoppers (species not distinguished), wheat stem maggot, cereal leaf beetle and barley thrips. All 53 counties in North Dakota were surveyed in 2008. A total of 972 wheat and 222 barley fields were examined. This approximately represents one field surveyed per 7,000 acres of wheat or barley. The survey was initiated on May 29 and continued through August 14, 2008. Crops were surveyed from the 1-leaf stage through hard kernel (ripening) stage. Field scouts surveyed for insect pests of winter wheat, hard red spring wheat, durum wheat, 2-row barley and 6-row barley. All other judgments of pest problems encountered in 2008 are based on reports from County Extension Agents and farmers.

Grain aphids

Grain aphids occurred at relatively low levels in 2008. Due to the cool temperatures aphids were found in only 10% of the fields surveyed. The average percent stem infestation was <1% with ranges from 0 to 40% (based on a 100 stem sample per field). Peak population densities occurred during early July. In North Dakota the treatment threshold is when 85% of the stems have one or more aphids present, prior to the completion of heading. There was little insecticide spraying for control of wheat aphids in 2008.

Grasshoppers

Scouts swept for grasshoppers using a 15-inch sweep net in field edges in 2008. Populations of nymphs and adults were generally below treatment levels throughout the state. The cool spring weather delayed grasshopper emergence and slowed the development of nymphs.

Wheat stem maggot

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Maggots were counted on 100 stems per field (20 stems at 5 locations in each field). Maggots boring in the stem cause characteristic 'white heads'. These heads fail to develop seeds, and are found in otherwise uniformly green fields. White heads were observed from late June to the end of July. Maggots were found in 17% of fields surveyed. In fields in which stem maggots were present, the average percent of stems infested was 5.8%, with ranges from 1 to 40%. The incidence of wheat maggot damage was higher in 2007 where 17% of stems in fields with wheat stem maggots present were infested.

Preliminary research done at North Dakota State University indicates that tank-mixing insecticides (lambda-cyhalothrin "Warrior II") with late herbicide applications (5-leaf to jointing stage) helps to reduce the incidence of white heads and increases yields when large numbers of wheat stem maggots are present. The use of insecticides may partially be responsible for the reduction in maggot damage observed in 2008.

Cereal leaf beetle

Cereal leaf beetle is an export concern for the shipment of hay from ND to California and is monitored for state regulatory purposes. No cereal leaf beetles were detected in ND in 2008.

Barley thrips

Barley thrips per stem were counted from a sample of 40 plants per field. The average number of thrips per stem was 10 with a range of 10 to 50 in fields where thrips were present. Overall the number of barley thrips was higher in 2008 than 2007. Populations were highest in the central and northeastern regions of the state where it was dry early in the summer. Numerous barley fields were sprayed due to the high thrips populations and the high market price for barley.

2008 Hessian fly survey

In 2008, Dr. Marion Harris conducted field trials on a five component sex-pheromone developed in collaboration with Ylva Hillbur, a chemical ecologist in Sweden who specializes in cecidomyiid sex pheromones. Previous field and semi-field tests indicate that the five-component blend might be useful for monitoring Hessian fly in the field. Objectives for 2008 were to determine if the sex-pheromone blend would be useful for monitoring Hessian fly in a field setting, and to establish the geographic distribution, seasonal abundance and number of generation per year of the Hessian fly in North Dakota. Six locations that represent the various climatic and cropping regions of the state were selected for the project. Three traps containing the sex-pheromone lure were deployed at each location. The traps were located in or near wheat plots with each trap positioned 30-60 cm above the ground. The sticky cards on the floor of each trap were replaced each week and the pheromone lure was replaced every 2-4 weeks. Ideally the traps were deployed immediately after the spring thaw and maintained in the field until fall freeze-up.

From the 2008 trap collections it was determined that:

- North Dakota Hessian fly responded to the 5-component pheromone lure.

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- Male flies were recovered from all six locations, which suggests that Hessian fly are distributed state-wide.
- Seasonal abundance of Hessian fly in 2008 was generally characterized by emergence taking place from early May into October, with 1 or 2 peaks in emergence occurring midsummer and a smaller peak in late September.
- Peak fly emergence generally occurred in late July to early August, this indicates that North Dakota Hessian fly did not go into aestivation during the warmest part of the growing season.
- In 2008, the fall emergence of Hessian fly coincided with or was slightly after the suggested planting period for winter wheat in North Dakota, this seems to imply that a fly-free date may not have worked.

In 2009, Dr. Harris will continue evaluating the Hessian fly sex-pheromone. We are interested in using the sex-pheromone to further study the phenology of North Dakota Hessian fly. In addition, we are interested in determining the optimal height for placing the pheromone traps, how best to arrange the traps in the field, determine how often the pheromone lures need to be replaced and find out if all five components are needed to attract male Hessian fly.

2008 Orange wheat blossom midge survey

Additional information on the orange wheat blossom midge as well as maps of the wheat midge survey for the last 13 years is available at the following address:

(www.ag.ndsu.nodak.edu/aginfo/entomology/entupdates/Wheat_Midge/owbm.html)

Our annual soil survey was started immediately after harvest. Sampling was done by the county extension agents taking soil samples from the current year's wheat fields. After collection, the samples are sent to NDSU for processing. At this time the 2008 data is still being analyzed. However, it does appear that the wheat midge is still prevalent across much of the northern part of the state, but generally at low levels. The hot spots for the 2008 growing season will likely be in the north central and northwestern portions of the state.

A new alternative to night field scouting for wheat midge is through the use of sex pheromone traps or yellow sticky traps. Traps are placed into wheat fields at heading and collected every three to five days. Three sex pheromone traps (\$7.20 a trap) per 160-acre wheat field or 10 yellow sticky traps (60 cents per trap) are recommended. Action thresholds are nine wheat midge per sex pheromone trap and five to 20 midge per yellow sticky trap. Sex pheromone traps are available through PheroTech International (<http://pherotech.com/>). Yellow sticky traps are available through Great Lakes IPM (<http://greatlakesipm.com/>) or PheroTech International.

Wheat stem sawfly in 2008

Wheat stem sawfly continues to be a concern for farmers in the southwestern and south-central parts of the state. Sweep-net samples from some locations yielded more than 150 sawfly per 100 sweeps. Although sawfly are usually considered a problem isolated to the western part of the state, reports from numerous growers indicate that it is moving

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eastward across the state. The most common method for controlling sawfly is with solid stemmed varieties. Farmers generally do not like using the solid stem varieties because of yield drag associated with the solid stem trait. NDSW0449, a new hard red spring wheat variety which has a semi-solid stem is in the process of being released. This variety has sawfly resistance and appears to yield better than most of the current sawfly-resistant varieties including Ernest which is the last NDSU variety released with sawfly resistance. The use of insecticides to control sawfly is also being considered. Dr. Janet Knodel is studying the use of insecticidal seed treatments such as Cruiser as a planting time management option. In addition to seed treatments, Dr. Knodel is also evaluating the application of insecticides at the 4-6 leaf and flag leaf stage to control adult sawfly as they emerge. Other strategies to control adult sawfly are underway in the neighboring state of Montana. Currently they are studying Braconid wasps as potential Biological control agents. If the researchers in Montana are successful, the use of parasitic wasps might also improve the sawfly situation in North Dakota.

Background on North Dakota Survey for Diseases and Pests

For the last ten field seasons, aphid monitoring has been carried out as part of a larger effort to survey diseases and insect pests in North Dakota cereals. The state is covered by 5-6 scouts who monitor fields within a county every 1-2 weeks from May through August. The insects that are monitored in cereals include: aphids, grasshoppers, and cereal leaf beetle. Results from these surveys can be found on the NDSU extension entomology website. For aphids, a plant is scored as infested if one or more insects are found. Scouts record any aphid found on the plant rather than separating different aphid species. In North Dakota, common aphids in cereals are bird cherry oat aphid, English grain aphid, corn leaf aphid, and greenbug. Scouts also provide qualitative information on species composition and have been instructed to report the occurrence of the Russian wheat aphid. The cereal aphids that are found in North Dakota are assumed to fly north from breeding sites in Kansas and Nebraska. These same winds are believed to bring rust pathogens to the state. Natural enemies of aphids are not monitored in North Dakota.

Background on North Dakota Autumn Survey for Wheat Midge

In the mid 1980s, a major wheat midge outbreak began in northern Canada and subsequently spread in the 1990s to large areas of Manitoba, Saskatchewan, North Dakota, and northwestern Minnesota. Although wheat midge numbers have declined in recent years the North Dakota Wheat Commission is still concerned enough about wheat midge to pay for an annual soil survey that provides estimates of overwintering wheat midge populations. For this survey county agents send in soil samples in September and October from the current years wheat fields. In our lab, we examine these soil samples for wheat midge cocoons. Cocoons contain overwintering third instar larvae. When wheat midge cocoons are found, larvae are dissected to estimate parasitism levels. A map of wheat midge larval numbers, which takes into account expected mortality from parasitism, is made available to wheat farmers in February/March each year.

It should be noted that the wheat midge is an insect that is easy to miss unless wheat spikelets are opened and developing seeds examined. The wheat midge is very small and has an adult stage that lives for only a few days. Because the adult hides in the canopy

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during the day and is only seen on wheat heads at dusk (when high mosquito populations make it unpleasant to be out examining wheat fields) the wheat midge is rarely reported except when third instar larvae are noticed in large numbers during wheat harvest. Yet even at harvest, observations of wheat midge larvae only occur under particular conditions. If the weather is very dry in August (after larvae finish feeding and before wheat is harvested), larvae remain in the wheat head and are threshed from the heads as the grain passes through the combine. At this time the orange larvae are quite apparent covering the harvesting equipment and can frequently be found in large numbers in the harvested grain loaded onto trucks.

Oklahoma

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I. PERSONNEL

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II. OVERVIEW OF CURRENT RESEARCH AND ACCOMPLISHMENTS

A. Barley Breeding Program (Dr. Do Mornhinweg)

A total of 50 RWA-resistant barley germplasm lines have been released to date. STARS 0501B – STARS 0507B are 6-rowed winter germplasm lines in a feed barley background, STARS 0601B - STARS 0619B are 6-rowed, spring germplasm lines in 4 malting barley backgrounds, STARS 0620B - STARS 0636B are 2-rowed spring barley germplasm lines in 4 malting barley backgrounds, and STARS 0637B- STARS 0643B are 2-rowed spring barley germplasm lines in 3 feed barley backgrounds. These germplasm lines encompass 36 different sources of resistance. These lines were developed by USDA-ARS in Stillwater, and evaluated and selected in Idaho, Colorado and/or Nebraska with assistance of. Phil Bregitzer and Don Obert, USDA-ARS, Aberdeen, ID, Frank Peairs, Colorado State University, and Gary Hein, University of Nebraska. Increases have been made prior to release of 10, RWA-resistant, 6-rowed, winter, feed barley germplasm lines resistant to both Greenbug and RWA.

Four RWA-resistant barley cultivars are now available. Sidney and Stoneham are 2-rowed feed barleys bred for the high and dry plains of eastern CO and western NE. Burton and RWA1758 are 2-rowed spring feed barleys developed for dryland or irrigated production from ID to CO. Sidney and Burton have resistance from STARS 9301B and Stoneham and RWA1758 have resistance from STARS 9577B. QTL analysis done in cooperation with Shipra Mittal and Lynn Dahleen, USDA-ARS, Fargo, identified 3 QTL's associated with RWA resistance in STARS 9301B. Only 2

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of these three were associated with resistance in STARS 9577B one of which showed a different gene action than in STARS 9301B.

A cooperative project with Texas AgriLife is ongoing to map RWA resistance genes in 3 winter barley germplasm lines and *Rsg1* and *Rsg2* greenbug resistance genes.

A breeding program has been initiated to develop winter, hulless, feed barleys resistant to both RWA and Greenbug, adapted to Oklahoma, and suitable for ethanol production. Hulless winter barleys, selected for adaptation to OK as well as percent starch of grain, were crossed as males to RWA and greenbug resistant lines developed by USDA-ARS in Stillwater. Crossing and backcrossing of hulless lines to selected females is ongoing. 4,000 F₄ RWA/GB resistant hulless head rows are in the field this year for evaluation. 4,000 RWA/GB resistant F₃ are being increased to F₄ in the greenhouse and 104 hulless F₂ bulks were planted in Woodward in the fall of 2008 for head selection.

A seedling screening test for BCOA resistance has been developed and tested for repeatability. Two replications of the Barley Core Collection (960 accessions) were screened with this new technique in the summer of 2006. Survivors were grown in pots in the greenhouse and data collected for plant height, grain yield, and yield components. Five seed each of 364 survivors were screened with BCOA in the summer of 2007. An aphid free set of identical flats was also grown. Selected survivors from the screening were rescued and, along with their matching non-infested checks, transplanted into pots in the greenhouse. Infested and non-infested pots for each line were placed side by side on greenhouse benches for increase. Yield and yield components were measured in the spring of 2008. Results indicated that the proposed 1-7 rating scale should be downsized to 1-4. Further confirmation of the rating scale is planned for the summer of 2009.

B. Wheat Breeding Program (Cheryl Baker)

We have continued to work on the purification of wheat differentials for use in Russian wheat aphid biotype screening trials. This material is available to other locations for use in screening tests. Usually the original resistance source for these differentials is a Plant Introduction received from the Germplasm Resources Information Network (GRIN); this USDA facility stores, maintains, and propagates material in a manner that retains as much of its original diversity as possible. Because most of the RWA resistant Plant Introductions are landraces that are highly heterogeneous, researchers cannot simply go back to GRIN to request another sample of a Plant Introduction and expect that sample to be the same as a previous sample. Inherent differences from location to location, and even within a single location, make the use of standardized differentials imperative.

We have continued in the quest to find additional sources of RWA resistance that are resistant to RWA1 and RWA2. In our tests, germplasm release STARS-0601 has continued to have the strongest resistance available to all RWA biotypes against which it has been tested.

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C. Sorghum Breeding Program (Dr. Yinghua Huang)

In the recent years, one of our research emphases focused on sorghum and greenbug interaction, which resulted in some interesting results. In this study, plant-mediated interactions with greenbug, a cereal aphid, were investigated at the gene expression level. Sorghum plants were infested by allowing the aphids to feed on seedlings. Molecular responses of the host plants were analyzed to determine the induced expression of resistance genes and related regulatory factors. More specifically, the extent and level of genes involved in several signal pathways were assessed in parallel, infested resistant genotype and the same genotype without infestation. Among aphid-induced gene expression profiling, signal molecules involved in the global signal transduction pathways, such as salicylic acid (SA), jasmonic acid (JA) and ethylene (ET) pathways, were identified in the host plants in response to greenbug feeding. Then, a variety of functional analysis tools are being used to associate these differentially expressed genes with key signal or metabolic networks that regulate interactions between the host plants and the aphids. Our results indicate that when aphid attacks plant, host plants activate certain genes to express resistance products. The molecular data from this study also provide clear evidence that these major signaling (SA, JA, and ET) pathways modulate a range of host defense responses to greenbug feeding. Overall, these findings provide new insights into the complex pathways governing host defense against phloem-feeding aphids.

D. RWA Biotype Diversity and Ecology (Dr. Gary Puterka)

Studies have been ongoing since 2006 on the ecology of overwintering RWA in diverse environments to determine if and how sexual reproduction (holocycle) occurs and its role in biotype development. Only a few populations collected from the wheat belt east of the Rocky Mountains produced only oviparae, thus were not holocyclic. In 2006, this research was further expanded to include the Colorado Plateau where three other *Diuraphis* spp. endemic to the U.S.A., *D. tritici*, *D. nodulus*, and *D. frequens*, were known to produce overwintering eggs. In the spring of 2007, a small holocyclic population of RWA was located in Dolores County, Colorado. Ninety-three fundatrices were collected from grasses and wheat and identified by morphological traits at this site and evaluated on 16 plant entries that included *DnI-Dn9*, CI 2401, and 2414-11 RWA resistance sources, two susceptible wheat entries, and the resistant barely sources Stars 9577B, STARS 9301, and susceptible Schyler. These evaluations determined that the population was comprised of 47 phenotypes which included biotypes RWA1, 2, 6, 7, and 8. Terry Randolph and Frank Peairs (CSU), independently evaluated 9 fundatrices from this site and identified 5 unique phenotypes. Although we were not able to locate this site in time to verify the presence of RWA males, dead oviparae were found in wheat collected from this site.

We concluded that RWA can go holocyclic and lay overwintering eggs that hatch in late March. The genetic recombination that resulted from sexual reproduction is responsible for the high degree of variation in the population we sampled. Studies are ongoing to further characterize the phenotypic and genetic diversity of the RWA populations from the Colorado Plateau region. Future efforts will include monitoring locations throughout the USA to characterize RWA biotypic diversity and identify

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other ecological zones where RWA may go holocyclic and generate the biotypic diversity that is currently being found in the field.

E. Molecular Ecology of Cereal Aphids and their Natural Enemies (Dr. Kevin Shufran)

The cytochrome oxidase subunit I (COXI) mtDNA gene was sequenced from the following species which are all autoecious holocyclic on grasses; *D. noxia*, *D. tritici*, *D. frequens*, *D. mexicana*, *Schizaphis graminum*, *Sipha flava*, *S. elegans*, and *Sitobion avenae*.

Based on the COXI, DNA barcoding successfully identified and separated the 4 *Diuraphis* aphids at any life stage, including eggs. This assay can be used in field surveys to accurately determine eggs, nymphs, adults, and sexuales.

In research with Dr. Jack Dillwith (Oklahoma State University), it was determined that although individual clones of *Diuraphis noxia* could be distinguished by cuticular hydrocarbon (CHC) profiles, biotypes 1 and 2 could not. There was continuous overlap in CHC profiles between the two biotypes. CHC's are another character in which *D. noxia* shows phenotypic plasticity, even within biotypes.

F. Greenbug Ecology and Biotypic Diversity (Dr. John Burd)

A regional survey of Hymenopterous parasitoids reared from mummified cereal aphids has been completed. Parasitoids were collected from the western slope of the Rocky Mountains in New Mexico, Colorado, and Wyoming. Parasitoids recovered included, *Aphelinus albipodus*, 2 unknown A. spp., *Diaeretiella rapae*, *Lysiphlebus testaceipes*, *Aphidius colemani*, and *Ephedrus plagiator*. Parasitoids attacked aphids on a variety of host plants, including wheat, barley, mountain brome, lovegrass, and crested wheatgrass. Collaborative work with Texas AgriLife Research, Amarillo, (Y. Weng, H. Lu, and J. Rudd) focused on mapping greenbug resistance genes *Gb2* and *Gb6* continues, and thus far, eight markers linked with *Gb2* and *Gb6* have been identified. In 2008, Collaborative research with the Energy Biosciences Institute, University of Illinois (J. Prasifka and J. Bradshaw) was initiated to assess the role of potential biofuel crops as hosts/reservoirs for agricultural pests (primarily aphids).

G. Remote sensing of cereal aphids (Dr. Norm Elliott)

In conjunction with collaborator Georges Backoulou from Oklahoma State University we are reporting on progress in remote sensing for monitoring Russian wheat aphids and greenbugs in wheat. The objectives were to assess whether variation in light reflectance from plants infested with varying densities of greenbugs could be detected using airborne imaging obtained with a multi-spectral digital camera mounted in a fixed wing aircraft. In a replicated experiment where greenbug density was manipulated in 1-m² plots of two winter wheat varieties ('Jagger' and 'OK 101') planted in a field, we found that wheat infested with greenbugs exhibited different reflectance responses for the two varieties. Both varieties showed a reduction in the normalized differenced vegetation index (NDVI) as greenbug density increased as indicated by a negative slope for the regression of NDVI on greenbug density. The slope for Jagger was greater in magnitude (-0.0031) than the slope for OK 101 (-0.0011). The regressions indicated that NDVI decreased more rapidly as

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greenbug density increased for Jagger than for OK 101. A second vegetation index, Green NDVI, responded similarly to NDVI with increasing greenbug density. In large plots in four production winter wheat fields we found significant negative correlations between greenbug density and the above mentioned vegetation indices for three of the four fields.

For the Russian wheat aphid we assessed the potential of using multispectral imagery and a spatial pattern recognition approach to identify and spatially quantify *D. noxia* infestations within wheat fields. Data used included multispectral imagery acquired from April - May 2005, and 2007, in the vicinity of Boise City, OK. Stress to wheat in fields was grouped into categories: *D. noxia*, drought and cultural issues. ERDAS Imagine software was used to process and analyze images. FRAGSTATS was used to quantify spatial pattern. Ten landscape metrics were computed at the class level for each stress factor. The analysis of variance of each landscape metric revealed that the shape of each kind of stress was different. The combination of multispectral data and landscape metrics made it possible to distinguish areas in fields infested by *D. noxia* from areas affected by drought or cultural issues.

III. PUBLICATIONS

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Projects in 2008

Intraguild Interactions among *Schizaphis graminum*, *Lysiphlebus testaceipes*, and Coccinellidae in Winter Wheat. Christopher Mullins Kristopher Giles and Tom Royer. C. Mullins is expanding on this work, and document the level of intraguild predation in wheat fields.

Predator Movement in Wheat Cropping Systems. Sara Donelson, Kristopher Giles. Studies have been initiated to examine the colonizing ability of Carabidae. Trapping and molecular techniques will be used to describe movement.

Evaluations of Variety and Insecticidal Seed Treatments for Hessian Fly Management. Dayna Alvey, Kristopher Giles, Tom Royer Brett Carver. Field evaluations of elite lines of wheat for control of Hessian fly. In addition, a survey of wheat fields was conducted through the Cooperative Agricultural Pest Survey (CAPS). Hessian fly was becoming more widely distributed, but Russian wheat aphid was not present in any of the fields checked. Barley Yellow Dwarf virus and Wheat Streak Mosaic virus were also detected.

Synopsis of Arthropod Pest Activity in Wheat, 2007-2008

Overall, pest pressure in 2007-08 was mild. An outbreak of armyworms occurred in spring of 2007 and an outbreak of fall armyworms occurred in fall 2008. Greenbug pressure is building as of now. Hessian fly is increasing in Oklahoma. Some fields were severely infested. Problem is growing.

Publications

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\$225,731.00 NRCS, Conservation Innovation Grants. 2008-2011. An innovative approach to conservation and integrated pest management in Oklahoma cropping systems. J. Edwards, R. Kochenower, G. Strickland, K. Giles and T. Royer

\$171,932.00 USDA-PMAP. 2007-2009. Implementation and economic evaluation of the Glance 'n Go greenbug+parasitism sampling and management plan on winter wheat in KS, OK, and TX. Giles, K, T. Royer, F. Epplin, A. Knutson, G. Michels, R. Bowling, P. Sloderbeck, J. Whitworth and N. Elliott.

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Overview of Cereal-Aphid Research Activities and Accomplishments

The rice root aphid, *Rhopalosiphum rufiabdominalis* (Sasaki), is distributed worldwide and colonizes a wide range of plants. However, relatively little is known about the suitability of different host plants, optimal rearing techniques, and the aphid's impact on plant fitness. To improve understanding of these factors, laboratory experiments were conducted to compare the abundance of rice root aphid on plants grown using three different soil-surface media and among selected monocotyledonous and dicotyledonous plants. Rice root aphid was more abundant on plants grown with a sandy soil surface than a surface with fine wood chips or only bare non-sandy soil. Rice root aphid was more abundant on 'Elbon' rye than on 'Bart 38,' 'Dart,' 'Fletcher' and 'Ramona 50' wheat. More winged rice root aphids were produced on Elbon rye than on Dart wheat, but the number of winged aphids on Elbon rye did not differ from that on other wheat lines. Rice root aphid was more abundant on Elbon rye and 'TAM 110' wheat than on 'Marmin,' 'Marshall' and 'Sharp' wheat. Additional observations with monocotyledonous plants showed that abundance of rice root aphid on 'Kivu 85' triticale was comparable to that on Elbon rye. Rice root aphid did not reproduce on potato or soybean, although winged adults persisted up to 24 days on caged potato plants. The differential abundance of rice root aphid on plants has implications with regard to colony rearing, future experiments and pest management.

Publications

Hesler, L.S. & S.D. Kindler. 2007. Abundance of rice root aphid among selected plant species and on plants grown with different soil-surface media. *Great Lakes Entomol.* 40:83-90.