

WESTERN EXTENSION/EDUCATION RESEARCH ACTIVITY – 066

MINUTES OF THE ANNUAL MEETING SEPTEMBER 19-20, 2005 FORT COLLINS, COLORADO

Report submitted by DW Mornhinweg

List of participants:

1. Kevin Shufran, USDA-ARS, Stillwater, OK
2. Phil Sloderbeck, Kansas State University, Garden City, KS
3. Do Mornhinweg, USDA-ARS, Stillwater, OK
4. Mustafa Mirik, Texas Ag. Exper. Station, Amarillo, TX
5. Rick Meyer, USDA, CSREES, Washington DC
6. Yiqun Weng, Texas Ag. Exper. Station, Amarillo, TX
7. Frank Peairs, Colorado State University, Fort Collins, CO
8. Louis Hesler, USDA-ARS, Brookings, SD
9. Cheryl Baker, USDA-ARS, Stillwater, OK
10. Mike Smith, Kansas State University, Manhattan, KS
11. Norm Elliot, USDA-ARS, Stillwater, OK
12. Sue Blodgett, Montana State University, Bozeman, MT
13. Judith Brown, University of Arizona, Tuscon, AZ
14. Gary Hein, University of Nebraska, Scotts Bluff, NE
15. Tom Royer, Oklahoma State University, Stillwater, OK
16. Tom Holtzer, Colorado State University, Fort Collins, CO
17. J.P. Michaud, Kansas State University, Hays, KS
18. Gearriett Cuperus, Oklahoma State University, Stillwater, OK

Minutes

Sept. 19.

8:07 AM - Kevin Shufran, WERA-066 Chair, opened the meeting. Dr. Shufran thanked Frank Peairs and Terri Randolph for the local arrangements and welcomed participants to the meeting. Attention was called to the handouts for the meeting available at the registration table, i.e. agenda, compiled state reports, and attendance sheet.

8:15 AM - Tom Holtzer, administrative co-advisor, spoke about the importance of documenting our accomplishments and their impacts for the WERA - 066 report. He reminded the group about the upcoming renewal petition due in 2006 and suggested the formation of a subcommittee just for the impacts section of the renewal petition. He said that May 15 was the last possible date for the submission of the renewal but suggested an earlier target date of Jan. 15 for a rough draft to members to ensure timely submission. He suggested we discuss at this meeting our key objectives

for the future. Not all objectives have to change if we are still working towards some of the previous objectives.

8:20 AM – Rick Meyers, CSREES, National Program Leader for Entomology, also emphasized the importance of inputs, outcomes and impacts in new logic models utilized by ARS as PART, program assessment rating tools, of all USDA-ARS programs. He discussed some of the budget concerns for FY06 and FY07 grant funding. In the future it appears that there may be a decrease in formula based funding and an increase in competitive funding. For FY06, funding will be maintained but may be reshaped in the future, perhaps as early as FY07. He stated that budget cuts due to hurricane Katrina costs will most likely come out of Health and Education but possibly some out of Research as well. Congressional earmarks may also be zeroed out. He suggested visiting www.AAAS.org for updates on Research and Development in the government.

8:35 AM – State reports from, Colorado, Kansas, Nebraska, Montana, Oklahoma, South Dakota, and Texas.

9:45 AM – Break

10:00 AM – Presentation by invited speaker, Dr. Judy Brown, University of Arizona, Update on Biotypes and the *Bemisia tabaci* complex.

12:00 PM – Break for lunch

1:15 PM – Continuation of state reports from Texas and USDA-ARS, Stillwater, OK.

Dr. Norm Elliot was asked to give an overview of the progress of the area wide pest management project. He said it is currently in the last year of phase 2. This coming year will involve documentation of the impact of the study. Focus groups with growers helped in convincing them to diversify cropping systems. There was a good diffusion of ideas among growers. There were economic surveys of 150 growers which included socioeconomic issues as well. Funding for another year to get all the data together.

Sue Blodgett suggested a compilation of all state RWA biotype surveys to construct a biotype map of the western US.

Frank Peairs began discussion a of Mike Brewer's replacement at University of Wyoming, Dr. Alex Latchinsky, and his fit to this group.

2:10 PM – Dr. JP Michaud, Kansas State University, began a discussion on research he had conducted involving the performance of RWA2 vs RWA1

on Dn7c under differing temperatures. Questions were fielded by J.P. and suggestions were made concerning the best conditions for future such studies.

2:45 PM – Kevin Shufran distributed a copy of a draft proposal “Proposed Guidelines for Identifying Biotypic Variation and Designating of *Diuraphis noxia* Biotypes for Use by the WERA-066” for group discussion. Kevin asked for a subcommittee to be responsible for the final draft and that this be added to the new petition as one of our new objectives and statements. Phil Sloderbeck volunteered to post the final draft to the KSU website for comments and suggestions from the group so we could all access it as we develop our protocol. The group decided instead, to tackle the proposed guidelines at this meeting during the subcommittee breakout session for aphid ecology and plant-insect interactions. Because so many people were eager to discuss this topic it was decided that the subcommittee breakouts would be held consecutively instead of concurrently so that the whole group could have input to both. Subcommittee chairs and recorders were nominated as follows: Biocontrol subcommittee, J.P. Michaud-chair, Tom Royer-recorder; Aphid Plant Interaction subcommittee, Mike Smith-chair, Cheryl Baker-recorder.

3:10 PM – Break

3:25 PM – Subcommittee on Biological Control.

4:00 PM – Subcommittee on Aphid Ecology and Plant-Insect Interactions

5:00 PM – Kevin appointed a nominating committee to solicit nominations for incoming secretary and a site selection committee for next years meeting. They were asked to report on the following day. Closing remarks for the day were offered as well as announcements.

Sept. 29

8:10 AM – Minutes from 2004 meeting were circulated for approval. Aphid Ecology and Plant-Insect Interaction subcommittee discussion continued on RWA biotyping proposal. A consensus was reached for the corrected proposal by the group. Oral subcommittee reports were not necessary as the group was all in attendance for each subcommittee. Written reports provided after the meeting can be found at the end of the minutes.

11:00 AM – Minutes for the 2004 WERA-066 meeting were approved as corrected, moved and seconded. Kevin Shufran began discussion on the renewal petition. He stated that the outgoing chair, incoming chair, and incoming secretary are responsible for writing and submitting the renewal. It was decided that a draft of the renewal should be sent out to the whole

group for review by mid Jan. Each objective of the petition was individually discussed. Kevin brought to the attention of the group that interest had been expressed by several individuals to add Hessian fly to the petition goals. Discussion followed with a consensus that such a change would only strengthen the group. The emphasis would still be RWA but the scope broadened to include Hessian Fly. It was suggested the title of the group could be changed to “Integrated Management of Russian Wheat Aphid and Other Cereal Arthropods”. Gary Hein suggested a new objective dealing with biotypes. Cheryl and Gary drafted a new objective: Enhanced effect of development of resistant varieties by coordinating the identification, monitoring, and characterization of RWA biotypes. This was approved by consensus of the group. Sue Blodgett and Tom Royer put forward some suggestions for the Education Plan as part of the renewal document.

11:45 AM – Nominations were made for incoming secretary. Phil Sloderbeck was nominated, moved and seconded. The 2006 selection site committee proposed Manhattan, Kansas in Mid Sept. 2006. The proposal was accepted. Discussion followed suggesting Monday Sept. 18 travel instead of the normal Sunday travel with the meeting to follow on Tues. the 19 and ending at noon on Wed. the 20. Final date approval will be made by the hosts.

Tom Holtzer added that once the petition renewal is entered in NIMSS extension directors in all states will receive a message to allow new members to join the committee. Everyone currently in the group must also respond in order to remain on the committee.

12:00 PM - Meeting adjourned.

SUBCOMMITTEE REPORTS

Biological Control Subcommittee Report

Chair: J.P. Machaud
Secretary: Tom Royer

J.P. Michaud opened the session with a discussion of what, if any, are the effects of no-till/conservation tillage on biological control of cereal aphids, especially on sorghum.

- In the 1980’s Dr. Burton did work on landing rates of aphids, but did not look at the effects on aphid survival due to biological control.

- A question was posed: How should such research be coordinated? Can we develop a protocol at a regional level to determine these effects (no-till vs. conventional till on biological control).
- One suggestion: we access information from Gerry Michaels. (corn leaf aphid and greenbug relationship).
- J.P. is looking at *H. convergens* and what they do after they leave a wheat crop. How do they bridge the “famine” times before sorghum is available to provide a food source?
- He has been looking at reproductive diapause. He thinks that *H. convergens* goes into a maintenance mode, getting water and survival food source from sunflower. Then when a aphid food source becomes available, they can reproduce in response to food availability.
- They conducted a study where they developed a “maintenance diet” and fed females. The beetles basically “dribbled” out a few eggs at different times during their time on the maintenance diet, then when they were fed greenbugs, they produced massive quantities of eggs.

Tom Royer brought up a discussion of the “Glance’n Go” sampling system and asked for suggestions on how to measure impact, how to measure adoption, how to increase adoption.

- Overview: scouting time was monitored, takes an average of 9 minutes 20 seconds to scout fields. Longest is 22 minutes if all samples are needed. Shortest is 3 minutes if minimum samples are needed to make a decision.
- A survey of SW Oklahoma producers indicated that 80+ percent scout their field at least 2-3 times for greenbugs, but many have not yet heard of Glance’n Go.
- Various Suggestions were offered to further promote and educate the public about Glance’n Go:
 - Promote by emphasizing time and money savings to growers.

- Relate time to ordinary activities, then demonstrate how much money this would be worth if Glance'n Go were used.
- Promote Glance'n Go by, news releases, running advertisements in agricultural publications and newspapers, and promote to agricultural suppliers and (time) link with first alerts for insect outbreaks.

Aphid Ecology and Plant-Insect Interactions Subcommittee Report

Chair: Mike Smith

Secretary: Cheryl Baker

Guidelines for Identifying Biotypic Variation and Designation of *Diuraphis noxia* Biotypes.

Rationale: In order to achieve the goals set forth by the WERA-066 concerning research and management of *Diuraphis noxia* and its biotypes, the members came to agreement as to what a biotype is and how it is identified and designated. Without such agreement, it would be extremely difficult to coordinate research efforts and achieve the objectives of the project. This agreement pertains to *D. noxia* only, and was based on lessons learned from designating greenbug, brown planthopper, Hessian fly and biotypes of other species. In addition, these guidelines are specifically for use in attaining the objectives set forth by this committee. Most importantly, we have strived for our decisions to be based on reputable science and facts, not hypothesis, theories, traditions or dogmatic views.

A primary objective for the future coordination of WERA-066 will be the development and deployment of new resistant wheat and barley to mitigate the injury caused by the population of *D. noxia* which is now injurious to *Dn4* wheat (Haley et al. 2004), as well as other unpublished and yet to be described populations injurious to other wheat and barley resistance genes. The following guidelines for the identifying and naming of *D. noxia* biotypes were established at the September 2005 WERA-066 meeting in Fort Collins, CO. Our challenge as members of WERA-066 was to develop a structured system which, is **most applicable and useful for attaining this objective of delivering *D. noxia* resistant crops to the producer**, for characterization and designation of *D. noxia* biotypes. Input into this document was from both entomologists and plant breeders.

What is a *D. noxia* biotype? A population (independent of geographic location) that is able to injure a cultivated plant containing a specific gene(s) which was previously resistant to known aphid populations. The above definition prescribes that the biotypic status of *D. noxia* be solely based on the phenotypic response of the plant as a result of the aphid's feeding.

In the above definition of *D. noxia* biotypes, there is no presumption of the genetic basis within the aphid for the ability to cause injury, nor is any evolutionary or taxonomic status implied. Certainly, there are genetic differences among, and even

within, biotypes that affect the phenotypic response of the plant. However, the term biotype does not describe those differences. The biotype classification does not require knowledge of the specific biological traits of the aphid that cause the observable symptoms of the plant. Characters measurable or observable in the aphid can be used to further characterize biotype-plant interactions, but not to designate biotype status. Again, biotype is not an evolutionary or taxonomic classification. It is merely, a convenient and very applicable way to describe an array of resistant and susceptible plant responses, or the insect-plant interaction that leads to the injury of a plant resistant source.

The guidelines for naming and testing *D. noxia* biotypes are:

1. Biotypes will be named sequentially beginning with RWA1, RWA2, RWA3 etc.
 - a. RWA1 will be the laboratory colony maintained at the USDA-ARS in Stillwater, OK; this colony was founded in 1987. *Dn4* wheat is resistant to this colony. It is the base line that all other biotypes will be compared/contrasted to.
 - b. RWA2 has been used to describe the *Dn4* injurious population first reported by Haley et al. (2004). This name was agreed upon at the WERA-066 meeting in 2004. A reference that has utilized this designation is Porter et al. (2005). Biotype designations will be independent of the crops upon which they are virulent.
2. The designation of biotypes will be done using publicly available plant genotypes, including designated *Dn* genes or defined genotypes of interest. Source seed of all of the differential *Dn* genes will be available from Stillwater-ARS. Additional differentials may be added as they are identified and become available in sufficient quantity.
 - a. For the purposes of the matrix, using genotypes with a similar background should decrease the likelihood of differences in plant reaction occurring for other reasons. Current *Dn* wheat genes that meet the current criteria and are currently available in a ‘Yuma’ background include:
 - i. *Dn1* (CO03797), resistance originally derived from PI137739;
 - ii. *Dn2* (CO03804), resistance originally derived from PI262660;
 - iii. *dn3* (CO03811), resistance originally derived from *Triticum tauschii* line SQ24, and
 - iv. *Dn4* (Yumar), resistance originally derived from PI372129.
 - b. Other *Dn* genes that meet the current criteria are:
 - i. *Dn5* (tentatively available in Colorado breeding line CO950043), resistance originally derived from PI294994;
 - ii. *Dn6* (currently available in “RWA Matrix-6501” and Colorado breeding line CO960223), this gene was originally identified in PI243781,
 - iii. The first published source of *Dn7* “RWA Matrix-Dn7”; purified for RWA1 resistance (at Colorado State University) from 94M370 (a wheat-rye translocation line developed in South Africa by G. Marais),
 - iv. *Dn8* (available in a selection from a South African wheat

- germplasm line, Karee-Dn8 (PI634775)), resistance originally derived from PI294994;
- v. *Dn9* (available in a selection from a South African wheat germplasm line, Betta-Dn9 (PI634770)), resistance originally derived from PI294994;
 - vi. *Dny* (available in Stanton wheat, developed at Kansas State University; RWA resistance originally derived from PI220350).
- c. Other differentials that are currently being purified for future use include:
- i. “RWA Matrix-2401” (a Stillwater ARS selection from CIt2401),
 - ii. “RWA Matrix-2414-11-2” and “RWA Matrix 2414-11-5” (from a Stillwater ARS breeding line derived from a cross with PI366515, with further selection done at Colorado State University), and
 - iii. *Dnx* (tentatively available in two Kansas State University breeding lines; KS041149, and KS00HW152-2-6, both derived from crosses with PI220127).
- d. Currently available *Dn* genes in crops other than wheat include:
- i. The barley genes that meet the criterion in #2 above are *Dnb1* and *Dnb2*, available together in STARS-9301B (Mornhinweg et al. 1995).
3. Where newly discovered differences exist in the resistance of plant genotypes, results will be reported to the WERA-066 for discussion and potential addition to the existing biotype test matrix.
4. New putative biotypes will be tested against the full array of the above plant genotypes. An aphid clone causing a differential reaction in plants with at least one resistance gene will be considered a new biotype.
5. Rating Scale and testing conditions
- a. A 1-9 rating scale for chlorosis similar to that described by Webster et al (1991) shall be adapted.
 - b. A 1-3 rating scale for leaf rolling shall be used.
 - c. A chlorosis rating of greater than 5 and/or a rolling rating of 2-3 will be designated as susceptible for biotype designations.
 - d. A susceptible check (Yuma wheat and Morex barley) is used. Tests will be rated when the susceptible variety is rated 8-9 for chlorosis and 3 for rolling.
 - e. A resistant check is used as long as all resistance genes have not been overcome.
 - f. Appropriate controls are used and environmental conditions standardized:
 - i. Lighting conditions are adequate to support healthy plant growth (full sunlight or supplemental light may be needed). Daylength is best maintained at 16L:8D to minimize alate development and maximize plant growth.
 - ii. Temperature conditions will be recorded for the test duration (daily max/min). The target range should be approximately 20-26°C.
 - iii. Plants should be infested when 1-2” tall.
 - iv. A demonstrated ability to reproduce results obtained in multiple locations is vital. A second confirmation test should be conducted

prior to a new biotype designation.

Citations

- Haley, S.D., F. B. Peairs, C. B. Walker, J. B. Rudolph, and T. L. Randolph. 2004a. Occurrence of a new Russian wheat biotype in Colorado. *Crop Sci.* 44: 1589-1592.
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Appendix: State Reports Submitted For 2005

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Reports below

Annual Report to WERA-66: 2005

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II. Sub-Committee Objectives Addressed

A. Biological Control

1. Seek ways to improve biological control of the Russian wheat aphid through the use of diversified dryland cropping systems.

B. Host Plant Resistance

1. Incorporate genetic resistance to Russian wheat aphid into commercially acceptable winter wheats for Colorado.
2. Categorize the mechanisms of known genetic sources of resistance to Russian wheat aphid in order to determine the best combinations for stable resistance.
3. Test experimental wheat lines and varieties that are resistant to Russian wheat aphid at multiple locations for level and stability of quality, yield and resistance.

C. Biology and Management

1. Refine economic injury levels and thresholds for Russian wheat aphid in small grains to incorporate additional factors such as cultivar, cropping system, and presence of other pests. Monitor economic impact of Russian wheat aphid in

Colorado.

2. Conduct studies on the field biology and ecology of the Russian wheat aphid to improve understanding and management of Russian wheat aphid.
3. Determine the influence of modified cultural practices, including grazing, planting date and volunteer control, on Russian wheat aphid densities.
4. Improve application technology including safer and more effective insecticides and more efficient application techniques.

III. Current Accomplishments

A. Biological control

1. Pitfall traps have been established at three cropping systems sites. Carabids and spiders are being collected and identified. A manuscript on carabid results is being prepared (Objective A1)
2. Uniform aphid natural enemy observations are taken at all three locations (Objective A1)

B. Host Plant Resistance

1. Russian wheat aphid resistant wheat cultivars are now planted on more than 25% of Colorado's wheat acreage.
2. Lines with multiple resistance genes were included again in preliminary yield tests. New genes are being combined and backcrossed with adapted wheat cultivars. Efforts continue to incorporate RWA resistance from goatgrass, rye and triticale into bread wheats. (Objective B1)
3. Resistant feed barley lines were tested in 2001 - 2004. Some lines have resistance to RWA as well as the ability to perform well agronomically under dryland conditions. (Objective B3)
4. Surveys were conducted to determine the presence of Dn4-virulent Russian wheat aphids. Of the 122 samples taken, 99 were Dn4-virulent. No virulence to Gamtooz-R (Dn-7) or 2414-11 was detected.
5. Approximately 7,500 of 12,000 new accessions from the national collection have been screened with RWA-2. Resistant lines were rescreened with RWA-1. Lines resistant to both were identified.

C. Biology and Management

1. Dryland cropping systems studies are ongoing at three locations in eastern Colorado. Both resistant (Dn4) and susceptible cultivars are used in treatments containing wheat. (Objectives A1, B3 and C3)
2. Aphid flights were monitored at four locations by means of suction traps. (Objective C2)
3. Ten foliar insecticide treatments were compared to commercial standard insecticide treatments for control of Russian wheat aphid. None were superior

to what is currently available to Colorado wheat producers. (Objective C4)

IV. Publications

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Rudolph, J. B., T. L. Randolph, S. M. Walters, F. B. Peairs, and A. Gebre-Amlak. 2005. 2004 Colorado field crop insect management research and demonstration trials. Colorado State Univ. Agric. Exp. Sta. Tech. Rep. TR03-01, 43 pp.

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CEREAL APHID RESEARCH
2005 STATE REPORT
IDAHO

I. CURRENT RESEARCH AND ACCOMPLISHMENTS

Aphid population monitoring

In 2005, the five suction traps in Idaho were put into operation early in June. Traps were located in Aberdeen, Arbon, Rexburg, Rockland, and Soda Springs. Rockland was not in operation until mid-July. Trapped aphids were collected weekly and mailed to the Research and Extension Center at Aberdeen, where they were sorted, identified and counted. The total collection count for the season was 2,346 aphids belonging to 19 species.

The purpose of this project is to provide timely information to potato and cereal producers about risks of aphid pests and virus epidemics. According to these preliminary results, adjusting fall potato vine-killing dates to avoid aphid flights would be impossible. However, the identification of the PVY aphid vectors would allow us the establishment of better management tools to reduce the spread of PVY and all non-persistently transmitted viruses. Adjusting planting dates for winter wheat, however, would be possible based on these data.

Comparison of aphid composition and phenologies across suction trap localities

The relative composition of aphid fauna in southeastern Idaho is very consistent. Bird cherry oat aphid (*Rhopalosiphum padi*) was the most abundant aphid species in four out of the five localities examined. And most of these localities displayed the same proportionate abundance of cereal aphid species. The most common cereal aphid was bird cherry oat aphid. Rose grass aphid (*Metopolophium dirhodum*) and Russian wheat aphid (*Diuraphis noxia*) were the second and third most active aphids in the field respectively. And English grain aphid (*Sitobion avenae*) was the least common of the cereal grain aphids flying near the suction trap localities.

The aphid composition in Rockland was slightly different from other localities. Russian wheat aphid, not bird cherry oat aphid, was the most prominent and abundant aphid in Rockland. Although constituting a large proportion of the aphid fauna and almost as abundant as Russian wheat aphid, the bird cherry oat aphid was the second most captured aphid. English grain aphid was never recovered.

The low numbers of bird cherry oat aphid could be an artifact of collecting effort. The suction trap in Rockland was not in operation until mid-July. The traps in the other collecting sites were in operation one month before. These other collecting traps could have an extra month's worth of bird cherry oat aphids.

The results from Rockland are more likely a reflection of its true aphid composition. Bird cherry oat aphid was not populous early in the summer. Searching for flying bird cherry oat aphids during this period would have been a wasted effort. Across all four sites, *Rhopalosiphum padi* was actively flying during those final weeks of July and early weeks of August when aphids were being collected in Rockland. Furthermore, the bird cherry oat aphid numbers during this seasonal peak in Rockland were the lowest of the four collecting sites. And Russian wheat aphid was more abundant in Rockland than any other test site.

Results of the trap in Rexburg are of special interest to potato growers, considering that most of the Idaho seed potato growers are located in this region. It is generally accepted that the most important vectors of PVY are actually aphid species that do not colonize potatoes. Eighteen of the caught species in Rexburg do not colonize potatoes. The only colonizing species was the green peach aphid. Potentially, all 18 species caught could vector non-persistently transmitted viruses into a potato crop. Two of the most abundant species (bird cherry-oat aphid, rose-grass aphid) in Rexburg were all previously confirmed as vectors of PVY in Idaho. Though the transmission rates of bird cherry-oat aphid are not as high as that of green peach aphid, the number of the bird cherry-oat aphid caught compared to the number of green peach aphid (one female collected at the end of June) suggest that cereal aphids in general contribute more to the spread of PVY than green peach aphid.

Biology of bird cherry-oat aphid

The response of the bird cherry-oat aphid, *Rhopalosiphum padi*, to wheat plants infected with *Barley yellow dwarf luteovirus* (BYDV) was evaluated in the laboratory. Significantly more aphids settled onto virus-infected than non-infected plants when aphids were able to contact the leaves of varieties Lambert and Caldwell. Additionally, more aphids congregated on screens above headspace of virus-infected plants than above non-infected ones of both varieties. Further tests demonstrated that the response of aphids is due to attraction (immigration) rather than arrestment emigration). The concentration of headspace volatiles was greater on virus-infected Lambert, than on non-infected plants of this variety.

Further bioassays were performed to assess responses of the aphid to BYDV-induced volatiles during disease progression in wheat plants. Treatments included non-transformed (Lambert) and transgenic (103.1J, which expresses the BYDV-PAV coat protein gene) wheat genotypes infected with BYDV and sham-inoculated, and a paper leaf model. Sham-inoculated plants were challenged with non-viruliferous aphids as a control to assess responses induced by aphid feeding. Observations were taken 15, 22, 29, and 36 days after virus inoculation for every treatment simultaneously. We measured immigration rates of virus-free apterae at intervals of 5 minutes for 60 minutes in darkness. Aphids on screens above leaves were considered immigrants. Results are being analyzed.

II. PERSONNEL

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III. PUBLICATIONS

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Kansas State University

WERA66 2005 Annual Report

C. Michael Smith (representative), Elena V. Boyko, Leslie R. Campbell, Andrea Ray-Chandler, Ming Chen, John Diaz-Montano, Xuming Liu, John C. Reese, Sharon Starkey, Priyamvada Voothuluru and Lieceng Zhu, Department of Entomology, Manhattan, KS 66506; J. P. Michaud, J. L. Jyoti, Tom Harvey, J.A. Qureshi, Department of Entomology & Ken Kofoed, Department of Agronomy, Kansas State University, Agricultural Research Center, Hays, KS 67601;

Alan Fritz, Bill Schapaugh, Department of Agronomy;
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Russian Wheat Aphid Feeding Behavior as Determined by Electrical Penetration Graph Analysis

Andrea Ray-Chandler, John C. Reese and C. Michael Smith

To determine more about how the feeding behavior of *D. noxia* biotype 1 on resistant and susceptible wheat plants correlates with resistance, the Electrical Penetration Graph (EPG) technique was used to obtain information about the potential expression of plant resistance in the first few hours after infestation. A prime parameter of interest was how long aphids took to establish feeding on a resistant or susceptible line. These data may also help establish if resistance factors are expressed inter- or intra-cellularly, or within the phloem as is often seen in aphid resistance on wheat and barley. EPG creates an electrical circuit between the aphid and plant. Changes in voltage over time result in recognizable waveforms correlated to different stylet penetration behavior. The identification and quantification of these activities gives plant resistance research important information regarding the physical locations of resistance within the plant and the times of responses of the insects to those plants.

The amount of behavioral variation in these recordings was broad but consistent. There were no differences between *D. noxia* feeding on resistant and susceptible lines in the total pathway time (all probing activity excluding phloem feeding). However, the available time for phloem feeding was significantly reduced on the resistant line compared to the susceptible line ($Z_{(17,17)} = 0.0245$, $P < 0.05$) (Mann-Whitney U test). There is an even more distinct difference in the total duration of phloem ingestion, where feeding was reduced significantly on the resistant line ($Z_{(17,17)} = 0.0176$, $P < 0.05$). The lack of difference in total pathway time between aphids feeding on resistant and susceptible lines gives strong indication that resistance factors are likely not inter- or intra-cellular within the general leaf tissue, but rather in the sieve element cells or phloem sap, as indicated by the differences in the available and total time spent feeding. It was interesting that successful data sets for aphids feeding on resistant plants were more difficult to acquire, because aphids either did not probe or became unglued from their gold recording wire tether. This trend suggests the possibility for a generalized antixenotic reaction, although the aphids cannot easily leave an undesirable plant, but rather can reduce probing and feeding.

Characterization of Soybeans for Resistance to the Soybean Aphid, *Aphis glycines*

John Diaz-Montano, John C. Reese, William T. Schapaugh, C. Michael Smith and Leslie R. Campbell

A total of 240 soybean entries were screened for resistance to the soybean aphid and 11 entries showed significantly lower numbers of offspring than the susceptible checks at 7 days after infestation. The low populations found in these 11 entries indicate that antibiosis and/or antixenosis could be conferring resistance to the soybean aphid. When these 11 entries were studied in no-choice tests all of them showed antibiosis at different levels. But when choice tests were performed only two of them showed antibiotic effects against the soybean aphid. These two entries had significantly fewer nymphs than the other entries in all experiments, suggesting that they are highly resistant to the soybean aphid.

Biological Performance of Russian Wheat Aphid ‘Biotype 2’

J. P. Michaud, J. L. Jyoti and J.A. Qureshi

We have completed a series of experiments testing the responses of both alate and apterous RWA colonizing different arrays of wheat cultivars. We found that different proportions of aphids responded when different combinations of cultivars were offered to apterous fourth instars. More biotype 2 RWA responded (settled on plants) when more Dn4- expressing cultivars were present in the lineup, and fewer biotype 1 RWA. This was shown to be a function of differential rates of plant abandonment after plants were ‘sampled’ for 24-48 h. However, we did not obtain significantly different rates of aphid colonization among cultivars in any combination save one, in which biotype 2 apterae colonized Yumar and Akron at a higher frequencies than Yuma or Stanton. These results suggest that young wheat plants appear to lack any meaningful antixenosis toward *D. noxia*, even though the aphids appear to perceive, and sometimes respond to, certain differences in cultivar suitability.

The performance of biotype 2 Russian wheat aphid, *Diuraphis noxia* (Mordvilko), was compared with that of the original biotype 1 on eight wheat cultivars at two constant temperatures and the plants evaluated for overall damage and leaf rolling. Colonies of biotype 2 grew an average of 2.3 and 24.9 times faster in the first and second generation, respectively, than did their biotype 1 counterparts at 20 C, reaching 80-125 aphids per plant after 20 days, compared to 10-31 for biotype 1. The numbers of aphids per plant at 10 d and 20 d after infestation displayed a significant biotype-temperature interaction. There was also a biotype x temperature interaction for plant damage at 10 d, and for damage and leaf rolling at 30 d. After 20 d at 24 C, damage ratings ranged from 7.3-8.6 on a scale of 1.0-9.0, and leaf rolling ranged from 2.4-2.9 on a scale of 1.0-3.0 for biotype 2, whereas values for biotype 1 ranged from 2.8-5.1 and 1.4-2.2, respectively. There were no notable differences among cultivars in either plant damage or leaf rolling induced by biotype 2 and ratings of both were higher than for biotype 1 in all cultivar-temperature combinations. The results indicate that biotype 2 *D. noxia* is more virulent than biotype 1 to all the cultivars tested, and induces plant injury that develops more rapidly than that induced by biotype 1, especially at higher temperatures.

We compared the development and reproduction of biotype 2 *D. noxia* at 21.7 ± 0.12 °C on ‘Trego’ (PI 612576), a susceptible commercial cultivar, and on lines CI 2401 and 03GD1378027 that represent putative resistance sources. CI 2401 is a pure wheat line originating in USSR (Tajikistan), whereas 03GD1378027 is a line derived in South Africa that carries a large rye translocation conferring *D. noxia* resistance. Both solitary apterous virginoparae of biotype 2 and their progeny had reduced survival and prolonged development times on CI 2401 and 03GD1378027 compared to Trego, but the former lines did not differ significantly with respect to either measure of aphid performance.

Progeny developed faster than did their foundress mothers on CI 2401 and Trego, but not on 03GD1378027. Mean foundress fecundity did not differ between CI 2401 and 03GD1378027 but was reduced on these lines relative to Trego. Foundresses were also more often found off plants of CI 2401 and 03GD1378027 than Trego. Estimates of intrinsic rate of increase were higher on Trego than on either CI 2401 or 03GD1378027, the latter two lines yielding similar values. The negative impacts of CI 2401 and 03GD1378027 on development and reproduction of biotype 2 indicate that these lines represent sources of resistance effective against this novel biotype.

We are now approaching the end of a year-long effort to quantify group-feeding benefits in both biotype 1 and biotype 2 RWA feeding on three wheat cultivars (Trego, Stanton and Halt) at two temperatures (20° and 24° C). This series of experiments, that has involved following almost 10,000 individual aphids and aphid colonies on wheat plants, have been surprisingly successful. We have measured substantial benefits (of a group size of ten aphids) on nymphal survival, developmental time, reproductive rate, and lifetime fecundity. However, these benefits are absent on some cultivars (depending on biotype) and are evident only at 20° C for biotype 1 and only at 24° C for biotype 2. For example, when biotype 2 aphids held at 24° C develop in a group of 10 and reproduce alone (on the same plant) as an adult, they reproduce 32% faster and achieve a 44% increase in lifetime fecundity compared to controls that developed in solitude. The fact that we destroyed the groups at onset of reproduction shows (1) that the plant is mediating this effect and that (2) our estimates of the benefits are conservative and could well be greater if the group could had been preserved throughout the reproductive period. These findings have important implications for calculation of intrinsic rates of increase in aphid populations.

We completed a study last fall that demonstrated the mechanism of diapause induction in *Hippodamia convergens*, our primary cereal aphid predator in western Kansas. Adult beetles were collected from sunflower plants and held in four treatments: 1) access to water only, 2) access to sunflower stalks only, 3) eggs of *Ephestia kuehniella* provided ad libitum + water and, 4) greenbug, *Schizaphis graminum* Rondani provided ad libitum. Most females fed greenbug matured eggs in less than a week and only a few entered reproductive diapause. In contrast, more than half of the females fed *Ephestia* eggs, an inferior diet, entered reproductive diapause, and those that matured eggs required an average of almost three weeks to do so. Time to 50 % mortality was 7 days for beetles receiving only water, and 12 days for those receiving only sunflower stalks, whereupon all survivors were fed greenbug. Even after feeding on greenbugs for a month, less than half of the surviving females in these two treatments produced eggs. We conclude that reproductive diapause is an important adaptation for improving *H. convergens* survival during summer when aphids are scarce, although females will forgo diapause if they have continuous access to high quality prey.

We are currently following 300 pairs of *H. convergens*, collected in the same manner as last year, to observed their lifetime fecundity and fertility schedules when provided various temporal patterns of food availability. The population contains a huge range of variation in terms of female reproductive schedules. Some females reproduce immediately without diapause, on a very basic, sub-optimal diet (*Ephestia* eggs once every 3 days, followed by pollen once every 3 days). Others begin reproduction weeks and even many months later, although provision with an optimal diet (greenbugs) will induce rapid and profuse egg production almost immediately. The result is that a large, synchronous cohort emerges from the maturing wheat every spring, having fed on Russian wheat aphid. This is followed by multiple, overlapping and highly skewed generations throughout the rest of the year, culminating in the hibernation of the latest cohorts of adults to mature.

Categories of Resistance in Wheat Cereal Introduction (CI)Tr 2401 to Russian Wheat Aphid Biotype 2 and Identification of New Sources of Biotype 2 Resistance from CIMMYT *Triticum dicoccum*-Derived Synthetic Wheats

Priyamvada Voothuluru, C. Michael Smith, Alan Fritz and Sharon Starkey

Wheat cereal introduction (CI)tr 2401 contains two genes that confer resistance to *D. noxia* biotype 2 (B2). CItr 2401 has a strong antibiosis effect that is exhibited as a reduced intrinsic rate of increase of B2. CItr 2401 plants also exhibit a low level of tolerance to leaf rolling and chlorosis. No antixenosis was detected in CItr 2401. Of 157 *Triticum dicoccum*-derived synthetic lines were evaluated for B2 resistance, two were highly resistant and sustained the same low damage score as the resistant control CItr 2401, and 12 lines were moderately resistant. Interestingly, the 12 *T. dicoccum*-derived resistant lines are also resistant to the Mexican *D. noxia* population and four of the same lines are also resistant to biotype G greenbug, *Schizaphis graminum*. These lines are strong candidates for use in improving the genetic diversity in bread wheat for resistance to different biotypes of *S. graminum* and *D. noxia*. Replicated screening experiments validating the 14 resistant synthetic wheat lines identified and their *Ae. squarrosa* parents are in progress to confirm the sources of this potential B2 resistance.

Russian Wheat Aphid a Potential Concern in Western Kansas in Spring 2005

P. E. Sloderbeck, C. Michael Smith, Sharon Starkey and J. P. Michaud

Surveys of wheat fields in western regions of Kansas indicated that *D. noxia* were present in many fields in the spring of 2005. Infestations were heavier than the several years and spotty infestations were reported as far east as Hays. Samples of plants and aphids were collected from Finney, Greeley and Hamilton Counties, and aphids from each sample were placed on plants of *Dn4* (biotype 1 resistant control), CItr2401 (biotype 2 resistant control), and Jagger (susceptible control) to determine the biotype present in each collection. Aphids from samples collected in Greeley and Finney counties were identified as biotype 1 (B1). Samples from Hamilton County did not survive to establish on test plants. These results indicate that the *D. noxia* collected in Greeley and Finney counties are B1, and that B1 continues to predominate at these locations. B2 is expected to become present in counties near the Colorado border, based on the predominance of B2 in reports from Eastern Colorado.

Inheritance and Mapping of New Greenbug Resistance Genes in *A. tauschii* Germplasm

Lieceng Zhu, C. Michael Smith, Alan Fritz, Elena V. Boyko, and Bikram S. Gill

Genetic mapping of greenbug resistance genes in crop plants using molecular markers provides a powerful way to characterize these genes. The use of molecular markers linked to resistance genes will facilitate selection of greenbug resistant traits in wheat breeding through marker-assisted selection. The physical mapping of greenbug resistance genes using aneuploid and deletion lines will aid in locating the specific chromosome position of these genes and provide information on gene cloning by using map-based procedures. In the present study, we genetically mapped *Gbx*, a wheat gene conferring resistance to current prevalent greenbug biotypes, and physically mapped the microsatellite markers linked to *Gbx* and the related gene *Gbz*. Our results indicated that

Gbx was inherited as a single dominant gene, and this gene was flanked by microsatellite markers *Xgdm150* and *Xwmc157* at distances of 3.3 and 2.7 cM, respectively. Both *Gbx* and *Gbz* were assigned to physical bins of the distal 18% region of the long arm of wheat chromosome 7D by using aneuploid and deletion lines of Chinese Spring wheat.

Categories of Resistance to Greenbug Biotype K in *Aegilops tauschii* Wheat Lines

Liceng Zhu, C. Michael Smith, and J. C. Reese

The wheat lines Largo, TAM110, KS89WGRC4, and KSU97-85-3 conferring resistance to greenbug, *Schizaphis graminum* Rondani, biotypes E, I and K 3 were evaluated to determine the categories of resistance in each line to greenbug biotype K. Our results indicated that Largo, TAM110, KS89WGRC4, and KSU97-85-3 expressed both antibiosis and tolerance to biotype K. Largo, KS89WGRC4, and KSU97-85-3, which express antixenosis to biotype I, did not demonstrate antixenosis to biotype K. The results indicate that the same wheat lines may possess different categories of resistance to different greenbug biotypes. A new cage procedure for measuring greenbug intrinsic rate of increase (r_m) was developed, using both drinking-straw and Petri-dish cages, to improve the efficiency and accuracy of r_m - based antibiosis measurements.

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- Qureshi, J.A., Jyoti, J.L. and J.P. Michaud. Selection of wheat varieties by two biotypes of the Russian wheat aphid (Homoptera: Aphididae). *Ins. Sci.* (accepted).
- Zhu, L. C., C. M. Smith, and J. C. Reese. 2005. Categories of resistance to greenbug (Homoptera: Aphididae) biotype K in wheat lines containing *Aegilops tauschii* genes. *J. Econ. Entomol.* 98: (Accepted).
- Zhu, L., C. M. Smith, A. Fritz, E. V. Boyko, and B. S. Gill. 2005. Inheritance and molecular mapping of new greenbug resistance genes in wheat germplasms derived from *Aegilops tauschii*. *Theor. Appl. Genet.* Online First: <http://dx.doi.org/10.1007/s00122-005-0003-6>.

Montana
2005 State Report: Western Coordinating Committee 66: Cereal Aphids

I. Designated Representatives and Collaborators:

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II. Situation:

A mild winter allowed an overwintering RWA population to persist and infest winter wheat and later spring wheat and barley during the 2005 production season. Montana produces winter wheat, spring wheat, durum wheat and barley. Not only do these crops overlap geographically, often there are several phenological stages of a small grains crop represented in an area available for pest infestation. An estimate of small grain acreage treated for Russian wheat aphid is approximately 50,000 to 75,000 acres in 2005.

III. Biology and Management:

Breeding of Russian wheat aphid varieties is a primary emphasis in states where the RWA is a consistent pest with significant economic impacts. However, RWA in Montana is sporadic in occurrence, causing variable widespread economic damage (Table 1). Therefore, the emphasis of research and extension programs in Montana is on RWA monitoring and treating as populations reach economic thresholds. Management guidelines including monitoring, economic thresholds and treatment guidelines are published in the High Plains Integrated Pest Management Guide <highplainsipm.org>.

Insecticides labeled for RWA control in wheat offer effective and reasonable cost for control alternatives. However, the situation with barley is more difficult. The insecticides labeled for RWA control in barley are either not readily available in Montana, are quite costly or have toxicities that render them undesirable for ground or aerial application. Field trials of newer insecticide products are being examined for potential use through special labels for RWA control on barley (Blodgett).

Table 1. Summary of Russian wheat aphid treated acreage in Montana, 1987-present.

Year	Estimated acreage treated for RWA
2005	50,000 – 75,000
2004	<2,500
2003	<1,000
2002	50,000
2001	10,000
2000	150,000
1993 – 1999	<1,000
1992	250,000
1987-1989	100,000+

IV. Current Accomplishments

Biological Control: Pitfall traps were established at a cropping system site located at the Roosevelt-Sheridan County Water Conservation District farm in Froid, MT. Carabid beetles were collected and are being identified. A publication is in preparation.

Host Plant Resistance: Russian wheat aphid samples from the Judith Basin and the Gallatin Valley were submitted to Colorado State University (F.B. Pears) for biotyping. Both samples were identified as RWA biotype 2.

V. Publications

Tharp, C.I., S. Blodgett and K. Kephart. 2004. Susceptibility of cereal leaf beetle (*Oulema melanopus*) in malting barley with various foliar insecticides, Huntley, MT 2004. *In* 2004 IPM Research at Montana State University, Department of Entomology. < <http://scarab.msu.montana.edu/IPM/Crop%20Research%20Bulletin%202004.htm>>.

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Nebraska Report to the WCC-066 Committee - 2005
Integrated Management of Russian Wheat
Aphid and Other Cereal Aphids

Nebraska Representative:

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Russian wheat aphid status in 2005:

Through the last three years, we have seen increasing presence and problems with Russian wheat aphid activity in western Nebraska. In 2005 Russian wheat aphid infestations were again light in most fields through early spring; however, a number of economic infestations were present in winter wheat fields in the Panhandle. By late May, we had seen an influx of winged Russian wheat aphids that infested late developing wheat fields and spring barley fields. As in the previous couple years, winter wheat fields that were delayed in development and spring barley fields were most likely to be heavily infested with aphids.

Several locations in the Nebraska panhandle were sampled to determine the biotype of Russian wheat aphid present. Aphids from these samples were transferred to pots containing three wheat varieties, Yuma (susceptible), Yumar (biotype 1 resistant) and Gamtoos (biotype 2 resistant). Four of seven samples were found to be biotype 2 (or a mixture of 1 and 2) and the remaining to be biotype 1. None were able to severely damage Gamtoos.

We received more normal precipitation through the spring and summer of 2005, and as a result, the aphid's alternate host grasses have stayed much greener. We expect that this will increase the current risk from Russian wheat aphids as we move into the fall.

Research/Extension Activities:

Areawide IPM Project:

Project personnel: Gary Hein, Drew Lyon, Paul Burgener, John Thomas, Rob Higgins, Dave Christian

We are evaluating diversified cropping systems incorporating aphid-resistant cultivars compared to the wheat-fallow systems with regard to economic, agronomic, and pest management parameters. In western Nebraska and eastern Wyoming, we are monitoring two pairs of diversified and wheat-fallow fields. Better growing conditions the past year have resulted in a more normal year. For the first time in four years, wheat yields were close to normal yields. Russian wheat aphids were present in all fields but field-wide economic infestations were not seen in any of the fields.

During the spring we used field cages to differentially screen out natural enemies to determine their effect on aphid populations in two of these fields. Fine-mesh (52 mesh) cages were used to exclude all predators and parasites, 20-mesh screen was used to exclude predators only, regular cages left open at the bottoms and no cage controls were used to allow free exchange of natural enemies. Greenhouse colony aphids were used to infest the plots early in the spring, and aphid and natural enemy populations were monitored through the rest of the season. Preliminary data indicate that significant aphid populations built up in all plots but greater aphid numbers were seen in the screened cages where predators and parasites were excluded. Further evaluation of the data will indicate the extent of differences found in the two fields sampled.

Extension activities have been focused on getting the word out that the Russian wheat aphid is a significant threat again after a number of years of it being very low in numbers. The use of resistant varieties was never high in Nebraska, but because few economic infestations were seen for over ten years, most growers do not think about scouting for the aphid. We are strongly encouraging growers to scout again for aphids, both in the fall and early spring, to catch infestations before they get to be extreme and much more difficult to control. We have begun to update our Extension publications with the new information on biotypes and the potential difficulties with previously resistant varieties. We have recently rewritten and reprinted the following NebGuide:

Hein, G. L., J. A. Kalisch, and J. Thomas. 2005. Cereal aphids. Univ. of Nebraska, Cooperative Extension, G96-1284.

Physiological Impacts of Cereal Aphid Feeding:

Project Personnel: Leon Higley, Tiffany Heng-Moss, Gautam Sarath, Lisa Franzen, and Andrea Gutsche

Differential Gene Expression of Barley in Response to Aphid Injury

Research Objectives:

- To use the impaired transport/end product inhibition model of plant-aphid interaction to establish temporal patterns of barley physiological responses to injury by various aphid species (including initial injury and recovery)
- To initiate differential gene expression of aphid injured and uninjured barley through the use of microarray analysis
- To confirm differential expression of genes associated with aphid injury through standard molecular techniques documenting changes at the mRNA level.

Our long-term goals are to extend these findings to other aphid-crop interactions (Russian wheat aphid-wheat, greenbug-sorghum, and soybean aphid-soybean) and the model plant system of green peach aphid-Arabidopsis.

Physiological and Biochemical Responses of Resistant and Susceptible Wheat to the Russian Wheat Aphid

Research Objectives:

To document changes in photosynthesis as a major mechanism for plant resistance to the aphids and investigate the impact aphid feeding has on the photosynthetic responses of wheat.

To explore the role of plant proteins in the defense response of susceptible and resistant wheat cultivars to aphid treatments and investigate protein and oxidative enzyme levels of plants after aphid feeding.

WCC-66 Cereal Insect Pests North Dakota Report 2005

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North Dakota Aphid Population Monitoring

For the last eight 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 of these surveys can be found at:

www.ag.ndsu.nodak.edu/aginfo/ndipm/05IPMSur/HTML/WheatIPMSurvey.htm.

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.

The 2005 growing season had few unusual features. North Dakota had a relatively warm spring which allowed many cereal farmers, especially those in the south, to plant early. Temperatures and moisture were good throughout the season throughout the state. The latter meant that scab levels were increased over 2004, especially in the SE part of the state (see maps). The quality and yields of the 2005 crop are predicted to be good except for moisture-related problems.

In 2004 aphids were not considered to be a problem in North Dakota and in 2005 they were even less of a problem (see maps). Phil Glogoza (Minnesota Extension) reported that the highest levels of aphids he saw all summer were 25-50% of stems with aphids. A number of predator species were present in fields throughout the summer. Regular heavy rain showers and high humidity may also have contributed to suppression of aphids. Barley yellow dwarf virus was observed at higher levels in some fields in SE North Dakota (see maps).

The second subject of the NDSU Survey, the cereal leaf beetle, was not observed by any scouts in 2005.

Grasshoppers, the third subject of the NDSU survey, were also not a problem in 2005. Only a few fields in the western crop reporting districts showed more than 15 adults per square yard. The majority of sweeps in our survey indicated from 1-7 adults per sweep, and 1 to 24 nymphs per sweep.

One other pest of wheat that was mentioned in 2005 by several wheat growers was wheat stem maggot. This pest usually occurs at low levels of 1-5% in North Dakota wheat fields. In some areas it seemed to occur at higher levels in 2005.

However, by the time damage from wheat stem maggots is detected (“whiteheads”) it is too late to treat the crop.

Other Insect Pests of 2005 Cereals in North Dakota

In recent years North Dakota wheat commodity groups (e.g. North Dakota Wheat Commission and Dakota Growers) and North Dakota State University wheat breeders have shown little interest in developing aphid-resistant cereals. This lack of concern about aphids, and insects in general, probably arose because of more serious problems with cereal diseases, e.g. wheat scab.

The only insects that have competed with wheat scab for the attention of wheat farmers are the orange wheat blossom midge, *Sitodiplosis mosellana* (Gehin), the wheat stem sawfly, *Cephus cinctus* (Norton), and the Hessian fly, *Mayetiola destructor* (Say). None of these three insects is part of the NDSU summer survey of insects in cereals. Qualitative information about the wheat stem sawfly and Hessian fly comes mostly from county agents and from wheat breeders who sometimes notice when their research plots have been attacked. In the 2005 field season, we received no reports of sawfly or Hessian fly damage.

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.

The autumn 2004 survey of overwintering wheat midge larvae (see maps at www.ag.ndsu.nodak.edu) included samples from northern counties of North Dakota as well as four counties in the northeastern corner of Montana. Wheat midge larval numbers were extremely low across the surveyed regions. Parasitism rates (probably *Platygaster* spp.) continued to be high. Low estimates of wheat midge numbers from the autumn 2004 soil survey created an expectation in the farming community that wheat midge numbers in the 2005 field season would also be low. There were no reports of high wheat midge populations and few reports of spraying.

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 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 but become active when harvested wheat is loaded into trucks. At this time, the orange larvae are seen dropping from harvested wheat in trucks to the soil.

It also should be noted that, relative to 2004, we had no trouble finding a wheat field with wheat midge-infested heads in 2005. Each August our lab collects up to 50,000 third instar wheat midge larvae from the field to be used the following spring for testing resistance in wheat. Levels of larvae in this 2005 easily-found field north of Minot, ND were above the economic threshold. There also were some 2005 reports of wheat midge levels above threshold and growers adding an insecticide to fungicide sprays for scab. Clearly, because the area planted to wheat in North Dakota is so vast and because the wheat midge, wheat stem sawfly and Hessian fly are difficult to detect, additional tools for monitoring are needed.

A female-produced sex pheromone for the wheat midge has been identified (Gries et al., 2000) and in 2005 became available commercially. Benefits of pheromone-based monitoring program for gall midges include sensitivity and convenience (Harris and Foster, 1999). We tested the pheromone trapping system for wheat midge and caught small numbers of males on campus where no mention has ever been made of wheat midges as pests. A limitation is that a female-produced sex pheromone only attracts males. Male wheat midges do not appear to move from eclosion sites to new wheat fields (as do females) and thus may not reflect the degree to which the crop will be attacked by ovipositing females.

The major component of the Hessian fly sex pheromone was identified in 1990 but is not an effective lure in the field (Harris and Foster, 1999). We are working in collaboration with two European labs, one in Sweden (electrophysiologist: Ylva Hilbur) and one in Germany (chemist: Wittko Franke) to identify the additional component(s) of the Hessian fly pheromone. In 2005 we tested, in small wheat plots, a three component blend of the main component and two other components identified by Dr Hilbur as being electrophysiologically-active (by EAG). This blend caught significantly more males than a control or the main component alone. In October 2005, David Buntin of the Georgia Experiment Station will run a field test of the three component blend, testing for significant activity and type of formulation.

Applied and Basic Research on Wheat Pests at NDSU

The theme of our research is the interactions that occur between insects and plants. The interactions we study are of two types, first, antagonistic interactions

in which the insect parasitizes the plant and second, mutually beneficial interactions in which the plant benefits via insect-mediated pollination and the insect benefits via a food source. Our goal is to document behavioral, ecological, and physiological mechanisms underlying insect-plant interactions. The questions we ask range from fundamental questions of interest to scientists who study behavior, ecology, physiology, evolutionary biology and molecular genetics, to applied questions of interest to plant breeders, pest management specialists, and conservation land managers. Only research on antagonistic interactions will be discussed here.

Antagonistic relationships between insects and plants are considered to be evidence of antagonistic coevolution as defined by the following events. The insect increases its fitness by attacking and successfully exploiting a plant. The reduced fitness of the attacked plant favors the selection of a novel plant defense that spreads through the plant population. This reduces the fitness of the insect and selects for a virulent insect genotype that spreads through the insect population. The rates at which plant defense and insect virulence reciprocally evolve depend on the genetics and ecology of the plant and insect, as well as the costs of defense and virulence.

In our research we study antagonistic relationships between wheat and two insects that attack during different stages of wheat development, the Hessian fly, which typically attacks during the seedling stage, and the orange wheat blossom midge (hereafter referred to as wheat midge), which attacks during early development of the seed. Both the Hessian fly-wheat and the wheat midge-wheat systems are fortunate in that one of the earliest stages of the interaction is mediated by avirulence (*avr*) genes of the insect and resistance (*R*) genes of the plant.

The Hessian fly-wheat interaction is the best characterized all of known *R* gene/*avr* gene-mediated insect-plant interactions. Thirty-one *R* genes for Hessian fly resistance, designated *H1-H31*, have been identified in wheat and its various relatives. The plant defense associated with these *R* genes has a dramatic and easily quantifiable effect on the fitness of the Hessian fly, i.e. the first instar larva dies within days of first attack. Hessian fly adaptation to *R* gene defense occurs via modifications in a corresponding larval *avr* gene and has a dramatic and easily quantifiable effect on the fitness of the insect, i.e. the first instar larva now survives and grows on the *R* gene plant. Wheat *R* genes and Hessian fly *avr* genes have been the subjects of classical genetic studies for over 50 years and now are the subjects of mapping and cloning studies as well as plant and insect gene expression profiling (USDA-ARS and Purdue).

Many of the hypotheses we test in our lab come from two models/concepts. The first is the gene-for-gene concept, centered on the interplay between a major *R* gene in a host plant and a corresponding *avr* gene in a plant parasite (Flor 1946). With few exceptions, the genetics of the Hessian fly-wheat interaction fits this

model well. The second important model is the “elicitor-receptor” model. In this model, *R* alleles encode “receptor” proteins capable of triggering inducible defense reactions when they bind to an appropriate ligand. *A* alleles in the parasite encode this ligand, the so-called “elicitor” of the defense response. The binding of an *A*-encoded elicitor to an *R*-encoded receptor triggers a defense response in the plant, killing the parasite. Plant genotypes lacking the dominant *R* allele fail to produce the ‘receptor’ and thus do not initiate a defense response. Similarly, when the parasite lacks the functional *A* allele, no elicitor is made, and the parasite avoids detection and succeeds in establishing a feeding site. An alternative model of *R* gene/*avr* gene interactions proposes an indirect interaction between the *R* gene product and the *avr* gene product, with the *R* gene product guarding a target of the *avr* gene product (the guard hypothesis, Dangl and Jones 2001).

Within the well-established group of researchers studying the Hessian fly-wheat interaction, our niche is phenotyping. Thus while molecular biologists at Purdue and USDA-ARS map and clone *avr* and *R* genes and study gene expression, we have documented and measured larval behavior and plant responses. Plant and insect responses have been documented both by simple plant/insect growth studies and by light and electron microscopy studies through collaboration with Dr Tom Freeman, NDSU Microscopy Lab. Key discoveries from our NDSU Hessian fly research are:

1. virulent Hessian fly larvae manipulate nutrient allocation within the wheat plant, establishing a nutritive tissue at the base of the plant where they feed (see attached Abstract of manuscript in press for Annals of Ent Soc),
2. *R* gene-defended wheat plants block the establishment of the nutritive tissue,
3. *R* gene-defended plants suffer growth deficits and therefore do not block all effects of Hessian fly attack,
4. *R* gene-defended plants do not all exhibit the same response to attack,
5. *R* gene-defended plants suffer little if any yield loss from the joint effects of larval attack and plant defense,
6. the adult female cannot distinguish between plants with and without *R* genes but can distinguish between plants with and without larvae, and between the responses of different *R* gene plants to larval attack.

Our approach has been strongly supported by molecular biologists studying Hessian fly-wheat interactions. We are in regular contact and have developed our research in parallel by studying the same interactions, i.e. using the same *avr/R* gene pairs. Dr Freeman and I have a formal cooperation with Drs Jeff Stuart (Purdue) and Ming-shun Chen (USDA-ARS, Kansas State) through a recently-awarded USDA/CSREES NRI research grant with four hypotheses (3-yr grant):

HYPOTHESIS ONE: Across all incompatible interactions there is a single type of plant defense and a single time of larval death.

HYPOTHESIS TWO: When larvae lacking the AVR product attack a R gene plant they do not encounter localized defenses and establish a normal nutritive tissue.

HYPOTHESIS THREE: When a R gene plant is attacked by both larvae lacking and larvae producing the AVR product, both types of larvae survive because defense against larvae producing the AVR product is localized.

HYPOTHESIS FOUR: During the early stages of Hessian fly attack, larvae inject secreted salivary proteins into plant epidermal cells.

The situation for our research on a second pest of wheat, the wheat midge, is quite different. While Hessian fly has been studied by both ecologists and geneticists for over 50 years, the wheat midge was studied rarely until the mid 1990s when a research group in Canada discovered the first effective trait for resistance. Subsequently this resistance was shown to be conditioned by a single gene, now referred to as *Sm1*. Because the wheat midge is a devastating pest of hard red spring wheat and durum wheat throughout the Northern Great Plains of Canada and the USA, this was an important discovery but one that would need to be managed carefully to avoid the evolution of virulence to *R* genes seen in related insects, e.g. Hessian fly. A major research effort on wheat midge and *Sm1* was initiated by Dr Robert Lamb and his colleagues at the Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg.

By the time I arrived at NDSU in 2000, many basic research questions about wheat midge-wheat interactions had been answered by Lamb et al. Thus, our focus for the wheat midge has been more applied, emphasizing practical considerations for the wheat growers of North Dakota. In collaboration with Drs Berzonsky, Elias, and Mergoum of NDSU Plant Sciences we have achieved the following in our research on the wheat midge:

1. determined that effective resistance against wheat midge was not present in North Dakota spring and durum wheats or advanced breeding lines,
2. developed a methodology, using greenhouse-grown plants and double haploids, to move *Sm1* into North Dakota adapted hard red and white spring wheats,
3. incorporated *Sm1* into NDSU wheat germplasm lines,
4. developed plant populations that are being used by USDA-ARS scientists (BRL, Fargo) to look for genetic markers for *Sm1*,
5. determined that plant traits affecting host selection have a strong effect and might be used to complement resistance conferred by *Sm1*, and
6. developed methodologies for studying feeding mechanism of wheat midge larvae and the mechanism of resistance conferred by *Sm1*.

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Annals of the Entomological Society of America (2005, in press)

Virulent Hessian Fly (Diptera: Cecidomyiidae) Larvae Induce a Nutritive Tissue During Compatible Interactions with Wheat

M.O. Harris, T.P. Freeman, O. Rohfritsch, K.G. Anderson, S.A. Payne, and J.A. Moore

ABSTRACT The compatible interaction between virulent Hessian fly *Mayetiola destructor* (Say) larvae and susceptible wheat *Triticum aestivum* L. plants was investigated at the light-microscope and ultrastructural levels. During the first day of larval attack of the abaxial surface of the sheath of the third leaf of a wheat seedling, small punctures of the appropriate size (0.1 μm diam) and spacing of the paired larval mandibles were found in the outer wall of epidermal cells. Inside epidermal cells, nuclei and cytoplasmic organelles appeared to be breaking down, and the number and size of cytoplasmic vacuoles had increased. Two to three days after initial larval attack, cells at the base of the third leaf showed distinct modifications: epidermal and mesophyll cells showed signs of becoming nutritive.

Nutritive cells were identified at the light-microscope level by an increase in cytoplasmic stainability, and at the ultrastructural level by increased numbers of cellular organelles (e.g. mitochondria, proplastids, Golgi, rough endoplasmic reticulum, numerous small vacuoles) and an enlarged nucleus. Breakdown of the nutritive tissue began soon after it was first observed. One of the first indications of this breakdown was a change in the shape and density of the nucleus. Prior to the rupture of nutritive cells, cell walls appeared to be thinner. Structural changes were not restricted to the leaf directly fed upon by larvae. The sixth leaf, a leaf more recently initiated by the shoot apical meristem, was found to consist primarily of well-developed epidermal layers, with poorly-developed mesophyll cells. The implications of these findings for understanding incompatible interactions between avirulent Hessian fly larvae and *R* gene-defended plants are briefly discussed.

WERA 2005 Report from Oklahoma

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Collaborating agencies:

ARS-PSWCRL, Stillwater, Texas A&M University.

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Cooperating OSU Scientists:

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Makuena Lebusa (M.S. Royer)

Matt Rawlings (M.S. Giles)

Sara Donelson (Ph.D, Giles)

Christopher Mullins (M.S. Giles)

Projects in 2005

Overwintering Ecology of *Lysiphlebus testaceipes* in winter wheat. Doug Jones, Kristopher Giles. Field and laboratory experiments were completed to determine the ability of *Lysiphlebus testaceipes* to function and survive a range of temperatures common during winters in Oklahoma. Results indicate that *L. testaceipes* is able to survive temperatures as low as -6 Celsius and successfully oviposit. Most interestingly, however, *L. testaceipes* successfully attacks greenbugs when ambient temperatures are at 4 Celsius, which is below the developmental threshold for the wasp and aphid.

Rice Root Aphid. Matt Rawlings, Kristopher Giles. Field and laboratory studies are continuing to evaluate the pest potential of *Rhopalosiphum rufiabdominalis* Sasaki in winter wheat. Sampling data demonstrated that the aphid is present throughout the Oklahoma. Initial field and greenhouse evaluations of aphid level versus wheat growth and yield indicate that this aphid has little to no effect on wheat. This data contradicts recently published data; additional work is continuing to refine experimental approaches.

Intraguild Interactions among *Schizaphis graminum*, *Lysiphlebus testaceipes*, and Coccinellidae in Winter Wheat. Makuena Lebusa, Tom Royer; Christopher Mullins Kristopher Giles. Laboratory and field experiments are continuing to evaluate the interactions among common coccinellid beetles and *Lysiphlebus testaceipes* in relation to their common prey, *Schizaphis graminum*. Makuena Lebusa completed laboratory studies demonstrating reduced suitability of mummified aphids for coccinellid predators and published a thesis entitled: **Suitability of *Lysiphlebus testaceipes*-parasitized greenbugs (*Schizaphis graminum*) as a food source for predatory coccinellidae.** C.

Mullins will expand 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.

USDA-ARS. Areawide Pest Management Program. Elliott, N. et al.

In cooperation with postdoctoral fellow Sean Keenan, continued studies examining the management characteristics of wheat producers throughout the central plains. Results have been summarized and submitted as a book chapter. In cooperation with Dr. Norman Elliott and Postdoctoral Fellows Dr. Mpho Phoofolo and Vasile Catana continued studies examining the ecology arthropod predators in simple versus diverse agricultural systems. Continuing in 2005.

Synopsis of Arthropod Pest Activity in Wheat, 2004-05

Aphid numbers were relatively low, but early season *Rhopalosiphum padi* populations were of concern to some growers. *Mayetiola destructor* was reported in several locations, especially in those fields that were produced under conservation tillage.

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SOUTH DAKOTA, WCC-66 ANNUAL REPORT, 2005

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Overview of Cereal-Aphid Research Activities and Accomplishments:

1. Meaningful sources of plant resistance are needed against the bird cherry-oat aphid, *Rhopalosiphum padi*, a worldwide pest of wheat. Moderate levels of resistance to this aphid were found in several lines of triticale and low levels of resistance in two wheat accessions. Follow-up studies with triticale accessions are in progress.

2. Research continues on host suitability, rearing, and economic impact of the rice root aphid, *Rhopalosiphum rufiabdominalis*, on small grains. Collaborative research on rice root aphid continues with the USDA-ARS Plant Science Laboratory in Stillwater, OK.

Publications

Hesler, L.S. and C.I. Tharp. 2005. Antibiosis and antixenosis to *Rhopalosiphum padi* among triticale accessions. *Euphytica* 143:153-160.

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Report from Texas

Understanding the Mechanisms of Host Resistance to Greenbug in Wheat

Yiqun Weng, G. Jerry Michels, Revindra N. Devkota, Jackie C. Rudd
(Texas Agricultural Experiment Station, Amarillo)

1. Induced resistance in greenbug resistant and susceptible wheat lines

Interaction between biotype E greenbugs and two near isogenic lines of Gb3 (TXGBE273 and TXGBE 281) was examined for 65 days after infestation. We demonstrated that systemic acquired resistance (SAR) was inducible in the resistant, but not the susceptible genotype by preconditioning with biotype E greenbugs for 48 hours. *Gb3*-mediated SAR reduced the size and buffered the fluctuation of the aphid population and extended the life of host plants. Expression of SAR in resistant plants was spatially and temporally dynamic. SAR was able to reduce the aphid population size, delay aphid density peaks and extend the life of the leaves. These effects were stronger in younger leaves. This work has been published in the *Journal of Economic Entomology* (JEE, 2005, 98:1024-1031).

2. Molecular mapping of greenbug resistance Genes in wheat

A new source of *Aegilops tauschii*-derived greenbug resistance was identified in a synthetic hexaploid wheat line W-7984. Genetic analysis suggested that a single, dominant gene governs the greenbug resistance in W-7984, which was placed in chromosome arm 7DL by linkage analysis with molecular markers. Allelism tests revealed that the greenbug resistance in W-7984 and Largo (*Gb3*) was controlled by two different genes in 7DL. Using a target mapping strategy, a genetic map of *Gb3* was constructed. One co-segregating and four closely linked markers with *Gb3* were identified. Deletion mapping placed *Gb3* into the telomeric 18% region of 7DL. We suggest that the greenbug resistance gene in W-7984 be designated as *Gb7*. Details of this work is available at Theoretical and Applied Genetics (2005, 110:462-469).

3. Molecular defense response to greenbug feeding in wheat

Gene expression profiling was conducted using cDNA-AFLP in *Ae. tauschii* accession PI268210, the donor of *Gb3* in wheat. Of 141 transcript-derived fragments (TDFs) cloned and sequenced, 122, 15 and 4 were up, down or transiently regulated, respectively within 48h of greenbug infestation. Many of the upregulated genes encode proteins or enzymes that are of importance in defense responses and signal transduction pathways against aphid feeding. No PR genes were identified. A significant portion (15%) of the upregulated genes belong to retroelements. Over one quarter of TDFs didn't find either BALSTx or BLASTn or both matches. It seems that JA and ethylene play important roles in signaling defense responses against the phloem-feeding greenbug. Sequences of 121 cDNAs from this study have been submitted to NCBI dbEST (GenBank IDs: CX244546 to CX244666).

4. Roles of plant volatile emission in host resistance against the greenbug in wheat

Previous studies indicated that antixenosis is an important component of *Gb3*-mediated host resistance to the greenbug. We reasoned that plant volatiles are the players in this process. We examined the volatile profiles of the uninfested resistant and susceptible NILs of *Gb3* using the SPME (solid phase micro-extraction)/GC-MS technology. No qualitative difference in volatile components in the two lines was found, but quantitative difference was significant. We also compared the volatile profiles of the two NILs before and after aphid feeding, and qualitative difference in volatile components was found in each line before and after infestation, and between the two lines after infestation.

Spectral Measurement of Greenbug (Homoptera: Aphididae) Density and Damage to Wheat Growing under Field and Greenhouse Conditions

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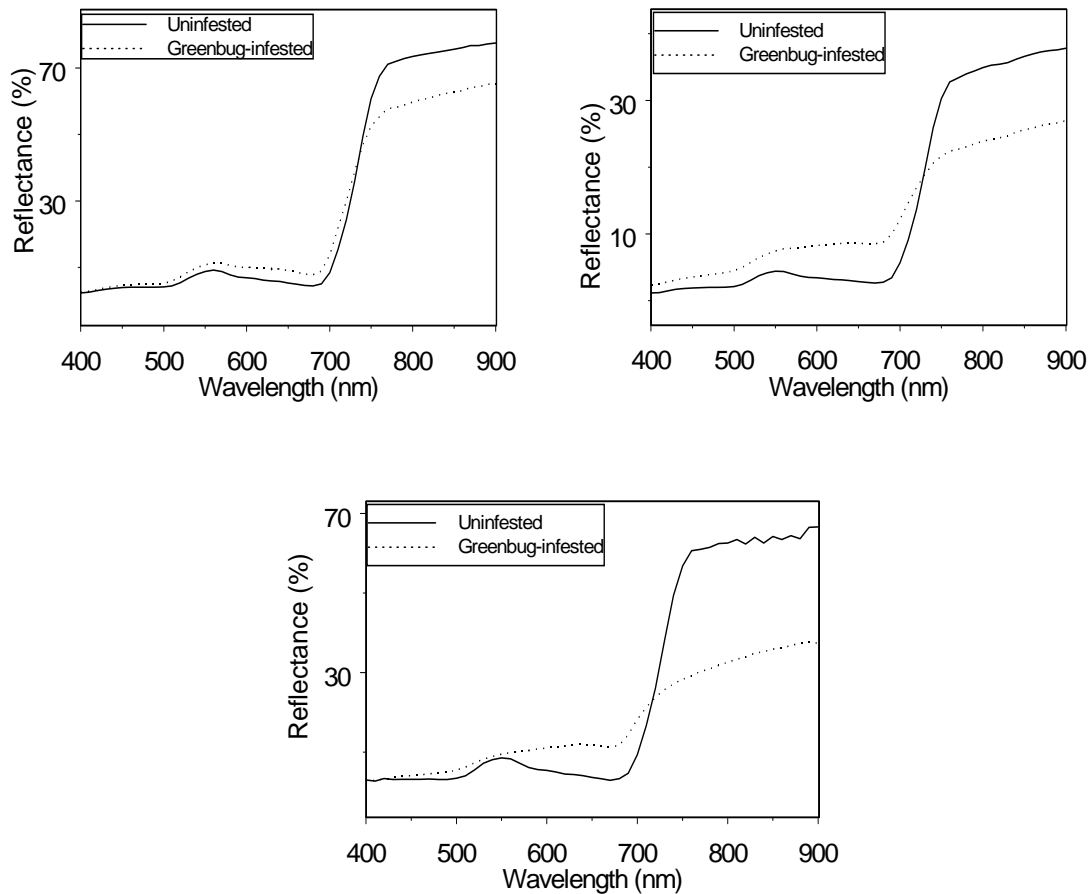
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Although spectral remote sensing techniques have been used to study many ecological variables and biotic and abiotic stress to agricultural crops over decades, the potential use of these techniques for greenbug (*Schizaphis graninum* Rondani) infestations and damage to wheat (*Triticum aestivum* L.) under field conditions is unknown. Hence, this research was conducted to investigate: 1) the applicability and feasibility of using a portable narrow-banded (hyperspectral) remote sensing instrument to identify and discern differences in spectral reflection patterns (spectral signatures) of winter wheat canopies with and without greenbug-damage, and 2) the correlation between miscellaneous spectral vegetation indices and greenbug density in wheat canopies growing in two fields and under greenhouse conditions. Both greenbug and reflectance data were collected from 0.25, 0.37, and 1 m² plots in one of the fields, greenhouse, and the other field, respectively. Regardless of the growth conditions, greenbug-damaged wheat canopies had higher reflectance in the visible range and less in the near infrared regions of the spectrum when compared with undamaged canopies. In addition to percentage reflectance comparison, over 150 spectral vegetation indices drawn from the literature were calculated and correlated with greenbug density. Correlation analyses revealed moderate to high correlations (r ranged from minus 0.94 to 0.41) between greenbug density and spectral vegetation indices. Hyperspectral remotely sensed data with an appropriate pixel size have the potential to portray greenbug density and discriminate its damage to wheat with repeated accuracy and precision.

Figure 1: Percentage spectral reflectance (400-900nm range) of greenbug-infested and uninfested winter wheat canopies associated with two field and one greenhouse experiments. Field 1 – Moore County, near Dumas, TX (upper left); Field 2 - Hardeman County near Chillicothe, TX (upper right); and a greenhouse experiment at the Texas Agricultural Experiment Station facilities at Bushland, TX (bottom).



Using digital image analysis and spectral reflectance data to quantify greenbug (Homoptera: Aphididae) damage in winter wheat

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The usefulness of digital image analysis and spectral reflectance data to quantify greenbug damage (*Schizaphis graminum* Rondani) was evaluated for two winter wheat (*Triticum aestivum* L.) fields, three field experiments, and one greenhouse setting in Oklahoma and Texas. A hyperspectral field spectrometer and a digital camera were used to record reflectance and to acquire images over 0.25 m², 0.37 m², and 1 m² greenbug-

damaged wheat canopies. A large number of spectral vegetation indices compiled from the literature were calculated and their relations with greenbug damage were investigated. The mean percentage greenbug damage estimated through digital image analysis varied from $13 \pm 1/0.25 \text{ m}^2$ to $73 \pm 7/0.37 \text{ m}^2$. The mean greenbug density ranged from $191 \pm 22/0.25 \text{ m}^2$ to $54,209 \pm 7,908/0.37 \text{ m}^2$. Correlation analyses showed that there were very strong associations between greenbug damage in wheat and spectral vegetation indices. Correlation coefficient ranged from 0.82 to -0.98. Thus, remote sensing using spectral reflectance and digital images can be nondestructive, rapid, cost-effective, and reproducible techniques to determine greenbug damage in wheat with repeated accuracy and precision. Together with the existing spectral indices, two versions of a new index algorithm are also suggested in this paper.

Reflectance characteristics of Russian wheat aphid (Homoptera: Aphididae) stress and density in winter wheat

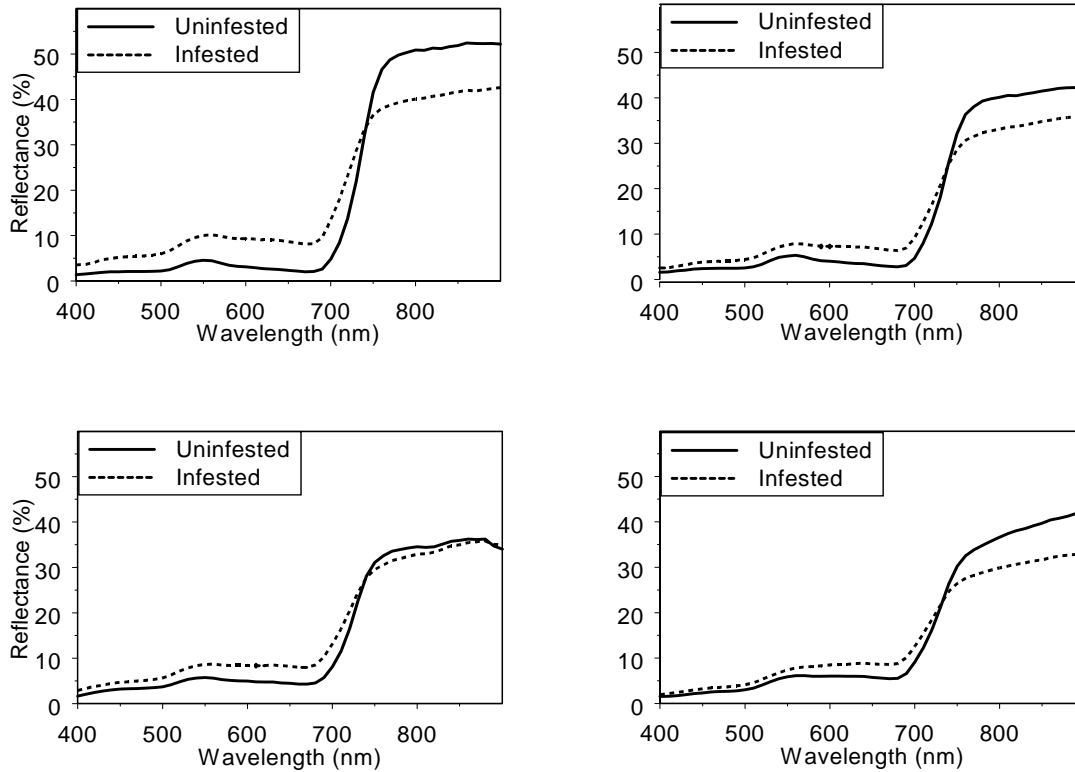
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The Russian wheat aphid (*Diuraphis noxia* Mordvilko) infests wheat (*Triticum aestivum* L), barley (*Hordeum vulgare* L.), and other certain small grains and grasses. Russian wheat aphid infestations are unpredictable in time and space. In favorable conditions, Russian wheat aphid feeding can result in heavy destruction of wheat and barley in a short period of time. Cumulative economic losses from Russian wheat aphid infestation in wheat and barley in the US have been estimated nearly at \$1 billion since its discovery in Texas in 1986. Of this damage, 60% has occurred in the Texas and Oklahoma Panhandles, northeastern Colorado, western Kansas, and southwestern Nebraska. A monitoring strategy that allows for the rapid assessment of aphid infestation and damage repeatedly over the growing season is critically needed. Tracking the irregular infestation patterns of Russian wheat aphid in order to optimize control efforts is center to the successful management of this aphid. One method that has been shown over number of years to be useful for monitoring insect outbreaks is to measure the light reflected by the infested canopy, plant, or leaf. Hence, this research was designed to investigate: 1) the potential use of remotely sensed data to discern and identify differences in spectral reflection patterns (spectral signatures) of winter wheat canopies with and without Russian wheat aphid infestation, and 2) the relationship between a large number of vegetation indices and Russian wheat aphid density in wheat canopies growing in field conditions. Russian wheat aphid-infested wheat canopies had significantly lower reflectance in the near infrared region and higher in the visible range of the spectrum when compared with uninfested canopies. Linear regression analyses resulted in poor ($R^2 = 0.26$) to strong ($R^2 = 0.90$) relationships between vegetation indices and Russian wheat aphid density. These results indicate that remotely sensed data with an appropriate pixel size have the potential to describe Russian wheat aphid density and distinguish its damage to wheat with repeated accuracy and precision.

Figure 2: Percentage spectral reflectance (400-900nm range) of Russian wheat aphid-infested and uninfested winter wheat canopies associated with three fields. Field 1 – Deaf Smith County near Amarillo, TX, sampled on 21 April (upper left); n =15 and sampled on 17 May 2004 (upper right), n = 15. Field 2 – Prowers County near Lamar, CO, Sampled on 20 May, 2004 (bottom left); n = 25. Field 3 – Cimarron County near Boise City, OK, Sampled on 18 May 2005 (bottom right); n = 20.



WCC-066 REPORT for the Period of Sept. 2004 through Sept. 2005
(Compiled by Dolores W. Mornhinweg)

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II. OVERVIEW OF CURRENT RESEARCH AND ACCOMPLISHMENTS

A. Remote sensing of cereal aphids

We have shown that multi-spectral remotely sensed data was sensitive to variation in the density of Russian wheat aphids in production wheat fields. In four fields studied over two years, there were lower NDVI values for highly infested plots than for less infested plots. Despite the fact that the fields were drought stressed, Russian wheat aphid presence could still be identified using the NDVI values for each plot. There were significant correlations between Russian wheat aphid density and NDVI for all fields. Lower NDVIs were found in plots with higher Russian wheat aphid densities indicating that the additional stress caused by Russian wheat aphids in the drought stressed field was evident in the imagery. Results of this study were encouraging, and indicate that further research is warranted to determine whether multi-spectral remote sensing can be used for detecting Russian wheat aphid infested fields in operational pest management programs for the pest.

B. Characterization of RWA Biotypes, their Ecology, and Biotypic Diversity

The reactions of two primary sources of Russian wheat aphid (RWA) resistance in barley, STARS 9301B and 9577B, to Biotypes RWA1 to RWA5 were studied to determine if new sources of RWA resistance in barley were needed. We found that STARS 9301B was universally resistant to all of the current RWA biotypes and 9577B showed susceptibility to certain RWA biotypes. We recommend that STARS 9301B be used in barley breeding programs aimed at developing RWA resistant barley cultivars. Approximately 70 sites were established in 2004 and 2005 in Texas, Oklahoma, New Mexico, Colorado, Nebraska and Wyoming to collect RWA from wheat, barley and wild grass hosts in order to establish clonal colonies for biotype determinations and to study seasonal ecology. These studies are ongoing.

C. Genetics of Russian wheat aphid biotypes in the US

RUSSIAN WHEAT APHID (HOMOPTERA: APHIDIDAE): WITHIN BIOTYPE RWA2 VARIATION FEEDING REACTION OF DN4 WHEAT AND STARS-9301B BARLEY

Since 2003, many areas of the world (including the USA) has seen the occurrence of *Diuraphis noxia* (Mordvilko) populations capable of injuring wheat containing the Dn4 resistance gene. These have been termed biotype RWA2, and are identified by observations of plant reaction by *D. noxia* feeding. Most research has focused on variation of feeding damage by a biotype to different resistance genes or sources. Little is known about variation occurring within biotypes in the ability or degree to cause damage to a single resistance source. We evaluated five single maternal lineages (clones) of the *D. noxia* biotype RWA2 from Colorado and Texas virulent to Dn4 wheat, for variation in ability to cause plant injury on both susceptible and resistant wheat and barley. Variation to cause injury on both susceptible and resistant varieties of wheat and barley was found within RWA2. These results argue against aphid biotypes as being single genotypes, and suggest that gene flow occurs within and between biotypes, or there were multiple origins of the Dn4 injurious populations.

LACK OF GENETIC VARIATION BETWEEN TWO RUSSIAN WHEAT APHID (HOMOPTERA: APHIDIDAE) BIOTYPES IN THE U.S.

Two Russian wheat aphid (*Diuraphis noxia*) biotypes occur in the US and have been named biotypes A and B (non-damaging and damaging to Dn4 resistant wheat, respectively). Fifteen Russian wheat aphid clones (10 biotype A and 5 biotype B) were collected from 5 different states and were studied for genetic variation. No variation was detected between or within the two biotypes based on random amplified polymorphic DNA (RAPD) and a 500 bp fragment of the cytochrome oxidase subunit I mtDNA gene. This lack of variation suggests the origin of biotype B is the extant population and argues against the introduction of the B biotype from another country. However, further comparisons of published sequences of Russian wheat aphids from

other countries and reviewing the literature cannot rule out the possibility of a second introduction form outside the US.

D. Genetics of *Lysiphlebus testaceipes* and Its Secondary Parasitoids

MOLECULAR MARKERS FOR IDENTIFICATION OF THE HYPERPARASITOID DENDROCERUS CARPENTERI AND ALLOXYSTA XANTHOPIS IN LYSIPHLEBUS TESTACEIPES PARASITIZING CEREAL APHIDS

Molecular markers have been developed to detect the presence of primary parasitoids in cereal aphids for use in estimating parasitism rates. However, the presence of secondary parasitoids (hyperparasitoids) may lead to underestimates of primary parasitism rates. Therefore, molecular markers to detect hyperparasitoids were developed. The 16S ribosomal RNA mitochondrial gene was amplified by polymerase chain reaction (PCR) and sequenced from two secondary parasitoid species, *Dendrocercus carpenteri* (Curtis) and *Alloxysta xanthopa* (Ashmead), four geographic isolates of the primary parasitoid, *Lysiphlebus testaceipes* (Cresson), and six cereal aphid species: bird cherry-oat aphid, *Rhopalosiphum padi* (L.); corn leaf aphid, *Rhopalosiphum maidis* (Fitch); English grain aphid, *Sitobion avenae* (F.); greenbug, *Schizaphis graminum* (Rondani); Russian wheat aphid, *Diuraphis noxia* (Kurdjumov); and yellow sugarcane aphid, *Sipha flava* (Forbes). Species-specific PCR primers were designed for each insect on the basis of these 16S rRNA gene sequences. Amplification of template DNA, followed by agarose gel electrophoresis, successfully distinguished *D. carpenteri* and *A. xanthopa* from all four isolates of *L. testaceipes* and all six cereal aphid species.

ESTIMATION OF HYMENOPTERAN PARASITISM IN CEREAL APHIDS BY USING MOLECULAR MARKERS

Polymerase chain reaction (PCR) primers were designed and tested for detection and identification of immature parasitoids in small grain cereal aphids. The PCR technique was evaluated for (1) greenhouse reared greenbugs, *Schizaphis graminum* (Rondani) parasitized by *Lysiphlebus testaceipes* (Cresson) and (2) aphids collected from winter wheat fields in Caddo county Oklahoma. For greenhouse samples, parasitism frequencies for greenbugs examined by PCR at 0, 24 and 48 h after removal of *L. testaceipes* parasitoids, were compared to parasitism frequencies as determined by greenbug dissection. PCR was unable to detect *L. testaceipes* in greenbugs at 0 and 24 h post-oviposition, but was able to detect parasitoids 48h post-oviposition at frequencies that were not significantly different from dissected samples. Field collected samples were analyzed by rearing aphids from each sample, and comparing frequencies of mummies developed with samples examined by PCR. Aphid samples included corn leaf aphids, *Rhopalosiphum maidis* (Fitch), bird cherry-oat aphids, *R. padi* (L.), English grain aphids, *Sitobion avenae* (F.), and greenbugs.

Mummies were isolated until adult emergence, where upon each parasitoid was identified to species (*L. testaceipes* was the only parasitoid species found). Parasitism detection frequencies for PCR were not statistically different from parasitism frequencies of reared aphids. These results indicate that PCR is a useful tool for providing accurate estimates of parasitism and for identification of immature parasitoids to species.

E. Frequency and Distribution of *Schizaphis graminum* mtDNA Haplotypes

F. Russian Wheat Aphid Resistant Barley

The first RWA-resistant barley cultivar, 'Burton', was released jointly by USDA-ARS, Aberdeen, ID, USDA-ARS, Stillwater, OK, Colorado State University, University of Nebraska, and New Mexico State University. Burton is a 2 row spring feed barley deriving its RWA resistance from STARS 9301B.

Three, RWA-resistant, 2-row, spring feed barleys, Stoneham, Sidney, and Xeris, were increased for cultivar release by both Colorado and Nebraska Foundation Seeds. These cultivars were bred for the extremely droughty high plains of eastern Colorado and western Nebraska. Stoneham has resistance from STARS 9577B while Sidney and Xeris have resistance derived from STARS 9301B. Although STARS 9301B and STARS 9577B were developed for resistance to RWA1, they have been shown to be resistant against RWA1, RWA2, RWA3, RWA4, and RWA5 as well. Seed should be available to growers this spring.

RWA-resistant, 6 row, winter feed barley germplasm lines STARS 0501B, STARS 0502B, STARS 0503B, STARS 0504B, STARS 0505B, STARS 0506B, and STARS 0507B, were released to breeders in the summer of 2005. These lines are in a Schuyler background with resistance from seven unadapted germplasm lines developed by USDA-ARS in Stillwater, each with resistance from different barley accessions. These lines were selected in Aberdeen, Idaho and are adapted to northern US growing conditions.

Increases have been made prior to release of 19, RWA-resistant, 2 row, spring, malting barley germplasm lines as well as 17, RWA-resistant, 6 row, spring malting barley germplasm lines and 7, RWA-resistant, 2 row, spring feed barley germplasm lines.

A breeding program has been initiated to develop winter, hullless, feed barleys resistant to both RWA and Greenbug, and adapted to Oklahoma.

Progress has been made toward a seedling screening test for BCOA resistance in barley.

G. Russian Wheat Aphid Resistant Wheat

Russian wheat aphid ‘biotypes’ have continued to be identified based on differential host plant responses. This has led to an increasing level of complexity for the wheat germplasm development program. In 2004, it was reported here that resistance to RWA-2 was found in some of the RWA-1 resistant breeding lines in the Stillwater ARS wheat genetics/breeding program, as well as in many RWA-1 resistant Plant Introductions (seed supplied by GRIN). As additional RWA ‘biotypes’ were identified, it was of interest to determine if the breeding lines that were resistant to RWA-1 and RWA-2 were also resistant to any newly identified ‘biotypes’. Therefore, during the past year, large-scale screening tests have been conducted with RWA-1, RWA-2 and RWA-3.

Screening tests with the new ‘biotypes’ were conducted in three separate greenhouses, using large, walk-in cages for RWA-2 and RWA-3 (the cages were intended to limit aphid dispersal as well as cross contamination). Initial tests included multiple lines derived from 16 different Plant Introductions (seed provided by GRIN), which had previously been screened to RWA-2, rescued and grown for seed increase; 60 Iranian wheat landraces (original source was Cal Qualset) resistant to RWA-2; and all of our breeding lines that had also proved to be resistant to RWA-1 and RWA-2.

From all of the material screened, we were able to identify Plant Introductions, Iranian landraces and advanced breeding lines that are resistant to all three RWA biotypes. In most cases, resistance was either segregating or heterogeneous in reaction; in a few lines, all plants that were screened were resistant. Resistance ranged from high to moderate levels. While a high level of resistance is certainly more impressive when looking at resistant plants, it should be noted that a moderate level may provide adequate protection under field conditions, especially when the plants are able to maintain flat leaves with infestation. The breeding lines derived from the different sources are at varying stages of purity and advancement within the program. Germplasm releases are planned as soon as purity of the seed is determined. Small samples will also be available for distribution.

It is important to note that there was some discrepancy in segregation when screening against the three different RWA ‘biotypes’. For example, breeding lines that were developed by screening with RWA-1 and are homozygous resistant to that ‘biotype’, may be segregating for resistance to another ‘biotype’. In addition, with sister lines derived from the same source of resistance, where both lines were resistant to RWA-1, only one may be resistant to another ‘biotype’. Unfortunately, this means that no generalizations can be made about whether or not a breeding line is resistant to a new ‘biotype’ based on its source of resistance, and screening tests must be done with each line. These results may indicate that more than one gene is playing a role in conferring resistance, but it was impossible to determine that with RWA-1 alone. Previously published information concerning RWA resistance gene designations may have to be rethought.

III. PERSONNEL CHANGES

A. S. Dean Kindler, Research Entomologist, will be retired in December after 40 years of service. He earned his Ph.D. in Entomology during 1967 at the University of Nebraska. Dean has had a long and productive career in the ARS. He began his service in 1964 with USDA-ARS in Lincoln, Nebraska. In 1987, Dean transferred to the Stillwater Unit. He contributed greatly to Stillwater's research effort in cereal aphid management, as well as the overall mission of the ARS. Some of his notable contributions were in forage entomology, greenbug biotypes, Russian wheat aphid host ecology, cereal aphid management, and the rice root aphid biology and economic impact. We wish him a long and happy life after retirement. Thank you, Dean. It has been an honor and pleasure to have worked with you.

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