

October 28-31, 2012

Hotel Valley Ho Scottsdale, Arizona

Welcome to Scottsdale!

Dear Colleagues,

On behalf of the organizing committee, I want to take this opportunity to welcome you to the Third International Lygus Symposium. In February of 2005, the first such conference was held in Ottawa, Canada. It was organized to better coordinate the effort of solving common research problems in the management of Lygus bugs. The original objectives were to: 1) Enhance national and international collaboration; 2) Exchange current research results relevant for the management of Lygus plant bugs; 3) Find solutions to practical problems and highlight information gaps to establish research priorities; 4) Determine an integrated management approach for Lygus control in different crop commodities. There is little difference between that first symposium and this; our objectives have stayed the same and the research that we'll share is just as relevant.

The current incarnation of the Lygus Symposium has once more gathered researchers from several countries, representing universities, public agencies, and private industry. Together we shall discuss the full range of mirid biology, behavior and ecology, as well as the most recent advances for controlling their populations. Most importantly, we again have the opportunity to readily share advice and insights with one another, enhancing our collective research efforts.

The venue for this meeting, the Hotel Valley Ho, is a landmark resort designed by architect Edward Varney in the mid-century modern style. Since opening in 1956, this chic vacation spot has hosted many trendsetters and movies stars. Our group should fit in quite well. The Valley Ho offers numerous amenities and is centrally located in Scottsdale, one of the premier resort towns in the United States. This setting was chosen to provide a relaxed atmosphere, fostering the kinds of exchanges that build a sense of community and common purpose.

We hope that you have a stimulating and rewarding experience during this symposium. We also encourage you to take full advantage of the many attractions that are readily available to you.

With sincere regards,

Color Drut

Colin S. Brent USDA-ARS Arid Land Agricultural Research Center Maricopa, Arizona October, 2012

Acknowledgements

Organizing Committee:

Colin Brent (Chair), Peter Ellsworth, Alfred Fournier, Ryan Kurtz, Ayman Mostafa, Steve Naranjo, Patricia O'Leary, Nancy Pierce

Scientific Committee:

Jeff Gore (Chair), Colin Brent, Jeff Fabrick, Joe Hull

Keynote Speakers:

Peter Goodell - University of California, Cooperative Extension, Statewide IPM Program Yves Carrière - Department of Entomology, University of Arizona

Web Design:

Colin Brent, Alfred Fournier, Wayne Dixon

Sponsors:

Cotton Incorporated Dow AgroSciences – Gala Dinner FMC – Breakfasts, Breaks & Lunches Monsanto – Opening Reception University of Arizona, Arizona Pest Management Center USDA-ARS, Arid-Land Agricultural Research Center

Moderators:

Colin Brent, John Byers, Peter Ellsworth, Al Fournier, Peter Goodell, Jeff Gore, James Hagler, Joe Hull, Jeff Fabrick, Steve Naranjo, Charlie Pickett

Special Thanks:

To Dan Langhorst for assistance with preparing conference materials, Colin Brent for designing the logo, and Al Fournier for serving as the communications coordinator.

Schedule of Events

Locations: Breakfast and lunch will be served in the small conference room (#1). All oral and poster presentations will occur in the large conference room (#2). Breaks will be held in the courtyard between these rooms.

Sunday, October 28

Time	Event
6:00-7:30p	On-Site Registration Desk Open - Palm Court lawn
7:00-9:00p	Opening Reception - Palm Court lawn, Sponsored by Monsanto

Monday, October 29

Time	No.	Event / Presentation Title
8:00a		Continental Breakfast Buffet
8:55a		Welcome and Introduction
		Session 1 Moderators: Colin Brent and Jeff Gore
9:00a	1	It takes a village to manage Lygus Peter Goodell , Gallegos, Gibbs
9:40a	2	Management strategies of cotton mirid bugs in different planting patterns in northern China Yanhui Lu , Wang, Wu
10:00a	3	Cotton plant response to selected Lygus infestation levels in the Texas High Plains Megha Parajulee, Shrestha, Carroll
10:20a		Break
10:40a	4	Managing lygus bugs in strawberries and the potential of microbial control Surendra Dara
11:00a	5	Seasonal dynamics of <i>Lygus hesperus</i> and its predators as mediated by differential irrigation and insecticide treatment Peter Asiimwe , Naranjo, Ellsworth
11:20a	6	Establishment, spread, and impact of <i>Peristentus relictus</i> in California, a Lygus nymphal parasitoid from southern Europe Charles Pickett , Swezey, Nieto, Bryer, Stadtherr, Erlandson

11:40a	7	Movement and parasitism of Lygus spp. by the imported parasitoid <i>Peristenus relictus</i> in trap-cropped strawberries Diego Nieto , Swezey, Bryer, Pickett, Hagler, Machtley
12:00p		Lunch
		Session 2 Moderators: Charlie Pickett and James Hagler
1:30p	8	Effect of <i>Lygus lineolaris</i> on navy beans in Manitoba Tharshinidevy Nagalingam , Holliday
1:50p	9	The Impact of planting date and varietal maturity selection on tarnished plant bug management in Midsouth Cotton Brian Adams , Gore, Catchot, Dodds, Musser, Cook
2:10p	10	Protein mark-capture monitoring of lygus bug (<i>Lygus hesperus</i> Knight) movement between an organic strawberry field and alfalfa trap crops in California Sean Swezey , Nieto, Bryer, Hagler, Machtley, Pickett
2:30p	11	Evolving pest complexes and IPM strategies for the mid-South Scott Stewart , Roberts, Catchot, Gore, Lorenz
2:50p		Break
3:10p	12	Seasonal occurrence of mirid bugs in edible beans and soybeans in Manitoba Tharshinidevy Nagalingam , Holliday
3:30p	13	Impact of Lygus spp. on damage, yield and quality of lesquerella (<i>Physaria fendleri</i>), a potential new oil-seed crop Steven Naranjo , Ellsworth
3:50p	14	Using social media in IPM extension programs Jeff Gore , Catchot, Dodds, Irby, Larson, Beuhring, Allen, Bond, Eubank, Walker, Krutz, Jackson, Musser, Golden, Cook, Henn, Oldham, Riley
4:30p		Poster Session (Papers 15-34) and Casual Reception
		Tuesday, October 30

Time	No.	Event / Presentation Title
8:00a		Continental Breakfast Buffet
		Session 3 Moderators: John Byers and Peter Goodell
9:00a	35	A novel messaging system for passive mate guarding in <i>Lygus hesperus</i> Colin Brent , Byers
9:20a	36	Thermal ecology of <i>Lygus hesperus</i> development Dale Spurgeon , Cooper

9:40a	37	Production and predator-induced release of defensive chemicals by the plant bug <i>Lygus hesperus</i> John Byers
10:00a	38	Monitoring and control of pest Mirid species in the UK using sex pheromones Michelle Fountain , Cross, Hall, Farman
10:20a		Break
10:40a	39	Tarnished plant bug, <i>Lygus lineolaris</i> , a potential biotype difference in the Mississippi delta Brian Adams, Gore, Catchot, Musser, Dodds
11:00a	40	Transcriptomics and genomics of the tarnished plant bug, <i>Lygus</i> <i>lineolaris</i> OP Perera, Snodgrass, Jackson, O'Leary
11:20a	41	Pyrosequencing of the adult tarnished plant bug, <i>Lygus lineolaris</i> , characterization of messages important in metabolism and development, and RNAi insect control Michael Roe , Magalhaes, Van Kretschmar, Donohue
11:40a	42	Polyphyly of mtDNA vs. species identification in western pest Lygus: evidence of cryptic species and interspecies hybridization or shortcomings of morpho-species taxonomy Richard Roehrdanz , Burange, Foottit, Maw, Boetel, Sears
12:00p		Lunch
		Session 4 Moderators: Jeff Fabrick and Joe Hull
1:30p	43	Identification of <i>Lygus hesperus</i> by DNA barcoding reveals insignificant levels of genetic structure among distant and habitat diverse populations Zhou, Kandemir, Walsh, Zalom, Laura Lavine
1:50p	44	Polygalacturonase gene expression in field collected <i>Lygus lineolaris</i> Daniel Fleming, Krishnan, Musser
2:10p	45	When a receptor isn't what it looks like: The <i>Lygus hesperus</i> "sex peptide receptor" Joe Hull, Brent
2:30p	46	Characterization of aquaporins from the western tarnished plant bug, <i>Lygus hesperus</i> and the whitefly, <i>Bemisia tabaci</i> Jeff Fabrick , Yool
2:50p		Break
3:10p	47	Insecticide resistance status and mechanisms in field populations of the tarnished plant bug <i>Lygus lineolaris</i> Yu Cheng Zhu , Luttrell

- 3:30p **48** Development of Sulfoxaflor for control of Lygus spp. and related mirids **James Thomas,** Siebert, Richardson, Castro, Huang, Annetts, Lee
- 3:50p **49** Response of *Lygus lineolaris* to sulfoxaflor and neonicotinoids in laboratory assays **Don Cook**, Snodgrass, Gore
- 4:10p **50** Discovery and optimization of hemipteran-active proteins for transgenic plant applications **Robert Brown,** Anilkumar, Baum, Bowen, Clark, Pleau, Shi, Sukuru, Wollacott
- 4:30p **51** In-planta efficacy evaluation of Lygus-active Bt proteins **Waseem Akbar,** Penn, Anilkumar, Hagerty, Brown, Chen, Clark

Wednesday, October 31

Time 8:00a	No.	Event / Presentation Title Continental Breakfast Buffet
		Session 5 Moderators: Steve Naranjo and Al Fournier
9:00a	52	Predicting effects of local and regional factors on population dynamics of <i>Lygus hesperus</i> Yves Carrière , Goodell, Ellers-Kirk, Larocque, Dutilleul, Naranjo, Ellsworth
9:40a	53	Using spatial metrics to assess crop stressed by pests: Case of <i>Diuraphis noxia</i> in wheat fields Georges Backoulou , Elliott, Giles, Willers
10:00a	54	Application of FRAGSTATS metrics to categorical imagery products used for tarnished plant bug sampling and management Jeffrey Willers , Backoulou
10:20a		Break
10:40a	55	Successful Lygus management as a stabilizing element of cotton IPM in Arizona Peter Ellsworth , Naranjo, Fournier, Brown, Dixon
11:00a	56	Western tarnished plant bug, <i>Lygus hesperus</i> Knight, management in San Joaquin Valley cotton: trends and Implications Larry Godfrey , Pierce
11:20a	57	Field scale movement of Lygus bugs in Arizona cotton Ayman Mostafa , Ellsworth, Hagler, Naranjo
11:40a	58	Development of Lygus management strategies for Texas cotton Ram Shrestha, Carroll, Parajulee
12:00p		Lunch

		Session 6 Moderators: Colin Brent and Peter Ellsworth
1:30p	59	The Lygus simulation training environment: An interactive Workshop & Discussion Peter Ellsworth, Goodell, Fournier, Brown, Kerns, Parajulee
3:30p		Break
3:45p		Panel Discussion and Wrap-up
7:00p		Gala Dinner – Tower Rooftop, Sponsored by Dow AgroSciences

Thursday, November 1 (Optional Tour)

*Early departures for return to the airport must be recorded on sign-up sheet at registration desk or by emailing your intent to participate and special needs for transportation to Dr. Al Fournier (Fournier@cals.arizona.edu) (count on 40 minutes drive from MAC to the Sky Harbor Airport in PHX).

Time	Event
7:55a	Board tour bus (bring your hat and sunglasses!) at Hotel Valley Ho
8:00a	Bus departs for Salt River Pima-Maricopa Indian Community
8:25a	Arrive Rousseau Farms organic vegetable production fields (<u>www.farmfreshaz.com</u>), Lin Evans, Independent Pest Control Advisor
9:40a	Depart for University of Arizona Maricopa Agricultural Center (MAC)
10:35a	Arrive MAC/ Break
10:50a	 Load plot trailer, Tour Lygus Studies Drs. Ayman Mostafa (UA), Peter Ellsworth (UA), Steven Naranjo (USDA-ALARC) Lygus control & non-target effects in cotton Local movement of Lygus within a cotton field Evaluation of transgenic cotton lines for Lygus control
12:00p*	Authentic Northern Mexican Carne Asada Lunch, al fresco (MAC Park) Grilled carne/chicken/vegetables (zucchini, squash, peppers, eggplant), fresh salsa, hot salsa, refried beans, homemade flour tortillas, corn tortillas, guacamole, salad, iced tea
1:00–2:30p	Optional bus tour of the UA-MAC farm (Dr. Bob Roth, Resident Director)
	Optional tour of USDA-ARS, Arid-Land Agricultural Research Center (ALARC) (Dr. Steven Naranjo, Center Director)
2:45p	Depart for Hotel Valley Ho
3:45p	Arrive Hotel Valley Ho

Third International Lygus Symposium Abstracts of Papers and Posters (indexed to schedule for easy reference)

It Takes a Village to Manage Lygus

Peter B. Goodell¹, Luis Gallegos², Marcia Gibbs²

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Lygus hesperus is polyphagous, mobile pest of crops in the San Joaquin Valley of California. Populations build on cultivated crops and uncultivated areas and move as hosts become unsuitable. Recent work supports the observations that some crops are better sources of *Lygus* than others in supplying *Lygus* to neighboring cotton. The landscape interactions between *Lygus* sources and sinks is extremely complex depending on the strength of the source (how many *Lygus* are produced), the timing of the population release (susceptibility of cotton), distance between source and cotton field, and crop assemblage in which the cotton field is located.

Developing regional plans to maximize our understanding of *Lygus* in the landscape requires community cooperation. Two different cases will be presented. In one case, very large landowners worked together to coordinate planting schemes and the timing of *Lygus* population controls in safflower. This landscape consists of large production units, less diversified cropping patterns, but strongly motivated management and good regional communication to support coordination across multiple landowners. Annual consultations with key community stakeholders provided a forum to discuss opportunities and challenges in local *Lygus* management and assess past outcomes in order for the community to develop and implement a sophisticated area management program for *Lygus*.

In another case, many smaller landowners have to be supported in approaching regional planning through community based programs such as web based GIS, supplemental scouting, demonstration projects and participatory field research and meetings. In this community, land ownership consisted of smaller properties, greater landscape diversity (riparian mixed with agricultural and rural towns), more independent decision-making within the landscape, reluctance in considering issues crossing farm borders but more local experimentation and flexibility in approach. Local meetings and demonstration projects have been conducted to increase the comfort level of this community to embrace landscape approaches to *Lygus* management.

Management strategies of cotton mirid bugs in different planting patterns in northern China

Lu Yanhui, Wang Guirong, Wu Kongming Institute of Plant protection, Chinese Academy of Agricultural Sciences

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Abstract not available.

Cotton Plant Response to Selected *Lygus* Infestation Levels in the Texas High Plains

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A thorough understanding of insect pest populations and crop plant interactions is critical for successful pest management. Our research program has conducted experiments characterizing cotton plant and Lygus hesperus interactions in various production systems. Research goals were to determine how much *Lygus* injury cotton plants can tolerate without significantly compromising yield and fiber quality, plus characterizing the impacts of varied Lygus infestations on plant growth and development, particularly on the fruiting pattern. Additional objectives targeted the development of a dynamic economic threshold and pesticide application rules for Texas High Plains Lygus management. Among various production inputs, irrigation rate and cotton cultivar are two important inputs that can significantly affect cotton growth and development. The effects of differing Lygus infestations on cotton growth and development were determined under selected irrigation levels (dryland, low, medium, high) and with differing cotton cultivars (long-/short-season) and at different crop phenological stages (pre-bloom, early bloom). Various Lygus infestation levels were evaluated by manually augmenting known numbers of Lygus onto cotton plants (0, 1-2, 3-6, 6-8 nymphs/plant in control, low, medium and high density treatments, respectively). Cotton responses to varied *Lygus* infestations were significantly different among the irrigation levels, and between the pre-bloom and early bloom stages. The high irrigation and pre-blooming cotton treatments displayed higher crop compensation capacities to Lygus injury compared to dryland and early bloom cotton. This suggests Lygus management recommendations should differ between dryland and irrigated fields and also between pre-bloom and early bloom stage cottons. In general, a 25-30% early season square loss can be compensated by irrigated cotton. This is accomplished via later season production of new lateral position squares; therefore, it has been recommended that pesticide applications for Lygus control are most likely not justified if the square retention is 70-75% at pre-blooming stage for irrigated Texas High Plains cotton.

Managing Lygus Bugs in Strawberries and the Potential of Microbial Control

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Strawberries are an important cash crop in California. Lygus bug (Lygus spp.) is a major pest of strawberries causing significant losses to the yield and quality. Strawberry industry primarily depends on chemical insecticides for managing this pest. Fall and summer planted strawberries along with second year crop increase the availability of the host plants for lygus bugs and the need for regular treatments. This will in turn increases the risk of pesticide resistance and harm beneficial arthropod populations. Minimizing excessive use of effective chemicals both to extend their life and improve environmental safety is important. It is also essential to identify and implement environmentally safe management options to improve current IPM practices. Microbial control with entomopathogenic fungi such as Beauveria bassiana is underexplored for managing strawberry pests in the California Central Coast. Various studies were conducted to evaluate the potential of using *B. bassiana* for managing strawberry pests with a particular emphasis on lygus bugs. Laboratory bioassays indicated that B. bassiana in combination with certain chemicals at reduced rates was effective against adult Lygus hesperus. Endophytic colonization of *B. bassiana* in some crops has been reported to adversely affect herbivores. In a greenhouse study, B. bassiana colonized strawberry plants and persisted for up to 9 weeks in various plant parts. Impact of colonized B. bassiana on lyous bugs and other pest is yet to be determined. Recent large plot field studies with *B. bassiana*, azadirachtin, chemical pesticides, and their combinations showed a promise for microbial control in strawberries especially for lygus bugs.

Seasonal Dynamics of *Lygus hesperus* (Hemiptera: Miridae) and its Predators as Mediated by Differential Irrigation and Insecticide Treatment

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Irrigation effects on plant characteristics have the potential to affect population dynamics of pests and their natural enemies. We evaluated the impact of varying irrigation regimes on densities of Lygus hesperus Knight and the associated arthropod predator community in cotton. We also evaluated how the responses to plant quality due to differential irrigation might be mediated by two important insecticides used to control L. hesperus. Irrigations were done at 20%, 40% and 60% soil water depletion (SWD) while L. hesperus and predators were manipulated using the broad spectrum insecticide, acephate, the selective insecticide, flonicamid, and an untreated check. Densities of *L. hesperus* were significantly influenced by irrigation regime and were significantly lower in the deficit-irrigated plants compared with the well-irrigated plants. Densities of the generalist predators that are known to prey on *L. hesperus* did not vary with irrigation regime but were significantly lower in the acephate sprayed plots. Predator:prey ratios reflected patterns of predator and prey densities with the lowest ratios in the acephate sprayed plots. Predator function, as inferred from a predation index, was not significantly affected by irrigation regime but was significantly lower in the acephate plots. The efficacy of the two Lygus control insecticides was not influenced by irrigation regime. These results indicate that irrigation regime is an important factor in seasonal dynamics of L. hesperus in cotton, but predator density and function as a factor of insecticide selectivity is more important in L. hesperus dynamics and management.

Establishment, Spread, and Impact of *Peristentus relictus* in California, a Lygus Nymphal Parasitoid from Southern Europe

Charles H. Pickett¹, Sean Swezey², Diego Nieto², Janet Bryer², Marypat Stadtherr¹, Martin Erlandson³

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Two nymphal parasitoids were imported into California from southern Europe beginning in 1998. For a period of ca. 7 years both Peristenus relictus and P. digoneutis sustained populations in the original Sacramento release site of managed alfalfa. We have not recovered the latter since 2007, however populations of *P. relictus* have persisted and spread south up to 250 km, and north 35 km. Both populations were released at several locations in the Monterey Bay region of California beginning in 2002, a major strawberry growing region where Lygus spp. are a key pest of this crop. Only P. relictus was recovered from this region, and it has spread at least 150 km south, and 93 km north. Parasitoids were released into wild vegetation supporting Lygus populations near or bordering conventionally managed strawberries. They were also released into organically produced strawberries intercropped with alfalfa to trap and control this pest. Populations of *P. relictus* have thrived in the wild vegetation, and in the strips of alfalfa, despite routine vacuuming and annual replacement of these strips. They have survived insecticide applications in the same kind of strips grown in conventionally produced strawberries, and in pesticide treated strawberry fields adjacent to wild vegetation colonized by *P. relictus*. Parasitism in the two wild vegetation release sites has steadily increased since 2005, seasonal averages reaching 71% and 59%. Lygus numbers at these sites have declined over the same period from a seasonal average high of 0.8 and 2.5 nymphs per sweep to less than 0.1 and 0.2 nymphs per sweep, respectively. The density of nymphs in the alfalfa and strawberries at the organic farm have declined over 10 fold during the same period of time, but their numbers have remained constant at the conventional site bordering a wild vegetation release site.

Movement and Parasitism of *Lygus* spp. by the Imported Parasitoid *Peristenus relictus* in Trap-Cropped Strawberries

Diego Nieto¹, Sean Swezey¹, Janet Bryer¹, Charles Pickett², James Hagler³, Scott Machtley³

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The European parasitoid *Peristenus relictus* has been established on the California Central Coast for the biological control of *Lygus* spp. (or lygus bugs) in strawberries. This establishment was facilitated by the high host densities provided by alfalfa trap crops, which are used for cultural management of lygus bugs in organic strawberries. The retention of *Lygus* spp. in alfalfa creates aggregated distributions that provide improved opportunities for biological control. The abundance and distribution of P. relictus between two trap crops separated by 50 strawberry rows was analyzed in 2008 and 2010. Parasitism of Lygus spp. nymphs by P. relictus (measured by larval abundance and % parasitism) was greatest in alfalfa trap crops when compared with strawberry rows. The spatial distribution of nymphs and P. relictus larvae in and between trap crops was significantly correlated. The movement of *P. relictus* adults from a marked alfalfa trap crop into adjacent strawberry rows or trap crops was also studied in 2008 and 2009 using a chicken egg albumin enzyme-linked immunosorbent assay (ELISA) mark-capture technique. In 2008 and 2009, 85% and 49% of proteinmarked wasps were captured from central trap crops, indicating that alfalfa trap crops act as host-density-anchors in organic strawberries that allow for spatial density dependence between parasitoid and host. Other parasitoids, however, dispersed from trap crops out into strawberry rows, indicating that alfalfa can also serve as a source of parasitoids for adjacent strawberries. Parasitoid activity persists despite active trap crop management in both organic and conventional systems using tractor-mounted vacuums and insecticides.

Effect of Lygus lineolaris on Navy Beans in Manitoba

Tharshinidevy Nagalingam, Neil J. Holliday

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Effects of lygus bug (*Lygus lineolaris*) infestations on seed quality and quantity of navy beans were studied from 2009 to 2011 in field cages in Manitoba, Canada. Two experiments were conducted at each of with three different growth stages of navy beans. Each experiment had a randomized complete block design, in which different known numbers of lygus adults or nymphs were introduced into 1 m² cages containing two crop rows of navy beans at a specific growth stage. Insects were left in the cages until harvest, when the seed was harvested, graded and weighed.

When insects were introduced at the mid-flowering to early pod stage (R2–R3) in an experiment in 2009, the yield of undamaged seed was reduced by 0.5 g/ adult lygus bug. In contrast, when the experiment was repeated in 2011, there was an increase in weight of undamaged seed with increasing lygus bug densities. In the two experiments where lygus bugs were introduced at the mid-pod set to early seed fill stage (R4–R5), there was a reduction in weight of undamaged seed weight but there was no evidence of linear response to number of lygus bugs. In cages infested with \geq 30 lygus at R4–R5, weight of undamaged seed was reduced by 42 g/ m² when adults were introduced, and by 85 g/m² when nymphs were introduced. In the two experiments where lygus bugs were introduced at the time or seed maturation (R6–R7), there was no evidence of reduction in yield quantity. Regardless of the stage of insect introduction, no significant reduction in quality of seed yield was detected. These results will be discussed in light of laboratory studies of plant growth stage-specific responses of plants to infestation by lygus adults and nymphs.

The Impact of Planting Date and Varietal Maturity Selection on Tarnished Plant Bug Management in Midsouth Cotton

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A field experiment was conducted at the Delta Research and Extension Center in Stoneville, MS during 2010 and 2011 to investigate the impact of varietal maturity and planting date on tarnished plant bug, Lygus lineolaris (Palisot de Beauvois), in cotton. Four planting dates were selected to encompass the standard cotton planting window for the Delta region of Mississippi. An early maturing variety and a late maturing variety were planted at each planting date. Each plot was split into sprayed for tarnished plant bug and unsprayed. Plots were sampled weekly for tarnished plant bug densities from first square until physiological maturity. Plots were harvested at the end of the season and lint yields were determined. Planting dates one and two had significantly lower densities of tarnished plant bug than the latter two planting dates. Fewer foliar insecticide applications were needed at the earlier planting dates. Planting date one had the highest yields and each successive planting date decreased in yield significantly. Earlier planting dates also sustained less yield loss from tarnished plant bug than later planting dates. The early maturing variety yielded significantly more than the late maturing variety. Tarnished plant bug also had less impact on yield of the early variety as a result of tarnished plant bug damage than on the late variety. A significant spray by variety interaction occurred as a result of the larger difference in yield between sprayed and unsprayed plots of the late variety compared to the early variety. These data demonstrate that later plantings of cotton in the Mississippi Delta are more impacted by tarnished plant bug than early plantings. Late season tarnished plant bugs have a greater impact on yield losses in cotton than earlier in the season. As a result, growers should manage cotton for earliness through planting date and varietal selection.

Protein Mark-Capture Monitoring of Lygus Bug (*Lygus hesperus* Knight) Movement Between an Organic Strawberry Field and Alfalfa Trap Crops in California

Sean L. Swezey¹, Diego J. Nieto¹, Janet A. Bryer¹, James A. Hagler², Scott A. Machtley², Charles H. Pickett³

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Alfalfa (*Medicago sativa* L.) is a highly attractive plant host to *Lygus* spp. in the western United States and is used as a trap crop to reduce damage to organic strawberries caused by lygus bug on the California Central Coast. To determine the influence of alfalfa trap crops on the movement patterns of adult lygus bugs into adjacent strawberries and other nearby trap crops, a protein mark-capture technique using a chicken-egg albumin enzyme-linked immunosorbent assay (ELISA) was used in a replicated, two-week field experiment. Results of this experiment showed that a large majority of marked-captured L. hesperus adults were found in alfalfa trap crops compared with adjacent strawberry rows at all post-marking time intervals. Low percentages of total marked-captured adults dispersed from trap crops into adjacent strawberry rows. A small percentage of marked adults were also captured in neighboring alfalfa trap crops located 62 m from the point of protein application, highlighting the dispersal capacity of some adults in a trap-cropped field system. However, the average distance of adult movement into adjacent strawberry rows was less than 0.5 m in this study. The strong attenuation of adult lygus bug movement by alfalfa in organic strawberries is a key operational feature of a targeted trap-cropping approach. Physical and cultural suppression measures can be more efficiently and economically directed at lygus bug adults in alfalfa trap crops than at more extensive adjacent strawberry acreage.

Evolving Pest Complexes and IPM Strategies for Cotton in the Midsouth and Southeast

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Significant changes during the last 10-15 years have changed the face of IPM in cotton. New insecticide chemistries, boll weevil eradication and the wide scale adoption of Bt transgenic cotton have dramatically changed the key insect pest complexes that occur across the US Cotton Belt. While some pests have been eradicated or relegated to a relatively minor status, others have emerged as major IPM issues. This paper will address new pest complexes and IPM strategies have evolved in a cotton production system dominated by Bt cotton, with special emphasis on the midsouthern and southeastern U.S. and the increasing pest status of plant bugs, including *Lygus lineolaris*, and stink bugs. Insecticide selection, cultural controls, action thresholds and other IPM tools for the management of tarnished plant bug will be discussed.

Seasonal Occurrence of Mirid Bugs in Edible Beans and Soybeans in Manitoba

Tharshinidevy Nagalingam, Neil J. Holliday

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Production of dry edible beans and of soybeans has greatly increased in Manitoba in the last decade, and, despite concerns about plant bugs, there was no information on the species composition and seasonal occurrence of mirid bugs in these crops. In this study we sampled commercial pinto bean, navy bean and soybean fields from 2008 to 2010 to investigate species composition and seasonal patterns of abundance of *Lygus* bugs and their relatives. Sampling was primarily by sweep netting and was conducted at weekly intervals throughout the growing season.

Four mirid species were found in all three of the crops, and their relative abundance was similar in the different crops. Based upon catches of adults, species composition was: *Lygus lineolaris* (91%), *L. borealis* (3%), *L. elisus* (1%) and *Adelphocoris lineolatus* (4%). *Lygus* adults were first found in the fields when the plants are in the flowering and pin pod stage which occurred during late July to early August. Nymphs of *Lygus* were found in all three crops from July to September (flowering to maturity), suggesting that these crops were suitable hosts for development, and that a single generation of *Lygus* bug was completed in them. A pronounced peak of *Lygus* adults numbers occurred in late August to early September, at the time of the mature seed stage of the crops. The late season peak of adults coincided with harvesting of the many canola fields in the area, suggesting that there is migration of *Lygus* bugs from harvested canola to beans. *Lygus* numbers were low in edible beans and soybeans compared to the other host crops in Manitoba: the highest number recorded was 78 per 100 sweeps in September 2010. In none of three years of the study was there evidence that mirids reduced quality or quantity of yield in the crops.

Impact of Lygus spp. on Damage, Yield and Quality of Lesquerella (Physaria fendleri), a Potential New Oil-Seed Crop

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Lesquerella, Physaria fendleri is native to the western U.S. and is currently being developed as a commercial source of valuable hydroxy fatty acids that can be used in a number of industrial applications. The plant is cultivated as a winter-spring annual and in the desert southwest it harbors large populations of arthropods, several of which could be significant pests once production expands. Lygus spp. are common in lesquerella and are known pests of a number of agronomic and horticultural crops. A four-year study was undertaken to evaluate the probable impact of Lygus spp. on production of this new crop. Plant damage and subsequent seed yield and quality were examined relative to variable and representative densities of *Lygus* spp. resulting from variable frequency and timing of insecticide applications. Increasing damage to fruiting structures was significantly associated with increasing pest abundance, particularly the abundance of nymphs, in all years. This damage, however, did not consistently translate into reductions in seed yield, individual seed weight or seed oil content, and pest abundance generally explained relatively little of the variation in crop yield and quality. Negative effects on yield were not sensitive to the timing of pest damage (early vs. late season), but were more pronounced during years when potential yields were lower due to weed competition and other agronomic factors. Results suggest that if the crop is established and managed in a more optimal fashion, Lygus spp. may not significantly limit yield. Nonetheless, additional work will be needed once more uniform cultivars become available and yield effects can be more precisely measured. Densities of Lygus spp. in unsprayed lesquerella are on par with those in other known agroecosystem level sources of this pest (e.g., forage and seed alfalfa). Thus, lesquerella production may introduce new challenges to pest management in crops such as cotton.

Using Social Media in IPM Research and Extension Programs

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Agricultural research and extension programs at Land Grant Universities have faced numerous challenges over the years. The means for transferring important information to growers. consultants, and the agricultural industry is a constantly evolving process. Historically, each county within a state had an agricultural extension agent that met with individual growers face to face on a regular basis and personal relationships were developed. Those ties have been severed over the years through reductions in funding and personnel. In light of these changes, many extension programs transitioned to a paper based newsletter that was distributed through the postal service and later through email. This proved to be an effective means for extension personnel to transfer important information about pest outbreaks, new registrations, and etc. to their clientele. However, these newsletters were usually only delivered on a weekly basis. In terms of hard copy newsletters that were distributed through the mail, there was often a 10-14 day delay in getting information to the agricultural industry. When the newsletters transition to an email distribution system, the delay was reduced, but the potential still existed for up to a week delay. More recently, the computer revolution has begun to have significant impacts on extension programs at many universities. Social media sites such as Facebook, Twitter, and Linkedin are connecting people in ways that have never been done before. In addition, internet based Blogs are becoming more and more popular in extension programs. These outlets, combined with smart phones, are putting the world at each person's fingertips and making extension programs more efficient in an ever changing agricultural environment.

The Mississippi agricultural research and extension programs have followed a similar path. The newsletter based system was started as the "Cotton Insect Situation" newsletter and was very successful. In 2003, the newsletter was expanded to include multiple crops and eventually all crops and disciplines by 2008 as the "Mississippi Crop Situation" newsletter. The newsletter was primarily distributed through email and in 2010 became interactive with clickable links to other resources. During the spring of 2011, the newsletter was replaced with an online blog (www.mississippi-crops.com). This avenue has proven to be very successful for getting up to date information to the agricultural industry in a timely manner. Additionally, the Mississippi Crop Situation blog has a link to Twitter (#mscrops) and the fan page on Facebook. This allows growers and consultants to follow the blog and receive updates on their phone almost immediately after they are posted. The success of these outlets in Mississippi is due to the fact that the extension personnel use them as a tool to transfer important information to their clientele, not as a replacement for traditional extension programs. Personal relationships are the foundation of a successful extension program. Social media outlets have been used successfully in Mississippi to compliment rather than replace more traditional extension roles, most notably, on farm visits.

Use of an Insect Growth Regulator Applied at the 2nd Week of Cotton Squaring for Management of *Lygus* Bugs: A Cotton Insecticide Efficacy Trial – 2011-2012

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Efficacy trials were conducted in 2011-2012 utilizing various insecticides and rates to determine their impact on Lygus bug infestations in commercial cotton fields located in California's southern desert. Cotton fields (var. 'DP 164') were located adjacent to alfalfa fields grown for forage. Alfalfa is more attractive food source to Lygus than cotton. However at harvest, Lygus bugs are forced to move to available plants to feed upon, often, it is nearby cotton. Severity of *Lygus* bug populations moving from alfalfa at harvest into cotton varies by year and can negatively impact cotton yield and quality. Treatments were applied with a high clearance sprayer to a field using a split plot (n = 2)design with sub-plots (n = 8) applied to replicated (n = 3) eight row treatments. Lygus bug (both life stages) and selected natural enemies (Zelus spp., green lacewing complex (Chrysoperla and Chrysopa spp.), Geocoris spp.) were quantified every ≈7d using sweep net sampling (n = 50/treatment) for 21d following treatments. Cotton square and boll counts were quantified every ≈7d from 10 randomly selected plants/treatment plots until seasons end. Main plot treatments were applied during the 2nd week of cotton squaring on 7/1/2011. Sub-plots were treated on 7/29/2011 & 8/6/2011. Use of the insect growth regulator, Diamond 0.83 EC (266.16 ml/0.40 ha) as a main treatment yielded significantly more (P = 0.0001; df = 40) cotton on average than use of Carbine 50 WG (62.37 gm/0.40 ha) (1,286.47 Kg and 1,120.59 Kg respectively).

Genetic Variation of *Lygus lineolaris* in Eastern North America Observed with Mitochondrial and Nuclear DNA Sequences

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Lygus lineolaris is the pest Lygus species of note in North America from the Great Plains to the Atlantic Coast and from southern Canada to the Gulf of Mexico. DNA sequences from three regions have been used to explore its genetic diversity. Examination of a 768-bp segment of mtDNA spanning the cox1-cox2 genes has recovered 60 haplotypes from 258 insects. Four of the haplotypes are comparatively common while the remaining 56 were observed only once or twice. Phylogenetic trees and haplotype networks reveal two genetic clades with somewhat different geographic distribution. Although the two clades are sympatric, one is much more numerous in the northern part of the species' range. A 658-bp portion of the mitochondrial cox1 barcode region supports the two clades but lacks the haplotype diversity. It was easier to screen and was employed to provide further details regarding the distribution of the clades. A nuclear fragment comprising 452 bases flanking a microsatellite locus (MSFR) separates into two common clades and a third clade that is rare. The two common clades differ by 10 substitutions. The third clade differs by 12 and 17 substitutions from the other two. The mitochondrial and nuclear clades are not coincident, although 90% of clade A MSFR have been mitochondrial clade 2 and 72% of MSFR clade B have been mitochondrial clade 1. There is no apparent link between the mitochondrial or nuclear genotypes and the host plant origins.

Overview of *Lygus* Research and Outreach Program in the Texas High Plains: Serving the Clientele of the World's Largest Cotton Patch

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The Texas A&M AgriLife Cotton Entomology Program is located in the world's largest cotton patch- the Texas High Plains. At the local level, the Program serves 25 cotton producing counties, including about 1.8 million ha of cotton in 2011. Insect pests are one of the major crop limiting factors that cause approximately a 4% annual crop loss costing about 80 million dollars for Texas High Plains cotton producers. Program goals include developing knowledge, skills, tools and technologies for sustainable insect pest management and improve the yield and quality of Texas High Plains cotton. Texas A&M cotton entomology research in the Texas High Plains began in 1937, but the focus on Lygus research was started in 2002. Our program has used Lygus as the model insect to answer various ecological questions in cotton pest management. In the past 11 years (2002-2012), more than 20 research projects were conducted on various aspects of Lygus biology, behavior, and ecology. Experiments were conducted in the laboratory, greenhouse, research farms, and growers' fields. Lygus researches conducted in our program includes host-plant survey, life table, host preference, intercrop movement, feeding biology, cotton plant-Lygus interaction, sampling, insecticide resistance, pesticide evaluation, overwintering biology, morphology, molecular biology, cultural control, landscape structure, and economic threshold development. Included were some basic research projects but most were focused on applied issues to improve Texas cotton Lygus management practices. Some major program outputs consist of alternate host identification, feeding and movement biology characterization, pesticide spray initiation and termination rules development, molecular marker development, genetic structure determination, pesticide resistance monitoring, morphological characterization, life table investigation, and crop protection product evaluation. The Cotton Entomology Program has contributed significantly to assisting Texas cotton producers, crop consultants, Extension agents and the scientific community by expanding our cotton pest management knowledge and skills through research and outreach.

Expression Analyses of ATP-binding Cassette Transporter Proteins in the Western Tarnished Plant Bug, *Lygus hesperus* Knight

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The western tarnished plant bug Lygus hesperus Knight is a pest of many crops in the western US, including cotton, alfalfa, numerous fruit and vegetable crops. and potentially of several emerging biofuel and natural product feedstocks. Control strategies have traditionally relied on broad-spectrum insecticides; however, reports of resistance in field populations of the sister species, L. lineolaris, suggest that the arsenal available for effective Lygus management may become more limited. Further exacerbating its pest status, little is known concerning the molecular mechanisms underlying L. hesperus biology, in particular those associated with insecticide resistance. ABC (ATP-binding cassette) transporter proteins, which play important functions in cellular transport by actively shuttling small molecules out of cells, have been linked to insecticide resistance. To better clarify the role these proteins have in Lygus, we used an adult *L. hesperus* transcriptome to identify potential ABC transporter homologs. More than 30 transcripts were identified. Those that had highest similarity with multidrug resistance (MDR) transporters, a subset of ABC transporters linked to xenobiotic clearance, were screened via semi-quantitative PCR for sex specific head, thorax, or abdominal expression. Abdominal/thoracic predominant expression was observed for one ABCA-like transporter and four MDR-like transporters. A second screen of abdominal specific tissues revealed that all of the MDR-like transcripts were expressed in male accessory glands and that the ABCA-like transcript was most highly expressed in testes. Malpighian tubule expression was also observed for two of the MDR-like genes, LhMDR-5 and LhMDR-7. Because Malpighian tubules are involved in xenobiotic detoxification, these two gene products have been selected for further study.

Feeding Behavior and Injury Caused by Lygus hesperus

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Despite the importance of *Lygus hesperus* as a crop pest in the western United States, key aspects of its feeding behavior and consequent injury to cotton are poorly understood. Previous studies of Lygus stage-dependent injury to cotton produced conflicting results. We sought to clarify these relationships by employing an experimental strategy that integrated observations of feeding behavior and within-plant distribution with traditional assays of plant injury. Video-based assays indicated that prereproductive adults allocated more time to feeding and less time to trivial movement compared with reproductive adults. These observations were consistent with results of greenhouse assays, where prereproductive adults more often resided on vegetative or reproductive buds, and produced higher numbers of injured and abscised squares, compared with reproductive adults. Direct comparisons of feeding injury to intact plants by third and fifth instars, and prereproductive adult females, indicated plants exposed to third instars had more injured squares attached to plants whereas the plants exposed to fifth instars had more abscised squares. These differences were not explained by respective feeding behaviors observed in video assays, but were associated with differences in within-plant distribution corresponding to insect age class. Fifth instars were often observed feeding in plant terminals containing small squares that are highly susceptible to abscission. Third instars were most often found sheltered within the bracteoles of larger squares that could better tolerate Lygus feeding. In contrast, prereproductive adults were less frequently associated with fruiting structures than were either nymphal instar. Our results indicate that age-dependent differences in Lygus injury to cotton can be explained by corresponding differences in behavior. These findings emphasize that variability in studies of plant injury can be minimized by controlling Lygus age class, and suggest the need to include estimates of nymph populations in treatment thresholds.

Measuring Up! Involving Stakeholders in Assessment of an Industry's IPM Revolution

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The availability of accurate, real-world data on pest management practices, crop pest losses, and associated costs are critical to assessing the adoption and impact of IPM programs. We engage agricultural stakeholders through annual survey workshops to develop data on crop pest losses, control costs, target pests, and pesticide use. These data, now spanning over 30 years for cotton, are useful in documenting adoption of IPM practices, economic savings to growers, and large-scale changes in pest management practices. The workshops encourage and reward stakeholder input, foster collaborative relationships with key stakeholder groups, and provide high quality data on pest management practices and their economic impacts. For example, the last 5 years have shown the lowest insecticide use in cotton on record (32 years) at just 1.5 sprays season-long, reducing insecticide loads on the environment by more 1.6 million pounds of active ingredient annually and saving growers over \$10 million per year. In addition to quantitative data, stakeholders identify the specific intent or intended targets of pesticide inputs, so the resulting data provide unique insights into the decision-making experience of each pest manager. These insights help guide existing and new programs in IPM research, implementation, and outreach. Our dialog with stakeholders helps us identify emerging pest issues and changing needs of stakeholder communities. The ability to measure impacts and industry practices is useful for generating interest in and sustaining support for our IPM programs, which in turn have produced great economic benefits for growers.

Preliminary Data on the Survival and Development of *Lygus lineolaris* Collected from the Delta and Hills Regions of Mississippi

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Lygus lineolaris feeds on most of the crops grown in Mississippi and has been the most important pest in Mississippi cotton for the past several years. Though polyphagous, they seem to preferentially feed and build large populations on a few crops and weed hosts. Recent research has shown that L. lineolaris from the Delta and Hills regions of Mississippi may respond differently to cotton. To further explore differences between these regions, four colonies of L. lineolaris from the Delta (2 colonies) and Hills (2 colonies) regions of Mississippi were used to compare survival and development times on corn, cotton, soybean, and artificial diet. Data indicate that there is little or no difference between the regions for either survival or development time. Over both regions, survival showed a trend of artificial diet>corn>cotton>soybean. The trend for overall development time indicated that development rate was slowest for *L. lineolaris* reared on corn and soybean and fastest on artificial diet and cotton. These preliminary data conflict with previous data and suggest that L. lineolaris populations in different regions of Mississippi may not be different after all; however the food source does affect survival and development time of L. lineolaris from both the Delta and Hills regions of Mississippi.

Refined Management of Western Tarnished Plant Bugs in Baby Lima Beans

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The western tarnished plant bug, Lygus hesperus Knight, is the most challenging insect pest for the production of several types of beans in California. Bean yield and quality can be severely impacted. Studies from 2010-11 were conducted to improve management options for this pest on baby lima beans. Experimental and registered insecticides were evaluated against this pest in small plots. Rimon[®], a recently registered product, was tested in grower fields and found to suppress lygus nymphs. However, since Rimon does not control adult lygus, it must be tank-mixed with another insecticide, which may not be economical in lima bean production. 'Haskell', a vine baby lima variety with a level of resistance to lygus bugs, was grown with various insecticide regimes intended to supplement lygus management under high pressure. The lygus bug pressure in 2011 was too low and too late in the season to complete this objective. Results from 2010 under higher pest pressure were more compelling; in untreated plots the yield was more than doubled in 'Haskell' compared with 'Mezcla', an unimproved line. Haskell's vigorous growth and host plant resistance traits allow it to partially withstand the infestation compared with the unimproved line. Indoxacarb, the "soft" insecticide approach, resulted in a ~50% yield increase compared with untreated plots for both 'Mezcla' and 'Haskell'. However, use of the "harsher" insecticide (Mustang Max[®]) resulted in a doubling of yields compared with the untreated plots for both varieties. Therefore, the soft approach did prove beneficial and was likely cost-effective but to maximize yields the conventional insecticide was justified. Studies were conducted to determine the appropriate method to sample lygus bug populations for treatment decisions in vine baby lima beans (such as 'Haskell'). The dense vine bean canopy inhibits the effectiveness of the standard sweep net sampling tool.

Quantifying Consumption of Protein-Tagged *Lygus hesperus* by the Cotton Predator Complex

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A new method for post-mortem quantification of predation on prey items marked with protein antigens is described. First, short-term protein marking retention tests were conducted on the targeted prey, immature Lygus hesperus. Chicken IgG, rabbit IgG, or soy milk proteins were readily detectable by a suite of protein specific enzyme-linked immunosorbent assays (ELISA) on the L. hesperus. Then, predator gut content assays were conducted on chewing and piercing-sucking type predators that consumed a 3rd instar L. hesperus marked with rabbit IgG. The rabbit IgG gut content ELISA detected the marked prey in the vast majority of both types of predators for up to 24 h after feeding. Finally, field cage studies were conducted to quantify predation rates of the natural cotton predator assemblage on protein marked *L. hesperus* nymphs. Each 4th instar L. hesperus marked with rabbit IgG, chicken IgG, and soy milk was released into one of 360 field cages containing a cotton plant and the natural population of predators. After 7 h, each caged plant was pulled from the field, the number of predaceous arthropods in each cage were tallied, and each individual predator was assayed for the presence of marked prey by a suite of protein-specific ELISAs. A procedural error with the soy mark application negated the anti-soy ELISA data, but the anti-rabbit IgG and anti-chicken IgG ELISAs pinpointed exactly which predators preyed on the IgG marked nymphs. The protein-specific gut ELISAs revealed that various members of Araneae, Heteroptera, and Coleoptera were the most common predators of the marked prev items. In all, 74 predation events were recorded in the guts of the 556 predators encountered in the field cages. Of these 26, 23, and 14 marked individuals were eaten by various members of Araneae, Heteroptera, and Coleoptera, respectively. This study verifies that prey immunomarking is a simple, versatile, and effective method for quantifying predation rates on *L. hesperus*.

Transcriptome Analysis and Expression Profile of Odorant Binding Proteins in the Tarnished Plant Bug, *Lygus lineolaris*

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L. lineolaris (tarnished plant bug) is an agronomically important pest species of numerous cropping systems, and is currently considered to be one of the principal pests of cotton in the southeast US. Similar to other insects, a number of L. lineolaris behaviors are driven by the perception and discrimination of semiochemical cues. Odorant binding proteins (OBPs), relatively small (~15-20 kDa) water-soluble proteins that transport and deliver semiochemicals to odorant receptors in sensory neurons, are thought to comprise an initial discriminatory filter. To begin to clarify the role of individual OBPs in L. lineolaris olfaction, we used a L. lineolaris transcriptome to identify potential OBP homologs. From this search, we identified 33 L. lineolaris OBPs (LylinOBP), including the previously characterized Lygus antennal protein (LAP). Based on the characteristic cysteine spacing patterns, the LylinOBPs were classified into two groups "classic" OBPs (six cysteine motif) and "Plus-C" OBPs (more than six cysteine residues in conjunction with a highly conserved proline). Phylogenetic analyses indicated that the LylinOBP transcripts clustered with OBP sequences derived from other Mirid species, suggesting that they have been evolutionarily conserved and may discriminate similar odorant compounds. Microarray-based expression analyses of chemosensory tissues (i.e. antennae, proboscis, and legs) from 2-day-old male and female adults revealed >10-fold increase in LylinOBP expression compared to controls for 21 transcripts in antennae, 17 in proboscis, and 11 in legs. Expression data for a subset of the transcripts were verified by quantitative PCR.

Retrospective Analysis of the Successful Establishment of a *Lygus* biocontrol Agent Using Microsatellite Data

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Since the 1970s, insecticide-resistant populations and environmental concerns have motivated attempts to manage tarnished plant bugs (Lygus spp.) by classical biological control. Peristenus digoneutis, a western Palearctic nymphal parasitoid of L. rugulipennis and related species in Europe, was introduced repeatedly from 1979 to 1988 into northern New Jersey (USA) against the North American L. lineolaris. It successfully established and now occurs in at least 11 northeastern U.S. states and three Canadian provinces. Establishment followed successive introductions of genetically differentiated geographic populations from various locations in France and Austria. Historical knowledge of the introduction and colonization of *P. digoneutis* relies on detailed collection, importation and release records and recovery data. These cannot answer questions about the ancestry and genetic composition of populations that established. New genetic analysis tools allow researchers to extract historical information from molecular data, making it possible to answer such questions. To analyze established *P. digoneutis* populations, microsatellites were developed *de novo* using pyrosequencing technology. Twenty-one polymorphic microsatellite loci were obtained that are suitable for assessing genetic variation on a fine scale and for reconstructing the most likely demographic scenario that contributed to the successful establishment of *P. digoneutis* in NJ. Results based on a limited sampling show substantial genetic variability in established NJ populations with at least 4 alleles per locus, comparable to what was observed in Drôme, a region of origin in France. At each locus NJ and Drôme share most alleles (though these alleles could also occur elsewhere). Most of the alleles present in NJ were also found in Pennsylvania, where P. digoneutis was not released but subsequently dispersed. However, several alleles were found in Pennsylvania that were not seen in NJ samples. Genotyping will continue with larger samples from NJ and other populations in Europe and the USA using historical vouchers and new collections.

Effect of Lygus lineolaris on Different Growth Stages of Navy Beans (Phaseolus vulgaris L.)

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Little information is available on the nature of injury and the damage caused to dry edible beans by feeding of Lygus lineolaris. In controlled environment growth rooms, we investigated the relative vulnerability of navy beans at different growth stages to L. *lineolaris*. Adults or 5th instar nymphs were confined for five days to a single reproductive structure of a navy bean plant at either R2 (mid- to full-flowering), R4–R5 (mid-pod to early seed fill) or R6-R7 (mid-seed fill to seed maturity) growth stages and. The types of injury were characterized by light microscopy immediately after insect removal, and the effect on yield was determined by growing the plants through to harvest, and assessing seed weight and quality. Different types of injury occurred at each growth stage. The major effects of feeding at each stage were: pod abortion at R2, injury to the funiculus and placenta at R4-R5, and seed pitting at R6-R7. Seed weight losses due to insect treatments were significant when feeding occurred at the R2-R3 and R4-R5 stages but not at the R6-R7 stage; where losses were significant, nymphs reduced seed weight more than adults. Yield quality loss due to seed pitting was significant when feeding occurred at the R6-R7 stage, but feeding at other stages did not cause detectable reduction in seed quality.

A Novel Bioassay to Evaluate *Beauveria bassiana* Strain NI8 and the Insect Growth Regulator, Novaluron, Against *Lygus lineolaris* on a Non-Autoclaved Solid Artificial Diet

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A new method to evaluate the potential of *Beauveria bassiana* strain NI8 and the growth regulator novaluron (Diamond ®) for tarnished plant bug (TPB), Lygus lineolaris Palisot de Beauvois (Hemiptera: Miridae) control was developed using a non-autoclaved solid diet. Diet ingredients include toasted wheat germ, ground lima bean meal, soy flour, yolk chicken eggs, inhibitor and agar. The diet was prepared in one step by blending the ingredients in heated boiled water, and it was suitable for bioassays of TPB from second instar to adult that required continuous observation over a period of several weeks. Fourth instar (97.5 ± SE 0.02), fifth instar (95.0 ± SE 0.03) and adults (95 ± SE 0.03) of TPB were more susceptible (infection %) than second (52.5 ± SE 0.07) and third instar (85.0 ± SE 0.05) to *B. bassiana*; while, second instar (100%), third instar (100%) and forth instar (97.5 ± SE 0.02) nymphs had higher growth inhibition (mortality %) than fifth instars (92.5 ± SE 0.04) after ten days of exposure to novaluron. No effects in longevity (days) were observed in adults $(21.57 \pm SE 0.9)$ treated with novaluron when compared with control insects exposed to water alone (20.47 ± SE 1.2), but both had highly significant greater longevity than adults exposed to B. bassiana (5.2 ± SE 0.2). Adults of TPB were maintained for over a month without changing the diet. The non-autoclaved diet is semi-liquid before it cools which facilitate the mechanics of diet packaging similar to food packaging or lepidopteran diet preparation. The solid artificial diet for Lygus bugs provides improved research capacity for studying the ecology and susceptibility of the insect to a number of different control agents include beneficial organisms, insect pathogens, and insecticidal toxins being developed for transgenic technologies.

Complete Mitochondrial Genomes of Lygus lineolaris and Lygus hesperus

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The mitochondrial genome of Lygus lineolaris is 17027 bp long. By comparison the Lygus hesperus genome is 17747 bp in length. Both genomes contain 13 protein coding regions, 22 tRNA genes, and two ribosomal RNA genes. The gene arrangement corresponds to the common order found among insect mtDNAs. When all comparable positions excluding the control region (14709 bp) are considered, the difference between the two genomes is 3.3%. If only the ORFs (7450 bp) are compared, the sequence divergence is 1.4%. The net 720 bp difference in genome lengths is completely accounted for by differences in the control regions. The ten indels in the remainder of the sequences are equally divided between the two species and do not affect the total length. The control regions contain an array of repetitive sequences. There are eighteen 61 bp repeats in the control regions of *L. hesperus* that are composed of eight slightly different sequences. The single copy of this region from in L. lineolaris differs by ten substitutions (16.4%) from the L.hesperus consensus. A long 161 bp repeat unit consists of several copies of the long sequence plus an additional truncated piece from one end of the long segment. In L. lineolaris there are five copies of 161 bp and one of 160 bp. The partial copy is 59 bp. For *L. hesperus* four full copies were detected. Three are 162 bp and the fourth one is 161 bp. The short piece is only 47 bp. The shortest repeat at 24 bp was found in seven copies in *L. lineolaris* and six copies in L. hesperus. Two substitutions separate the 24 bp consensus sequences of the species. The control region could be a source of species diagnostic sequences.

Variation in Bacterial Communities Associated with the Cotton Fleahopper on Two Host Plants Across Texas, USA

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Herbivorous insects are known to harbor microbial symbionts that can provide several ecological advantages. For example, in some insects bacterial symbionts allow them to use otherwise inadequate host plants. Such mutualistic associations can lead to the formation of genetically divergent host-associated populations of insects. Previous studies have indicated the presence of host-associated genetic differentiation (HAD) in the cotton fleahopper (CFH), Pseudatomoscelis seriatus (Miridae), associated with two host plants: cotton and horsemint. We investigated the potential role of microbial diversity in HAD using 454 pyrosequencing of bacterial 16s ribosomal subunit DNA to characterize the bacterial communities associated with CFH from populations utilizing cotton and horsemint across Texas. We detected both geographic and host-plant associated patterns of bacterial community diversity, variation and composition. Notably, we also found that the facultative endosymbiotic bacteria, Wolbachia, thought to be widespread in many insects, was absent in all populations except one in Weslaco, TX. Further phylogenetic analysis of Wolbachia sequences from this site indicated the presence of two different Wolbachia strains in cotton-associated and horsemintassociated CFH populations. This preliminary study indicates that symbiotic associations of insects might play an important role underlying genetic structure in herbivorous populations.

Infestation Levels of Tarnished Plant Bug, *Lygus lineolaris*, in Different Cotton Varieties in Arkansas

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The tarnished plant bug is one of the most important pests of cotton in the Mid-South. Applying recommended insecticides when bugs reach treatment level is the most commonly used option to control this pest. However, increasing levels of resistance to insecticides are beginning to make some chemistries less effective. Previous studies have shown that some cotton varieties sustain significantly lower damage from TPB in small, 2-row, plots. Utilizing varietal resistance to manage TPB populations in cotton would reduce the number of insecticide applications made annually for this pest and should help delay the further development of insecticide resistance. Varieties exhibiting resistance in small plots were evaluated in large plot studies conducted at the Northeast Research and Extension Center, Keiser, AR in 2010-2012. Each variety was grown under 2 treatment regimes, 1) treated when tarnished plant bugs reached threshold of 3 bugs per 5 row feet, 2) untreated control. Surveys of grower fields were also conducted in 2011-2012. In most instances in large plot studies, resistant varieties reached treatment threshold fewer times when compared to a susceptible variety. In surveys of grower fields, resistant varieties averaged half as many applications for tarnished plant bugs as susceptible varieties in 2011. Tarnished plant bug populations were extremely high in 2012 and although resistant varieties in grower fields reached threshold fewer times than susceptible varieties, this difference was not as big as in 2011 when populations were more moderate.

Tarnished Plant Bug (*Lygus lineolaris* (Palisot de Beauvois) Host Plant Resistance Evaluations in Midsouth Cotton

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Tarnished plant bug (Lygus lineolaris Palisot de Beauvois) is an important pest of U.S. cotton in the Midsouth production region. Chemical control is the principal tactic used in tarnished plant bug pest management programs; however, there is a heightened interest among cotton producers in accessing commercially acceptable cultivars with host plant resistance to Lygus. Efficient techniques for screening cotton lines for resistance are key to the success of breeding programs emphasizing host plant resistance. On-going research in Arkansas, funded through Cotton Incorporated, has included an annual survey of commercially available cotton cultivars for resistance to tarnished plant bug by monitoring white flower anther injury. Additional work has focused on development of screening techniques for eliminating the preference factors of survey materials for plant bug resistance. This work has included use of a simple laboratory arena test that allows evaluation of feeding preferences of tarnished plant bug nymphs. Survival levels of plant bug nymphs among cultivars has been contrasted using caged field plants as well as caged main stem terminal cuttings from field plants. Results show promise for expanding further host plant resistance screening protocols. If confirmed, resistance of cultivars and breeding lines could be compared by taking plant cuttings from existing field tests.

Sulfoxaflor - Mode of Action and Basis for Efficacy on Resistant Insects

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Sulfoxaflor, the first product from the new class of sulfoximine insecticides, is a broadspectrum sap-feeding insecticide with excellent efficacy against aphids, whiteflies, hoppers, scales, mealybugs, psyllids and Lygus. Sulfoxaflor acts on the nicotinic acetylcholine receptors (nAChRs) of insects in a manner that is distinct compared to all other insecticides, including the neonicotinoids. Importantly, sulfoxaflor is effective against pest insect strains that are resistant to a wide range of insecticides, including the neonicotinoids. The lack of cross-resistance to sulfoxaflor in these insecticide resistant strains is correlated with metabolic stability to cytochrome P450 monooxygenases, which available information suggests is the primary resistance mechanism in most neonicotinoid-resistant species. CYP6G1, a monooxygenase associated with resistance to neonicotinoids and other insecticides readily metabolizes a variety of neonicotinoids, but not sulfoxaflor. Computer modeling studies of a Drosophila monooxygenase CYP6G1 suggest that the unique structure of sulfoxaflor limits its ability be metabolized. In addition, Drosophila strains possessing mutant Da1 and D_β2 nAChR subunits, conferring resistance to a variety of neonicotinoids, show little cross resistance to sulfoxaflor. Similarly, a Drosophila strain exhibiting resistance to spinosad (altered Da6 nAChR subunit) also lacked cross-resistance to sulfoxaflor. The novelty of the sulfoximine chemistry coupled with the lack of metabolism based resistance, provided the rationale for the placement of sulfoxaflor in mode of action subgroup 4C, by the Insecticide Resistance Action Committee (IRAC), differentiating it from other Group 4 insecticides (Group 4A). Consistent with IRAC guidelines, sulfoxaflor should not be rotated with other Group 4 insecticides except where no other options are available. In summary, sulfoxaflor is a new, effective tool for managing Lygus and other sap-feeding pest insects, including those resistant to the currently used insecticides.

Oviposition Choices by Lygus lineolaris Affect Egg Parasitism by Anaphes iole

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Natural enemies are important mortality factors for herbivores. In response to natural enemy pressure, herbivores can alter life history decisions, such as oviposition behavior, so that offspring are better protected from natural enemies. An example is oviposition into structures where vulnerability to natural enemies is reduced, i.e., use of enemy-free space. We investigated this phenomenon using a well-known herbivore-parasitoid association. *Lygus lineolaris* (Palisot de Beauvois) is native to North America and has a broad host range (>350 plant species). This bug's eggs are attacked by a native parasitoid, *Anaphes iole* Girault, and parasitism levels may vary greatly among host plants. We evaluated the egg distribution pattern of *L. lineolaris* on several crop and non-crop species, and subsequent parasitism by *A. iole* to determine whether oviposition choices by *L. lineolaris* females protect their eggs from parasitism. Our results indicate that the reproductive structures of several host plants provide a refuge from parasitism for *L. lineolaris* eggs. Oviposition site choice by female bugs appears to be a selective strategy to take advantage of enemy-free space.

Salivary gland gene profile and potential association with feeding damage from tarnished plant bug *Lygus lineolaris*

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In an attempt to identify genes responsible for feeding and damaging to field crops, cDNA library was constructed with salivary glands dissected from the tarnished plant bug, *Lygus lineolaris*. Approximately 7,000 clones were sequenced, and the sequences were assembled to 666 contigs. By using Blast searching of GenBank databases, the identities of 432 cDNAs were revealed, and the other 234 cDNAs have not been identified. These 432 sequences code at least 60 different enzymes.

Endopolygalacturonases, involving degradation and remodeling of the plant cell wall, are the most abundant enzymes in the salivary glands. Other enzymes, such as endo-1 4-beta-xylanases and trypsins, are also abundantly present. In addition, several cytochrome P450 and glutathione S-transferase cDNAs were identified from the salivary glands. Of the 432 identified genes, 32% of the genes are associated with metabolic processes (mostly for primary metabolic, macromolecular metabolic, and cellular metabolic processes), and 47% of the genes are involved in catalytic (mainly hydrolase) activities.

A Novel Messaging System for Passive Mate Guarding in Lygus hesperus

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Male Lygus hesperus deliver a large spermatophore to females during copulation. Among the materials transferred is an odorant molecule, myristyl acetate (MA), which renders females less attractive to other males as a form of passive mate guarding. This antiaphrodisiac is produced in the male accessory glands, has a dose-dependent effect, and over time its effect decreases. To better understand what other accessory glandderived factors might contribute to the suppressive effect, and to elucidate how female regain their attractiveness, we examined MA and two other compounds, A and B, which are also found in females after mating. We found that the concentration of MA detectable in females decreases steadily after mating, just as their attractiveness to males increases. Compound A also follows this pattern of decline. Compound B appears to be a degradation product of compound A and its concentration in the female increases over time. Neither compound A nor B, by themselves or in combination with each other or MA, enhanced the antiaphrodisiac effect. However, compound B topically applied to a female already expressing MA was able to render those females attractive once again, and this anti-antiaphrodisiac effect was dose dependent. The results suggest that *L. hesperus* males may have evolved the capacity to monitor female status by the relative proportion of MA and compound B. This dual approach would allow potential suitors to more accurately judge when a female might be ready to mate again compared to relying on the concentration of a single chemical that is delivered in varying quantities by males.

Thermal Ecology of Lygus hesperus Development

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Astute understanding of the interactions between a pest organism and its abiotic environment is necessary for development of improved, ecologically-based management strategies. Relationships between temperature and development of an important pest of crops in the western United States, the western tarnished plant bug (Lygus hesperus Knight), were previously described. However, these descriptions relied on linear models that inadequately represented influences of temperature on Lygus development. We reexamined development of L. hesperus eggs, nymphs, and adult reproductive organs at constant temperatures, and described observed temperature dependencies using nonlinear biophysical development rate models. Egg development was observed at 10C within green beans and an agarose oviposition substrate, but hatch occurred only for eggs developing in agarose. Developmental abnormalities were observed for eggs in both substrates at 37.8C, and no nymphs hatched. Newly-emerged nymphs did not survive to adulthood at 10 or 37.8C. However, nymphs of each instar initially reared at 26.7C and transferred to 10 or 37.8C developed to the subsequent instar, except for fifth instars transferred to 37.8C. Both eggs and nymphs exhibited low- and high-temperature inhibition of development, and a developmental optimum temperature of about 32C. When adults were held at temperatures from 12.8 to 35C, all phases of reproductive development exhibited mild high-temperature inhibition, except the completion of seminal vesicle filling was strongly inhibited by high temperature. Optimum temperatures for reproductive development varied among the organs examined, and among developmental phases within an organ. Interpretation of these optima is not straightforward because initial development of some reproductive organs occurs during the nymphal stage, and observed development time to later phases of organ development incorporates temperature influences on earlier developmental phases. Our collective results provide new insights into the interactions between L. hesperus and its environment that will permit more meaningful interpretation of seasonal ecological patterns.

Production and Predator-Induced Release of Defensive Chemicals by the Plant Bug Lygus hesperus

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Both sexes of adult western tarnished plant bug, Lygus hesperus Knight (Heteroptera: Miridae), released three volatile chemicals in relatively large amounts when attacked by ants (Pogonomyrmex rugosus and Solenopsis xyloni) or when grabbed by forceps [Byers (2006) J. Chem. Ecol. 32:2205-2218]. The defensive volatiles were detected by solid-phase microextraction (SPME) and gas chromatography-mass spectrometry (GC-MS). The relative amounts of the volatile compounds, hexyl butyrate, (E)-4-oxo-2hexenal, and (E)-2-hexenyl butyrate, absorbed by SPME were similar in males and females (about 100%, 44%, and 4%, respectively, from females, and 83%, 37%, and 3% from males). Ants of both species upon attack of L. hesperus adults were repelled by the bug's defensive discharges (confirmed by SPME). Sexually mature L. hesperus were individually extracted in pentane to determine the mean amounts of hexvl butvrate (14.9 µg/female; 10.3 µg/male), (E)-4-oxo-2-hexenal (2.7 µg/female; 3.1 µg/male), and (E)-2-hexenyl butyrate (1.2 µg/female; 0.6 µg/male). (E)-4-Oxo-2-hexenal was unstable in solvent when kept in contact with a macerated adult. The production of the three major volatile components began about 2 d after molting to the adult and amounts increased for about 5-10 d with little or no increase thereafter. A cost of defensive secretion is suggested for females but not for males, because heavier females produced more volatile compounds than lighter females. The initial discharge percentage (IDP), defined as the proportion of volatile compounds initially present that is discharged to defend against predation, was estimated at about 60% (50% in males and 70% in females).

Monitoring and Control of Pest Mirid Species in the UK Using Sex Pheromones

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Mirid bugs are important pests of several high-value horticultural crops in the UK. The most important species include the common green capsid, Lygocoris pabulinus, and the European tarnished plant bug, Lygus rugulipennis. L. pabulinus is a sporadic but very damaging pest of apples, pears, blackcurrants, strawberries and increasingly cane fruits and L. rugulipennis is an important pest of late season strawberries and of various glasshouse salad crops, notably cucumber. Crop invasion by mirids is sporadic and unpredictable. They cause damage at low population densities and are difficult to detect at such levels in normal crop inspections.

Virgin females of both these species produce sex pheromones attracting males. We have shown that individual, undisturbed females produce three compounds in species-specific blends. The compounds are hexyl butyrate, (E)-2-hexenyl butyrate and (E)-4-oxo-2-hexenal. These compounds have been reported to be sex pheromone components of several other Lygus species, but an effective lure has never been developed. We have now designed dispensers which release specific blends of these compounds for at least four weeks under field conditions. Trap design is also important. Green cross-vane traps are best for L. rugulipennis but proved ineffective at catching L. pabulinus for which blue sticky traps are required. Traps baited with appropriate blends of the three pheromone components caught as many males as traps baited with live females of the same species.

Extensive trapping in affected crops has shown the traps provide an excellent early warning system of invasion into the crops. L. rugulipennis is detected in the pheromone trap at least two weeks before detection in cucumber glasshouses and more than two weeks before detection in strawberry plantations using traditional monitoring methods.

Tarnished Plant Bug, *Lygus* lineolaris, A Potential Biotype Difference in the Mississippi Delta

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A laboratory experiment was performed to compare fitness parameters of tarnished plant bug populations collected from the Hills and Delta regions of Mississippi. Each population was split into two cohorts to be reared on cotton or artificial diet to make comparisons of food source as well as region of collection. Data were analyzed using analysis of variance and regression analysis. Populations were collected from pigweed in four locations in each region. Each population was maintained separately and allowed to mate. Progeny from the F1 generation of each population were compared from each region and food source. Parameters measured included development times to fourth instar, fifth instar and adult, total nymphal survivorship, fecundity, and fertility. Populations collected from the Delta region and reared on cotton developed significantly faster to all life stages than other populations while populations from the Hills reared on cotton were significantly slower than other populations except Hills populations reared on artificial diet. There were no significant differences for percent survivorship for region of collection; however, populations on diet had significantly higher survivorship than those reared on cotton. Populations of tarnished plant bug from the Delta region laid significantly more eggs per female per day than populations from the Hills region. Populations reared on cotton also laid significantly more eggs per female per day than those reared on diet. Populations collected in the Delta region laid significantly more viable eggs per female per day than those from the Hills region. Populations reared on cotton produced significantly more nymphs per female per day than those reared on diet. There were no significant differences in mean percent hatch of total eggs laid for region or food source. These data indicate there are differences in several fitness parameters between tarnished plant bug populations from the Hills and Delta regions of Mississippi.

Transcriptomics and Genomics of the Tarnished Plant Bug, Lygus lineolaris.

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Messenger RNA pooled from tarnished plant bugs that included developmental stages from embryos (12 hour intervals) to 5th instar nymphs, and 3rd to 5th instar nymphs and adults treated with 5 different classes of insecticides (to induce genes involved in detoxification) was used to obtain nucleotide sequence reads (over 500,000 Roche 454 sequence reads and 18 million Illumina GAII reads) of expressed genes. The initial assembly resulted in a total of 79,444 contigs, of which 52,054 were 80 nucleotides or longer. The longest contig assembled was 7,318 nt with 20.5-fold coverage. A total of 3,833,035 sequence reads containing 317,782,637 nucleotides contributed to the assembly of contigs ≥80 nt with an N50 of 973 nt. The coverage of contigs ≥80 nt ranged from 1482.9 to 1.3-fold with an average of 10.4. The cumulative length of assembled contigs was 29,644,105 nt. Refinement of the transcriptome assembly with additional Illumina HiSeq2000 reads resulted in 37,934 unique transcripts of 300 nucleotides or longer, of which 19,460 sequences had one or more BLAST description and 13,788 sequences were mapped to at least one gene ontology (GO) term. Bacterial artificial chromosome (BAC) library containing 36,500 genomic clones were sequenced using Illumina HiSeg 2000 platform and assembled to obtain partial genomic sequence data. Illumina sequence reads generated from individual insect genomic DNA were assembled using BAC genomic sequence contigs were used as reference to identify polymorphic genetic markers suitable mapping and population genetic studies. Efforts are under way to sequence, assemble, annotate the ~900 Mbp genome of L. lineolaris.

Pyrosequencing of the Adult Tarnished Plant Bug, *Lygus lineolaris*, Characterization of Messages Important in Metabolism and Development, and RNAi Insect Control

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The adoption of Bt transgenic cotton has eliminated most lepidopteran pests from this crop and has produced a secondary pest problem, with piercing-sucking insects such as the tarnished plant bug, Lygus lineolaris (Palisot de Beauvois). The future of cotton genetic pest management is threatened by these insects and their development of resistance to chemical insecticides. L. lineolaris is also a pest of more than 100 other crops. The development of transcriptome data for this insect should be transformative in essentially all aspects of research on plant bug biology and the development of control strategies. Using 454 pyrosequencing, tarnished plant bug whole body (WB) and gut (G) transcriptomes were constructed (half plate for each). A total of 116,163,527 bases were obtained representing 262,555 WB and 229,919 G reads (SRA048217) of which 232,058 (SRS280903) and 168,069 (SRS280894) reads, respectively, were assembled into contiguous sequences (contigs). The average read length was 233.1 and 208.5 bps for WB and G, respectively. The whole body and gut reads were assembled together (WB-G) to produce the most complete transcriptome possible from our sequencing effort and resulted in 6,970 contigs with an average length of 393 bps. The gut transcriptome alone was assembled into 3,549 contigs with an average length of 349 bp. The smallest contig was 55 bp and the largest was 3466 bp, and there were 62,484 sequences that could not be assembled (singletons) among both transcriptomes. Blast2GO® was used to annotate the sequences by BLAST to entries with Gene Ontology (GO), enzyme commission (EC) and InterPro (IPR) data. We further characterized metabolic systems and transcripts associated with development, and developed a promising strategy for RNAi-based insect control.

Polyphyly of mtDNA vs. Species Identification in Western Pest Lygus: Evidence of Cryptic Species and Interspecies Hybridization or Shortcomings of Morphospecies Taxonomy

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Mitochondrial cox1 (barcode) sequences were used to assess the genetic diversity and phylogeny of four important Lygus pest species of western North America (L. hesperus, L. elisus, L. borealis, and L. keltoni). For three of the species the phylogenetic inferences assigns the barcodes to clusters that are predominantly single species. Most (95%) of *L. elisus* fall into two monophyletic clusters on the phylogenetic tree. Similarly 95% of *L. borealis* are in a single cluster. The pattern for *L. hesperus* is slightly more complex, with 94% of the samples distributed in three clusters, two predominant closely related clusters and a third more divergent smaller one containing insects from the Pacific Northwest. The remaining ~5% of individuals identified as one of these three species are scattered somewhat randomly on the tree. Insects identified as L. keltoni do not belong to a cluster of their own. About 2/3 of them are embedded in the most numerous L. hesperus cluster. 12% are associated with the divergent L. hesperus cluster from the northwest and another 12% are indistinguishable from *L.borealis*. *L.* keltoni also shares a haplotype with a small divergent group of L. elisus. Altogether 37/42 (88%) of *L.keltoni* share a haplotype with one of the other three species. Possible explanations for the discordance between mtDNA data and morpho-species are: 1) interspecies hybridization in regions of sympatry leading to mitochondrial introgression; and/or 2) overlapping morphological characters. Either scenario could serve to blur species identifications and obscure the potential existence of cryptic species in the Lygus complex.

Identification of *Lygus hesperus* by DNA Barcoding Reveals Insignificant Levels of Genetic Structure Among Distant and Habitat Diverse Populations

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The western tarnished plant bug Lygus hesperus is an economically important pest that belongs to a complex of morphologically similar species that makes identification problematic. The present study provides evidence for the use of DNA barcodes from populations of L. hesperus from the western United States of America for accurate identification. This study reports DNA barcodes for 134 individuals of the western tarnished plant bug from alfalfa and strawberry agricultural fields in the western United States of America. Sequence divergence estimates of <3% reveal that morphologically variable individuals presumed to be L. hesperus were accurately identified. Paired estimates of F_{st} and subsequent estimates of gene flow show that geographically distinct populations of L. hesperus are genetically similar. Therefore, our results support and reinforce the relatively recent (< 100 years) migration of the western tarnished plant bug into agricultural habitats across the western United States. This study reveals that despite wide host plant usage and phenotypically plastic morphological traits, the commonly recognized western tarnished plant bug belongs to a single species, Lygus hesperus. In addition, no significant genetic structure was found for the geographically diverse populations of western tarnished plant bug used in this study.

Polygalacturonase Gene Expression in Field Collected Lygus lineolaris

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Polygalacturonase (PG) enzymes in the salivary glands of Lygus spp. have been shown to breakdown plant pectin to allow for easier plant tissue uptake and possible oviposition site preparation. However, there is a paucity of information about their expression in geographically distinct populations as well as regulation at the transcriptional level on different host plants. Lygus lineolaris adults were collected from cotton, horseweed, and pigweed from the Delta and Hills regions of Mississippi. Quantitative real time-polymerase chain reaction (gRT-PCR) was used to measure expression levels of three PG genes (PG1, PG2, PG3). The data indicated differences between the regions, host plants, and the PG genes. L. lineolaris collected from cotton in the Delta expressed PG1, PG2, and PG3 lower than those collected from pigweed or horseweed. L. lineolaris collected from cotton and horseweed in the Delta also expressed lower levels of PG3 than PG2, while those collected from pigweed expressed higher levels of PG1 than PG2. L. lineolaris collected from cotton in the Hills showed higher expression of PG1 than those collected from horseweed and pigweed and those collected from horseweed expressed lower levels of PG3 than those collected from cotton or pigweed. L. lineolaris from cotton in the Hills expressed PG1 higher than PG2 or PG3 on and those collected from horseweed and pigweed expressed PG3 lower than PG1 or PG2. Taken together, this data indicates that some genetic differences in PG expression exists between the two populations (Delta and Hills) of L. lineolaris in Mississippi and is its regulation is also affected by host plant.

When a Receptor Isn't What it Looks Like

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The western tarnished plant bug, Lygus hesperus Knight, is a pest of many traditional and emergent crops in the western US. Despite its economic importance, our knowledge of the molecular mechanisms underlying critical *L. hesperus* behaviors is limited, in particular those that govern female reproductive behavior. Mating has been shown to trigger a behavioral switch that decreases female receptivity but enhances oviposition rate. A similar switch has been described in Drosophila melanogaster that is regulated by an accessory gland-derived peptide, termed sex peptide, and its cognate receptor (sex peptide receptor; SPR), which is highly conserved across insect species. We consequently examined the potential role sex peptide may have in mediating the L. hesperus female behavioral switch. Similar to Drosophila, the Lygus factor localizes to the accessory gland, is heat labile, and is soluble. Characterization of a potential receptor using homology-based PCR methods identified a transcript encoding a G protein-coupled receptor with high sequence similarity (62% identical) to the Drosophila SPR. Initial end-point PCR indicated that L. hesperus SPR (LhSPR) was present in female reproductive tissues. Furthermore, in vitro analyses using a fluorescent LhSPR-Venus chimera confirmed cell surface localization and activation by accessory gland homogenates. A red fluorescent analog of the Drosophila sex peptide, however, failed to bind to either LhSPR-Venus or LhSPR. More extensive analysis of LhSPR expression via quantitative PCR revealed that it is most highly expressed in the hindgut and Malpighian tubules of both sexes. In addition, injections of synthetic *Drosophila* sex peptide had no effect on female receptivity. Taken together, our data suggest that, despite high sequence similarity to the Drosophila SPR, the endogenous ligand for the Lygus "sex peptide receptor" is different than the Drosophila sex peptide, and that the primary function of LhSPR is likely related to smooth muscle contraction.

Characterization of Aquaporins from the Western Tarnished Plant Bug, Lygus hesperus and the Whitefly, Bemisia tabaci

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Sap-feeding hemipterans, including aphids, whiteflies, plant bugs, and stink bugs are major arthropod pests of agriculture, damaging crops by direct feeding or in some cases, indirectly by transmission of plant diseases. The western tarnished plant bug, Lygus hesperus, and the whitefly, Bemisia tabaci, are phytophagous hemipterans with highly adapted piercing-sucking mouthparts for specialized feeding. Whereas L. hesperus pre-orally digests plant tissue after injection of saliva, whiteflies use modified mouthparts or stylets to penetrate and extract phloem from sieve elements. Furthermore, their alimentary tracts differ, with *B. tabaci* using a highly evolved filter chamber to maintain osmotic potential and remove excess dietary fluid. Aquaporins represent a large family of proteins known to facilitate the transfer of water across cell membranes for essential physiological processes in most living organisms. Numerous aquaporins exist in arthropods, with diverse functions, such as water (and sometimes other small solutes) transport, cryoprotection, anhydrobiosis, and ion channel function in nervous tissue. Water-specific aquaporins play important roles in insect digestion, excretion, as well as stress responses under extreme environmental conditions. Previous reports indicate the presence of five to seven distinct aquaporins from insects with completed genomes. Here, we mined *L. hesperus* and *B. tabaci* transcriptomes for aguaporin-like sequences, uncovering five and twelve putative aguaporin gene transcripts, respectively. Two aquaporins, L. hesperus aquaporin 1 (LhAQP1) and B. tabaci aquaporin 1 (BtAQP1) belong to the insect DRIP subfamily and function as water-specific channel proteins when expressed in an Xenopus oocyte expression system. Both aquaporins appear to play important roles within alimentary and/or excretory systems. Furthermore, using the *Xenopus* system, we identified a novel natural plant extract containing both blocking and potentiating activity against LhAQP1. Development of chemical agents that adversely affect insect aquaporin function, particularly in pests requiring high maintenance of water osmotic balance, could provide novel pest management opportunities.

Insecticide Resistance Status and Mechanisms in Field Populations of the Tarnished Plant Bug *Lygus lineolaris*

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Control of the tarnished plant bug mainly relies on chemical insecticides. High selection pressure in cotton system prompted resistance development in field populations to many different insecticide classes. During the past a few years, we conducted extensive monitoring of insecticide resistance in field populations in the Delta regions of Mississippi, Arkansas, and Louisiana using spraying dose-response assay and major detoxification enzyme activity assays. Bioassay results indicated that different populations showed significantly different survival rates (10-100%) to both organophosphate and neonicotinoid insecticides. High resistance levels are usually associated with high spray intensity, particularly in continuing cotton cropping system. To better understand resistance and explore ways to monitor resistance in field populations, we examined acephate susceptibility and the activities of two major detoxification enzymes in nine field populations. Regression analysis of LC₅₀s with kinetic esterase activities revealed a significant polynomial guadratic relationship with R^2 up to 0.89. Glutathione S-transferases also had elevated activity in most populations, but the variations of GST activities were not significantly correlated with changes of acephate susceptibility. Microarray analysis of 6,688 genes revealed 329 up- and 333 down-regulated genes in an acephate-resistant strain. Six esterase, three P450, and one glutathione S-transferase genes were significantly up-regulated. All vitellogenin and eggshell protein genes and most protease genes were significantly down-regulated in the resistant strain. The large portion of metabolic or catalysis genes with significant upregulations indicated a substantial increase of metabolic detoxification in the resistant strain. Significant increase of acephate resistance, increases of esterase activities and gene expressions, and variable esterase sequences consistently demonstrated a major esterase-mediated resistance development, which was functionally provable by abolishing the resistance with esterase inhibitors. In addition, significant elevation of P450 gene expression and reduced susceptibility to imidacloprid in acephate-resistant strain indicated a concurrent resistance risk that may impact other classes of insecticides.

Development of Sulfoxaflor for Control of Lygus spp. and Related Mirids

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Sulfoxaflor is the first member of a new chemical class of insecticides, the sulfoximines, and was discovered by Dow AgroSciences. It is currently in global development for control of a broad spectrum of sap feeding pests on many different crops. In addition to outstanding *Lygus* spp. activity, sulfoxaflor has demonstrated excellent control of many different species of aphids, planthoppers, whiteflies, mealybugs and scales. Initial registrations were obtained in the first half of 2012 in several countries, with a US registration decision anticipated for 2012. Global trade names include TransformTM WG and Closer TM SC.

Sulfoxaflor has been broadly characterized for control of *Lygus* bugs, particularly in cotton. In the US, it demonstrates commercial levels of efficacy at 50-75 g ai/ha, against both *L. lineolaris* and *L. hesperus*. In China, rates of 38-75 g ai/ha have provided control of *L. lucorum* in cotton. Although *Lygus* spp. are not cotton pests in Australia, the closely related mirid, *Creontiades dilutus*, is a key pest and similar rates of sulfoxaflor provide excellent control. Sulfoxaflor has also demonstrated efficacy against *Lygus* spp. in alfalfa, vegetable and strawberry crops. In addition to reduction of *Lygus* spp. populations, strong positive yield responses have been observed in many research trials, and no outbreaks of secondary pests, such as mites, have been observed in sulfoxaflor-treated crops.

Response of *Lygus lineolaris* to sulfoxaflor and neonicotinoids in laboratory assays

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The tarnished plant bug, Lygus lineolaris (Palisot de Beauvois), has become a major pest of cotton, Gossypium hirsutum (L.), within the Mid-Southern United States over the last several years. Lygus lineolaris has become the target of more insecticide applications than any other insect pest of cotton in the Mid-South with some growers making up to 15 foliar insecticide applications for Lygus control. A limited number of insecticides are available for Lygus management representing the organophosphate, carbamate, neonicotinoid, pyridine carboxamide, benzylphenol urea, and pyrethroid classes of chemistry. Also, the level of activity against L. lineolaris varies among products. In addition, L. lineolaris is becoming resistant to many of the products currently available for control. Populations of *L. lineolaris* exhibiting varying levels of tolerance/resistance to some organophosphates, pyrethroids, cyclodienes, and carbamates have been reported in the Mid-South. Sulfoxaflor (Transform[™] WDG) is a new insecticide for L. lineolaris management that has been shown to perform equal to or better than currently available products. Sulfoxaflor is considered to be in a different class than other insecticides and will be an additional tool to help manage L. lineolaris infestations. Laboratory assays to determine the baseline response of L. lineolaris to sulfoxaflor were initiated during 2010. Also, monitoring the response of L. lineolaris populations from 20 to 25 locations in Arkansas, Louisiana, and Mississippi to the neonicotinoids, imidacloprid and thiamethoxam, was conducted during 2007 to 2012 using similar procedures. Feeding assays using formulated product mixed at different concentrations in 10% honey-water solution were conducted using wild L. lineolaris adults. For imidacloprid and thiamethoxam monitoring, a L. lineolaris population collected from a non agricultural area in Arkansas was used as a reference. The mean LC₅₀ values for imidacloprid ranged from 4.24 to 5.22 µg/ml during 2007 to 2011. Mean resistance ratios ranged from 2.49 to 3.07. The mean LC_{50} values for thiamethoxam ranged from 2.02 to 4.42 µg/ml during 2007 to 2012. Mean resistance ratios ranged from 0.67 to 1.47. For sulfoxaflor 20 collections from Arkansas, Louisiana, Mississippi, and Tennessee were tested during 2010 to 2012. The LC₅₀ values ranged from 0.2 to 5.74 µg/ml.

Discovery and Optimization of Hemipteran-active Proteins for Transgenic Plant Applications

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Following the eradication of the Boll Weevil, and adoption of *Bt* technologies for lepidopteran control, Lygus has become the #1 pest for many cotton producers in the US. With the increased cost to control Lygus, there is increasing support to develop biotechnology solutions to control this pest. Our discovery platform has identified numerous Lygus-active proteins from *Bacillus thuringiensis*, and optimization efforts have increased activity of Lygus-active proteins against both *Lygus hesperus* and *Lygus lineolaris* species. We have demonstrated that transgenic cotton plants, expressing these optimized proteins, cause increased mortality and stunting of Lygus nymphs, and show excellent potential for improved yield performance in the field. These results provide a basis for the development of cotton varieties protected from Lygus, and the potential exists that such novel traits could be extended within the Bt crop protection paradigm to other hemipteran insect species such as Stink bugs.

In-planta Efficacy Evaluation of Lygus-active Bt Proteins

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Lygus spp have become number one insect pests of cotton in the USA and management is becoming more difficult as these insects have developed resistance to many insecticides. Therefore, there is a need for alternative technology for managing Lygus spp. Here we report results from in planta efficacy evaluation of Lygus-active Bt proteins for development of alternative options for *Lygus* control. Transgenic events expressing different Bt proteins were tested in growth chamber caging, field caging and open field efficacy trials. Caging trials rely on rearing Lygus nymphs to adult stage on plant material and then releasing two pairs of each sex on plants that are enclosed by a cloth cage. The adults are allowed to mate and reproduce and data on next generation Lygus is collected after >3 weeks. More than 7-fold reductions in second generation L. lineolaris and L. hesperus numbers were recorded between the best transgenic events and the negative control at multiple field sites in caging efficacy trials. Open field efficacy trials were conducted at multiple locations which relied on natural insect pressure and data on insect numbers and square retention were recorded. In this trial, significantly fewer L. lineolaris and increased square retention were recorded in transgenic events compared to negative control.

Predicting Effects of Local and Regional Factors on Population Dynamics of Lygus hesperus

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We evaluated how local and landscape characteristics affect population density of the western tarnished plant bug, Lygus hesperus (Knight), in cotton fields of the San Joaquin Valley in California. During two periods covering the main window of cotton vulnerability to Lygus attack over three years, we examined the associations between abundance of six common Lygus crops and uncultivated habitats surrounding cotton fields and *Lygus* population density in these cotton fields. We also investigated impacts of insecticide applications in cotton fields and cotton flowering date. Consistent associations observed across periods and years involved abundances of cotton and uncultivated habitats that were negatively associated with Lygus density, and abundance of seed alfalfa and cotton flowering date that were positively associated with Lygus density. Safflower and forage alfalfa had variable effects, possibly reflecting among-year variation in crop management practices, and tomato, sugar beet and insecticide applications were rarely associated with *Lygus* density. Using data from the first two years, a multiple regression model including the four consistent factors successfully predicted *Lyqus* density across cotton fields in the last year of the study. Our findings indicate that manipulation of the spatial arrangement of a few habitats with consistent effects and of cotton planting date has potential for managing *L. hesperus* populations in cotton.

Using Spatial Metrics to Assess Crop Stressed by Pests: Case of *Diuraphis noxia in* Wheat Fields

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A variety of spatial pattern metrics can be generated from multispectral imagery data. These spatial metrics can be used to characterize stress induced by pests in field crops. This presentation is an ongoing study that has been conducted in wheat fields. The study explores spatial metrics to differentiate stress induced by *Diuraphis noxia* from other stress factors. We project that this approach can be applied to survey the impact of Lygus bugs and other mirid pests in cotton fields, and improve cotton fields' management.

Application of FRAGSTATS Metrics to Categorical Imagery Products Used for Tarnished Plant Bug Sampling and Management

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An algorithm that fuses a digital elevation model of topographical relief and a vegetative index, such as the Normalized Difference Vegetation Index (NDVI), has been developed for building maps of crop variability. The key features of this algorithm are briefly presented. In cotton pest management for the Tarnished Plant Bug (Lygus lineolaris), the fused map produced by this process for these two remote sensing layers, is found useful for selection of sample sites and development of variable-rate pesticide applications. Considering that such maps indicate different spatial patterns for different fields and within and among different production seasons, it is necessary to be able to quantify the information of these types of imagery products over different acquisition dates, locations, and crop rotations in order to build additional algorithms that seek to describe landscape community structure derived from remote sensing techniques for wider applicability to understanding Tarnished Plant Bug population ecology. As a first step in the development of such a kind of processing capacity, an investigation of FRAGSTAT metrics of fused categorical image data products is proposed as a valuable and important collateral image processing technique. Illustration of how FRAGSTATS is applied to imagery products and what metrics can be obtained from them is described.

Successful Lygus Management as a Stabilizing Element of Cotton IPM in Arizona

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Advances in selective management of lepidopteran and homopteran pests over the last two decades have created new opportunities in IPM of arthropod complexes. These advances involve introduction of new technologies including transgenics, selective seed treatments, reduced-risk biorationals and even pest eradication; however, the success of these technologies has often hinged on the improved role of conservation biological control and the detailed knowledge to properly incorporate all of these elements into working IPM. What remain as key pests and key disruptors of IPM are often hemipterans (Pentatomidae, Miridae, Plataspidae, Lygaeidae / Oxycarenidae) in many cropping systems including cotton, which is specifically challenged by *Lygus spp*. This presentation will detail the elements of successful Lygus management in the cotton system with descriptions of measured outcomes and long-term impacts that have helped stabilize the overall cotton arthropod IPM program in Arizona.

Western Tarnished Plant Bug, *Lygus hesperus* Knight, Management in San Joaquin Valley Cotton: Trends and Implications

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The western tarnished plant bug, Lygus hesperus Knight, continues to be a key arthropod pest in cotton systems in the west. Yield losses and control costs affect profitability but Lygus management decisions also affect populations of spider mites and cotton aphids in San Joaquin Valley cotton. Alfalfa and safflower are important sources for lyous that potentially move and damage other crops. Chemical control is key to minimizing damage from lygus bugs as other forms of management are not fully developed. Because of insecticide resistance concerns, it is important to utilize active ingredients from several chemical classes for lygus management. Organophosphates and carbamates were the standard for lygus bug control in cotton in the 1990's; presently Vydate® is the only carbamate still used in SJV cotton. Pyrethroid use increased in the early-mid 1990's and continued as a primary tool through mid-2000's. The development of resistance has reduced the efficacy of pyrethroids. Clothianidin (Belay®), available in 2011, provides a higher level of control than other neonicotinoids. Since ~2007, flonicamid (Carbine®) has been the standard for lygus management in SJV cotton. Studies comparing efficacy of registered and experimental insecticides, conducted from 2009 to 2012, were conducted. Lygus nymphs and adults were quantified in these studies, as well as numbers of natural enemies. Secondary pests (cotton aphids and spider mites) were assessed for the potential of treatments to flare populations. Fruit retention and seed cotton yield were recorded. The second aspect of this project was to monitor susceptibility of lygus bugs to representatives of organophosphate, carbamate, and pyrethroid chemistry. These bioassays were done from collections made in June and in August (2008 to 2012) from three locations in the SJV; the surface residue (inside surface of small plastic bags) method was used. Bioassays with flonicamid, utilizing a floral foam method adapted from studies on tarnished plant bug, were conducted for *L. hesperus*.

Field-Scale Movement of Lygus Bugs in Arizona Cotton

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Lygus bugs are the major insect pest of cotton in Arizona with potential to affect yield quantity and quality. Inter-field movements of *Lygus* bugs over large areas have been examined in the western U.S. (Carriere et al. 2006, 2012). They identified "source" hosts like alfalfa and weeds that readily supply Lygus to "sink" cotton fields. These immigrating adults impact cotton by direct feeding and by initiating the growth of nymphal populations. Their feeding result in abortion of young squares and can interfere with normal fertilization of the flower, leading to poorly formed, undersized or light bolls. However, very little is known about smaller scale *Lygus* movement within cotton fields or precisely how they colonize and develop once arriving in individual cotton fields. Research has shown that egg white, cow milk, or soy proteins can be sprayed in the field using conventional spray equipment to mark insects. In turn, protein-marked insects can be tracked to measure their movements in the field. The assays used to detect a protein mark on an insect have been optimized for mass screening and fieldscale studies. By spraying different cotton areas within one field, we can follow the fieldscale movement of Lygus in cotton. Understanding within-field movement of Lygus in cotton may help us improve our research approach for determining "hot spot" areas in the field where *Lygus* populations may be concentrated and causing greater damage. We may be able to then target different tactics of Lygus management based on their movement in the field, e.g., targeted sampling efforts in expected hot spots or apply controls only to these "hot-spot" areas of the field. This approach may increase field scouting and control efficiencies, saving growers money and protecting yields in the management of this key cotton pest in Arizona.

Development of Lygus Management Strategies for Texas Cotton

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Lygus hesperus (Knight) (Hemiptera: Miridae) is an economically important pest of Texas High Plains cotton. Approximately 1,566 metric tons of cotton is lost annually due to Lygus infestations in our region. Lygus are typically a mid- to late season pest of Texas cotton; therefore, management practices are mostly focused toward the mid- to late cotton growing season. Lygus management activities are less aggressive in this region because Texas High Plains infestations in cotton are generally low as compared to other U.S. cotton production regions. Lygus management decisions have typically been formulated based upon information from individual farms, but when Lygus become an area-wide pest, larger-scale regional pest management strategies would likely be better suited for long term Lygus management. Essential components for successful pest management programs include strong research and extension components including efficient IPM tools, a reliable pest forecasting system, efficient communication network, resource management plan, and periodic monitoring and evaluation system. Our program has focused on the development of knowledge-based resources for ecologically intensive Lygus management programs. Examples of specific studies in our Program include: identifying host sequences, population movement, overwintering biology, interaction of *Lygus* populations within landscape level habitat structure, *Lygus* genetic diversity, assisting in the development of Lygus resistant cotton cultivars, monitoring pesticide resistance, and evaluating relevant pesticide chemistries. Review of Lygus literature revealed most past studies focused on its biology, ecology and chemical control. Although the published Lygus information is very extensive, it has not been consolidated for easier access and best utilization. Future Lygus research efforts need to focus on consolidation of the published information, development of a population forecasting system and resistant cotton cultivars, promotion of other pest control strategies such as enhancement of natural control services. Multi-state collaborative efforts of sharing knowledge and resources are needed in order to develop successful Lygus management programs.

The Lygus Simulation Training Environment, An interactive Workshop & Discussion

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Landscape ecology is an area of rapidly expanding investigation with the potential for improving the management of spatial assets in agroecosystems. New understanding, however, must be translated into pragmatic working knowledge, accepted and applied by growers and pest managers. This is especially relevant when new skills of communication, cooperation and collaboration are necessary to encourage (behavioral) change. Crop selection and placement within a grower's holdings represents a major area for ecological engineering so as to minimize pest risks and maximize ecosystem services. However, larger individual benefits are possible with cooperation and coordination of members of the larger community. We have developed a platform for simulating risks of damage to cotton by *Lygus hesperus* based on source-sink relationships empirically measured and inferred from large scale studies in CA, AZ and TX. This workshop will include a discussion of the project and a working demonstration of a grower training with audience participation in this multi-user gaming environment. We will also cover the use of serious games as teaching tools and for evaluating outcomes. This approach has potential applications to any spatial phenomena where growers need to perceive and understand risks as a function of the spatial ecology of their crop and non-crop assets (e.g., pest resistance to insecticides; recruitment of pollinators and other beneficials; strategic placement of structured refugia for resistance management, off-site movement of pollutants, etc.).

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