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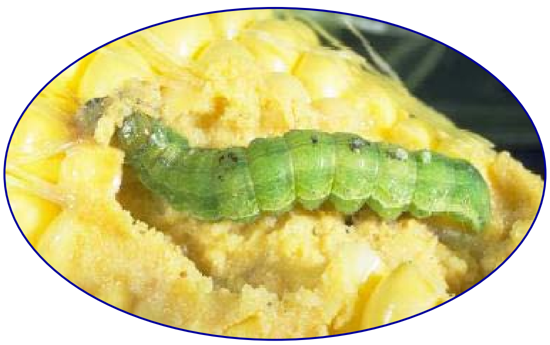

Southern Insect Management Research Unit

Stoneville, Ms

Annual Report on Progress (2010) and Plans (2011)

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Mission Statement

Southern Insect Management Research Unit
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MISSION

To develop sustainable, cost efficient, environmentally safe pest control methods for U.S. southern row crops. Current efforts include: (1) improving integrated pest management systems for pests of cotton, corn, soybean, and sweetpotato; (2) discovering new entomopathogens and establishing fundamental principles for their use in row crop pest control; and (3) gaining new knowledge that impacts management decisions and policy related to transgenic crop risk assessment and resistance management for Bt-cotton's target pests. Specifically:

(*1) Identify and characterize new sources of maize, soybean, and sweetpotato resistance to insects, e.g. corn earworm, stink bug, and sugarcane beetle.*

(2) Develop and enhance biological control strategies for suppression of plant bugs and stink bugs in area-wide pest control systems for southern row crops.

(3) Improve and continue to evaluate an area-wide pest management system for control of tarnished plant bugs in cotton.

(4)Comparison of tarnished plant bug and stink bug injury to cotton and comparison of various sampling methods to predict yield losses from these insects in flowering cotton.

(4) Entomopathogenic fungi will be developed for controlling tarnished plant bug populations developing on wild host plants.

(5) Sterile insect technique will be developed targeting these same tarnished plant bug populations.

(6) Epizootics of the cotton aphid fungus, Neozygites fresenii will be augmented to enhance cotton aphid control.

(7) Conduct fundamental research to generate baseline data on population genetic parameters to facilitate research on mechanisms of resistance to Bt proteins at the molecular level in Lepidopteran pests of cotton.

(8) Characterize the injury potential of Lepidopterous pests (esp. Helicoverpa zea) to new transgenic cottons to develop accurate action thresholds.

(9) Determine the effects that Bt corn (Yieldgard®) has on bollworm biology including: pupal diapause, reproduction, flight potential, physiology, and the ability of progeny to survive and damage transgenic Bt cottons (Bollgard® and Bollgard II®).

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Disclaimer and Purpose of Report

This report summarizes progress made on research objectives for 2010 and plans for research activities in 2011.

Many of the results are preliminary and others are being released through established channels. Therefore, this report is not intended for publication and should not be referred to in literature citations.

Intent of this report is to give the reader an overview of the Southern Insect Management Research Unit (SIMRU) activities. The activities (progress and plans) address the research unit mission. Formal annual reports of research progress as submitted to the CRIS system are included in the summary.

Overall Summary and Perspective of 2010 SIMRU Activities

The past year was a period of transition for the Southern Insect Management Research Unit. The Unit recruited and employed a new Research Leader; finalized research on three five-year projects; successfully proposed and finalized three new major research projects for appropriated funds; and developed position announcements and recruitment plans for two additional Category 1 scientists. During this transition, the five Category I scientists provided leadership and administrative support well beyond the expectations of their research positions. Dr. Gordon Snodgrass is recognized for his leadership as Acting Research Leader. Drs. Jackson, Perera and Snodgrass provided exceptional leadership in finalizing the previous research projects and organizing plans for three new projects. The Unit continued to make research progress and provide meaningful information on biological control of tarnished plant bug, insect resistance to chemical insecticides and Bt toxins, and biology, ecology and management of major pests of cotton, corn, soybean and sweetpotato. The cooperative spirit and team atmosphere of SIMRU is a great strength from the perspective of the new Research Leader. SIMRU has a long and proud history of service to the agricultural systems of the Mississippi Delta, and to creative management systems for suppressing insect pests across the region. In addition to the Category I scientists, the Unit received significant contributions from two Category II scientists and two Category III scientists. The technical support staff and the administrative team are strong and mature, and the Unit enjoys strong collaborations with university partners across the South, especially Alcorn State University and Mississippi State University within the state. The ingredients for a nationally recognized research effort on applied entomology will be in place with successful recruitment and hiring of two additional Category I scientists in 2011. This will be fueled by a careful evaluation of the past and a dialogue of challenging and relevant research problems for the future.

Randy Luttrell
Research Leader

Research Accomplishments

Summary Accomplishments -- Internal Research Programs

Adams Research Program

In 2010, we completed a study evaluating ten insecticides that were incorporated into the row bed prior to planting. Also, the application method (pre-plant incorporation versus transplant water application) of five insecticides was evaluated to determine whether various classes of insecticide performed differently based on application method. Insect damage, yield and quality of the sweetpotatoes from each treatment was recorded and analyzed. **(L. C. Adams, R. Jackson, C. Johnson)**

During the 2010 growing season, we compared four products to suppress the reniform nematode in Mississippi Delta sweetpotato fields. Soil samples were taken twice during the season to asset reniform populations. Yield and quality of the sweetpotatoes from each treatment was recorded and analyzed. **(L. C. Adams, R. G. Luttrell, C. Johnson)**

Since the 2006 growing season, our group has participated, in cooperation with researchers from Louisiana State University, North Carolina State University and Mississippi State University, in the National Sweetpotato Collaborators Group variety trials. During 2010 SIMRU submitted yield and quality results from five check lines and three numbered research varieties to the NSCG Annual Report. **(L. C. Adams, R. G. Lutrell, C. Johnson)**

For the second year, we evaluated Monty's Plant Food foliar feed products for sweetpotatoes, cotton and soybeans grown in the Mississippi Delta. Monty's Plant Food products 8-16-8, 2-15-15 and Liquid Carbon were applied during the seasonal growth stages of each crop to evaluate yield. Increased yield was observed with some treatments. **(L. C. Adams, C. Johnson)**

We have trapped, charted, summarized and reported results of insect pheromone trapping of *H. zea* (18^{th} year) and *H. virescens* (19^{th} year). Long-term trapping of these pests provides valuable information on the potential influence of major technological developments (i.e. Bt cotton) on overall population dynamics of these pests in the Mississippi Delta. **(L. C. Adams, C. Johnson)**

Allen Research Program

A survey of looper species present in soybean fields in Mississippi and Arkansas was conducted. Soybean fields were sampled with a sweep net on a weekly basis in four counties of Mississippi and Arkansas. Only two species of loopers was collected and successfully reared to adults during 2010. Soybean looper and the gray looper or mint looper, which has seldom been recorded on soybean plants, were collected. The gray looper was the dominant species during May and June, whereas soybean loopers comprised the majority of the population during July and August. The importance of indentifying the caterpillar species before insecticide application was verified in a laboratory insecticide study. The gray loopers were found to be approximately 25-fold more susceptible to pyrethroid insecticides compared to a population of soybean loopers. **(C. Allen and A. Walters).**

The potential of using pollen as a natural marker for studying the movement of tarnished plant bugs was examined. Field populations of tarnished plant bugs were collected from a known host plant and then frozen after collection. Additionally, a laboratory colony was allowed to feed on pigweed plants for 3 days and then removed from the plants. The colony of plant bugs was frozen at different intervals from 0 – 24 hours after feeding. Individual plant bugs were placed in a chemical solution that dissolves the insect, but does not destroy any pollen grains. Pigweed pollen was recovered from plant bugs up to 24 hours after removal from the pigweed plants. (**C. Allen, G. Wilson and A. Walters).**

Laboratory experiments were conducted to examine the susceptibility of the nymphal stages and adult stage of the tarnished plant bug to a pyrethroid (permethrin), organophosphate (methamidophos), and a neonicotinoid (thiamethoxam) insecticide. The susceptibilities of 5th instar and adult stages of the green, southern green, and redbanded stink bugs were examined using the pyrethroid, bifenthrin, and organophosphate, acephate. For tarnished plant bugs, estimated LC_{50}'s of the 4th and 5th instar nymphs to permethrin and methamidophos were 1.2 to 3.3 times higher than that of adults for the insecticides examined, while the first through third instar nymphs were less than that of adults. There was no real difference in the response of 3rd through 5th instar tarnished plant bug nymphs and adults to thiamethoxam. The LC_{50}'s of the 5th instars of the various stink bug species were 1.6 to 10.5 times as high as that of adults depending on stink bug species and insecticide tested. **(C. Allen, F. Musser and G. Snodgrass).**

Jackson Research Program

An analytical technique was used to evaluate infestations of tarnished plant bug (TPB) adults in Mississippi Delta cotton fields during June, July, and August in an effort to determine the proportion of those adults that were immigrants into these fields. The stable carbon isotope technique can distinguish whether a TPB adult developed as a nymph on a C_3 versus a C_4 plant host. The two primary C_4 hosts of TPB during this period of the growing season are corn and various pigweed species. C_3 hosts are more numerous, but during this period, cotton is the predominant C_3 host. Two years of research show that during late June and early July, 70-90% of tarnished plant bug adults collected from cotton fields developed as nymphs outside of cotton. By mid-July, 50% of TPB adults collected in cotton fields were immigrants, whereas virtually no TPB adults were immigrating to cotton fields by early-to-mid August. These findings indicate that TPB scouting in cotton should be intensified during late June and early July so that control measures can be more precisely timed in order to reduce the amount of TPB reproduction in cotton. This practice should have the greatest impact on populations found in cotton during mid-July through Mid-August. **(R. Jackson, G. Snodgrass, C. Allen)**.

Pyrethroid resistance in bollworm populations has been monitored within a 9-state region from Texas to Virginia during the last 4 years using adult vial tests. During this time, pyrethroid resistance in bollworms has not changed significantly. However, the proportion of moths tested that survived the 5µg dose of cypermethrin during this project was 122% higher than that previously reported for these states during 1998-2000. In addition, stable carbon isotope analysis was used to determine if there were differences between pyrethroid resistant and susceptible bollworm moths with regard to host origin. No differences in host origin were apparent between resistant and susceptible moths, but the data indicated that the bollworm generation that developed on corn in nearly all states had higher survival in the presence of cypermethrin than any other generation. These data indicate that host plant suitability of bollworm plays a role in response to pyrethroid insecticides. Although a change in resistance has not been evident during the last few years, comparison to historical data indicates a shift in response to pyrethroids. These findings should be taken into consideration when making decisions on rate selection and timing of pyrethroid applications targeting bollworms in mid-South row crops. **(R. Jackson)**.

Various colonies of bollworms were collected from plants expressing different Bt cotton and corn technologies (e.g. Bollgard II®, WideStrike®, and SmartStax®) and tested for susceptibility to Cry1Ac and Cry2Ab through diet-incorporation, dose-mortality bioassays. Resistance ratios for Cry1Ac indicated that bollworm populations collected from pyramided-gene Bt crops were 3-8X less susceptible to Cry1Ac compared to a laboratory-susceptible colony. The susceptibility of these colonies to Cry1Ac was comparable to many of those generated during 2002-2008 in Arkansas where resistance ratios ranged from about 0.1 to >500. Similar to Cry1Ac susceptibility estimates, Cry2Ab resistance ratios ranged from 2-12. Average mortality ratio for the colonies tested against Cry1Ac in 2010 was 0.8, which was comparable to a range of 0.8-1.0 from 2002-2008 in Arkansas. These data suggest that Cry1Ac-susceptibility has remained unchanged over this period of time. However, the mortality ratio for Cry2Ab in 2010 was 0.4, which was lower than the range of 0.6-1.0 from 2002-2008 in Arkansas. These data indicate that if a reduction in susceptibility is being observed, it is likely due to reduced susceptibility to the Cry2Ab protein. This, however, would not explain increased survival of bollworms on WideStrike cotton varieties, as these produce the Cry1Ac + Cry1F Bt proteins. Although field observations and limited laboratory data point to the potential for reduced Bt susceptibility in bollworm populations, there is also another factor that can affect the numbers of surviving larvae observed in Bt crops. In studying pheromone trap captures of bollworm adults in Mississippi from 2006-2010, bollworm moth numbers at their peak in 2010 were 2x higher than those for the previous 4 years. In addition, the seasonal average number of bollworm moths captured per trap per week was <50 for 2006-2009. Bollworm adult populations in 2010 averaged >100 moths per trap per week. Thus, higher populations going into systems with selection generated higher numbers of survivors coming out of these systems. Based on this information, unusual survival of bollworms in pyramided-gene Bt crops in 2010 was primarily a function of high bollworm population numbers. **(R. Jackson, R. Luttrell)**.

Luttrell Research Program

Variations in bollworm (*Helicoverpa zea*) and tobacco budworm (*Heliothis virescens*) responses to Bt toxins expressed in transgenic cottons (Cry1Ac, Cry2Ab2, Cry1F and Vip3A) were measured and related to crop, ecological and seasonal traits of field populations collected for the laboratory assays. Most of this work was done at the University of Arkansas from 2002-2009 through Cooperative Agreements with the USDA ARS Southern Insect Management Research Unit. Efforts were continued in 2010 to summarize and publish important benchmark, monitoring and ecological descriptions of this work. (**R. Luttrell, Ibrahim Ali, K. Dixon).**

Mississippi populations of bollworm and tobacco budworm were collected and assayed for resistance (or reduced susceptibility) to Cry1Ac and Cry2Ab2. Cohorts of the colonies were also exposed to cotton leaf tissue from conventional, Bollgard, Bollgard II and Widestrike cottons. Most colonies showed elevated LC50s, but responses were within the range of those previously measured at the University of Arkansas. A colony collected from Bollgard II cotton by J. Gore and a colony originating from a single larvae found surviving on Bollgard II cotton by P. McKibben and reared by J. Willers had less than 50% mortality at pupation when fed Bollgard cotton for the entire larval period. These same colonies had 99 and 80% mortality at pupation on Bollgard II. Previous benchmark studies at the University of Arkansas recorded 94 to 100% mortality at pupation on Bollgard and greater than 98% mortality on Bollgard II cotton. (**R. Luttrell, K. Dixon, J. Gore, R. Jackson, J. Willers**).

Perera Research Program

Transcriptome of tobacco budworm (TBW), *Heliothis virescens*, was assembled and bioinformatic analysis was completed in collaboration with Dr. Kent Shelby, USDA-ARS, Columbia, MO, Dr. Juan Luis Jurat Fuentes, University of Tennessee, Knoxville, TN, Dr. Fred Gould, North Carolina State University, Raleigh, NC, and Dr. Michael Adang, University of Georgia, Athens, GA. The assembly contained over 2 billion nucleotides contributed by the collaborators. The aim of this project was to identify the complete expressed gene repertoire of the TBW for use in various studies, including evaluation of response to Bt toxins and development of genetic markers for mapping and population genetic studies. Microarrays developed using the transcriptome sequences were used to study physiological response in TBW larvae from resistant and susceptible strains challenged with various Bt toxins. Using the results of the microarrays and other studies, modulation of a membrane-bound alkaline phosphatase gene in resistant strains of TBW, bollworms (*Helicoverpa zea and H. armigera*), and fall armyworm (*Spodoptera frugiperda*) was identified and its use as a potential biomarker for resistance was proposed. In addition, several other genes modulated in response to intoxication were identified by microarray data analysis and are under investigation. **(O. P. Perera).**

High-throughput sequencing of the transcriptome of the tarnished plant bug (TPB) was carried out using Roche 454-Titanium and Illumina Genetic Analyzer II platforms. RNA extracted from adults and nymphs of TPB collected from several field locations in Mississippi, Arkansas, and Louisiana were pooled to increase the detection of polymorphisms in the sequences. Insects were also treated with sub-lethal doses of several insecticides to induce genes that respond to intoxication. The assembly of the transcriptome resulted in 52,054 sequences longer than 80 nucleotides. Over 220 sequences belonging to the three major classes of detoxification (ie. Cytochrome p450 monooxygenases, esterases, and glutathion-s transferases) were identified and submitted to GenBank database. Microarrays were developed to profile physiological response in TPB to major classes of insecticides. **(O. P. Perera).**

A picorna-like virus infecting TPB was identified and characterized. Genome organization of this virus resembles those of the genus *Iflavirus* (Picornavirales:Iflaviridae), that also includes that resembles sacbrood virus of the honey bee and the infectious flacherie virus of silkworm. Structural proteins of the viral capsid, the cystiene protease, helicase, and RNA-dependent RNA polymerase (RdRp) were identified using database searches and homology based modeling using solved crystal structures of similar viruses. Preliminary surveys of TPB populations from the

Delta region indicates low incidence of this virus in natural populations ranging from 0-8%. **(O. P. Perera).**

Portilla Research Program

We maintained *L. hesperus* and *L. lineolaris* colonies for consecutive generations in order to continue with the diet modification research and fitness parameter evaluation for both species. The use of this diet will facilitate the rearing of these insects within the bio-control industry. **(M. Portilla)**.

Bioassays were conducted in order to construct life tables and to calculate demographic parameters for 16 iso-families of fall armyworm. Using a novel method, I selected the more resistant colonies to evaluate the fitness parameters using traditional and transgenic corn and cotton varieties. A series of Bt resistance experiments was conducted under greenhouse and field conditions. These investigations also led me to develop a preliminary Bt foliage-based diet formulation to assay Bt-resistant colonies of FAW and other Bt-resistant lepidopterans. **(M. Portilla)**.

I concluded the identification of polygyne and monogyne fire ants colonies in Southwest Mississippi. **(M. Portilla)**.

Snodgrass Research Program

Tarnished plant bug populations were tested for resistance to imidacloprid during May at 6 locations in southeastern AR and 15 locations in the MS Delta. Results found no unusually high LC_{50} values in any of the populations. **(G. Snodgrass)**.

Plant bug populations were tested for resistance to thiamethoxam (Centric) during June at 4, 6, and 20 locations in LA, AR, and the MS Delta. The average LC_{50} values were 0.93, 2.0, and 2.5 ug/vial for the LA, AR, and MS populations, respectively. The LC_{50} value for susceptible bugs from Crossett, AR in 2010 was 1.68 ug/vial. On average, little resistance to thiamethoxam was found, however, one population from Belzoni had an LC_{50} value of 11.90 the first time it was tested and a value of 13.46 when it was re-tested. Efforts to rear the population for further testing failed. The occurrence of a population like the one from Belzoni will often signal that a change in resistance in other populations could occur in the near future. **(G. Snodgrass)**.

Plant bug populations from 4, 6, and 15 locations in LA, AR, and the MS Delta were tested for resistance to pyrethroids, acephate (Orthene), and thiamethoxam during July. Any population with percent mortality less than 70% in a discriminating-dose glass-vial bioassay using permethrin had a high level of resistance to pyrethroid insecticides. Average percent mortalities in the test were 58.5, 54.3, and 49.6% for the plant bug populations from LA, AR, and MS, respectively. In testing acephate, a resistance ratio of 3.0 or higher indicated a high level of resistance. The average resistance ratios were 3.14, 3.09, and 3.4 for the populations from LA, AR, and MS, respectively. Resistance to thiamethoxam was determined by feeding a single dose (7.5 ug) to 30 individuals from each population. Previous work had found that the only population ever tested that had less than 80% mortality at this dose was the Belzoni population tested this year. If any of the populations had had less than 80% mortality they would have been tested with the full range of doses. However, all of the populations had mortalities greater than 80% and no population with an unusual level of resistance was found. To perform the resistance tests during May-July required the collection and testing of over 15,000 adults. **(G. Snodgrass)**.

A replicated field test in cotton was performed from July13 to 28 July in which the NI8 isolate of *Beauveria bassiana* (which was discovered and propagated at SIMRU) was tested for control of tarnished plant bug alone and in combination with novaluron (Diamond). Test plots were 16 rows by 50 ft, and these sized plots were found to be too small to prevent movement of adults between the plots. Adults collected from the plots after treatment and held in the laboratory were found to have infection rates as high as 65.7% after two applications of NI8 spores. This infection rate occurred at average high and low air temperatures of 94.6 and 74.3^0 F. This rate of infection compares favorably with control with conventional insecticides, but death by infection with NI8 is slower than with standard insecticides. The novaluron and NI8 treatments, alone and in combination, did not have a significant effect on beneficial arthropod populations found in the cotton plots. **(G. Snodgrass and R. Jackson)**.

Preliminary tests were conducted during October to determine if novaluron (Diamond) and the NI8 isolate of *B. Bassiana* were effective against diapausing and reproductive plant bug adults and nymphs found on pigweed, *Amaranthus* spp., and tall goldenrod, *Solidago altissima*. These test results are still being summarized but some results are available. These include the findings that novaluron and NI8 were active against plant bug adults and nymphs on the wild hosts. The species of beneficial arthropods found on the hosts were identified along with their life stage. Some of the most abundant beneficial arthropods were collected or reared and tested in the laboratory. These include the goldenrod crab spider and a species of jumping spider. **(G, Snodgrass, R. Jackson and M. Portilla).**

The efficacy of NI8 for diapausing and reproductive plant bugs was determined at 10, 11.5, 15, and 20^0 C using conidia concentrations of 1×10^4, 1×10^5, 1×10^6, and 1×10^8 per ml. Conidia did not germinate at 10^o C. At the other three temperatures the highest mortalities occurred at the highest conidia rates. Mortality occurred a little more rapidly in the reproductive adults than in those in diapauses. No differences in mortality between reproductive and diapausing adults were found. Nymphs (1-3 instar) were also tested with the four rates of conidia and held at 25^o C. Mortality among nymphs was lower than that obtained with adults. In a few cases, cast skins were found 24 h after treatment (which showed that the nymphs had molted). These nymphs later died and sporulated which suggested that NI8 could infect nymphs entering ecdysis. Statistical analyses of these laboratory tests have not yet been conducted. **(G. Snodgrass and G. Gipson)**.

A laboratory test was conducted that looked at germination of NI8 conidia in a novaluron + Silwet (surfactant) water solution as compared to conidia in a water + Silwet solution. No difference in germination of the conidia in the two solutions was found. **(G. Snodgrass and G. Gipson)**.

Zhu Research Program

A biochemical approach was developed and implemented for characterization and monitoring of insecticide resistance in the tarnished plant bug. More than 40 populations were collected from the delta areas of Arkansas, Louisiana, and Mississippi and were used to establish detoxification enzyme activity baselines, including esterase, glutathione S-transferase, and acetylcholinesterase. Results indicated that metabolic detoxification is a major resistance mechanism to chemical insecticides, and that detoxification enzymes are currently suppressible with corresponding enzyme inhibitors. (**Y.C. Zhu, G.L. Snodgrass, R. Luttrell**).

Molecular cloning and sequencing were conducted to identify and characterize genes responsible for insecticide resistance in the tarnished plant bug. Transcriptome sequencing coped with microarray analysis has identified many genes associated with chemical insecticide resistance in the tarnished plant bug. In addition to resistance candidate genes, several new genes were identified with potential association to insecticide resistance development in the tarnished plant bug. (**Y.C. Zhu, Z. Guo, R. Luttrell**).

Extra-oral digestion by sucking insects may be detrimental and responsible for cotton square/boll shedding. A molecular and biochemical study was initiated to investigate mechanisms of feeding damage to host plants caused by tarnished plant bug. More than 5,000 cDNA clones from salivary glands were sequenced for identification of detrimental genes associated with feeding damage to cotton. Approximately 40 enzymes were putatively annotated by using Blast similarity search of GenBank. Several of those enzymes are associated with the protein, semicellulose, and other nutrient digestion and absorption. This study established a foundation for further functional studies to identify key genes associated with feeding damage to field crops. (**Y.C. Zhu, Z. Guo, R. Luttrell**).

A comparative study was conducted to investigate molecular and biochemical mechanisms of Bt resistance in the sugarcane borer. Major Bt resistance-related cDNAs were cloned and sequenced, including 3 aminopeptidases, 3 alkaline phosphatases, 1 cadherin, and numerous trypsin and chymotrypsin cDNAs. Gene structures, gene expression levels, and enzyme activities were comparatively characterized between susceptible and resistant strains. Functional study with RNAi indicated that altered aminopeptidase gene expressions might be the major Bt resistance mechanism in the sugarcane borer. (**Y. Yang, Y.C. Zhu, R. Leonard, F. Huang**).

Microarray analysis was conducted to explore new resistance genes to Bt in the sugarcane borer. cDNA libraries were constructed and sequenced. Approximately 6,000 unique cDNAs (contigs and singletons) were assembled. DNA chips were manufactured, and microarrays were performed to compare and analyze global gene expressions between Bt-susceptible and –resistant strains. Preliminary microarray data identified a few new enzymes that may be associated with detoxification and Bt resistance development in the corn borer. (**Z. Guo, Y.C. Zhu, F. Huang**).

Midgut proteinases are involved in Bt protoxin activation and toxin degradation, then mediate Bt specificity, toxicity, and resistance development in the target insect. To enhance Bt performance and delay resistance development, experiments were conducted to examine the interaction of Bt toxin with bio-active reagents, proteinase inhibitors, in the tobacco budworm. Several proteinase inhibitors were evaluated for their insecticidal activity and interaction with Cry1Ac. Results indicated that many trypsin and chymotrypsin inhibitors could significantly suppress larval growth and development and synergize Bt toxicity. **(Y.C. Zhu).**

Insect midgut plays an important role in overcoming host plant defenses and in detoxifying insecticides. Sequencing and analysis of midgut transcriptomes in major cotton insects may facilitate identification of genes responsible for insecticide resistance development. More than 5,000 cDNAs were sequenced from midgut tissue of the tobacco budworm. Approximately 143 contigs and singletons were putatively identified as insecticide resistance-related gene transcripts, encoding a nearly complete list of candidate genes for insecticide detoxification and resistance evolution. Approximately 70% of the resistance gene transcripts are new resistance candidate gene transcripts in GenBank. Profiling midgut gene transcripts in this study will certainly facilitate further study to examine and compare global gene expression levels between biological/physiological samples to identify the most important gene for future gene knockdown or protein inhibition. **(Y.C. Zhu).**

Expressed sequence tags (EST) from larval gut of the European corn borer (ECB) were sequenced to explore candidate genes potentially involved in Bt toxicity and resistance. A total of 12,519 ESTs with an average length of 656 bp were sequenced. EST analysis revealed 52 candidate genes that potentially have roles in Bt toxicity and resistance. Comparisons of expression profiles of 41 candidate genes between Cry1Ab-susceptible and resistant strains of ECB by RT-PCR showed 10 candidate genes apparently down-regulated and 12 genes were up-regulated in the resistant strain. **(C. Khajuria, Y.C. Zhu, M.S. Chen, K.Y. Zhu).**

Bt resistance allele frequency was monitored in field populations of *Helicoverpa armigera* using F_2 screen. Results showed that the resistance allele frequency in field populations of *H. armigera* ranged from 0.0119 to 0.0297 with an overall frequency of 0.0146 and a 95% confidence interval of 0.0084–0.0225. (**F.Y. Liu, Z.P. Xu, Y.C. Zhu, F. Huang, Y.H. Wang, H.L. Li, H. Li, C.F. Gao, W.J. Zhou, J.L. Shen**).

F_1/F_2 screen, dose-response bioassays, and field survey were used to monitor resistance to the Cry1Ac-expressing cotton in a field population of *Helicoverpa armigera*. Field survey showed an increased trend of egg populations of *H. armigera* on Bt cotton in recent years. By using the F_2 screening procedure, the resistance allele frequency in 2007 was estimated to be 0.075 (95% CI: 0.053 - 0.100), which was 12-times greater than that estimated 9 years ago. Dose-response bioassay with the field population collected from the same area showed a significant resistance level (11-fold) to Cry1Ac toxin compared to a laboratory susceptible strain. (**Z.P. Xu, F.Y. Liu, J. Chen, F. Huang, D.A. Andow, Y.H. Wang, Y.C. Zhu, J.L. Shen**).

Accomplishments – Cooperative Agreement Research

Chukwuma, Frank. Alcorn State University. Pest identification and the development of IPM systems for sweetpotato in the Mississippi Delta. Cooperative Agreement 58-6402-6-079. Agreement Period: 8/1/2006 – 7/31/2011

Sweetpotato root damage by insect feeding and crop yields were evaluated in field plots with a 3-year history of ryegrass and by incorporating HumaSoil based soil conditioner. Insect populations were monitored by weekly sweep net sampling and biweekly counts of insects collected on two different sizes of purple sticky traps and Pherocon™ AM yellow sticky traps. The sticky traps attracted more insect pests than those captured by sweep net sampling. Several species of click beetles (adult wireworms, mostly *Conoderus vespertinus*), *Cerotoma trifurcata*, *Lygus lineolaris*, *Diabrotica undecimpunctata*, tortoise beetles and flea beetles were collected in purple/yellow sticky traps or sweep net samples. Overall, mean insect damage was not significantly different ($P<0.001$) between control and test plots with the history of ryegrass or incorporated with soil conditioner. But mean weight of marketable sweetpotatoes significantly differed under different growing conditions. Different sweetpotato varieties showed significantly different ($P<0.001$) susceptibility to insect damage. Marketable yield of sweetpotatoes also significantly differed ($P<0.001$) among varieties. **(T. Rashid, L. Adams)**

Glycoside analysis samples were collected by digging one sweetpotato plant from each of 4 replicated rows. Roots were dried at room temperature. Periderm from each undamaged root was separated. Samples were lyophilized and stored at -80^{o}C. Tissue material was sequentially and successively extracted with solvents with increasing polarity (i.e. Hexane, 100% methanol, 50% methanol (v/v), and water) and centrifuged in preparation for glycoside analysis. **(T. Rashid, L. Adams)**

Spatial distribution and abundance of foraging imported fire ants was examined in sweetpotato fields in the Mississippi Delta. Foragers were collected in baited vials on a 10 m grid throughout the field. Other ant species were also identified and counted. Distribution of foraging ants appeared to be related to annual cycles of disturbance in the field. Movement of foragers into the field likely corresponded to movement of colonies into the field, considering typical foraging distance for fire ants. Data are being analyzed for determining the edge effect on ant foraging. **(T. Rashid)**

Gore, Jeffery. Delta Research and Extension Center, Mississippi State University. Bt risk assessment for lepidopterous pests of cotton. Cooperative Agreement 58-6402-8-13 Agreement Period: 7/1/2008 – 7/01/2013

The impact of bollworms, *Helicoverpa zea* (Boddie), on yields of Bollgard II cotton was evaluated in 2010. Treatments were in a randomized complete block design with three replications and included 16-row sprayed and unsprayed strips. Coragen (chlorantraniliprole) was applied at 0.09 lbs ai/A three times during the season. Based on drop cloth samples, bollworm densities were significantly lower in the sprayed strips than the unsprayed strips. Additionally, boll damage was significantly lower in the sprayed strips than in the unsprayed strips. Cotton lint yields were significantly higher in the sprayed plots compared to the unsprayed plots. Yields in the sprayed plots averaged 982.4 lbs lint per acre compared to 849.4 lbs lint per acre for the unsprayed plots. These results indicate the need for supplemental insecticide applications on Bollgard II cotton to prevent yield losses from bollworms in some situations. **(J. Gore, D. Cook, R. Jackson).**

Bollworm, *Helicoverpa zea* (Boddie), survival, feeding, and injury in Bollgard II cotton washigher in 2010 than in previous years. Collections of bollworms were made from multiple commercial cotton fields and corn fields across Mississippi. The larvae were transported to the laboratory and reared for one generation on artificial diet. Individuals from each colony were tested against currently registered dual gene cottons (Bollgard II and Widestrike) and non-Bt cotton. Larvae were fed excised plant tissues in the laboratory. Larval mortality was rated at 72 and 96 hours. Mortality of the larvae collected from Bt crops was compared to mortality of colonies collected from non-Bt crops. Mortality of larvae collected from Bt crops was significantly lower on Bollgard II and Widestrike cotton than larvae collected from non-Bt corn on each of those cottons. Mortality for all colonies was significantly higher on Bollgard II cotton than non-Bt cotton. Mortality of bollworms collected from a Bollgard II field was not significantly different than the mortality of bollworms collected from non-Bt corn on Widestrike cotton. Each of the colonies was given to the USDA, Southern Insect Management Research Unit for further testing. **(J. Gore, D. Cook).**

An experimental dual-gene cotton was evaluated in 2010. Plots of the experimental cotton were planted at the Delta Research and Extension Center in Stoneville Mississippi. The plots were arranged as a split plot design with four replications. The main plot factor was foliar insecticide and included sprayed for Lepidoptera or unsprayed. The subplot factor was cotton genotype and included two experimental lines and a non-Bt line. Bollworm and fall armyworm densities were significantly lower in the Bt lines compared to the non-Bt lines. Additionally, sprayed plots had lower densities of Lepidoptera than the non-sprayed plots. These results suggest that this new cotton line will perform similar to commercially available dual gene cottons. **(J. Gore, D. Cook).**

The role of corn hybrids containing multiple proteins that target above ground pests (i.e. corn borers, corn earworms, and armyworms) were evaluated for their efficacy against corn earworm and the impact it will have on infestations in cotton. In 2010, bollworm populations were higher during June than in previous years. These dual-protein corn hybrids had significantly less ear feeding compared to single gene corn hybrids. However, some larvae still developed to the later instars on these corn hybrids. Future research will need to focus on the role of dual-protein corn hybrids on corn earworm population dynamics. **(J. Gore, D. Cook).**

Insecticide resistance in all insect pests of cotton threatens the economic viability of dual-gene Bt cottons. Increased costs of foliar insecticides associated with resistant insects compounded with technology fees of dual-gene cottons results in a significant economic cost for growers. Surveys of cotton aphid susceptibility to neonicotinoid insecticides were conducted from 2008 to 2010.

Cotton aphids were collected from commercial cotton fields across Mississippi, Louisiana, Arkansas, and Tennessee. The aphids were collected from fields where control failures with foliar neonicotinoids were reported. Additional collections were made from cotton fields in these states where no foliar applications of neonicotinoid insecticides had been made. Leaf dip bioassays were used to measure their susceptibility to neonicotinoids. Cotton aphids collected from fields where control failures had occurred showed significant levels of resistance each year. Cotton aphids collected from non-treated fields in 2010 were 10-fold more resistant than cotton aphids collected from non-treated cotton fields in 2008. These data indicate that cotton aphid resistance to neonicotinoids is increasing in the mid-South. **(J. Gore, D. Cook).**

Gore, Jeffery. Delta Research and Extension Center, Mississippi State University. Refining the area-wide suppression of the tarnished plant bug in Mississippi. Cooperative Agreement 58-6402-8-275. Agreement Period: 4/10/2008 – 3/31/2011.

The tarnished plant bug is the most important pest of cotton in the mid-South. Widespread resistance to pyrethroid and organophosphate insecticides has made control marginal at best. An area-wide tarnished plant bug suppression program developed at SIMRU has been used in an overall tarnished plant bug management program. On grower farms, selective herbicides were sprayed during the spring to control broadleaf weeds. In-season management practices were studied to complement the area-wide suppression program. On the farms where herbicide applications were made, different varieties of Bollgard II cotton with different levels of maturity were planted. Additionally, early season Diamond applications were made during the third week of squaring to manage tarnished plant bugs. On early maturing varieties, fewer insecticide applications were needed to control tarnished plant bugs. In fields where early applications of Diamond were used, subsequent applications were needed on a 10-14 day interval. In contrast, fields that did not receive the early Diamond applications required 5-7 day application intervals to get similar levels of control. Overall, these management strategies used in an integrated pest management system reduced the numbers of insecticide applications needed for tarnished plant bugs. **(J. Gore, D. Cook).**

Tarnished plant bug densities were determined weekly throughout 2009 and 2010 in a 10 square mile area in Stoneville, MS. Non-cultivated hosts were sampled with a 15 in diameter sweep net in line transects along non-crop areas. During the cropping season, representative fields of cotton, soybean, and corn were sampled on a grid pattern so that each field was sampled at 6 locations regardless of field size. These data have not been fully analyzed at this time. **(J. Gore, D. Cook).**

Gould, Fred. North Carolina State University. Bt-resistance frequency detection. Cooperative Agreement 58-6402-6-048. Agreement Period: 8/30/2006 – 3/31/2011.

Our lab collaborated with Juan Luis Jurat-Fuentes, O.P. Perera, and others in evaluating whether expression of a gut membrane-bound alkaline phosphatase (ALP) could be involved in resistance to Bt toxins and if monitoring ALP gene expression levels could be used for resistance monitoring. One paper from that collaboration was recently accepted by PLoS One. (Jurat-Fuentes, J. L., Karumbaiah, L., Jakka, S.R.K., Ning, C., Liu, C., Wu, K., Jackson, J., Gould, F., Blanco, C., Portilla, M., Perera, O. P., and M. J. Adang. 2011. Reduced levels of membrane-bound alkaline phosphatase are common to lepidopteran strains resistant to Cry toxins from *Bacillus thuringiensis*. PLoS One (accepted).) We recently sent our new 454 Roche transcriptome data to O. P. Perera and Kent Shelby for inclusion in a genomic database. **(F. Gould)**

Other work in our lab has involved archiving of collected *H. zea* and *H. virescens* moths from the cotton belt as well as more Northern areas. This reference collection can serve as a means of investigating resistance evolution. We have also been maintaining a number of *H. virescens* strains with different Bt resistance mechanisms. **(F. Gould)**

Huang, Fangneng. Louisiana State University. Characterizing mechanisms of *Bacillus thuringiensis* resistance in sugarcane corn borer. Cooperatvie Agreement 58-6402-6-035. Agreement Period: 7/14/2006 – 7/14/2011.

No report.

Luttrell, Randall. University of Arkansas. Monitoring *Helicoverpa zea* populations for susceptibility to Cry1Ac. Cooperative Agreement 58-6402-6-059. Agreement Period: 8/31/2006 – 3/31/2011.

No research to report. Active research concluded in 2009.

Martin, Steve. Delta Research and Extension Center, Mississippi State University. Economic analysis of Bt corn technologies and associated refuge systems. 58-6402-0-459. Agreement Period: 4/01/2010 – 4/01/2013.

Economists are waiting on field data for this project. Plans are to conduct an economic analysis comparing the single gene, 50% corn refuge system with the stacked gene, 20% corn refuge system. Hopefully, the research will be completed by late summer. **(S. Martin, R. Jackson)**.

Musser, Fred. Mississippi State University. Bt risk assessment for Lepidopterous pests of cotton. Cooperative Agreement 58-6402-8-274. Agreement Period: 3/3/2008 – 3/2/2013

In cooperation with EPA, models have been developed to address changes in Bt crops being planted, new Bt events being developed and changes in Bt-crop refuge requirements being proposed. In particular, models have evaluated the impact of reduced refuges required for multi-gene corn coupled with increased selection pressure on corn earworm on the risk of developing resistance to the Bt toxins. Our model indicates that the risk of resistance from dual-gene corn using similar toxins to those found in Bt-cotton combined with reduced refuges may be somewhat higher than for single-gene corn with a larger refuge. Mixing non-Bt and Bt-plants (refuge in a bag) rather than keeping the refuge and Bt areas distinct further reduces the benefit of the refuge. **(F. Musser, M. Caprio)**.

Musser, Fred. Mississippi State University. Development of a tarnished plant bug and stink bug management system. Cooperative Agreement 58-6402-6-061. Agreement Period: 8/22/2006 – 8/22/2011.

Tarnished plant bug resistance to 3 major insecticide classes has been monitored annually since 2006 from 12 areas in central and northeastern Mississippi. Over this time resistance levels have been stable and low for pyrethroid and neonicotinoid insecticides. Resistance levels to organophosphate insecticides were stable but moderate. There was no indication that areas nearer the delta region had any more resistance than bugs collected farther from the delta. **(Musser)**

Payne, Gregory. University of West Georgia. Monitoring for potential recessive resistance in bollworm/tobacco budworm to protein toxins in Bt cotton. 58-6402-8-318. Agreement Period: 7/19/2008 – 7/10/ 2010.

No report.

Publications and Presentation

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Ali, I., Luttrell, R.G., Susceptibility of *Helicoverpa zea* and *Heliothis virescens* (Lepidoptera: Noctuidae. Journal of Invertebrate Pathology **(submitted)**

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Guo, Z., Zhu, Y.C., Luttrell, R.G. cDna microarray analysis of gene expression in insecticide-susceptible and resistant tarnished plant bug. National Cotton Council Beltwide Cotton Conference **(submitted)**

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Gore, J., D. Cook, A. Catchot, F. Musser, B. R. Leonard, G. Lorenz and S. Stewart. "Impact of *Lygus lineolaris* management on biodiversity in cotton IPM" Entomol. Soc. Amer. Annual meeting, San Diego, CA, Dec. 14, 2010.

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Oppert, C., Perera, O.P., Castagnola, A., Gould, F., Jurat-Fuentes, J.L. Proteomic analysis of resistance to diverse Cry toxins from *Bacillus thuringiensis* in strains of *Heliothis virescens* National Entomological Society of America Annual Meeting

Perera, O.P., Shelby, K., Jurat-Furentes, J.L., Draft transcriptome assembly of the tobacco budworm, *Heliothis virescens* (L.) and expression profiles of gut specific genes. National Entomological Society of America Annual Meeting

Perera, O.P., Snodgrass, G.L., Gore, J., Jackson, R.E. Insecticide resistance in *Lygus lineolaris*: a population genomics approach Entomological Society of America Annual Meeting

Rashid. T. Insect Pests of Sweetpotatoes in Mississippi (Alcorn State University Extension Program) Managing Agricultural Insects Pests. (Fact Sheet, Alcorn State University Extension Program) Managing Fire Ants. (Fact Sheet, Alcorn State University Extension Program)

T. Rashid, C. A. Abel and L. C. Adams, Insect Population Monitoring and Damage to Sweetpotatoes. National Sweetpotato Collaborators Group Meeting, Orlando, FL, February 6-7, 2010.

Zhu, Y. C., Examination of esterase gene expression and enzyme activity in field populations of the tarnished plant bug, *Lygus lineolaris*. Proc. 6th Asia-Pacific Congress of Entomol. APCE2009: 49-50 (2009).

Zhu, Y. C., Extensive implementation of transgenic crop: potential risk of resistance development and mechanisms in target insects. Proc. 6th Asia-Pacific Congress of Entomol. APCE2009: 326 (2009).

Zhu, Y.C., Organophosphate Resistance in the Tarnished Plant Bug: Population Variation in Major Detoxification Enzyme Activity and Gene Expression. 56th Conference of the MEA. Oct. 27-28/2009, Starkville, MS.

Zhu, Y. C., Snodgrass, G., Monitoring Insecticide Resistance in Field Populations of the Tarnished Plant Bug Using Biochemical and Molecular Approaches. Proc. 2010 Beltwide Cotton Conf. CD-ROM. Jan. 4-7, 2010. New Orleans, LA 2010.

Yunlong Yang , Yu Cheng Zhu , James Ottea , Claudia Husseneder , B. Rogers Leonard , Fangneng Huang , Analyses of sequences, gene expressions, and RNAi of three Cry1Ab-resistance related aminopeptidase genes in the sugarcane borer, *Diatraea saccharalis*. Monday, December 14, 2009 Hall D, First Floor (Convention Center) ESA 2009 2009 ESA Annual Meeting, December 13-16, 2009

Zhize Zhang , Richard Shukle , Omprakash Mittapalli , Y.C. Zhu , John C. Reese, Bao-Zhen Hua , Ming-Shun Chen , A gut transcriptome of the Hessian fly (Mayetiola destructor), a member of gall midges ESA 2009 Wednesday, December 16, 2009 Hall D, First Floor (Convention Center) 2009 ESA Annual Meeting, December 13-16, 2009

Zhu, Y.C. "High selection pressure and Bt resistance development in target insects". Special seminar invited by Dr. Marianne P. Carey, Dept. of Biochemistry, Case Western Reserve University. 7/28/2010.

Zhu, Y.C. "Insecticide Resistance genes, Unwanted and/or Wanted?" Special seminar invited by Dr. Zachary Huang, Dept. of Entomol, Michigan State University. 7/29/2010.

Research Plans

2011 Research Plans – Internal Research Program

Adams Research Program

Continue to evaluate products to suppress reniform nematodes and asset root-knot nematode populations in sweetpotatoes grown in the Mississippi Delta. **(L. C. Adams, R. G. Luttrell and C. Johnson)**

Develop procedures to determine spring populations of wireworms, rootworms, white grubs and sugarcane beetles in sweetpotato fields. **(L. C. Adams, R. G. Luttrell and C. Johnson)**

Participate in the 2011 NSCG Sweetpotato variety trials. **(L. C. Adams, R. G. Luttrell and C. Johnson)**

Continue to monitor and report populations of *H. zea* and *H. virescens* in the Mississippi Delta through pheromone trapping. **(L. C. Adams and C. Johnson)**

Continue collaborative research with Alcorn State University scientists studying sweetpotato insect identification, sampling and damage in the Mississippi Delta.
(L. C. Adams, R. G. Luttrell and C. Johnson)

Allen Research Program

A survey of looper and stink bug species on soybean fields in the Mississippi Delta will be conducted and continued over the next few years to evaluate the temporal and spatial occurrence of these insects **(C. Allen, L. Andrews and A. Walters).**

Cold tolerances of various Lepidopteran and Hemipteran insect pests under diapausing and non-diapausing conditions will be examined **(C. Allen, R. Jackson, and G. L. Snodgrass).**

Evaluation of economic thresholds for bollworms and stink bugs on maturity group IV and V soybeans will be conducted. **(C. Allen and R. Jackson)**

Susceptibilites of various stink bugs species to commonly used insecticides will be evaluated on field collected insects from various locations within the MS Delta **(C. Allen, A. Walters and L. Andrews).**

Presence and type of pollen on stink bugs and tarnished plant bugs collected in light and sticky traps will be examined and the practicality of using this data for indications of movement will be explored **(C. Allen and G. Jones).**

Jackson Research Program

Tarnished plant bug infestations in cotton fields in the Mississippi Delta will be compared with that of the MS Hills region with regard to infestation levels and the proportion of the TPB population immigrating into cotton throughout the growing season. **(R. Jackson, G. Snodgrass, C. Allen)**.

Pyramided-gene, Bt field corn will be evaluated for its effects on bollworm population ecology and dynamics. Increased adoption of these Bt technologies have the potential to reduce bollworm population levels, which could also impact populations infesting cotton later in the growing season. **(R. Jackson, C. Allen, R. Luttrell)**.

Bollworm populations will be tested for susceptibility to various Bt toxins/technologies. We will attempt to identify a relationship between laboratory assay results and field performance of these populations on Bt technologies. **(R. Jackson, R. Luttrell, C. Allen)**.

Sentinel plots will be placed throughout the MS Delta to evaluate the benefit of pyramided-gene, Bt cotton varieties for bollworm control. Economic returns based on differences between insecticide-treated and non-treated plots of the same variety will be the basis of comparison among Bt technologies. These Bt varieties will also be compared to non-Bt varieties at these sites. **(R. Jackson, R. Luttrell)**.

Luttrell Research Program

Statistical explorations of a large dataset on bollworm and tobacco budworm susceptibility to a number of different Bt toxins (Cry1Ac, Cry2Ab2, Cry1F, Vip3a) will be completed and published. This summative examination of ~10 years of bioassays should be useful to others interested in tracking changes to Bt susceptibility over time. Seasonal dynamics of pest occurrence, generation time of different conventional and transgenic host plants and ecological traits of the field populations sampled for the Bt assays will be examined. Additional field populations will be sampled and assayed if field resistance is expected in local populations. **(R. Luttrell, Ibrahim Ali, R. Jackson)**

A multiple-year effort to compile and study crop, insect and management production records for Wildy Farms in Leachville, Arkansas will be completed and published. **(R. Luttrell, C. Allen, T. Teague)**.

Efforts to produce large numbers of Lepidotera (corn earworm, tobacco budworm, soybean looper, fall armyworm and southwestern corn borer) for field infestation studies on cotton, soybean and corn will be initiated. Given sufficient numbers of insects for large cage and field infestations, studies will be conducted to test contemporary transgenic crops and insecticides against varying densities of artificially infested pests. **(Luttrell, Jackson, Allen, Ford, Houston).**

Field experiments will be initiated to compare insect control and profitability of conventional and transgenic cottons. **(Luttrell, Jackson, Allen)**.

Perera Research Program

Conduct studies on the genetic variation of detoxification genes to identify loci associated with insecticide resistance. Single nucleotide polymorphisms identified from the transcriptomes will be used in the population genomics studies. **(O. P. Perera).**

Characterization of immune response in TPB infected with *Beauveria bassiana* will be carried out using TPB infected with NI-8 strain of *B. Bassiana*. Insects sprayed at a rate of 10^9 conidia/ml were collected at 6 hour intervals for 72 hours. RNA extracted from the insects will be used to profile expression patterns of known immune response pathways during the time course. In addition, we will be able to profile the gene expression in *B. Bassiana* during the course of infection when the genome sequence is available. These studies will help us understand the host-pathogen interactions between these two organisms and will provide insights into enhancing the efficacy of biological control by *B. Bassiana.* **(O. P. Perera).**

Portilla Research Program

Develop a protocol to conduct bioassays using artificial diet to evaluate the effect of *B. Bassiana* on non target insects: Pirate bugs, big eye bugs, lady bugs, green lacewing, jumping spiders, crab spiders and other arthropods present in the area at the moment of the applications. Possibly fire ants and stink bugs will be included in the evaluation. Experiments will be conducted under laboratory, field and green house conditions. **(M. Portilla)**.

Method for determining the effect of the entomopathogenic fungus, *Beauveria bassiana* and the insect growth regulator Diamond on fecundity and growth inhibition of the tarnished plant bug, *Lygus lineolaris*. **(M. Portilla)**.

Effect of different concentration of *B. Bassiana* on the reproductive rates of Lygus lineolarias and *Lygus hesperus*. **(M. Portilla)**.

Snodgrass Research Program

During May, June, and July resistance levels in tarnished plant bug populations found in the delta of AR, LA, and MS to permethrin, acephate, imidicloprid, and thiamethoxam will be determined. At least 20 different locations will be used for each insecticide. Any population found with an unusually high level of resistance to imidicloprid or thiamethoxam will be field tested in cotton to determine what the increased resistance means in terms of field control with recommended rates of the insecticide. **(Snodgrass and Jackson).**

The NI8 strain of *Beauveria bassiana* discovered and propagated at SIMRU will be tested in a replicated large plot (1 acre) test in cotton alone and in combination with novaluron for tarnished plant bug control. Treatments will include NI8, Novaluron, NI8 + novaluron, acephate, and an untreated check. The effect of the treatments on beneficial arthropod populations will also be determined. An effort will be made to conduct the test in June and again in July. **(Snodgrass and Jackson)**.

Laboratory studies on the efficacy of NI8 alone and in combination with novaluron will be conducted in the laboratory using reproductive and diapausing plant bug adults and nymphs. The studies will be aimed at determining possible rates at which to use NI8 and novaluron in the field. **(Snodgrass, Gipson, and New Hire).**

Laboratory studies of the effect of novaluron, NI8, and novaluron + NI8 treatment on beneficial arthropods will be conducted. As many beneficial arthropod species as possible (selected from the most common ones found on cotton and weeds in the mid-South) will be tested. **(Portilla, Luttrell, and Snodgrass)**.

A replicated field test to determine the effect of treatment of plant bug populations on their main weed hosts in September and October with novaluron, NI8, or novaluron + Ni8 to reduce the overwintering (diapausing) generation will be conducted in September and October. The effect of the treatments on beneficial arthropod populations will also be determined. **(Snodgrass, Jackson, New Hire)**.

Zhu Research Program

Characterization of insecticide resistance mechanisms in the tarnished plant bug. (1) Continue to collect tarnished plant bug populations from Mississippi, Arkansas, and Louisiana. Select the colonies/populations with Orthene and neonicotinoid insecticides. Apply microarray analysis of gene regulations in resistant bug. (2) Continue to survey and monitor detoxification enzyme activities in Delta areas (12-15 locations). Establish relationship between enzymatic activity and biological response (LC50 or dose response). (3) Analysis of fitness cost in resistant bug.

Investigation of Bt resistance in the sugarcane borer. (1) Continue to conduct comparative study of Bt resistance candidate genes, including midgut cadherin, alkaline phosphatase, and proteinase genes. (2) Exploratory analysis and evaluation of other detoxification and immune-related genes in Bt resistant strain.

Examination of insecticide resistance in the fall armyworm. Select colony with conventional insecticide or Bt. Apply microarray to analyze gene regulation. Clone and sequence major resistance cDNAs. Measure major detoxification enzyme activities.

2011 Research Plans – Cooperative Agreements

Chukwuma, Frank. Alcorn State University. Pest identification and the development of IPM systems for sweetpotato in the Mississippi Delta. Cooperative Agreement 58-6402-6-079. Agreement Period: 8/1/2006 – 7/31/2011

Pheromone traps will be placed in sweetpotato fields in different parts of Mississippi (Delta, Vardaman and Lorman) to detect the presence of sweetpotato weevil (*Cylas formicarius*). **(Rashid)**

Rearing methods for sugarcane beetle (*Euetheola humilis*) will be developed. Biological and behavioral study will be conducted. Chemical profiles of different life stages will be studied. **(Rashid)**

Gore, Jeffery. Delta Research and Extension Center, Mississippi State University. Bt risk assessment for lepidopterous pests of cotton. Cooperative Agreement 58-6402-8-313. Agreement Period: 7/1/2008 – 7/01/2013

Experiments evaluating the impact of bollworms on yields of Bollgard II and Widestrike cotton will be continued. **(J. Gore and D. Cook).**

Collections of bollworms from Bt crops will be continued in 2011. **(J. Gore and D. Cook).**

As part of a graduate student project, a series of experiments will be conducted to evaluate the impact of Bt corn hybrids on bollworm, *Helicoverpa zea* (Boddie), population dynamics, fitness, and subsequent damage in cotton. The first component of the project will be a survey of commercial corn fields planted with VT Triple Pro corn hybrids. The second experiment will evaluate the overall fitness of bollworm larvae completing development on non-Bt corn, one-gene Bt corn, and two-gene Bt corn hybrids. The final component will use field and laboratory experiments to quantify damage caused by bollworms coming from different corn technologies. **(J. Gore, D. Cook, A. Catchot, and B. Von Kanel).**

An experiment that was initiated in 2010 will be continued in 2011 that examines the impact of volunteer corn on bollworm population dynamics. In this experiment, seed were collected from plots that were planted to different corn technologies. The seeds were picked up by hand after harvest to determine the Bt expression levels in volunteer corn. The corn has been planted in the greenhouse and bioassays will be conducted within the next month to measure survival levels. **(J. Gore and D. Cook).**

Gore, Jeffery. Delta Research and Extension Center, Mississippi State University. Refining the area-wide suppression of the tarnished plant bug in Mississippi. Cooperative Agreement 58-6402-8-275. Agreement Period: 4/10/2008 – 3/31/2011.

Experiments supporting area-wide management of tarnished plant bugs will be continued in 2011. **(J. Gore and D. Cook).**

Gould, Fred. North Carolina State University. Bt-resistance frequency detection. Cooperative Agreement 58-6402-6-048. Agreement Period: 8/30/2006 – 3/31/2011.

None reported.

Huang, Fangneng. Louisiana State University. Characterizing mechanisms of *Bacillus thuringiensis* resistance in sugarcane corn borer. Cooperatvie Agreement 58-6402-6-035. Agreement Period: 7/14/2006 – 7/14/2011.

None reported.

Luttrell, Randall. University of Arkansas. Monitoring *Helicoverpa zea* populations for susceptibility to Cry1Ac. Cooperative Agreement 58-6402-6-059. Agreement Period: 8/31/2006 – 3/31/2011.

No research to report. Active research concluded in 2009.

Martin, Steve. Delta Research and Extension Center, Mississippi State University. Economic analysis of Bt corn technologies and associated refuge systems. 58-6402-0-459. Agreement Period: 4/01/2010 – 4/01/2013.

Plans are to complete the economic analysis in late summer. **(S. Martin, R. Jackson).**

Musser, Fred. Mississippi State University. Bt risk assessment for Lepidopterous pests of cotton. Cooperative Agreement 58-6402-8-274. Agreement Period: 3/3/2008 – 3/2/2013

Modeling efforts will continue to address the risk of insects developing resistance to Bt toxins for an increasingly complex array of Bt toxins and refuge requirements available in corn and cotton. Furthermore a new graduate student will begin research evaluating the role of volunteer corn emerging from former Bt-corn fields on the risk of Bt resistance development in a corn-cotton landscape. **(F. Musser, M. Caprio)**.

Musser, Fred. Mississippi State University. Development of a tarnished plant bug and stink bug management system. Cooperative Agreement 58-6402-6-061. Agreement Period: 8/22/2006 – 8/22/2011.

Resistance monitoring will continue at the same locations as in previous years. **(F. Musser)**.

Payne, Gregory. University of West Georgia. Monitoring for potential recessive resistance in bollworm/tobacco budworm to protein toxins in Bt cotton. 58-6402-8-318. Agreement Period: 7/19/2008 – 7/10/ 2010.

None reported.

IPM Project

Previous Research Project Through 2010: ***Integrated Pest Management for Mid-South Area Row Crops***

Project Scientists: Ryan Jackson (Lead Scientist), Clint Allen, R. G. Luttrell, O. P. Perera, G. L. Snodgrass, Y. C. Zhu, Vacant (vice-Blanco), Jeff Gore (left ARS)

Project Number: 6402-22000-045-00D
Project Type: Appropriated

Start Date: Sep 01, 2005
End Date: Aug 31, 2010

Objectives and Approach

Identify and characterize new sources of maize, soybean, and sweetpotato resistance to insects, e.g. corn earworm, stink bug, and sugarcane beetle. Genetic studies will be conducted using inbred lines derived from two backcross populations of maize that are resistant to silk-feeding corn earworm. A stink bug trap crop will be designed that will produce high populations of the pest to evaluate soybean germplasm for resistance. Field studies will be conducted to collect and identify damaging soil insects of sweetpotato in the Lower Mississippi Delta and sweetpotato germplasm will be screened for resistance to these pests. Develop and enhance biological control strategies for suppression of plantbugs and stink bugs in area-wide pest control systems for southern row crops. Experiments will be conducted in the laboratory, greenhouse, and field to determine the effect of herbivore-induced volatiles on the behavior and life history of natural enemies of tarnished plant bug and stink bugs. Field exploration for new egg and nymphal parasitoids of plant bugs and stink bugs will be conducted. Improve and continue to evaluate an area-wide pest management system for control of tarnished plant bugs in cotton. The research project has demonstrated that a single, March application of a broadleaf herbicide in cotton field margins enclosed in 9 mi2 areas will suppress tarnished plant bug populations during the crop's growing season. Research will be conducted to determine the optimal and minimal-effective sizes for the area-wide program and to determine the economic and environmental costs and benefits of this program. Comparison of tarnished plant bug and stink bug injury to cotton and comparison of various sampling methods to predict yield losses from these insects in flowering cotton. Field experiments will be conducted to develop accurate sampling methods for tarnished plant bugs and stink bug in flowering cotton. Additionally, injury potential of these insects will be determined on cotton to refine current thresholds or develop new thresholds based on those sampling methods

2010 Annual Report

A new source of corn earworm resistance was successfully pyramided with an established resistance trait (maysin) that resulted in the development of corn lines with high levels of resistance to the pest. These lines were used to develop resistant corn hybrids that should increase protection of corn hybrids from corn earworm damage and reduce the incidence of mycotoxins in corn. Various sampling techniques were used to monitor insect populations in sweetpotato grown in the MS Delta. Wireworms were the most consistent insect pest of sweetpotato in this region. Flea beetles, southern corn rootworms, and white grubs were also occasional pests that damaged harvestable potatoes. The most yield-limiting pest of sweetpotato identified in the MS Delta was reniform nematode. Nematicides were shown to improve marketable sweetpotato yields by nearly 40%. Sweetpotato varieties were evaluated for resistance against insect pests and nematodes, and the Covington variety showed promise in reducing injury caused by flea beetle species. Area-wide control of tarnished plant bugs in cotton was tested in large areas approximately 9 square miles across MS, AR, LA, and TN. This method involved applying a selective herbicide in March to broad-leaf weed hosts growing near cotton. Eliminating early-season host plants of tarnished plant bug reduced tarnished plant bug densities during the cotton growing season. For every dollar spent in controlling wild plant hosts of the tarnished plant bug, growers realized an average benefit of $7.49 in reduced insecticide costs for control of this pest in their cotton. Other tests indicated herbicide-treated areas must be at least $5mi^2$ in order to achieve this benefit. Sterile releases of tarnished plant bug adults were shown to reduce tarnished plant bug populations in laboratory and large field cage experiments. This technique had potential as a tool to augment the herbicide applications used in the area-wide control program for tarnished plant bugs. Tarnished plant bug thresholds in cotton were evaluated by comparing sampling methods and various thresholds for initiation of control measures in cotton. Plant-based scouting procedures that incorporated fruiting form injury and general plant vigor provided the best correlations with yield. Insect counts using various sampling methods were more variable and less reliable than plant-based methods. These experiments were conducted in commercial situations across the Mid-South, and control recommendations have been amended in the region. A new threshold of 10% dirty squares (a medium-sized flower bud with a yellow discoloration caused by tarnished plant bugs) was added to the recommendations. Insecticide resistance monitoring of tarnished plant bug populations throughout the Mid-South identified acephate resistance in 2005. Acephate resistance was semi-dominant, which led to the spread of this resistance across field populations in the Mid-South. A bioassay for detecting resistance in tarnished plant bug populations to the neonicotinoid insecticides, imidacloprid and thiamethoxam, was developed in 2006. To date, only low levels of tolerance to these insecticides have been found.

Project Accomplishments

Rapid Dissemination of Results from Tarnished Plant Bug Insecticide Resistance Monitoring Efforts. The discovery of acephate resistance in tarnished plant bug populations allowed us to warn consultants, growers, and Cooperative Extension Service personnel about control problems. As a result, Cooperative Extension Service control guides have recommended 2007 in AR, LA, MS, and TN, that alternation of insecticide classes be used in tarnished plant bug control to reduce resistance development. The bioassay developed to monitor resistance to neonicotinoid insecticides is the only rapid method of determining the resistance level in a population to these insecticides. These insecticides are the only major class to which resistance has not developed and managing resistance by alternation of insecticides depends on these insecticides being effective.

Host Plant Utilization of Tarnished Plant Bug as Measured by Stable Carbon Isotope Analysis. Insecticide applications targeting tarnished plant bugs have risen dramatically in recent years. During this period, changes to the cropping landscape and production practices have led to knowledge gaps related to tarnished plant bug ecology. Because no synthetic pheromone exists for tarnished plant bug, population level ecological studies have been difficult. An analytical tool was used to establish “proof of concept” for evaluating host plant utilization of tarnished plant bug during the cotton growing season. Information from this study will enable researchers to identify the importance of crop hosts, such as field corn, and wild hosts, such as pigweed, on tarnished plant bug populations infesting cotton fields.

Development of Genetic Markers for Population Genetic Studies of Tarnished Plant Bug, *Lygus lineolaris*. Multiple insecticide resistance in tarnished plant bug is becoming more prevalent in the MS Delta. Lack of population genetic data is a major drawback in understanding genetic structure, allele frequencies, and gene flow among tarnished plant bug populations. By using genetic markers developed for tarnished plant bug, we previously detected substantial gene flow between tarnished plant bug populations separated by approximately 10 miles. An expanded 3-year study of 5 populations across the Mississippi Delta along a 200-mile transect was completed in 2009. These data demonstrated the potential for rapid spread of insecticide resistance in tarnished plant bug populations due to the high level of gene flow among populations. Transcriptome of Lygus (L.) lineolaris was sequenced using high-throughput nucleotide sequencing. Single nucleotide polymorphisms in expressed genes, especially those responsible for detoxification mechanisms have been identified for use in population genetic studies to determine the genetic diversity of natural populations. In addition, several sensory receptors (odorant binding proteins) were identified and characterized. Tissue expression patterns of these odorant binding proteins were also studies. Odorant binding proteins identified in this project will be studied further to develop repellents or attractants for tarnished plant bug.

Relating Corn Earworm Pyrethroid Susceptibility to Plant Host Ecology. The corn earworm is a serious pest of cotton and corn production in the U.S. One of the main control strategies for corn earworm in these crops is synthetic insecticides, primarily the pyrethroid class of chemistry. From 1990 to 2007, corn earworm resistance to cypermethrin increased over 20-fold. Corn earworm populations were monitored from May through September in 10 states for susceptibility to pyrethroids. Through the use of analytical techniques, pyrethroid susceptibility in moths was related to larval plant hosts. Peak pyrethroid resistance coincided with corn earworm populations completing a generation on field corn. When wild plant hosts or cotton and soybean contributed corn earworm numbers to the general population, the level of pyrethroid resistance was lower. Detecting problem areas in the Mid-South and southeastern U.S. for pyrethroid resistance selection will facilitate changes in management strategies to avoid high-level resistance development. Because pyrethroids are used in most management programs for corn earworm, this information will benefit the agricultural community as well as the general public through avoidance of high-level pyrethroid resistance development.

Susceptibilities of Tarnished Plant Bug and Stink Bug Nymphs to Insecticides. Tarnished plant bugs and various stink bug species are some of the most serious pests of cotton and soybean, respectively, in the Mid-South. Most of the measurements of susceptibility to insecticides are conducted using the adult stage of these insects, although field applications target both adult and immature stages. Multiple colonies of tarnished plant bugs and stink bugs were used to examine the susceptibilities of the different nymphal instars and adult stages to a pyrethroid and an organophosphate insecticide. Generally, the fourth and fifth instars of the species tested were more tolerant to both permethrin (pyrethroid) and methamidophos (organophospate) compared to the adult stage. First and second instars were more susceptible than adults and the other instars tested. The results of this study indicate that the success of an insecticide application for control of these insects can be improved by targeting populations during the more vulnerable life stages of development. Making successive applications of an insecticide at short intervals (a few days) may be a practical approach for control since a proportion of the more tolerant, later instars, will develop into adults which are more susceptible to the insecticides tested at the time of the second application.

Monitoring for Tarnished Plant Bug Populations with Metabolic Resistance to Insecticides. Organophosphate insecticides have predominantly been used against tarnished plant bugs in recent years due to reduced efficacy of other insecticides. A biochemical approach was developed to survey organophosphate resistance related enzyme levels in field populations of the tarnished plant bug. A susceptible laboratory strain was established and was used as standard strain for enzyme level comparison in different years. More than 40 populations were collected in from the delta areas of Arkansas, Louisiana, and Mississippi, and 12 populations, represent thought to different levels of resistance, were also collected and assayed. Three esterase substrates and one each of glutathione S-transferase (GST) and acetylcholinesterase (AChE) were used to examine corresponding detoxification enzyme activities in different populations. Compared with a laboratory susceptible colony, up to 5.29-fold of esterase, 1.96-fold of GST, and 1.97-fold of AChE activities were detected in the populations. In addition to a survey of enzyme activities in different populations, the impact of enzyme inhibitors which could be used in formulation to synergize insecticide toxicity against target pests were evaluated. As much as 52-76% of esterase, 72-98% of GST, and 93% of AChE activities were inhibited in vitro. By revealing variable esterase and GST activities among the field populations, background information has been established which may lead to a better understanding of underlying resistance mechanisms in the tarnished plant bug. This study also provided information for suppression of detoxification enzymes which can be incorporated into resistance management strategies.

2010 IPM Project Publications

Zhu, Y., Fang, Q.Q. 2009. The Phylogenetic Relationships of Introduced Aphelinus (Hymenoptera: aphelinidae), Biological Control Agents of the Russian Wheat Aphid (Homoptera: aphididae). Insect Science. 16:277-286

Jia, B., Liu, X., Liu, Y., Zhu, Y., Gao, C., Shen, J. 2009. Inheritance, Fitness Cost, and Mechanism of Resistance to Tebufenozide in Spodoptera exigua (Hubner) (Lepidoptera: Noctuidae). Pest Management Science. 65(9):996-1002
Lee, K.C., Sun, J., Zhu, Y., Mallette, E.J. 2009. A Case Study of the Formosan Subterranean Termite, Coptotermes formosanus Shiraki(Isoptera: Rhinotermitidae) Transported with a Non-Cellulosic Commercial Carrier in South Mississippi. Sociobiology. 53(3):619-630.

Wang, Y., Liu, X., Zhu, Y., Zhou, W., Shen, J. 2009. Inheritance Mode and Realized Heritability of Resistance to Imidacloprid in the Brown Planthopper, Nilaparvata lugens (Stal) (Homoptera: Delphacidae). Pest Management Science. 65:629-634.

Zhu, Y., Sun, J., Luo, L.L., Liu, X.F., Lee, K.C., Mallette, E.J., Abel, C.A. 2010. Identification of Two Haplotypes of Cytochrome Oxidase Sub-unit II (COII) Gene of the Formosan Subterranean Termite, Coptotermes formosanus Shiraki (Isoptera: Rhinotermitidae) in South Mississippi. Pest Management Science. 66(6):612-620.

Wang, Y., Chen, J., Gao, C., Zhou, W., Zhu, Y., Shen, J. 2009. Dynamics of Imidacloprid Resistance and Cross-Resistance to Other Insecticides in the Brown Planthopper, Nilaparvata lugens (stal)(Homoptera: Delphacidae). Entomologia Experimentalis et Applicata. 131:20-29.

Snodgrass, G.L., Jackson, R.E., Abel, C.A., Perera, O.P. 2010. Utilization of Early Soybeans for Food and Reproduction by the Tarnished Plant Bug (Heteroptera: Miridae) in the Delta of Mississippi. Environmental Entomology. 39(4): 111-121 DOI: 10.1603/EN09379

Abel, C.A., Snodgrass, G.L., Jackson, R.E., Allen, K.C. 2010. Oviposition and Development of the Tarnished Plant Bug (Hemiptera: miridae) on Field Maize. Environmental Entomology. 39(4):1085-1091 DOI: 10.1603/EN10010

New Research Project Through 2015: ***Insecticide Resistance Management and New Control Strategies for Pests of Corn, Cotton, Sorghum, Soybean, and Sweet Potato***

Project Scientists: Ryan Jackson (Lead Scientist), Clint Allen, R. G. Luttrell, O. P. Perera, G. L. Snodgrass, Y. C. Zhu, Vacant (cive-Blanco)

Project Number: 6402-22000-063-00D
Project Type: Appropriated

Start Date: Sep 01, 2010
End Date: Aug 31, 2015

Objectives

The long-term objective of this project is to develop an improved understanding of how the changing cropping landscape impacts insecticide resistance development and management of various insect pest species in order to increase profitability and sustainability of mid-South row crops. Objective 1: Improve tarnished plant bug control and insecticide resistance management by gaining new information on the pest’s ecology and biology using multi-disciplinary approaches, e.g. molecular genetic tools, stable carbon isotope analysis, gene expression and proteomics, and insecticide resistance assays coupled with field sampling. Objective 2: Determine the effect of bollworm ecology (corn earworm) on resistance to pyrethroid insecticides by developing and utilizing genetic markers linked to resistance traits, stable carbon isotope analysis, gossypol detection in adult insects, and insecticide resistance monitoring. Objective 3: Develop pest control strategies for the U.S. Mid-South’s Early Soybean Production

System by determining accurate treatment thresholds, understanding the impact of changing cropping systems on farm-scale pest ecology, and developing effective insecticide resistance management practices for the stink bug complex, three-cornered alfalfa hopper, bean leaf beetle and soybean looper. Objective 4: Improve low input systems of pest control for sweet potato by evaluating the efficacy and proper use of newly registered insecticides to enhance their integration with crop rotation and other low cost control strategies.

Approach

We plan to improve tarnished plant bug control and insecticide resistance management by gaining new information on the pest¿s ecology and biology using multi-disciplinary approaches. Analytical techniques, such as stable carbon isotope analysis, will be used to determine the influence of C4 host plants, such as field corn or pigweed, on populations of tarnished plant bug adults infesting cotton fields. This information will identify sources of tarnished plant bugs that may lead to alternative control measures prior to infestations into cotton fields. Tarnished plant bug populations will be monitored for resistance to various classes of insecticides commonly used by mid-South producers. This will provide real-time information to decision makers that will allow them to adjust their control recommendations based on the type of resistance that is found in their area of the mid-South. Detoxification enzyme activity surveys will be conducted in an effort to correlate and quantify insecticide resistance levels in field populations of the tarnished plant bug. Molecular genetics techniques will be conducted on tarnished plant bug populations that could lead to assays to evaluate the extent of field resistance in tarnished plant bug populations and provide input for insect management decisions. We also plan to determine the effect of bollworm ecology (corn earworm) on resistance to pyrethroid insecticides. Analytical techniques, such as stable carbon isotope analysis and a gossypol detection technique, will be used to determine the impact of bollworm larval plant host on pyrethroid resistance levels measured in adults collected from pheromone traps. Molecular genetics tools will be used to identify candidate genes and biological pathways associated with insecticide resistance in bollworm populations. Successful identification of loci associated with insecticide resistance and the development of genetic markers for those will provide a method to obtain quantitative estimates of field evolved resistance by estimating the allele frequencies via population studies. We will also develop pest control strategies for the U.S. Mid-South¿s Early Soybean Production System by determining accurate treatment thresholds and developing effective insecticide resistance management practices for the stink bug complex and bollworm. Field studies will be conducted to evaluate treatment thresholds for stink bugs and bollworms in early season soybeans. Stink bug populations will be monitored for potential resistance to various classes of insecticides, and this effort will provide real-time information to decision makers regarding the proper use of insecticides for control of these pests. We also plan to improve low input systems of pest control for sweet potato by evaluating the efficacy and proper use of newly registered insecticides to enhance their integration with crop rotation and other low cost control strategies. Field and laboratory studies will be conducted to determine the impact of crop rotation on populations of insect pests of sweet potatoes, as well as information of insecticide efficacy and proper application techniques.

IPM Project Publications Last Five Years

Temporal and spatial distribution of *Helicoverpa zea* and *Heliothis virescens* (Lepidoptera:

noctuidae) moths in pheromone traps across agricultural landscapes in Arkansas - (Peer Reviewed Journal)
(22-Mar-11)

Major Pesticide Receptors, Detoxification Enzymes, and Transcriptional Profile in Midgut of the Tobacco Budworm, *Heliothis virescens* (Lepidoptera: Noctuidae) - (Peer Reviewed Journal)
(25-Oct-10)

Electrical Penetration Graphic Evidence of Pymetrozine Toxicity Mainly via Inhibition of Phloem Feeding in the Rice Brown Planthopper, Nilaparvata lugens (Stal) - (Peer Reviewed Journal)
(21-Oct-10)

Molecular characterization and RNA interference of three midgut aminopeptidase N isozymes from bacillus thuringiensis-susceptible and -resistant strains of sugarcane borer diatraea saccharalis - (Peer Reviewed Journal)
Yang, Y., Zhu, Y., Abel, C.A., Ottea, J., Husseneder, C., Leonard, R.B., Huang, F. 2010. Molecular characterization and RNA interference of three Midgut Aminopeptidase N Isozymes from bacillus thuringiensis-susceptible and -resistant strains of sugarcane borer, diatraea saccharalis. Insect Biochemistry and Molecular Biology. 40:592-603.

Utilization of Early Soybeans for Food and Reproduction by the Tarnished Plant Bug (Heteroptera: Miridae) in the Delta of Mississippi - (Peer Reviewed Journal)
Snodgrass, G.L., Jackson, R.E., Abel, C.A., Perera, O.P. 2010. Utilization of Early Soybeans for Food and Reproduction by the Tarnished Plant Bug (Heteroptera: Miridae) in the Delta of Mississippi. Environmental Entomology. 39(4): 111-121 DOI: 10.1603/EN09379

Oviposition and Development of the Tarnished Plant Bug (Hemiptera: miridae) on Field Maize - (Peer Reviewed Journal)
Abel, C.A., Snodgrass, G.L., Jackson, R.E., Allen, K.C. 2010. Oviposition and Development of the Tarnished Plant Bug (Hemiptera: miridae) on Field Maize. Environmental Entomology. 39(4):1085-1091 DOI: 10.1603/EN10010

Comparison of Three Insect Sampling Methods in Sweetpotato Foliage in Mississippi - (Peer Reviewed Journal)
Abel, C.A., Adams, L.C. 2010. Comparison of Three Insect Sampling Methods in Sweetpotato Foliage in Mississippi. Journal of Entomological Science. 45(2):11-128.

Spatial and Temporal Variability in Host Use by *Helicoverpa zea* as Measured by Analyses of Stable Carbon Isotope Ratios and Gossypol Residues - (Peer Reviewed Journal)
Head, G., Jackson, R.E., Adamczyk Jr, J.J., Bradley, J.R., Van Duyn, J.W., Gore, J., Hardee, D.D., Leonard, B.R., Luttrell, R.G., Ruberson, J., Mullins, W., Orth, R.G., Sivasupramaniam, S., Voth, R. 2010. Spatial and Temporal Variability in Host Use by *Helicoverpa zea* as Measured by Analyses of Stable Carbon Isotope Ratios and Gossypol Residues. Journal of Applied Ecology. 47(3):583-592.

Influence of Within-season Densities of Heliothines and Tarnished Plant Bugs on Variability in End-of-season Cotton Yield Mapping - (Peer Reviewed Journal)
Allen, K.C., Luttrell, R.G., Parker, Jr., C.D. 2009. Influence of Within-season Densities of Heliothines and Tarnished Plant Bugs on Variability in End-of-season Cotton Yield Mapping .

Journal of Cotton Science. 13:11-22

Cloning & Characterization of the Cry1Ac-binding Alkaline Phosphatase (HvALP) from *Heliothis virescens* - (Peer Reviewed Journal)
Perera, O.P., Willis, J.D., Adang, M.J., Jurat-Fuentes, J. 2009. Cloning & Characterization of the Cry1Ac-binding Alkaline Phosphatase (HvALP) from *Heliothis virescens*. Insect Biochemistry and Molecular Biology.39:294-302.

Mycotoxigenic Fungi, Mycotoxins, and Management of Rice Grains - (Peer Reviewed Journal)
Reddy, K., Reddy, C., Abbas, H.K., Abel, C.A., Muralidharan, K. 2008. Mycotoxigenic Fungi, Mycotoxins, and Management of Rice Grains. Journal of Toxicology Toxins Reviews. 27:287-317.

Resistance to Acephate in Tarnished Plant Bug (Heteroptera: Miridae) Populations in the Mississippi River Delta - (Peer Reviewed Journal)
Snodgrass, G.L., Gore, J., Abel, C.A., Jackson, R.E. 2008. Resistance to Acephate in Tarnished Plant Bug (Heteroptera: Miridae) Populations in the Mississippi River Delta. Journal of Economic Entomology.102:699-707

Predicting Field Control of Tarnished Plant Bug (Heteroptera: Miridae) Populations with Pyrethroid Insecticides by Use of Two Glass-Vial Bioassays - (Peer Reviewed Journal)
Snodgrass, G.L., Gore, J., Abel, C.A., Jackson, R.E. 2008. Predicting Field Control of Tarnished Plant Bug (Heteroptera: Miridae) Populations with Pyrethroid Insecticides by Use of Two Glass-Vial Bioassays. Southwest Entomology. 33:181-190

A Bioassay for Determining Resistance Levels in Tarnished Plant Bug Populations to Neonicotinoid Insecticides - (Peer Reviewed Journal)
Snodgrass, G.L., Abel, C.A., Jackson, R.E., Gore, J. 2008. A Bioassay for Determining Resistance Levels in Tarnished Plant Bug Populations to Neonicotinoid Insecticides. Southwestern Entomologist.33:173-180

Characteristics of Eleven Polymorphic Microsatellite Markers in the Red Imported Fire Ant, Solenopsis invicta Buren - (Peer Reviewed Journal)
Garlapati, R.B., Cross, D., Perera, O.P., Caprio, M. 2009. Characteristics of Eleven Polymorphic Microsatellite Markers in the Red Imported Fire Ant, Solenopsis invicta Buren. Molecular Ecology Notes. 9:822-824.

Role of Diapause in the Utilization of Fall and Winter Hosts by the Tarnished Plant Bug in the Mid-South - (Abstract)
Snodgrass, G.L. 2008. Role of Diapause in the Utilization of Fall and Winter Hosts by the Tarnished Plant Bug in the Mid-South. Mid-South Entomologists online journal.

Comparison of Resistance Levels in Four Population of the Rice Stem Borer, Chilo suppressalis (Lepidoptera: Pyralidae) - (Peer Reviewed Journal)
Yue, P., Cong, F., Wen, M., Li, Q., Wei, J., Xu, G., Jin, L., Zhu, Y. 2008. Comparison of Resistance Levels in Four Population of the Rice Stem Borer, Chilo suppressalis (Lepidoptera: Pyralidae). Pest Management Science. 64:308-315

Assessment of Impact of Insecticides on Anagrus nilaparvatae (Pang et Wang) (Hymenoptera: Mymanidae), an Egg Parasitoid of the Rice Planthopper, Nilaparvata lugens (Hemiptera: Delphacidae) - (Peer Reviewed Journal)

Wang, H., Yang, Y., Ya Su, J., Shen, J., Gao, C., Zhu, Y. 2008. Assessment of Impact of Insecticides on Anagrus nilaparvatae (Pang et Wang) (Hymenoptera: Mymanidae), an Egg Parasitoid of the Rice Planthopper, Nilaparvata lugens (Hemiptera: Delphacidae). Crop Protection Journal.27:514-522

Survey of Susceptibilites to Monosultap, Triazophos, Fipronil, and Abamectin in Chilo suppressalis (Lepidoptera: Crambidae) - (Peer Reviewed Journal)
He, Y., Cao, M., Gao, C., Chen, W., Huang, L., Zhou, W., Liu, X., Shen, J., Zhu, Y. 2007. Survey of Susceptibilites to Monosultap, Triazophos, Fipronil, and Abamectin in Chilo suppressalis (Lepidoptera: Crambidae). Journal of Economic Entomology.100:1854-1861.

Chararacteristics of Thirteen Polymorphic Microsatellite Markers in the Corn Earworm, Helicovepa zea (Lepidoptera : Noctuidae) - (Peer Reviewed Journal)
Perera, O.P., Blanco C.A., SchefflerB.E., and Abel C.A. 2007. Characteristics of 13 ploymorphic microsatellite makers in the corn earworm, Helicovepa zea (Lepidoptera: Noctuidae). Mol. Ecol. Notes. doi: 10.1111/j.147-8286.2007.01806.x

Differential Susceptibilities to Pyrethroids in Field Populations of Chilo suppressalis (Lepidoptera: Pyralidae) - (Peer Reviewed Journal)
Yue, P., Jin, L., Wen, M., Cong, F., Li, Q., Wei, J., Xu, G., Zhu, Y. 2007. Differential Susceptibilities to Pyrethroids in Field Populations of Chilo suppressalis (Lepidoptera: Pyralidae). Pesticide Biochem. Physiol. 89:12-19

A Laboratory Bioassay for Monitoring Resistance in Tarnished Plant Bug Populations to Neonicotinoid Insecticides - (Proceedings/Symposium)
Snodgrass, G.L. 2007. A Laboratory Bioassay for Monitoring Resistance In Tranished Plant Bug Populations to Neonicatinoid Insecticided. National Cotton Council Beltwide Cotton Conference. Available: http://www.cotton.org/beltwide.

Current Status of Insecticide Resistance in Tarnished Plant Bug Populations in the Mid-South in 2006 - (Proceedings/Symposium)
Snodgrass, G.L. 2007. Current Status of Insecticide Resistance in Tarnished Plant Bug Poplations in the Mid-South in 2006. National Cotton Council Beltwide Cotton Conference. Available: http://www.cotton.org/beltwide.

Managing Difficult to Control Tarnished Plant Bugs in Cotton - (Review Article)
Gore, J., Snodgrass, G.L., Jackson, R.E., Catchot, A. 2006. Managing Difficult to Control Tarnished Plant Bugs in Cotton. Mid America Farm Grower. September 1, 2006

Biochemical and molecular study on insecticide resistance mechanisms - (Other)
Zhu, Yu Cheng, 2006. Biochemical and molecular study on insecticide resistance mechanisms. Invited Symposium of Nanjing Agric. Univ. Sep 6, 2006.

Potential use of proteinase inhibitors and avidin for insect control and Bt resistance management - (Abstract)
Zhu, Yu Cheng, 2006. Potential use of proteinase inhibitors for insect control and Bt resistance management. Program and Abstract of The 9th International Colloquium on Invertebrate Pathology and Microbial Control and VIII International Conference on Bacillus thuringiensis. P165.

Comparative Study on Glutathione S-Transferase Activity, CDNA, and Gene Expression

Between Malathion Susceptible and Resistant Strains of the Tarnished Plant Bug, *Lygus lineolaris* - (Peer Reviewed Journal)
Zhu, Y., Snodgrass, G.L., Chen, M. 2006. Comparative Study on Glutathione S-Transferase Activity, CDNA, and Gene Expression Between Malathion Susceptible and Resistant Strains of the Tarnished Plant Bug, *Lygus lineolaris*. Journal of Pesticide Biochemistry and Physiology. 87:62-72.

Multi-State Evaluation of Bug Sampling Methods in Blooming Cotton - (Experiment Station)
Bagwell, R., Musser, F., Catchot, A., Robbins, J., Burris, E., Cook, D., Lorenz, G., Studebaker, G., Greene, J., Gore, J., Stewart, S. 2006. Multi-state evaluation of bug sampling methods in blooming cotton. Extension Publications.

Interaction of Bt Toxin Cry1ac and Proteinase Inhibitors on the Growth, Development, and Midgut Proteinase Activities of the Bollworm, *Helicoverpa zea* - (Peer Reviewed Journal)
Zhu, Y.C., Abel, C.A., Chen, M.S. 2007. Interaction of Cry1Ac toxin (Bacillus thuringiensis) and proteinase inhibitors on the growth, development, and midgut proteinase activities of the bollworm, *Helicoverpa zea*. Pesticide Biochem. Physiol. 87:39-46.

Current Status of Insecticide Resistance in Tarnished Plant Bug Populations in the Mid-South - (Proceedings/Symposium)
Snodgrass, G.L., Gore, J., Abel, C.A. 2006. Current status of insecticide resistance in tarnished plant bug populations in the mid-south. National Cotton Council Beltwide Cotton Conference.

Comparative Study on Glutathione S-Transferase Activity, Cdna, and Gene Expression Between Malathion Susceptible and Resistant Strains of the Tarnished Plant Bug, *Lygus lineolaris* - (Proceedings/Symposium)
Zhu, Y., Snodgrass, G.L., Chen, M. 2006. Comparative study on glutathione s-transferase activity, cdna, and gene expression between malathion susceptible and resistant strains of the tarnished plant bug, *Lygus lineolaris*. National Cotton Council Beltwide Cotton Conference.

Bt Resistance Project

Previous Research Project Through 2010: *Resistance Management and Injury Potential of Lepidopterous Pests to Transgenic Cottons*

Project Scientists: O. P. Perera (Lead Scientist), Carlos Blanco (left ARS), C. A. Abel (moved to different location), Ryan Jackson, Clint Allen, R. G. Luttrell, Vacant (vice Blanco)

Project Number: 6402-22000-047-00D
Project Type: Appropriated

Start Date: Oct 01, 2005
End Date: Sep 30, 2010

Objective:
Conduct fundamental research to generate baseline data on population genetic parameters to facilitate research on mechanisms of resistance to Bt proteins at the molecular level in Lepidopteran pests of cotton. Characterize the injury potential of Lepidopterous pests (esp. *Helicoverpa zea*) to new transgenic cottons to develop accurate action thresholds. Determine the effects that Bt corn (Yieldgard®) has on bollworm biology including: pupal diapause, reproduction, flight potential, physiology, and the ability of progeny to survive and damage transgenic Bt cottons (Bollgard® and Bollgard II®).

Approach:
Population genetic studies for characterizing natural populations of *H. virescens* and *H. zea* are in need of a comprehensive set of genetic markers. Well defined genetic markers will also facilitate the development of linkage maps which in turn will serve as valuable tools in studying inheritance of traits. Simple sequence repeats (SSR) or microsatellites found in genomes as well as single nucleotide polymorphisms (SNP) found in gut specific genes will be used as genetic markers for *H. virescens* and *H. zea*. Genomic DNA libraries enriched for microsatellite sequences will be used to develop SSR markers. Subtraction cDNA libraries enriched for gut specific genes will be used to identify SNP markers.

2010 Annual Report

This is the final report for this project. Significant progress was made during the five years of the project. Behavior of bollworm larvae on transgenic Bt cotton varieties was compared with those on non-Bt cotton. Differences in larval movement within plants and feeding behavior were observed and recommendations were made to consultants and growers to adopt scouting techniques dependent upon the cotton technology being evaluated. Bollworm colonies established from field collections were used to determine injury potential on Bollgard, Bollgard II, and WideStrike cotton. Bollworms significantly delayed maturity of Bollgard cotton across a range of infestation levels and intervals. Yields of Bollgard cotton declined with increased larval infestation. A significant negative relationship between cumulative numbers of white flowers infested and seed cotton yields in Bollgard cottons was also observed. The resulting model indicated a 1.69 g reduction in yield for every white flower infested by bollworms. These results will be important for refining action thresholds for bollworms on Bollgard cotton and for predicting yield reductions caused by bollworm infestations in white flowers of Bollgard cotton. For Bollgard II and WideStrike cotton, yield reductions were only observed when 100% of white flowers were infested over a period of several weeks, and this reduction was only observed in some instances. The resulting model for Bollgard II indicated a 0.77 g reduction in yield for every white flower infested by bollworms. No significant relationships were observed for cumulative numbers of white flowers infested on yields of Widestrike cotton. Results indicated that bollworms will rarely cause yield losses of Bollgard II and Widestrike cotton. Fecundity of moths was not affected by larval development on Bt corn or Bt cotton. In addition, development of bollworms on Bt corn containing Cry1Ab protein did not impact ability of the subsequent larval generation to survive on Bt cotton (Bollgard and Bollgard II). However, bollworm moths that developed as larvae on Bt field corn had reduced flight muscle performance compare to those that developed on non-Bt corn. This reduced flight muscle performance could negatively impact dispersal of bollworms that develop on Bt corn, which may influence the impact of spatial refuges on Bt resistance levels in bollworm populations. This information was transferred to extension agents, growers, and consultants through on-farm tests and through presentations at grower and consultant meetings. Methods for screening unknown, recessive alleles conferring resistance to Bt toxins were developed by collecting data on methodological, logistical, and economic aspects of second generation (F2) screening of field samples. These data served as a model for monitoring insect resistance in Bt crops. Genetic markers developed for the tobacco budworm and the bollworm were used to evaluate genetic variation in field populations in time and space. Expressed gene sequences (transcriptome) of these insects were obtained to identify single nucleotide polymorphism (SNP) markers.

Accomplishments

Effects of Cropping Systems on Oviposition Behavior of Heliothine Moths. Farm records from a 4,000 ha farm in Arkansas over a five year period were used to examine the influence of the surrounding crop structure on populations of heliothine eggs in cotton fields. Crop types within a 0.4 km buffer surrounding each cotton field were estimated. Crop types included Bt and non-Bt varieties of corn and cotton, soybean and rice. Bt cotton and corn have the potential to reduce overall populations of heliothines in an area by diminishing populations of locally produced adults. However, the scale of insect dispersal is important when determining the impact of local suppression on overall infestation potential. In this study, there was no relationship between the area of Bt crops surrounding a particular cotton field and numbers of heliothine eggs in that field for the spatial scale examined (0.4 km). This information is important in resistance management strategies to Bt transgenic crops with regards to refuge placement and local dispersal of moths.

Impact of Pyramided-toxin Bt Corn Hybrids on Bollworm Populations. Over 90% of the second generation of bollworm utilizes field corn as a host. Because this crop is a funnel for bollworm populations, the impact of corn hybrids that produce multiple Bt toxins on bollworms was examined. Pyramided-toxin corn hybrids reduced bollworm larval numbers in ears and the level of kernel damage compared to single-toxin and non-Bt corn hybrids. This reduction in ear feeding will likely also reduce mycotoxin levels in these hybrids. Information on bollworm adult emergence in large field cages also indicated that pyramided-toxin Bt corn hybrids have the potential to reduce overall bollworm populations in the near future if this technology is readily adopted by growers. This potential population reduction could dramatically lower bollworm infestation levels in cotton, which would lead to a reduction in insecticide applications targeting this pest.

Identification of a Biomarker for Resistance to Transgenic Bt Crops. Membrane bound forms of alkaline phosphatase (mALP) were demonstrated to be binding targets for Bacillus thuringiensis toxin Cry1Ac. Laboratory selected strains of tobacco budworm, resistant to Cry1Ac toxins contained reduced levels of mALP in the midgut tissues compared to susceptible strains. This reduction in ALP activity levels is an efficient biomarker to detect Cry1Ac-resistant larvae. Considering the unique resistance and cross-resistance phenotypes of the tobacco budworm strains used in this work, the alterations in mALP expression levels as an effective biomarker for resistance to Cry toxins in lepidopteran pests was proposed.

2010 Bt Resistance Project Publications

Allen, K.C., Luttrell, R.G. 2009. Spatial and Temporal Distribution of Heliothines and Tarnished Plant Bugs Across the Landscape of an Arkansas Farm. Crop Protection Journal. 28(9):722-727.

Groot, A.T., Inglis, O., Bowdridge, S., Santangelo, R., Blanco, C.A., Lopez, J., Teran Vargas, A., Gould, F., Schal, C. 2009. Geographic and Temporal Variation in Moth Chemical Communication. Evolution. 63(8):1987-2003.

Blanco, C.A., Andow, D.A., Gould, F., Abel, C.A., Sumerford, D.V., Hernandez, G., Lopez, J., Adams, L.C., Groot, A., Leonard, R., Parker, R., Payne, G., Perera, O.P., Teran-Vargas, A.P. 2009. Bacillus thuringiensis Cry1Ac Resistance Frequency in Tobacco Budworm (Lepidoptera: Noctuidae). Journal of Economic Entomology. 102:381-387

Blanco, C.A., Gould, F., Groot, A., Abel, C.A., Perera, O.P. 2010. Offspring from Sequential Matings between Bacillus thuringiensis-Resistant and Bacillus thuringiensis-Susceptible Heliothis virescens Moths (Lepidoptera: Noctuidae). Journal of Economic Entomology. 103:861-868

Head, G., Jackson, R.E., Adamczyk Jr, J.J., Bradley, J.R., Van Duyn, J.W., Gore, J., Hardee, D.D., Leonard, B.R., Luttrell, R.G., Ruberson, J., Mullins, W., Orth, R.G., Sivasupramaniam, S., Voth, R. 2010. Spatial and Temporal Variability in Host Use by Helicoverpa zea as Measured by Analyses of Stable Carbon Isotope Ratios and Gossypol Residues. Journal of Applied Ecology. 47(3):583-592.

New Research Project Through 2015: *Effect of Resistance on Insect Pest Management in Transgenic Cotton*

Project Scientists: O. P. Perera (Lead Scientist), Kerry Clint Allen, Ryan Jackson, R. G. Luttrell, Vacant (vice-Blanco)

Project Number: 6402-22000-065-00D
Project Type: Appropriated

Start Date: April 26, 2011
End Date: March 31, 2016

Objectives

Determine the impact of a changing cropping landscape on host plant ecology and insect resistance management practices for bollworm using analytical techniques. Determine gene flow and migration patterns by analyzing tobacco budworm and bollworm populations in temporal and spatial scales using genetic and/or empirical/mathematical approaches. Identify possible mechanisms of resistance to Bt toxins by profiling gene expression patterns and develop a marker based genetic linkage map.

Approach

More than 95% of the second generation bollworm within each growing season utilizes field corn as a host. Impact of corn plants expressing multiple Bt toxins on the bollworm populations will be studied by comparing historical pheromone trap data with current and future population estimates influenced by increased acreages of Bt corn expressing multiple Bt toxins. Stable carbon isotope analysis will be used to identify bollworms using corn as a host plant. Influence of local cropping landscape on bollworm populations will be studied using sentential plots of conventional and Bt corn and cotton and early maturing soybeans. Large field cages will be used to evaluate the impact of pyramided-gene Bt corn hybrid/refuge system on resistance management strategies. Expressed genes of tobacco budworm and bollworm will be identified by transcriptome sequencing, and genetic markers developed from polymorphic nucleotide regions will be used in ecological genetic studies of tobacco budworm and bollworm populations. Gene expression profiles will be used to identify biological processes involved in physiological response to ingestion of Bt toxins. Markers developed for candidate loci associated with resistance to Bt toxins will be used to estimate allele frequencies in natural populations. Genetic loci under selection will be identified using statistical methods. A genetic linkage map of the bollworm developed using polymorphic markers will be used to study inheritance of loci of interest to Bt resistance.

Bt Resistance Project Publications Last Five Years

Microsatellite variation in *Helicoverpa zea* (Boddie) populations in the southern United States **- (Peer Reviewed Journal) (31-Mar-11)**

Susceptibility of *Spodoptera frugiperda* (Lepidoptera: noctuidae) isofamilies to Cry1Ac and Cry1F proteins of Bacillus thuringiensis **- (Peer Reviewed Journal)**
Blanco, C., Portilla, M., Jurat-Fuentes, J., Sanchez, J.F., Viteri, D., Vega-Aquin, P., Teran-Vargas, A.P., Azuara-Dominguez, A., Lopez, J., Arias De Ares, R.S., Zhu, Y., Barrera, D.,

Jackson, R.E. 2010. Susceptibility of *Spodoptera frugiperda* (Lepidoptera: noctuidae) isofamilies to Cry1Ac and Cry1F proteins of Bacillus thuringiensis. Southwestern Entomologist. 35(3):409-415.

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Offspring from Sequential Matings between Bacillus thuringiensis-Resistant and Bacillus thuringiensis-Susceptible *Heliothis virescens* Moths (Lepidoptera: Noctuidae) **- (Peer Reviewed Journal)**

Blanco, C.A., Gould, F., Groot, A., Abel, C.A., Perera, O.P. 2010. Offspring from Sequential Matings between Bacillus thuringiensis-Resistant and Bacillus thuringiensis-Susceptible *Heliothis virescens* Moths (Lepidoptera: Noctuidae). Journal of Economic Entomology. 103:861-868

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Bollworm (Lepidoptera: noctuidae) behavior on transgenic cotton expressing Cry1Ac and Cry1F proteins **- (Peer Reviewed Journal)**

Jackson, R.E., Gore, J., Abel, C.A. 2010. Bollworm (Lepidoptera: noctuidae) behavior on transgenic cotton expressing Cry1Ac and Cry1F proteins. Journal of Entomological Science. 45(3):252-261.

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Identification of Two Haplotypes of Cytochrome Oxidase Sub-unit II (COII) Gene of the Formosan Subterranean Termite, Coptotermes formosanus Shiraki (Isoptera: Rhinotermitidae) in South Mississippi **- (Peer Reviewed Journal)**

Zhu, Y., Sun, J., Luo, L.L., Liu, X.F., Lee, K.C., Mallette, E.J., Abel, C.A. 2010. Identification of Two Haplotypes of Cytochrome Oxidase Sub-unit II (COII) Gene of the Formosan Subterranean Termite, Coptotermes formosanus Shiraki (Isoptera: Rhinotermitidae) in South Mississippi. Pest Management Science. 66(6):612-620.

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A Case Study of the Formosan Subterranean Termite, Coptotermes formosanus Shiraki(Isoptera: Rhinotermitidae) Transported with a Non-Cellulosic Commercial Carrier in South Mississippi **- (Peer Reviewed Journal)**

Lee, K.C., Sun, J., Zhu, Y., Mallette, E.J. 2009. A Case Study of the Formosan Subterranean Termite, Coptotermes formosanus Shiraki(Isoptera: Rhinotermitidae) Transported with a Non-Cellulosic Commercial Carrier in South Mississippi. Sociobiology. 53(3):619-630.

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Response of Tobacco Budworm (Lepidoptera: Noctuidae) to Bacillus thuringiensis Cry1Ac Incorporated into Different Insect Artificial Diets **- (Peer Reviewed Journal)**

Blanco, C.A., Gould, F., Vega-Aquino, P., Jurat-Fuentes, J., Abel, C.A., Perera, O.P. 2009. Response of Tobacco Budworm (Lepidoptera: Noctuidae) to Bacillus thuringiensis Cry1Ac Incorporated into Different Insect Artificial Diets. Journal of Economic Entomology. 102(4):1599-1606.

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Inheritance, Fitness Cost, and Mechanism of Resistance to Tebufenozide in Spodoptera exigua (Hubner) (Lepidoptera: Noctuidae) **- (Peer Reviewed Journal)**

Jia, B., Liu, X., Liu, Y., Zhu, Y., Gao, C., Shen, J. 2009. Inheritance, Fitness Cost, and Mechanism of Resistance to Tebufenozide in Spodoptera exigua (Hubner) (Lepidoptera: Noctuidae). Pest Management Science. 65(9):996-1002

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Incidence of *Heliothis virescens* on Garbanzo Varieties in Northwestern Mississippi **- (Peer Reviewed Journal)**

Blanco, C.A., Teran-Vargas, A.P., Lopez, J., Abel, C.A. 2009. Incidence of *Heliothis*

***virescens* on Garbanzo Varieties in Northwestern Mississippi. Southwestern Entomologist.34:61-67**

Monitoring of Resistance Development to Bt Cotton in Field Populations of Helicoverpa armigera (Lepidoptera: noctuidae) **- (Proceedings/Symposium)**
(02-Jan-09)

Dynamics of Imidacloprid Resistance and Cross-Resistance to Other Insecticides in the Brown Planthopper, Nilaparvata lugens (stal)(Homoptera: Delphacidae) **- (Peer Reviewed Journal)**
Wang, Y., Chen, J., Gao, C., Zhou, W., Zhu, Y., Shen, J. 2009. Dynamics of Imidacloprid Resistance and Cross-Resistance to Other Insecticides in the Brown Planthopper, Nilaparvata lugens (stal)(Homoptera: Delphacidae). Entomologia Experimentalis et Applicata. 131:20-29.

Inheritance Mode and Realized Heritability of Resistance to Imidacloprid in the Brown Planthopper, Nilaparvata lugens (Stal) (Homoptera: Delphacidae) **- (Peer Reviewed Journal)**
Wang, Y., Liu, X., Zhu, Y., Zhou, W., Shen, J. 2009. Inheritance Mode and Realized Heritability of Resistance to Imidacloprid in the Brown Planthopper, Nilaparvata lugens (Stal) (Homoptera: Delphacidae). Pest Management Science. 65:629-634.

The Phylogenetic Relationships of Introduced Aphelinus (Hymenoptera: aphelinidae), Biological Control Agents of the Russian Wheat Aphid (Homoptera: aphididae) **- (Peer Reviewed Journal)**
Zhu, Y., Fang, Q.Q. 2009. The Phylogenetic Relationships of Introduced Aphelinus (Hymenoptera: aphelinidae), Biological Control Agents of the Russian Wheat Aphid (Homoptera: aphididae). Insect Science. 16:277-286

Size and Chemical Composition of *Heliothis virescens* (Lepidoptera: Noctuidae) Spermatophores **- (Peer Reviewed Journal)**
Blanco, C. A., Rojas, M. G., Groot, A., Morales Ramos, J. A., Abel, C. A. 2009. Size and Chemical Composition of *Heliothis virescens* (Lepidoptera: Noctuidae) Spermatophores. Ann. Entomol. Soc. Am. 102(4): 629-637.

Study on Bt Susceptibility and Resistance Mechanisms in the Sugarcane Borer, Diatraea saccharalis **- (Proceedings/Symposium)**
Zhu, Y., Wu, X., Yang, Y., Ottea, J., Leonard, R.B., Abel, C.A., Huang, F. 2008. Study on Bt Susceptibility and Resistance Mechanisms in the Sugarcane Borer, Diatraea saccharalis. Society for Invertebrate Pathology Annual Meeting. 2008:52.

Susceptibility to Neonicotinoid and Other Insecticides and Potential Risk of Resistance Development in the Brown Planthopper, Nilaparvata lugens (stal) (Homoptera: Delphacidae) **- (Peer Reviewed Journal)**
Wang, Y., Chen, J., Gao, C., Zhou, W., Zhu, Y., Shen, J. 2008. Susceptibility to Neonicotinoid and Other Insecticides and Potential Risk of Resistance Development in the Brown Planthopper, Nilaparvata lugens (stal) (Homoptera: Delphacidae). Pest Management Science.64(12) 1278-1284

Bacillus thuringiensis Cry1Ac Resistance Frequency in Tobacco Budworm (Lepidoptera: Noctuidae) **- (Peer Reviewed Journal)**
Blanco, C.A., Andow, D.A., Gould, F., Abel, C.A., Sumerford, D.V., Hernandez, G., Lopez, J., Adams, L.C., Groot, A., Leonard, R., Parker, R., Payne, G., Perera, O.P., Teran-Vargas,

A.P. 2009. Bacillus thuringiensis Cry1Ac Resistance Frequency in Tobacco Budworm (Lepidoptera: Noctuidae). Journal of Economic Entomology. 102:381-387

Plant Host Effect on the Development of *Heliothis virescens* (F.) (Lepidoptera: noctuidae) **- (Peer Reviewed Journal)**
Blanco, C.A., Teran-Vargas, A.P., Abel, C.A., Portilla, M., Rojas, M.G., Morales Ramos, J.A. 2008. Plant Host Effect on the Development of *Heliothis virescens* (F.) (Lepidoptera: noctuidae). Environmental Entomology. 37(6):1538-1547.

F1 Screening for Resistance Gene Alleles to Bt Cotton in Helicoverpa armigera: How to Differentiate S and R Genotypes **- (Peer Reviewed Journal)**
Zhu, Y., Liu, F., Xu, Z., Chang, J., Abel, C.A., Shen, J. 2008. F1 screening for resistance gene alleles to bt cotton in helicoverpa armigera: how to differentiate s and r genotypes. Journal of Entomological Science. 43(3): 311-319

Yield Response of Dual-toxin Bt Cotton to *Helicoverpa zea* Infestations **- (Peer Reviewed Journal)**
Gore, J., Adamczyk Jr, J.J., Catchot, A., Jackson, R.E. 2008. Yield Response of Dual-toxin Bt Cotton to *Helicoverpa zea* Infestations. Journal of Economic Entomology. 101:1594-1599

An Empirical Test of the F2 Screen for Detection of Bacillus thuringiensis-Resistance Alleles in Tobacco Budworm (Lepidoptera: Noctuidae) **- (Peer Reviewed Journal)**
Blanco, C.A., Perera, O.P., Gould, F., Sumerford, D.V., Hernanderz, G., Abel, C.A. 2008. An Empirical Test of the F2 Screen for Detection of Bacillus thuringiensis-Resistance Alleles in Tobacco Budworm (Lepidoptera: Noctuidae). Journal of Economic Entomology. 101(4):1406-1414.

Soybean Flour and Wheat Germ Proportions in Insect Artificial Diet and Their Effect on the Growth Rates of *Heliothis virescens* (F.) (Lepidoptera: Noctuidae) **- (Peer Reviewed Journal)**
Blanco, C.A., Portilla, M., Abel, C.A., Winters, H.E., Ford, R.L., Streett, D.A. 2009. Soybean Flour and Wheat Germ Proportions in Insect Artificial Diet and Their Effect on the Growth Rates of *Heliothis virescens* (F.) (Lepidoptera: Noctuidae). Journal of Insect Science. 9:52

Buprofezin Resistance Selection, Susceptibility Survey, and Analysis of Fitness Costs in Nilaparvata lugens (Homoptera: Delphacidae) **- (Peer Reviewed Journal)**
Wang, Y., Gao, C., Shen, J., Zhang, J., Huang, Y., Li, W., Dai, D., Zhuang, Y., Zhou, W., Zhu, Y. 2008. Buprofezin Resistance Selection, Susceptibility Survey, and Analysis of Fitness Costs in Nilaparvata lugens (Homoptera: Delphacidae). Pest Management Science. 64:1050-1056

Resistance Allele Frequency to Bt Cotton in Field Population of Helicoverpa armigera (Hübner) Lepidoptera: Noctuidae) in China **- (Peer Reviewed Journal)**
Liu, F., Xu, Z., Chang, J., Chen, J., Meng, F., Zhu, Y., Shen, J. 2008. Resistance Allele Frequency to Bt Cotton in Field Population of Helicoverpa armigera (Hübner) Lepidoptera: Noctuidae) in China. Journal of Economic Entomology. 101(3):933-943

Geographic and Temporal Variation in Moth Chemical Communication **- (Peer Reviewed Journal)**
Groot, A.T., Inglis, O., Bowdridge, S., Santangelo, R., Blanco, C.A., Lopez, J., Teran

Vargas, A., Gould, F., Schal, C. 2009. Geographic and Temporal Variation in Moth Chemical Communication. Evolution. 63(8):1987-2003.

Imidacloprid Susecptibility Survey and Selection Risk Assessment in Field Populations of the Brown Planthopper, Nilaparvata lugens(Stal)(Homoptera: Delphacidae) **- (Peer Reviewed Journal)**

Wang, Y., Gao, C., Li, W., Shen, J., Zhuang, Y., Dai, D., Zhou, W., Ma, C., Yang, Y., Zhu, Y. 2008. Imidacloprid Susecptibility Survey and Selection Risk Assessment in Field Populations of the Brown Planthopper, Nilaparvata lugens(Stal)(Homoptera: Delphacidae). Journal of Economic Entomology. 101(2):515-522.

Sweet Potato Yield Reduction Caused by Reniform Nematode in the Mississippi Delta **- (Peer Reviewed Journal)**

Abel, C.A., Adams, L.C., Stetina, S.R. 2007. Sweet Potato Yield Reduction Caused by Reniform Nematode in the Mississippi Delta. Plant Health Progress. 10:1094/PHP-2007-1115-01-RS.

Regional Assessment of *Helicoverpa zea* (Lepidoptera: Noctuidae) Populations on Cotton and Non-Cotton Crop Hosts **- (Peer Reviewed Journal)**

Jackson, R.E., Bradley, J., Van Duyn, J., Leonard, B.R., Allen, C., Luttrell, R., Adamczyk Jr, J.J., Gore, J., Hardee, D.D., Voth, R., Sivasupramaniam, S., Mullins, W., Head, G. 2008. Regional Assessment of *Helicoverpa zea* (Lepidoptera: Noctuidae) Populations on Cotton and Non-Cotton Crop Hosts. Entomologia Experimentalis et Applicata. 126:89-106

Baseline Suscepitibility of the Tobacco Budworm (Lepidoptera: Noctuidae) to the CrylF Toxin from Bacillus thuringiensis **- (Peer Reviewed Journal)**

Blanco, C.A., Storer, N.P., Abel, C.A., Jackson, R.E., Leonard, R. 2007. Baseline Suscepitibility of the Tobacco Budworm (Lepidoptera: Noctuidae) to the CrylF Toxin from Bacillus thuringiensis. Journal of Economic Entomology. 101(1): 168-173

Characterization of Microsatellite Loci in the Western Tarnished Plant Bug, Lygus hesperus Knight (Hemiptera: Miridae) **- (Peer Reviewed Journal)**

Shrestha, R.B., Parajulee, M.N., Perera, O.P., Scheffler, B.E., Densmore, L.S. 2007. Characterization of Microsatellite Loci in the Western Tarnished Plant Bug, Lygus hesperus Knight (Hemiptera: Miridae). Molecular Ecology Notes. 7:1342-1344. (doi:10.1111/j.147-8286.2007.01875.x)

Monitoring Bacillus thuringiensis-Susceptibility in Insect Pests That Occur in Large Geographies: How to Get the Best Information When Two Countries are Involved **- (Peer Reviewed Journal)**

Blanco, C.A., Perera O. P., Boykin D., Abel C.A., Gore J., Matten S. R., Ramirez-Sagahon J. C., Teran-Vargas A. P. 2007. Monitoring Bacillus thuringinesis-susceptibility in insect pests that occur in large geographies: How to get the best information when two countries are involved. J. Inv. Pathol. 95:201-207

Impact of Fall Armyworm Survival in Bt Crops on Survival and Damage Potential of Subsequent Generations **- (Proceedings/Symposium)**

Jackson, R.E., Gore, J., Catchot, A. 2007. Impact of Fall Armyworm Survival in Bt Crops on Survival and Damage Potential of Subsequent Generations. In: Proceedings of National Cotton Council Beltwide Cotton Conference, January 9-12, 2007, New Orleans, Louisiana .

p. 1634-1637

Differentiating Tobacco Budworm and Corn Earworm Using Near-Infrared Spectroscopy **- (Peer Reviewed Journal)**

Jia, F., Maghirang, E.B., Dowell, F.E., Abel, C.A., Ramaswamy, S. 2007. Differentiating Tobacco Budworm and Corn Earworm Using Near-Infrared Spectroscopy. Journal of Economic Entomology. 100:759-764

Characterization of Eight Polymorphic Microsatellite Markers from the Tarnished Plant Bug, *Lygus lineolaris* (Palisot de Beauvois) **- (Peer Reviewed Journal)**

Perera O. P., Snodgras G.L., Scheffler B.E., Gore J., and Abel C. A. 2007. characterization of eight ploymorphic microsatelliet markers in the tarnished plant bug, *Lygus lineolaris* (Palisot de Beauvois). Mol. Ecol. Notes. doi: 10.1111/j.1471-8286.2007.01747.x

Paternity allocation in a mutant *Heliothis virescens* colony **- (Peer Reviewed Journal)**

Blanco, C.A., Perera, O.P., Groot, A., Hernandez, G., Teran-Vargas, A.P. 2008. Paternity Allocation in a Mutant *Heliothis virescens* Colony. Southwestern Entomologist. 33:253-263.

Cross-Resistance Responses of Cry1Ac-Selected *Heliothis virescens* (Lepidoptera: Noctuidae) to the Bacillus thuringiensis Protein Vip3A **- (Peer Reviewed Journal)**

Jackson, R.E., MarcusM.A., Gould F., Bradley, J. R. Jr., and Van Duyn J.W. 2007.cross-resistance responses of cry1Ac-selected *Heliothis virescens* (Lepidoptera: Noctuidae) to the Bacillus thuringiensis protein Vip3a. J. Econ. Entomol. 100: 180-186.

Genetic Variation for Resistance to Bacillus thuringiensis Toxins in *Helicoverpa zea* (Lepidoptera: Noctuidae) in Eastern North Carolina **- (Peer Reviewed Journal)**

Jackson, R.E., Gould, F., Bradley Jr., J.R., Van Duyn, J.W. 2006. Genetic Variation for Resistance to Bacillus thuringiensis Toxins in *Helicoverpa zea* (Lepidoptera: Noctuidae) in Eastern North Carolina. Journal of Economic Entomology. 99:1790-1797

Prediction of Masked Chafer (Coleoptera: Scarabaeidae) Capture in Light Traps Through a Degree-Day Model **- (Peer Reviewed Journal)**

Blanco, C.A., Hernandez, G. 2006. Prediction of masked chafer (coleoptera: scarabaeidae) capture in light traps through a degree-day model. Journal of Insect Science. 6:36

Alternative Tarnished Plant Bug Management

Previous Research Project Active Through 2010: *Entomopathogens and Sterile Insect Technique for Control of Sucking Insect Pests of Cotton*

Project Scientists: G. L. Snodgrass, Todd Ugine (no longer with ARS), Eric Villavaso (retired), R. G. Luttrell, Vacant (vice-Ugine)

Project Number: 6402-22000-060-00D
Project Type: Appropriated

Start Date: Jun 07, 2010 (Original Project Began in 2004)
End Date: Jul 31, 2011

Objectives

Changes in cotton insect pest management practices (i.e., Bt cotton, boll weevil eradiation, and Lepidopteran-specific insecticides) have resulted in reduced broad spectrum insecticide use, which coincidentally controlled secondary insect pests. This combined with insecticide resistance have lead to increased concerns over sucking insect pests. Three alternative management strategies will be developed to control tarnished plant bug and cotton aphid. First, entomopathogenic fungi will be developed for controlling tarnished plant bug populations developing on wild host plants. Second, sterile insect technique will be developed targeting these same tarnished plant bug populations. These first two objectives will contribute to the area wide management of tarnished plant bug. Finally, epizootics of the cotton aphid fungus, Neozygites fresenii will be augmented to enhance cotton aphid control.

Approach

Entomopathogenic fungi isolated from native tarnished plant bug populations will be selected for characteristics relevant to microbial biopesticide development. Formulations will be developed for improving persistence and efficacy of fungi. A selected formulated isolate will be incorporated into ongoing area wide management programs in wild host plants. Tarnished plant bugs will be sterilized by irradiation and fitness evaluated. Once the sterile insect technique is developed, sterile insects will be released into populations developing on wild host plants. Diapause models will be developed and used to strategically time sterile insect release. Factors leading to the initiation and spread of cotton aphid fungus epizootics will be identified. Based on these factors cultural and pest management practices will be identified to augment epizootics and improve the establishment of epizootics through inoculative biocontrol.

2010 Annual Report

Research on the project over the five-year period was affected by the retirement and the departures of a number of scientists between January 2007 and January 2009. During the time period, mass production of the NI8 isolate was greatly improved and conidia from the isolate can now be produced in amounts needed on a scale practical for research in area wide management demonstrations. Basic data needed for using the NI8 isolate were developed by an ARS scientist who determined the time for development of all the tarnished plant bug life stages at a range of temperatures (18-32ºC). This will allow us to investigate the role of temperature on fungal infection for various life stages in the laboratory which will ultimately suggest life stages to target with NI8 in the field. Detailed reproductive curves for tarnished plant bugs have also been generated at a range of temperatures of insects infected with the NI8 isolate and noninfected insects. This work showed that because *Beauveria (B.) bassiana* is slow acting, reproductive females can replace themselves many times before the fungus kills them. This makes treatment of overwintering adults (which are in diapause and nonreproductive) the best method for using the fungus to reduce plant bug populations. The best time to treat the overwintering generation is during September and October in the Mid-South when nymphs that develop on wild hosts produce diapausing adults. In order to target overwintering adults, we conducted research in 2010 to determine if diapausing adults, and nymphs that produce diapausing adults, were as susceptible to the NI8 isolate as reproductive adults and nymphs. The fungus was found to be equally effective against diapausing and reproductive plant bugs at 15, 20 and 25ºC. Field tests using the NI8 isolate to control the overwintering generation will be conducted during the fall and winter of 2010. Research on rearing tarnished plant bugs on artificial diet found that first instars had high mortality due to difficulty in feeding on the diet packets. If this problem could be corrected rearing large numbers would be much more cost effective. A mass rearing program would be needed if control measures such as the release of sterile adults were ever used in plant bug control. One field test using the NI8 isolate for tarnished plant bug control in cotton was conducted in August 2006. The results were promising in that the number of nymphs found in the NI8 treatment were 12 times lower than the number found in the check and were not significantly different from the number of nymphs found in the standard acephate treatment. Tests with NI8 in cotton for plant bug control are being conducted in 2010. The genome of the NI8 *B. Bassiana* isolate was sequenced and is currently being assembled. It will provide between 100-150 fold coverage of the 40 Mb genome. It will be a basic source of genetic information of use to many researchers that work with the fungus.

Accomplishments

Bi-phasic Mass Production of Novel *B. Bassiana* Isolate NI8. Samples from several kilograms of conidia from the NI8 isolate being held in cold storage were tested for germination and pathogenicity to tarnished plant bugs. These conidia were found to have lower germination and pathogenicity than the original NI8 isolate and were destroyed. Conidia from the original NI8 isolate were used to produce 6 kilograms of isolate. Each batch of the new conidia was tested to ensure that no contamination had occurred and the conidia were pure. All of the batches of conidia were found to be equal to the original NI8 isolate in germination and pathogenicity. These conidia are being used in field and laboratory tests in 2010.

Incorporation of the NI8 *Beauveria (B.) bassiana* Isolate into Area Wide Management. Previous research showed that tarnished plant bug nymphs which develop on wild hosts at the day lengths found in the Mid-South in September and October produce overwintering adults (in diapause). In addition, in mild winters adults overwintering on blooming henbit, Lamium amplexicaule L., break diapause during December. To reduce the overwintering population, we will begin testing the use of the NI8 isolate on wild hosts this fall. To help evaluate if this treatment will be effective, we conducted laboratory tests to determine if the NI8 isolate was as pathogenic to diapausing adults as it was to reproductive adults. Nymphs developing at a diapause-inducing day length of 10 h (which will produce diapausing adults) were also tested. The NI8 isolate was found to be equally effective against diapausing and reproductive adults and nymphs at temperatures of 15, 20, and 25ºC. At 10ºC the conidia would not germinate. During the study a few nymphs molted within 24 h of being treated with the conidia and later died and sporulated. This indicated that NI8 could infect nymphs when they were near ecdysis (shedding their exoskeleton). The cadavers of these nymphs were saved for future experiments, i.e., conidia will be isolated from the cadavers and mass produced to test and determine if they have increased virulence as compared to the stock NI8 isolate.

2010 Alternative Tarnished Plant Bug Management Publications

None

New Research Project Active Through 2015: ***Control of Tarnished Plant Bugs by Biocontrol and Other Methods***

Project Scientists: G. L. Snodgrass, R. G. Luttrell, Vacant (vice-Ugine)

Project Number: 6402-22000-064-00D
Project Type: Appropriated

Start Date: Jan 03, 2011
End Date: Jan 02, 2015

Objectives

Determine the effect of temperature and reproductive state on susceptibility of tarnished plant bugs to *Beauveria (B.) bassiana* (ARSEF 8889). Determine the effect of exposure to insect growth regulators (IGRs) and *B. Bassiana* (ARSEF 8889) on immature tarnished plant bug survival. Determine the effect of host plant and application timing (season) on susceptibility of tarnished plant bugs treated with ARSEF 8889 and IGRs (in situ).

Approach

The effect of temperature and reproductive state on the susceptibility of tarnished plant bugs to *Beauveria (B.) bassiana* (ARSEF 8889) will be determined in replicated laboratory tests. The two reproductive states tested will be normal reproductive adults and nymphs and diapausing adults and nymphs that produce diapausing adults. Temperatures tested will range from 10°C to 30°C. Insect growth regulators (IGRs) will be tested with nymphs in replicated laboratory tests to determine which IGRs are effective and the rate at which to use them. The most effective IGR(s) will be tested in laboratory tests in combination with ARSEF 8889 to determine the most effective combination treatment. Results from the laboratory tests will be tested in the field in replicated tests in cotton (for in-season plant bug control) and in the fall and winter on wild host plants (for control of the diapausing overwintering generation). The effect of IGRs and ARSEF 8889 treatment on beneficial arthropod populations will be evaluated in the field tests and with additional laboratory tests.

Alternative Tarnished Plant Bug Management Project Publications Last Five Years

Jin, X., Ugine, T. A., Chen, J., Streett, D. A. 2009. Method for determining the best hydrophilic-lipophilic balance (HLB) number of a compatible non-ionic surfactant in formulation development for areal conidia of metarhizium anisopliae (Deuteromycotina: Hyphomycetes). Biocontrol Science and Technology. 19(3): 341-347.

Leland, J.E. 2005. Characteristics of *Beauveria bassiana* isolates from *Lygus lineolaris* populations of Mississippi. Journal of Agricultural and Urban Entomology 22:57-72.

Leland, J.E., M.R. McGuire, J.A. Grace, S.T. Jaronski, M. Ulloa, Y. H. Park, and R.D. Plattner. 2005. Strain selection of a fungal entomopathogen, *Beauveria bassiana*, for control of plant bugs (*Lygus* spp.) (Heteroptera:Miridae). Biological Control 35:104-114.

Leland, J.E. and G.L. Snodgrass. 2005. Prevalence of naturally occurring *Beauveria bassiana* in *Lygus lineolaris* (Heteroptera: Miridae) populations from wild host plants of Mississippi. Journal of Agriculture and Urban Entomology 23:157-163.

Lund, J., T. G. Teague, D. C. Steinkraus, and J. Leland. 2006. Control of tarnished plant bugs using *Beauveria bassiana* and the insect growth regulator, novaluron. Summaries of Arkansas Cotton Research 2005, Arkansas Agricultural Experiment Station Research Series, pp. 249-254.

Villasvaso, E. J. 2005. A non-sticky trap for tarnished plant bug (Heteroptera: Miridae). J. Entomol. Sci. 40 (2):136-142

SIMRU's FY2010 S.T.E.P Employees

- **3rd row (L to R): Julian Beamon (Mississippi Delta Community College), Andrew Adams (Mississippi State University), John Bates (Mississippi State University), Thomas Sherman (St. Joseph Catholic School).**

- **2nd row(L to R): Chris Morris (Mississippi State University), Bailey Tubertini (East Mississippi Community College:), Kentavius Mason (Hinds Community College), Jesse King (Mississippi Valley State University), David Liang (Mississippi School for Math and Science), Shelby Reister (Washington School), Emily Mosow (Millsaps College), Michael McCain(University of Mississippi).**

- **1st row(L to R) Cavishia Roberson (Rust College), Gwendolyn Lee (Jackson State University), Ri'Carya Horton (Alcorn State University), Lakeldren Williams (Mississippi State University), Jasmine Warren (University of Mississippi), Jordan Tullos (Mississippi State University),Breanna Pennington (O'Bannon High School).**

- **Not shown pictured: Jason Greenlee (Mississippi State University), Julian Henry (Mississippi Delta Community College), Dustin Pickelmann (Delta State University), Sarah Taylor (Mississippi State University), Tashanika Knight (Delta State University), D'Anice Dishmon (O'Bannon High School), Jasmine Williams (O'Bannon High School), Corey Douglas (Delta State University), Dana May (Delta State University), Wade Cody Burns (Delta State University), Flenadia Moore (Mississippi Valley State University)**

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