TEXAS A&M AGRILIFE RESEARCH & EXTENSION

COTTON ENTOMOLOGY RESEARCH REPORT 2014

TECHNICAL REPORT 15-4

TEXAS A&M AGRILIFE RESEARCH, CRAIG NESSLER, DIRECTOR
THE TEXAS A&M SYSTEM, COLLEGE STATION, TEXAS

COTTON ENTOMOLOGY PROGRAM

RESEARCH ACTIVITY ANNUAL REPORT

2014

SUBMITTED TO:

PLAINS COTTON IMPROVEMENT COMMITTEE
PLAINS COTTON GROWERS, INC.

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FUNDING AND LOGISTICAL SUPPORT

USDA NIFA

Cotton Incorporated Core program

Cotton Incorporated State Support Committee

Texas A&M AgriLife Research

Texas A&M AgriLife Extension Service

Plains Cotton Growers, Inc.

Monsanto Company, Bayer CropScience, BASF, StollerUSA

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Introduction

Plains Cotton Growers, Inc. (PCG) has been a strong supporter of cotton insect research and extension activities in west Texas for many years. Most notably, PCG was instrumental in securing state funds for the Boll Weevil Research Facility at the Lubbock Center, and provided both financial and political support to conduct boll weevil biology and ecology research even before the boll weevil became a significant economic pest of the High Plains region. After the initial entry of the boll weevil into the eastern edge of the High Plains, PCG promoted and along with USDA-APHIS administered the boll weevil diapause suppression program involving a team effort that continued to include Texas A&M University. PCG also supported Texas Cooperative Extension (now Texas A&M AgriLife Extension Service) efforts to annually evaluate the diapause suppression program, conduct applied research trials to develop boll weevil management practices that would enhance the diapause suppression program's efforts and in the 1990s supported an annual survey of High Plains overwintering sites and grid trapping of cotton across the High Plains area. Under the strong and cooperative leadership of PCG, the boll weevil eradication program for the High Plains area progressed much more rapidly than anticipated. Now, the successful boll weevil eradication program has eliminated the boll weevil from this region for over a decade. The team effort of PCG, Texas A&M AgriLife Research and AgriLife Extension Service over many decades has resulted in a comprehensive understanding of boll weevil ecology and behavior.

With a successful boll weevil eradication program and increased adoption of the Bollgard® technology (now >70%), the cotton insect research and extension program focus has changed considerably during the last 10 years. Our current research/extension focus is on developing ecologically intensive management strategies for cotton pest management. Our research has demonstrated the need for continuing investigation of basic behavior and life patterns of insects while having a strong field-based applied research to bridge the gap between basic, problem-solving science and producer-friendly management recommendations. We have assembled a strong group of people to work as a team to examine multiple disciplines within the broad theme of Cotton IPM. We invest our considerable time and manpower resources in investigating the behavior and ecology of major cotton pests of the High Plains with the goal of developing management thresholds based on cotton production technology. Our Program has successfully leveraged research funds based on the funding provided by PCIC to support our Technician position. We are excited about and greatly value our Cotton Entomology research and extension partnerships with multidisciplinary scientists at the Texas A&M AgriLife Center, together with seasoned IPM Agents we have in the region, to continue this partnership as we challenge ourselves to deliver the best cotton insect-pest management recommendations to our Texas High Plains producers.

Texas A&M AgriLife Research & Extension Center at Lubbock

COTTON ENTOMOLOGY PROGRAM

Megha N. Parajulee, Ph.D.
Professor, Faculty Fellow, and Texas A&M Regents Fellow

PROGRAM OVERVIEW: The Cotton Entomology Program at Lubbock combines basic and applied research with strong outreach, industry, and grower partnerships to produce information to enhance the ability of the cotton industry in the Texas High Plains to mitigate cotton yield losses due to insect pests through the use of ecologically intensive integrated pest management. Selected projects of the Program are briefly highlighted in this exhibit.

COTTON FLEAHOPPER POPULATION DYNAMICS AS AFFECTED BY NITROGEN FERTILITY; HALFWAY, TEXAS

A multi-year study investigating the effects of differential nitrogen fertility on cotton fleahopper population dynamics in a typical drip-irrigation Texas High Plains cotton production system has been initiated from the 2014 growing season. Differential nitrogen fertility (0, 50, 100, 150, and 200 lbs N/acre) is being examined for its effect on cotton plant physiological parameters, thereby influencing cotton fleahopper injury potential and plant compensation.

Side-dressing variable rates of nitrogen fertilizer and resulting phenotypic variation in cotton

INVESTIGATION OF GENETICALLY MODIFIED COTTON CONFERRING INSECT TOLERANCE *(WITH MONSANTO)*

As part of an ongoing Monsanto program to develop commerically available *Lygus*-tolerant cotton germplasm, numerous cotton lines, genetically modified to confer *Lygus* tolerance via protein expression (similar to *Bt* technology), were evaluated for effectiveness under whole-plant cage and open field conditions. Other species including cotton fleahoppers and western flower thrips were also studied for their tolerance to these transgenic events. This 4-year industry-funded project completed in 2014.

Field cage evaluation of transgenic cotton events

UNDERSTANDING COTTON FLEAHOPPER OVERWINTERING EMERGENCE BIOLOGY AND POPULATION DYNAMICS

Cotton fleahoppers are minor but significant pests of cotton in the Texas High Plains. They generally overwinter in woolly croton. Dead croton twigs, containing overwintered cotton fleahopper eggs, were collected from the Brazos Valley during the winter months and stored in a walk-in cooler. Current study is evaluating the influence of amount, frequency, and method of soaking of these croton twigs on fleahopper diapause breakdown, nymphal emergence, and survivorship. We plan to develop a climatic data-based model to predict the fleahopper emergence and likely pest risk on cotton based on rainfall patterns.

DEVELOPMENT OF ECONOMIC THRESHOLD AND MANAGEMENT RECOMMENDATIONS FOR LYGUS BUG

Texas A&M AgriLife **Cotton Entomology Program** has been providing a unique leadership in *Lygus* research across the United States cottonbelt since 2002. We have quantified the compensation ability of cotton to *Lygus*-induced fruit loss and the recommendation has been made to our producers that pesticide applications prior to 30% pre-flower and 25% early flower fruit shed may not be necessary. We also have developed a late-season insecticide termination guideline for Texas High Plains cotton, according to which, insecticide intervention for *Lygus* control may not be warranted when harvestable bolls accumulate ≥350 heat units or the boll is ≥3 cm in diameter after crop cut-out. Current effort concentrates on developing economic threshold-based management recommendations for *Lygus* in Texas High Plains cotton, thereby aiming to minimize economic losses to producers. Specific objectives are to: determine the maximum potential for *Lygus* to inflict damage to cotton bolls at various boll maturity levels, characterize the cotton boll feeding biology and behavior of *Lygus*, and establish the *Lygus* economic threshold for Texas cotton.

Single-plant cages in *Lygus* economic threshold study

THRIPS MANAGEMENT IN TEXAS HIGH PLAINS COTTON: THRESHOLD DEVELOPMENT AND PRODUCT EVALUATION

Two research projects, funded by USDA NIFA and Cotton Incorporated, are investigating ecological attributes of thrips and their management recommendations in Texas High Plains cotton. Primary goals of these projects are to characterize the cotton crop response to various levels of thrips injury and to develop/validate new economic thresholds. Comparative evaluation of available thrips management products should help growers in making informed and economically sound thrips management decisions.

Greenhouse and field-cage investigation of cotton to thrips injury

EFFECT OF NITROGEN FERTILIZER ON COTTON FLEAHOPPER DAMAGE POTENTIAL AND CROP RESPONSE TO INJURY

M.N. Parajulee, A. Hakeem, R. Norman, S.C. Carroll, J.P. Bordovsky

Objective: The objective was to evaluate the effect of nitrogen fertilizer application rates on cotton fleahopper damage potential and cotton's response to fleahopper injury.

Methodology: A high-yielding FiberMax® cultivar, FM 9063B2F, was planted at a targeted rate of 54,000 seeds/acre on May 19, 2014. The experiment was a split-plot randomized block design with five nitrogen fertility rate treatments as main plot, two insect augmentation treatments as sub-plots, and five replications. The five main-plot treatments included pre-bloom side-dress applications of augmented nitrogen fertilizer rates of 0, 50, 100, 150, and 200 lbs N/acre using a soil applicator injection rig on 23 July. Pre-treatment soil samples (consisting of three 0 to 24-inch depth soil cores; each subdivided into 0 to 12-inch and 12 to 24-inch sections and bagged according to depth), were collected from each of the 25 experiment plots on July 10, 2014. Two 10-ft. sections of uniform cotton were flagged in the middle two rows of each 16-row main-plot that served as two insect treatment sub-plots. The sub-plot treatments included two cotton fleahopper augmentation levels (5 nymphs per plant vs. no fleahoppers augmented as control) applied to each of the five nitrogen rates two weeks into cotton squaring to simulate an acute infestation of cotton fleahoppers. Crop growth and fruiting patterns were monitored during the crop season and the treatment plots were harvested for lint yield and fiber analysis.

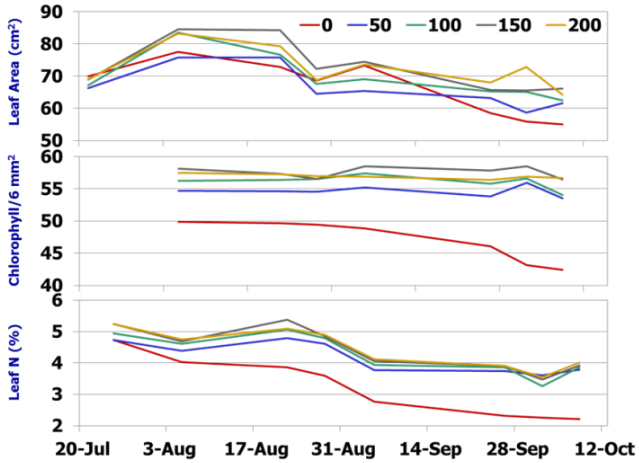

Fig. 1. Temporal dynamics of leaf growth (leaf area), chlorophyll, and % leaf nitrogen content measured on fifth mainstem leaf as influenced by the variable rates of augmented nitrogen (lb N/acre), 2014.

Results: Plant growth response varied significantly to variable rates of N (Fig. 1). Leaf size was slightly smaller in zero and 50 lb N plots, but the leaf chlorophyll in zero N plots was much lower than that for other plots throughout the growing season. The leaf chlorophyll content in zero N plots declined precipitously beginning in late August, when plants began allocating much of their resources to boll maturation, whereas this phenomenon did not occur in plots that received ≥50 lb N/acre. Percentage leaf nitrogen declined as season progressed, especially when plants began diverting its energy to fruit maturation. However, the leaf nitrogen content in zero N plots began to decline soon after cotton began flowering.

The lint yields in 50 and 100 lb N plots were significantly lower (Fig. 2) in fleahopper augmented plots (25% square loss) compared to that in control plots, suggesting that the plant response to cotton fleahopper injury is greatly influenced by nitrogen fertility.

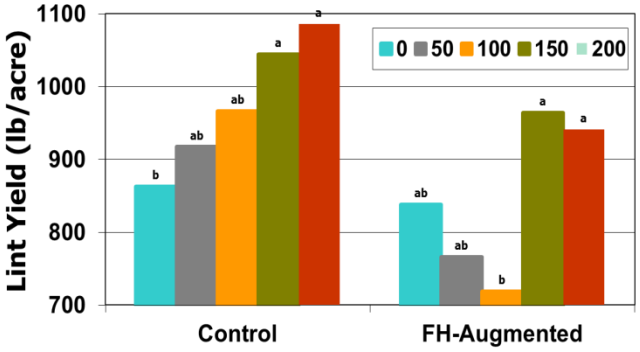

Fig. 2. Effect of nitrogen augmentation rates on lint yield following a single acute infestation of cotton fleahopper versus uninfested control, 2014.

TITLE:

Cotton yield response to cotton fleahopper acute infestations as influenced by irrigation level treatments, Lamesa, TX, 2014.

AUTHORS:

Megha Parajulee, Abdul Hakeem, Stanley Carroll, and Wayne Keeling; Professor, Research Associate, Research Scientist, and Professor, Texas A&M AgriLife Research

MATERIALS AND METHODS:

Plot Size:	4 rows by 300 feet, 3 replications
Planting Date:	May 16, 2014
Cultivar:	DP 1454 B2RF
Fertilizer:	120-40-0
Pre-plant Irrigation:	Low = 5.05 inches; High = 5.05 inches
In-season Irrigation:	Low = 3.0 inches; High = 6.0 inches
Herbicides:	Prowl® – 3 pt/A (April 14); Roundup PowerMax® – 1 qt/A + Dual® 1 pt/A (June 13); Roundup PowerMax® – 1 qt/A (July 8)
Insect Treatments:	Control (zero cotton fleahopper); Cotton fleahopper infested (5 nymphs per plant)
Insect Release Date:	July 10, 2014 (fleahopper susceptible stage)
Harvest Date:	October 20, 2014 (hand-harvested)

Cotton fleahopper feeding injury was evaluated in a high yielding cotton cultivar, DP 1454 B2RF, as affected by irrigation level. Two seasonal irrigation levels were evaluated, High (11.05") and Low (8.05"), under a center pivot irrigation system. The experiment consisted of 2 irrigation levels (high and low) and two cotton fleahopper augmentation treatments (5 fleahopper nymphs per plant versus no fleahopper augmentation as control). Each treatment plot consisted of 5 plants and the entire test was replicated three times, with a total of 12 experimental units.

Conditions conducive to cotton fleahopper emergence were simulated in a laboratory environment in order to induce hatching of overwintered eggs embedded in the woolly croton stems that were collected from the Texas Brazos Valley, and emerged cotton fleahoppers were subsequently reared using fresh green beans as a feeding substrate. A single release of nymphal cotton fleahoppers was timed to simulate the acute infestation of cotton fleahoppers while cotton was highly vulnerable to the fleahopper injury, which is approximately around the second week of cotton squaring. The cotton fleahopper release was conducted on July 10, immediately following the pre-release plant mapping, by aspirating third- to fourth-instar cotton fleahopper nymphs from the laboratory colony, transferring them into 0.75" X 1.5" plastic vials, then cautiously and methodically depositing them onto the terminals of plants in each treatment plot at the rate of 5 nymphs per plant; the control plots received no fleahoppers. There was no natural infestation of cotton fleahoppers at the experimental farm, so the control plots did not require any insecticidal intervention. Post-release data collection included plant mapping on July 17 and 25, leaf chlorophyll measurements on July 25, and a pre-harvest complete plant mapping and harvesting on October 20, 2014.

RESULTS AND DISCUSSION:

Although the crop was at a highly cotton fleahopper susceptible stage, the augmented cotton fleahopper density of 5 nymphs per plant caused much lower levels of fruit abscission than we

had anticipated. It is likely that a higher level of cotton fleahopper mortality occurred immediately after the release. It is generally expected that 20% of the released insects survive and feed on plants to cause the injury impact. Thus, we had expected 1 cotton fleahopper nymph per plant to cause the injury, which is much above the currently practiced treatment threshold. Nevertheless, it was clear that augmentation of fleahoppers caused significant injury to cotton squares and fruit abscission rates were 16% and 9% for 'Low' and 'High' water regimes, respectively (Fig. 1). It is also evident that the fleahoppers caused higher levels of injury under 'Low' water regime compared to that under a 'High" water regime, suggesting that the ability of cotton fleahoppers to inflict injury to water-stressed plants is greater than that for fully water-turgid plants or the water-stressed plants may be more susceptible to cotton fleahopper injury. Lint yield was not significantly impacted by the fleahopper augmentation treatment, but the yield was numerically lower in fleahopper augmented plots compared to that in control plots (Fig. 2). Lint yield values were 1,030 and 918 lbs per acre for 'Low' water regime and 1,638 and 1,579 lbs/acre for 'High' water regime in control and fleahopper augmented plots, respectively (Fig. 2). The effect of fleahopper on lint yield was numerically more pronounced under 'Low' water regime compared to that for 'High' water regime, indicating plants' greater ability to compensate for fleahopper-induced fruit loss under high irrigation production system.

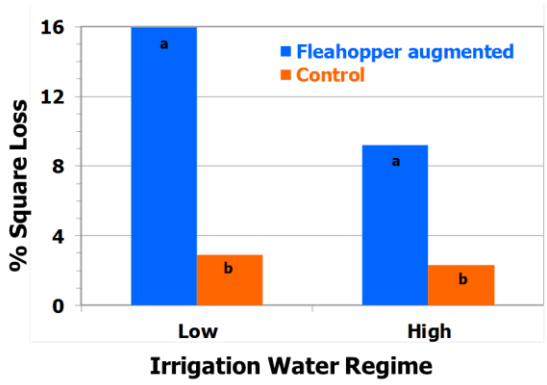

Fig. 1. Average percentage square loss following a simulated acute infestation of cotton fleahoppers, achieved by augmenting 5 nymphs per plant during the second week of squaring, under low and high irrigation regimes, Lamesa, Texas, 2014.

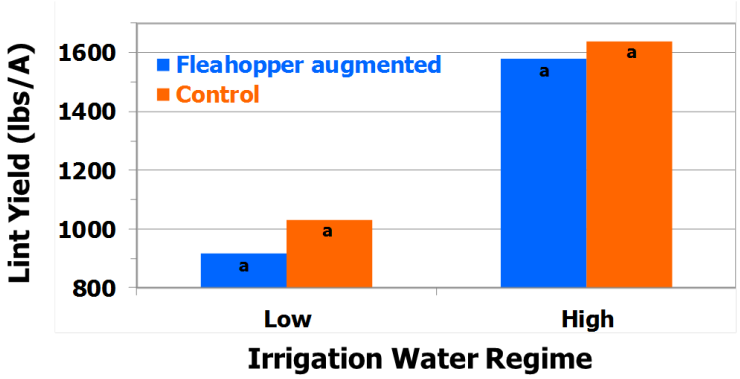

Fig. 2. Average lint yield following a simulated acute infestation of cotton fleahoppers, achieved by augmenting 5 nymphs per plant during the second week of squaring, under low and high irrigation regimes, Lamesa, Texas, 2014.

ANNUAL REPORT 2014

Cotton Incorporated Core Program

Project Number: 14-457

Evaluation of Cotton Fleahopper Damage Potential and Crop Response to Injury under Variable Nitrogen Fertility Level

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Evaluation of Cotton Fleahopper Damage Potential and Crop Response to Injury under Variable Nitrogen Fertility Level

Project Summary

The cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter), is a significant economic pest of cotton in the Texas High Plains. Injury by cotton fleahoppers to squaring cotton often causes excessive loss of small squares during the early fruiting period of plant development (first 3 weeks of squaring). Both adults and immatures feed on new growth, including small squares. Greater damage is observed on smooth leaf varieties than on hirsute varieties, which may extend the susceptible period into early bloom, especially under a high-input production regime. Cotton is affected by cotton fleahopper injury from about the fifth true-leaf through first week after initiation of flowering. Squares up to pinhead size are most susceptible to damage, and yield loss is most likely from feeding during the first three weeks of fruiting. Cotton fleahopper damage also delays crop maturity and thus increases the vulnerability of cotton to late season pests such as heliothine caterpillars and *Lygus* bugs. The objective of this study was to evaluate the cotton crop growth parameters and lint yield following cotton fleahopper acute infestations under a range of nitrogen fertility rates. The five main-plot treatments included pre-bloom side-dress applications of augmented nitrogen fertilizer rates of 0, 50, 100, 150, and 200 lbs N/acre using a soil applicator injection rig on 23 July 2014. The sub-plot treatment included two cotton fleahopper augmentation treatments (5 cotton fleahopper nymphs per plant versus no fleahopper augmentation as control) applied to each of the five nitrogen fertility rates two weeks into cotton squaring, the most critical phenological stage of cotton for cotton fleahopper management in the Texas High Plains. Cotton fleahopper infestation caused significant crop maturity delay, as measured by number of unopened bolls (non-harvestable bolls) at harvest. Averaged across all N treatments, percentage unopened bolls were 7.7% in cotton fleahopper augmented plots compared with 1.8% unopened bolls in uninfested (control) plots; N augmentation levels did not significantly influence the percentage boll opening at the time of harvest. As expected, lint yield varied with N level regardless of the cotton fleahopper infestation. In uninfested control plots, the lowest lint yield (862 lb/acre) was observed in zero N and highest lint yield (1,081 lb/acre) in 200 N treatments, with numerical increase in lint yield for each incremental nitrogen application of 50 lb/acre. However, combined over all N treatments, the acute infestation of cotton fleahoppers, with 14-27% square abscission during the third week of squaring, rendered the lint yield reduction from 975 lb/acre in the uninfested control to 846 lb/acre in fleahopper augmented treatments. In fleahopper augmented treatments, 50 and 100 lb/acre N plots had numerically lower lint yield, compared to that in uninfested control plots, whereas 150 and 200 lb/acre N plots had higher lint yield than in uninfested control plots. It is noteworthy that zero-N plots fully compensated the 27% square loss and the lint yield between control and fleahopper infested plots were similar. However, the lint yields in 50 and 100 lb/acre N plots were significantly lower in fleahopper augmented plots (25% fleahopper-induced square loss) compared to that in control plots, clearly suggesting that the plant response to cotton fleahopper injury is greatly influenced by nitrogen fertility.

Introduction

The cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter), is a significant economic pest of cotton in the Texas High Plains. Injury by cotton fleahoppers to squaring cotton often causes excessive loss of small squares during the early fruiting period of plant development (first 3 weeks of squaring). Both adults and immatures feed on new growth, including small squares. Greater damage is observed on smooth leaf varieties than on hirsute varieties, which may extend the susceptible period into early bloom, especially under a high-input production regime. Cotton is affected by cotton fleahopper injury from about the fifth true-leaf through first week after initiation of flowering. Squares up to pinhead size are most susceptible to damage, and yield loss is most likely from feeding during the first three weeks of fruiting. Cotton fleahopper damage also delays crop maturity and thus increases the vulnerability of cotton to late season pests such as heliothine caterpillars and *Lygus* bugs, particularly when natural enemies are destroyed by insecticides directed against cotton fleahoppers.

Predominantly, cotton fleahoppers feed upon pinhead-sized or smaller squares, which results in abortion of these young fruits, thereby impacting yields. While cotton fleahopper feeding preferences serve as a baseline for their management in cotton fields, a detailed understanding of cotton plant responses to fleahopper damage remains unachieved. Cotton plant growth is sensitive to numerous environmental and management input factors, particularly irrigation and nitrogen fertility. Cotton growth responses to various input factors are well-documented and growth models have been developed. However, the specific cotton plant responses to cotton fleahopper injury under a range of nitrogen fertility remain uninvestigated. This study was designed to evaluate the cotton crop growth parameters and lint yield following cotton fleahopper acute infestations under a range of nitrogen fertility rates.

Materials and Methods

This study was conducted at the Texas A&M AgriLife Research farm near Plainview, Texas. A 5-acre subsurface drip irrigation system has been in place for 12 years and nitrogen fertility treatments have been applied in a randomized block design with five replications since 2002 (Fig. 1). The present study utilized the same experimental set up for nitrogen application rates as for the last 12 years. Pre-plant land preparations on the field of 30-in row-spacings included an application and incorporation of Treflan® (trifluralin) @ 2 pints/acre on 19 February 2014. The field did not receive pre-plant fertility applications. Prior to planting (1 January to 19 May), the study field had received a total of only 0.52 inches of rain. On 19 May the field was planted to FiberMax 9063 B2F at a targeted rate of 60,000 seeds/acre followed by an 'over-the-top' Caparol 4L® (prometryn) @ 3 pints/acre) application immediately after planting. Following a 24-25 May rain event (2.50 inches), the soil surface was treated with a 'rotary hoe' implement on 28 May to combat plant damaging blowing sands. The weather patterns during much of this growing season had been highly unpredictable and erratic, which was characterized as unusually cool and wet. The study site received 2.94 inches of rain, accompanied with high damaging winds, on a single rain event of June 6-8 weekend, which was also associated with a hailstorm. The plant stand was so severely damaged by the hailstorm that the field was replanted with FiberMax 9063 B2R (54,000 seeds/acre) on 16 June 2014, which is considered a very late planting for the study site.

The second planting resulted in a very good plant stand that received numerous additional rains. Plant growth was slower than normal but fruiting started the third week of July which coincided

with the applications of the various nitrogen augmentation treatments. The second cotton planting received one cultivation on 30 June plus additional herbicide treatments on 27 June (Crop Smart® @ 32 oz/acre; Warrant® @ 3 pints/acre) and 7 July (Crop Smart® @ 40 oz/acre). Since the time of the first planting, the study site received a total of 11.49 inches of rain in less than two months, with the accompanying cool weather hampering the study's cotton crop plant growth rate. Although summer plant development was slower than normal due to numerous cloudy and cool rain events, all twenty-five of the experimental treatment plots developed into full uniform plant stands during this growing season.

Experimental plots were 16 rows wide x 120 ft long and 5 ft alleys separated the plots. The experiment was a split-plot randomized block design with five nitrogen fertility rate treatments as main plot, two insect augmentation treatments as sub-plots, and five replications. The five main-plot treatments included pre-bloom side-dress applications of augmented nitrogen fertilizer rates of 0, 50, 100, 150, and 200 lbs N/acre using a soil applicator injection rig on 23 July. The individual plots have been receiving the same nitrogen augmentation rates for the past 12 years. 2014 pre-treatment residual nitrogen soil samples were pulled on 10 July from each of the 25 experimental plots. The soil samples were quickly placed into an unused greenhouse to quickly remove the soil moisture. These dried samples were processed through a soil grinder prior to shipment to Ward Laboratories (Kearney, NE) for residual nitrogen analyses. Two 10-ft. sections of uniform cotton were flagged in the middle two rows of each 16-row main-plot that served as two insect treatment sub-plots. The sub-plot treatment included two cotton fleahopper augmentation treatments (5 cotton fleahopper nymphs per plant versus no fleahopper augmentation as control) applied to each of the five nitrogen fertility rates two weeks into cotton squaring, the most critical phenological stage of cotton for cotton fleahopper management in the Texas High Plains, in these designated row sections to simulate an acute infestation of cotton fleahoppers.

Woolly croton was harvested from rangeland sites near College Station, Texas, in early February and then placed into cold storage. Forty 1-gallon sheet metal cans, each containing 4 ounces of dry croton twigs per can, were initiated to generate the required number of cotton fleahopper nymphs for the experiment. Conditions conducive to cotton fleahopper emergence were simulated in a laboratory environment in order to induce hatching of overwintered eggs embedded in the croton stems, and emerged cotton fleahoppers were subsequently reared on fresh green beans. The single release of nymphal cotton fleahoppers mentioned above was timed to simulate the acute heavy infestation of cotton fleahoppers (4-5 days of feeding) while cotton was highly vulnerable to the fleahopper injury. It was planned so that this arrangement would ensure significantly high levels of fleahopper-induced square damage on treatment plots to quantify the variation in damage potential as influenced by soil applied N. The release was accomplished on 30 July by aspirating third-instar fleahopper nymphs from the laboratory colony, transferring them into 0.75" X 1.5" plastic vials, then cautiously depositing them onto the terminals of plants in each treatment plot at the rate of 5 nymphs per plant; the control plots received no fleahoppers and were kept fleahopper-free during the entire study period. Because natural infestations of cotton fleahoppers did not occur at our site due to the severe crop delay, the control sections within each of the 25 plots actually did not receive supplemental insecticidal interventions until an Orthene® 97 insecticide application was applied on 7 August to all experimental units (both fleahopper release sections and control sections within each of the 25 main-plots) to ensure complete removal of all cotton fleahoppers following their release and feeding period. All control and fleahopper-augmented sections were monitored for fleahopper-

induced fruit loss on 14 August. The entire test was kept insect-free for the remainder of the study to isolate the effect of cotton fleahopper injury only.

Additional data collected included monitoring of plant height, leaf chlorophyll content, leaf nitrogen content, and squaring patterns in all 50 experimental units (5 N rates x 2 insect treatments x 5 replications), starting from the first week of squaring (pre-release data) and approximately weekly thereafter well into the fall crop developmental period. The dates in which ten 5th main stem leaves (from the plant top) were collected for chlorophyll readings, leaf area measurements, leaf dry weights, and end-of-study laboratory leaf nitrogen analysis included 25 July; 5, 22, and 28 August; 5 and 26 September; and 2 and 8 October. In-season plant mapping and plant height data from five randomly selected plants per plot were collected on 5 and 26 August. On 26 August, five randomly selected plants in each of the 25 experimental plots (125 total plants) were dug-up and returned to the laboratory for measurement of detailed individual plant biomass of the following: 1) root, 2) shoot, 3) leaves, and 4) fruits. Later on 26 September, 15 randomly selected bolls were collected from the 5th mainstem node from the top of the plants and then the 375 total bolls (15 bolls per plot X 25 plots) were placed into an ice chest and returned to the laboratory to measure boll parameters including: 1) boll diameter, 2) boll fresh weight, 3) boll carpel wall puncture pressure, and 4) boll dry weight following placement into a drying oven.

The timing of crop 'cut-out' within individual plots was estimated by counting the Nodes Above White Flower (NAWF) on a series of randomly selected plants per plot on 28 August; and 5 and 19 September. The 19 September inspection results indicated that most of the plants of all fertility treatment plots had reached the physiological 'cut-out' developmental stage. The entire test was prepared for harvest by first spraying a boll opener (Boll Buster® 1 quart per acre) and a defoliant [ET® (pyraflufen) 1.25 oz per acre] in a tank mix on 23 October, followed by an application of a desiccant (Helmquat® 3SL 1 quart per acre) as a boll opener on 3 November. Final plant mapping and harvesting of test sections were performed on 20 November and the ginned lint samples were sent to Cotton Incorporated for fiber quality analysis.

0	50	200	50	200
100	100	0	100	50
200	150	50	150	0
50	200	100	200	100
150	0	150	0	150

Figure 1. Helms Farm nitrogen study experimental plot layout following a five-treatment x five-replication randomized block design. Each of the 25 plots received one of the five nitrogen augmentation treatments including 0, 50, 100, 150, or 200 lbs N/acre, Hale County, TX.

Results and Discussion

Influence of N fertility level on cotton plant growth parameters. Soil residual N levels were significantly higher in plots that received the two highest application rates of N fertilizer versus plots receiving 50 lb/acre N applications or no N augmentation; plots that received 100 lb N/acre had an intermediate level of residual nitrogen (Fig. 2). The two highest N augmentation plots (150 and 200 lb/acre) resulted in three-times higher amount of soil residual N compared to that in zero and 50 lb/acre plots. These plots had been receiving same densities of applied N for the previous 12 years and the relationship between applied N rates and resulting residual N has generally followed this trend for all previous years. Variation in residual N did not show significant variable effect on early cotton growth parameters, such as plant height, leaf area, and chlorophyll content. However, the effect of N application rate was more pronounced as season progressed (Fig. 3). Also, the effect of N application rate was less pronounced in leaf surface area compared to that for chlorophyll concentration and leaf N content of the fifth mainstem node leaf. Measured leaf chlorophyll content varied with nitrogen application level, and leaf chlorophyll contents from cotton in those plots which received 0 lb N/acre or 50 lb N/acre were significantly lower than all others (Fig. 3). Chlorophyll concentration in zero N plots was 5 or more units lower than that for 50 lb N/acre plots throughout the growing season, while the concentration further declined as the season progressed. Cotton in plots which received the three highest nitrogen application rates (100, 150, and 200 lb N/acre) exhibited relatively consistent leaf chlorophyll readings and the values in these three N rates did not significantly vary (Fig. 3). It is noteworthy that the leaf chlorophyll content in zero N treatment plots declined precipitously beginning in late August, when plants began allocating much of their resources to boll maturation, whereas this phenomenon did not occur in plots that received ≥50 lb N/acre. Percentage leaf nitrogen declined as season progressed, especially when plants began diverting its energy to fruit maturation (mid- to late August). However, the leaf nitrogen content in zero N plots began to decline soon after cotton began flowering, but it declined much more rapidly in zero N plots than for N augmented plots when plants began allocating much of their resources to boll maturation (Fig. 3).

Plant parameter values such as plant height, leaf area (leaf size), leaf chlorophyll concentration, and percentage leaf nitrogen were much lower in zero N plots compared to that in all N augmented plots by the time crop attained full maturity (Figs. 3-5), indicating a high degree of physiological stress on plants receiving zero pounds of augmented nitrogen. Lower rates of N augmentation resulted in lower plant parameter values compared to that for high rates of N augmentation.

Variable rates of N augmentation affecting plant height, leaf size, leaf chlorophyll, and leaf nitrogen content correspondingly impacted leaf dry weight and boll dry weight at full crop maturity. Fifth mainstem leaf dry weight was significantly lower at zero N plots (Fig. 6). Leaf dry weight values were similar in all N augmented plots although the two lower N augmented treatments (50 and 100 lb/acre) had numerically lower leaf dry weight compared to that for two highest N rates. Nitrogen fertility level also influenced boll maturity. Plants in zero N plots advanced to reproductive phase earlier and bolls formed and matured significantly earlier than in N augmented plots. As a result, dry weight of fifth mainstem node bolls was significantly greater in zero N plots compared to that for N augmented plots (Fig. 6). Laboratory measurement of boll exocarp penetrability showed that the fifth mainstem node bolls from zero N augmented plots

required significantly greater pressure to puncture the exocarp versus that required to do so for bolls from N augmented plots (Fig. 7).

Variation in soil residual N levels, coupled with variable N application, resulted in phenotypic expression of nitrogen deficiency in cotton across treatment plots, more pronouncedly between zero N plots and N augmented plots, which were reflected on temporal chlorophyll contents of the fifth leaf (Fig. 3).

N fertility level and cotton fleahopper infestation. Cotton plants were two weeks into squaring when an acute infestation of 5 cotton fleahopper nymphs per plant was deployed on 30 July. Pre-release monitoring of squaring profiles showed that plants had ~6 squares per plant across all N treatments. Total square density did not vary with N treatments prior to cotton fleahopper infestation (Fig. 8). This density is considered equivalent of 1 cotton fleahopper nymph per plant, with 20% field survivorship and visual observation retrieval of released nymphs. The density is also equivalent to 3-4 times current cotton fleahopper threshold (25-30 cotton fleahoppers per 100 plants) for the Texas High Plains.

One week of cotton fleahopper infestation resulted in significant square abscission in cotton fleahopper augmented plots, but negligible square abscission (2% or less) was observed in uninfested control plots (Fig. 8). While total square density did not vary across N treatments, cotton fleahopper-induced square abscission level varied significantly with N application rates. In general, higher N rate favored lesser impact of cotton fleahopper injury. Square abscission rate was numerically highest at zero N plots, followed numerically by 50 and 100 lb N/acre plots, yet all values were statistically similar. However, abscission rates were reduced to 19 and 14% in 150 and 200 N treatments, respectively (Fig. 8). These data suggest that the application of excessive N may make cotton plants less susceptible to cotton fleahopper injury. However, additional research is required to ascertain this observation. No biological or physiological reasons are speculated for reduced square abscission observed in the two highest N rate plots.

Cotton fleahopper infestation caused significant crop maturity delay, as measured by number of unopened bolls (non-harvestable bolls) at harvest. Averaged across all N treatments, percentage unopened bolls were 7.7% in cotton fleahopper augmented plots compared with 1.8% unopened bolls in uninfested (control) plots; N augmentation levels did not significantly influence the percentage boll opening at the time of harvest (Fig. 9). Nevertheless, because the level of square abscission was not excessive (14-27%) for pre-flower cotton (75% fruit set is considered a lower limit for Texas High Plains cotton into the third week of squaring), the crop did not suffer a major crop maturity delay due to cotton fleahopper infestation.

As expected, lint yield varied with N level regardless of the cotton fleahopper infestation. In uninfested control plots, the lowest lint yield (862 lb/acre) was observed in zero N and highest lint yield (1,081 lb/acre) in 200 N treatments, with numerical increase in lint yield for each incremental nitrogen application of 50 lb/acre (Fig. 10). However, combined over all N treatments, acute infestation of cotton fleahopper, with 14-27% square abscission during the third week of squaring, rendered the lint yield reduction from 975 lb/acre in uninfested control to 846 lb/acre in fleahopper augmented treatments (Fig. 10.). In fleahopper augmented treatments, 50 and 100 lb/acre N plots had numerically lower lint yield, compared to that in uninfested control plots, whereas 150 and 200 lb/acre N plots had higher lint yield than in uninfested control plots. It is noteworthy that zero-N plots fully compensated the 27% square loss and the lint yield between control and fleahopper infested plots were similar (Fig. 11). However, the lint yields in

50 and 100 lb/acre N plots were significantly lower in fleahopper augmented plots (25% fleahopper-induced square loss, Fig. 8) compared to that in control plots, clearly suggesting that the plant response to cotton fleahopper injury is greatly influenced by nitrogen fertility.

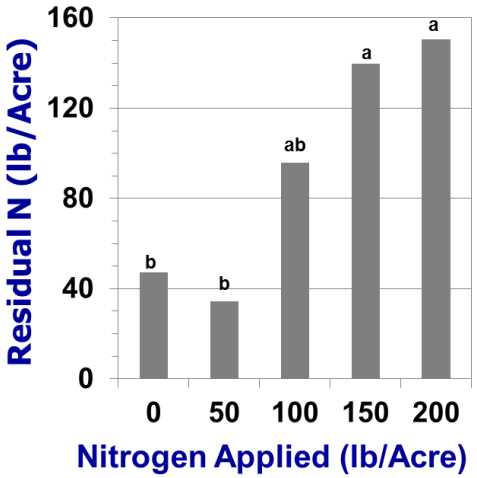

Figure 2. Effect of prior years' N application (0, 50, 100, 150, and 200 lb per acre) on residual N accumulation for the current crop year, 2014.

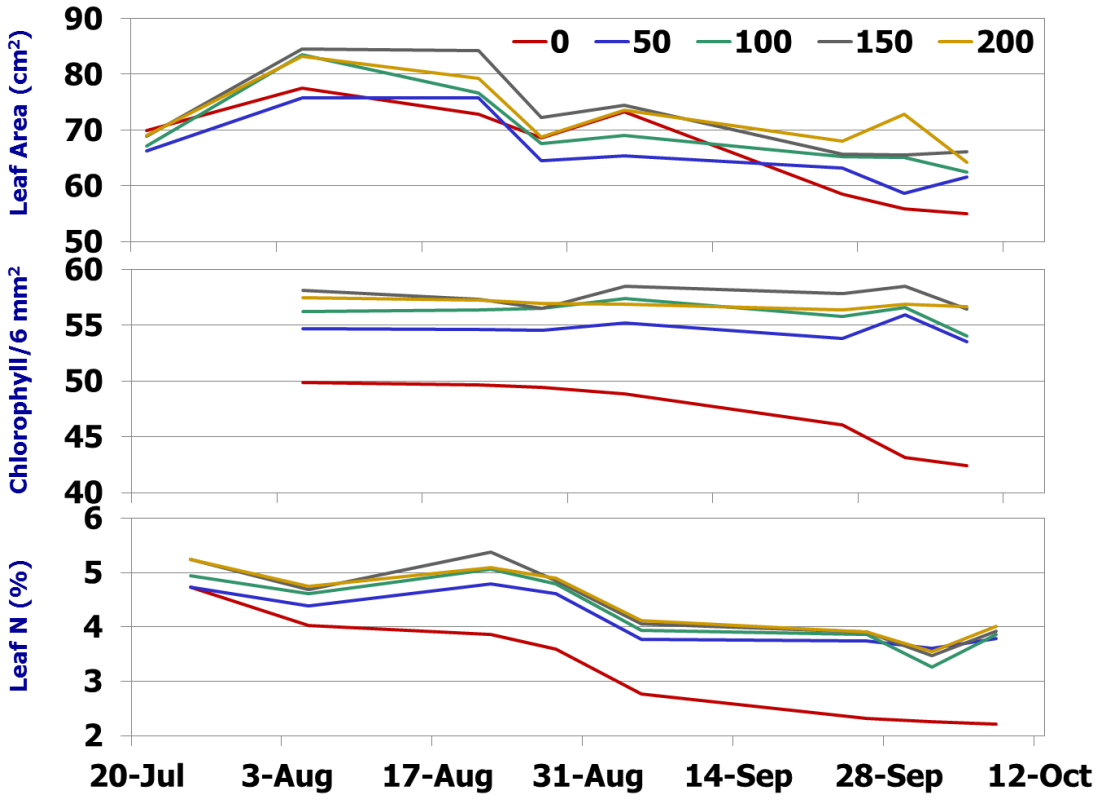

Figure 3. Temporal dynamics of leaf growth (leaf area), chlorophyll concentration, and percentage leaf nitrogen content measured on fifth mainstem leaf as influenced by the variable rates of augmented nitrogen (lb N/acre), 2014.

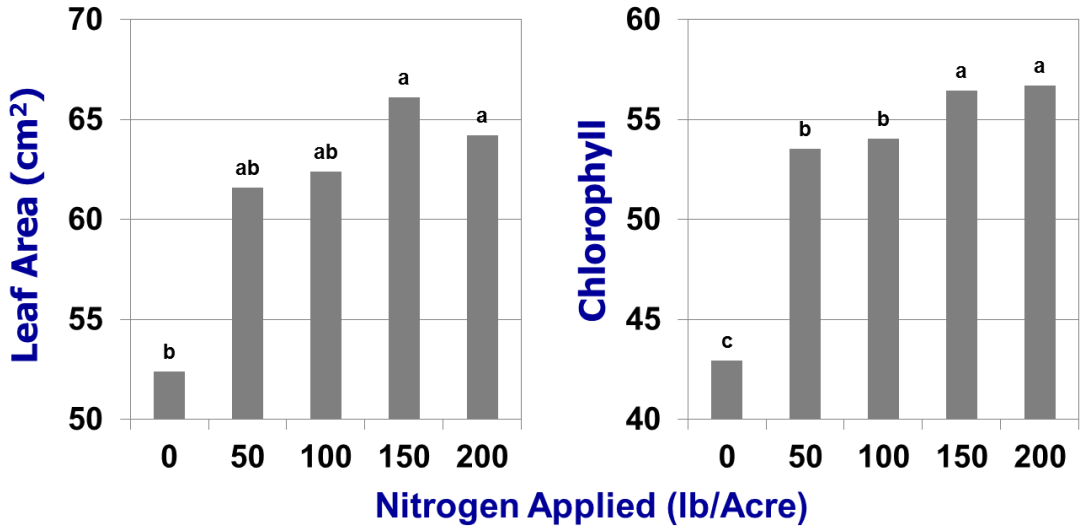

Figure 4. Average leaf surface area (left) and chlorophyll concentration or SPAD values (right) of the fifth mainstem node leaf on a full-grown crop as affected by N treatments, September 26, 2014.

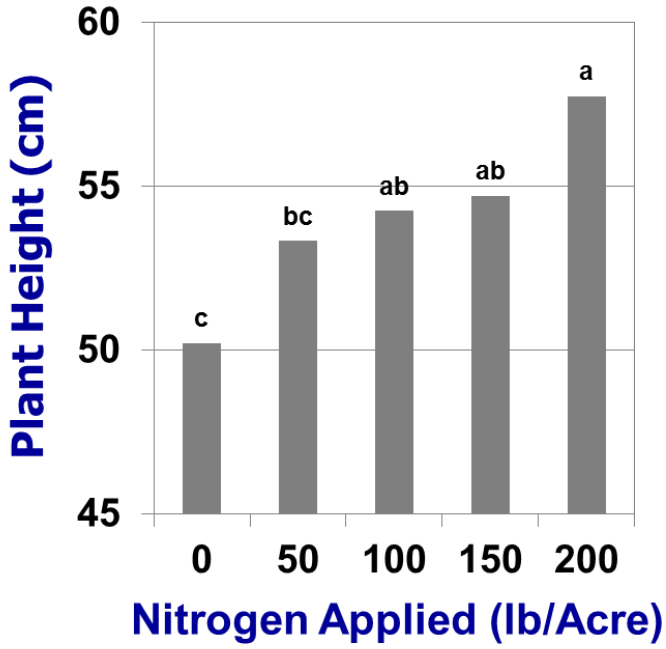

Figure 5. Effect of variable nitrogen treatments on cotton plant height at full crop maturity, September 26, 2014.

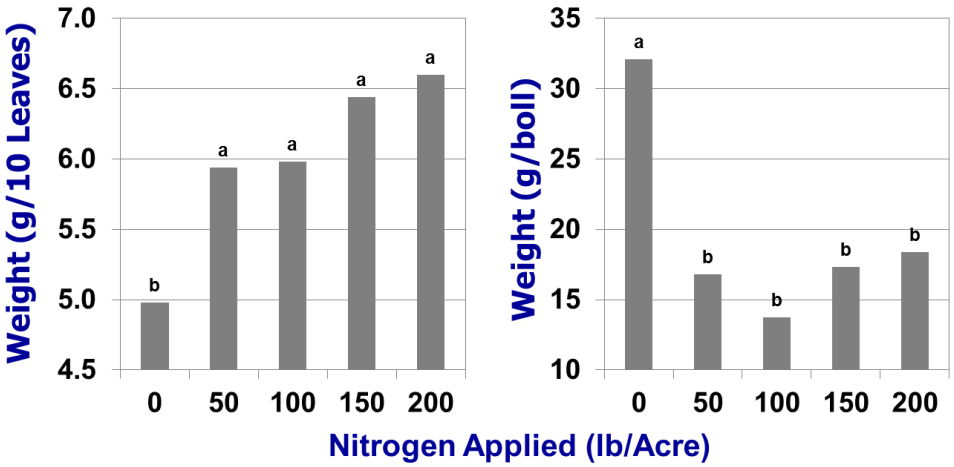

Figure 6. Effect of variable nitrogen on fifth mainstem leaf dry weight and fifth mainstem node boll dry weight at full crop maturity, September 26, 2014.

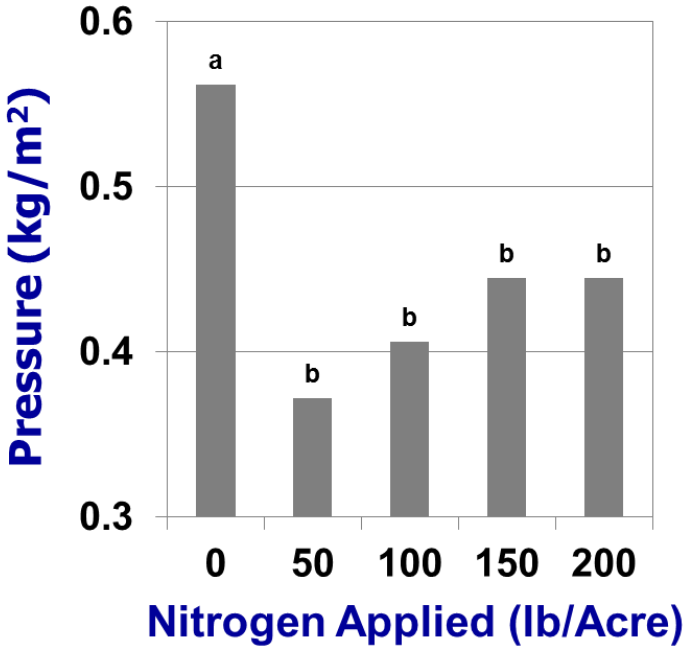

Figure 7. Effect of variable nitrogen on boll maturity as measured by the pressure required to puncture the carpel wall of the fifth mainstem node position bolls, September 26, 2014.

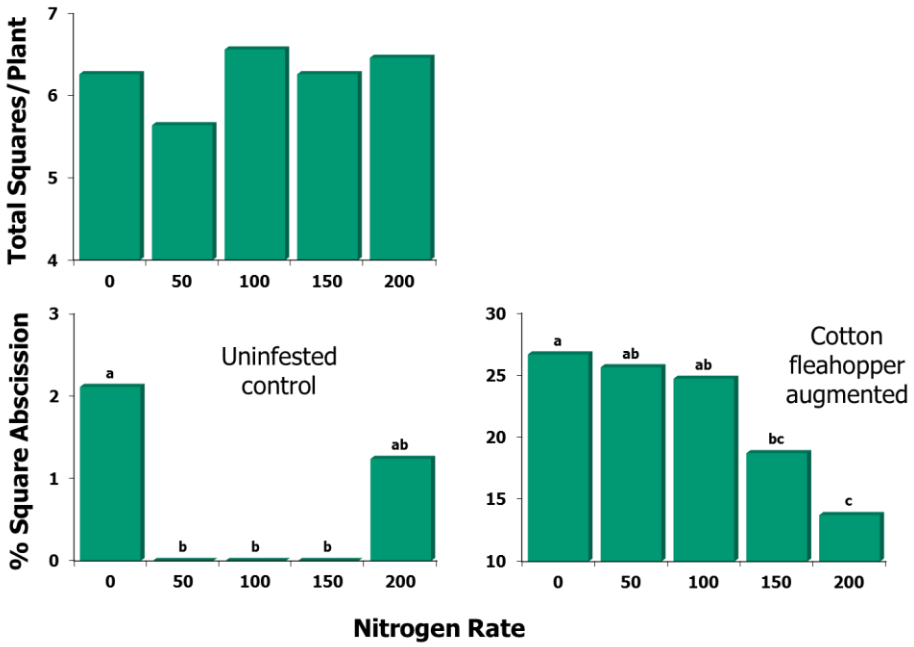

Figure 8. Total square density (number of squares set per plant) at the time of cotton fleahopper augmentation (top panel) and percentage square abscission (bottom panel) in control versus cotton fleahopper augmented treatments, as influenced by variable rates of nitrogen application (0, 50, 100, 150, and 200 lb per acre), 2014, Hale County, TX.

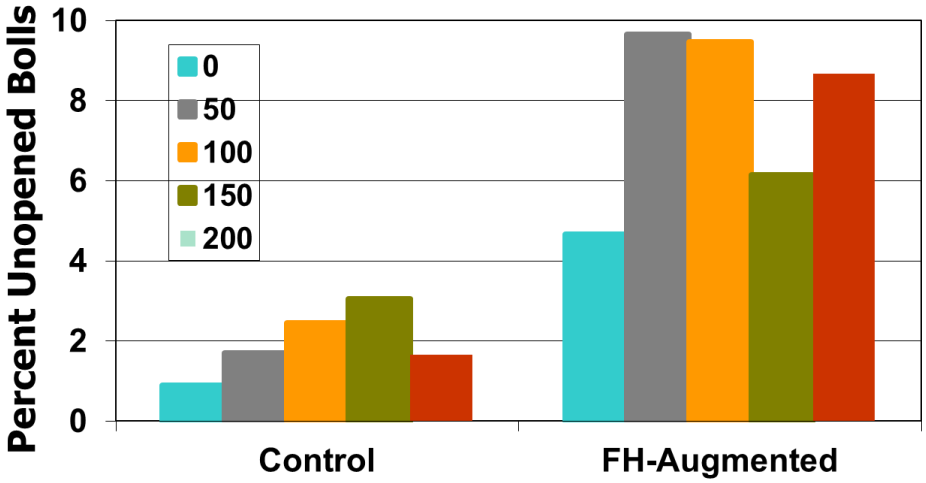

Figure 9. Effect of nitrogen augmentation rates (0, 50, 100, 150, and 200 lb per acre) on cotton maturity as measured by number of unopened (non-harvestable) bolls at harvest, November 20, 2014, Hale County, TX.

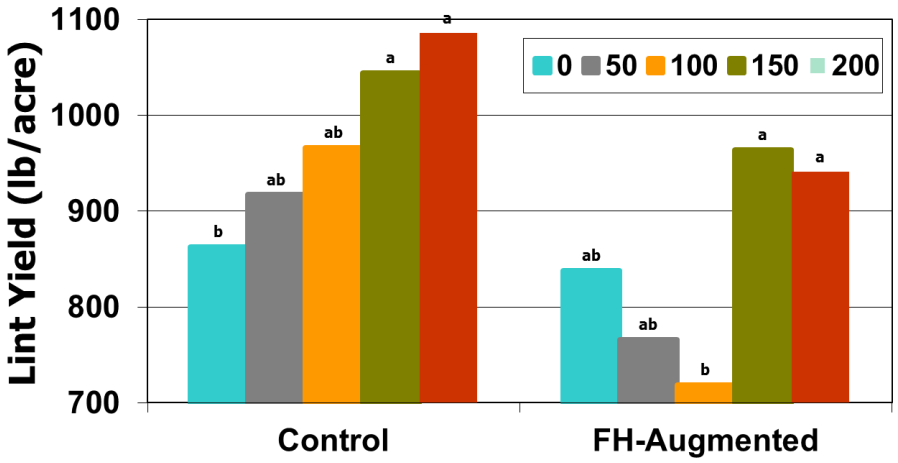

Figure 10. Effect of nitrogen augmentation rates (0, 50, 100, 150, and 200 lb per acre) on lint yield following a single acute infestation of cotton fleahopper versus uninfested control, 2014, Hale County, TX.

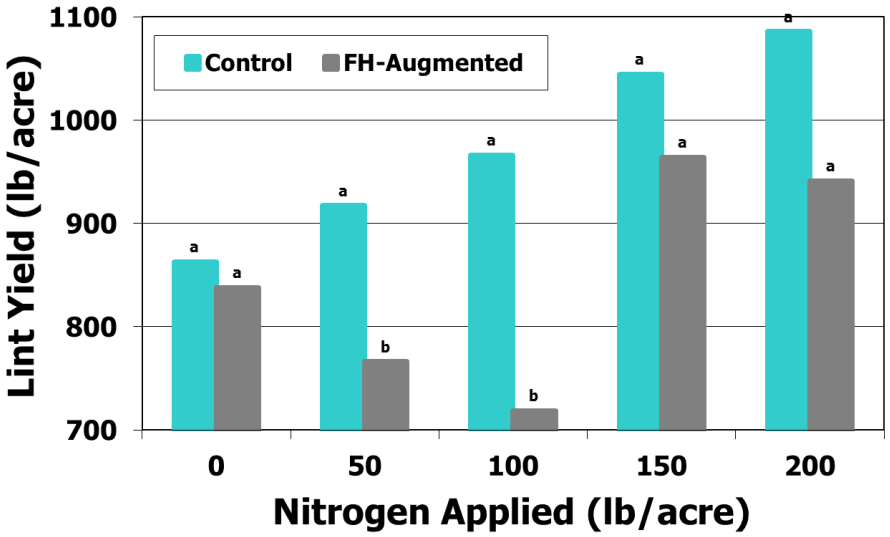

Figure 11. Effect of nitrogen augmentation rates (0, 50, 100, 150, and 200 lb per acre) on cotton lint yield following a single acute infestation of cotton fleahopper versus uninfested control, 2014, Hale County, TX.

Acknowledgments

Cotton Incorporated Core Program and Plains Cotton Growers, Inc. provided funding for this study.

2014 ANNUAL REPORT (YEAR 3)

Cotton Incorporated/Texas State Support Program

Project Number: 12-112TX

Development of Economic Threshold and Management Recommendations for *Lygus* in Texas High Plains Cotton

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Development of Economic Threshold and Management Recommendations for *Lygus* in Texas High Plains Cotton

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PROJECT SUMMARY

Western tarnished plant bug, *Lygus hesperus,* is the primary *Lygus* species inhabiting cotton and several other crop hosts in the Texas High Plains. In Texas High Plains cotton, *Lygus* is generally more pestiferous in the boll development stage than in early squaring stage. Our recent study on boll damage assessment based on heat unit-delineated maturity provided a boll-safe cutoff value of 350 heat units (~2-3 weeks from flowering), although *Lygus* adults and nymphs both cause external lesions on bolls throughout boll development and may give farmers a false impression of *Lygus* damage. A four-year State Support funded project revealed that late-instar nymphs caused significantly more damage to maturing bolls than adults, and inflicted 23, 29, and 15% more loss in lint yield, seed weight, and seed counts per boll, respectively, versus adults. Nevertheless, no economic threshold for *Lygus* boll management has been developed for Texas cotton. This project aims to conduct a comprehensive threshold study for *Lygus* in Texas cotton.
The major goal of this project was to develop economic threshold-based management recommendations for *Lygus* in Texas High Plains cotton, thereby aiming to minimize economic losses to producers. Specific objectives were to: 1) determine the maximum potential for *Lygus* to inflict damage to cotton bolls at various boll maturity levels (ages), 2) characterize the cotton boll preference behavior of *Lygus*, and 3) establish the *Lygus* economic threshold for Texas cotton. Boll damage potential of *Lygus hesperus* was determined in a no-choice cup-cage study. Ten cohorts of cup-caged single bolls (1-20 days old) were each exposed to a *Lygus* adult for 48 hours and the boll damages were quantified. After bolls reached 13 days of age, *Lygus* caused very little seed damage, which as expected, also did not result in significant lint yield loss. Cotton bolls were safe from *Lygus* damage when they reached >28 mm diameter or their carpel wall hardness was 0.7 lb per square foot or greater. Cotton boll feeding preferences of *Lygus hesperus*, within-plant boll distribution profile, and *Lygus* damage to cotton bolls at various *Lygus* densities were determined in a whole-plant cage field study. Individually caged cotton plants were exposed to 4 levels of *Lygus* (0, 1, 2 and 4 or 6 adults per cage) for one week when plants were at two selected boll development stages (350 and 550 HU after first flower). When the crop matured from 350 HU to 550 HU after first flower, the percentage of bolls vulnerable to *Lygus* feeding damage was reduced from 50% to 30%. Internal warts were mostly limited to the bolls measuring <35 mm in diameter. In this open-choice boll feeding situation, *Lygus* preferred to feed on bolls that were 10-30 mm in diameter. Averaged over three years, artificial augmentation of 1, 2, 4, and 6 *Lygus* per plant at 350 HU after first flower reduced the cotton lint yield by 137, 313, 422, and 516 lb/acre, respectively, whereas the yield reduction values for the same *Lygus* densities were 66, 191, 213, and 415 lb/acre during the late season (550 HU from first flower). Thus, the *Lygus* yield reduction potential decreased by 52, 39, 50, and 20% for 1, 2, 4, and 6 *Lygus* per plant infestation when cotton matured from 350 HU to 550 HU. A detailed understanding of *Lygus* boll feeding biology and behavior will be highly valuable in improving *Lygus* management decisions during the different boll developmental stages. A detailed understanding of *Lygus* boll feeding biology and behavior will be highly valuable in improving *Lygus* management decisions during the different boll developmental stages.

Introduction

Cotton, *Gossypium hirsutum* L., is a major cash crop in the U.S. and worldwide. The U.S. is the world's third largest cotton producer and the U.S. cotton industry is valued at more than 25 billion dollars per year. In Texas, approximately six million acres of cotton have been planted annually in recent years, and Texas is the largest cotton producing state (Williams 2013). *Lygus hesperus* is an important economic pest of cotton in some regions of the United States and it is an emerging pest of Texas High Plains cotton. In 2012, a 2.04% reduction in U.S. cotton yields was attributable to arthropod pests – 0.7% due to *Lygus* species, which was ranked top among other yield-reducing pests (Williams 2013) and also cost more per infested acre because multiple applications were often required. In Texas, over 2 million acres of cotton were infested by *Lygus* in 2012 (Williams 2013). *Lygus* can cause severe cotton square loss, anther damage, and seed damage depending upon the crop growth stage the infestation occurs. Both adult and nymphal stages of *Lygus* can inflict damage to cotton fruiting structures. *Lygus* late-instar nymphs are capable of inflicting greater internal damage to maturing bolls than are adults, and this was especially true for 1-2 week old (150-250 HU) bolls (Jubb and Carruth 1971, Parajulee *et al.* 2011). In the Texas High Plains region, *Lygus* generally infest cotton fields during the latter part of the cropping season, thus causing mostly damage to the cotton bolls. Following the introduction of *Bt*-technology (Bollgard cotton), outbreaks of lepidopteran pests have been drastically reduced, and in recent years, secondary piercing-sucking pests such as *Lygus* are of increasing concern to Texas High Plains producers (Parajulee *et al.* 2008).

Cotton boll profiles change as crop matures, and as a result, the number of *Lygus* susceptible and/or tolerant bolls to *Lygus* damage also change. As boll maturity profiles change, *Lygus* boll selection and feeding behavior may also change which can result in different levels of crop injury and yield loss. There is a strong relationship between boll maturity and *Lygus* feeding damage, thus understanding the boll maturation profile and characterizing *Lygus* damage risk dynamics is very important. Because reliable *Lygus*-resistant or tolerant cotton cultivars are unavailable, cotton producers primarily rely on pesticides for *Lygus* management. Current pesticide application decisions are based on field scouting, whereby spray applications are typically warranted when *Lygus* populations exceed locally established economic threshold (ET) levels.

Oosterhuis and Kim (2004) reported that cotton bolls that accumulated 350-450 heat units were safe from piercing-sucking insects. It is expected that *Lygus hesperus* may also be unable to damage cotton bolls once a certain boll maturity level has been reached, after which pesticide applications would not be necessary. However, the actual boll damage potential of *Lygus hesperus* is largely unknown. One important question in this study was: At what point do maturing bolls or the entire crop become "safe" from *Lygus* feeding damage, and, consequently, when does insecticide use become unnecessary? Given the availability of tools to identify when the bolls are safe, timing of insecticide use termination may be refined to minimize unnecessary economic and ecological costs.

The objectives of our field experiments were to: 1) determine the maximum potential for *Lygus* to inflict damage to cotton bolls at various boll maturity levels (ages), 2) determine the cotton boll maturity profile during two boll development stages (at 350 and 550 HU After First Flowering [AFF]), 3) determine the boll feeding preference of *Lygus hesperus* adults as affected

by the change in boll maturity profile as the crop matures from 350 HU to 550 HU AFF, and 4) quantify the yield loss caused by 4 different levels of *Lygus* infestations (0, 1, 2 and 4 or 6 *Lygus* adults per plant). The overall goal is to better understand the boll feeding biology and behavior of *Lygus hesperus* in order to further develop a dynamic economic threshold for improved *Lygus* management in Texas High Plains cotton.

Materials and Methods

Estimating *Lygus* Boll Damage Potential

A field study to quantify adult *Lygus hesperus* cotton boll damage potential was conducted at the Texas A&M AgriLife Research and Extension Center farm in Lubbock, Texas. Cotton cultivar ST5458B2RF was planted on May 18 (2012), May 22 (2013), and May 15 (2014) in a drip-irrigated field with 40-inch row spacing. The targeted seeding rate was 56,000 seeds per acre. On June 2, the 2012 study was treated with Orthene® 97S for thrips at a rate of 3.0 oz per acre and with Cornerstone Plus® herbicide (41% glyphosate) at 32 oz per acre for weed management, whereas the 2013 and 2014 study plots did not receive insecticide interventions for thrips control and weeds were removed via hand-hoeing.

2012 Study. The experimental design was a split-plot randomized block with three replications. Ten cotton boll age cohorts (1 to 20 days from flowering at 1-day increment) served as the main plot and two *Lygus* infestation levels (I: one adult *Lygus* feeding for 48 hours, and II: control or zero bugs) served as subplots. Thus, there were 30 main plots (3 blocks x 10 boll age cohorts), each of which consisted of 100 ft long cotton rows. In each main plot, 20 randomly selected white flowers were individually cup-caged using modified polystyrene foam and cloth-net "cup cages" (Fig. 1). Thus, a total of 600 white flowers were cup-caged (30 main plots x 20 flowers per main plot). Two treatment levels (control and single *Lygus* infestation) were applied in each main plot. Each plot contained 20 cup-caged bolls of which 5 bolls were used as controls, and the remaining 15 bolls were exposed to *Lygus* feeding. Cotton bolls in the Texas High Plains region typically accumulate 14-30 HU per day in August; thus, in ten days following cup-caging the fruit, on August 20, the August 1st cup-caged bolls received about 450 HU, whereas the August 10th cup-caged bolls had accumulated approximately 200 HU. Once the cotton bolls received 200-450 HU, individual *Lygus* adults were released in the appropriate cages and allowed to feed for 48 hours. *Lygus* adults were initially reared on artificial diet, but were "trained" on fresh green beans and cotton squares for a week prior to using them for the boll feeding experiment. Prior to release into the cup-cages, the *Lygus* adults were starved for 4-5 h. Five *Lygus* infested bolls from each plot were used for boll size, weight, carpel wall hardness and *Lygus* damage assessment (internal and external *Lygus* damage lesions), while the remaining ten *Lygus* infested bolls were kept for yield assessments. Both control bolls and the bolls kept for yield assessment were harvested during the first week of November, 2012.

2013 Study. The study was deployed in a split-plot randomized block design with three replications (blocks) to quantify the effect of *Lygus* density and infestation timing on cotton yield and quality. The study consisted of two *Lygus* infestation levels (one adult *Lygus* feeding for 48 hours versus zero bugs) as main plot factors and ten cotton boll age cohorts (every-other-day caging of bolls from Day 1 to Day 20) as subplot factors. Thus, there were 60 experimental units. Each experimental unit had eight individually caged bolls as subsamples, thus, this study comprised of a total of **480 individually caged cotton bolls** (three blocks x two *Lygus*

infestation levels x ten boll age cohorts x eight subsamples).

Cotton field was divided into three blocks. Each block consisted of 10 cotton rows, representing 10 boll age cohorts. Every two days for a period of 20 consecutive days (July 29 to August 18), one cotton row (a main plot) was randomly selected and twenty randomly selected new, white flowers were individually tagged, yielding 10 cotton boll age cohorts. On Day 21 (August 19), all 480 bolls were caged using modified polystyrene foam and cloth-net "cup cages" and individual *Lygus* adults were released in the appropriate cages and allowed to feed for 48 hours. Control cages received zero insect augmentation. After 48 hours, released *Lygus* bugs were killed in all cages and 50% of the infested bolls from each boll age cohort were retrieved and processed in the laboratory to evaluate internal and external *Lygus* damage lesions, boll weight, diameter, and boll hardness. The remaining 50% of the infested bolls were kept for harvest to determine yield and lint quality.

2014 Study. The study was deployed in a split-plot randomized block design with three replications (blocks) to quantify the damage potential of *Lygus* adults and late-instar nymphs with respect to cotton boll development stage. The study consisted of three *Lygus* infestation levels (one adult *Lygus* feeding for 48 hours, one late-instar nymph feeding for 48 hours, and zero bugs per boll) as main plot factors and ten cotton boll age cohorts (every-other-day caging of white flowers, also referred to as 1-day old bolls, from Day 1 to Day 20) as subplot factors. Thus, there were 90 experimental units. Each experimental unit had four individually caged bolls as subsamples, thus, this study comprised of a total of 360 **individually caged cotton bolls** (three blocks x three *Lygus* infestation levels x ten boll age cohorts x four subsamples).

Cotton field was divided into three blocks. Each block consisted of 10 cotton rows, representing 10 boll age cohorts. Every two days for a period of 20 consecutive days (4 August to 22 August), one cotton row (a main plot) was randomly selected and fifty randomly selected new, white flowers were individually tagged, yielding 10 cotton boll age cohorts. On Day 20 (23 August), all 360 bolls were caged using modified polystyrene foam and cloth-net "cup cages" and individual *Lygus* adults or nymphs were released in the appropriate cages and allowed to feed for 48 hours. Control cages received zero insect augmentation. After 48 hours, released *Lygus* bugs were killed in all cages and 25% of the infested bolls from each boll age cohort were retrieved and processed in the laboratory to evaluate internal and external *Lygus* damage lesions, boll weight, diameter, and boll hardness. The remaining 75% of the infested bolls were kept for harvest to determine yield and lint quality. These individual bolls were harvest on October 22.

Figure 1. Deployment of cup-cages to enclose age-specific bolls for *Lygus* damage potential study, Lubbock, TX, 2012-2014.

Determination of Boll Maturation Profile, Feeding Preference and Economic Threshold

A field study was conducted to quantify the effect of *Lygus* density and infestation timing on cotton yield and fiber quality. Cotton cultivar ST5458B2RF was planted on May 18 (2012), May 22 (2013), and May 15 (2014) in a drip-irrigated field with 40-inch row spacing at the Texas A&M AgriLife Research farm located near Lubbock, Texas. The targeted seeding rate was 56,000 seeds per acre. On June 2, the 2012 study was treated with Orthene® 97S for thrips at a rate of 3.0 oz per acre and with Cornerstone Plus® herbicide (41% glyphosate) at 32 oz per acre for weed management, whereas the 2013 and 2014 study plots did not receive insecticide interventions for thrips control and weeds were removed via hand-hoeing.

The field study was laid out in a split-plot randomized block design with three replications, two main plot factors (two cotton boll developmental stages [early boll development and late boll development]), and four subplot factors (four levels of *Lygus* infestation [control or zero bugs, one bug/plant, two bugs/plant, and four or six bugs/plant]). There were a total of 24 experimental units. Each experimental unit had 8 cotton plants as subsamples (3 used for damage assessment and 5 for yield and quality assessment). A total of 192 whole-plant sleeve-caged cotton plants (three blocks x two cotton boll stages x four *Lygus* densities x eight subsamples) were used for this study (Fig. 2).

The cotton field study site was closely monitored and kept virtually arthropod pest-free until cages were deployed on July 24, July 29, and July 28 in 2012, 2013, and 2014, respectively. When the cotton plants reached the target maturity level (350 HU >60 °F after first flower on August 7, August 13, and August 17 in 2012, 2013, and 2014, respectively, and 550 HU >60 °F after first flower on August 21, August 29, and August 27 in 2012, 2013, and 2014, respectively), field-collected *Lygus* were released into the whole-plant sleeve-cages at the rates of 0, 1, 2, and 4 *Lygus*/plant in 2012 and 2013; the infestation densities were changed to 0, 2, 4, and 6 *Lygus*/plant in 2014 to increase the damage intensity. *Lygus* adults were collected from nearby alfalfa field or from adjacent counties and acclimatize in the laboratory for 48 hours before using them for the boll feeding experiment. Cotton plants were exposed to the *Lygus* adults for ~7 days, after which time, the insects were killed via a pesticide application. Three randomly selected cotton plants from each plot were cut and brought to the laboratory on August 13, August 19, and August 27 for the 350 HU and August 29, September 2, and September 5 for the 550 HU plots in 2012, 2013, and 2014, respectively. The cotton crop was defoliated by spraying FOLEX® 6EC (12 oz per acre) and a boll opener (Ethephon® 6; 32 oz per acre) in a tank mix in all three years of the study. After the crop was ready to harvest, the remaining 5 caged plants from each plot, which had been maintained pest-free, were harvested manually to evaluate the lint yields and fiber quality. Harvested single-plant samples were ginned individually via table-top gin and samples were analyzed for fiber quality (HVI) parameters at Cotton Incorporated. Data from the whole-plant cage study were summarized by calculating average and standard errors. ANOVA, GLM model in SAS, 2010 were used to evaluate the treatment effects (α=0.1) and treatment means were compared by LSMEAN procedure.

Figure 2. Field deployment of whole-plant cages for threshold study, Lubbock, TX, 2012-2014.

Results and Discussion

Boll Development vs. *Lygus* Damage Potential

The Lubbock area cotton crop during the August 1-20 period in 2012 received ≈24 HU per day and bolls developed rapidly. The diameter of the cotton bolls grew at an average rate of 1.2 mm per day and gained an average of 1.4 grams of weight per day. As the bolls matured and became larger, the carpel walls became harder as evidenced by the pressure required to puncture the carpel wall, increasing at a rate of 0.018 lb per square foot per day (Fig. 3). The 2013 and 2014 boll development patterns were similar to that for 2012. When forced to feed on a single boll, each *Lygus* adult inflicted, averaged across all boll age cohorts, 10-28 external lesions per boll in 48 hours. Numerous external lesions were found in all bolls, irrespective of their age. It indicates that in a "no-choice" feeding situation *Lygus* can cause external feeding injury to all bolls, but the actual number of damaged seeds was significantly reduced as bolls became older, larger and tougher to puncture. When bolls reached an age of 16 days (2012) or 13 days (2013), *Lygus* caused very little seed damage (<2 seeds per boll) that did not result in significant lint yield reductions (Figs. 4-5). We were unable to derive this relationship for 2014 data due to field management failure prior to harvest. When cotton bolls received >350 HU after first flower, they were safe from *Lygus*-induced fiber yield loss. Cotton bolls were observed to be safe from *Lygus* damage when the bolls: 1) exceeded >28 mm in diameter, 2) weighed >14 g, or 3) carpel wall puncture force exceeded 0.7 lb per square foot (Figs. 3-5).

Boll damage potential significantly increased as bolls mature from Day 1 to Day 7, demonstrating that the 1-wk old bolls are the most sensitive to *Lygus* injury. The damage potential begins to decrease after 7 days, but bolls are still susceptible to *Lygus* injury for about another 5-6 days. Considering year-to-year variations, it appears that the maturing bolls are no longer susceptible to *Lygus* injury two weeks after white flower (Figs. 4 and 5).

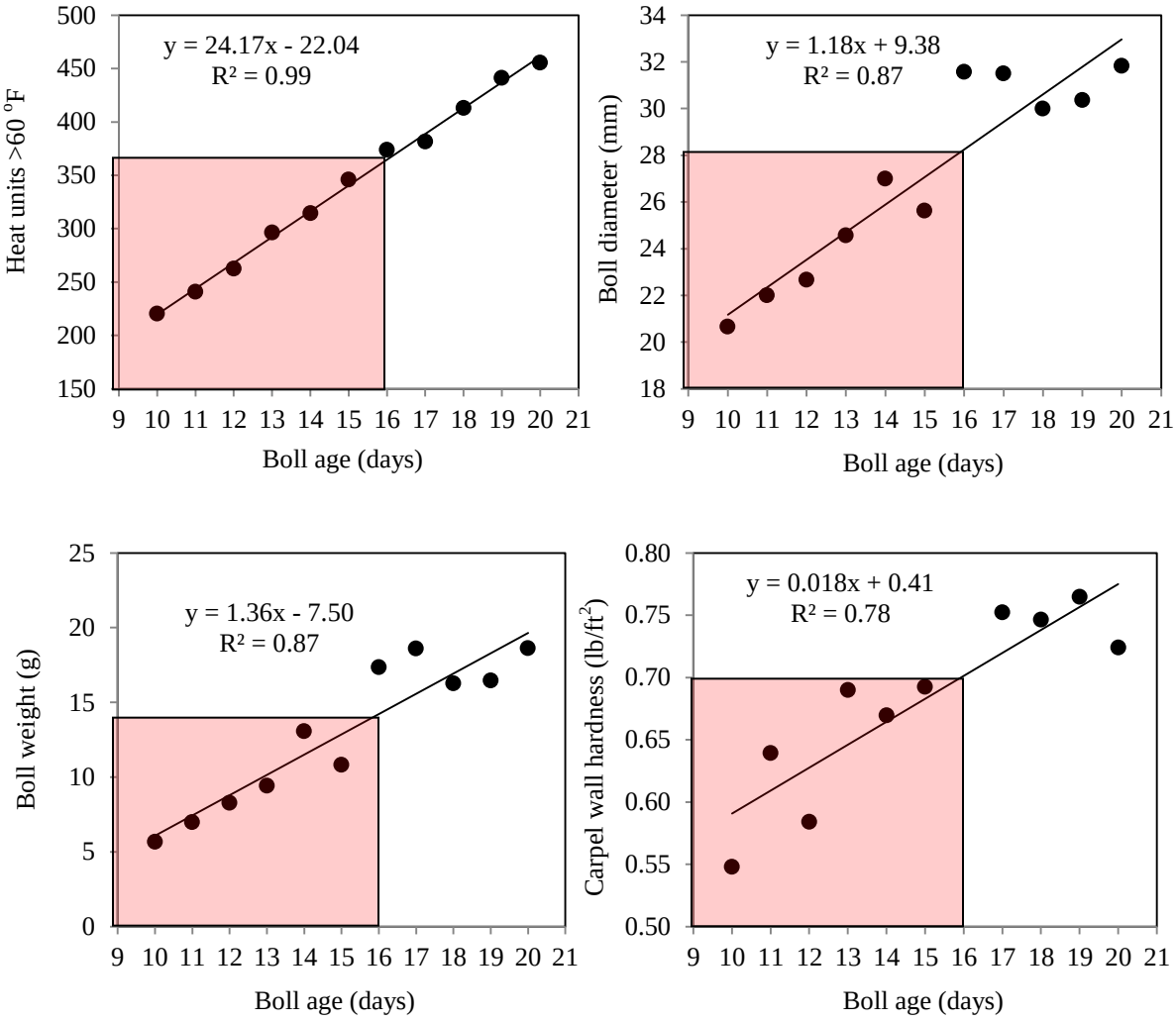

Figure 3. Cotton boll age relationships as associated to heat unit accumulations, boll size, boll weight, and carpel wall hardness, Lubbock, Texas, 2012.

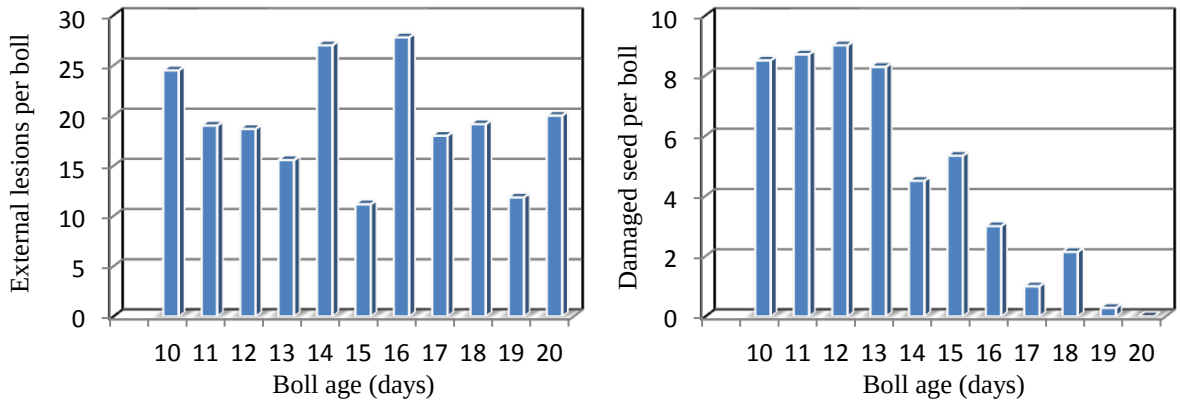

Figure 4. Cotton boll injury (external lesions and damaged seeds) at various boll ages following a 48-h feeding of a single *Lygus* adult, Lubbock, TX, 2012.

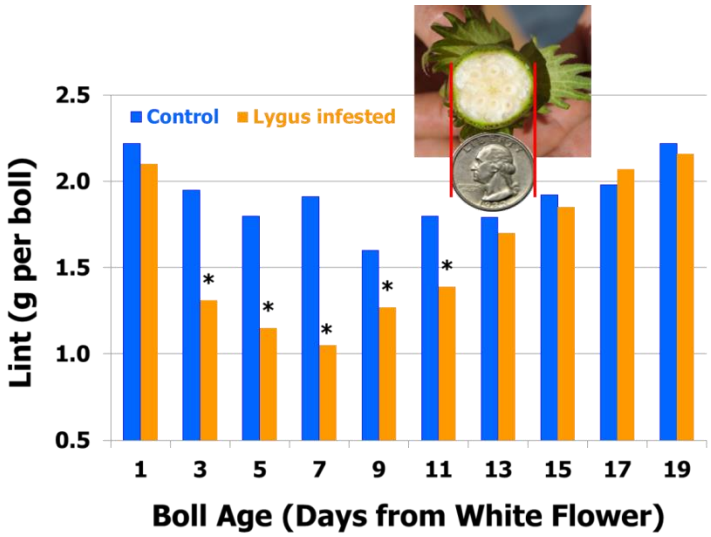

Figure 5. Single-boll lint yield (grams per boll) following 48 hours of feeding by a single *Lygus* adult versus uninfested boll at boll ages ranging from Day 1 to Day 19, Lubbock, TX, 2013. * indicates that the *Lygus*-infested bolls resulted in significantly lower yield than unfested bolls.

Fruiting Profile

At 350 HU after first flower, an average of 57% fruit retention was observed, but fruit retention was decreased to 37% when cotton reached 550 HU after first flower. Cotton plants at 350 HU were observed to have 84% bolls, 14% squares and 2% flowers, while at 550 HU, the cotton plants had 99% bolls, 1% squares, and no flowers. Although there were a higher percentage of cotton bolls on 550 HU plants, the actual number of bolls per plant decreased from an average of 8.8 bolls per plant at 350 HU to 6.3 bolls at 550 HU. Approximately 28.4% of the bolls were naturally aborted from the plants as they matured from the 350 HU to 550 HU stage (Fig. 6).

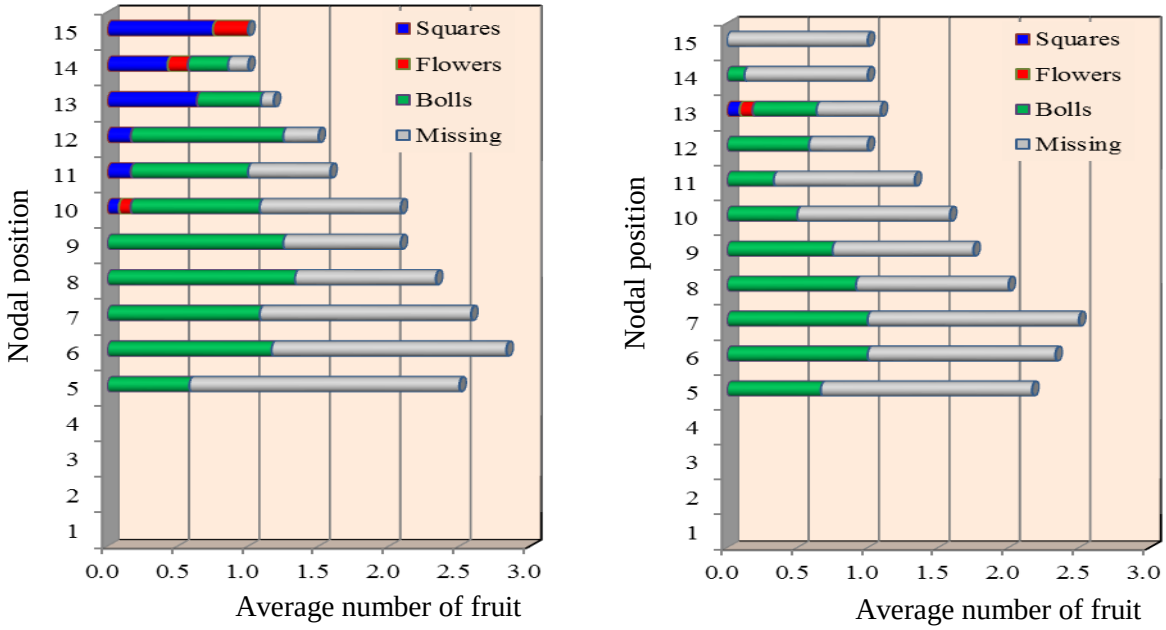

Figure 6. Fruiting profile at 350 (left) and 550 (right) HU after first flower, Lubbock, TX, 2012.

Most of the bolls were from first fruiting positions of the sympodial branches. At 350 HU, 66%, 24%, 8%, and 2% bolls were from the first, second, third and fourth sympodial branch fruiting positions, respectively; while at 550 HU, 81%, 16%, 3%, and 0% bolls were from the first, second, third and fourth sympodial branch fruiting positions, respectively (Fig. 7). When the cotton plants matured from 350 HU to 550 HU, they dropped all of the 4th fruiting position and most of the 3rd fruiting position bolls. Since 97% of the bolls were on first and second fruiting positions on the cotton plants at the 550 HU stage, our sampling and crop protection efforts should be focused on protecting primarily the first and second position bolls at this stage. However, fruiting profiles may vary with cotton cultivar, cotton growing region, and crop management practices and input use patterns.

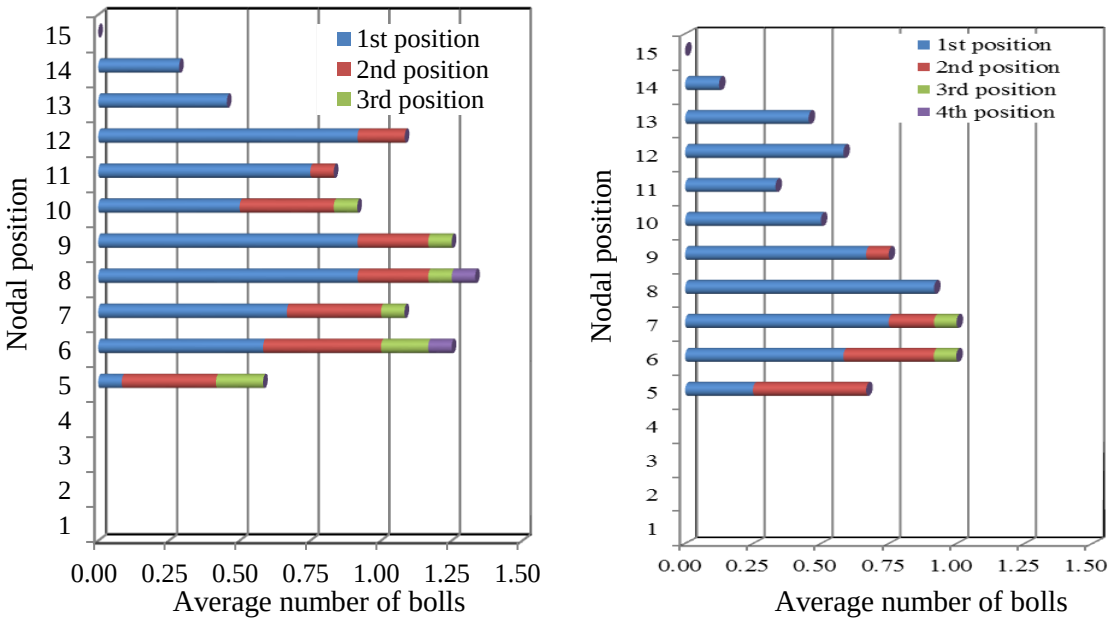

Figure 7. Boll distribution on sympodial branches at 350 (left) and 550 (right) HU after first flower, Lubbock, TX, 2012.

Boll Maturation Profile

Thirty-two cotton plants were harvested (16 plants each from 350 HU and 550 HU plots) from which 643 bolls were retrieved. Boll diameter was measured using a Vernier caliper and bolls were categorized into 6 boll size groups (5-10, 11-15, 16-20, 21-25, 26-30 and 31-35 mm). Our past research indicates >25 mm diameter sized cotton bolls are safe from *Lygus* damage. Plants at 350 HU had 47% of the bolls safe from *Lygus* damage (larger than 25 mm diameter), whereas after 2 additional weeks, cotton in the same field had 70% of the bolls safe from *Lygus* damage. When the cotton crop matured from 350 to 550 HU, the proportion of bolls vulnerable to *Lygus* feeding damage was reduced from 53% to 30%. Therefore, it is likely that with a similar level of *Lygus* infestation, *Lygus* may cause a greater amount of cotton yield loss when infesting a mid-season crop (350 HU) compared to that for a late season infestation (550 HU).

For our 2012 cotton crop, within-plant cotton boll maturation profile shows that bolls distributed from the 5th to 13th nodes (Fig. 8). At the 350 HU stage, the top 4 bolls (from 10-13th node) were <25 mm diameter size and were vulnerable to *Lygus* damage if bugs were present. When the

cotton reached 550 HU, only the top 3 bolls (nodes 11-13) were <25 mm diameter size and therefore vulnerable to *Lygus* damage, if present. Bolls from the 5th to 9th nodes were larger and less vulnerable to *Lygus* feeding damage. There was a very strong positive relationship between boll size (diameter) and the hardness of the boll carpel wall. As we move from the top to bottom nodes of a cotton plant, as expected, we found larger bolls with harder carpel walls (Fig. 8). The vertical boll profile suggests that cotton growers or crop consultants need to focus their *Lygus* damage evaluations primarily during the 350-550 HU, and mostly on the top 3-4 bolls, since they are the most vulnerable to *Lygus* feeding injury. The 2013 data also showed similar trends in terms of within-plant boll maturation distribution.

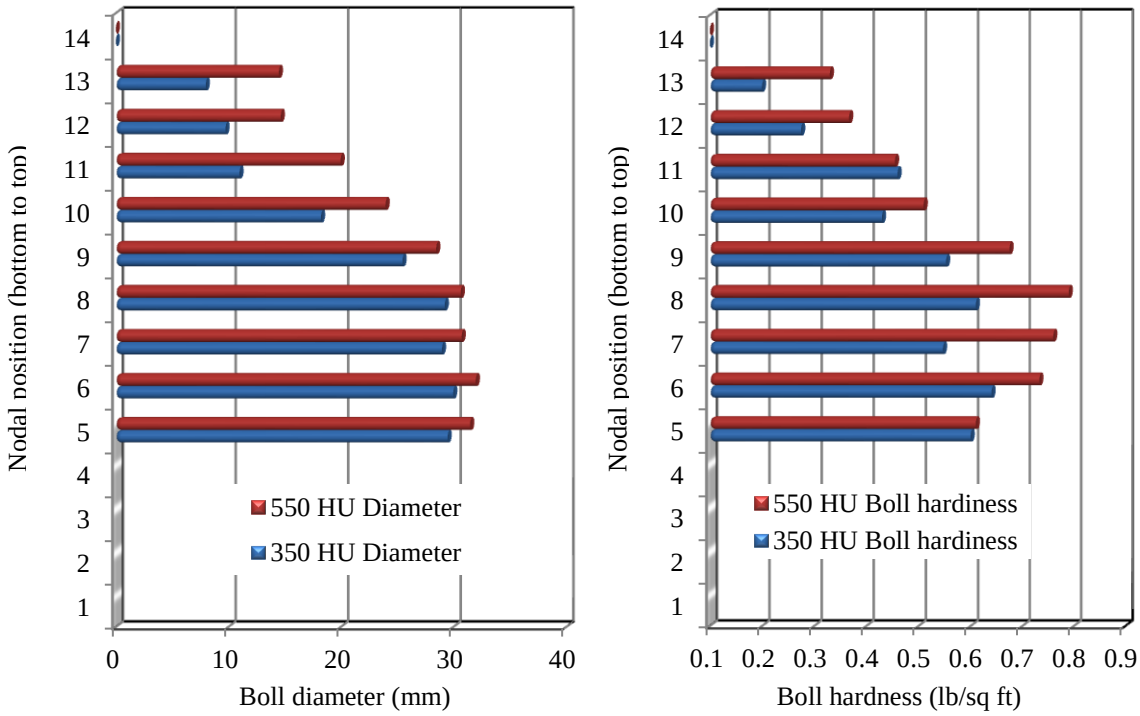

Figure 8. First position boll size profiles of 350 and 550 HU cotton after first flower, Lubbock, TX, 2012.

Lygus Boll Feeding Preference and Boll Damage

In the whole-plant caging study, *Lygus* external feeding lesions were found in bolls of all sizes, indicating *Lygus* attempted to feed on cotton bolls irrespective of boll size. Nevertheless, successful punctures and the resulting internal warts were limited to the bolls <35 mm in diameter. A significantly higher proportion of bolls had internal warts (>20% of bolls) for <30 mm bolls, indicating that in an open-choice situation, *Lygus* preferred to feed on bolls that were <30 mm in diameter (Fig. 9). Cotton plants at the 350 HU had 90% of the bolls measuring <30 mm in diameter, whereas plants at the 550 HU had 78% of the bolls at <30 mm diameter (Fig. 9). The no-choice cup-cage study showed bolls that are >25 mm diameter were safe from *Lygus* damage, whereas in the open-choice whole-plant caging study, *Lygus* preferred to feed on bolls

up to 30 mm in diameter. This slight discrepancy might be due to differences in cotton boll development inside cup-cages versus whole-plant cages, or due to differences in *Lygus* behavior in the presence of different boll size options and containments. Evaluation of internal lesions and internal warts suggests there is not a significant relationship between external *Lygus* feeding lesions and actual seed damage due to *Lygus* feeding (Fig. 10), but there were strong relationships between the number of internal warts and number of *Lygus* damaged seed. It clearly indicates that estimating *Lygus* damage by using external lesions can be misleading; therefore, it is best to use the number of internal warts to estimate the degree of *Lygus* crop damage.

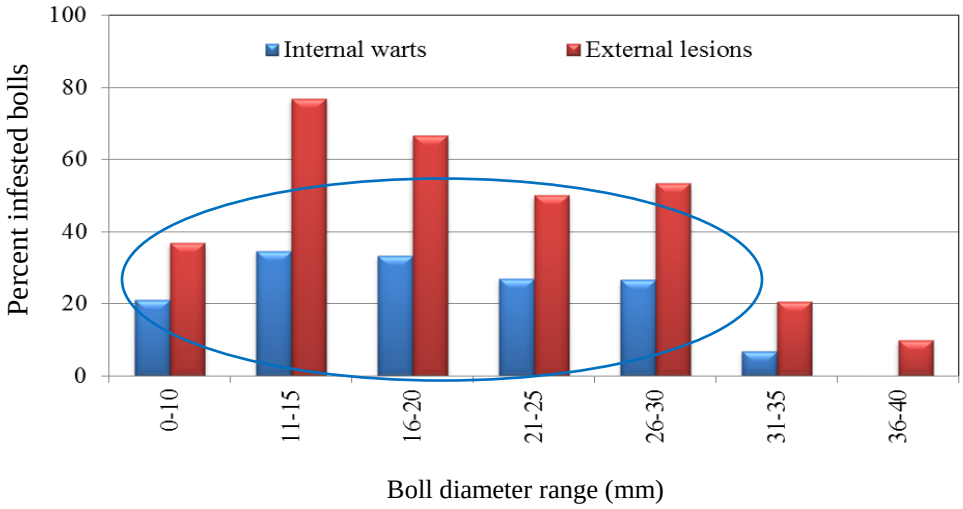

Figure 9. Boll feeding preference of *Lygus* in whole-plant cages based upon the proportion of external and internal boll damage. Lubbock County, TX, 2012.

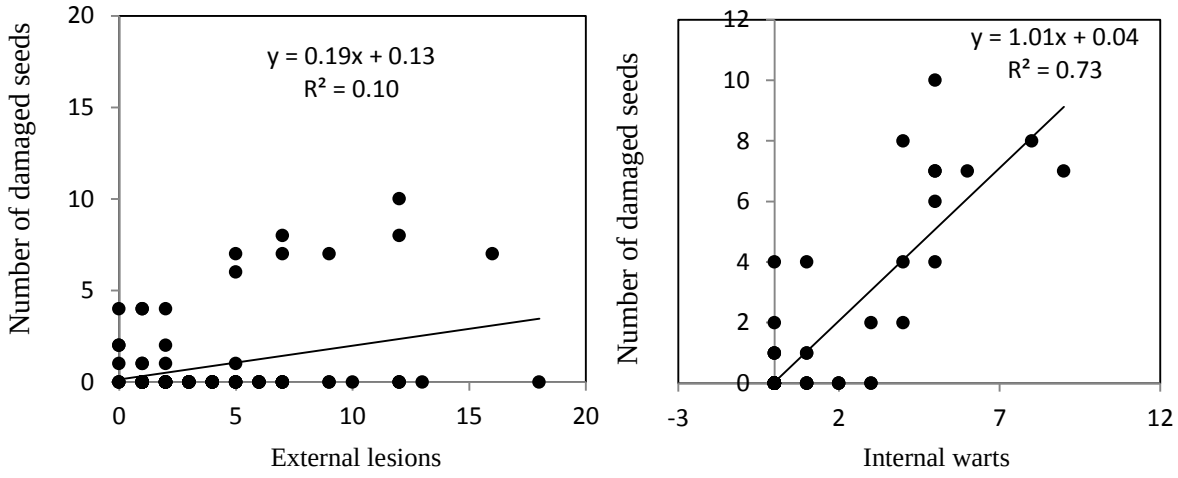

Figure 10. Relationships between the number of damaged seeds per boll and the number of external lesions or internal warts, Lubbock, TX, 2012.

Yield Loss

In general, as expected, *Lygus* augmentation reduced the lint yield compared to that in uninfested control cages (Figs. 11-13). However, the damaging effect of *Lygus* was more pronounced during mid-season (350 HU from first flower) compared to that in late season (550 HU from first flower) for all three years of the study.

In 2012, artificial augmentation of 2-4 *Lygus* bugs per plant at 350 HU after first flower significantly reduced the cotton lint yield, but the same level of *Lygus* infestation at 550 HU did not result in significant lint yield reduction compared with that in uninfested control plants (Fig. 11). Augmentation of 1, 2, and 4 *Lygus* bugs per plant at 350 HU after first flower reduced the cotton lint yield by 116, 425, and 580 lb/acre, respectively, whereas the yield reductions for the same *Lygus* densities were 125, 149, and 185 lb/acre during the late season (550 HU from first flower).

These data suggest that the maturing bolls are much more tolerant to *Lygus* injury when the plant attains 550 HU from first flower. It is also possible that *Lygus* bugs may choose to feed on superfluous bolls or squares and the yield contributing fruits may not be significantly impacted by such late infestations. Because potential yield loss risks due to certain *Lygus* density infestations vary with boll maturation profile, the *Lygus* management economic threshold should be optimized for a dynamic ET to accommodate for within-plant fruit maturity profiles.

In 2013, cotton lint yields in mid-season plots (cages) were much lower than in 2012, but the augmentation of 1, 2, and 4 *Lygus* bugs per plant reduced the cotton lint yield by 157, 106, and 281 lb/acre, respectively (Fig. 12). While these lint yield reduction values were not statistically significant, owing to greater variation in data, the trend was convincingly supportive of a clear influence of *Lygus* augmentation on yield reduction and the data trend was similar to that in 2012. Overall, lint yield was higher in late-season test plants compared to that in mid-season test plants, but the augmentation of 1 *Lygus* per plant did not result in significant yield reduction, whereas 2 and 4 *Lygus* per plant reduced 143 and 159 lb/acre, respectively (Fig. 12).

In 2014, augmentation of 2, 4, and 6 *Lygus* per plant at 350 HU after first flower reduced the cotton lint yield by 407, 406, and 516 lb/acre, respectively, whereas the yield reductions for the same *Lygus* densities were 282, 295, and 415 lb/acre during the late season (550 HU from first flower) (Fig. 13). Overall yield in 2014 was higher than in 2012 and 2013, but the damage inflicted by 2 and 4 *Lygus* per plant on mid-season cotton was comparable to that for 2012, whereas the damage inflicted in late season cotton was higher in 2014 compared to that in 2012 or 2013.

Lygus-induced lint yield reduction for a given *Lygus* density was lower for late season compared to that for mid-season infestations in all three years of the study (Figs. 11-13). These data clearly suggest that the maturing bolls are more tolerant to *Lygus* injury when the plant attains 550 HU from first flower. It is also possible that *Lygus* bugs may choose to feed on superfluous bolls or squares and the yield contributing fruits may not be significantly impacted by such late infestations. Because potential yield loss risks due to certain *Lygus* density infestations vary with boll maturation profile, the *Lygus* management economic threshold should be optimized for a dynamic ET to accommodate for within-plant fruit maturity profiles. Regression analysis of our current three-year data suggests that *Lygus* adults could inflict maximum lint losses of 125 and 78 lb/acre, respectively, for mid- versus late season infestations of per unit (1 adult) Lygus per

plant (Fig. 14). Additional data will be generated in 2015 and a more robust ET values would be calculated for late season Lygus management in the Texas High Plains.

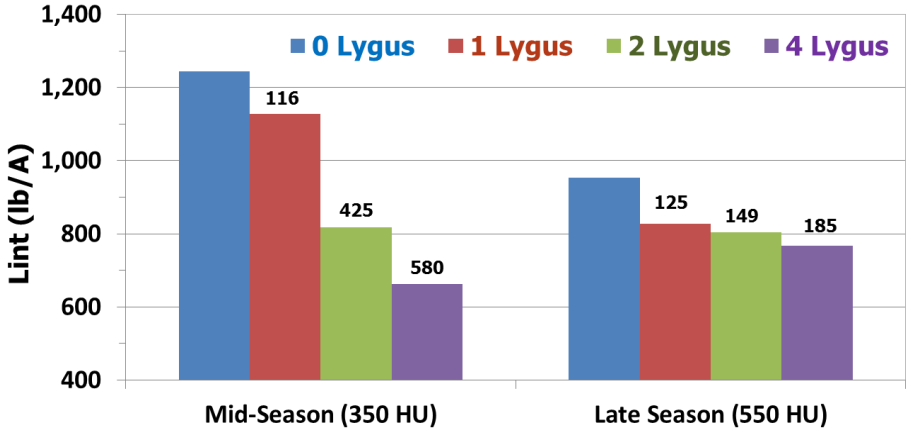

Figure 11. Influence of varying levels of *Lygus* infestations on lint yields at two crop phenological stages, as measured by heat-unit accumulation beyond first white flower, Lubbock County, TX, 2012.

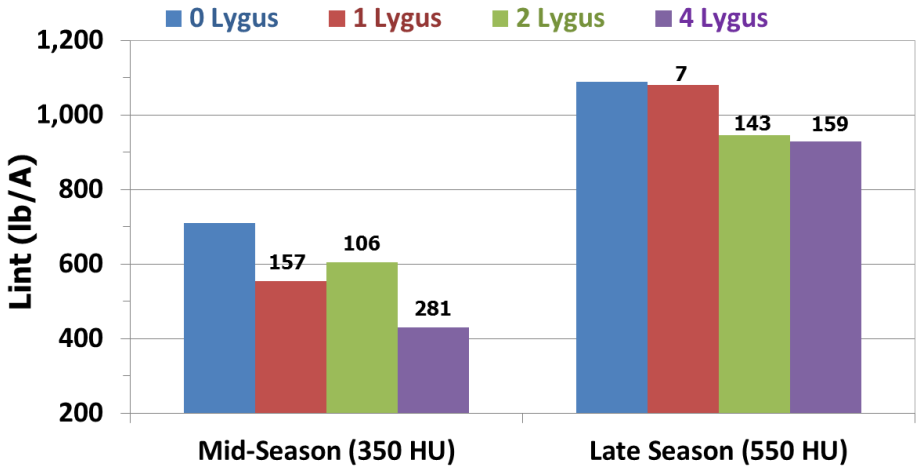

Figure 12. Influence of varying levels of *Lygus* infestations on lint yields at two crop phenological stages, as measured by heat-unit accumulation beyond first white flower, Lubbock County, TX, 2013.

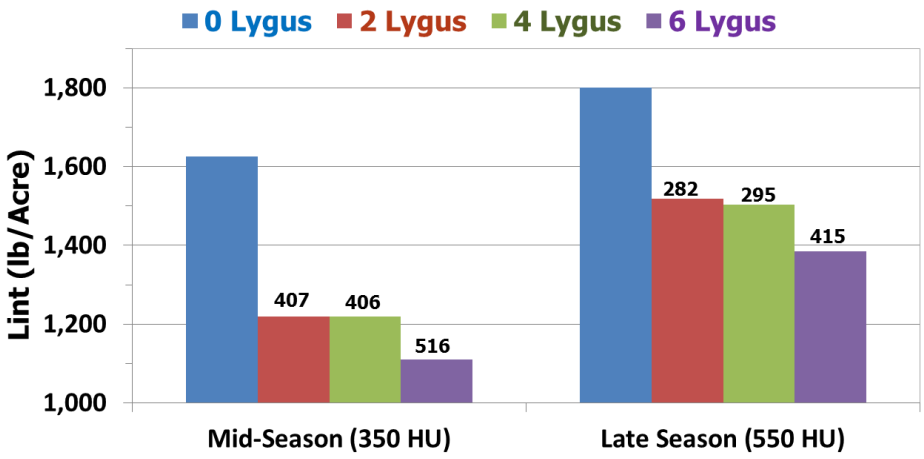

Figure 13. Influence of varying levels of *Lygus* infestations on lint yields at two crop phenological stages, as measured by heat-unit accumulation beyond first white flower, Lubbock County, TX, 2014.

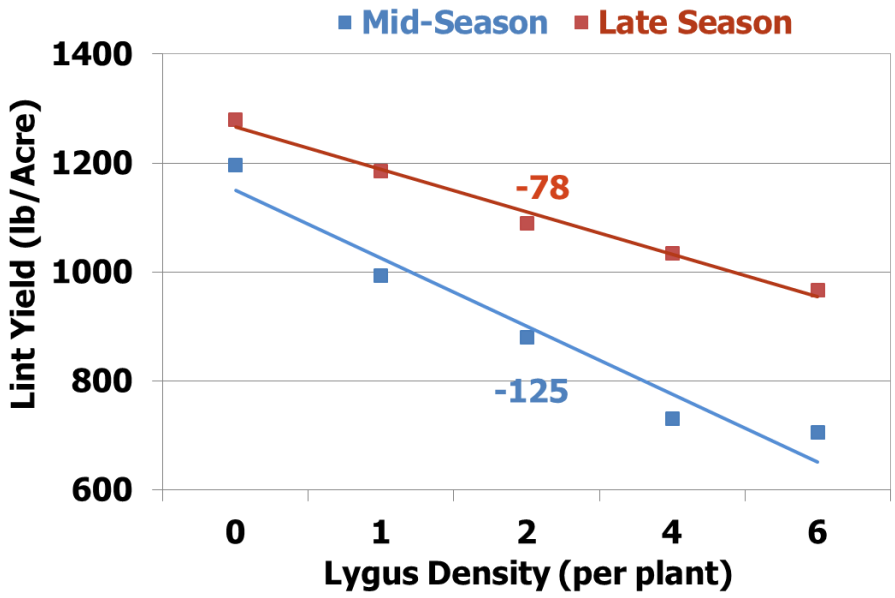

Figure 14. Regression analyses of three years of *Lygus* whole-plant cage study data (2012-2014) showing the relationship between the amount of lint yield reduction and *Lygus* density augmentation per plant. Mid-season infestation, $y=1275.1 - 124.7x$ ($R^2 = 0.95$); Late season infestation, $y=1345-78x$ ($R^2=0.99$); x=Number of adult *Lygus* bugs augmented per plant, y=Lint yield (lb/acre).

Summary

There was a significant change in boll composition (boll profile) between the cotton plants at 350 and 550 HU from first flower. Despite a subtle variation between no-choice (cup-caged single boll feeding) versus choice (whole-plant cage with access to all boll types for feeding) situations, it appeared that bolls were relatively safe at 28-30 mm diameter size or 350 HU, which was approximately equivalent to 2-wk old bolls. While year-to-year variation exists and the variation in boll susceptibility is expected across cropping system management (irrigation, planting date, fertility, etc.), maturing bolls should generally be safe from *Lygus* injury two weeks after white flower, especially for *Lygus* adults. *Lygus*-induced lint yield reduction for a given *Lygus* density was lower for late season compared to that for mid-season infestations in all three years of the study. These data clearly suggest that the maturing bolls are more tolerant to *Lygus* injury when the plant attains 550 HU from first flower. It is also possible that *Lygus* bugs may choose to feed on superfluous bolls or squares and the yield contributing fruits may not be significantly impacted by such late infestations. Because potential yield loss risks due to certain *Lygus* density infestations vary with boll maturation profile, the *Lygus* management economic threshold should be optimized for a dynamic ET to accommodate for within-plant fruit maturity profiles.

Acknowledgements

We thank our funding agencies including Cotton Incorporated Texas State Support Committee and Plains Cotton Growers, Inc. The authors also are indebted to Betsy Perafan, Sydnee Woodman, Christy Hogue, Jason Saiz, Jeffrey Drymalla, and Carley Covert, whose technical assistance in the field was invaluable.

References

Jubb, Jr. G. L., and L. A. Carruth. 1971. Growth and yield of caged cotton plants infested with nymphs and adults of *Lygus hesperus.* J. Econ. Entomol. 64: 1229-1236.

Oosterhuis, D. M., and M. Kim. 2004. Structural and biochemical changes in the boll wall in support of the NAWF 5+350 heat units rule, pp. 285-295. *In* Proc., Beltwide Cotton Conferences, National Cotton Council, Memphis, TN.

Parajulee, M. N., R. B. Shrestha, A. K. Barman, and S. C. Carroll. 2008. Ecologically intensive pest management in cotton agroecosystems: *Lygus hesperus* as a model system. Egyptian Journal of Agricultural Research 86: 57-81.

Parajulee, M. N., M. B. Adhikari, D. L. Kerns, R. B., Shrestha, and S. C. Carroll. 2011. Insecticide termination rule for western tarnished plant bug in the Texas High Plains, pp. 1204-1208. *In* Proceedings, Beltwide Cotton Conferences, National Cotton Council, Memphis, TN.

Williams, M. R. 2013. Cotton insect losses-2012, pp. 546-586. *In* Proceedings, Beltwide Cotton Conf., National Cotton Council, Memphis, TN.

2014 ANNUAL REPORT (YEAR 3)

Cotton Incorporated Core Program

Project Number: 12-364

Characterization of Cotton Crop Response to Thrips Injury for Improved Thrips Management in Texas High Plains Cotton

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Introduction

Thrips are economically important pests in Texas cotton. Thrips can be found in cotton throughout the growing season, but cotton is most vulnerable to thrips damage for the first thirty days following planting and cotyledon emergence. In the U.S., thrips infested a cumulative area equaling 8.9 million acres in 2012 while thrips infested 3.8 million acres in Texas which caused a loss of approximately 9,000 bales in Texas (Williams 2013). Excessive feeding of thrips leads to the browning of leaves on the edges, development of a silvery leaf surface color, or curling upward from the edges (Fig. 1). Western flower thrips, flower thrips, soybean thrips, onion thrips, and tobacco thrips are five common thrips species found in U.S. cotton (Cook *et al*. 2011). Albeldaño *et al.* (2008) have reported nine species of thrips from Texas cotton. Western flower thrips [*Frankliniella occidentalis* (Pergande)] is a primary pest in Texas cotton (Parajulee et al. 2006) and causes severe damage to cotton seedlings in infested fields, which are generally vulnerable to thrips damage up to the 4-5 true leaf stage (Cook *et al*. 2011). Thrips cause leaf area destruction, delayed maturity, retarded plant growth and loss of apical dominance (Sadras and Wilson 1998, Harp and Turner 1976). Previous thrips surveys revealed at least eight thrips species in Texas cotton, but *Frankliniella occidentalis* (western flower thrips) and *Thrips tabaci* (onion thrips) are the most common species, comprising more than 75% of the thrips found in Texas cotton. The various thrips species in Texas, being difficult to identify, have typically been managed as a single complex, with a single approach being broadly applied. Differential damage potential and pesticide susceptibility among these species remain unexamined, but with the recent aldicarb (Temik®) discontinuation, their examination may be critical.

Lacking thrips-tolerant cotton cultivars, cotton growers primarily use insecticides to control thrips. While several seed treatment options are available, soil-applied aldicarb had been the most reliable and common method used for cotton seedling thrips control. With the discontinuation of aldicarb, cotton growers will need alternative thrips management techniques, especially in the Texas High Plains. Ideally, cotton growers should be empowered with the capability to estimate the daily cost of delaying foliar insecticide applications for controlling thrips, further empowering them to finely adjust and achieve their acceptable, sustainable economic injury level for maximum benefits and minimum costs. Proposed project outputs include information such as the specific relationship between the degree of thrips injury to cotton seedlings and the resulting plant response in terms of final yield and fiber quality, the specific cotton growth stage most vulnerable to thrips infestation, an accurate economic threshold for initiating thrips management actions, and the effect of infestation duration on cotton development and lint yield, all of which would be valuable to empower growers with such a capability, given EPA-mandated aldicarb discontinuation.

Foliar insecticide applications are likely to replace aldicarb, and are likely to increase in number. Given such an increase, and since information regarding specific thrips species, their damage potential, and how cotton responds is unavailable, the risk of excessive or inadequate insecticide use is likely to increase as well. Further, while Texas A&M AgriLife Extension currently provides general thrips management thresholds, such broadly-applicable thresholds are insufficient to address specific thrips species, different injury levels, infestation duration, and their effects on the cotton crop growth response and final yield potential. Therefore, the goal of this project is to develop applicable information which will empower producers to optimize the timing and extent of management actions to mitigate thrips damage while protecting the agroecosystem, maximizing yields, and minimizing production costs. In addition to benefitting

producers, the outcome of this study will aid crop consultants and county IPM agents in making recommendations to improve thrips management in Texas High Plains cotton.

The manipulation of thrips populations in a cotton field setting is very challenging and maintaining selected thrips densities on cotton seedling in an open field condition is unmanageable. Nevertheless, we must use field cages and confine known number of thrips per caged plant to obtain a desired thrips density. Specific objectives of the second year of this study were to: 1) evaluate cotton varietal response to natural colonization of thrips in open field studies, 2) greenhouse evaluation of cotton varietal response to thrips augmentation, and 3) design a field cage prototype to determine the cotton crop damage potential of the western flower thrips for developing an economic threshold. The ultimate goal of the research project is to develop new economic thresholds for thrips based upon plant response characteristics, validating or revising the current Texas High Plains thrips treatment threshold recommendations, and precisely characterizing the cotton crop response to various levels of thrips injury at different cotton seedling ages.

Figure 1. A) Adult western flower thrips, *Frankliniella occidentalis*, B) Severe damage caused by *F. occidentalis* to seedling cotton, C) Stunted cotton seedlings due to thrips injury.

Materials and Methods

Objective 1. Cotton cultivar response to natural colonization of thrips in the field

This study was conducted at the Texas A&M AgriLife Research farm in Lubbock, Texas. The study was deployed in a randomized block design with four replications and six cultivar treatments. Experimental plots were eight 40-inch rows wide x 90 ft long and 5 ft alleys separated the plots. Six cotton cultivars (SSG-HQ-212-CT, DP 353, FM 1740B2F, 07-7-1407, 07-7-1001, and PHY 367 WRF) were planted on May 9 (2013) and June 3 (2014). Each 8-row plot was further divided into two 4-row plots and each of the two 4-row plots was randomly assigned to a 'control' or 'sprayed' treatment. Thus, the entire study consisted of 48 experimental units (six cultivars x two treatments x four replications).

2013 Study. Cotton germination was delayed due to cooler soil temperatures, but the plant emergence was satisfactory in most plots. A poor crop stand on some experimental plots may be attributed to variations in cultivar seed vigor rather than to the soil conditions. Plant stand counts were performed on May 23 and June 3 by counting all plants in 3 row-ft per row in all 48 plots. Thrips densities were monitored in all 48 plots using a ten-plant thrips washing technique. Thrips sampling dates were May 23, May 25, June 3, June 10, and June 17. An insecticide (Orthene® 97S @ 3.0 oz/acre) was sprayed in all 24 'sprayed' treatment plots after each thrips sampling

event on May 24, May 30, June 11, and June 18, and the entire test (all 48 plots) was sprayed with this insecticide on June 26. Insecticide treatment application was skipped after the thrips sampling event on June 3 due to spray logistic issues, but the residual insecticides from previous week's application kept the thrips populations suppressed until the insecticide application on June 11. Plant response to thrips injury was monitored by measuring plant height, shoot length, root length, total leaf area, and total dry biomass of cotton seedlings from each plot on June 24. A 10-ft section was marked on each of the two center rows within each plot and the flowering profile was monitored 2-3 times per week. This type of phenological monitoring began prior to the initiation of flowering and continued until crop cut-out. Flowering profiles were monitored on July 10, 12, 16, 19, 22, 24, 26, 30, August 13, and 30. The two 10-ft sections from the middle two rows (20 total row-ft/plot) that were designated for plant fruiting response were harvested and ginned to estimate the cotton lint and seed yields from each experimental plot.

Plant response to thrips injury was monitored by measuring shoot length, root length, shoot biomass, root biomass, total leaf area, and total dry biomass of cotton seedlings from each plot. The study area received approximately 3.0 inches of rain on July 16-17 which provided a much needed break from an extended drought. Nevertheless, the test plots received a full complement of irrigation and the test had not been exposed to a water-stress situation. Frequent cultivations kept the weeds under control as well. The crop received harvest-aid chemicals on October 9 and the crop was harvested on November 4, followed by sample ginning on November 20.

2014 Study. The study site received frequent rains that cooled the soil temperature. Thus, the early crop growth was extremely slow. Plant stand counts were performed on 13 June by counting all plants in 3 row-ft on each of the four rows in all 48 plots. Thrips densities were monitored in all 48 plots using a five-plant thrips washing technique in each plot. Thrips sampling dates were June 19 and 26, and July 8. An insecticide (Centric® @ 3 oz. per acre) was sprayed in all 24 'sprayed' treatment plots on 23 June, but there were no visible thrips in subsequent sampling events to warrant additional spray applications. Harsh weather conditions followed by some unexplained herbicide drift injury on most of the conventional cotton lines in the region around the third week of June and first week of July prevented the test crop from achieving normal growth. Test plots were ranked using a 1-10 scale (1 being dead and 10 being healthy plants) on 16 July for crop vigor. Cultivar PHY 367 WRF and FM 1740B2F scored 8 and 7.5, respectively, while SSG-HQ-212-CT scored the lowest at 3.25. Overall, the stand counts were poor. Monsanto representatives visited the test site to ascertain the herbicide injury. Similar symptoms had been observed in other parts of Texas High Plains region on cotton cultivars that did not possess the Roundup Ready technology. The crop vigor did not improve during much of the growing season. No further pesticide was applied because of low thrips presence and stunted growth of the crop. Nevertheless, the crop was harvested and ginned.

Objective 2. Cotton cultivar response to different thrips densities in the greenhouse

2013 Study. A greenhouse study was conducted to determine the maximum potential effect of different densities of thrips on seedling cotton. Six cotton varieties (07-7-1001, 07-7-1407, PHY 367 WRF, SSG-HQ-212-CT, FM 1740B2F and ST 5458B2RF) were planted in 16-oz Styrofoam® cups on October 8, 2013. At the bottom of the Styrofoam® cups, 1-3 small holes were made to allow for water drainage from the potting soil. The study was deployed in a completely randomized block design with four replications, six cultivars, and four thrips densities. Each experimental unit contained 6 plants. Thrips were field-collected from cotton and

reared on green beans in the laboratory. Immature thrips were transported to the greenhouse in containers with green beans. A brush was used to dislodge thrips from the green beans onto the cotton seedlings. Every effort was made to release only immature thrips to avoid unintentional movement of thrips between treatments. Thrips densities released included: no thrips (control), ½ thrips per plant (e.g., 1 thrips per two plants), one thrips per plant, and two thrips per plant at the 1- to 2-true leaf stage. An automatic sprinkler system was programmed to water the plants three times per week for 8 minutes. In addition, supplemental water was manually applied as needed.

The greenhouse ambient air temperatures were recorded using a small iButton® data logger (Maxim Integrated, San Jose, CA). Visual leaf tissue damage rankings of all plants were recorded prior to clipping. Ranking was based on a scale of 1-10 (1 = healthy plants with no damage symptoms and 10 = plants killed by thrips). Chlorophyll readings were also recorded using a chlorophyll meter to determine if treatments (thrips densities) and/or tested cotton varieties had an impact on chlorophyll levels. Leaf area from each treatment was recorded using a leaf area meter to test the influence of thrips on leaf surface areas.

Thrips were allowed to feed and reproduce for three weeks (the duration that is equivalent to the western flower thrips lifecycle) before plants were clipped near the soil surface and placed into denatured ethyl alcohol. Later, the adult and juvenile thrips were quantified via a plant washing technique as follows: All six plants per unit were placed on a fine sieve and rinsed with water until all thrips could be dislodged from the leaves onto a very fine sieve (No. 150)**, and** then thrips were washed in a salt solution. Sand and heavy materials were first removed from the bottom opening of the separatory funnel, followed by any thrips which were placed on a filter paper. A vacuum system was used to remove extra water. Adults and juveniles were counted using a microscope at a 10X or higher magnification. The number of thrips from each treatment and variety were recorded and used in the statistical analysis. Analysis of variance was used to determine the effect of thrips densities on cultivars.

2014 Study. An identical greenhouse study to 2013 was repeated in 2014. Six cotton cultivars were planted in 16-oz Styrofoam® cups. The study was deployed in a complete randomized block design with four replications. Each experimental unit contained six plants. Thrips were reared on green beans purchased from local grocery stores. Four densities of thrips released onto seedling cotton included: no thrips (control), ½ thrips per plant (one thrips per two plants), one thrips per plant, and two thrips per plant at the 1- to 2-true leaf stage. On the control plants, Orthene® 97 was applied twice weekly. Twenty-two days following the thrips releases, the plants were clipped near the soil surface and stored in 90% ethyl alcohol for thrips washing. Prior to clipping the plants, visual leaf damage rankings were conducted which were based on a 1-10 scale (1 = normal healthy plants and 10 = plants killed by thrips). Chlorophyll readings were recorded using a chlorophyll meter and leaf area from each treatment was recorded using a leaf area meter. Adult and juvenile thrips were quantified via washing technique and counted using a microscope at 10X or higher magnification. Analysis of variance was conducted to test whether thrips densities had an effect on tested variables.

Objective 3. Developing thrips economic threshold for seedling cotton

Several field cage models were designed in 2012 and 2013 for economic threshold studies, but none were effective in containing the thrips or allowing adequate ventilation inside the cage for thrips survival. Therefore, a new rectangular cage was designed and a threshold study was conducted in 2014, but we plan to generate a second year of data in 2015, if the project is funded.

Density-dependent threshold studies were conducted in seedling cotton. Rectangular wooden-frame cages [98 cm (L) x 30 cm (W) x 44 cm (H)] with No-Thrips® screen were constructed and deployed in the field, with each cage enclosing 8-13 cotton seedlings (Fig. 2). Silicone caulk was used to attach No-Thrips® screen to the wooden frame. A thin metal flashing (1-1.5") was attached at the bottom of the cage to restrict thrips movement from the bottom of the cage. A temperature sensor was kept inside the cage to record the internal cage temperatures.

Freshly collected adult thrips, primarily western flower thrips, were released at various densities to generate a damage gradient across density treatments. After the thrips were released and the plants caged, thrips were allowed to feed for 5-10 days and then the cages were removed. Two plants from each cage were removed and washed to retrieve thrips to estimate the thrips survival. Within 24 h of cage removal, thrips augmented plots were sprayed with Orthene® 97 to kill all remaining thrips. Remaining plants were kept insect-free throughout the remainder of the growing season; these plants were harvested and ginned for lint yield estimation.

Three separate studies were conducted to capture within-season variation in seedling response to various thrips density treatments. Experimental protocols were identical in all three tests.

Figure 2. Wooden-framed No-Thrips® field cage for threshold study (left); Installation of thrips cages in the field and release of thrips densities (right).

Test I. Cultivar ST 5458B2RF, without the seed treatment for thrips management, was planted on May 15, 2014. Field cages were deployed and six thrips density treatments were released onto plants on June 6 when the cotton seedlings were at the 1-2 true-leaf stage. Six density treatments included 0, 1, 2, 4, 6, and 10 thrips per plant, replicated five times (total 30 cages). Within four days of thrips release, the test site received 2.5" of rainfall. Therefore, we allowed the thrips feeding exposure to continue for about 10 days before removing the cages. Two plants from each cage were harvested and washed to retrieve thrips. After removal of cages, thrips augmented rows were sprayed with Orthene® 97.

Test II. Only 100 m from the *Test I* site, *Test II* was conducted on the same cotton cultivar using the same cages. This cotton cultivar ST 5458B2RF was planted on June 3 and the test was deployed on June 18 at the 1-2 true-leaf stage. Thrips densities included 0, 1, 2, 4, 6, and 10 thrips per plant plus an uncaged control. Cages were removed on June 23 and two plants from each cage were harvested and washed to retrieve thrips. After removal of cages, thrips augmented rows were sprayed with Orthene® 97.

Test III. Immediately after *Test II* was terminated, *Test III* was deployed in the same experimental field only 50 feet away from *Test II* using the same cages. Thrips cages were

deployed and thrips were released on June 25. Because the seedlings were at the 5-6 true-leaf stage, thrips densities were increased. Treatments included 0, 2, 4, 10, 20 and 30 thrips per plant. Cages were removed on July 1, followed by the removal of two plants per cage to retrieve thrips via plant washing, and then sprayed Orthene® 97 to kill augmented thrips on remaining plants.

Following removal of the cages, all plots were regularly monitored such that the study site could remain relatively pest-free for the remainder of the season. Plant-mapping data were collected and plants were harvested to quantify the crop response to various levels of thrips infestations.

Results and Discussion

Objective 1. Cotton cultivar response to natural colonization of thrips in the field

2013 Study. Visual thrips counts did not significantly vary between treatments or cultivars. Stand counts between treatments were also non-significant; however, plant counts were significantly higher in CT1205, CT1206, DP 353 and PHY 376 WRF compared to FM 1740B2F and SSG HQ 212 CT. Cultivars DP 353 and PHY 367 WRF had significantly more thrips in control plots than sprayed plots (Fig. 3). No significant thrips population densities or lint yield differences were found between the insecticide-treated and untreated control portions of the other four cultivars. Cultivar DP 353 had the longest flowering period and peak flowering occurred later in the season compared with the other cultivars examined (Fig. 4). Also, the flowering of SSGHQ and FM 1740 peaked on the same date, although not as high, plus both of these cotton lines/cultivars had longer flowering periods than DP 353 displayed. In both treated and control plots, the highest number of white flowers were observed in PHY 367 WRF on July 30 (Figs. 4 and 5) and peak flowering continued from mid-July through August. Several significant differences were observed between plant biomass and cultivar treatments ($P<0.1$) in control and sprayed plots (Tables 1-2); however, interactions between insecticide and cultivar treatments were non-significant. Significantly lower lint yield in untreated control plots ($P<0.1$) was observed between sprayed and control plots in DP 353 and PHY 367 WRF which might be due to presence of significantly more thrips in control plots than insecticide-sprayed plots in these two cultivars (Fig. 6). Significant differences in seed yield was observed between sprayed and control plots in DP 353; however, no significant differences in seed yield were observed between sprayed and control plots in other cultivars tested (Fig. 7).

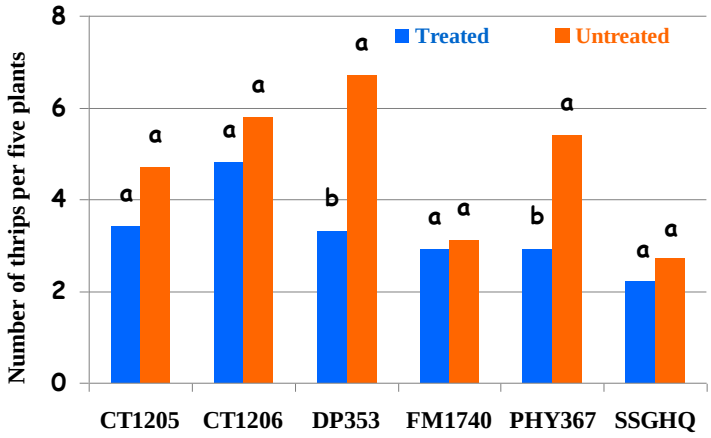

Figure 3. Thrips densities recovered using whole-plant washing procedure, 2013.

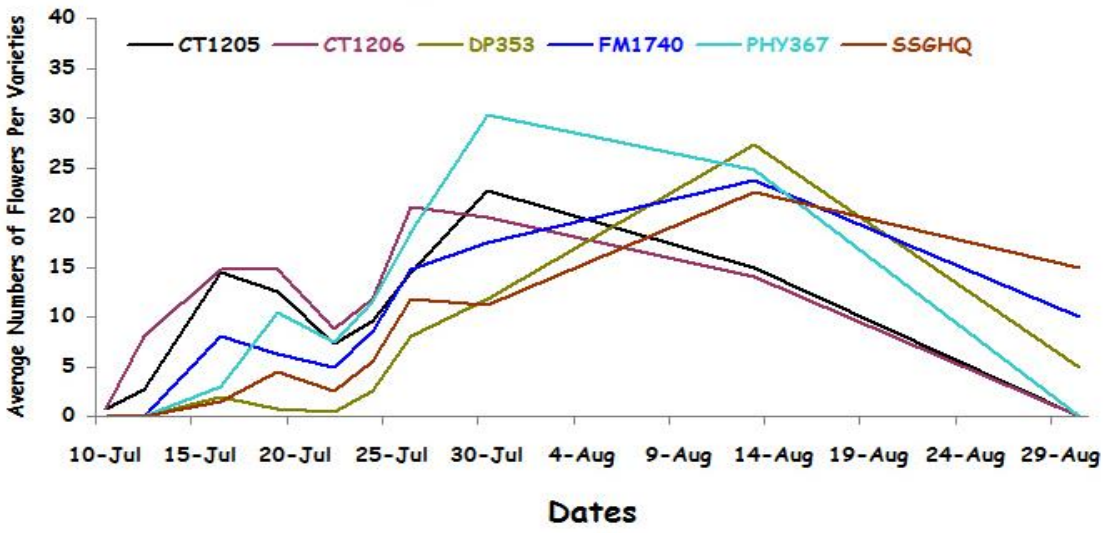

Figure 4. Flowering profile of cotton cultivars in untreated control plots, 2013.

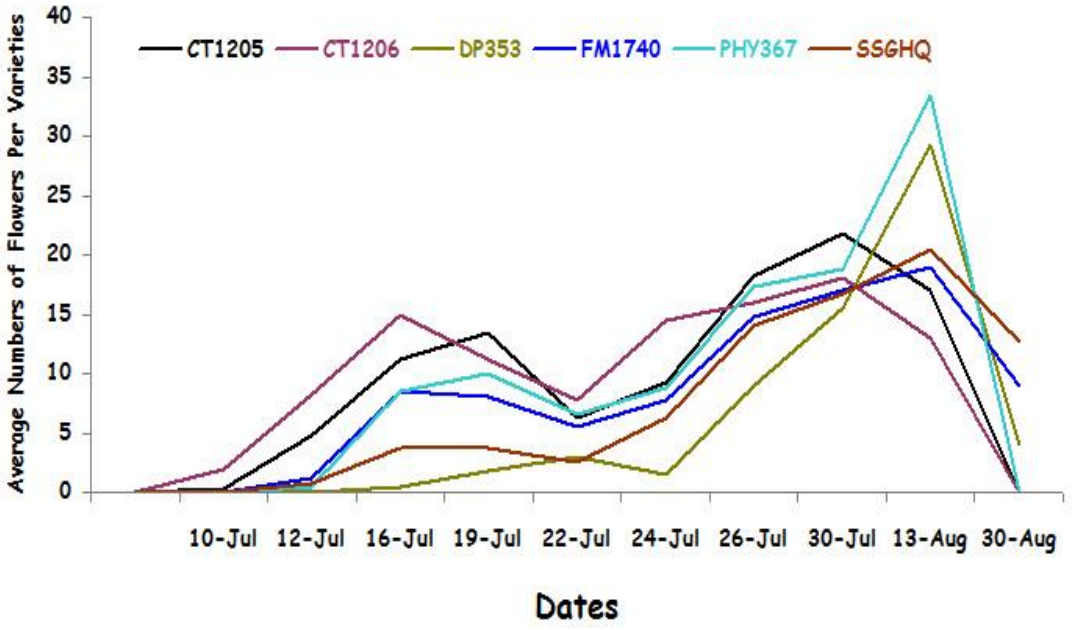

Figure 5. Flowering profile of cotton cultivars in insecticide sprayed plots, 2013.

Table 1. Variation in selected plant parameters across tested cultivars and lines in control plots, Lubbock, TX, 2013.

	Cultivars/Lines					
Plant Parameters	CT1205	CT1206	DP353	PHY367	FM1740	SSGHQ
Shoot length (cm)	9.10a	8.97a	8.32a	8.37a	7.90a	6.52a
Root length (cm)	17.35a	16.47a	14.32a	16.37a	16.25a	14.07a
Shoot biomass (g)	2.06a	2.36a	1.42ab	1.31ab	1.67ab	0.94b
Root biomass (g)	1.76ab	2.05a	1.06bc	1.20bc	1.49abc	0.93c
Leaf biomass (g)	4.69ab	5.50a	3.73ab	3.04b	3.94ab	2.56b
Leaf area (cm^2)	135.6ab	163.41a	134.19ab	103.22ab	114.86ab	85.15b
Leaf chlorophyll	54.39a	53.60a	49.75a	55.12a	55.24a	51.14a

Table 2. Variation in selected plant parameters across tested cultivars and lines in insecticide sprayed plots for thrips management, Lubbock, TX, 2013.

	Varieties/Lines					
Plant Parameters	CT1205	CT1206	DP353	PHY367	FM1740	SSGHQ
Shoot length (cm)	8.32ab	8.97ab	8.72ab	9.47a	8.25ab	6.22b
Root length (cm)	19.57a	19.19ab	15.35b	17.50ab	15.90ab	16.10ab
Shoot biomass (g)	2.88a	2.47a	1.90ab	2.23ab	1.58ab	0.88b
Root biomass (g)	2.44a	2.15a	1.40ab	2.02a	1.56ab	0.91b
Leaf biomass (g)	6.61a	6.29a	4.77ab	4.59ab	3.85ab	2.70b
Leaf area (cm^2)	163.83a	170.01a	162.86a	128.96a	111.14a	73.19a
Leaf chlorophyll	53.91a	54.38a	51.47a	54.64a	53.30a	51.10a

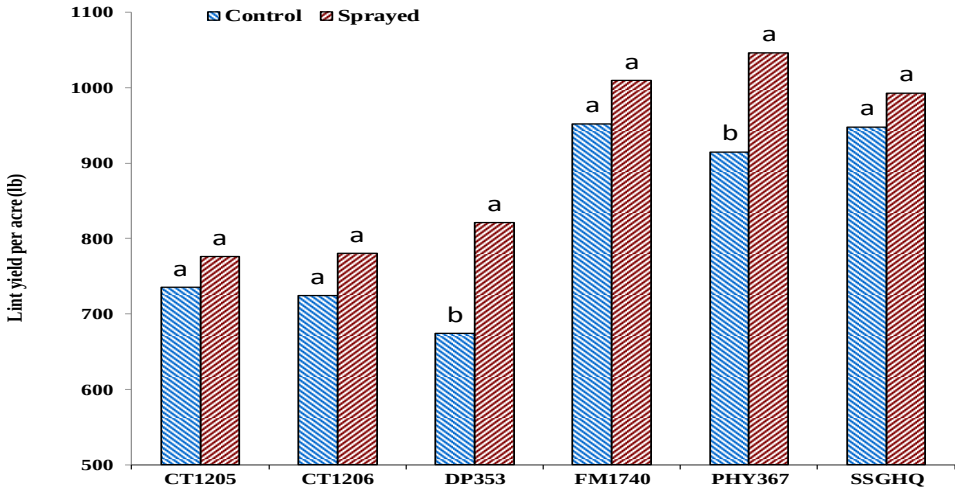

Figure 6. Lint yield (lb per acre) across tested cultivars and breeding lines, 2013.

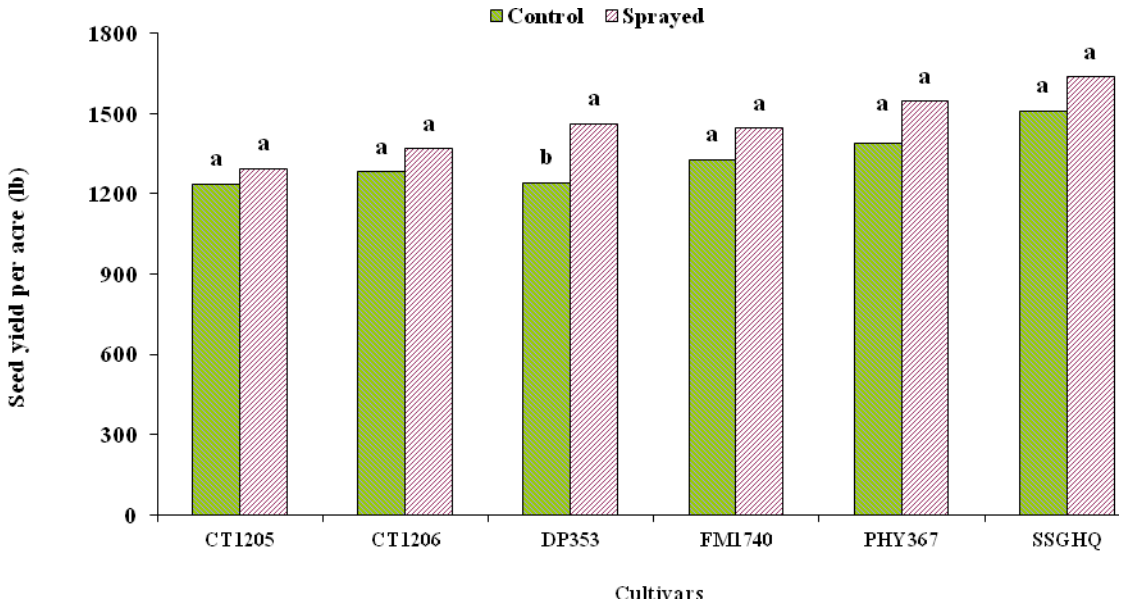

Figure 7. Seed yield (lb per acre) across tested cultivars and breeding lines, 2013.

During this study, we observed that field colonization of thrips was low and varied with cultivars, with DP 353 attracting the most adult thrips and lowest densities observed in FM 1740B2F and SSG HQ 212 CT. However, drastic cultivar difference in plant growth and yield masked the subtle difference in thrips tolerance across these tested cultivars or lines.

2014 Study. Because the plant growth was compromised (see Material and Method section above) for this study for much of the early growing season, thrips colonization did not occur. As a result, the study was reduced to a simple agronomic comparison of tested cultivars and germplasms. The test plots were harvested on December 15, 2014 and ginned on January 12, 2015. Commercial cultivars PHY 367 WRF and FM 1740B2F produced significantly higher yield than another commercial cultivar DP 353 (Fig. 8). Experimental germplasms showed average lint yield performance. As expected, seed yield followed the similar trend as for lint.

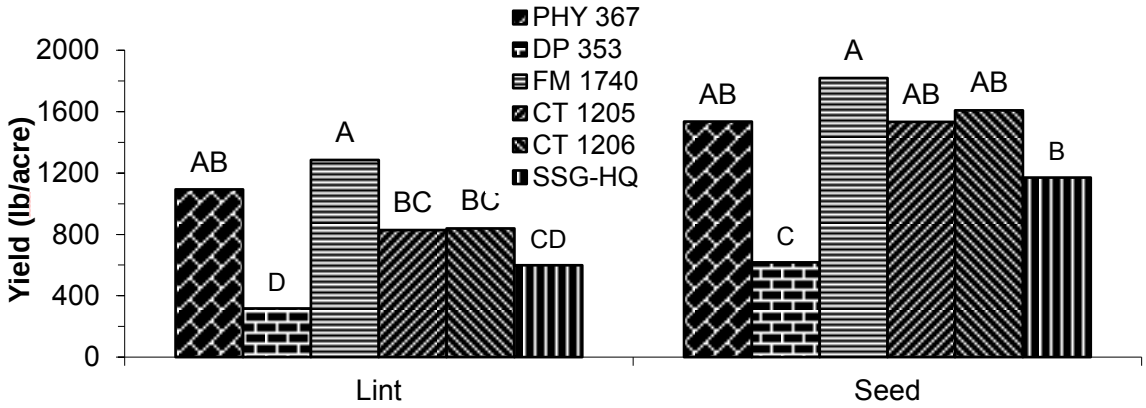

Figure 8. Lint and seed yield (lb per acre) across tested cultivars and breeding lines, 2014.

Objective 2. Cotton cultivar response to different thrips densities in the greenhouse

In 2013, several factors were significant between released thrips densities and thrips numbers recovered. Statistically different numbers of thrips (adults + immatures) were recovered between densities 0, 0.5, 1 and 2 (Fig. 9). For both adult and immature thrips numbers, thrips release density 0 had the lowest numbers of thrips retrieved compared to the thrips augmented treatments, indicating that the thrips movement across treatments was minimal. Total thrips retrieved were the highest at 1 thrips per plant treatment, followed by 2 thrips per plant, 0.5 thrips per plant, and the lowest number in uninfested treatment, all significantly different from each other (Fig. 9).

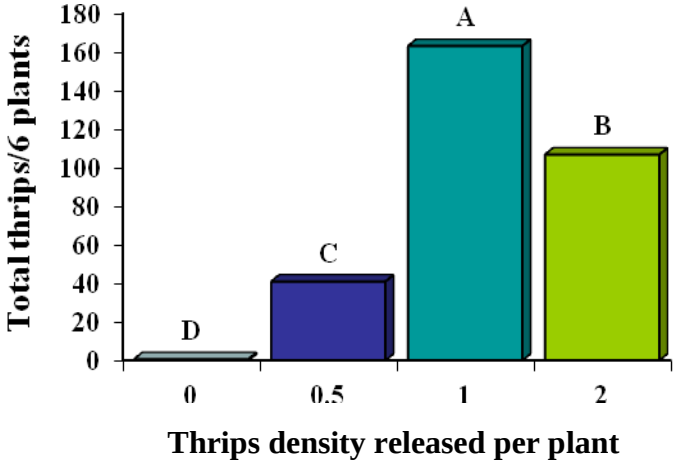

Figure 9. Recovery of total thrips (adult and immature) from seedling cotton using a plant washing technique in a greenhouse study, 2013.

Adult thrips numbers retrieved after three weeks of study were highest in the 0.5 and 1 density treatments, followed by 2 thrips released per plant, and the lowest numbers in the uninfested treatment (Fig. 10). Immature thrips densities increased to 157 and 104 per 6-plant treatments, respectively, in three weeks, whereas 0.5 thrips per plant augmentation resulted in 32 thrips per 6-plant (Fig. 11). No significant differences were found between cultivars and recovered total number of thrips (adults + immatures), immatures only or adults only. In 2014, total thrips were significantly higher in 2 thrips per plant treatment, followed by 1 and 0.5 thrips per plant treatments, and non-significant number in the uninfested treatment (Fig. 12).

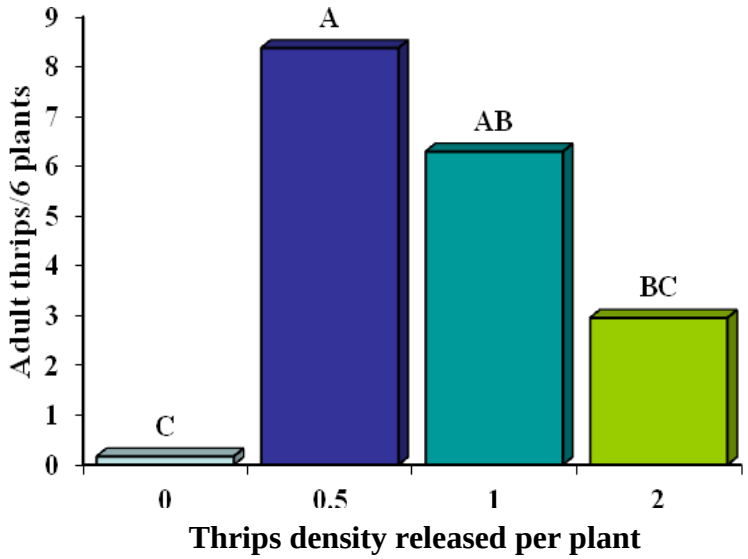

Figure 10. Recovery of adult thrips (22 days after initial thrips releases) from seedling cotton using a planting washing technique in a greenhouse study, 2013.

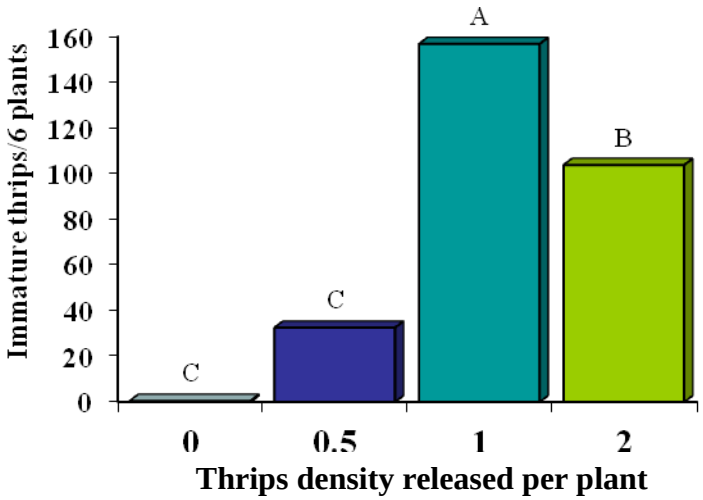

Figure 11. Recovery of immature thrips from seedling cotton using a washing technique in a greenhouse study, 2013.

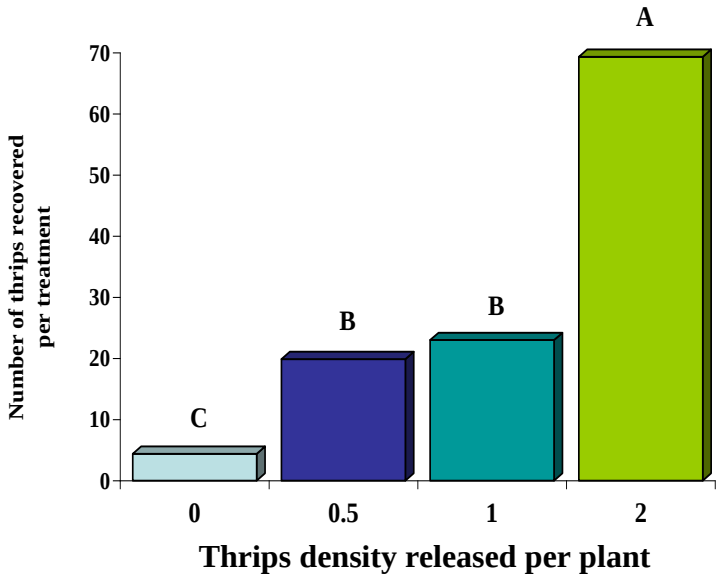

Figure 12. Recovery of total thrips (adult and immature) from seedling cotton using a plant washing technique in a greenhouse study, 2014.

*Leaf area***.** Leaf surface area measurements were significant between thrips densities 0, 0.5 and 2; however, no significant differences in leaf area were recorded between thrips release densities of 0 and 1 per plant; and densities 1 and 2 (Fig. 13). Additionally, no significant differences were found in leaf area reduced by thrips among the cultivars tested. There was a clear indication that thrips infestations, regardless of the densities, tended to reduce the leaf surface area in seedling cotton.

Visual ranking. Significant differences were observed in visual ranking of the cotton seedlings between thrips densities released (P = 0.0001); however, no significant differences (P>0.05) were recorded in visual ranking between cultivars. Visual injury ranking was significantly lower (less injury) in thrips densities 0 and 0.5 compared with that in released thrips densities 1 and 2; however, no significant differences (P>0.05) were recorded in visual ranking between thrips densities 1 and 2 (Fig. 14). It is noteworthy that 0.5 thrips per plant exerted significantly higher injury, based on visual ranking, compared with that in no-thrips control plants.

Chlorophyll readings. No significant differences were observed in chlorophyll readings of the indicator leaf (4th mainstem leaf) on seedlings between thrips densities released (P>0.05) but various significant differences (P<0.05) were recorded in chlorophyll readings between cultivars tested. In 2013, cultivar CT-1206 showed the highest chlorophyll readings, which were significantly different from ST 5458B2RF, PHY 367 WRF and SSG HQ 212 CT (Fig. 15). No significant differences (P>0.05) in chlorophyll levels were recorded among cultivars CT-1205, CT-1206 and 1740B2RF. Also, no significant differences (P>0.05) in chlorophyll levels were recorded among ST 5458B2RF, PHY 367 WRF and SSG HQ 212 CT (Fig. 15). In 2014, chlorophyll readings were higher on CT lines and FM 1740B2F compared with the remaining lines/cultivars. Chlorophyll readings, on general numerical trends, were consistent between 2013 and 2014 across cultivars tested, except for SSG HQ 212 CT (Fig. 15).

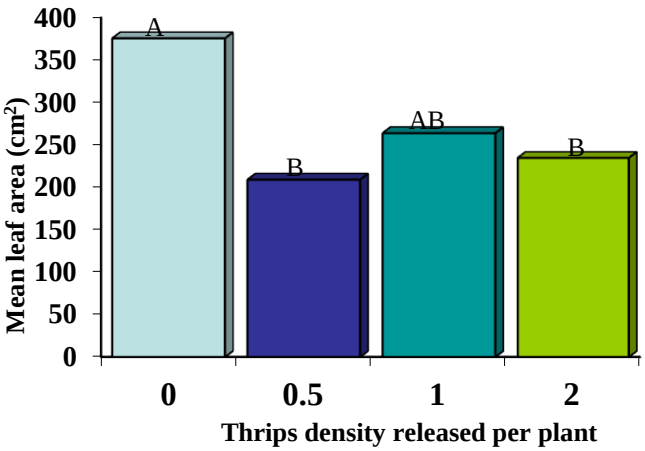

Figure 13. Effect of western flower thrips injury on leaf surface area of the cotton seedlings at various thrips densities in a greenhouse study, 2013.

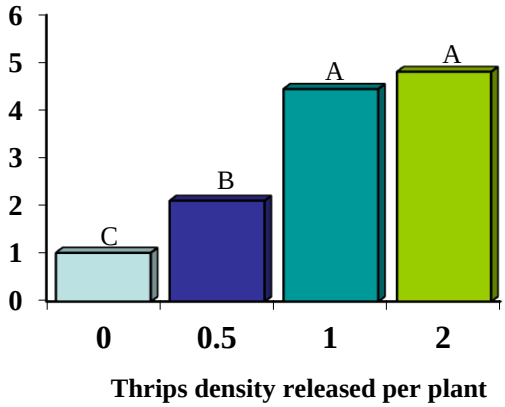

Figure 14. Effect of western flower thrips injury on visual leaf damage ranking of the cotton seedlings at various thrips densities in a greenhouse study, 2013.

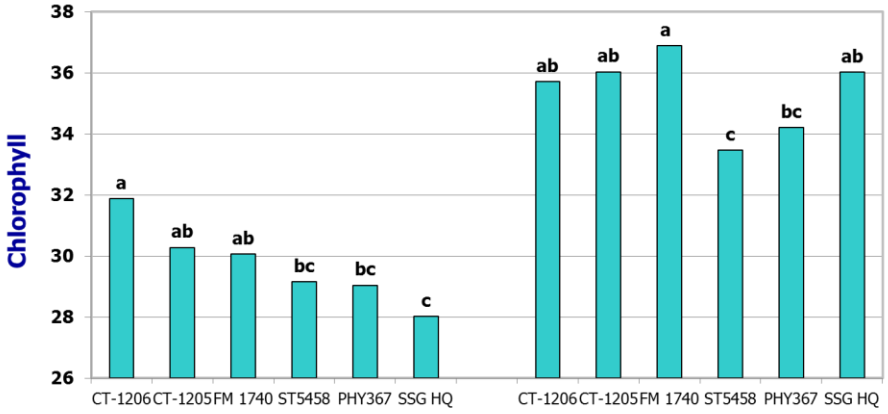

Figure 15. Effect of western flower thrips injury on chlorophyll readings of the cotton seedlings of selected cultivars in a greenhouse study, 2013 (left panel) and 2014 (right panel).

Objective 3. Determine the cotton crop damage potential of the western flower thrips for developing an economic threshold

No-thrips® cages appeared to contain thrips in the field cages better than any of the other field cage materials (fabrics) that we have used in previous studies. Different materials and designs were used in the past, including 1) transparent plastic cup cage, 2) wire mesh sleeve cage, 3) opaque plastic cylinder, 4) transparent plastic jar without ventilation, and 5) transparent plastic jar with ventilation (Fig. 16). None of these methods were suitable for thrips studies in the field because of the excessive temperature buildup inside the cages, plus material of the screen was unable to contain the thrips. However, the No-Thrips® cage design provided a satisfactory performance.

Figure 16. Cage types evaluated previously: 1) transparent plastic cup cage, 2) wire mesh sleeve cage, 3) opaque plastic cylinder, 4) transparent plastic jar without ventilation, and 5) transparent plastic jar with ventilation.

Despite our preliminary study showing a satisfactory thrips retention in the No-Thrips® cage, 5-day post-release thrips retrieval was much lower than expected in all three studies. We speculate that a frequent rain and cool/wet weather might have attributed to this lower thrips retrieval from the cages. It is also possible that there might have been a greater mortality once they were released into the cages. We do not believe that the large number of thrips escaped from the cages, but a small number of escapes is always a possibility. Despite the low rate of retrieval, it appears that the thrips feeding had exerted some effect on the plants, resulting in reduced yield. On the first test, all five thrips augmented treatments had lower average lint yields (749 lb/acre in 6 thrips/cage treatment to 964 lb/acre in 4 thrips/cage treatment) compared to that in control cages (1145 lb/acre), although the values were not statistically significant owing to a large variance in the data (Fig. 17). Test II also suggested that thrips feeding occurred, resulting in lower plant height and smaller main-stem diameter in all thrips augmented treatments compared to that in control cages (Fig. 18). Nevertheless, the thrips feeding, if any, during the seedling stage in this study did not significantly impact lint yields (Fig. 18). Test III was conducted when plants were near the end of the thrips tolerant stage: 5-6 true-leaf stage with good crop health. Therefore, a significant yield-reducing effect of thrips augmentation was not expected. Nevertheless, thrips augmented treatment cages had numerically lower yield compared to that in control cages (Fig. 19).

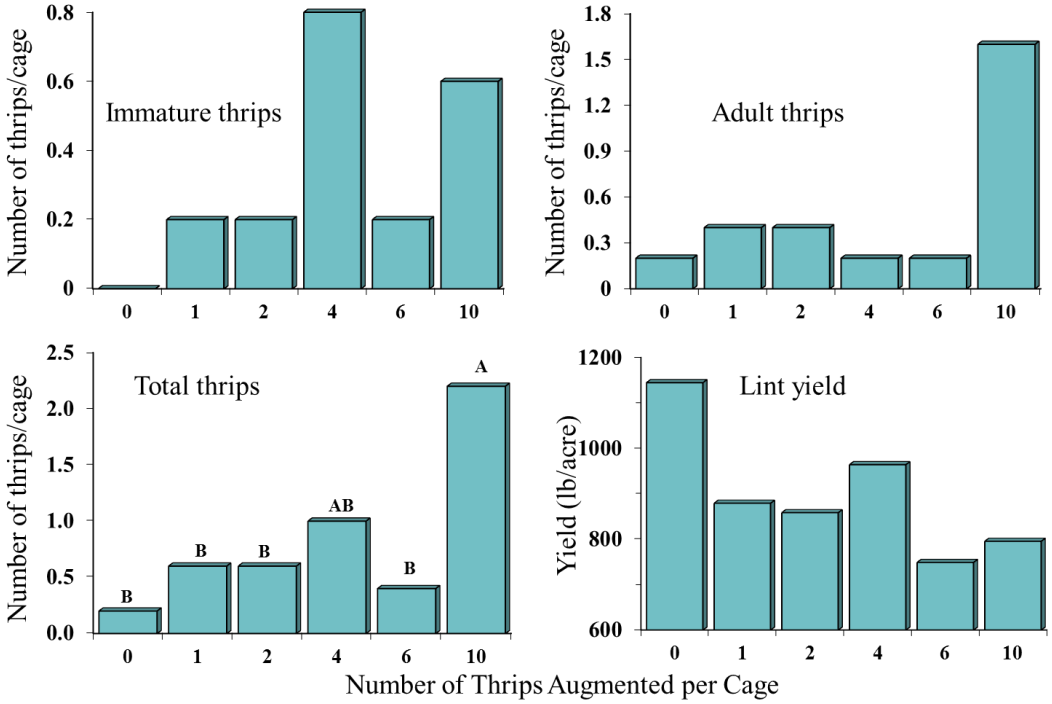

Figure 17. Number of thrips recovered at 5-day post-release into field cages and lint yield from cotton infested with varying densities of thrips in No-Thrips® cages during the 1-2 true-leaf stage, Lubbock, Texas, 2014 (*Study I*).

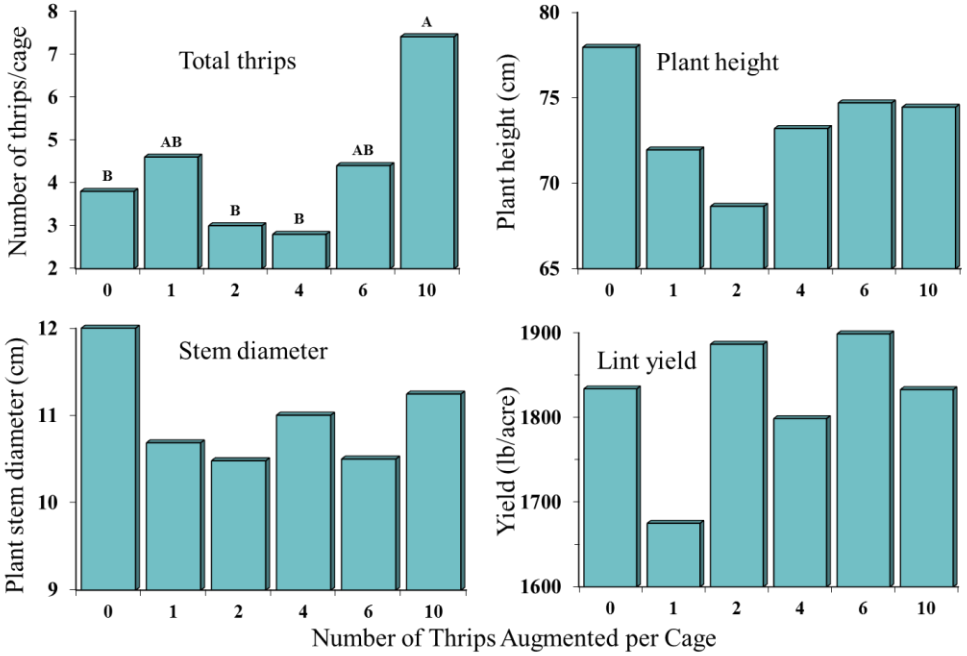

Figure 18. Number of total thrips (immatures plus adults) recovered at 5-day post-release into the field cages, plant height, stem diameter, and lint yield from cotton infested with varying densities of thrips in No-Thrips® cages during the 1-2 true-leaf stage, Lubbock, Texas, 2014 (*Study II*).

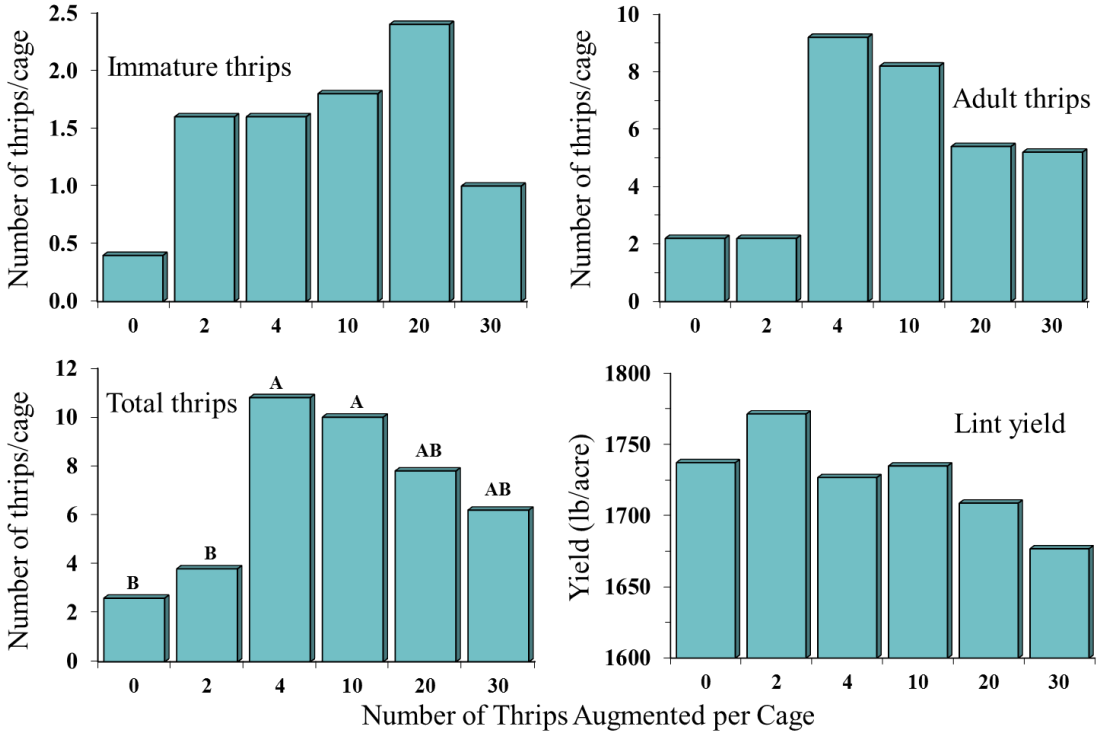

Figure 19. Number of thrips (immature, adult, and total) recovered at 5-day post release into the field cages and lint yield from cotton infested with varying densities of thrips in No-Thrips® cages during the 5-6 true-leaf stage, Lubbock, Texas, 2014 (*Study III*).

The 2014 threshold studies showed that thrips density-dependent threshold studies can be conducted in the Texas High Plains using the No-Thrips® cages. However, several design modifications may be necessary to accomplish the stated objectives in 2015. We plan to repeat the 2014 study with identical protocol for adult thrips, except that the thrips will be pre-conditioned in the laboratory on seedling cotton before they are released into the field cages. In 2014, thrips were collected from adjacent alfalfa and directly released onto cotton seedlings. It is possible that the thrips from alfalfa were not adapted to cotton seedlings, in addition to other weather factors that contributed to lower thrips survival/feeding performance. Also, we plan to conduct the threshold studies using immature thrips via releasing thrips on select cotton rows in open field without caging the plants. For this purpose, thrips will be collected from adjacent cotton and immature thrips will be separated from the collection, provision them on seedling cotton for 1-2 days, and then release them into cotton seedlings in open field. The thrips released sections of cotton rows will be sprayed with insecticides 5 days after thrips augmentation to kill the thrips. Both cage studies (adults) and open-field studies (immatures) should allow us to generate appropriate density-dependent feeding data to develop thrips management thresholds for Texas High Plains cotton.

References

Albeldaño, W. A., J. E. Slosser, and M. N. Parajulee. 2008. Identification of thrips species on cotton in the Texas Rolling Plains. *Southwestern Entomologist* 33: 43-51.

Cook, D., A. Herbert, D. S. Akin, and J. Reed. 2011. Biology, crop injury, and management of thrips (Thysanoptera: Thripidae) infesting cotton seedlings in the United States. *Journal of Integrated Pest Management* 2: 1-9.

Parajulee, M. N., R. B. Shrestha, and J. F. Leser. 2006. Influence of tillage, planting date, and *Bt* cultivar on seasonal abundance and within-plant distribution patterns of thrips and cotton fleahoppers in cotton. *International Journal of Pest Management* 52: 249-260.

Sadras, V. O., and L. J. Wilson. 1998. Recovery of cotton crops after early season damage by thrips. *Crop Science* 38: 399-409.

Harp, S. J., and V. V. Turner. 1976. Effects of thrips on cotton development in the Texas Blacklands. *Southwestern Entomologist* 1: 40-45.

Williams, M R. 2013. Cotton Insect losses-2012, pp. 546-586. *In* Proceedings, Beltwide Cotton Conf., National Cotton, San Antonio, TX.

COTTON FLEAHOPPER DAMAGE ON WATER-STRESSED COTTON

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Abstract

Cotton fleahopper, *Pseudatomoscelis seriatus*, can cause excessive loss of cotton squares, resulting in reduced yield and harvest delays. Field testing during drought conditions provided opportunity to assess insect activity in a high contrast of dryland and irrigated conditions. Plant water stress affected natural cotton fleahopper populations (South Texas study: increasing more in irrigated plots) and water stressed plants were more sensitive to equal cotton fleahopper pressure (High Plains study: lint loss and possibly boll load decreasing more in low irrigation plots). As seen last year, plant development stage at the time of initial cotton fleahopper infestation was crucial, with early squaring cotton having higher densities than cotton at early bloom in the infestation (South Texas study). For field application, detection of fleahoppers in early planted cotton may serve as early warning of cotton fleahoppers in later-planted cotton. As the infestation progresses, fleahoppers may persist better in cotton with low water stress. But the greatest potential for yield decline from cotton fleahopper was when cotton was water stressed and infestations occurred during pre-bloom squaring. Understanding how water stress contributes to cotton fleahopper fluctuations may allow better estimation of cotton risk from cotton fleahopper damage.

Introduction

Cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter) (Hemiptera: Miridae), has been documented to cause excessive loss of cotton squares in Texas and Oklahoma, resulting in reduced yield and harvest delays. Cotton fleahopper is also an occasional pest in New Mexico, Arkansas, Louisiana, and other mid-south states. Within Texas, regional average cotton fleahopper induced yield loss estimates vary, reaching up to 6% (Williams 2000). A challenge to management is that square loss and subsequent yield loss to individual fields varies considerably as populations build.

This variability has been partly associated with cultivar differences and other host plant factors (Holtzer and Sterling 1980, Knutson et al. 2009, Barman et al. 2011), with the stage of cotton development when movement into the field occurs (Parajulee et al. 2006), and with environmental stressors, in particular plant water stress (Stewart and Sterling 1989). Even though foliar insecticide application may control the population, benefits to control may depend on these factors.

Understanding the degree to which these factors contribute to cotton fleahopper fluctuations and subsequent plant damage may allow better estimation of cotton risk from cotton fleahopper leading to improved in-season management (i.e., insecticides).



From left to right: cotton fleahopper, a blasted square (damage), and a healthy square. Photos provided by authors and Texas AgriLife Research, Lubbock and Corpus Christi.

Materials and Methods

We hypothesize that plant water stress and plant vigor, and plant development at the time of infestation are main factors that affect cotton fleahopper population fluctuation and plant response/yield loss. These factors were considered in two studies, one in South Texas, and the second in the Texas High Plains.

Field testing in 2013 during drought conditions provided opportunity to assess insect activity in a high contrast of dryland (with supplemental irrigation due to severe drought) and irrigated (irrigation targeting 90% crop ET replacement) water regimes. The South Texas location focused on following a natural cotton fleahopper population and subsequent yield in a plot with two water regimes, two planting dates, two cultivars, and controlled with insecticide or not. The Texas High Plains location focused on plant response using an augmented population of cotton fleahopper under two water regimes. Details of the experimental layout at each location follow:

South Texas - Corpus Christi - Texas A&M AgriLife Research & Extension Center

A split-split-split plot design was implemented with 5 replications. The main plot was two water regimes, 1) low irrigation during drought (6.1 acre-inch for the earlier planting, 7.9 acre-inch for the later planting) and 2) high irrigation during drought (10.4 acre-inch for the earlier planting, 13.8 acre-inch for the later planting). The 1st split was two planting dates; Earlier (May 6) and Later (May 31), with both planting dates being agronomically late for the region. The 2nd split was two cotton cultivars; PhytoGen 367 WRF (Dow AgroSciences) and Stoneville 5458 B2RF (Bayer CropScience). The 3rd split was insecticide treatment using Centric 40 WG (thiamethoxam, Syngenta Crop Protection) at a rate of 1.25 oz/acre on June 11, 1, July 3, and 15. Irrigation was delivered by above ground drip. Insect counts were made on a weekly basis for 9 weeks after fleahopper numbers exceeded 10 bugs per 100 plants using a beat bucket technique. A total of 20 plants were sampled per plot. Plant data included yield (lbs. lint/A) as well as boll load and plant height for the unsprayed plots.

Texas High Plains - Lamesa

The plot design was a 2 by 2 factorial with 3 replications. The 1st factor was irrigation at 2 levels: a low rate in drought (4.5 acre-inch) and a high rate in drought (9.0 acre-inch). The 2nd factor was infestation rate: a control (no infestation) and 5 nymphs/plant at the 3rd week of squaring. Infestations were applied to uniform-sized plants. Plot size was 45 ft by 4 rows, and irrigation was by center pivot. Because cotton fleahopper populations were very low, the infestation was augmented with a specific and acute insect feeding pressure of 5 nymphs/plant at the 3rd week of squaring. Plant data included yield (lbs. of lint/A) and boll load (bolls/plant).

All measurements were analyzed with ANOVA, conforming to a split-split-split plot design in Corpus Christi, and a 2 by 2 factorial in Lamesa. Count data were transformed by the square root of the count + 0.5.

Results

South Texas

Fleahoppers exceeded an ET of 15% of plants infested. More cotton fleahoppers were seen on earlier planted cotton ($P < 0.0001$), especially early in the infestation (June 27 when the earlier planted cotton was at 3rd week of squaring and the later planted cotton was at the 1st week of squaring). Cotton fleahopper density did not differ between dryland and irrigated plots at the beginning of the infestation (June 27, $P = 0.24$) (Fig. 1), but as the infestation progressed more fleahoppers were detected in irrigated plots on July 3 ($P = 0.04$) (Fig. 2) and on irrigated plots of the earlier planted cotton on July 11 ($P = 0.009$) (Fig. 3). Cultivar differences were also detected, supporting historical claims of cultivar effects ($P = 0.005$) (Figs. 1-3). The insecticide Centric controlled fleahopper well across most conditions ($P < 0.0001$) (Figs. 1-3), including the very high populations found on June 27 in the earlier planting during the 3rd week of squaring (Fig. 1).

There was a good yield response with the best yields seen under irrigation for both cultivars, planting dates, and with or without insecticide protection ($P = 0.0008$) (Fig. 4). The benefits of good soil moisture were seen on unsprayed plots, which had higher bolls loads (Fig. 5) on taller plants (Fig. 6). Yield also increased when plots were sprayed, but to a much smaller degree ($P = 0.05$), and the later planted cotton (which had fewer cotton fleahoppers) had higher yield than earlier planted cotton ($P = 0.006$) (Fig. 4).

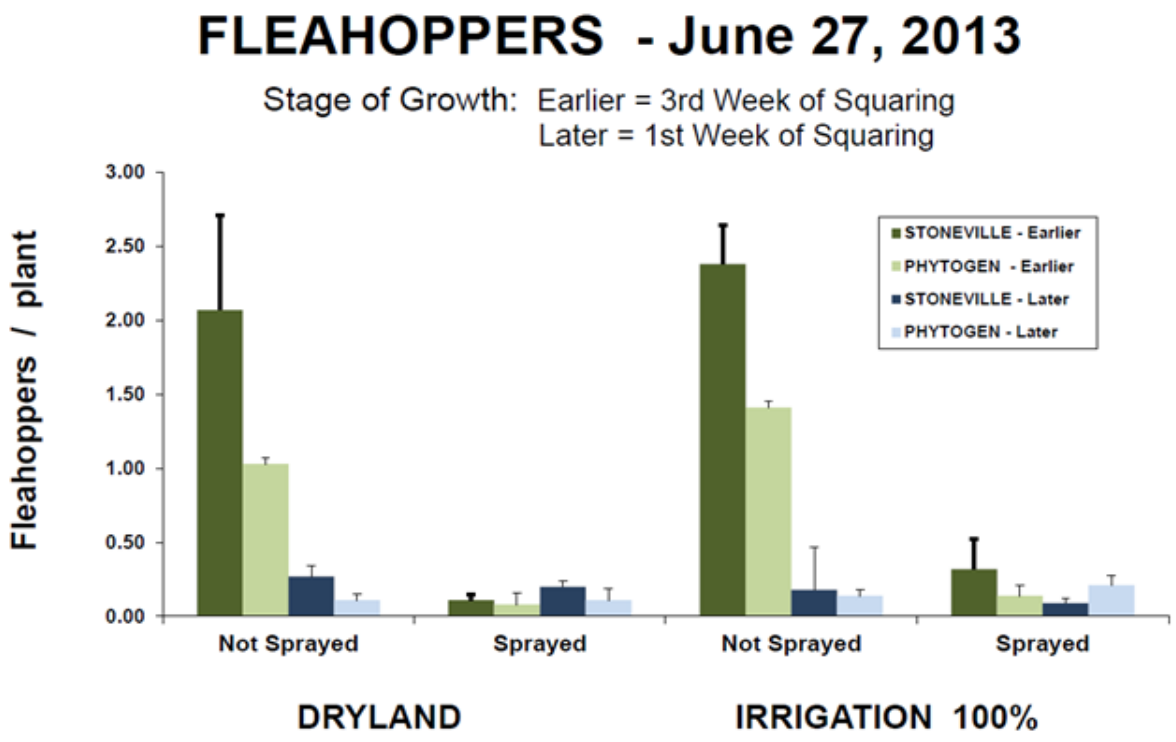

Figure 1. Number of cotton fleahoppers per plant for two sprayed and not sprayed cotton cutivars under two water regimes and two planting dates on June 27, 2013, Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas, 2013.

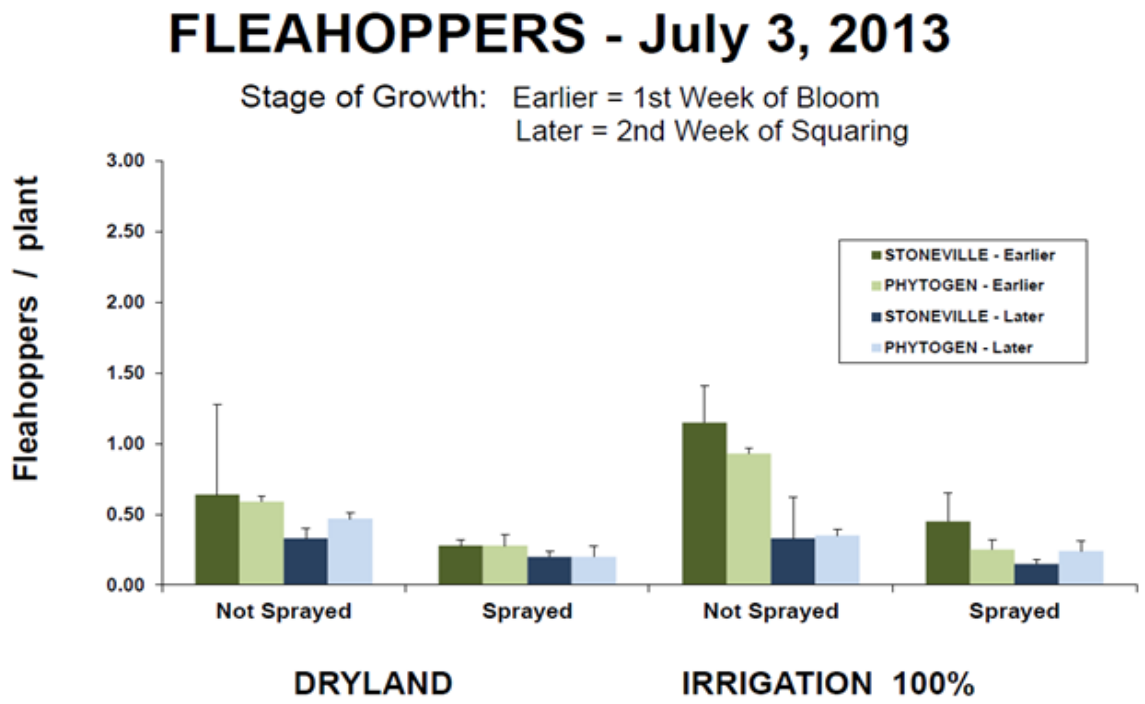

Figure 2. Number of cotton fleahoppers per plant for two sprayed and not sprayed cotton cutivars under two water regimes and two planting dates on July 3, 2013, Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas, 2013.

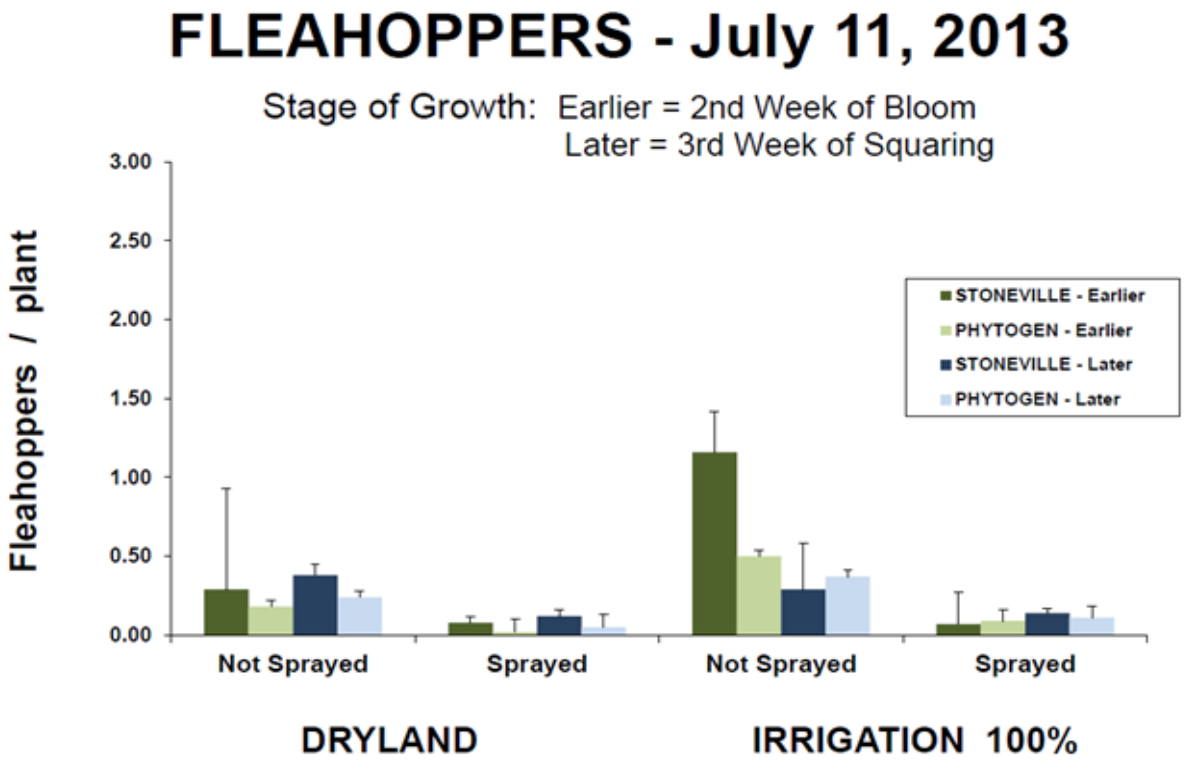

Figure 3. Number of cotton fleahoppers per plant for two sprayed and not sprayed cotton cutivars under two water regimes and two planting dates on July 11, 2013, Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas, 2013.

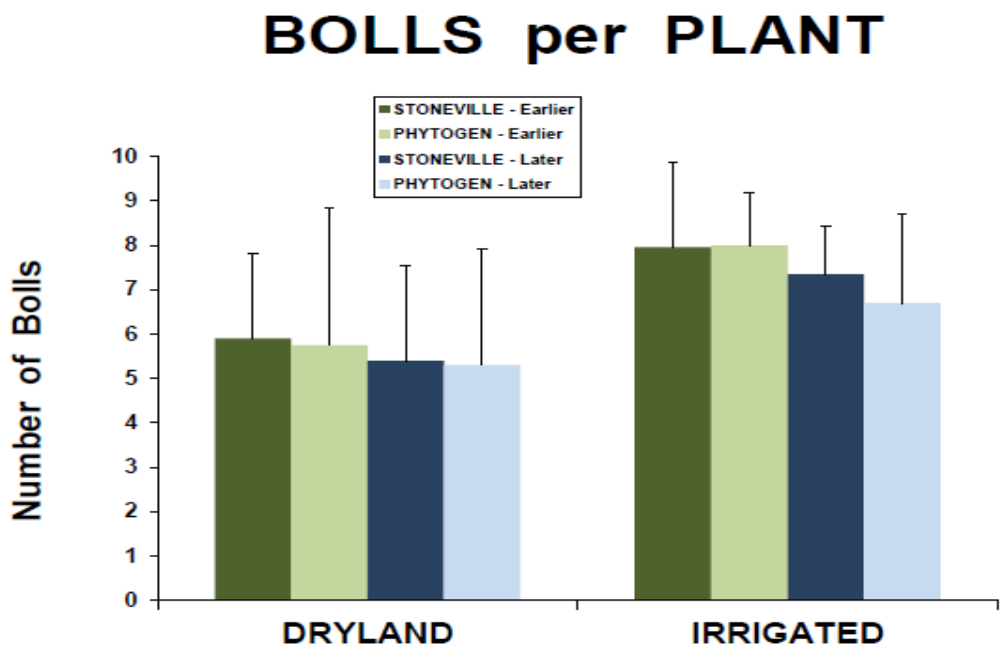

Figure 4. Number of bolls per plant for cotton cutivars under two water regimes, two planting dates, and not sprayed with insecticide, Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas, 2013.

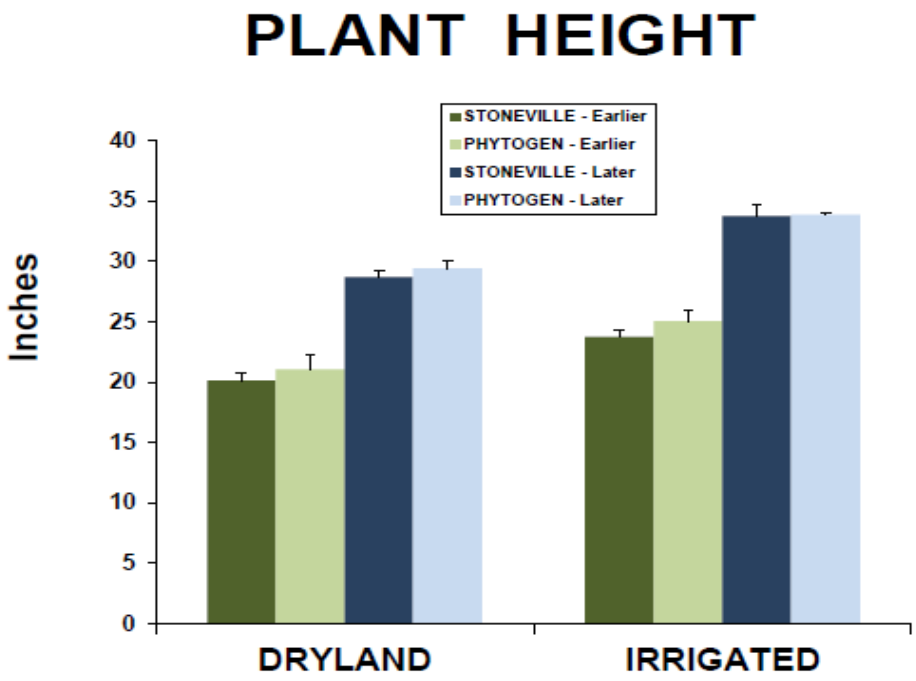

Figure 5. Plant height of two cotton cutivars under two water regimes and two planting dates, and not sprayed with insecticide, Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas, 2013.

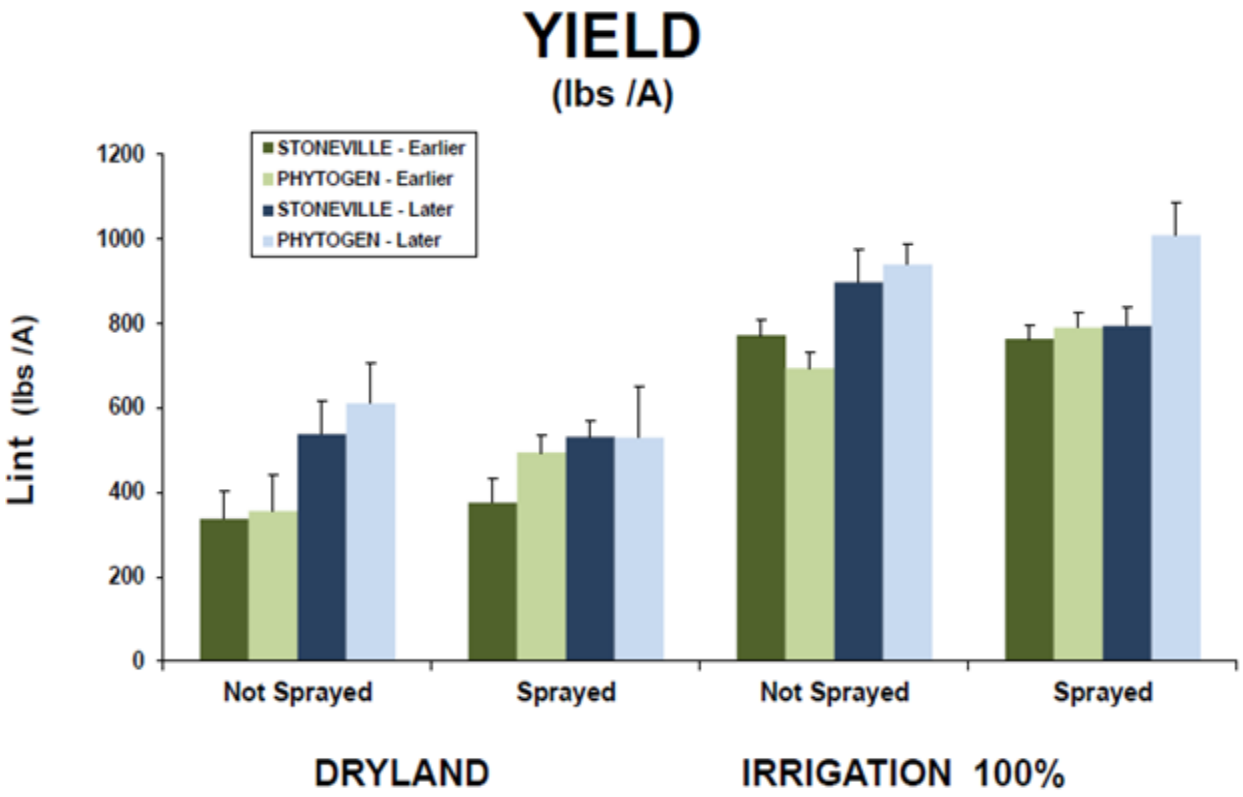

Figure 6. Yield (lbs. of lint/acre) for two sprayed and not sprayed cotton cutivars under two water regimes and two planting dates, Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas, 2013.

Texas High Plains

Natural populations of cotton fleahopper were low at this site which allowed field comparison of plant response to a specific and acute cotton fleahopper insect feeding pressure of 5 nymphs/plant at the 3rd week of squaring (fleahopper augmented) and a control (no augmentation of cotton fleahopper). This plant growth stage has been shown to host cotton fleahopper well. When plants were not water stressed (high irrigation), there was no effect of cotton fleahopper pressure based on boll load (Fig. 7) and lint yield (Fig. 8). But under water stress (low irrigation during a drought year), there was yield loss due to cotton fleahopper pressure ($P < 0.05$) (Fig. 8), which was also reflected in reduced boll load (although not significantly different) (Fig. 7).

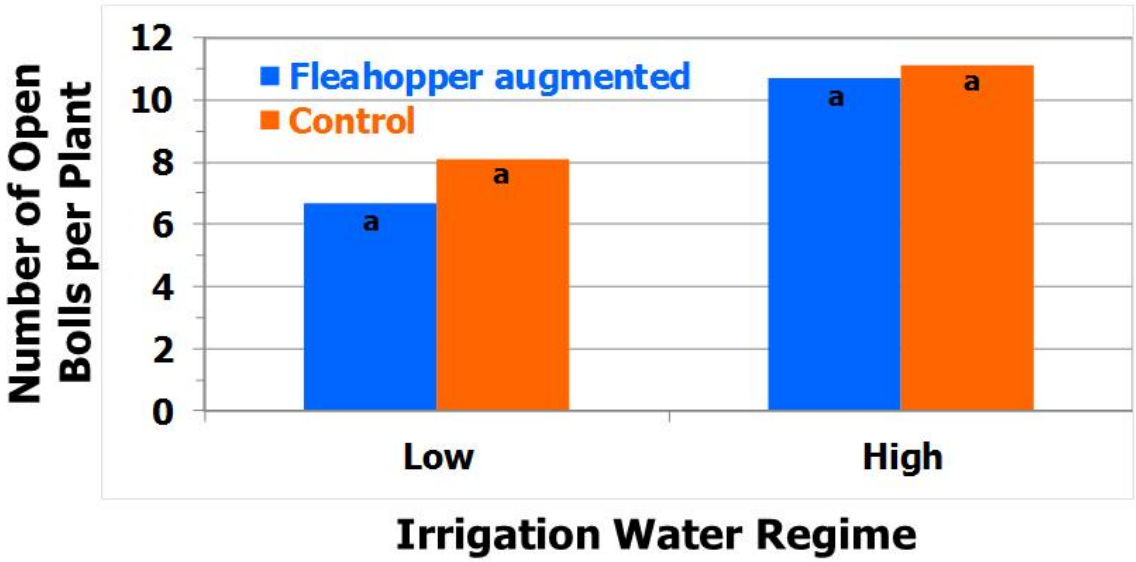

Figure 7. Number of open bolls per plant under low and high irrigation with and without (control) an augmented population of cotton fleahopper (fleahopper augmented) of 5 nymphs/plant released at the 3rd week of squaring, Texas A&M AgiLife Research, Lamesa, Texas, 2013.

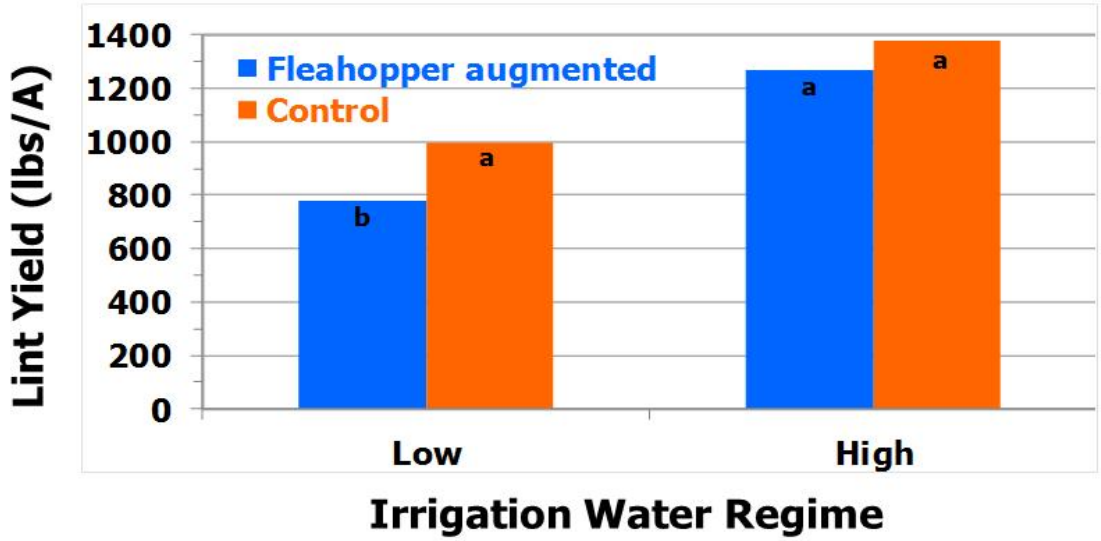

Figure 8. Yield (lbs. of lint/acre) under low and high irrigation with and without (control) an augmented fleahopper population (fleahopper augmented) of 5 nymphs/plant released at 3rd week of squaring, Texas A&M AgiLife Research, Lamesa, Texas, 2013.

Conclusions

We live in a climate that produces highly variable weather, as seen in drought conditions in Texas the last two years. Plant water stress affects natural cotton fleahopper populations (South Texas study: increasing more in irrigated plots) and water stressed plants are more sensitive to equal cotton fleahopper pressure (High Plains study: lint loss and possibly boll load decreasing more in low irrigation plots). As seen last year, plant development stage at the time of initial cotton fleahopper infestation is crucial, with early squaring cotton having higher densities than cotton

at early bloom in the infestation (South Texas study). For field application, detection of fleahoppers in early planted cotton may serve as early warning of cotton fleahoppers in later-planted cotton. As the infestation progresses, fleahoppers may persist better in cotton with low water stress. But the greatest potential for yield decline from cotton fleahopper was when cotton was water stressed and infestations occurred during pre-bloom squaring.

Acknowledgements

Many thanks to L. Pruter, J. Glover, C. Farias, and A. Cartwright for assistance in field data collection. We thank R. Kurtz for discussions as we developed this study. Cotton Inc. Core Program funds (project 11-952) were critical in launching this project.

References

Barman, A.K., M.N. Parajulee, C. Sansone, and R.F. Medina. 2011. Population level genetic variability of cotton fleahopper (*Pseudatomoscelis seriatus*) in the United States. Beltwide Cotton Conf., Atlanta, GA.

Holtzer, T.O., and W.L. Sterling. 1980. Ovipositional preference of the cotton fleahopper, *Pseudatomoscelis seriatus*, and distribution of eggs among host plant species. Environ. Entomol. 9: 236 240.

Knutson, A., C.W. Smith, and M. Campos. 2009. Project 02-266, Agronomic evaluation of intra and interspecific introgression populations of Upland cotton. Report to Cotton Inc.

Stewart, S.D., and W.L. Sterling. 1989. Causes and temporal patterns of cotton fruit ascission. J. Econ. Entomol. 82: 954-959.

Williams, M.R. 2000. Cotton insect loess estimates—1999, pp. 884-887. In Proc.-Beltwide Cotton Conf., San Antonio, TX, 4-8 Jan.

MANAGING THRIPS IN ORGANIC COTTON WITH HOST PLANT RESISTANCE AND SPINOSAD INSECTICIDE

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Abstract

Thrips are a recurring problem to seedling cotton in the Texas High Plains. It has been estimated that thrips impact to the High Plains cotton industry in 2010 was in excess of $6 million. A replicated trial, evaluating 4 cotton cultivars, 2 experimental cultivars, a susceptible check, and a commercial standard was conducted near Muleshoe, TX. Plots were split into 2 foliar regimes, spinosad (Entrust®) at 2 oz/acre and unsprayed. In general, thrips pressure was moderate. Spinosad insecticide reduced thrips pressure, and subsequent applications appear to be additive. Cultivars did not differ in thrips colonization, but the experimental cultivars did have a significant impact on thrips damage. These data suggest that these cultivars do not express host plant resistance but may have more tolerance to thrips compared to commercial varieties.

Introduction

Thrips are a recurring problem to seedling cotton in the Texas High Plains where the dominant species is western flower thrips, *Frankliniella occidentalis* (Pergande). More acres of cotton were infested by thrips than any other pest in 2012; in addition more cotton acres were treated for thrips than all other pests combined. It has been estimated that thrips impact to the High Plains cotton industry in 2010 was in excess of $6 million. In irrigated cotton where thrips populations are historically high (usually areas where there is a significant acreage of wheat), many conventional growers may choose to utilize preventative insecticide seed treatments and/or foliar remedial insecticide treatments to suppress thrips. One of the most challenging factors facing organic cotton producers in the Texas High Plains is the effective management of early-season thrips in an organic production system. In 2011 we investigated the efficacy of 13 Organic Materials Review Institute (OMRI) approved insecticides at various rates and combinations for thrips suppression in cotton (Aza-Direct, Bugitol, Cedar Gard, Ecotec, Entrust, Pest Out, Pyganic, Saf-T-Side, SucraShield, and Surround). In 2012 we continued the study but reduced the treatment list to only those products which showed potential to provide significant thrips suppression in 2011 (Aza-Direct, Bugitol, Entrust, and Saf-T-Side+Ecotec). Entrust proved to be most effective in suppressing thrips in 2012 and was selected for continued testing in 2013 along with 3 cultivars with varying degrees of host plant resistance (tolerance) to thrips and a susceptible check. Organic Materials Review Institute (OMRI) provides organic certifiers, growers, manufacturers, and suppliers an independent review of products intended for use in certified organic production, handling, and processing.

Materials and Methods

This trial was conducted in a commercial organic cotton field in Bailey County near Muleshoe, TX. Historically, western flower thrips have been the dominant thrips species infesting cotton in this area. The trial was planted 13 May, 2013 on 30-inch rows with a John Deere MaxEmerge planter equipped with cone planting units and irrigated using a low elevation spray application (LESA) center pivot irrigation system. Plots were 4-rows wide × 55 ft long and were arranged in a split-plot design with 4 replicates. Treatments included 4 cotton cultivars, two experimental, (07-7-1407 and 07-7-1020), a susceptible check (AT Atlas), and the industry standard (FM 958). Each cultivar plot was split into untreated and treated plots; spinosad (Entrust®) was applied to treated plots at 2 oz/acre. The insecticide application was applied in accordance with label recommendations at 26.4 gallons/acre (GPA) total volume and included AgAid, an OMRI approved adjuvant, at 8oz/100 gallons of water. Three insecticide applications were made weekly, beginning at near 100% emergence, 28 May. Treatments were applied in a 15 inch

band directly over the top of the crop row with a CO2 pressurized backpack sprayer and hand held boom equipped with hollow cone nozzles. The crop stage was noted and thrips were counted at crop emergence and 7, 14, 17, and 21 days after emergence (DAE); all counts were made prior to insecticide applications. Thrips counts were made by collecting ten plants/plot and washing in an alcohol solution; adult and immature thrips collected in solution were filtered out and counted under a dissecting stereo scope. Thrips samples collected were also separated by life stage. Plant damage ratings were assessed at 14 and 21 DAE, the rating scale ranged from 1 to 5, where a rating of 1 indicates no damage and a rating of 5 indicates severe damage. Leaf area was estimated 7, 14, and 21 DAE by collecting 10 plants per plot and measuring the leaf area per plant using a LI-COR, Inc. LI-3100 laboratory area meter. Data were subjected to analysis of variance (ANOVA) and when a significant F test was observed, mean separation was performed using the least significant difference (LSD) at the 5% probability level. Thrips days were calculated by following the methodology described by Ruppel (1983; J. Econ. Entomol. 76:2, pp. 375-377).

Results and Discussion

Environmental conditions at the trial site were windy with temperatures near normal to slightly above normal (Figure 1). Three separate rain events occurred June 3, 6, and 8; a nearby NOAA weather station recorded .38, .88 and .97 inches respectively. Thrips pressure, in general, was moderate. Much of the area wheat, which is an alternative host that normally supports and bridges thrips populations until cotton emergence, had desiccated prematurely due to extreme winter and early spring environmental conditions limiting early season populations.

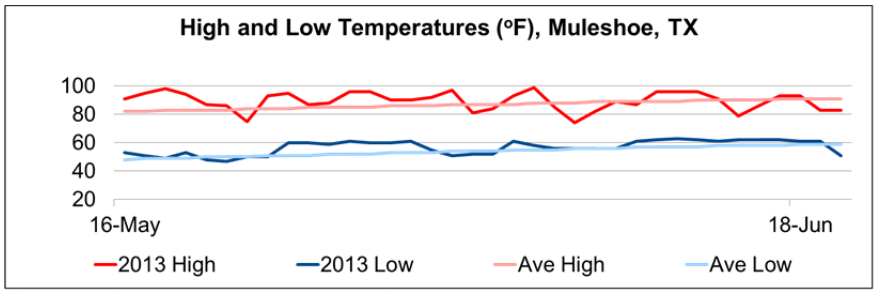

Figure 1. High and low temperatures from 2013 vs. the 30 year long term averages (1980-2010).

The cotton was slow to emerge, 15 days were required to attain near 100% emergence on 28 May and an additional 7 days from emergence until a trial average of 1.5 true leaves had developed on 4 June. Mean thrips numbers of untreated plots were less than 50% of action threshold when the initial insecticide application was applied (28 May, 100% emergence) but was over 2X the established action threshold of one thrips per true leaf by 7 DAE, and maximum pressure, 8X action threshold, was reached by 17 DAE (14 June) (Figure 2). No differences in thrips densities were observed at any sample date when comparing cotton cultivars within insecticide treatments. A significant difference was only observed when comparing all treatments at the 4 true leaf stage at 17 DAE (Figure 3). No statistical differences were noted in plant damage ratings at 14 DAE (data not presented), but by 21 DAE, significant differences were apparent (Figure 4). The untreated commercial cultivars exhibited the greatest thrips damage; injury was reduced in the experimental cultivars and plots treated with spinosad insecticide. Leaf area measurements revealed significant differences between treatments at 21 DAE, but no differences were observed on earlier sampling dates (Figure 5). The treated 7-07-1020 cultivar had most leaf area, and the untreated 7-07-1020 cultivar had similar leaf area as treated commercial and 7-07-1407 cultivars.

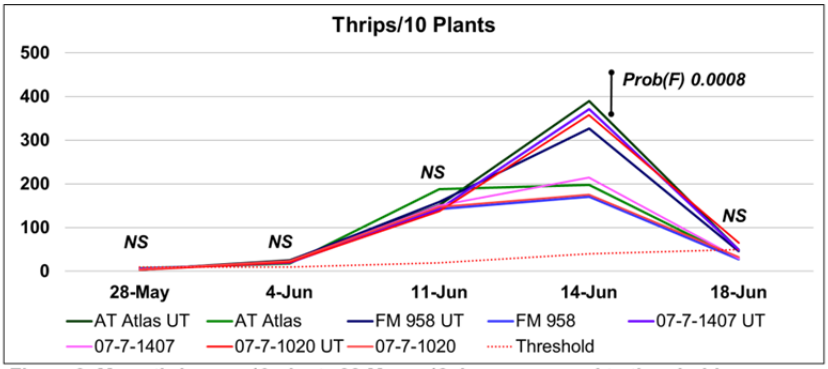

Figure 2. Mean thrips per 10 plants 28 May – 18 June compared to threshold.

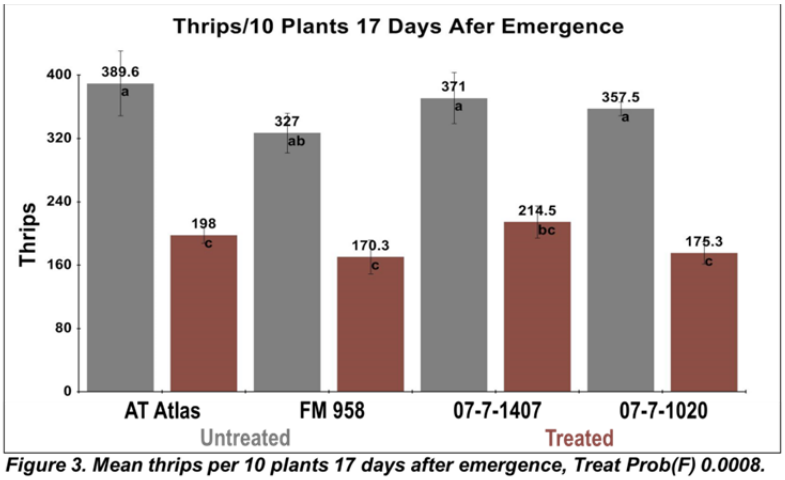

Figure 3. Mean thrips per 10 plants 17 days after emergence, Treat Prob(F) 0.0008.

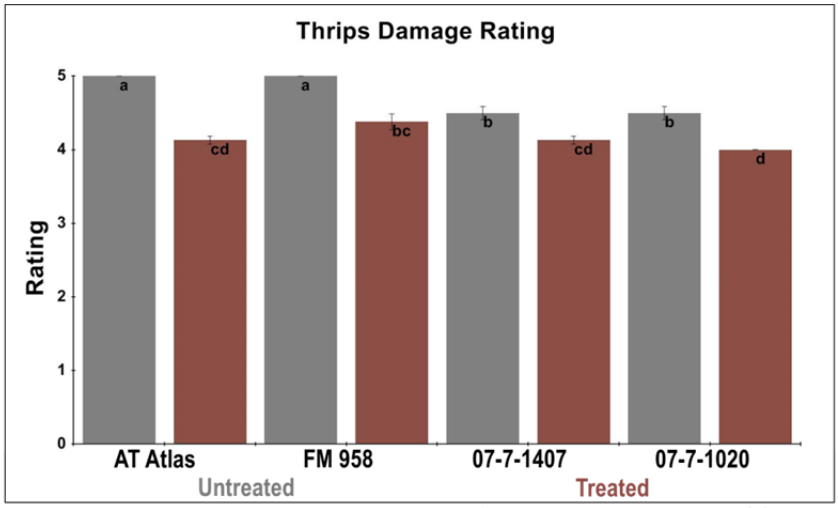

Figure 4. Mean thrips damage ratings 21 days after emergence, Treat Prob(F) 0.0005.

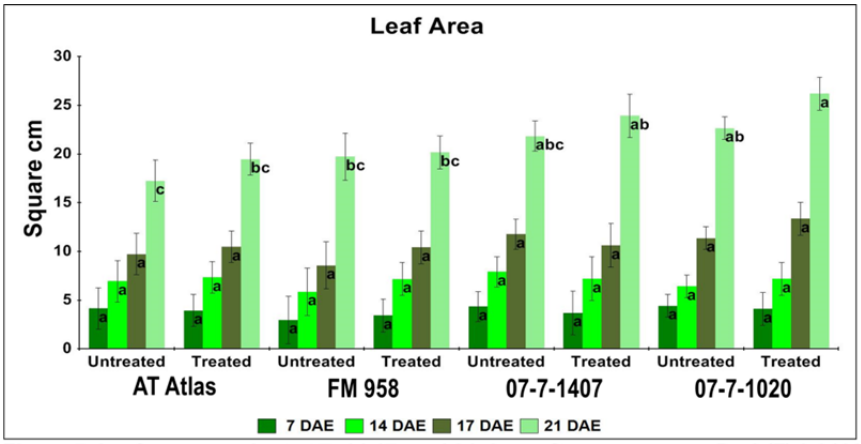

Figure 5. Leaf area per plant at 7, 14 17, and 21 days after emergence, Prob(F) 0.0166 21 DAE.

The percent of a thrips population which is immature is a good indicator of that population's ability to colonize; a higher percentage of immature thrips suggests a higher degree of colonization. When data from all post treatment sampling dates were combined and analyzed, cultivar had no impact on the percentage of the population which was immature (Figure 6). In 2 cultivars, Atlas and 07-7-1020, the Entrust insecticide significantly reduced the immature percentage but only provided slight numeric reductions in the other cultivars. Based on this data, Entrust appears to suppress colonization to a degree but cultivar did not have an impact.

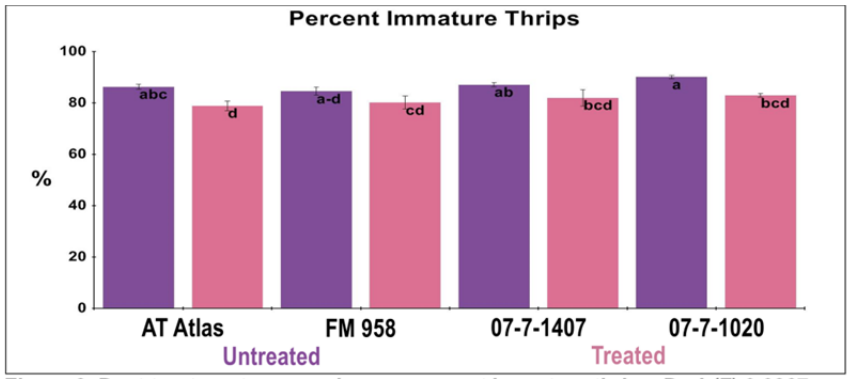

Figure 6. Post treatment seasonal mean percent immature thrips, Prob(F) 0.0367.

Cumulative thrips days can give an indication of thrips pressure over time. No differences in thrips days were observed when comparing cotton cultivars within insecticide treatments but a significant difference was observed when comparing all treatments (Figure 7). Spinosad reduced thrips days by 23.4% when comparing only insecticide treated vs untreated plots. This decrease is an indication of reduced overall thrips pressure and feeding duration.

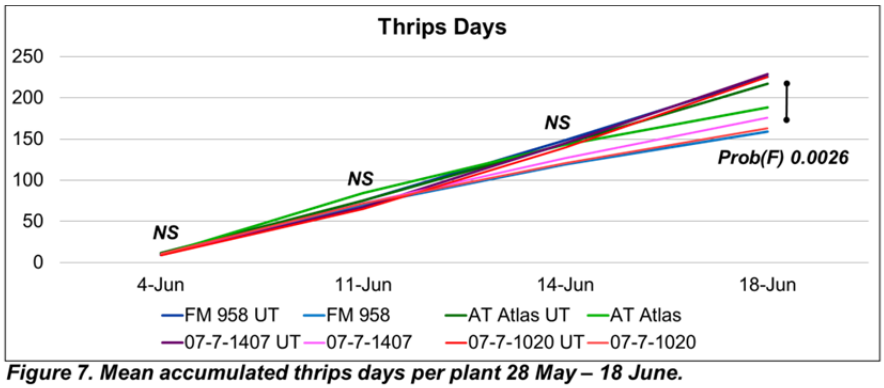

Figure 7. Mean accumulated thrips days per plant 28 May – 18 June.

Conclusions

Thrips pressure was moderate but exceeded action threshold throughout most of the seedling stage. Spinosad insecticide lowered the seasonal mean percent immature thrips, decreased thrips numbers at 17 DAE, and reduced accumulated thrips days. Cultivars did not differ in thrips colonization but had a significant impact on thrips damage and leaf area. These data suggest that the new cultivars do not express host plant resistance but may have more tolerance to thrips compared to commercial varieties.

Acknowledgements

The project site was provided by Jimmy Wedel, Muleshoe, TX. This project was funded by the USDA National Institute of Food and Agriculture. We also acknowledge and thank Ray White, Hayden Hadley, Austin Mason, and Cole Miller for their contribution in collecting and processing thrips samples

FIELD PERFORMANCE AND HERITABILITY OF THRIPS RESISTANCE FOR COTTON VARIETY DEVELOPMENT

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Abstract

In the absence of synthetic pesticide applications, thrips (Thysanoptera: Thripidae) management can be more problematic in organic production systems than conventional cotton systems. Additionally, nearly all organic cotton acreage on the Texas High Plains (THP) is planted with one or two conventional cultivars and seed-saving is near-ubiquitous, as these cultivars are no longer commercially available. Therefore, development of new thrips-tolerant, non-transgenic cultivars has the potential to greatly improve the availability and diversity of viable cultivars and overall production of organic cotton on the THP. Fifteen advanced breeding lines, 4 cultivars, and 1 newly-released germplasm line were planted at 2 field locations in 2013. Each genotype was evaluated for thrips resistance potential and overall field performance under organic management. Thrips resistance was assessed using visual injury ratings at both study sites. Yield and fiber quality data were collected to evaluate overall field performance of each genotype. Breeding lines '07-7-519CT', '07-7-1407CT', and '11-2-802GD' exhibited high field tolerance to thrips feeding. Lines '07-14-510FS' and 11-2-802GD and cultivars FiberMax® 'FM 958' and 'Tamcot 73' displayed the greatest lint yields among all evaluated genotypes. Both 07-7-519CT and 11-2-802GD exhibited a desirable combination of high thrips tolerance and yield potential, and would therefore be candidates for release as cultivars or parent material.

In addition, two broad-sense heritability trials were conducted to evaluate the inheritance of the thrips resistance trait and potential utility in variety development. Two separate families were evaluated, each originating from different interspecific *Gossypium hirsutum* L. and *Gossypium barbadense* L. crosses. The first family was derived from cold-tolerant *G. hirsutum* breeding line 07-7-1407CT and *G. barbadense* 'Cobalt'. Parents and the F_1 and F_2 generations were evaluated in a field study in 2012, and visual thrips injury ratings were conducted on individual plants for each genotype at 4-5 true leaves. The H^2 value for thrips resistance in this trial was 26.1%. The second family was derived from a CA 2266 (*G. hirsutum*) x TX 110 (*G. barbadense*) cross, and parents and F_1, F_2, and F_3 generations were evaluated in a greenhouse trial under elevated thrips pressure in 2013. H^2 values for F_2 CA 2266 x TX 110 and F_3 CA 2266 x TX 110 generations were 22.9% and 28.4%, respectively. These values support previous assumptions regarding the quantitative nature of thrips resistance. While these values were relatively low, they indicate that visual phenotyping for thrips resistance and subsequent selection is consistent between the field and greenhouse. More work is necessary to further validate these data at both the greenhouse and field level.

UPDATE ON BOLLWORM PYRETHROID RESISTANCE MONITORING

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Abstract

Polyphagous bollworms are potentially exposed to pyrethroid insecticides during each generation. Since cotton is a host during the latter part of the growing season, any resistance developed during the season will reduce control realized in cotton. Pheromone traps have been used sporadically since the late 1980s throughout the cotton belt to collect male moths for testing resistance to a pyrethroid insecticide. Testing was conducted across the cotton belt in a coordinated fashion from 2007-2014 using a concentration of 5 µg/vial of cypermethrin as the diagnostic dose. Overall survival during 2014 was 18.8%, which was somewhat higher than recent years. However, resistance was not uniform across all states. Louisiana and Virginia have regularly had higher survival than all other states during recent years. This year Georgia joined them with all having yearly average survivorship between 30 and 35%. In contrast, Missouri, South Carolina and Tennessee all had average survival of less than 10%. The other states fell between these extremes.

Introduction

Bollworm, *Helicoverpa zea*, is a pest in numerous crops where it may be exposed to pyrethroid insecticides. Since it can have 5 or more generations per year in the southern U.S., it has the potential to develop large populations and insecticide resistance has the potential to develop and spread rapidly. One to two of these generations occur in cotton, causing substantial economic loss. Because pyrethroid insecticides are relatively inexpensive, they are often the first choice of growers for foliar control of bollworms. Knowledge of the susceptibility of bollworms to pyrethroid insecticides is therefore critical to effective management of this pest.

Monitoring pyrethroid resistance in bollworms has been conducted for numerous years, beginning in 1988 in a few states and then coordinated throughout the cotton belt in 1989-1990 (Rogers et al. 1990). Since then monitoring has continued at various levels every year. Regional data from previous years can be found in earlier Beltwide cotton conference proceedings (Martin et al. 1999, 2000, Payne et al. 2001, 2002, Musser et al. 2010, 2011, 2013). During this time the bioassay methodology has remained consistent. Male moths are captured in a pheromone trap and placed in a glass vial that was previously treated with insecticide. Mortality is recorded after 24 h. A concentration of 5 µg cypermethrin / vial has been used with baseline survival generally less than 10% (Martin et al. 1999).

Materials and Methods

Hartstack pheromone traps were placed in various locations in ten states across the cotton belt from VA to TX. Pheromones (Luretape with Zealure, Hercon Environmental) were changed every 2 weeks. Some traps were monitored at least weekly from May until September, but most were monitored over a shorter period when bollworms were abundant and cotton was susceptible to bollworm feeding. Healthy moths caught in these traps were subsequently tested for pyrethroid resistance. Moths were individually placed in 20 ml scintillation vials that had been previously coated with 0 or 5 µg cypermethrin per vial. Vial preparation for all locations except Louisiana was done at Starkville, MS and shipped to cooperators as needed throughout the year. Louisiana data are from vials prepared in Louisiana. In addition to rates of 0 and 5 µg cypermethrin per vial, Louisiana also tested survival at 10 µg cypermethrin per vial. At all locations, moths were kept in the vials for 24 h and then checked for mortality. Moths were considered dead if they could no longer fly. Reported survival was corrected for control mortality (Abbott 1925).

Results and Discussion

A total of 8815 moths were assayed during 2014. The fewest moths (169) were tested in North Carolina while the most moths (2539) were tested in Louisiana. Average survival to the 5 µg cypermethrin / vial concentration was 18.8% in 2014 (Table 1), which was the highest rate of survival since 2007 (Fig. 1). As has been consistently observed in the past, survival during July was higher than during previous months. While late season moths are often more susceptible, survival rates during 2014 were maintained during August and September.

Table 1. Bollworm survival to 5 µg cypermethrin per vial in 24-h vial tests during 2014.

State	May	June	July	Aug	Sep	Overall	Total bollworms tested
AR	3.6	8.3	26.2	8.0	14.3	14.3	990
GA		8.3	20.8	42.1	21.6	30.4	787
LA	7.1	25.0	31.4	43.2	52.2	33.3	2539
MS	18.9	11.8	12.3	9.1		13.8	1178
MO				10.2	7.3	9.3	597
NC			24.4			24.4	169
SC		0.0	10.6	2.7	0.0	5.3	605
TN			7.6			7.6	261
TX		11.5	22.3	16.0	16.2	16.8	1220
VA			27.7	33.9	31.6	32.4	649
Average	9.9	10.8	20.4	20.7	20.5	18.8	8815

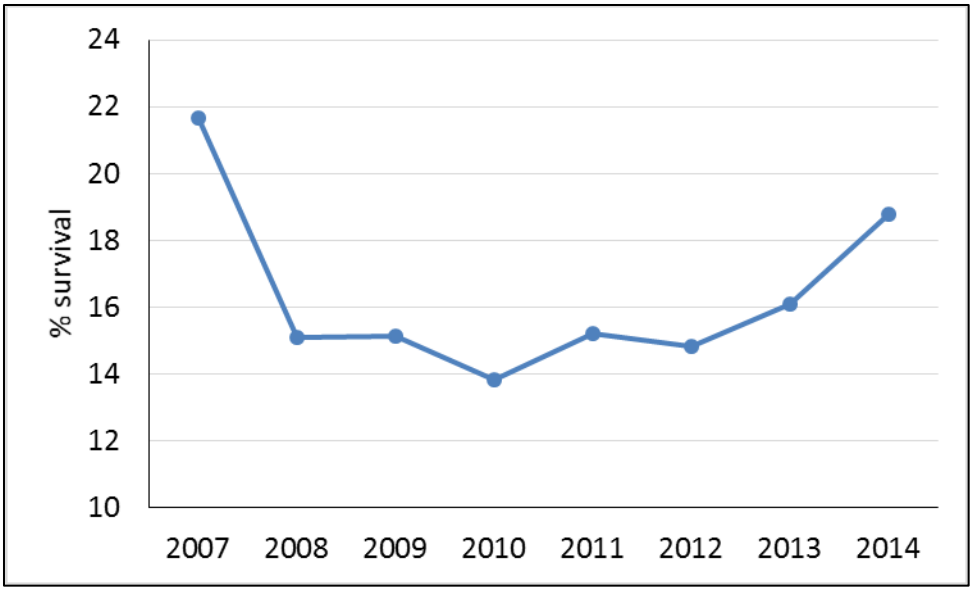

Fig. 1. Beltwide bollworm average survival per year at 5 µg cypermethrin per vial from 2007 – 2014.

Most states had survival rates similar to previous years, put survival in Georgia was sharply higher during 2014, making average survival in Georgia for the year similar to Louisiana and Virginia, the two states that have had the least susceptible moths during the last several years (Fig. 2). Whether this is a one-year spike like observed in 2007, or a long-term change in susceptibility remains to be seen. North Carolina has also had higher survival than most states each of the last two years, so it may be that pyrethroid resistance in bollworms is becoming more common along the eastern coast of the U.S.

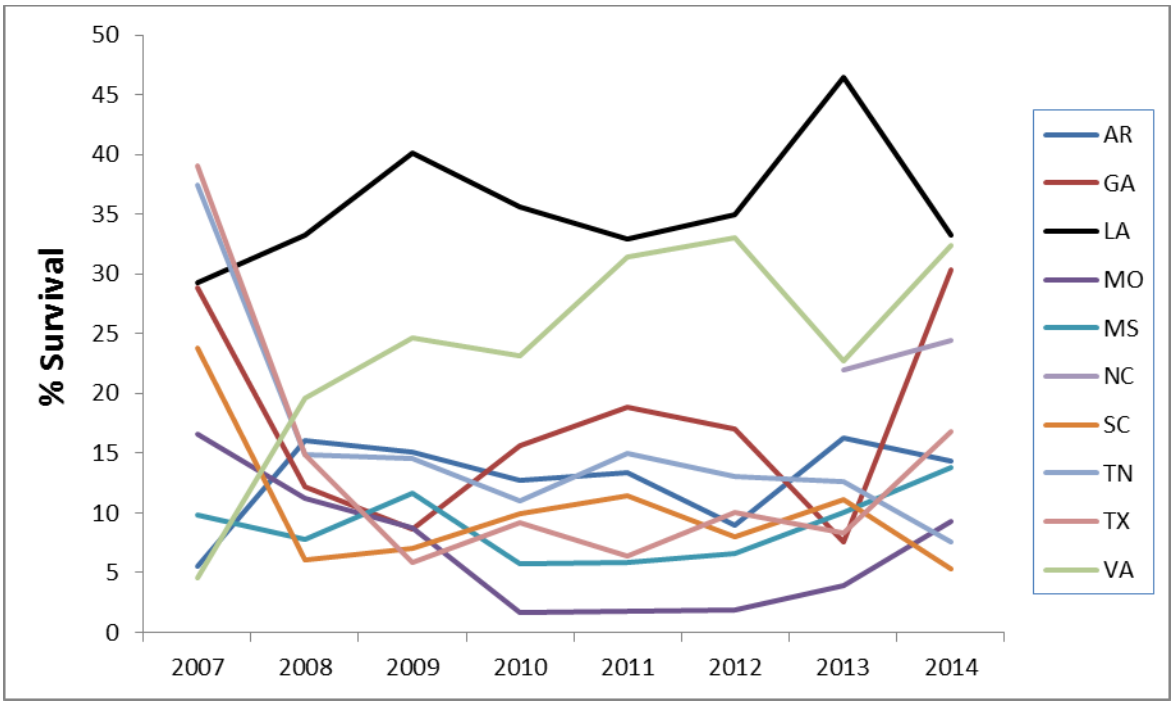

Fig. 2. Average bollworm survival by state per year at 5µg cypermethrin per vial from 2007 – 2014.

A comparison of bollworm susceptibility in Louisiana at both 5 µg and 10 µg cypermethrin, reveals that the relationship between these concentrations is not the same throughout the year. While survival during May and June was similar at both concentrations, survival continued to increase throughout the year at 5 µg, but stayed steady

between 20% and 30% survival at 10 µg (Fig. 3). For a point of reference, tobacco budworm was considered resistant to pyrethroids when there was 30% survival of the moths at the 10 µg concentration. Louisiana stayed near this line most of the year, and larval control of bollworms with pyrethroids is considered erratic.

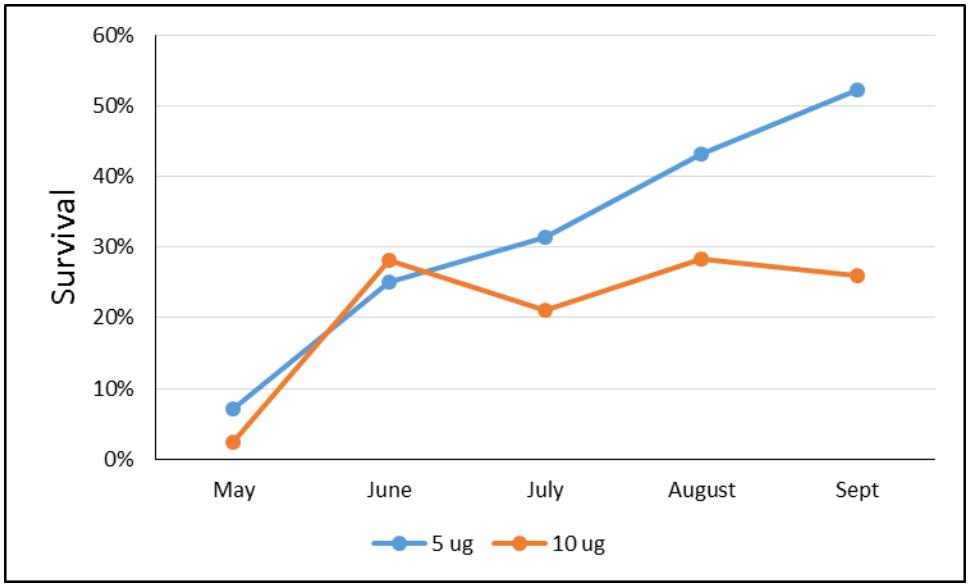

Fig. 3. Monthly bollworm survival at 5 µg and 10 µg cypermethrin per vial in Louisiana during 2014.

Bollworm adults are considered highly mobile (Lingren et al. 1994, Beerwinkle et al. 1995), which would suggest that pyrethroid resistance would quickly spread from one region to another. However, pyrethroid resistance has persisted in LA and VA for numerous years while populations in adjacent states remain largely susceptible. Field control of bollworm larvae is inconsistent throughout many parts of the cotton belt, so it is likely that numerous resistance genes are present in populations. It is likely that resistance is associated with high fitness costs, so resistance is reduced every winter, and spreads to new regions more slowly than expected. However, monitoring from 1998-2000 found average survival rates of less than 10%, while average current survival is approaching 20% and exceeds 30% in some states. Even though pyrethroids may not be applied as frequently in cotton as in the past, there are still enough applications made in the landscape to slowly decrease pyrethroid susceptibility, making the selection of this class of chemistry for targeting bollworms a risky decision.

Conclusions

Pyrethroid susceptibility in bollworms over the cotton belt appears to be slowly decreasing, but the rate of decline is not uniform. Louisiana and Virginia have had the lowest susceptibility for several years. Georgia has similar survival to pyrethroids during 2014. Average survival on 5 µg cypermethrin over the entire cotton belt rose to 18.8% during 2014, which was the highest survival observed since 2007.

Acknowledgements

The authors thank Cotton Inc. and all the cooperating universities for providing partial funding for this project.

References

Abbott, W. S. 1925. A method of computing the effectiveness of an insecticide. J. Econ. Entomol. 18: 265-267.

Beerwinkle, K. R., J. D. Lopez, D. Cheng, P. D. Lingren, and R. W. Meola. 1995. Flight potential of feral *Helicoverpa zea* (Lepidoptera: Noctuidae) males measured with a 32-channel, computer-monitored, flight-mill system. Environ. Entomol. 24: 1122-1130.

Lingren, P. D., J. K. Westbrook, V. M. Bryant, J. R. Raulston, J. F. Esquivel, and G. D. Jones. 1994. Origin of corn earworm (Lepidoptera: Noctuidae) migrants as determined by citrus pollen markers and synoptic weather systems. Environ. Entomol. 23: 562-570.

Martin, S. H., R. D. Bagwell, M. L. Boyd, B. L. Freeman, G. A. Herzog, D. R. Johnson, M. B. Layton, B. R. Leonard, N. Liu, G. T. Payne, P. V. Pietrantonio, M. E. Roof, M. J. Sullivan, J. W. Van Duyn, and J. R. Weeks. 1999. Status of bollworm, *Helicoverpa zea*, susceptibility to pyrethroids: IRAC-US 1998 update, pp. 867-871. *In:* Proceedings, Beltwide Cotton Conferences. 3-7 January, 1999. Orlando, FL. National Cotton Council, Memphis, TN.

Martin, S. H., J. S. Bachelor, R. D. Bagwell, M. L. Boyd, B. L. Freeman, G. A. Herzog, D. R. Johnson, M. B. Layton, B. R. Leonard, N. Liu, G. T. Payne, P. V. Pietrantonio, M. E. Roof, R. Seward, R. K. Sprenkel, M. J. Sullivan, J. W. Van Duyn, and J. R. Weeks. 2000. Status of bollworm, *Helicoverpa zea*, susceptibility to pyrethroids in the mid-south and southeast: IRAC-US 1999 update, pp. 1359-1365. *In:* Proceedings, Beltwide Cotton Conferences. 4-8, January, 2000. San Antonio, TX. National Cotton Council, Memphis, TN.

Musser, F. R., R. E. Jackson, J. K. Greene, B. R. Leonard, S. D. Stewart, M. N. Parajulee, J. J. Adamczyk, G. M. Lorenz, D. S. Akin, K. V. Tindall, D. A. Herbert, and P. M. Roberts. 2010. Pyrethroid resistance monitoring of bollworms, pp. 1259-1264. *In:* Proceedings, Beltwide Cotton Conferences. Jan. 4-7, 2010. New Orleans, LA. National Cotton Council, Memphis, TN.

Musser, F. R., R. E. Jackson, J. K. Greene, B. R. Leonard, S. D. Stewart, M. N. Parajulee, J. J. Adamczyk, G. M. Lorenz, D. S. Akin, K. V. Tindall, D. A. Herbert, and P. M. Roberts. 2011. 2010 pyrethroid resistance monitoring of bollworms, pp. 1111-1114. *In:* Proceedings, Beltwide Cotton Conferences. Jan. 4-7, 2011. Atlanta, GA. National Cotton Council, Memphis, TN.

Musser, F. R., R. E. Jackson, J. K. Greene, D. Kerns, S. D. Stewart, M. N. Parajulee, R. D. Parker, G. M. Lorenz, A. Jones, D. A. Herbert, and P. M. Roberts. 2013. Beltwide pyrethroid resistance monitoring of bollworms, pp. 897-900. *In:* Proceedings, Beltwide Cotton Conferences. Jan. 7-10, 2013. San Antonio, TX. National Cotton Council, Memphis, TN.

Payne, G. T., J. S. Bachelor, J. W. Van Duyn, R. D. Bagwell, B. R. Leonard, M. L. Boyd, B. L. Freeman, N. Liu, J. R. Weeks, A. Herbert, G. A. Herzog, D. R. Johnson, M. B. Layton, G. L. Lentz, R. Seward, S. H. Martin, P. V. Pietrantonio, M. E. Roof, M. J. Sullivan, and R. K. Sprenkel. 2001. U.S. Cottonbelt survey: testing the susceptibility of the bollworm, *Helicoverpa zea* (Boddie) to pyrethroid insecticides, pp. 782-785. *In:* Proceedings, Beltwide Cotton Conferences. 9-13 January, 2001. Anaheim, CA. National Cotton Council, Memphis, TN.

Payne, G. T., J. Adamczyk, J. S. Bachelor, R. D. Bagwell, M. L. Boyd, C. Brister, B. L. Freeman, A. Herbert, G. A. Herzog, D. R. Johnson, M. B. Layton, G. L. Lentz, B. R. Leonard, N. Liu, S. H. Martin, P. V. Pietrantonio, M. E. Roof, R. K. Sprenkel, M. J. Sullivan, J. W. Van Duyn, and J. R. Weeks. 2002. U.S. cottonbelt survey: testing the susceptibility of the bollworm, *Helicoverpa zea* (Boddie), to pyrethroid insecticides- 2001 update. *In:* Proceedings, Beltwide Cotton Conferences. 8-12 January, 2002. Atlanta, GA. National Cotton Council, Memphis, TN.

Rogers, B., H. D. Feese, S. L. Riley, C. A. Staetz, J. Whitehead, and W. Mullins. 1990. *Heliothis* resistance monitoring in the cotton belt: PEG-US update, pp. 163-164. *In:* Proceedings, Beltwide Cotton Conferences. Las Vegas, NV. National Cotton Council, Memphis, TN.

Long-term Survey of Bollworm Moth Flight Activity and Pyrethroid Resistance Monitoring in the Texas High Plains

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INTRODUCTION

The Texas High Plains is recognized as the most intensive cotton growing area in the world, with approximately 4 million acres of contiguous cotton grown in a 41-County production region. In this region, the bollworm, *Helicoverpa zea* (Boddie), is classified as an important economic pest of cotton. Seed from genetically modified cotton is available with Bollgard® II (*Bt*) technology which provides excellent crop protection from lepidopteran pests. Continued bollworm population monitoring is important because of the significant cotton acreage that is not planted with this technology, particularly on reduced-input dryland which accounts for approximately 60% of the cotton acreage. In addition, the percentage of lower-input dryland cropping is increasing as irrigation capacity is steadily decreasing across the region.

Trapping Studies. In 2002, an ongoing trapping study was initiated to investigate the weekly flight activity patterns of the cotton bollworm, tobacco budworm, *Heliothis virescens* (F.), and beet armyworm, *Spodoptera exigua* (Hübner) in the southern Texas High Plains region. Since the pyrethroid resistance study discussed below was conducted only for cotton bollworms, the trapping/flight data discussed in this report will also be limited solely to bollworms. Insect traps (Hartstack pheromone trap; Fig. 1) were used to measure the seasonal abundance and flight profiles of cotton bollworm adult males as they responded to baited pheromone traps.

Bollworm Pyrethroid Resistance Monitoring. Beginning in 2007, the Lubbock Texas AgriLife Cotton Entomology Program began cooperating in a multi-state cotton bollworm pyrethroid resistance monitoring study. Musser et al. (2013) provides a comprehensive review of the entire Beltwide Resistance Monitoring Program (2007-2012 time period). In this report, only the portion from the Texas Southern High Plains region will be highlighted and this version incorporates the results of the 2014 season.

Figure 1. Stanley Carroll servicing a Hartstack pheromone trap containing male cotton bollworm moths (left panel). After counting the moths for the flight profile portion of the study, the freshly captured moths were placed individually into glass vials (right panel) of two types, clean 20-ml vials (untreated controls) or vials treated with a concentration of 5-µg/vial of cypermethrin (diagnostic dose).

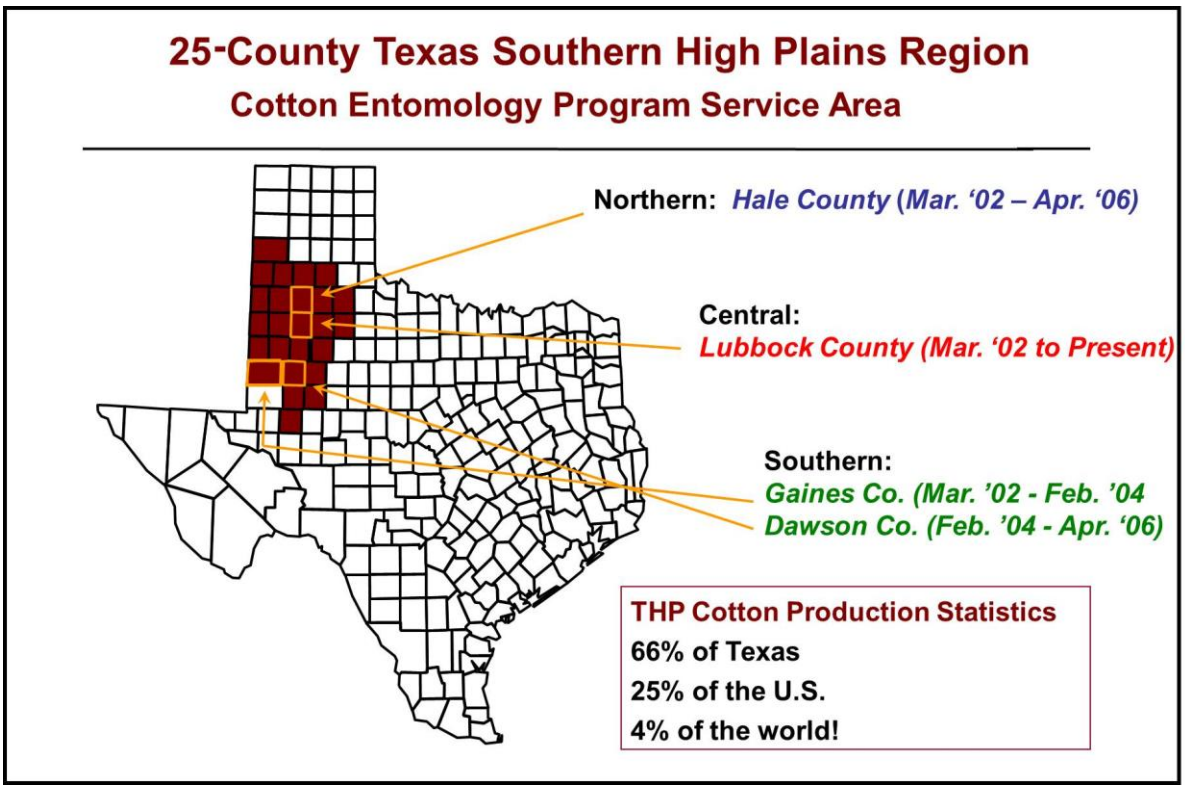

Figure 2. Selected counties and trapping durations for a pheromone trapping study conducted in the Southern Texas High Plains to investigate the seasonal moth flight patterns of cotton bollworm, 2002-2014.

MATERIALS and METHODS

Cotton Bollworm Trapping.

Study Duration: March 2002 to Present

Study Sites: Hale, Lubbock and Gaines/Dawson counties, Texas (Fig. 2)

Sampling Protocol:

- Three traps (Hartstack et al. 1979) baited with bollworm pheromone lures were placed in each of the selected counties representing the northern, central, and southern areas of the Texas High Plains (Fig. 2). Traps located in Gaines Co. were moved to neighboring Dawson Co. after the first year of the study to facilitate more frequent monitoring. Trapping sites within a county were geographically separated by a minimum distance of 5 miles.
- Traps were monitored throughout the year at intervals of approximately one week during active flight periods (spring, summer, fall) and bi-monthly during periods of low flight activity (winter).
- Cotton bollworm specific pheromone lures were replaced on the traps at two-week intervals.

Cotton Bollworm Pyrethroid Resistance Monitoring.

Study Duration: 2007 to Present

Study Sites: Lubbock County, Texas

Sampling Protocol:

- Freshly captured healthy male moths were taken from pheromone traps located at three Lubbock County sites (same sites as described above) and after return to the lab, placed into either clean 20-ml scintillation vials (untreated controls) or identical vials treated at Dr. Fred Musser's laboratory (Mississippi State University) with a concentration of 5-µg/vial of cypermethrin (diagnostic dose) (Fig.1).
- Moth survival/mortality was monitored 24-h later for the moths held in both untreated control vials and cypermethrin treated vials. Moths capable of controlled flight were counted as "alive", while dead and/or those unable to fly were classified as "dead".
- Moth survival observed from treated vials was corrected for control mortality as reported by Abbott (1925).

RESULTS

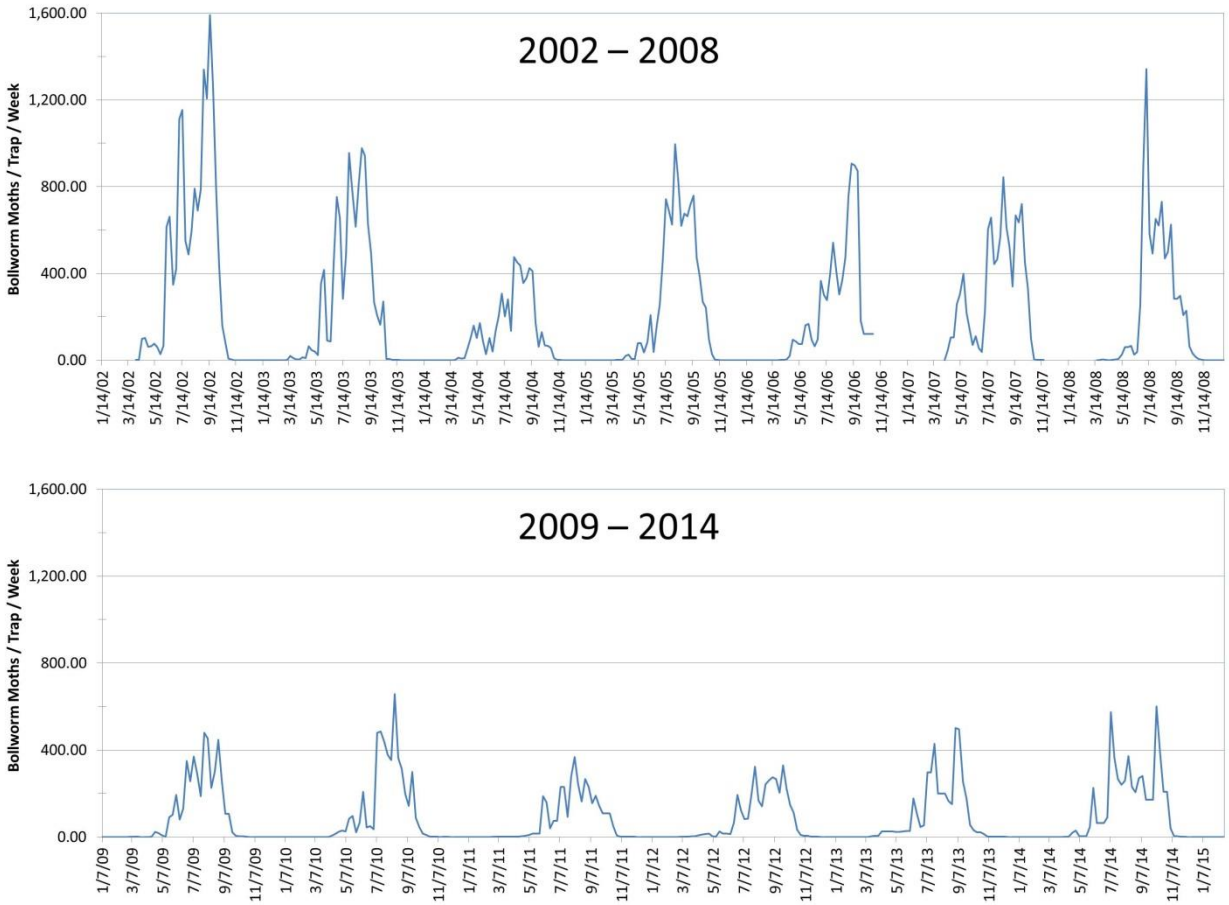

Figure 3. Annual seasonal flight profiles based upon average weekly cotton bollworm moths captured per pheromone trap positioned in rural cotton producing areas of Lubbock County, TX. 2002-2014.

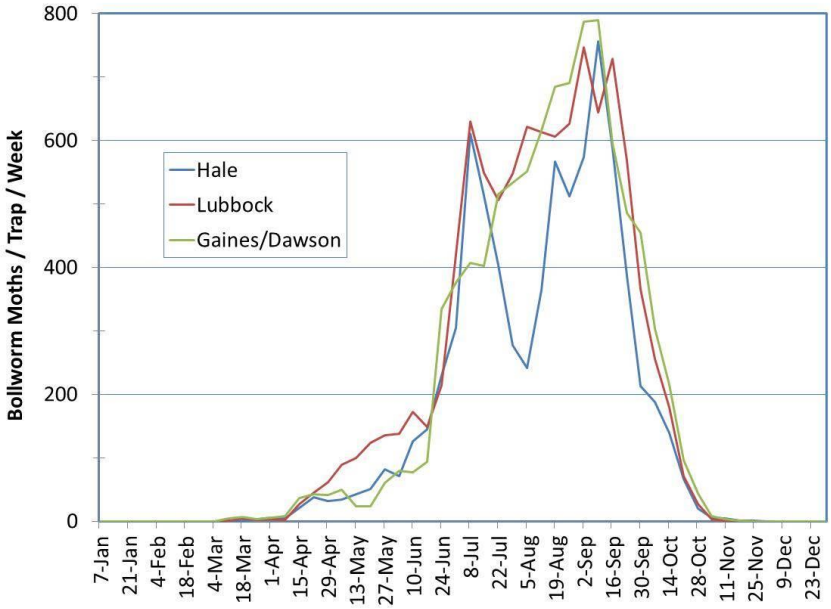

Figure 4. Cotton bollworm historical flight profiles (each colored-coded line represents the mean weekly trap captures averaged across four years). Multiple county flight profiles are shown so that comparisons can be made for areas roughly representing northern (Hale), central (Lubbock) and southern (Gaines/Dawson) regions of the Texas Southern High Plains. 2002-2005.

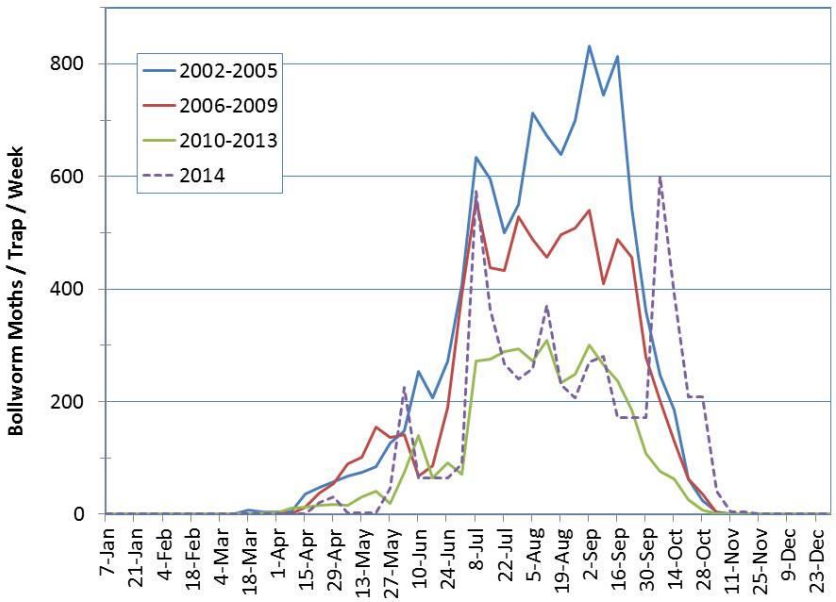

Figure 5. Average number of bollworm moths/trap/week, Lubbock County. The 13 years of male moth flight profiles (see Fig. 3) are grouped into three 4-year profiles representing boll weevil eradication/early Bollgard® adoption period (2002-2005), increased Bollgard® adoption (2006-2009), Bollgard® technology adoption peak (2010-2013), plus the most recent year (2014).

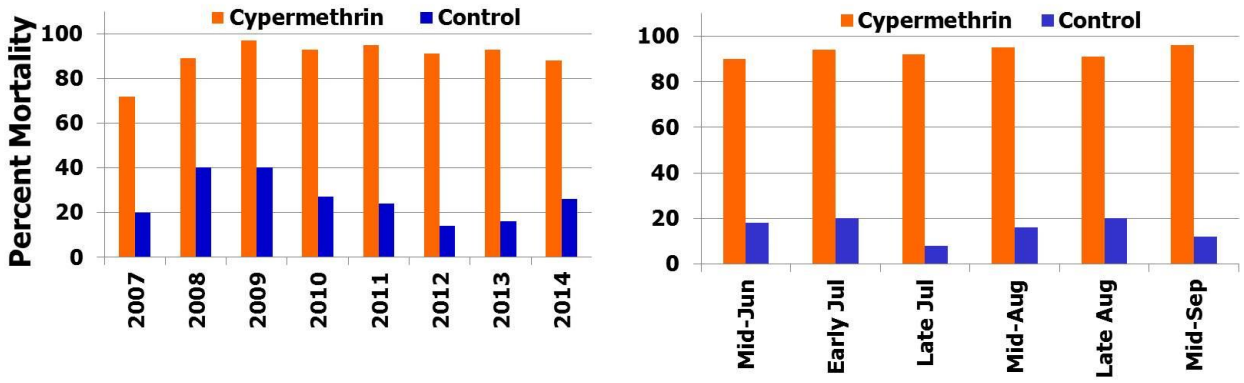

Figure 6. Cotton bollworm moth susceptibility (measure in terms of % mortality on y-axis) to cypermethrin in a vial bioassay, 2007-2014. The data in the left panel show the year-to-year variation in cypermethrin susceptibility of individual bollworm moths placed into vials treated with 5 µg cypermethrin (treated) and moth mortality in clean vials without cypermethrin (control). The data presented in the right panel show within-season variation in cypermethrin susceptibility of bollworm moths averaged across the 8 years of the study, Lubbock, TX.

DISCUSSION

Cotton Bollworm Flight Profiles.

Thirteen annual bollworm moth flight profiles for bollworms in Lubbock County are shown in Figure 3. Overall, the abundance of bollworms decreased over the study years. With the exception of 2004, bollworm trap captures during the first seven years of the study were noticeably higher than in the last six years. Bollworm male moth captures were relatively high during 2002 and reaching a peak of 1590 moths/trap/week during mid-September. Overall population levels detected in Lubbock County were relatively similar from 2003 to 2008, except for 2004 which exhibited a much reduced population similar to what was observed in later years (2009-2014).

Figure 4 illustrates the historical bollworm flight profiles (based upon pheromone trap captures) for the three Texas Southern High Plains counties. Bollworm flight activity in the region was low or non-existent during the period from mid-November to mid-March. An extended period of high bollworm moth activity occurred during the mid-June to mid-October time period which overlays the entire period that cotton is vulnerable to fruit damage. Within this extended period of activity, the highest numbers of moths responded to traps from early August to mid-September.

During the first four years (2002-2005), seasonal flight profiles were monitored in three areas representing northern (Hale County), central (Lubbock County), and southern (Gaines/Dawson counties) regions of the Texas High Plains. Although individual yearly flight profiles can vary greatly, Figure 4 clearly indicates that when averaged across several years, the flight profiles from the different north-south regions of the THP do not differ greatly in timing and/or magnitude of peak bollworm flight activities.

Figure 5 illustrates four 3-year bollworm flight profiles roughly representing the years immediately following boll weevil eradication and the beginning of Bollgard® adoption (2002-2005), the increased Bollgard® adoption years (2006-2009), Bollgard® adoption peak years

(2010-2013), plus the most recent year (2014; dotted line). The most recent years can be characterized by the presence of continued drought, some crop failure, low crop yields, and decreased irrigation capacity across the region. Generally speaking, the bollworm flight activity was heaviest in the earlier monitored years (2002-2005) and tended to progressively decrease during the 2006-2009 period and 2010-2013 years, both in terms of the peak activity numbers and duration of the active flight periods. The flight/trap response profile for 2014 was unusual in that the second peak of moth activity occurred approximately 3-4 weeks later than had been observed during the earlier years of the study. We are not able to accurately explain why bollworm moths were responding to traps so late in the season during 2014, but relatively cooler and wetter growing season might have altered the life cycle patterns of the bollworm.

Bollworm Pyrethroid Resistance Monitoring.

Bollworm moths in the Texas High Plains, specifically the Lubbock County populations, were highly susceptible to 5 µg cypermethrin in the vial bioassay, with 90-97% mortality in 7 of the 8 years of the study; the 2007 study showed an average of 74% seasonal mortality (Fig. 6).

Although vial bioassays were performed on fresh moths collected within a 24-hour trapping period, control vials had 20-40% mortality. Corrected mortality (Abbott 1925) due to cypermethrin (5 µg) ranged from 80 to 93%.

Averaged over 8 years, within-season mortality of cypermethrin-treated moths did not vary significantly. Mortality values were >90% throughout the season (Fig. 6).

ACKNOWLEDGMENTS

Funding for this study was provided by Texas A&M AgriLife Research Cotton Entomology Project 8810, Cotton Incorporated Core Program, and Plains Cotton Growers, Inc., Lubbock.

The bollworm cypermethrin resistance monitoring portion of this report is a subset of a multi-state, multi-year Beltwide program. All cypermethrin-treated and control vials used in this bioassay were furnished by Dr. Fred Musser of Mississippi State University.

REFERENCES

Abbott, W. S. 1925. A method of computing the effectiveness of insecticides. J. Econ. Entomol. 18: 265-267.

Hartstack, A. W., Jr., J. A. Witz, and D. R. Buck. 1979. Moth traps for the tobacco budworm. J. Econ. Entomol. 72: 519-522.

Musser, F. R., R. E. Jackson, J. K. Greene, D. Kerns, S. D. Stewart, M. N. Parajulee, R. D. Parker, G. M. Lorenz, A. Jones, D. A. Herbert, and P. M. Roberts. 2013. Beltwide pyrethroid resistance monitoring of bollworms, pp. 897-900. *In* Proceedings, Beltwide Cotton Conferences, National Cotton Council, Memphis, TN.

Metapopulation approach for landscape level management of western tarnished plant bug, *Lygus hesperus*, in Texas (Hemiptera, Miridae)

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Abstract: Insect source-sink dynamics are vital to ecologically intensive pest management. Maintaining sink plant hosts, or "trap crops", and destroying alternate hosts or breeding places adjacent to the field crop are effective pest management strategies for some arthropods. However, determining whether a host acts as a source or a sink is challenging, especially when the pest species is highly mobile and polyphagous. The western tarnished plant bug, *Lygus hesperus*, is highly polyphagous, and can utilize > 300 hosts. Its presence has been documented in 26 roadside weed hosts in the Texas High Plains. Previous studies demonstrated that *L. hesperus* prefer alfalfa over cotton and several alternate weed hosts. A four-year project involved surveying and sampling for *L. hesperus* in the agricultural landscapes of several sub-regions of the southwestern United States, including the Texas High Plains. In Texas, geographic information of the landscape vegetation complex was compiled from a 150 km radius in the Texas High Plains. In one study, fifty irrigated cotton fields representing the crop diversity within this region were sampled via sweep-net for 10 weeks. This effort also included sampling of up to six non-cotton insect habitats within a 3 km radius of each field. Seasonal average *L. hesperus* abundance data were regressed with 27 field characteristics (variables), including habitat-specific land cover, distance between focal cotton fields and non-cotton habitats, longitude, latitude, elevation, habitat heterogeneity index, and several environmental/ecological variables. Significant variables were selected using a stepwise regression at 15% probability rate. A 10-parameter linear model explained 93% of the variation in the data. Major parameters contributing significantly to variation in *L. hesperus* abundance in cotton were corn and sunflower acreages, focal cotton field distances from several non-cotton hosts, and habitat heterogeneity index. In addition, field marking-and-capture studies were conducted using protein markers and enzyme-linked immunosorbent assays to characterize *L. hesperus* intercrop movement behavior. The field marking-and-capture approach can be used to study the effects of various crop management practices on *L. hesperus* intercrop movement and can potentially be applied to other pests and cropping systems.

Key words: western tarnished plant bug; cotton IPM; ecological pest management; alternate host management

Cotton, *Gossypium hirsutum* (L.) is grown in 17 states in the United States with coverage exceeding 9 million harvested acres. The United States cotton farmers harvest about 3.5 million metric ton of cotton annually, valued at $ 7.5 billion (NASS, 2009 – 2013, last five year average data)[1]. Texas is the largest cotton-producing state in the U.S. In Texas, on an average 2.2 million hectares of cotton are planted and 1.2 million metric ton of cotton are produced (valued at $ 3.5 billion)[2]. Approximately 55% of U.S. cotton is produced in Texas, while 67% of Texas cotton is produced in the Texas High Plains. Thus, cotton produced in the Texas High Plains accounts for 37% of total U.S. cotton production.

Texas High Plains cotton production was a low-insecticide use system until boll weevil populations arrived in early 1990s. A significant shift in arthropod pest management approach was expected after eradication programs eliminated the boll weevil from the Texas High Plains. Increased adoption of transgenic cotton cultivars conferring tolerance to lepidopteran pests reduced the insecticide load in cotton insect manage-

基金项目: Cotton Incorporated Core Program, USDA CSREES RAMP

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收稿日期: 2014 – 11 – 15

ment. Despite this reduction, increased public concerns for environmental safety and questions regarding the sustainability of current crop protection practices have placed a premium on the development of integrated pest management (IPM) approaches.

The IPM approaches have been widely used to manage cotton pests. These approaches include biological, cultural, chemical, and plant resistance methods[3]. Integration of these approaches has been hindered due to a lack of understanding of the biology and ecology of the target pests, often resulting in the unilateral reliance on insecticides. The ecological effects of widespread insecticide use have often fueled other pest problems, environmental pollution, and affected non-target organisms. Recognition of the ecological changes and problems associated with reliance on insecticides fostered the development of IPM concepts which are now well-established in all cotton-production regions of the U. S. [4].

The western tarnished plant bug, *Lygus hesperus* (Knight), is highly polyphagous, and can survive on a broad range of hosts[5]. Its presence has been reported in 26 unique roadside weed hosts in the Texas High Plains[6]. Previous studies have demonstrated that *L. hesperus* prefer alfalfa over cotton and several weed hosts[7-8]. These data suggest that the insect source-sink dynamics could be a valuable component of *L. hesperus* management strategy.

Maintaining sink plant hosts, or "trap crops", and destroying source plant hosts (alternate hosts or breeding sites) adjacent to the field crop are effective strategies in managing many pest populations. Determining whether a particular host acts as a source or a sink is challenging, especially when the pest species is highly mobile and polyphagous. *L. hesperus* sub-populations in the crop field are continuously interacting with sub-population in nearby host habitats through intermigration. These strong interactions represent an important opportunity for exploitation in an integrated pest management system, particularly, since even if all *L. hesperus* are removed from a specific field, re-colonization pressure due to these interactions will continue. In fact, landscape-level management of metapopulation dynamics can be a sustainable, economical, and environmentally conscious approach of pest management.

Previous studies have indicated that *L. hesperus* move from alfalfa and other weed hosts into cotton[9-10]. *L. hesperus* management decisions in cotton might be greatly affected by advancing the understanding of host availability and *L. hesperus* source-sink dynamics. For example, *L. hesperus* population dispersal from alfalfa to adjacent cotton might be increased by government-enforced mowing of rural roadside weed hosts such as volunteer alfalfa. Researchers in California demonstrated that strip-cutting commercial alfalfa mitigates *L. hesperus* dispersal to cotton[11]. Similarly, an area-wide *L. hesperus* management project in Mississippi demonstrated that roadside weed management is an effective means of minimizing tarnished plant bugs, *Lygus lineolaris* (Palisot de Beauvois), in adjacent cotton[12]. *L. hesperus* intercrop movement behavior has not been fully characterized in cotton-alfalfa systems in the Texas High Plains. Doing so could improve *L. hesperus* management strategies in the region and the utility of such strategy could be expanded to other cotton producing regions of the United States.

Development of a sustainable and environmentally conscious *L. hesperus* management approach requires a greater understanding of metapopulation dynamics. Information on the *L. hesperus* metapopulation dynamics is lacking in current scientific literature. Therefore, the goals of this research were to 1) identify the *L. hesperus* sub-populations in Texas High Plains via habitat survey, 2) characterize the metapopulation dynamics through landscape-level studies, and 3) validate *L. hesperus* sub-population interactions (intermigration) in a field marking and intercrop movement study.

1 Materials and Methods

1.1 Materials

Survey site: The survey site consisted of 25-County Texas High Plains region that is serviced by Cotton Growers, Inc. This is the largest contiguous cotton growing season in the world, encompassing about 1.5 hm^2 of upland cotton. Sampling was conducted via "keep it simple" KIS vacuum sampling, and then detecting protein markers using indirect enzyme-linked immunosorbent assay (ELISA). Protein markers used in the included 10% egg white (EW) and 10% non-fat dry milk (NFDM) marker solutions.

Reagent: Reagents used in the ELISA assays included Tris-buffered saline (TBS), phosphate-buffered saline with tween 20 (PBST), and tetramethyl benzidine (TBM).

Techniques: Landscape level study utilized GPS technology in conjunction with manual cartographic techniques, high-resolution USDA-FSA-AFPO NAIP digitally orthorectified aerial imagery, and USDA-NASS cropland data layers. A high-throughput geographical information system (GIS), ESRI ArcGIS Desktop, was used for data storage and processing. ELISA assay data were recorded as absorbance reading at 650 nm using Stat Fax 3 200 plate reader (Awareness Technology Inc, FL).

1.2 Methods

1.2.1 *Lygus hesperus* habitat survey

A four-year (2002 – 2005) survey of cotton and non-cotton hosts was conducted to examine the role of

non-cotton hosts in supporting *L. hesperus* populations in cotton in the Texas High Plains. An area wide survey of prevalent non-cotton hosts was conducted in April (pre-season survey) and in late July/early August (to coincide with peak cotton blooming/fruiting) in each of the twenty-five counties comprising the Plains Cotton Growers, Inc. (PCG) service area in the Texas High Plains. At four locations in each county, two 100-sweep samples were collected per host habitat plant species using standard 15 sweep-nets, resulting in 800 sweeps per habitat per county. In each of the twenty-five PCG service area counties, cotton was sampled in late July/early August at a rate of approximately 500 sweeps per county.

1.2.2 Landscape-level study of *Lygus hesperus*

A two year (2008 – 2009) survey of cotton and non-cotton hosts was conducted to examine the effects of host habitat spatial distribution and cotton surrounding landscape composition heterogeneity on *L. hesperus* abundance in Texas High Plains cotton. During each year's cotton growing season, fifty irrigated cotton fields were selected from a six-county (about 1.3986 million hm^2) (2008) or seven-county (about 1.6317 million hm^2) (2009) area near Lubbock, Texas. These selected focal fields and their surrounding habitats were used for a late-season cartographic study and classification of agricultural fields and non-cotton host habitats within a 3-km radius and weekly sweep-net sampling. Of the fifty selected cotton focal fields, ten were chosen for more detailed sampling work, to consist of weekly sampling of six non-cotton host habitat patches within a 3-km buffer zone. Sweep-net samples were placed in cold storage and later processed to determine insect abundances.

Late-season host habitat classification and ground-truthing of the agricultural landscape surrounding cotton focal fields to a buffer radius of 3 km were performed with the aid of GPS technology and in conjunction with manual cartographic techniques. The accuracy of the resulting habitat maps were subsequently validated through meticulous matching and association with high-resolution USDA-FSA-AFPO NAIP digitally orthorectified aerial imagery, as well as USDA-NASS cropland data layers[1]. A high-throughput geographical information system (GIS), ESRI ArcGIS Desktop, was used for data storage and processing.

Twenty-five landscape variables or characteristics were calculated for each focal field surveyed in 2008 and 2009. The landscape characters analyzed werelatitude, longitude, corn area (hm^2), mixed weeds area (hm^2), non-habitat area (hm^2), playa area (hm^2), habitat heterogeneity (Shannon's H′), alfalfa area (hm^2), sorghum area (hm^2), alfalfa distance (m), cotton distance (m), cotton area (hm^2), conservation research program grass or CRP area (hm^2), mixed weeds distance (m), non-habitat distance (m), sunflower distance (m), urban area (hm^2), excess area (hm^2), sunflower area (hm^2), corn distance (m), urban distance (m), CRP distance (m), playa distance (m), and sorghum distance (m). Distances were measured between the closest boundaries of two habitat patches.

1.2.3 *Lygus hesperus* intercrop movement study

A two-year (2008 – 2009) experiment was conducted at the Texas A & M AgriLife Research farm located near Lubbock, Texas to assess bidirectional *L. hesperus* intercrop movement in a cotton-alfalfa system. *Lygus hesperus* intercrop movement was determined by field-marking *L. hesperus* insects in alfalfa and adjacent cotton using two different protein markers, capturing the insects via "keep it simple" KIS vacuum sampling, and then detecting protein markers using indirect ELISA. Field-marking and *L. hesperus* sampling in alfalfa and cotton was initiated at the "7 – 8 true leaf" cotton stage. Weekly spray applications of 10% egg white (EW) marker solution (185 L/hm^2) in alfalfa and 10% non-fat dry milk (NFDM) marker solution (185 L/hm^2) in cotton were made from cotton squaring to cotton boll maturation. KIS sampling was conducted following a 24-hour post-spray foraging period. Indirect ELISA was used to detect *L. hesperus* protein marker acquisition. Based on indirect ELISA results, *L. hesperus* samples were classified into four categories: residents, immigrants, visitors, and roamers. *L. hesperus* testing positive for a protein marker applied in the opposing host were categorized as immigrants. *L. hesperus* testing positive only for the protein marker applied to the capture source host were categorized as residents. *L. hesperus* testing negative for both protein markers were recorded as visitors. *L. hesperus* testing positive for both protein markers, evidence of protein acquisition in both hosts, were recorded as roamers.

1.3 Statistical analysis

Seasonal average *L. hesperus* abundance data from the landscape study were regressed with 27 field characteristics (variables), including habitat-specific land cover, distance between focal cotton fields and non-cotton habitats, longitude, latitude, elevation, habitat heterogeneity index, and several environmental/ecological variables. Significant variables were selected using a stepwise regression at 15% probability rate to develop a habitat association model. Intercrop movement data were analyzed by ANOVA using PROC MIXED, SAS 9.2. Means were separated using LSMEANS with 0.1 alpha levels. The relationship between *L. hesperus* abundance in cotton and the number of immigrants from alfalfa was evaluated via correlation and regression analyses of two-year data. The relationship between al-

falfa *L. hesperus* immigrants and roamers was also determined via correlation and regression analyses.

2 Results

2.1 Non-cotton host habitats of *Lygus hesperus*

Throughout surveying, *L. hesperus* bugs were collected from 23 host plants (Table 1). Among non-cotton host plants, wild mustards (flixweed, tumble mustard, black mustard, and London rocket) supported the highest number of *L. hesperus* adults and nymphs, but these hosts senesced well before cotton was available as a suitable host for *L. hesperus* (Table 1). In most of the High Plains region, London rocket was available as a *L. hesperus* host as early as in late January, whereas no other apparent host plants were available for *L. hesperus* until late February. Therefore, it appears that London rocket is responsible for supporting early-emerging *L. hesperus* (emerging from overwintering quarters) in the northern region of the High Plains.

Among the weed hosts surveyed, samples collected fromhuisache daisy, ragweed, and wavy gaura did not have any *L. hesperus* nymphs, indicating that these weed hosts may not be suitable for *L. hesperus* reproduction. Higher numbers of *L. hesperus* nymphs (>10 nymphs per 100 sweeps) were found in flixweed, tumble mustard, black mustard, London rocket, yellow sweetclover, and alfalfa.

Table 1 Number of 100-sweep samples and seasonal average abundance (number/100 sweeps) **of *Lygus hesperus* bugs in cotton and 22 non-cotton hosts in the Texas High Plains** (all 25 counties averaged), **2003**

Host common name	Scientific name	Number of samples	Adult	Nymph	Total
Flixweed	*Descurainia sophia*	144	170.2	50.6	220.8
Tumble mustard	*Sisymbrium altissimum*	13	105.1	51.6	156.7
Black mustard	*Brassica nigra*	15	102.2	20.6	122.8
London rocket	*Sisymbrium irio*	41	96.2	24.2	120.4
Yellow sweet clover	*Melilotus officinalis*	18	79.4	12.4	91.8
Alfalfa	*Medicago sativa*	222	45.8	10.3	56.1
Curly dock	*Rumex crispus*	3	47.0	1.7	48.7
Russian thistle	*Salsola iberica*	71	39.6	3.9	43.5
Pigweed	*Amaranthus* sp.	76	27.5	7.1	34.6
Redstem filaree	*Erodium cicutarium*	5	14.0	4.6	18.6
Prairie sunflower	*Helianthus petiolaris*	4	10.0	1.8	11.8
Scarletgaura	*Gaura coccinea*	4	9.5	0.5	10.0
Wooly leafbursage	*Ambrosia grayi*	36	9.4	0.5	9.9
Texas blueweed	*Helianthus ciliaris*	49	7.6	1.3	8.9
Kochia	*Kochia scoparia*	30	7.0	0.4	7.4
Huisache daisy	*Amblyolepis setigera*	3	7.3	0.0	7.3
Gumweed	*Grindelia squarrosa*	5	3.2	1.8	5.0
Ragweed	*Ambrosia artemisiifolia*	26	3.5	0.0	3.5
Silverleaf nightshade	*Solanum elaeagnifolium*	45	2.8	0.3	3.1
Blue mustard	*Chorispora tenella*	21	2.8	0.1	2.9
Cotton	*Gossypium hirsutum*	143	2.5	0.3	2.8
Wavygaura	*Gaura sinuata*	3	1.7	0.0	1.7
Wild sunflower	*Helianthus annuus*	20	0.5	0.1	0.6

2.2 Effect of landscape characteristics on *Lygus hesperus* populations in cotton

Of twenty-five landscape variables, only six (latitude, corn area, mixed weeds area, non-habitat area, playa area and habitat heterogeneity) were found to be significantly positively correlated with the number of *L. hesperus* collected from cotton focal fields (Table 2, Fig. 1). Five landscape variables (cotton area, CRP area, mixed weeds distance, non-habitat distance, and sunflower distance) were significantly negatively correlated with *L. hesperus* abundance in focal field cotton.

A stepwise regression analysis of all twenty-five landscape variables predicting *L. hesperus* abundance in focal field cotton showed that only nine variables were useful in a predictive model (Table 3). The multivariate regression model developed based on two years of focal field survey data was significant, with a 93% model R^2 (Fig. 2). Model validation and optimization is yet to be performed, but landscape metapopulation management based upon a strong predictive model is necessary to map potential risks for *L. hesperus* infestation and outbreak. Such a risk map will help in management decision-making and in directing the overall landscape-level pest management effort.

2.3 Intercrop movement dynamics of *Lygus hesperus*

2.3.1 Influx into cotton

Analysis of variance of *L. hesperus* influx into cotton (year and host combined) revealed significant

differences in the pattern of *L. hesperus* influx into cotton among the sampling weeks ($df = 6, 24$; $F = 5.2$; $P = 0.0015$). There were significant interactions between *L. hesperus* intercrop movement with week and year ($df = 6, 24$; $F = 3.74$; $P = 0.0091$). *L. hesperus* influx to a cotton field from nearby alfalfa was very low when cotton was in its vegetative growth stage (0 – 40 DAP, days after planting, prior to July). During the first five weeks following cotton planting, *L. hesperus* was not detected in cotton. Once cotton began squaring (40 – 45 DAP), *L. hesperus* began moving into cotton from alfalfa therefore obviously alfalfa was a source of *L. hesperus* to nearby cotton during the cotton squaring stage.

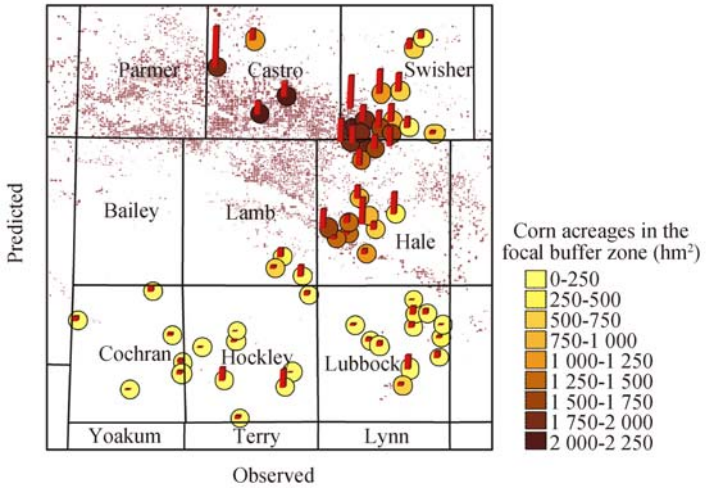

Fig. 1 Color-coded 2009 Texas High Plains corn hectares (per focal field buffer zone) **versus seasonal average *Lygus* abundances per cotton focal field** (indicated by red columns)
2009 corn crop coverage in the Texas High Plains is shown in the background.

Table 2 Relationships of average number of *Lygus hesperus* in cotton field (per 1 000 sweeps) **with various habitat abundance and proximity variables: the Pearson correlation analysis**

Variable	Correlation	Significance
Latitude	+	s
Corn area (hm^2)	+	s
Mixed weed area (hm^2)	+	s
Non-habitat area (hm^2)	+	s
Playa area (hm^2)	+	s
Habitat heterogeneity (Shannon's H′)	+	s
Alfalfa area (hm^2)	+	ns
Longitude	+	ns
Sorghum area (hm^2)	+	ns
Alfalfa distance (m)	+	ns
Cotton distance (m)	+	ns
Cotton area (hm^2)	–	s
CRP area (hm^2)	–	s
Mixedweed distance (m)	–	s
Non-habitat distance (m)	–	s
Sunflower distance (m)	–	s
Urban area (hm^2)	–	ns
Excess area (hm^2)	–	ns
Sunflower area (hm^2)	–	ns
Corn distance (m)	–	ns
Urban distance (m)	–	ns
CRP distance (m)	–	ns
Playa distance (m)	–	ns
Sorghum distance (m)	–	ns

At alpha = 0.1. s: Significant correlation, ns: non-significant correlation.

Table 3 Multivariate linear model variables and parameters predicting average *Lygus hesperus* density in focal field cotton based on various landscape parameters

Variables	Parameter	F value	$Pr > F$	Partial R^2	Model R^2
Intercept	162.2713	10.94	0.01	–	–
Corn area	0.0105	48.43	<0.0001	0.36	0.36
Sunflower area	0.1189	47.81	<0.0001	0.15	0.52
Playa distance	−0.0126	30.70	0.00	0.08	0.60
Mixed weed distance	0.0134	32.23	0.00	0.07	0.66
Corn distance	0.0070	33.97	0.00	0.10	0.77
CRP distance	−0.0101	19.48	0.00	0.05	0.82
Urban distance	0.0043	11.42	0.01	0.04	0.86
Longitude	−0.0002	10.44	0.01	0.04	0.90
Habitat heterogeneity	−2.2409	5.12	0.05	0.03	0.93

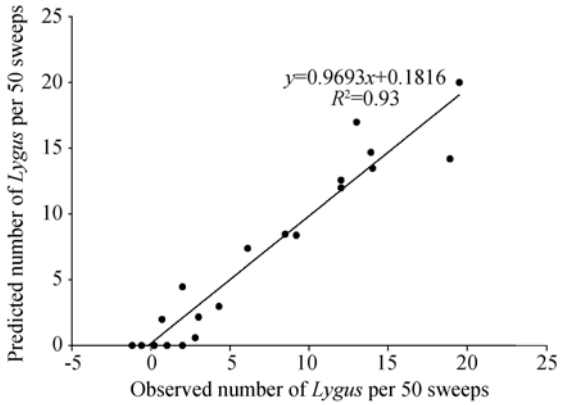

Fig. 2 Model prediction of average *Lygus* abundance in focal fields

In 2008, *L. hesperus* influx into cotton was highest at 83 DAP (fifth sampling week), followed by 97, 62, 76, 47, 69, and 104 DAP. In 2009, *L. hesperus* influx into cotton was highest at 56, 82, and 90 DAP (fifth sampling week), followed by 107, 65, 42, and 56 DAP. Peak *L. hesperus* influx into cotton in both years occurred during the second week of August (during the fifth sampling week), when cotton was in full bloom. *Lygus hesperus* influx was low in all weeks prior to this peak (during squaring). The first peak of *L. hesperus* influx in cotton in 2009 occurred at 56 DAP or during the third sampling week. It is unanticipated to have detected peak influx during squaring, as this is not considered to be the most favorable cotton stage with respect to *L. hesperus*. High influx during squaring in 2009 might be accounted for by temporarily reduced alfalfa quality due to poor irrigation timing as a result of an irrigation system backlog on the research farm and concurrently high temperatures and low relative humidity.

2.3.2 Influx into alfalfa

Analysis of variance of *L. hesperus* influx in alfalfa (both years and both hosts combined) revealed significant differences in the patterns of *L. hesperus* intercrop movement among the sampling weeks ($df = 6, 24$; $F = 10.65$; $P = 0.0001$). Week and year interacted significantly ($df = 6, 24$; $F = 10.78$; $P = 0.0001$) in terms of *L. hesperus* intercrop movement. In 2008, *L. hesperus* influx into alfalfa from nearby cotton was near-zero when cotton was in its vegetative growth stage (0 – 40 DAP, prior to July). *L. hesperus* was not detected in cotton during the first five weeks following cotton planting. Once cotton began squaring (40 – 45 DAP), *L. hesperus* began moving between alfalfa and cotton.

2.3.3 Net movement

L. hesperus net movement into cotton varied significantly ($df = 1, 2$; $F = 230.41$; $P = 0.0043$) between 2008 and 2009. Year and phenological stage affected *L. hesperus* net movement significantly ($df = 2, 32$; $F = 9.57$; $P = 0.0006$). In both 2008 and 2009, *L. hesperus* net movement was negative during cotton squaring, indicating net outflow from cotton. Net outflow peaked during the second week of July, at 49 – 62 DAP in 2008 and 2009 (Fig. 3). During this period, few *L. hesperus* (< 1 *L. hesperus*/12 m^2) were retained in cotton, and most *L. hesperus* visiting cotton moved back to alfalfa. In 2008, as cotton grew older, and cotton squares continued to grow and blooming began, *L. hesperus* net movement, with respect to cotton, gradually increased from negative toward zero, and became positive as cotton approached full bloom (76 DAP). Thereafter, *L. hesperus* net movement remained positive in cotton, indicating an increased cotton capability to retain more *L. hesperus* having moved from alfalfa. In 2009, *L. hesperus* net movement in cotton never became positive during the cotton growing season. This indicates that alfalfa remained more attractive than cotton throughout the cotton growing season. Unexpectedly, *L. hesperus* density in cotton increased continually, and cotton was able to retain some *L. hesperus* migrants from alfalfa, even when *L. hesperus* net movement was negative in cotton.

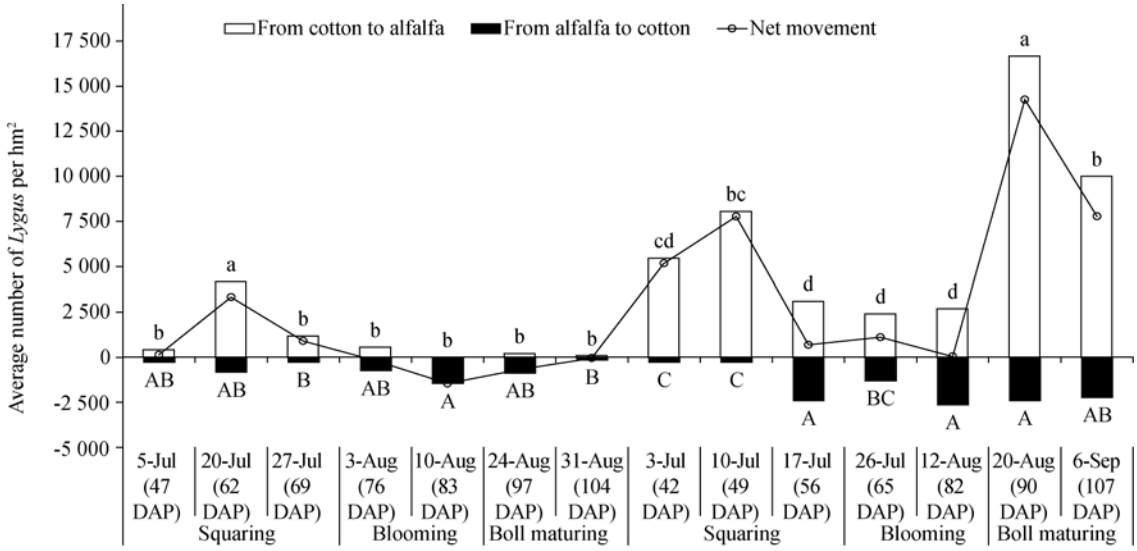

Fig. 3 Weekly average *Lygus* abundance in cotton and alfalfa and net *Lygus* intercrop movement between alfalfa and adjacent cotton, Lubbock, Texas, 2008 (left) **and 2009** (right)

DAP: Days after planting. 1 hm^2 = 833.3 KIS samples, the negative value suggest the opposite direction of *Lygus* movement (i.e., from alfalfa into cotton). The similar alphabet on the top or bottom of the bar among the sampling weeks within a specific host and year are not significantly different at alpha = 0.1 when means were separated by LSMEAN.

3 Discussion

In the northern region, five (Hale County) to 12 (Lubbock County) non-cotton host plants were observed to "bridge" the sequence between non-cotton host plants and cotton during the cotton squaring stage. However, in the southern region, alfalfa and Russian thistle were the only non-cotton hosts that bridged the host sequence with cotton during cotton fruiting season. Among the weed hosts surveyed, samples collected from huisache daisy, ragweed, and wavy gaura did not have any *L. hesperus* nymphs, indicating that these weed hosts may not be suitable for *L. hesperus* reproduction. Higher numbers of *L. hesperus* nymphs (>10 nymphs per 100 sweeps) were found in flixweed, tumble mustard, black mustard, London rocket, yellow sweetclover, and alfalfa. Identification of reproductively suitable host habitats for *L. hesperus* would later help in determining sub-populations and estimating the total *L. hesperus* population in Texas High Plains landscape.

The season-long cotton survey showed that the *L. hesperus* metapopulation is divided amongst many habitats and geographically specific sub-populations. Cotton growers are primarily concerned with managing *L. hesperus* sub-populations present in their cotton fields. Understanding metapopulation dynamics among these sub-populations is essential for successful management of *L. hesperus* in cotton. Promoting those landscape characteristics showing negative relationships with *L. hesperus* sub-populations in cotton and minimizing those showing positive relationships with *L. hesperus* in cotton is necessary for landscape-level *L. hesperus* metapopulation dynamics management. A study investigating physical tracking of *L. hesperus* intermigration dynamics between or among major habitats was conducted to validate this model and further characterize *L. hesperus* metapopulation dynamics.

In order to understand the effect of *L. hesperus* influx dynamics on a *L. hesperus* population in cotton, and to understand the potential for cotton injury due to *L. hesperus*, *L. hesperus* retention in cotton, rather than influx, is more important. For the purposes of this study, *L. hesperus* retention was calculated by subtracting outflow from inflow. Inflow was estimated via sampling, but estimating outflow is difficult. Assuming the cotton-alfalfa system as closed, *L. hesperus* outflow from cotton was estimated by sampling alfalfa and determining the quantity of influx into adjacent alfalfa. In this study, *L. hesperus* influx in cotton and alfalfa was quantified for each sampling week.

Net *L. hesperus* intercrop movement between cotton and alfalfa was calculated by subtracting cotton 'Total *L. hesperus* Influx' (EW-marked *L. hesperus* captured in cotton) from cotton 'Total *L. hesperus* Outflux' (NFDM-marked *L. hesperus* captured in alfalfa). Positive net movement values indicate net *L. hesperus* gains in cotton. Likewise, negative net movement values indicate net *L. hesperus* losses. *L. hesperus* net movement data have the potential to indicate the timing of host source-sink dynamics-information which may be of value in making pest management decisions.

Although differences in crop structure, combined with the chosen sampling method, may have led to overestimation of *L. hesperus* densities in alfalfa, it is

likely that naturally higher *L. hesperus* densities in alfalfa and naturally high *L. hesperus* intercrop movement between cotton and alfalfa may have contributed to this overestimation. Many *L. hesperus* from alfalfa may have visited cotton, but most returned to alfalfa. Only a few actually "settled" in cotton. Each time a large number of *L. hesperus* move from alfalfa to cotton, a few *L. hesperus* may remain and settle, which explains the steady, gradual *L. hesperus* population increase in cotton. However, since most returned to alfalfa, *L. hesperus* net movement calculations indicated high *L. hesperus* influx into alfalfa.

L. hesperus sub-populations in agricultural field crops and host habitats continuously interact, and these interactions represent an excellent opportunity for exploitation in *L. hesperus* metapopulation management. This is particularly true, given that even if all *L. hesperus* are removed from a specific crop field, *L. hesperus* source populations residing in nearby habitats will continue to exert considerable re-colonization pressure and pose an infestation risk. Managing pests at the landscape-level via intelligent exploitation of metapopulation dynamics may prove to be sustainable, economical, and environmentally conscious tool for use in conjunction with other methods in an integrated pest management system. In fact, development of elegant, environmentally conscious pest management approaches requires a greater understanding of metapopulation dynamics. Further detailed investigations of *L. hesperus* metapopulation dynamics in the Texas High Plains is necessary for continued development of landscape-level, sustainable, integrated approaches to *L. hesperus* management.

Acknowledgements

This project was partially funded by Cotton Incorporated Core Program, USDA CSREES RAMP, International Cotton Research Center, and Plains Cotton Growers, Inc. Several former graduate students, technicians, and numerous undergraduate research students are acknowledged for their technical service in several projects represented in this paper.

References

[1] National Agricultural Statistics Service (NASS). http://www.nass.usda.gov/Statistics_by_Subject/index.php? sector = CROPS
[2] Williams M R. Cotton insect losses - 2010. //Proceedings of the Beltwide Cotton Conferences, National Cotton Council, Memphis, TN, 2011: 896 – 940
[3] Frisbie R E, Crawford J L, Bonner C M, et al. Implementing IPM in cotton. //Frisbie R E, El-Zik K M, Wilson L T. Integrated pest management systems and cotton production. New York: John Wiley & Sons, 1989: 389 – 412
[4] Luttrell R G. Cotton pest management: Part 2. A U. S. perspective. Annual Review of Entomology, 1994, 39: 527 – 542
[5] Day W H. Evaluation of biological control of the tarnished plant bug (Hemiptera: Miridae) in alfalfa by the introduced parasite *Peristenus digoneutis* (Hymenoptera: Braconidae). Environmental Entomology, 1996, 25(2): 512 – 518
[6] Parajulee M N, Arnold M D, Carroll S C, et al. *Lygus* abundance on wild hosts: a survey across the Texas High Plains. // Proceedings of the Beltwide Cotton Conferences, National Cotton Council, Memphis, TN, 2003: 970 – 973
[7] Sevacherian V, Stern V M. Host plant preference of *Lygus* bugs in alfalfa-interplanted cotton fields. Environmental Entomology, 1974, 3(5): 761 – 766
[8] Barman A K, Parajulee M N, Carroll S C. Relative preference of *Lygus hesperus* (Hemiptera: Miridae) to selected host plants in the field. Insect Science, 2010, 17: 542 – 548
[9] Sevacherian V, Stern V M. Movement of *Lygus* bugs between alfalfa and cotton. Environmental Entomology, 1975, 4(1): 163 – 165
[10] Fleischer S J, Gaylor M J, Hue N V. Dispersal of *Lygus lineolaris* (Heteroptera: Miridae) adults through cotton following nursery host destruction. Environmental Entomology, 1988, 17(3): 533 – 541
[11] Mueller S C, Summers C G, Goodell P B. Strip cutting alfalfa for *Lygus* management: forage quality implications. Crop Management, 2005, 4(1): 105 – 112
[12] Snodgrass G L, Scott W P, Hardee D D. Results from two years of an experiment on tarnished plant bug control in cotton through reduction in numbers of early-season wild host plants. //Dugger P, Richter D. Proceedings of the Beltwide Cotton Conference, National Cotton Council, Memphis, TN, 2000: 1229 – 1233

(责任编辑:高 峰)

Agricultural and Forest Entomology

Agricultural and Forest Entomology (2014), **16**, 24–32 DOI: 10.1111/afe.12031

Intraspecies mixture exerted contrasting effects on nontarget arthropods of *Bacillus thuringiensis* cotton in northern China

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Abstract

1 Row-intercropping is a type of multiple cropping with two or more crops grown simultaneously in alternate rows in the same area. It is a traditional agronomic practice and is still prevalent in modern Chinese agricultural ecosystems. Many studies have proposed that intercropping at the crop species level can significantly contribute to pest management when properly managed. However, the performance of intercropping at the plant genotype level is still largely unknown.

2 A multiyear field experiment was conducted to examine the effects of intraspecies *Bacillus thuringiensis (Bt)*/non-*Bt* crop mixture on nontarget arthropods. Densities of dominant pests and predators were assessed via direct visual observations.

3 Cotton aphid population levels in monoculture *Bt* cotton fields were greater than that observed in non-*Bt* cotton, whereas the row-mixture planting of *Bt* and non-*Bt* suppressed the abundance of cotton aphids compared with that in monoculture of either genotype. Investigations also demonstrated that the intraspecies row-mixture increased whitefly abundance compared with monoculture of either genotype. However, the mixture exerted neutral effects on population sizes of mirid bugs and predators.

4 These results suggest that crop cultivation management is insufficient to control secondary pests of *Bt* cotton, and thus multiple pest suppression strategies are warranted.

Keywords *Bt* cotton, cotton aphid, intercropping, mirid bug, predator, whitefly.

Introduction

Cotton bollworm *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae), once the major cotton pest in northern China, has been effectively controlled by the adoption of transgenic cotton expressing a δ-endotoxin from *Bacillus thuringiensis* (*Bt*). However, cultivation of *Bt* cotton led to substantial variations in crop composition and pest management practices, which in turn changed the arthropod community structures within cotton ecosystems, resulting in a greater herbivore population size in *Bt* cotton compared with that in non-*Bt* cotton (Wilson *et al.*, 1992; Cui & Xia, 1998, 2000; Greene *et al.*, 1999; Herron *et al.*, 2000; Wu *et al.*, 2002; Deng *et al.*, 2003; Lu *et al.*, 2010). For example, Cui and Xia (1998, 2000) found that populations of *Aphis gossypii* Glover (Hemiptera: Aphididae), *Tetranychus cinnbarinus* Boisduval (Prostigmata: Tetranychidae), *Trialeurodes vaporariorum* Westwood (Hemiptera: Aleyrodidae) and *Empoasca biguttula* Ishida (Hemiptera: Cicadellidae) were elevated in *Bt* cotton fields compared with that in non-*Bt* cotton. Herron *et al.* (2000) and Deng *et al.* (2003) found that cotton aphid populations in *Bt* cotton were significantly larger compared with non-*Bt* cotton. Wu *et al.* (2002) and Lu *et al.* (2010) reported that the widescale use of *Bt* cotton has led to a frequent outbreak of mirid bugs in northern China. However, population densities of major predator species in *Bt* cotton fields were significantly greater than those in conventional cotton receiving pesticide applications (Wu & Guo, 2005; Sisterson *et al.*, 2007;

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Lu *et al.*, 2012). Finally, evidence suggests that *H. armigera* populations in northern China have developed field-evolved resistance to *Cry1Ac*-expressing *Bt* cotton (Liu *et al.*, 2010). Therefore, the longevity of *Bt* cotton is dependent on its control effect on the resistance development of target pests and outbreaks of nontarget pests.

Various refuge strategies have been field-tested for delaying the resistance development of target pests to *Bt* crops (Gould, 1998; Tabashnik *et al.*, 2005) with promising effects (Tabashnik *et al.*, 2008, 2009; Wu *et al.*, 2008). At the same time, much effort has been directed toward managing the secondary pest complex in *Bt* cotton, and increasing the biological control effect of natural enemies is an effective strategy for overall *Bt* cotton management. Numerous studies suggest that the enhancement of predator abundance and diversity through increasing plant diversity exerts positive effects on pest control in many cropping systems (Andow, 1991; Parajulee *et al.*, 1997; Parajulee & Slosser, 1999; Men *et al.*, 2004; Gardiner *et al.*, 2009).

In most agro-ecosystems, strip intercropping, namely the planting of two or more crops simultaneously in different strips in a manner to permit independent cultivation, as well as allowing the crops to interact agronomically (Vandermeer, 1992), is the principal strategy in plant diversity enhancement. Such strip intercropping could be achieved via interspecies or intraspecies row-mixtures. An interspecies row-mixture refers to the planting of two or more species of crops simultaneously in the same field, whereas an intraspecies mixture is the planting of two or more genotypes of the same crop species simultaneously in the same field. A few studies have documented the effects of intraspecies mixtures on the predator complex and any resulting pest control in cotton fields (Sisterson *et al.*, 2004; Yang *et al.*, 2012). For cotton fields, the intraspecies row-mixture of *Bt* and non-*Bt* cotton is equivalent to setting a structured refuge.

The present study aimed to explore the feasibility of utilizing a structured refuge to suppress nontarget pests of *Bt* cotton in small-holder agro-ecosystems of northern China. We hypothesized that a row-mixture planting of *Bt* and non-*Bt* cotton would exert a positive effect on pest control, and that this effect would be irrespective of cotton genotype. We also hypothesized that the effect of an intraspecies mixture on pest and predator abundance would be consistent across growing seasons.

Materials and methods

Field experimental design

Field experiments were conducted at the Langfang Experiment Station (39.538°N, 116.708°E) of the Chinese Academy of Agricultural Sciences (CAAS), located in the Jiuzhou County of Hebei Province. Before 2008, field corn was grown in the selected fields. Based on the current *Bt* cotton adoption rate of the Hebei Province and the refuge size for target pests recommended by Vacher *et al.* (2003), we set up three experimental treatments: (i) monoculture of a *Bt* cultivar; (ii) monoculture of a non-*Bt* cultivar; and (iii) intercropping of 75% *Bt* and 25% non-*Bt*. Intercropping plots were planted in a repeated pattern: one row of non-*Bt* and then three rows of *Bt*. The pattern continued until all rows within a plot were occupied. A randomized complete block design was used with four replications. Each plot within a block encompassed approximately 0.33 ha (length 20 m, width 16.5 m), which is a typical cotton field size in the Hebei Province. Seeding was performed at a rate expected to produce 40 000 plants per planted ha. A 3-m fallow space was left between plots and among blocks to decrease insect dispersion among treatments (Wu & Guo, 2003; Li *et al.*, 2010). Cotton was maintained with agronomic practices standard to northern China, although no fungicides or insecticides were applied to the experimental plots. Plot layout and management practices were identical across all three study years.

Cotton genotypes

The cotton genotypes used in the present study included a genetically modified *Bt* cotton (cv 'GK-12', expressing a δ-endotoxin from *Bt*) and a non-*Bt* cotton (cv 'Simian-3', the parental line of 'GK-12'). The seeds of the two genotypes were provided by colleagues from the Biotechnology Research Center of CAAS. Cotton genotypes exhibited marked differences in leaf trichome density (Xue *et al.*, 2008), *Bt* toxin content (Zhang *et al.*, 2006) and associated resistance to lepidopteran species.

Arthropod sampling

Arthropods sampled included three pest species groups [cotton aphid *A. gossypii*; mirid bug complex *Lygocoris lucorum* Meyer-Dur, *Adelphocoris suturalis* Jackson and *Adelphocoris fasciaticollis* Reuter; and whitefly *Bemisia tabaci* (Gennadius) biotype B] and four predator groups [ladybirds beetles *Coccinella septempunctata* L. and *Propylaea japonica* Thunberg; lacewing *Chrysoperla sinica* (Tjeder); spiders complex and *Orius similis* Zheng]. In each growing season, arthropod sampling was conducted every 10 days from early June until mid-September, corresponding to 4 weeks after cotton seedling emergence to plant defoliation for harvest preparation. Arthropod groups were sampled by visually inspecting 20 cotton plants at five randomly chosen sampling sites distributed across the two diagonal lines of the plot (100 plants per plot) *in situ*. Because of practical concerns as a result high densities, cotton aphid and whitefly populations were quantified by visually inspecting three leaves each from the upper, middle and lower main stem portions of the plant, respectively. In total, nine leaves per selected plant were investigated. For other arthropods, entire plants were visually inspected in the morning (8.00–10.00 h) or afternoon (16.00–18.00 h), with particular attention being paid to flowers and squares, which are likely hiding places for feeding insects.

Statistical analysis

Data obtained from the 100 total plants from the five sampling sites within each experimental plot were pooled to correct for data dependency, and so each plot was used as a replication

unit. Arthropod density responses to treatments were analyzed by two steps. First, the overall effects of these factors on pests and predator abundances during the 3-year study were analyzed with a linear mixed effect model using planting pattern and cotton genotype as a fixed factor, and year as a random factor (SAS Institute, 2003). Then, the effects of planting pattern (monoculture or row-mixture), cotton genotype (*Bt* or non-*Bt*), sampling date, and their interactions, on the abundance of natural enemies and herbivores in each growing season were further analyzed separately with a PROC MIXED procedure in repeated measures analysis of variance (SAS Institute, 2003). Differences in arthropod abundances on specific sampling dates were separated using Fisher's least significant difference. When necessary, the data were $\sqrt{(n+0.5)}$ transformed or $\log(n+1)$ transformed to satisfy assumptions of normality and homogeneity of variance before analysis of variance.

Results

Row-mixture intercropping arrested the abundance of cotton aphids

Cotton aphid population size varied significantly across years and sampling dates. The abundance of cotton aphid on *Bt* cotton was higher that that of non-*Bt*, and row-mixture intercropping markedly decreased the abundance of cotton aphid throughout all 3 years of the study (Figs 1 and 2). In addition, the interaction between planting pattern and cotton genotype was statistically significant (Table 1).

In each growing season, cotton aphid population levels varied significantly across sampling dates (Figs 1 and 2 and Table 2). Row-mixture intercropping significantly depressed the abundance of cotton aphid compared with the *Bt* or non-*Bt* monoculture (Fig. 1). At the same time, the densities of cotton aphid varied greatly within cotton genotypes in monoculture fields and across growing seasons (Fig. 2 and Table 2). The effect of cotton genotype on cotton aphid densities changed with sampling date, as did the effect of planting pattern. The impact of *Bt* cotton on population size of cotton aphid varied greatly among years. In 2008, the abundance of cotton aphid in *Bt* cotton was markedly higher than that in *non-Bt* cotton (Fig. 2A–D), whereas, in 2009 and 2010, no significant differences in cotton aphid were found between *Bt* and non-*Bt*. In addition, the interaction between cotton genotype and planting pattern was not significant for cotton aphid, except for the 2008 growing season (Table 2).

Row-mixture intercropping exerted a neutral effect on the abundance of mirid bugs

Abundances of mirid bugs showed significant variations across years and sampling dates, although planting pattern and genotype had no marked impact on mirid bugs activities. Yet, the interactions between year and planting pattern were statistically significant (Table 1).

Discernible fluctuations of mirid bugs abundance were found across the sampling dates for all years (Fig. 3 and Table 2), although comparable numbers of mirid bugs were found between *Bt* and non-*Bt* cotton fields at the same sampling date. Row-mixture intercropping showed no pronounced effect on the abundance of mirid bugs compared with the monocultures of

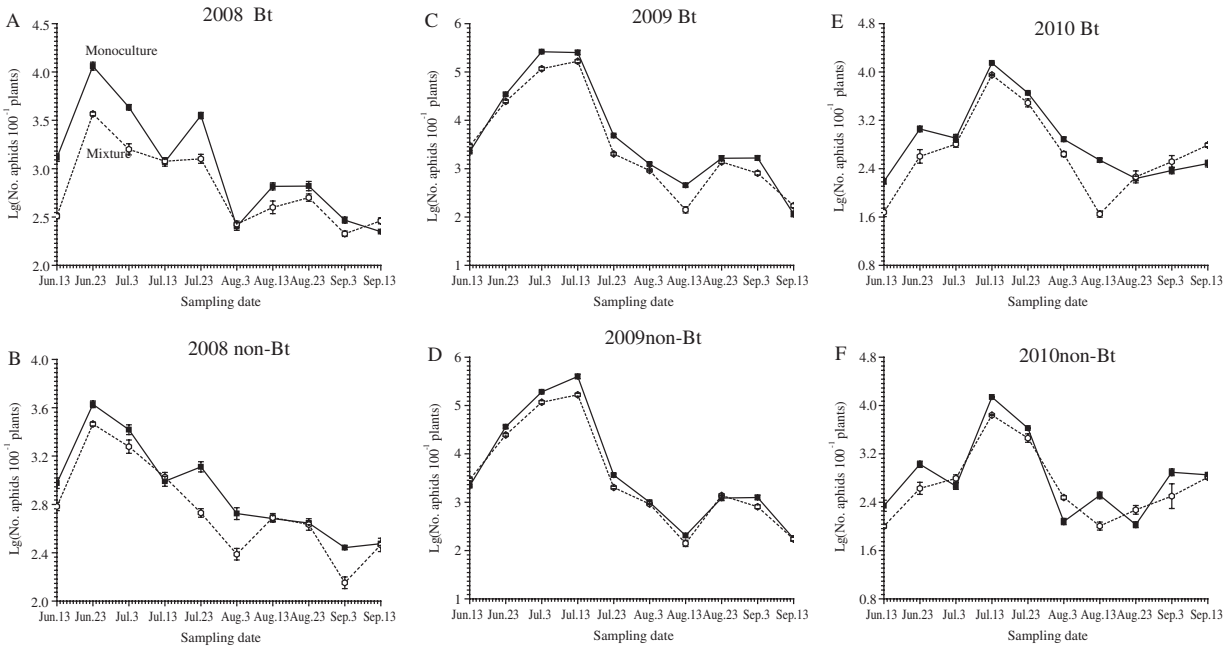

Figure 1 Dynamics of the cotton aphid on the same cotton genotype under different planting patterns [monoculture *Bacillus thuringiensis (Bt)* cultivar, monoculture non-*Bt* cultivar, and mixed-rows of same *Bt* and non-*Bt* cultivars] from mid-June to mid-September in (A, B) 2008, (C, D) 2009 and (E, F) 2010. Solid-lines on the line graphs represent population sizes (mean ± SE) of the monoculture fields, whereas the dotted-lines represent those of the mixture of *Bt* and non-*Bt* cotton at a row ratio of 75% to 25%, respectively.

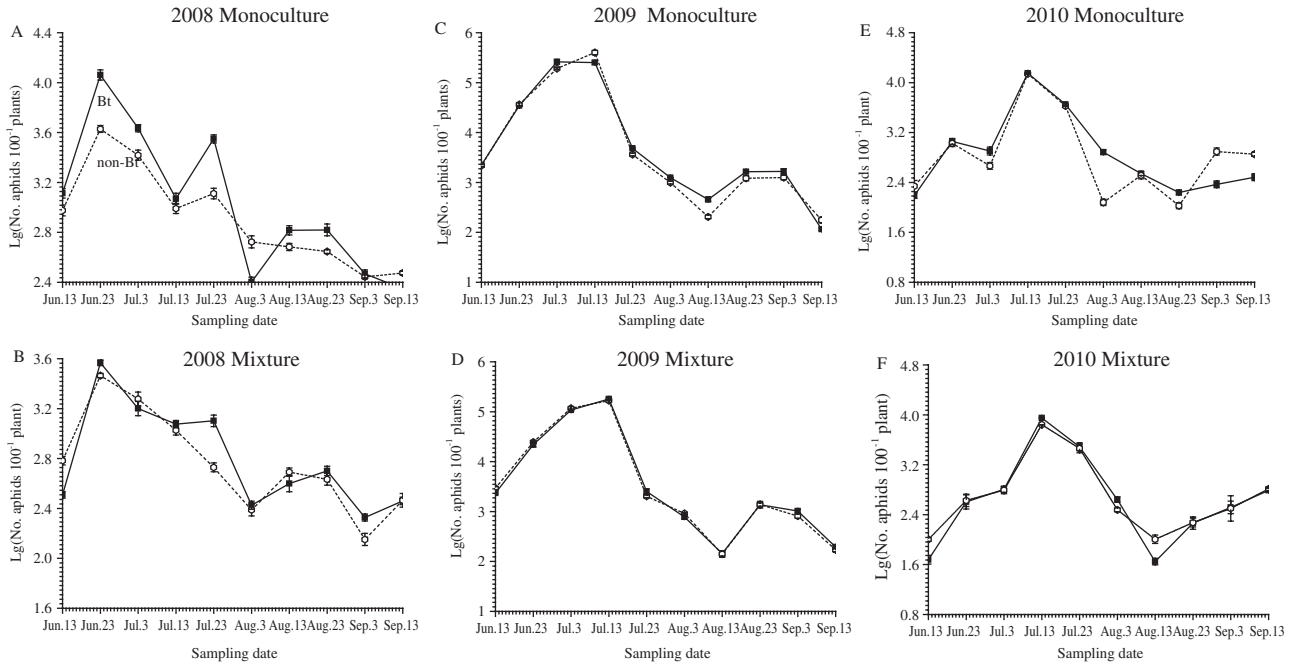

Figure 2 Dynamics of cotton aphid on *Bacillus thuringiensis (Bt)* cotton and non-*Bt* cotton under different planting patterns (monoculture *versus* mixture) from mid-June to mid-September in (A, B) 2008, (C, D) 2009 and (E, F) 2010. Solid-lines on the graphs represent population sizes (mean ± SE) of monoculture fields, whereas the dotted-lines represent those of the mixture of *Bt* and non-*Bt* cotton at a row ratio of 75% to 25%, respectively.

Table 1 *F*- and *P*-values from the linear mixed model estimated effect of cotton genotype, planting pattern, year and their interactions on population size of herbivores in northern China cotton fields in 2008, in 2009 and 2010

		Cotton aphid		Mirid bugs		Whitefly	
Factor	d.f.	*F*	*P*	*F*	*P*	*F*	*P*
Year	2,40	2300.90	< 0.0001	289.63	< 0.0001	51.28	< 0.0001
Genotype	1,40	10.66	0.002	0.08	0.781	0.49	0.488
Pattern	1,40	268.34	< 0.0001	3.85	0.057	17.64	0.001
Date	9, 459	82.77	< 0.0001	104.77	< 0.0001	618.25	< 0.0001
Year × Genotype	2,40	10.91	0.002	0.30	0.589	1.84	0.183
Year × Pattern	2,40	1.34	0.273	3.29	0.049	0.35	0.707
Genotype × Pattern	1,40	5.44	0.008	2.33	0.112	2.16	0.129
Genotype × Pattern × Year	2,40	0.18	0.835	1.57	0.221	1.01	0.372

either the *Bt* or non-*Bt* genotypes in the 2009 and 2010 growing seasons (Fig. 3 and Table 2). However, in 2008, the population size of mirid bugs was higher in intercropping fields compared with the corresponding cotton genotype in monoculture fields (Fig. 3C, D). The interaction between genotype and planting pattern was significant for the growing season of 2008 (Table 2). In conclusion, no clear trends were found because the effect of cotton genotype and planting pattern on the population size of mirid bugs changed with sampling date.

Row-mixture intercropping increased abundances of whiteflies

The results of a linear mixed model indicated that there were significant variations in the abundances of whiteflies across years and sampling dates. In addition, the row-mix planting pattern consistently showed increased whitefly densities. However, differences as a result of cotton genotype were not detectable. Furthermore, neither the interactions between each of two factors (year, planting pattern and genotype), nor the interactions of all the factors were statistically significant (Table 1).

Whitefly densities varied significantly across sampling dates. There were significant differences in abundance of whitefly between the two planting patterns (monoculture versus mixed-row plantings) in most of the investigating periods (Fig. 4 and Table 2). The row-mixture plantings increased the abundance of whitefly compared with the same genotype monocultures, whereas the effect of cotton genotype on whitefly abundance was negligible in most cases, whether under monoculture or mixture. Moreover, the interaction between planting pattern and

Table 2 *F*-values of the repeated measures analysis of variance testing the effects of planting pattern, cotton genotype and sampling date on population sizes of cotton aphid, mirid bugs and whitefly in northern China cotton fields in 2008, 2009 and 2010

Year	Factor	d.f.	Cotton aphid	Mirid bugs	Whitefly
2008	G	1,12	39.46***	4.7	5.93*
	P	1,12	238.67***	5.21*	16.45**
	D	9,108	501.33***	149.26***	2875.59***
	G × D	9,108	18.41***	4.14***	7.14***
	P × D	9,108	19.01***	14.34***	2.88**
	G × P	1,12	11.34**	7.65*	0.39
	G × P × D	9,108	9.98***	8.62***	2.36*
2009	G	1,12	3.59	1.11	0.85
	P	1,12	88.03***	3.55	1.52
	D	9,108	3829.37***	128.47***	389.24***
	G × D	9,108	6.09***	2.78**	0.32
	P × D	9,108	19.44***	4.90***	7.12***
	G × P	1,12	2.75	0.02	0
	G × P × D	9,108	6.26***	4.24***	0.92
2010	G	1,12	0.04	0.98	0.88
	P	1,12	42.55***	1.46	18.86***
	D	9,108	404.33***	218.71***	668.19***
	G × D	9,108	13.10***	3.01**	0.5
	P × D	9,108	20.35***	14.81***	11.72***
	G × P	1,12	1.79	0.75	2.59
	G × P × D	9,108	7.74***	2.59**	1.34

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

G, cotton genotype; P, planting pattern; D, sampling date; G × D, interaction between cotton genotype and sampling date; P × D, interaction between planting pattern and date; G × P, interaction between cotton genotype and planting pattern. G × P × D, interaction between cotton genotype, planting pattern and sampling date.

sampling date was significant for all growing seasons. However, the interactions between planting pattern and sampling date, and between cotton genotype, planting pattern and sampling date, were only significant for the 2008 growing season.

Row-mixture intercropping failed to enhance the abundance of predators

Overall, the predator abundance significantly varied between treatments among years and across sampling dates (Table 3). Cotton genotype and planting pattern contributed little to the variances in predator abundance, and this effect was consistent among growing seasons. However, the interactions between year and planting pattern were significant for most of the taxa group (Table 3).

The densities of all predator taxa fluctuated greatly across sampling dates (Table 4) but did so equally between monoculture and row-mixture intercropping fields, whether for *Bt* or non-*Bt* cotton fields in most cases, except for the growing season of 2008 (Table 4). The abundance of predators, such as adult ladybirds, *O. similis* Zheng and spiders, was higher in the non-*Bt* cotton field compared with that in the *Bt* field, whether for monoculture or mixture fields in 2008. The effect of planting pattern on the population size of adult ladybirds changed with sampling date for the 2008 and for 2010 growing seasons. At the same time, the effect of cotton genotype and planting pattern on spiders abundance changed with sampling date, and no clear trends were found for all the years tested.

Discussion

Impact of row-mixture as a Bt resistance management approach on cotton aphid

Cotton aphid abundance was higher on *Bt* cotton than on non-*Bt* cotton in 2008, whereas, in 2009 and 2010, the population size of cotton aphid in *Bt* fields was similar to that of the non-*Bt* fields. Many other studies have also reported that the abundance of cotton aphid in *Bt* cotton is higher compared with that in conventional non-*Bt* cotton (Wilson *et al.*, 1992; Cui & Xia, 1998; Greene *et al.*, 1999; Deng *et al.*, 2003). The discrepancy observed among the seasons in the present study may be a result of varying environmental conditions and arthropod complexes across study years.

Furthermore, intraspecies intercropping has suppressed the abundance of cotton aphid. This result supports our hypothesis that intraspecies mixtures would improve pest suppression. This finding is congruent with previous studies indicating that intercropping exerts strong positive effects on pest control (Litsinger & Moody, 1976; Risch, 1981; Andow, 1991; Altieri & Nicholls, 2004; Bomford, 2004; Shrewsbury & Raupp, 2006; Björkman *et al.*, 2010). However, the effects of mixed-row intercropping showed a significant variation among years and within genotypes. Xue *et al.* (2008) stated that the outbreak of cotton aphid was more frequently observed in transgenic *Bt* cotton because the lower leaf trichome density of transgenic *Bt* cotton facilitated aphid feeding compared with conventional non-*Bt* cultivars. Accordingly, we would have expected an

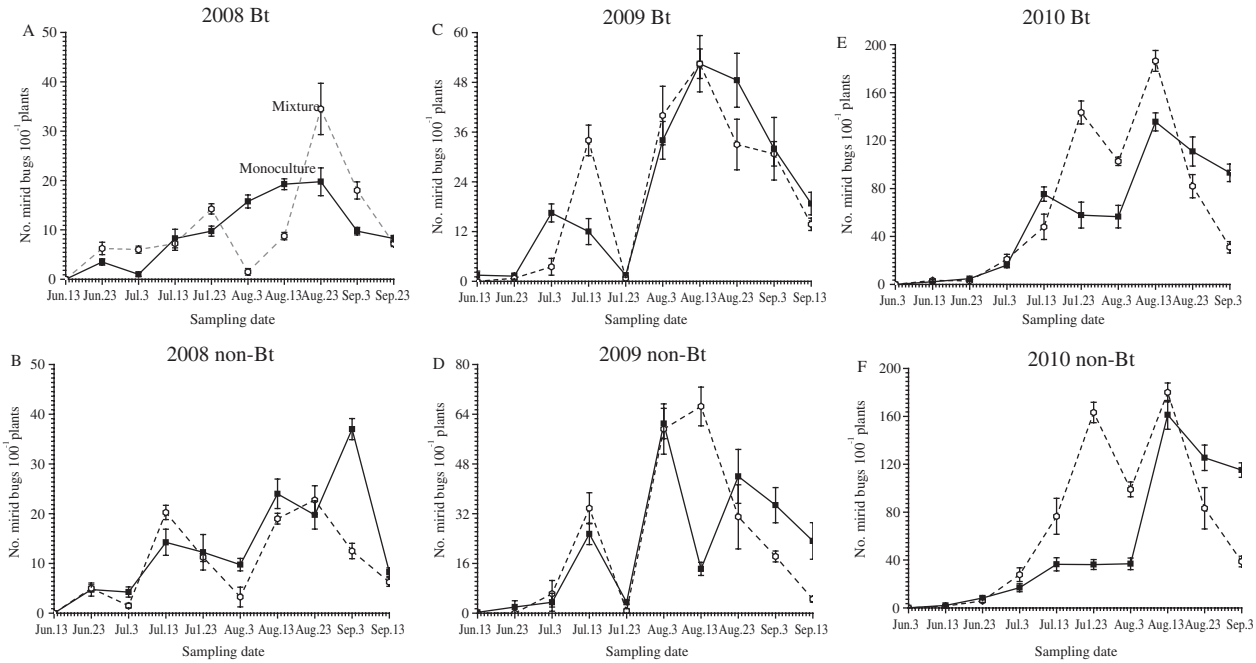

Figure 3 Dynamics of mirid bugs on *Bacillus thuringiensis (Bt)* cotton and non-*Bt* cotton with different planting patterns (monoculture *Bt* cultivar, monoculture non-*Bt* cultivar, and mixed-rows of same *Bt* and non-*Bt* cultivars) from mid-June to mid-September in (A, B) 2008, (C, D) 2009 and (E, F) 2010. Solid-lines on the line graphs represent population sizes (mean ± SE) of the monoculture fields, whereas the dotted-lines represent those of the mixture fields of *Bt* and non-*Bt* cotton at a row ratio of 75% to 25%, respectively.

intermediate cotton aphid population in mixture plots, with the highest densities in *Bt* plots and the lowest densities in non-*Bt* plots. However, the suppression effect of mixture on cotton aphid was similar between the two genotypes. This indicates that there may be other factors contributing to the observed phenomenon.

Influence of row-mix intercropping on whiteflies and mirid bugs

By contrast to our hypothesis, intercropping increased the occurrences of whitefly in the present study. At the same time, intercropping failed to alter the abundances of mirid bugs. The specific response of pests to intercropping may result from dispersion capability differences. Furthermore, the effect of intercropping on pests is partly determined by plant resistance, whereas plant resistance changes with the developmental age of plant (Barton & Koricheva, 2010). In addition, plants can modulate their defensive strategy based on neighbour identity (Broz *et al.*, 2010). The discrepancy of mixed-row plantings on mirid bugs among seasons may be the result of variation in climate and interactions among arthropods.

Neutral effects of row-mix intercropping on predator abundance

Planting pattern did not significantly influence the predator abundance in most of cases. Therefore, our expectation of increased predator activities in intercropped fields was rejected. Takizawa and Snyder (2011) suggested that higher predator biodiversity fostered the survivorship of juveniles, which in turn increased reproductive rates and contributed more offspring to succeeding generations, along with an increased foraging efficiency. In the present study, the abundances of predators, such as ladybirds and spiders, in intraspecies mixture cotton fields were higher than the corresponding genotype of monoculture cotton fields in 2008. However, this phenomenon was not observed in 2009 and 2010. In general, lower prey abundances are expected to aggravate intraguild predation and competition and thus lead to reduced activity and lower reproduction rates. Considering all of the factors noted previously, it is not unexpected that the intraspecies plantings in the present study did not enhance the occurrence of predators when prey is not sufficient.

Implications for future pest management

Although the widespread planting of *Bt* cotton has led to area-wide population suppression of key target pest species, such as *H. armigera* (Wu *et al.*, 2008), *Bt* cotton adoption has also led to the outbreak of mirid bugs (Wu *et al.*, 2002; Lu *et al.*, 2010) . Therefore, management of nontarget pests is a new requirement for the sustainable application of *Bt*-transgenic cotton. From the perspective of delaying resistance development in a target pest, Wu *et al.* (2008) argued that no structured refuge is advisable as a result of the presence of natural refuges provided by the wide diversity of crops in northern China. However, other studies report that the widescale planting of *Bt* cotton has led to an increased resistance frequency in target pests in some regions

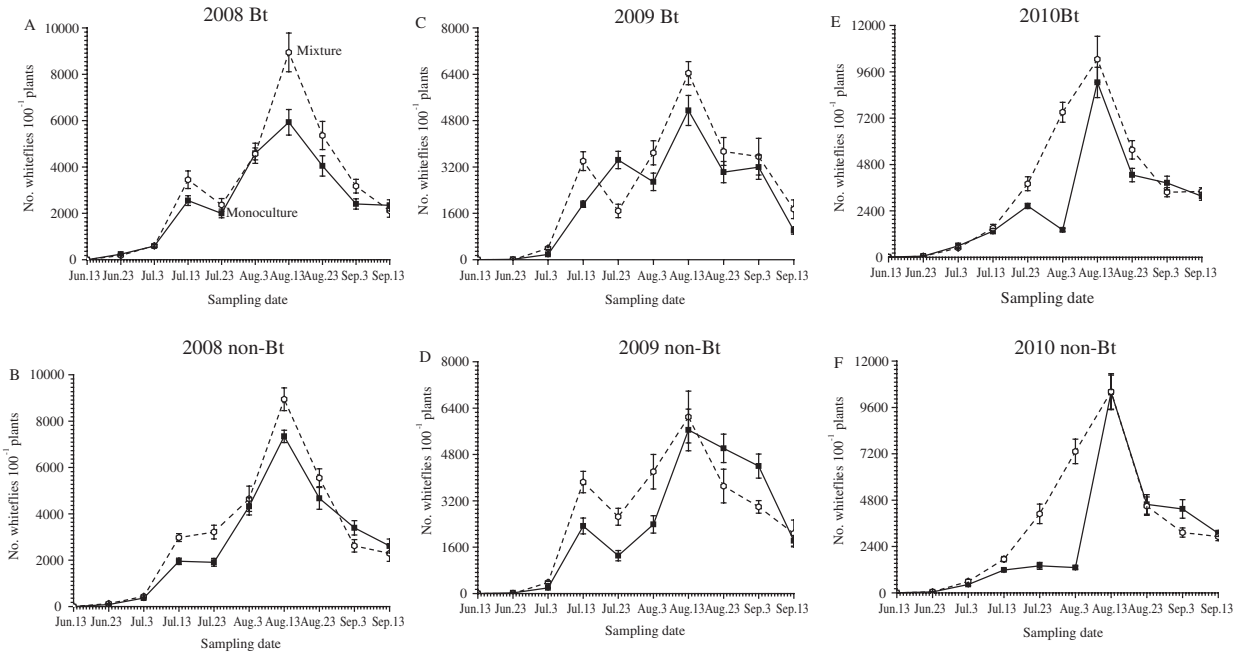

Figure 4 Dynamics of whitefly on *Bacillus thuringiensis (Bt)* cotton and non-*Bt* cotton with different planting patterns (monoculture *Bt* cultivar, monoculture non-*Bt* cultivar, and mixed-rows of same *Bt* and non-*Bt* cultivars) from mid-June to mid-September in (A, B) 2008, (C, D) 2009 and (E, F) 2010. Solid-lines on the line graphs represent population sizes (mean ± SE) of the monoculture fields, whereas the dotted-lines represent those of the mixture fields of *Bt* and non-*Bt* cotton at at a row ratio of 75% to 25%, respectively.

Table 3 *F*-values from the linear mixed model estimated effect of cotton genotype, planting pattern, year and their interactions on population size of predators in northern China cotton fields in 2008, 2009 and 2010

Factor	d.f.	Adult ladybirds	Larval ladybirds	Adult lacewing	Larval lacewing	*Orius similis*	Spiders
Y	2,40	410.97***	5.35**	60.04***	56.6***	264.99***	686.19***
G	1,40	0.85	1.50	0.87	6.91**	0.07	2.00
P	1,40	0.07	0.31	3.4	0.72	0.87	0.71
D	9,459	6.96***	9.32***	11.03***	5.01***	49.88***	101.28***
Y × G	2,40	0.00	0.01	0.15	11.93***	2.62	1.16
Y × P	2,40	7.89***	3.49*	0.59	4.78**	9.27***	43.04
G × P	1,40	0.69	0.02	0.09	8.04***	0.83	3.48*
G × P × Y	2,40	1.39	0.25	0.54	3.54*	3.08	7.17**

*P < 0.05; **P < 0.01; ***P < 0.001.

G, genotype; P, planting pattern; D, sampling date; G × D, interaction between cotton genotype and sampling date; P × D, interaction between planting pattern and sampling date; G × P, interaction between cotton genotype and planting pattern. G × P × D, interaction between cotton genotype, planting pattern and sampling date.

(Liu *et al.*, 2010; Wan *et al.*, 2012). To suppress secondary pest and to delay the resistance development of target pests, Wang *et al.* (2006) proposed that non-*Bt* crops should be planted concurrently with *Bt* crops.

The present study simulated the effects of structured refuge on secondary insects and their predators through intraspecies intercropping in northern China. The mixture in the present study's field scale (small scale) significantly suppressed the abundance of cotton aphid during the seedling and squaring (budding) stages and triggered higher whitefly densities, although it did not modify the population size of mirid bugs and the predator complex. These study results partly support the conclusion that the intraspecies mixture has a potential positive effect on pest control, although the effects are inconsistent with pest species and plant developmental stages. Therefore, future studies need to examine approaches that synchronize pest management regimes, pest species and plant developmental stages with respect to developing effective pest control programmes. In addition, a larger scale study may better determine the observed phenomenon to mimic the actual production scale. This is because the size and scope of intercropping can change the composition and diversity of landscape vegetation parameters. Because landscape structure dramatically influences the abundance, diversity and function

Table 4 *F*-values of the repeated measures analysis of variance testing the effects of cotton genotype, planting pattern, sampling date and their interactions on population sizes of predator in northern China cotton fields during in 2008, 2009 and 2010

Year	Factor	d.f.	Adult ladybirds	Larval ladybirds	Adult lacewing	Larval lacewing	*Orius similis*	Spiders
2008	G	1,12	8.35*	4.09	0.67	2.97	13.01**	5.69*
	P	1,12	11.45**	9.83**	3.2	2.07	1.8	60.69***
	D	9,108	14.42***	49.11***	14.51***	10.78***	25.25***	364.99***
	G × D	9,108	2.25*	10.38***	3.86***	2.09	5.74***	3.89***
	P × D	9,108	1.94	13.99***	0.43	3.41***	1.71	7.38***
	G × P	1,12	1.09	1.25	1.17	0.03	0.04	26.81***
	G × P × D	9,108	2.98**	0.9	1.65	2.48*	1.82	3.45***
2009	G	1,12	0.04	0.73	0.45	7.79*	1.94	4.46
	P	1,12	0.07	3.4	1.02	2.2	0.05	8.25*
	D	9,108	15.09**	18.58***	5.37***	12.53***	130.64***	408.59***
	G × D	9,108	1.23	1.03	1.01	1.77	8.68***	8.89***
	P × D	9,108	1.78	1.09	0.31	3.39***	23.18***	13.55***
	G × P	1,12	0.9	0.14	5.35*	4.80*	5.22	0.38
	G × P × D	9,108	0.71	0.6	1.3	1.91	3.50***	6.91***
2010	G	1,12	0.03	0.62	0.76	0.63	0.63	0.02
	P	1,12	2.63	2.78	3.93	1.19	2.51	0.37
	D	9,108	41.17***	16.4***	12.33***	8.49***	104.73***	0.85
	G × D	9,108	1.01	0.48	0.64	0.5	1.25	1.95*
	P × D	9,108	8.3***	1.17	1.77	1.99*	8.83***	1.67
	G × P	1,12	0.23	0.09	0	2.25	0.65	0.22
	G × P × D	9,108	0.95	0.21	0.64	0.39	1.41	2.87**

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

G, genotype; P, planting pattern; D, date; G × D, interaction between cotton genotype and sampling date; P × D, interaction between planting pattern and sampling date; G × P, interaction between cotton genotype and planting pattern. G × P × D, interaction between cotton genotype, planting pattern and sampling date.

of natural enemies within croplands, optimizing the landscape structure through a reasonable arrangement of crop species or variety is crucial for developing ecologically intensive pest management approaches. Therefore, broadening the species pool of beneficial insects supported by a complex landscape and optimizing their activity should help to realize the benefits of habitat management.

Acknowledgements

We are grateful to Marvin Harris, Owen McSpadden and Stanley Carroll from the Texas A&M University for reviewing an earlier draft of the manuscript and providing constructive editorial comments. We would like to extend special thanks to Barbara Ekbom and three anonymous reviewers for their valuable insights and helpful suggestions with respect to improving an earlier version of this article. This project was supported by National Nature Science Fund of China (No. 31030012) and National Science supported planning, China (No. 2012BAD19B05).

References

Altieri, M.A. & Nicholls, C.I. (2004) *Biodiversity and Pest Management in Agroecosystems*, 2nd edn. Haworth Press, New York, New York.

Andow, D.A. (1991) Vegetational diversity and arthropod population response. *Annual Review of Entomology*, **36**, 561–586.

Barton, K.E. & Koricheva, J. (2010) The ontogeny of plant defense and herbivory: characterizing general patterns using meta-analysis. *American Naturalist*, **175**, 481–493.

Björkman, M., Hamback, P.A., Hopkins, R.J. & Rämert, B. (2010) Evaluating the enemies hypothesis in a clover-cabbage intercrop: effects of generalist and specialist natural enemies on the turnip root fly (*Delia floralis*). *Agricultural and Forest Entomology*, **12**, 123–132.

Bomford, M. (2004) *Yield, pest density, and tomato flavor effects of companion planting in garden-scale studies incorporating tomato, basil, and brussels sprout*. PhD Thesis, International Federation of Organic Agriculture Movements IFOAM, West Virginia University.

Broz, A., Broeckling, C., De-la-Pena, C. *et al.* (2010) Plant neighbor identity influences plant biochemistry and physiology related to defense. *BMC Plant Biology*, **10**, 115.

Cui, J.J. & Xia, J.Y. (1998) Effects of *Bt* transgenic cotton (with early maturity) on population dynamic of main pests and their natural enemies. *Acta Gossypii Sinica*, **10**, 255–262.

Cui, J.J. & Xia, J.Y. (2000) Effects of *Bt* (*Bacillus thuringiensis*) transgenic cotton on the dynamics of pest population and their enemies. *Acta Phytophyl Sinica*, **27**, 141–145.

Deng, S.D., Xu, J., Zhang, Q.W., Zhou, S.W. & Xu, G.J. (2003) Effect of transgenic *Bacillus thuringiensis* cotton on population dynamics of non-target pests and natural enemies. *Acta Entomologica Sinica*, **46**, 1–5.

Gardiner, M., Landis, D., Gratton, C. *et al.* (2009) Landscape diversity enhances biological control of an introduced crop pest in the north-central USA. *Ecological Applications*, **19**, 143–154.

Gould, F. (1998) Sustainability of transgenic insecticidal cultivars: integrating pest genetics and ecology. *Annual Review of Entomology*, **43**, 701–726.

Greene, J.K., Turnipseed, S.G., Sullivan, M.J. & Herzog, G.A. (1999) Boll damage by southern green stink bug (Hemiptera: Pentatomidae) and tarnished plant bug (Hemiptera: Miridae) caged on transgenic *Bacillus thuringiensis* cotton. *Journal of Economic Entomology*, **92**, 941–944.

Herron, G., Powis, K. & Rophail, J. (2000) Baseline studies and preliminary resistance survey of Australian populations of cotton aphid *Aphis gossypii* Glover (Hemiptera: Aphididae). *Australian Journal of Entomology*, **39**, 33–38.

Li, G.P., Feng, H.Q., Chen, P.Y., Wu, S.Y., Liu, B. & Qiu, F. (2010) Effects of transgenic *Bt* cotton on the population density, oviposition behavior, development, and reproduction of a nontarget pest, *Adelphocoris suturalis* (Hemiptera: Miridae). *Environmental Entomology*, **39**, 1378–1387.

Litsinger, J. & Moody, K. (1976) Integrated pest management in multiple cropping systems. *Agro-Ecosystems*, **3**, 93–118.

Liu, F., Xu, Z., Zhu, Y. *et al.* (2010) Evidence of field-evolved resistance to Cry1Ac-expressing *Bt* cotton in *Helicoverpa armigera* (Lepidoptera: Noctuidae) in northern China. *Pest Management Science*, **66**, 155–161.

Lu, Y.H., Wu, K.M., Jiang, Y.Y. *et al.* (2010) Mirid bug outbreaks in multiple crops correlated with wide-scale adoption of *Bt* cotton in China. *Science*, **328**, 1151–1154.

Lu, Y.H., Wu, K.M., Jiang, Y.Y., Guo, Y.Y. & Desneux, N. (2012) Widespread adoption of *Bt* cotton and insecticide decrease promotes biocontrol services. *Nature*, **487**, 362–365.

Men, X.Y., Ge, F., Yardim, E. & Parajulee, M.N. (2004) Evaluation of winter wheat as a potential relay crop for enhancing biological control of cotton aphid. *BioControl*, **49**, 701–714.

Parajulee, M.N. & Slosser, J.E. (1999) Evaluation of potential relay strip crops for predator enhancement in Texas cotton. *International Journal of Pest Management*, **45**, 275–286.

Parajulee, M.N., Montandon, R. & Slosser, J.E. (1997) Relay intercropping to enhance abundance of insect predators of cotton aphid (*Aphis gossypii* Glover) in Texas cotton. *International Journal of Pest Management*, **43**, 227–232.

Risch, S.J. (1981) Insect herbivore abundance in tropical monocultures and polycultures: an experimental test of two hypotheses. *Ecology*, **62**, 1325–1340.

SAS Institute (2003) *Statistical Analysis Systems Institute Inc.,* Version 9.1. SAS Institute, Cary, North Carolina.

Shrewsbury, P.M. & Raupp, M.J. (2006) Do top-down or bottom-up forces determine *Stephanitis pyrioides* abundance in urban landscapes? *Ecological Applications*, **16**, 262–272.

Sisterson, M.S., Biggs, R.W., Olson, C., Carriere, Y., Dennehy, T.J. & Tabashnik, B.E. (2004) Arthropod abundance and diversity in *Bt* and non-*Bt* cotton fields. *Environmental Entomology*, **33**, 921–929.

Sisterson, M.S., Biggs, R.W., Manhardt, N.M., Carrière, Y., Dennehy, T.J. & Tabashnik, B.E. (2007) Effects of transgenic Bt cotton on insecticide use and abundance of two generalist predators. *Entomologia Experimentalis et Applicata*, **124**, 305–311.

Tabashnik, B.E., Dennehy, T.J. & Carrère, Y. (2005) Delayed resistance to transgenic cotton in pink bollworm. *Proceedings of the National Academy of Sciences of the United States of America*, **102**, 15889–15898.

Tabashnik, B.E., Gassmann, A.J., Crowder, D.W. & Carrère, Y. (2008) Insect resistance to *Bt* crops: evidence versus theory. *Nature Biotechnology*, **26**, 199–202.

Tabashnik, B.E., Van Rensburg, J.B.J. & Carrère, Y. (2009) Field-evolved insect resistance to *Bt* crops: definition, theory, and data. *Journal of Economic Entomology*, **102**, 2011–2025.

Takizawa, T. & Snyder, W.E. (2011) Predator biodiversity increases the survivorship of juvenil predators. *Oecologia*, **166**, 723–730.

Vacher, C., Bourguet, D., Rousset, F., Chevillon, C. & Hochberg, M.E. (2003) Modelling the spatial configuration of refuges for a sustainable control of pests: a case study of *Bt* cotton. *Journal of Evolutionary Biology*, **16**, 378–387.

Vandermeer, J.H. (1992) *The Ecology of Intercropping*. Cambridge University Press, U.K.

Wan, P., Huang, Y.X., Wu, H.H., Huang, M.S., Cong, S.B., Tabashnik, B.E. & Wu, K.M. (2012) Increased frequency of pink bollworm resistance to *Bt* toxin *Cry1Ac* in China. *PLoS One*, **7**, e29975.

Wang, S.H., Just, D.R. & Pinstrup-Andersen, P. (2006) Tarnishing silver bullets: *Bt* technology adoption, bounded rationality and the outbreak of secondary pest infestations in China. Agricultural and Applied Economics Association Annual Meeting, 22–26 July 2006. Long Beach, California.

Wilson, F.D., Flint, H.M., Deaton, W.R. *et al.* (1992) Resistance of cotton lines containing a *Bacillus thuringiensis* toxin to pink bollworm (Lepidoptera: Gelechiidae) and other insects. *Journal of Economic Entomology*, **85**, 1516–1521.

Wu, K.M. & Guo, Y.Y. (2003) Influences of *Bacillus thuringiensis* Berliner cotton planting on population dynamics of the cotton aphid, *Aphis gossypii* Glover, in northern China. *Environmental Entomology*, **32**, 312–318.

Wu, K.M. & Guo, Y.Y. (2005) The evolution of cotton pest management practices in China. *Annual Review of Entomology*, **50**, 31–52.

Wu, K.M., Li, W., Feng, H.Q. & Guo, Y.Y. (2002) Seasonal abundance of the mirids, *Lygus lucorum* and *Adelphocoris* spp.(Hemiptera: Miridae) on *Bt* cotton in northern China. *Crop Protection*, **21**, 997–1002.

Wu, K., Lu, Y., Feng, H., Jiang, Y. & Zhao, J. (2008) Suppression of cotton bollworm in multiple crops in China in areas with *Bt* toxin-containing cotton. *Science*, **321**, 1676–1678.

Xue, K., Deng, S., Wang, R.J., Yan, F.M. & Xu, C.R. (2008) Leaf surface factors of transgenic *Bt* cotton associated with the feeding behaviors of cotton aphid: a case study on non-target effects. *Science in China, Series C: Life Sciences*, **51**, 145–156.

Yang, B., Ge, F., Ouyang, F. & Parajulee, M.N. (2012) Intra-species mixture alters pest and disease severity in cotton. *Environmental Entomology*, **41**, 1029–1036.

Zhang, G.F., Wan, F.H., Lövei, G.L., Liu, W.X. & Guo, J.Y. (2006) Transmission of *Bt* toxin to the predator *Propylaea japonica* (Coleoptera: Coccinellidae) through its aphid prey feeding on transgenic *Bt* cotton. *Environmental Entomology*, **35**, 143–150.

Accepted 25 August 2013
First published online 22 October 2013

Effect of Selected Insecticides on *Lygus hesperus* (Heteroptera: Miridae) Oviposition Behavior in Cotton

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Environ. Entomol. 43(1): 83–90 (2014); DOI: http://dx.doi.org/10.1603/EN12181

ABSTRACT Oviposition behavior of western tarnished plant bug, *Lygus hesperus* Knight, as affected by residual insecticides, was studied in potted as well as field-grown cotton. In this study, we investigated the spatial distribution and phytotomical preference of *Lygus* oviposition in presquaring and blooming cotton, as affected by selected insecticides. Flonicamid, acephate, and cypermethrin were applied to cotton at 82, 516, and 114 g (active ingredient)/ha, respectively. At 3 d posttreatment, a gravid female *Lygus* was caged on the plants. After 4 d, caged plants were harvested and eggs were counted by whole plant dissection. Results indicated significantly greater egg deposition in untreated controls than treated plants in both potted and field-grown cotton. For untreated plants, *Lygus* preferred to oviposit on the pulvinus and leaf petiole, where 76 and 85% of eggs were laid in potted and field-grown cotton, respectively. For insecticide-treated plants, no plant structure preference was observed, although fewer eggs were laid. The upper stratum of the plant canopy had significantly more eggs than the lower or middle strata of untreated plants, while more eggs were observed in the middle strata of plants treated with acephate in field-grown cotton. Sublethal effects could not be adequately assessed in the cypermethrin treatment owing to high *Lygus* mortality.

KEY WORDS oviposition, spatial distribution, phytotomy, insecticide, plant structure

Miridae, the largest family in suborder Heteroptera, exhibits tremendous morphological diversity and trophic plasticity and consists of numerous key herbivores as well as predators (Wheeler 2001). *Lygus* spp. are among the most economically important mirid pests, feeding on a wide range of important crops such as cotton, alfalfa, strawberry, lentil, safflower, and other fruit, vegetable, and fiber crops. *Lygus hesperus* Knight, the western tarnished plant bug, is of major importance in western cropping systems, including cotton, especially in California (Gutierrez et al. 1977), Arizona (Ellsworth 2000, Blackmer et al. 2004), and in the Texas High Plains (Parajulee et al. 2008). *Lygus* movement into cotton is affected by proximity and proportions of adjacent and nearby hosts, weed host densities, harvest and senescence timing, and environmental factors such as rainfall (Sevacherian and Stern 1975, Stern 1976, Parajulee et al. 2008). Alfalfa is one of the most important hosts for *L. hesperus* population growth in the Texas High Plains during cotton squaring and flowering stages (Parajulee et al. 2008).

Fleischer and Gaylor (1988) studied *Lygus lineolaris* (Palisot de Beauvois) dispersal, and reported that some portion of a population migrating to cotton will feed and reproduce before they emigrate. Insect feeding and oviposition are two important behaviors affecting crop loss and influencing the types of control measures adopted. Relative to insects in many other orders, members of Hemiptera have peculiar feeding and oviposition preferences. In cotton, while both *Lygus* nymphs and adults cause plant injury (Mauney and Henneberry 1979, Leigh et al. 1988), *Lygus* adults prefer to feed on vegetative structures, whereas nymphs prefer reproductive structures (Snodgrass 1998).

Feeding and oviposition behaviors are closely related in mirids as with many other arthropods. Ferran et al. (1996) reported, in *Macrolophus melanotoma* (Costa), that oviposition behavior is signaled by rostral probing of the plant surface as a means of locating, recognizing, and marking the oviposition site, after which the ovipositor is thrust into the plant tissue for egg deposition. In related studies of *Lygus rugulipennis* Poppius, Romani et al. (2005) observed such probing behavior by females before oviposition.

Mirids exhibit endophytic egg laying (embedding or insertion of eggs within plant tissues), an adaptation which reduces egg desiccation, encourages egg survival in adverse conditions, and protects them from natural enemies (Wheeler 2001). *Lygus* oviposition is considered to be partially endophytic, such that only the egg operculum is exposed when deposited in the plant tissue epidermal cell layer. Preference for oviposition in particular plant species or plant structures depends on many factors, one of which is the ease with

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which the ovipositor can penetrate substrate plant tissue. Alvarado–Rodriquez et al. (1986) reported that *L. hesperus* and *L. lineolaris* prefer laying eggs in tender plant tissues, whereas Graham and Jackson (1982) reported that *Lygus* phytotomical oviposition preference is mostly influenced by host maturity. Previous studies have shown that *L. hesperus* egg distribution varies with plant characteristics such as trichomes (Benedict et al. 1983), variety, and also plant height (Tingey and Leigh 1974).

Lygus populations can develop in high numbers in alfalfa and, on harvesting, move into cotton (Sevacherian and Stern 1974, Graham et al. 1986). This phenomenon can prove detrimental to cotton if it occurs during the critical cotton stages of squaring and boll formation. In such a situation, insecticidal application may become necessary as a means of population control (Ellsworth and Barkley 2001). Two of the most popular recommended insecticides with action against *L. hesperus* are acephate (Ellsworth and Barkley 2001, Snodgrass et al. 2009) and flonicamid (Barkley and Ellsworth 2004). Many growers also prefer using pyrethroids against *Lygus* because of their longer residual activity than acephate (Grafton–Cardwell et al. 2000). After insecticidal applications, residual lethal effects decline temporally, evincing sublethal or nonlethal effects such as feeding or oviposition repellency. Desneux et al. (2007) suggested that pesticides disrupt the coordination between insect nervous and hormonal systems, resulting in breakdowns of behavioral and physiological events related to oviposition. An extensive review of available literature reveals little regarding the influence of sublethal insecticide exposure on *Lygus* oviposition. The paucity of information on the influence of sublethal insecticide exposure on *Lygus* oviposition necessitates experimentation as a means of elucidation, and such experimentation could readily and usefully examine oviposition phytotomical distribution.

The purpose of this study was to determine *L. hesperus* oviposition behavior in cotton as affected by sublethal insecticide exposure in greenhouse- and field-grown cotton. This study characterizes *Lygus* phytotomical oviposition distribution in cotton under various insecticide treatments.

Materials and Methods

Insects. *L. hesperus* adults were obtained from a laboratory colony maintained by the Cotton Entomology Program, Texas A&M AgriLife Research, Lubbock, TX. The environmental growth chamber (Precision Scientific Low Temperature Illuminated model 818 Incubator, Winchester, VA) during rearing was operated at 27 ± 1°C and ≈40% relative humidity (RH) with a photoperiod of 14:10 (L:D) h. Insects were reared on locally obtained fresh green beans until the second instar, at which point they were reassigned to artificial diet (Debolt 1982). More than 80% of colony insects reached adulthood in 16–17 d after emergence from eggs. The test subjects were 10- to 12-d-old adult females. Strong (1970) reported a preoviposition period in *L. hesperus* of 9 d and a subsequent female egg-laying period lasting approximately 3 wk. Female gravidity was verified before testing: 150 *Lygus* adult females were randomly selected from the main colony and confined individually in glass vials, each containing a ≈1-cm green bean segment as an oviposition and food substrate. Substrates were observed for evidence of oviposition to allow for selection of only gravid females for testing.

Insecticide Residue Tests in Cotton. Cotton cultivar 'DP 141 B2RF' (Monsanto Co., St. Louis, MO) was used for this study. Insecticide residual tests on presquaring greenhouse-grown cotton were conducted in December 2008. In this study, the presquaring cotton stage was defined as cotton plant development to three to five true leaves. In 2009, residual tests were conducted with mid-May planted, full-grown flowering cotton in both greenhouse (potted plants) and field settings. The greenhouse plants were grown in 1.9-liter pots in 2008 (presquaring cotton) and 3.8-liter pots in 2009 (full-grown and flowering cotton) with potting soil mix as a growth medium.

Insecticides used in the study included cypermethrin (Ammo 2.5 EC) at 114 g (active ingredient [a.i.])/ha (FMC Corp., Philadelphia, PA), flonicamid (Carbine 50 WG) at 82 g (a.i.)/ha (FMC Corp.), and acephate (Orthene 97s) at 516 g (a.i.)/ha (Amvac Chemical Corporation, Los Angeles, CA). The insecticides were applied using a pressurized-CO_2 handboom sprayer calibrated to deliver 95 liters/ha through TeeJet TX-6 hollow cone nozzles (two per row) at 276 kPa pressure. The control plants were sprayed with water. At the time of application, air movement was negligible, minimizing drift and ensuring uniform spray deposition on the leaf surface.

Preflower test plants (2008 study) were sprayed with insecticides on 22 December. Once the sprayed product dried on the test plants, cotton plants were transferred to the laboratory for 72 h at 22°C, after which the plants were caged (two plants per cage) using paper cylindrical cartons (30 cm in height by 7.5 cm in diameter) enclosed with sheer stretchable nylon hosiery cloth. Two gravid adult *Lygus* females (10–12 d old) were transferred via aspirator into cages (one female per plant). The total number of experimental units per treatment was five ($n = 5$ cages; two plants per cage). The bugs were confined in these cages for 5 d, and the caged plants were retained indoors at ≈ 24°C. Plants were watered on day 3. On day 5, the cages were opened for observation and insect mortality was recorded. Surviving insects were removed via aspirator. Whole plants were collected for dissection and egg counting. Eggs were counted with the aid of a stereo microscope (AO model 570 Stereo Microscope, AO Instrument Company, Buffalo, NY) at 10–20× magnification. The number of eggs from each plant was recorded for separate plant structures, including the pulvinus, distal leaf petiole (DP), basal leaf petiole (BP), squares, flowers, bolls, leaf scars, terminal, main stem (MS), and leaf blade (LB) at each node throughout the height of the plant.

Table 1. Average number (±SEM) of eggs laid (n = 10 females) in plant structures by *L. hesperus* under the influence of sublethal insecticide residues in greenhouse-grown presquaring cotton, 2008

Treatment	Average no. eggs per structure per female				Total no. eggs per female
	Leaf blade	Leaf petiole + pulvinus	Upper stem	Terminal	
Untreated	0.1 ± 0.1a	12.3 ± 1.6a	4.6 ± 1.2a	0.0 ± 0.0a	17.0 ± 2.4a
Flonicamid	0.5 ± 0.5a	7.3 ± 1.7b	2.9 ± 0.7ab	0.0 ± 0.0a	10.7 ± 1.6b
Acephate	1.5 ± 1.4a	6.0 ± 1.5b	1.6 ± 0.5bc	0.1 ± 0.1a	9.2 ± 1.4b
Cypermethrin	0.0 ± 0.0a	0.6 ± 0.4c	0.1 ± 0.1c	0.0 ± 0.0a	0.7 ± 0.4c

Means within a column followed by the same letter are not significantly different (LSD test, $P \geq 0.05$).

The residual tests on flowering cotton (2009 study) were conducted with modifications to cage design to accommodate larger plants and also to facilitate ease of movement of the confined bugs. Insecticides were applied at the rates previously described to cotton grown in field plots (4.4 by 10 m) and to cotton grown in 3.8-liter pots on 22 July 2009. At 72 h after insecticide treatment, 14 plants each from the field plots and pots were caged (n = 14 cages, 1 plant per cage). The cages consisted of a hand-sewn tube of sheer organdy cloth (75 by 60 cm) cinched terminally and basally using twist ties to enclose the plant canopy. One gravid female *Lygus* was introduced into each cage at 72 h. After confining the insects for 4 d, mortality was recorded, insects were removed, and whole plants were collected for dissection and counting of eggs deposited into the plant tissues. Only plants with survivors were used for dissection and assessment of eggs.

The harvested plants (n = 14 from field, n = 10 from pots) were labeled and stored in a large walk-in cooler (≈4–5°C) until processing to prevent egg hatching and to maintain plant freshness and integrity. However, plant freshness of the field-grown cotton was compromised on five to six plants per treatment, so the actual sample size for this data set was eight to nine plants. Per-female oviposition density was quantified for separate plant structures as described previously. Four potted plants from each treatment were maintained in the laboratory for eight additional days to observe egg hatching and nymphal emergence.

Data Analysis. The oviposition data on the presquaring cotton were analyzed for differences among treatments in terms of total number of eggs laid per plant (or per female) and also in terms of the various plant structures. For the flowering cotton, oviposition data were tabulated based on the number of eggs per plant structure and plant nodal distribution. Based on the number of plant nodes, total number of eggs per plant was divided into upper, middle, and lower strata. The statistical design was a split-plot design with 8–9 replications for the field-grown cotton and 10 for potted cotton. Differences between the numbers of eggs laid in each plant structure and strata within and among treatments were determined with analysis of variance (ANOVA) using PROC GLM (SAS Institute 2003). Means were separated using F-protected least significant difference (LSD; $P \leq 0.05$).

Results

Effect of Insecticide Residues on Oviposition. *Lygus* oviposition was negatively affected by the presence of insecticide residues, irrespective of plant age. In the presquaring cotton, untreated plants had the most eggs, with an average of 17.0 eggs per female, while cypermethrin-treated plants exhibited the fewest, at 0.7 eggs per female (F = 22.2; df = 3, 19; $P < 0.0001$; Table 1). *Lygus* mortality rates were high for the cypermethrin-treated (40%) and acephate-treated (50%) plants, whereas *Lygus* on the untreated and flonicamid-treated plants suffered 10 and 20% mortalities, respectively.

In flowering cotton, the average number of *L. hesperus* eggs per female on potted cotton was greater than on the field-grown plants, except for acephate and cypermethrin treatments. Untreated control plants had a significantly greater number of eggs per female than other treatments, with 21.8 ± 1.2 and 16.2 ± 2.4 (mean ± SE) for the potted and field-grown plants, respectively (Fig. 1). In the potted cotton, significantly more eggs were laid per female in the flonicamid treatment than in the acephate-treated plants (F = 35.1; df = 3, 81; $P < 0.0001$; Fig. 1). Similarly, acephate-treated plants had significantly higher densities of eggs than cypermethrin-treated plants, where <1 egg per plant was found. In the field-grown cotton (F = 7.6; df = 3, 62; $P < 0.0002$), the number of eggs for flonicamid (11.2 ± 1.3) and acephate (9.0 ± 1.3) were not significantly different, but both were significantly higher than egg counts observed for the cypermethrin-treated plants (4.8 ± 1.2). High mortality was observed in potted cotton plants treated with cypermethrin (92%) and acephate (92%) at 4 d, whereas mortality in field-grown plants was <10% across all treatments, except cypermethrin-treated plants, where 43% mortality was recorded.

Percent nymphal emergence was calculated only for potted plants from four experimental units. As a result of high test subject mortality in cypermethrin and acephate treatments, as well as difficulty in retrieving first instars, percent nymphal emergence is not presented. However, plant dissection revealed that untreated control plants and flonicamid-treated plants had total nymphal emergences of ≈61 and ≈42%, respectively, at 8 d after gravid female removal.

Effect of Insecticide Residues on Plant Strata. In flowering cotton, the analysis of egg counts as influenced by plant strata, insecticide treatments, and the

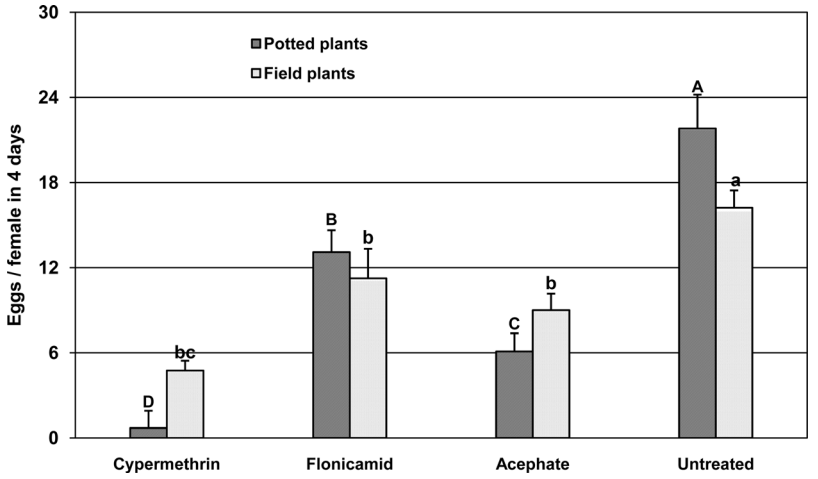

Fig. 1. Average number (±SEM) of eggs laid by *L. hesperus* on potted and field-grown flowering cottons treated with selected insecticides (n = 8–9 field-grown plants; n = 10 potted plants). Scale bars capped by the same letter within test (potted/field-grown cotton) are not significantly different (LSD; $P \geq 0.05$).

interaction of plant strata × insecticide treatments indicated significant differences: plant strata ($F = 31.8$; df = 2, 81; $P < 0.0001$), insecticide treatments ($F = 35.1$; df = 3, 81; $P < 0.0001$), and their interaction ($F = 13.1$; df = 6, 81; $P < 0.0001$).

In field-grown cotton, the numbers of eggs observed in the upper and middle strata of acephate-treated plants were statistically similar but significantly exceeded than observed in the lower stratum. In the untreated and flonicamid-treated plants, significantly more eggs were observed in the upper stratum than lower stratum (Fig. 2A). Oviposition was drastically reduced in cypermethrin-treated plants, with no ovipositional preference across plant strata (Fig. 2A). More *Lygus* eggs were found in the upper stratum of untreated plants than in any of the insecticide treatments (Fig. 2B). Although the upper stratum of cypermethrin-treated plants had the fewest eggs, the actual number did not significantly differ from acephate-treated plants. No significant differences were observed in middle and lower strata among insecticide-treated plants.

In potted cotton, although significantly more eggs were found in the middle stratum of flonicamid-treated plants than the lower stratum, neither of these differed significantly from the intermediate value observed in the upper stratum (Fig. 3A). Furthermore, significantly more eggs were found in the upper stratum of the acephate-treated plants compared with that in other strata. Untreated plants had significantly more eggs on upper stratum, followed by middle stratum, and the fewest number of eggs observed in the lower stratum (Fig. 3A). Furthermore, significant differences in the numbers of *Lygus* eggs were detected among treatments (Fig. 3B) within the upper ($F = 26.9$; df = 3, 36; $P < 0.0001$) and middle strata ($F = 11.8$; df = 3, 36; $P < 0.0001$) but not within the lower stratum ($F = 1.9$; df = 3, 36; $P = 0.14$). Within the upper stratum, the untreated plants had significantly greater numbers of eggs than the other treatments. Egg counts in flonicamid- and acephate-treated plants were statistically similar, but both had significantly more eggs than cypermethrin-treated plants.

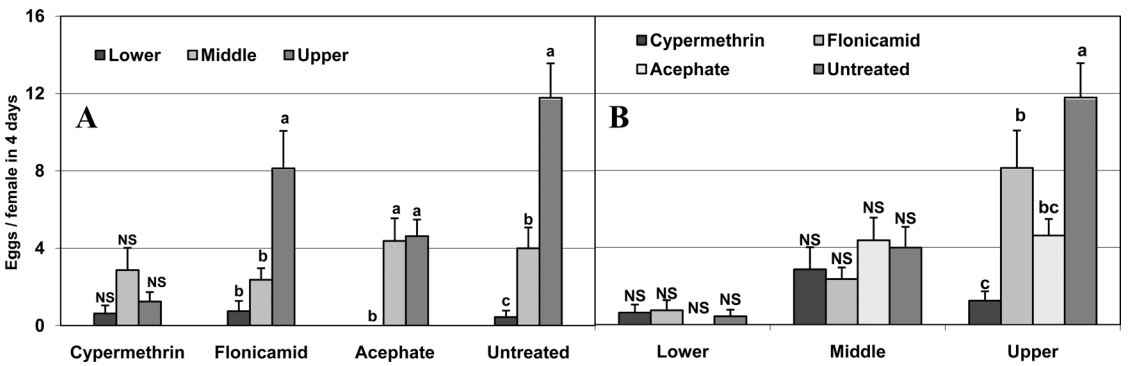

Fig. 2. Average number (±SEM) of eggs laid by *L. hesperus* on field-grown flowering cotton plants treated with selected insecticides at the lower, middle, and upper strata (n = 8–9). Scale bars capped by the same letter within each insecticide treatment (A) or plant stratum (B) are not significantly different (NS, not significant; LSD; $P \geq 0.05$).

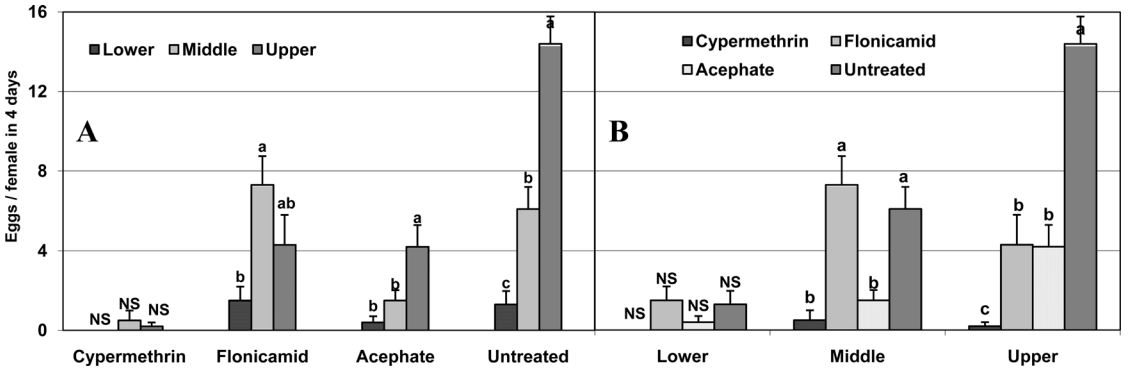

Fig. 3. Average number (±SEM) of eggs laid by *L. hesperus* on potted flowering cotton plants treated with selected insecticides at the lower, middle, and upper strata ($n = 10$). Scale bars capped by the same letter within each insecticide treatment (A) or plant stratum (B) are not significantly different (NS, not significant; LSD; $P \geq 0.05$).

Effect of Insecticides on Egg Placement in Plant Structures: Potted Cotton. In presquaring (three to five true leaf stage) potted cotton, *Lygus* preferred laying eggs on the leaf petiole and the pulvinus, where >70% of the eggs were found (Table 1). No eggs were found on the lower main stem, and few eggs were found on the terminal. In flowering potted cotton, there were significant differences in oviposition distribution among plant structures in all treatments except cypermethrin (Table 2). In the untreated control and flonicamid treatment, the pulvinus and distal leaf petiole had the highest oviposition compared with that in other plant structures. In addition, in the potted untreated controls, significantly more eggs were laid in the terminals than in the other remaining structures, with the exception of squares, in which egg-laying was similar. In acephate-treated plants, the distal leaf petiole had the most eggs, followed by the pulvinus. Egg-laying in plant terminals was observed only in untreated plants.

Effect of Insecticides on Egg Placement: Field-Grown Cotton. In the field plants, there were significant differences in oviposition among plant structures in all treatments (Table 2). In the untreated control and acephate treatments, pulvini and distal leaf petioles had greater numbers of eggs compared with that in other plant structures. In flonicamid-treated plants, oviposition was significantly greater in pulvini than distal leaf petioles and all other structures. In field-grown cotton, no eggs were found in leaf scars or leaf blades.

Discussion

In this study, >70% of *L. hesperus* eggs were found on leaves, mainly in the pulvini and distal leaf petioles of plant upper canopies. Similar results were found in studies on *L. hesperus* by Benedict et al. (1981), whose report suggested that 69–97% of all eggs laid in cotton were on the leaves—mostly leaf petioles. Jackson (2003) reported ≈82% egg-laying on leaf petioles. Similarly, Bariola (1969) reported, for *L. lineolaris*, that the highest numbers of eggs were laid in leaf petioles. By contrast, Fleischer and Gaylor (1988) reported that significantly more eggs were laid by *L. lineolaris* in cotton squares and terminals than on leaves. In the current study, it was determined that although the total number of eggs laid was reduced in the insecticide-treated plants, female ovipositional

Table 2. Average number (±SEM) of eggs laid by *L. hesperus* on various plant structures of insecticide-treated potted and field-grown cotton, 2009

Treatments	n	Average no. eggs (±SEM) per plant structure[a]									Average no. eggs per female[b]
		Leaf blade	Pulvinus	Distal leaf petiole	Basal leaf petiole	Squares	Flowers	Main stem	Terminal	Leaf scars	
Potted cotton											
Untreated	10	0.5 ± 0.2c	7.2 ± 1.1a	7.9 ± 1.3a	0.8 ± 0.3c	1.5 ± 0.5bc	0.0 ± 0.0c	0.5 ± 0.3c	2.8 ± 0.8b	0.6 ± 0.5c	21.8 ± 1.2A
Flonicamid	10	0.2 ± 0.1b	4.4 ± 0.8a	4.8 ± 1.5a	0.5 ± 0.2b	1.4 ± 0.7b	0.0 ± 0.0b	0.1 ± 0.1b	0.0 ± 0.0b	1.7 ± 0.5b	13.1 ± 2.1B
Acephate	10	0.0 ± 0.0c	2.1 ± 0.6b	3.3 ± 0.7a	0.3 ± 0.2c	0.0 ± 0.0c	0.0 ± 0.0c	0.2 ± 0.2c	0.0 ± 0.0c	0.2 ± 0.1c	6.1 ± 1.2C
Cypermethrin	10	0.0 ± 0.0ns	0.2 ± 0.2ns	0.5 ± 0.5ns	0.0 ± 0.0ns	0.0 ± 0.0ns	0.0 ± 0.0ns	0.0 ± 0.0ns	0.0 ± 0.0ns	0.0 ± 0.0ns	0.7 ± 0.7D
Field-grown cotton											
Untreated	9	0.0 ± 0.0b	6.7 ± 0.7a	6.2 ± 0.9a	0.9 ± 0.5b	0.7 ± 0.3b	0.2 ± 0.2b	0.8 ± 0.4b	0.8 ± 0.4b	0.0 ± 0.0b	16.2 ± 2.4A
Flonicamid	8	0.0 ± 0.0c	6.0 ± 0.7a	2.9 ± 1.0b	0.9 ± 0.3c	0.4 ± 0.2c	0.0 ± 0.0c	0.9 ± 0.3c	0.2 ± 0.2c	0.0 ± 0.0c	11.2 ± 1.5B
Acephate	8	0.0 ± 0.0b	2.9 ± 0.5a	3.9 ± 0.7a	0.9 ± 0.6b	0.5 ± 0.3b	0.0 ± 0.0b	0.6 ± 0.3b	0.3 ± 0.2b	0.0 ± 0.0b	9.0 ± 1.3BC
Cypermethrin	8	0.0 ± 0.0c	1.3 ± 0.5b	3.3 ± 1.0a	0.3 ± 0.3bc	0.0 ± 0.0c	0.0 ± 0.0c	0.0 ± 0.0c	0.0 ± 0.0c	0.0 ± 0.0c	4.8 ± 1.2C

[a] Means within a row among plant structures followed by the same lowercase letter are not significantly different (LSD test, $P \geq 0.05$); ns, not significant.

[b] Means in this column followed by the same uppercase letter are not significantly different (LSD test, $P \geq 0.05$).

preference for particular plant structures (leaf petiole and pulvinus) were unaffected.

Based on these results, in untreated plants, *L. hesperus* prefer to oviposit in the upper stratum. Other studies reported similar findings for *L. hesperus* (Benedict et al. 1981, Jackson 2003). It is plausible that the choice of oviposition substratum by *Lygus* depends primarily on tissue hardness rather than plant compounds such as semiochemicals. Constant et al. (1996) reported that tissue hardness plays a significant role in oviposition substrate selection by the mirid, *Macrolophus caliginosus* Wagner. The upper cotton canopy comprises primarily new tissues and is thus generally more tender and succulent. Alvarado–Rodriquez et al. (1986) reported that *L. hesperus* and *L. lineolaris* prefer to oviposit on tender plant tissues, and Graham and Jackson (1982) reported that plant structure preference by *Lygus* is influenced by host maturity.

In a penetrometer study on potted cotton, there were significant differences in hardness, specifically, wherein upper and middle leaves were significantly easier to penetrate than lower canopy leaves for distal petiole, basal leaf petiole, and main stem, whereas pulvinus and leaf blade penetration forces did not differ across plant strata (M.N.P., unpublished data). This observation makes it difficult to conclude that *Lygus* oviposition preference between canopy strata depends on tissue hardness alone, as the pulvinus, which contained the most observed eggs, did not differ between strata in terms of hardness. Nutritional and biochemical factors may influence oviposition preference as well, and require further investigation.

Oviposition preference in different strata among the insecticide treatments differed in field-grown and potted cotton. In response to cypermethrin and acephate treatments on field-grown cotton, the densities of eggs observed in middle strata approximately equaled or exceeded the densities of eggs in the upper strata, indicating that *Lygus* avoided the upper stratum. It appears that the insect moves to the middle canopy stratum for egg-laying in response to insecticide spray distribution, or more specifically, in response to greater upper canopy insecticide deposition, a phenomenon that is especially pronounced in fields with dense canopies. However, this phenomenon was not observed in flonicamid-treated field-grown plants. The reason for this is unclear, but it may be that rather than flonicamid being acutely toxic, it acts as an antifeedant. However, in potted cotton, with a lower canopy density and superior spray coverage, the eggs laid on middle and upper strata on flonicamid-treated plants did not differ.

The total number of eggs found in plants varied with each treatment and test. In the untreated control, 3.4, 5.4, and 4.1 eggs per female per day were observed in presquaring, potted, and field-grown cotton, respectively (estimated from Tables 1 and 2). These oviposition values are in general agreement with those reported by Benedict et al. (1983) where 2.0 and 2.9 eggs per day were recorded in smooth- and pilose-leaf cotton varieties, respectively. In addition, Mueller and Stern (1973) reported, in total, 136 eggs per female in $\approx$34 d (4.1 eggs per day) in green beans at 27°C. However, in the insecticide-treated plants of presquaring cotton, oviposition was affected, and fewer eggs were laid because of the presence of insecticide residues. This has important implications, as *Lygus* can cause damage to terminals and flower buds in presquaring cotton (Cook et al. 2003). Mortality was 50 and 40% in the acephate and cypermethrin treatments, respectively, even though the pesticides were allowed to weather on the plants for 3 d after spraying. This mortality appears to have affected the outcome of the test for these treatments. Reducing potential residual insecticide weathering agents by holding plants indoors may have inadvertently increased the mortality in tested insects.

The effect of insecticide residues on flowering cotton was analyzed independently for potted plants and field-grown plants because of differences in plant growth and vigor, although the tests were conducted simultaneously. Greenhouse conditions provided better control over experimental field settings, but did not provide a realistic environment for insecticide residue studies. The average number of nodes in field-grown cotton was 10, while the number in potted cotton was 8. Broader canopies and more branches were observed in field-grown cotton while the potted cotton was spindly with relatively few branches. Similar observations were made by Wilson et al. (1984) in their study on *L. hesperus* distribution, where they reported that greenhouse experiments involving plants with simple and spindly canopies provided biased information on *Lygus* behavior. Although insects of uniform age and reproductive potential were used in this test, the mean number of eggs observed in potted cotton plants was higher than in field-grown plants in the untreated control. This might be because of observational error inherent to this kind of experiment, particularly where large dense field-grown plants present a considerable difficulty in terms of visually searching for eggs. Another possibility for varied oviposition is the fluctuating environmental conditions in the field vs. indoors, where potted plants could be maintained under relatively constant controlled conditions. It is also expected that the insecticide efficacy in the field setting would be lower if the insecticides were applied by a ground rig or an aerial applicator compared with a hand-boom sprayer as used in the current study.

The treatment effects were highly significant for both potted and field-grown cotton. In the potted plants, cypermethrin and acephate induced >90% mortality, which was similar to results in the presquaring potted plant tests. Applying cypermethrin and acephate at "low" labeled rates and incubating plants for 72 h before insect infestation did not reduce test mortality below 30% in the potted plant study. The high mortalities for cypermethrin may have been avoided if insect releases for the tests were delayed until 14 d after initial treatment, as other studies have reported median lethal times of 14 d for cypermethrin on *L. hesperus.*

No mortality was observed in the untreated control and flonicamid treatments in both tests on flowering

cotton. Flonicamid is a novel target-specific insecticide with action against piercing-sucking pests, and is recommended for *Lygus* management (Barkley and Ellsworth 2004). Although the mode of action is unclear, flonicamid acts as a feeding inhibitor, subsequently starving the insects to death (Joost 2006, Morita et al. 2007). In the current study, flonicamid treatments did not impart high mortalities during any of the tests, but demonstrated effectiveness in oviposition suppression. Romani et al. (2005) studied the oviposition behavior in *L. rugulipennis* and reported that the oviposition site selection is primarily determined by specific stylet sensillae and the probing behavior exhibited by the female, which eventually results in oviposition site selection. As such, it is highly possible that in our study, *Lygus* exposure to flonicamid residues might have inhibited or reduced stylet sensitivity, leading to reduced feeding and subsequent oviposition. In addition, flonicamid is known to be much selective on natural enemy preservation compared with acephate and cypermethrin (Jalali et al. 2009).

In the field-grown cotton plants, mortality in all treatments was below 10% except for cypermethrin, where it was 43%. Acephate, applied at the recommended rate of 516 g (a.i.) per hectare, failed to prove lethal, but was effective in reducing oviposition.

To our knowledge, this is the first study investigating the impact of insecticide residues on *L. hesperus* oviposition behavior. Sublethal effects of insecticidal residues can influence the number of eggs laid, but were not observed to influence oviposition preference. In addition, these effects emphasize the plant structure preferences and spatial distribution of *L. hesperus* oviposition in cotton, and may be used as a tool aiding in host plant resistance studies. Sublethal insecticidal residual effects also contribute information useful for integration in *Lygus* management programs using the biocontrol agent and egg parasitoid, *Anaphes iole* Girault, where information on host (*Lygus*) egg distribution on the plant is essential. *Lygus* ovipositional preference may be affected by physical, biochemical, or evolutionary factors, and these topics need further investigation.

Acknowledgments

We thank Stanley Carroll, Ram Shrestha, and Mark Arnold for technical assistance during this research, Ram Shrestha for helping with statistical analysis, and Owen McSpadden for critical review of an earlier draft of this manuscript. U.S. Department of Agriculture–Cooperative State Research, Education, and Extension Service–Risk Avoidance and Mitigation Program (USDA–CSREES–RAMP) and Texas A&M AgriLife Cotton Entomology Program provided funding for this research.

References Cited

Alvarado–Rodriquez, B., T. F. Leigh, and K. W. Foster. 1986. Oviposition site preference of *Lygus hesperus* (Hemiptera: Miridae) on common bean in relation to bean age and genotype. J. Econ. Entomol. 79: 1069–1072.

Bariola, L. A. 1969. The biology of the tarnished plant bug, *Lygus lineolaris* (Beauvois), and its nature of damage and control on cotton, p. 102. Ph.D. dissertation, Texas A&M University, College Station, TX.

Barkley, V., and P. C. Ellsworth. 2004. Search for effective chemical controls for *Lygus* bugs and whiteflies in Arizona cotton, pp. 187–197. *In* J.C. Silvertooth (ed.), Cotton: A College of Agriculture and Life Sciences Report P-138. Publication No. AZ1335. University of Arizona, College of Agriculture and Life Sciences, Tucson, AZ.

Benedict, J. H., T. F. Leigh, J. L. Frazier, and A. H. Hyer. 1981. Ovipositional behavior of *Lygus hesperus* (Hemiptera, Miridae) on 2 cotton genotypes. Ann. Entomol. Soc. Am. 74: 392–394.

Benedict, J. H., T. F. Leigh, and A. H. Hyer. 1983. *Lygus hesperus* (Heteroptera: Miridae) oviposition behavior, growth, and survival in relation to cotton trichome density. Environ. Entomol. 12: 331–335.

Blackmer, J. L., C. Rodriguez–Saona, J. A. Byers, K. L. Shope, and J. P. Smith. 2004. Behavioral response of *Lygus hesperus* to conspecifics and headspace volatiles of alfalfa in a Y-tube olfactometer. J. Chem. Ecol. 30: 1547–1564.

Constant, B., S. Grenier, G. Febvay, and G. Bonnot. 1996. Host plant hardness in oviposition of *Macrolophus caliginosus* (Hemiptera: Miridae) J. Econ. Entomol. 89: 1446–1452.

Cook, D., B. R. Leonard, M. Willrich, J. Gore, and R. Gable. 2003. Residual toxicity of seed treatments and soil applied insecticides to tarnished plant bug, *Lygus lineolaris* (Palisot de beauvois), in cotton. J. Agric. Urban Entomol. 20: 184–192.

Debolt, J. W. 1982. Meridic diet for rearing successive generations of *Lygus hesperus.* Ann. Entomol. Soc. Am. 75: 119–122.

Desneux, N., A. Decourtye, and J. M. Delpuech. 2007. Sublethal effects of pesticides on beneficial arthropods. Annu. Rev. Entomol. 52: 81–106.

Ellsworth, P. C. 2000. *Lygus* control decision aids for Arizona cotton, pp. 269–280. *In* J. C. Silvertooth (ed.), Cotton: A College of Agriculture Report. Series P-121. Publication No. AZ1224. University of Arizona, College Agriculture and Life Science, Tucson, AZ.

Ellsworth, P. C., and V. Barkley. 2001. Cost effective management of *Lygus* in Arizona cotton, pp. 299–307. *In* J. C. Silvertooth (ed.), Cotton: A College of Agriculture and Life Sciences Report. Publication No. AZ1170. University of Arizona, College Agriculture and Life Science, Tucson, AZ.

Ferran, A., A. Rortais, J. C. Malausa, J. Gambier, and M. Lambin. 1996. Ovipositional behavior of *Macrolophus caliginosus* (Heteroptera: Miridae) on tobacco leaves. Bull. Entomol. Res. 86: 123–128.

Fleischer, S. J., and M. J. Gaylor. 1988. *Lygus lineolaris* (Heteroptera: Miridae) population dynamics, nymphal development, life tables, and Leslie matrices on selected weeds and cotton. Environ. Entomol. 17: 246–253.

Grafton–Cardwell, B., J. Christiansen, B. Striggow, G. Montez, and B. Brindley. 2000. Insecticide resistance trends in San Joaquin Valley *Lygus* bug. *In* Proceedings of the *Lygus* Summit, Visalia, CA.

Graham, H. M., and C. G. Jackson. 1982. Distribution of eggs and parasites of *Lygus* spp. (Hemiptera: Miridae), *Nabis* spp. (Hemiptera: Nabidae), and *Spissistilus festinus* (Say) (Homoptera: Membracidae) on plant stems. Ann. Entomol. Soc. Am. 75: 56–60.

Graham, H. M., C. G. Jackson, and J. W. Debolt. 1986. *Lygus* spp. (Hemiptera: Miridae) and their parasites in agricultural areas of Southern Arizona. Environ. Entomol. 15: 132–142.

Gutierrez, A. P., T. F. Leigh, Y. Wang, and R. D. Cave. 1977. An analysis of cotton production in California: *Lygus hesperus* (Heteroptera: Miridae) injury: an evaluation. Can. Entomol. 109: 1375–1386.

Jackson, C. G. 2003. Oviposition by *Lygus hesperus* and its egg parasitoid, *Anaphes iole,* in cotton, alfalfa, and a wild mustard. Southwest. Entomol. 28: 167–176.

Jalali, M. A., T. Van Leeuwen, L. Tirry, and P. De Clercq. 2009. Toxicity of selected insecticides to the two-spot ladybird, *Adalia bipunctata.* Phytoparasitica 37: 323–326.

Joost, H. 2006. Flonicamid: a novel mode of action for piercing and sucking insects, pp. 1583–1590. *In* Proceedings of the Beltwide Cotton Conference. National Cotton Council, Memphis, TN.

Leigh, T. F., T. A. Kerby, and P. F. Wynholds. 1988. Cotton square damage by the plant bug, *Lygus hesperus* (Hemiptera: Heteroptera: Miridae), and abscission rates. J. Econ. Entomol. 81: 1328–1337.

Mauney, J. R., and T. J. Henneberry. 1979. Identification of damage symptoms and patterns of feeding of plant bugs in cotton. J. Econ. Entomol. 72: 496–501.

Morita, M., T. Ueda, T. Yoneda, T. Koyanagi, and T. Haga. 2007. Flonicamid, a novel insecticide with rapid inhibitory effect on aphid feeding. Pest Manage. Sci. 63: 969–973.

Mueller, A. J., and V. M. Stern. 1973. Effects of temperature on the reproductive rate, maturation, longevity, and survival of *Lygus hesperus* and *L. elisus* (Hemiptera: Miridae). Ann. Entomol. Soc. Am. 66: 593–597.

Parajulee, M. N., R. B. Shrestha, A. K. Barman, and S. C. Carroll. 2008. Ecologically intensive pest management in cotton agroecosystems: *Lygus hesperus* as a model system. Egypt. J. Agric. Res. 86: 57–81.

Romani, R., G. Salerno, F. Frati, E. Conti, N. Isidoro, and F. Bin. 2005. Oviposition behavior in *Lygus rugulipennis:* a morpho-functional study. Entomol. Exp. Appl. 115: 17–25.

SAS Institute. 2003. Statistical Analysis Systems Institute Inc., version 9.1. SAS Institute, Cary, NC.

Sevacherian, V., and V. M. Stern. 1974. Host plant preferences of *Lygus* bugs in alfalfa-interplanted cotton fields. Environ. Entomol. 3: 761–766.

Sevacherian, V., and V. M. Stern. 1975. Movements of *Lygus* bugs between alfalfa and cotton. Environ. Entomol. 4: 163–165.

Snodgrass, G. L. 1998. Distribution of the tarnished plant bug (Heteroptera: Miridae) within cotton plants. Environ. Entomol. 27: 1089–1093.

Snodgrass, G. L., J. Gore, C. A. Abel, and R. Jackson. 2009. Acephate resistance in populations of the tarnished plant bug (Heteroptera: Miridae) from the Mississippi River Delta. J. Econ. Entomol. 102: 699–707.

Stern, V. M. 1976. Ecological studies of *Lygus* bugs in developing a pest management program for cotton pests in the San Joaquin Valley, CA, pp. 8–13. *In* D. R. Scott and L. E. O'Keeffe (eds.), *Lygus* Bug: Host Plant Interactions. Proceedings of the International Congress Entomology. University Press, Moscow, ID.

Strong, F. 1970. Physiology of injury caused by *Lygus hesperus.* J. Econ. Entomol. 63: 808–814.

Tingey, W. M., and T. F. Leigh. 1974. Height preference of *Lygus* bugs for oviposition on caged cotton plants. Environ. Entomol. 3: 350–351.

Wheeler, A. G., Jr. 2001. Biology of plant bugs: pests, predators, opportunists. Cornell University Press, Ithaca, NY, p. 507.

Wilson, L. T., T. F. Leigh, D. Gonzalez, and C. Foristiere. 1984. Distribution of *Lygus hesperus* (Knight) (Miridae: Hemiptera) on cotton. J. Econ. Entomol. 77: 1313–1319.

Received 15 June 2012; accepted 8 September 2013.

Life Table and Population Dynamics of the Cotton Aphid, *Aphis gossypii*, on Upland Cotton

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Abstract. Controlled field studies using clip cages were used to quantify development, reproduction, and survival, and generate life history characteristics and population growth parameters of the cotton aphid, *Aphis gossypii* Glover, on irrigated upland cotton, *Gossypium hirsutum* L. Newly born (<6-hour-old) aphid nymphs were individually placed on the underside of fifth mainstem cotton leaves (n = 30 plants). Aphids were monitored every 24 hours and the developmental stage, fecundity, and mortality recorded until the last aphid from the cohort died. Individual aphids were transferred to a new fifth leaf on the same plant when plants gained a newer leaf. Average daily temperature under the leaf surface was 28.1ºC. Nymphal durations were 38, 48, 37, and 34 degree-days above a development threshold of 6.3ºC for 1st, 2nd, 3rd, and 4th instars, respectively. Aphids began dying at 448 degree-days, and the last individual in the cohort died at 907 degree-days, with an average longevity of 674 degree-days. The gross reproductive rate and finite rate of increase were 62.24 and 1.43263, respectively. A complementary study was done by daily monitoring population dynamics and within-plant distribution of the aphid for two growing seasons. Cotton aphids were not typically in the field until mid-July or early August, and the population decreased after mid-September. A fifth mainstem leaf from the top of the plant canopy (T_5) and a leaf from the mid-canopy (M) consistently tracked population activity with whole-plant densities, suggesting that either T_5 or M should serve as a reliable indicator-leaf for monitoring cotton aphid population dynamics in the field.

Introduction

The cotton aphid, *Aphis gossypii* Glover (Hemiptera: Aphididae), is a common and in some years economic pest of cotton, *Gossypium hirsutum* L., in the Cotton Belt of the United States. On the Texas High Plains, the cotton aphid is an occasional or secondary pest of cotton in most years (Leser 1994), while in other years, aphids increase in number and become serious and economically significant (Leser et al. 1992). Parajulee et al. (2003) reported that although there has been no documentation, it is believed that large, widespread outbreaks of cotton aphids on the Texas High Plains occur because of aerial immigration from southern locations.

Many studies have examined the effect of environmental, cultural, and plant condition factors on abundance of cotton aphids. Development, reproduction, and survival of the cotton aphid are influenced by temperature and quality of food (Komazaki 1982, Liu and Peng 1987, Liu and Hwang 1991, Kocourek et al. 1994,

van Steenis and El-Khawass 1995, Ebert and Cartwright 1997, Slosser et al. 1998, Xia et al. 1999, Wang and Tsai 2000). The speed of physiological development and rate of reproduction of the aphid increase as the temperature increases to an upper limit (Parajulee 2007). Nevertheless, at extreme cold and hot temperatures, developmental rate and reproduction rate of the cotton aphid are less. Kersting et al. (1999) reported that the developmental rate for different stages of cotton aphid on cotton increased linearly as temperature increased within the range of 15 to 30ºC. Food quality and temperature play key roles in increase in cotton aphid abundance.

The effect of temperature on population growth of the cotton aphid on cotton was studied by Kersting et al. (1999) in Turkey and Xia et al. (1999) in China, and on other host plants such as *Cucumis sativus* L. (Kocourek et al. 1994, van Steenis and El-Khawass 1995), *Lagenaria siceraria* (Molina) (Liu and Peng 1987), *Citrus unshiu* Marcovitch (Komazaki 1982), and *Psidium guajava* L. (Liu and Hwang 1991). Parajulee (2007) quantified the temperature-dependent development, reproduction, and survival, and generated life history characteristics and population growth parameters of the cotton aphid on phenologically standardized greenhouse-grown cottons. He estimated the maximum rate of development for cotton aphids to occur at 32.2, 30.8, 30.4, 30.0, and 30.2ºC for first to fourth instars and for total nymphal development, respectively, while the optimum temperature for overall cotton aphid development, reproduction, and population increase was estimated to be 28.6ºC.

In the present study, a field cage experiment was used to generate life history characteristics and population growth parameters in cotton standardized for plant phenology and indirectly in the nutritional status of the host substrate. In addition, natural population dynamics of cotton aphids were examined to compare with population dynamics in controlled cages in the field.

Materials and Methods

A 2-year (2003-2004) study was done at the Texas A&M AgriLife Research farm at Lubbock, TX. Thirty experimental plots (4 rows x 30.5 m) with 1-m row spacing were planted with 'PM 2326RR' cotton in mid-May. Standard land and crop management procedures recommended for furrow-irrigated cotton on the Texas High Plains were used. Cotton plots were fertilized with 90-0-0 (N-P-K) kg/ha.

Field Cage Study. On 18 July 2004, 30 newly born aphid nymphs (<6 hours old) were transferred individually to the underside of fifth mainstem node cotton leaves (one aphid per fifth leaf of a cotton plant per plot). Leaves were at the fifth node from the top of the plant (Parajulee 2007). An individual nymph was confined in a clear plastic, hinged box (catalog # 203, Alpha Rho Inc., Fitchburg, MA) as a cage on a section of the fifth leaf. Spring-loaded stainless steel hair clips (L & N Sales and Marketing, Hatboro, PA) were reshaped to fit over the plastic cages and to hold cages tightly closed and securely attached to leaves. A 1.25-cm-diameter hole was drilled in the bottom of each plastic cage, and perforated muslin cloth was hot glued over the hole to provide ventilation to the cage interior while preventing escape of aphids. Test aphids were transferred to the new fifth leaf on the same plant when the plant produced a newer leaf (at approximately 5-day intervals) to standardize the leaf nutritional quality among test insects. The individual insect was transferred to the aphid cage, and the cage was fastened to the new leaf. Individually caged nymphs were observed for molting and survival every 24 hours

until the last individual from the cohort of 30 molted to the adult stage. Adult aphids were observed daily and newly born offspring counted and removed until the last adult from the cohort died.

Life history parameters of instar-specific and total developmental period (days or degree-days), age-specific survival (percentage) and fecundity (number of progeny per female), gross reproductive rate (mean number of offspring produced per female per lifetime without regard to female survival), net reproductive rate (mean number of offspring produced per female accounting for the female survival), finite rate of increase (rate of population growth), doubling time, and intrinsic rate of increase were measured. In this analysis, the first day as nymph was set as the first pivotal age and age increments were set to 1 day. The intrinsic rate of increase (r) was determined by iteratively solving the Euler equation, $\Sigma\, e^{-rx} l_x m_x = 1$, where x is the age in days (including immature stages), r is the intrinsic rate of increase, and l_x is the proportion of individuals alive at time x of an original cohort (including immature mortality). The variable m_x is the mean number of offspring produced per surviving aphid during the age interval x (1 day). The life table parameters, including gross reproductive rate ($GRR = \Sigma m_x$), net reproductive rate ($R_0 = \Sigma l_x m_x$), finite rate of increase ($\lambda = e^r$, a discrete form of the intrinsic rate of increase), and doubling time ($DT = ln2/r$) were calculated using the methods described by Andrewartha and Birch (1954). After computing the intrinsic rate of increase (r) for the original data (r_{all}), the jackknife method (Meyer et al. 1986) was used to estimate the standard error of the calculated life table statistics, including GRR, R_0, λ, GT, and DT. One-way analysis of variance and least significant difference procedure ($P < 0.05$) were used to measure variation in instar developmental periods and instar-specific survival.

To monitor the temperatures that aphids experienced, two, four-probe HOBO® weather dataloggers (model H08-006-04, Onset Computer Corp., Bourne, MA) were installed in the cotton field for the duration of the field study. The two dataloggers were installed on two separate plants, with temperature probes at four identical locations on each plant to estimate the actual temperature experienced by aphids across various strata within the plant. One of the temperature recording sites was on the undersurface of cotton leaves at the fifth node from the top of the plant. Degree-days were computed above a developmental threshold of 6.26ºC (Parajulee 2007).

Field Population Dynamics. Thirty-two homogeneous cotton plants were selected when plants began fruiting (all plants with a match-head square), and each plant was monitored daily for naturally colonized abundance of cotton aphids. Cotton aphid age structure was not quantified. Daily population dynamics of cotton aphids were monitored for 108 days (30 June to 16 October) in 2003 and 80 days (16 July to 4 October) in 2004. Test plants were monitored for cotton aphids at four specific positions within the plant canopy to examine the within-plant distribution of cotton aphids and identify an indicator leaf position for sampling aphids. Sampling positions were 1) second leaf from the top of the plant (T_2), 2) fifth leaf from the top of the plant (T_5), 3) a leaf approximately at the mid-canopy of the plant (M), and 4) second leaf from the base of the plant (B_2). In addition to the four selected potential indicator leaves on each of the 30 plants, six selected plants were monitored daily for whole-plant aphid counts for the entire monitoring period. It is to be noted that the sampling position within the plant canopy remained constant, but the actual leaf sampled changed as the season progressed and plants attained newer nodes.

Temporal patterns of cotton aphid population dynamics were evaluated by graphical representation and pairwise correlation analyses for each year.

Results and Discussion

Field Cage Study. Instar-specific nymphal duration did not significantly vary among instars. Nymphal durations, measured in terms of heat units, consisted of 38, 48, 37, and 34 degree-days above a development threshold of 6.26ºC for first, second, third, and fourth instars, respectively. Cotton aphids began to die at 448 degree-days (20 days after birth) and ended at 907 degree-days (44 days after birth) (Fig. 1).

Average longevity of individually caged cotton aphids (n = 30) in the field was 31.4 calendar days and 674 degree-days. Xia et al. (1999) reported a developmental threshold of 7.1ºC from a laboratory study and estimated aphid longevity to be approximately 275 degree-days. Aphid survival in the present study was more than two times that of the survival reported by Xia et al. (1999). Although the developmental threshold in this study was lower than that used by Xia et al. (1999), the difference was overshadowed by increased survival of aphids in this study. Kersting et al. (1999) reported a developmental threshold of 6.2ºC and average survival period of 25.2 days at 25-30ºC, >20% shorter survival period than in this study. Both previous studies were done in a laboratory and used excised leaves as rearing substrate, while the present study was done in the field and used plants *in situ*.

Results of the present study indicated that cotton aphids survived longer in their natural habitat (cotton plants in the field) than in a laboratory. One reason for

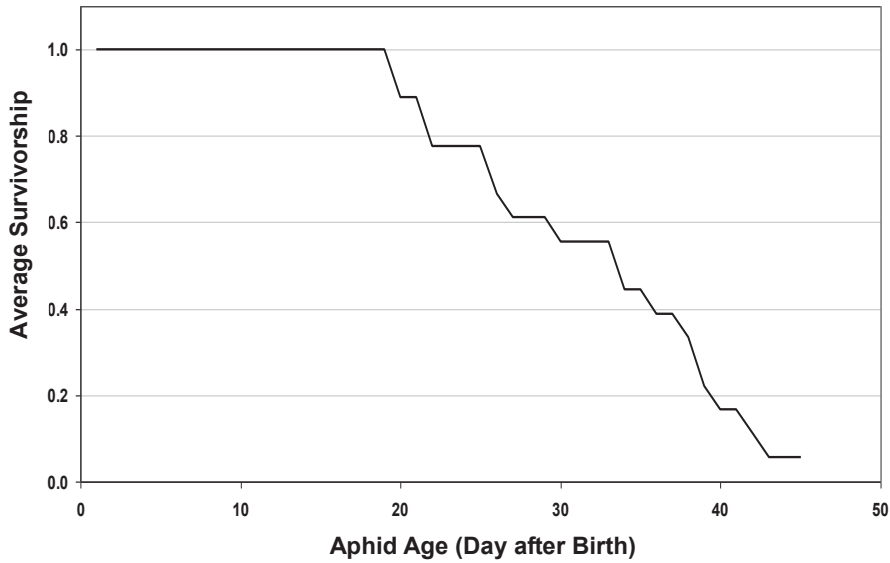

Fig. 1. Daily average survival of individually caged cotton aphids in the field (n = 30), Lubbock, TX.

the large difference between this and previous studies might be because aphids use turgor pressure of living plants to aid in intake of plant sap. Excised leaves might not provide the same amount of turgor pressure as leaves of a living plant. Further study is needed to quantify the difference in nutritional status and nutrient uptake by cotton aphids in the two scenarios.

Aphid reproduction began at 140 degree-days (6 days after birth), and the daily fecundity peaked at 241 degree-days (12 days after birth); reproduction ceased at 740 degree-days (35 days after birth) (Fig. 2). Cotton aphids produced an average of 63 offspring during a 30-day average reproductive lifespan. Because survival was 100% when 80% of reproduction had been completed (Day 19) (Figs. 1-2), the net reproductive rate was only slightly less than the gross reproductive rate. The gross reproductive rate, net reproductive rate, finite rate of increase, intrinsic rate of growth, and doubling time were 62.24, 59.06, 1.4326, 0.35971, and 1.94, respectively. Xia et al. (1999) reported gross and net reproductive rates of 28.3 and 24.4, respectively, while reared at 25ºC on excised cotton leaves in a laboratory.

Although the field temperature fluctuated during the study period in the current study, degree-days accumulated by caged aphids remained between 20 to 25 per day when 95% of reproduction was occurring (27 days), with average daily temperature of 28.1ºC (Fig. 3). Data from laboratory studies indicated cotton aphids produced the most nymphs on cotton seedlings in a laboratory at 25-27ºC (Akey and Butler 1989), a temperature range only slightly cooler than average temperatures during July-August on the Texas High Plains.

In the laboratory study (Parajulee 2007), gross and net reproductive rates and intrinsic rate of growth were 50.51, 44.75, and 0.37403 at 25ºC, and 58.93, 53.08, and 0.31218 at 30ºC, respectively. Aphid fecundity in the field study was greater than in the previously reported studies. As suggested for survival, less

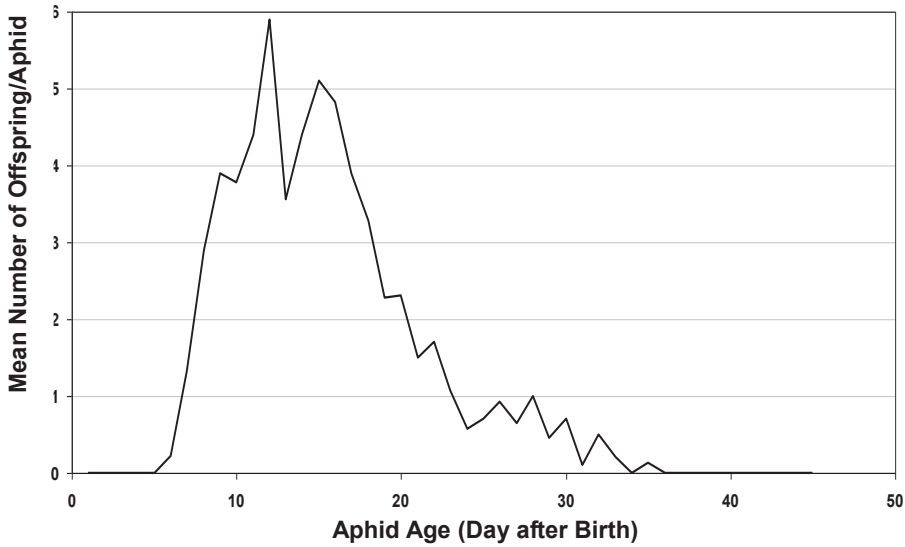

Fig. 2. Daily average fecundity (mean number of offspring per aphid) of individually caged cotton aphids (n = 30) in the field, Lubbock, TX.

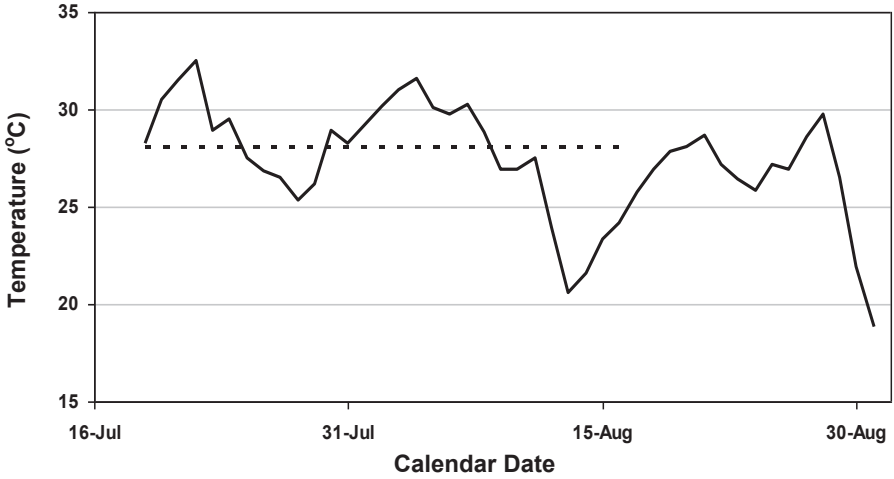

Fig. 3. Daily average temperature under the surface of cotton leaves next to caged cotton aphids in a life table study, Lubbock, TX, 2004. The dashed line indicates the average daily temperature (28.1ºC) when 95% reproduction occurred.

nutritional quality of plants or the use of excised leaves as rearing substrates in the studies may have influenced fecundity. However, in a study by Parajulee (2007), aphids were caged on living plants in a growth chamber, indicating that some other factor might also influence survival and fecundity of aphids. Effects of other factors such as solar radiation or humidity on development and survival of cotton aphids in the field should be investigated in future studies.

In summary, this field study with individual caged aphids revealed that the gross reproductive rate was numerically greater in the field (62.24) compared with that in a laboratory; greatest laboratory gross reproductive rate among the six constant temperatures was 58.93 (Parajulee 2007). The finite rate of increase (λ) of cotton aphids in the field was 1.43263, whereas the λ value for a comparable temperature in the growth chamber was 1.411435. This indicated that cotton aphids had slightly faster developmental and population growth rates in a natural habitat compared to a growth chamber. Nutrition might be a likely reason for this difference. Future research should specifically address this and other potentially influential factors such as solar radiation, daily fluctuations in field temperatures, humidity, and air condition and movement (natural vs. growth chamber).

Field Population Dynamics. Cotton aphid field population activity varied between the 2 years of the study (Figs. 4-5). In 2003, abundance of aphids began to increase in mid-July, but fluctuated around a mean of 0.2 aphid per leaf (single-leaf monitoring) or 2.5 aphids per plant (whole plant monitoring) and did not have a clear peak (Fig. 4). Nevertheless, the whole-plant observation of aphid density showed an apparent peak in late July and again in mid-August, but both of these superficial peaks resulted in characteristic cotton aphid population crashes within 1 week after the peaks.

In 2004, aphids began to increase in abundance in mid-August, approximately 1 month later than in the previous year, and showed a single peak in

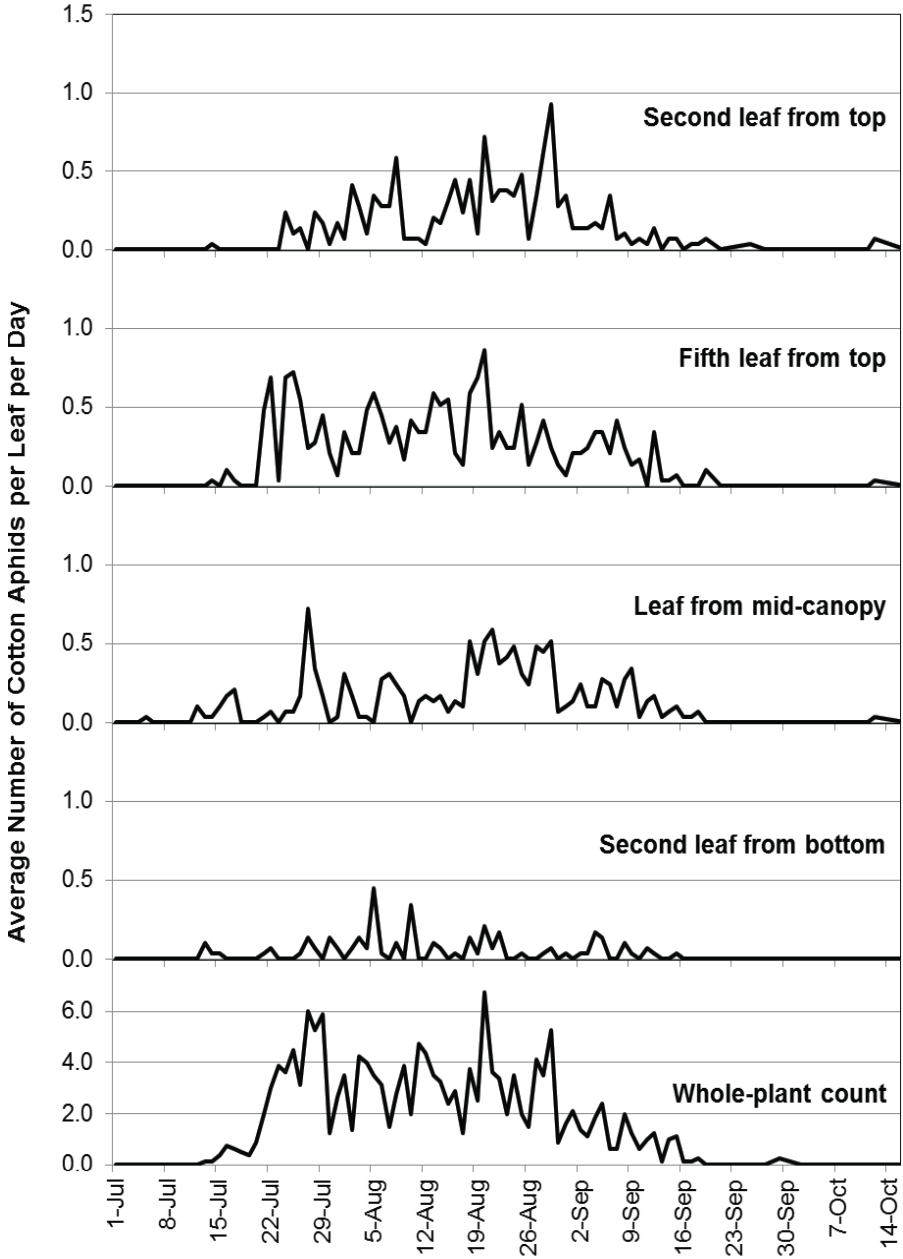

Fig. 4. Daily cotton aphid population dynamics on cotton leaves representing specific within-plant strata, 2003.

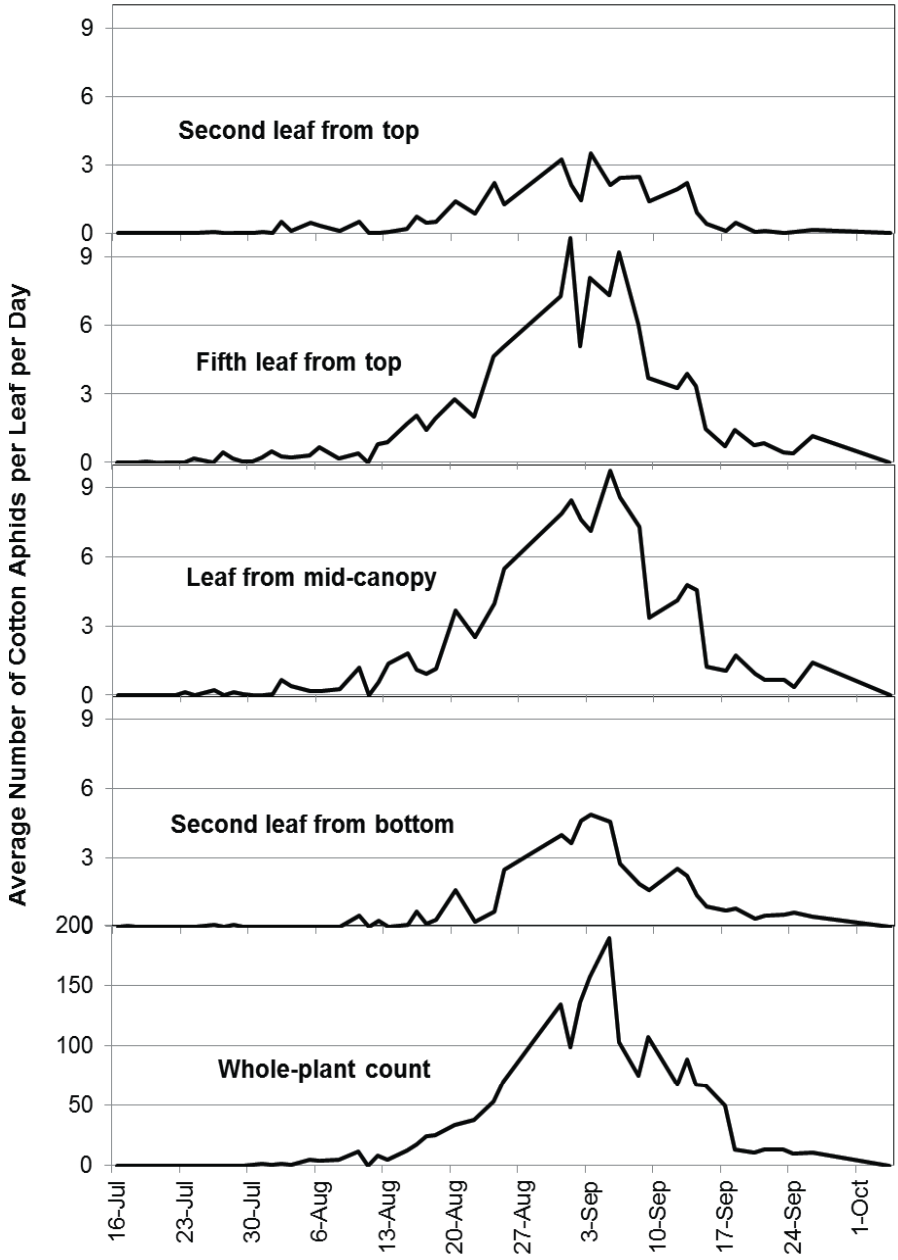

Fig. 5. Daily cotton aphid population dynamics on cotton leaves representing specific within-plant strata, 2004.

Table 1. Pairwise Correlation between Cotton Aphid Densities via Whole-Plant Counting (n = 6 Plants) and Specific Single-Leaf Counting (n = 32 Plants), Lubbock, TX, 2003-2004

	Whole-plant aphid density	
Indicator leaf within the plant canopy[a]	2003	2004
Second leaf from top of the plant (T_2)	0.33	0.86
Fifth leaf from top of the plant (T_5)	0.51	0.88
Leaf from approximately the middle of the plant (M)	0.46	0.92
Second leaf from the base of the plant (B_2)	0.21	0.94

[a]Correlation between cotton aphid counts on indicator leaf B_2 and whole-plant in 2003 was not significant.

late August/early September. In 2004, a year when cotton aphids were moderately abundant, numbers on indicator leaves T5 and M resembled aphid population dynamics patterns and peak abundance on whole plants (Fig. 5). Because aphids were scarce in 2003, abundance on potential indicator leaves was slightly correlated with numbers on whole plants (Table 1). Nevertheless, the number of aphids on indicator leaf T_5 was best correlated with that on whole plants, followed by M and T_2, with B_2 least correlated with the whole-plant estimate. In 2004, aphids were more abundant (18, 35, 51, and 92 times greater as indicated by T_2, T_5, M, and B_2 indicator leaves, respectively) than in 2003, and the aphid population activity patterns depicted by indicator leaves were similar to those observed on whole plants (Fig. 5). As a result, indicator-leaf estimates and whole-plant count were very correlated (Table 1). It is noteworthy that numbers of aphids on B_2 were not significantly related to whole-plant population dynamics in 2003, but showed numerically best correlation with whole-plant count when aphids were moderately abundant (2004). This suggested that aphid dispersion patterns on basal leaves might be more significantly influenced by aphid abundance on the plant. Parajulee (2007) and Parajulee et al. (2010) used T_5 as an indicator leaf to estimate cotton aphid population dynamics in greenhouse-grown plants and characterize cotton plant physiological parameters as influenced by various fertilizer rates. In the study, T_5 and M leaves consistently tracked population activity with whole-plant densities during years with scarce or moderate numbers of aphids, suggesting that either T_5 or M, a leaf from the mid-canopy, should serve as a reliable indicator leaf for monitoring cotton aphid population dynamics in the field.

References Cited

Akey, D. H., and G. D. Butler. 1989. Developmental rates and fecundity of apterous *Aphis gossypii* on seedlings of *Gossypium hirsutum*. Southwest. Entomol. 14: 295-299.

Andrewartha, H. G., and L. C. Birch. 1954. The distribution and abundance of animals. University of Chicago Press, Chicago, IL.

Ebert, T. A., and B. Cartwright. 1997. Biology and ecology of *Aphis gossypii* Glover (Homoptera: Aphididae). Southwest. Entomol. 22: 116-148.

Kersting, U., S. Satar, and N. Uygun. 1999. Effect of temperature on development rate and fecundity of apterous *Aphis gossypii* Glover (Hom. Aphididae) reared on *Gossypium hirsutum* L. J. Appl. Entomol. 123: 23-27.

Kocourek, F., J. Havelka, J. Berankova, and V. Jarosik. 1994. Effect of temperature on development rate and increasing rate of increase of *Aphis gossypii* reared on greenhouse cucumbers. Entomol. Exp. Appl. 71: 59-64.

Komazaki, S. 1982. Effects of constant temperatures on population growth of three aphid species, *Toxoptera citricidus* (Kirkaldy), *Aphis citricola* van der Goot and *Aphis gossypii* Glover (Homoptera: Aphididae) on citrus. Appl. Entomol. Zool. 17: 75-81.

Leser, J. F. 1994. Management of cotton aphids: Texas style, pp. 137-141. *In* Proceedings, Beltwide Cotton Conferences, Nat. Cotton Council of Amer., Memphis, TN.

Leser, J. F., C. T. Allen, and T. W. Fuchs. 1992. Cotton aphid infestations in west Texas: a growing management problem, pp. 823-827. *In* Proceedings, Beltwide Cotton Conferences, Nat. Cotton Council of Amer., Memphis, TN.

Liu, Y. C., and Y. B. Hwang. 1991. Life table of the cotton aphid, *Aphis gossypii* Glover, at various photoperiods. Chinese J. Entomol. 11: 106-116.

Liu, Y. C., and J. J. Peng. 1987. Population growth and temperature-dependent effect of cotton aphid, *Aphis gossypii* Glover, on bottle gourd cucurbit. Chinese J. Entomol. 7: 95-111.

Meyer, J. S., C. G. Ingersoll, L. L. McDonald, and M. S. Boyce. 1986. Estimating uncertainty in population growth rates: jackknife vs. bootstrap techniques. Ecology 67: 1156-1166.

Parajulee, M. N. 2007. Influence of constant temperatures on life history parameters of the cotton aphid, *Aphis gossypii*, infesting cotton. Environ. Entomol. 36: 666-672.

Parajulee, M. N., P. W. Kidd, D. R. Rummel, and W. P. Morrison. 2003. Aerial movements of the cotton aphid, *Aphis gossypii* Glover (Homoptera: Aphididae), and discovery of male morphs in the Texas High Plains. Southwest. Entomol. 28: 241-247.

Parajulee, M. N., S. C. Carroll, R. B. Shrestha, R. J. Kesey, D. M. Nesmith, and J. P. Bordovsky. 2010. Effect of nitrogen fertility on agronomic parameters and arthropod activity in drip irrigated cotton, pp. 1163-1169. Proceedings, Beltwide Cotton Conferences, National Cotton Council, Memphis, TN.

Slosser, J. E., W. E. Pinchak, and D. R. Rummel. 1998. Biotic and abiotic regulation of *Aphis gossypii* Glover in west Texas dryland cotton. Southwest. Entomol. 23: 31- 65.

Slosser, J. E., M. N. Parajulee, G. B. Idol, and D. R. Rummel. 2001. Cotton aphid response to irrigation and crop chemicals. Southwest. Entomol. 26: 1-13.

van Steenis, M. J., and K.A.M.H. El-Khawass. 1995. Life history of *Aphis gossypii* on cucumber: influence of temperature, host plant and parasitism. Entomol. Exp. Appl. 76: 171-131.

Wang, J. J. and J. H. Tsai. 2000. Effect of temperature on biology of *Aphis spiraecola*. Ann. Entomol. Soc. Amer. 93: 874-883.

Xia, J. Y., W. van der Werf, and R. Rabbinge. 1999. Influence of temperature on bionomics of cotton aphid, *Aphis gossypii*, on cotton. Entomol. Exp. Appl. 90: 25-35.