

**TEXAS A&M AGRILIFE RESEARCH & EXTENSION**

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# COTTON ENTOMOLOGY RESEARCH REPORT



**2012**

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

**2013**

# **COTTON ENTOMOLOGY PROGRAM**

## **RESEARCH ACTIVITY ANNUAL REPORT**

**2012**

### **SUBMITTED TO:**

**PLAINS COTTON IMPROVEMENT COMMITTEE**

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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 8 years. 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 and 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 behavior and ecology of major cotton pests of the High Plains with the goal of developing management thresholds based on cotton production technology. Some basic research is also underway to develop some molecular techniques to accurately identify some insect species, particularly *Lygus* bugs in a mixed population or to understand their movement behavior. That will allow us to recommend appropriate insecticide and dose for that specific insect. Our Program has successfully leveraged research funds based on the funding provided by PCIC to support our Technician position. We hope to continue this partnership as we challenge ourselves to deliver the best cotton insect-pest management recommendations to our Texas High Plains producers. The sudden departure of former Cotton Extension Specialist, David Kerns, posed some interim challenges for the 2012 summer, but exceptionally talented people on Dr. Parajulee's Program, together with seasoned IPM Agents we have in the region, transitioned this situation rather well, and we look forward to working with our soon-to-be hired Extension Specialist.

### 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 ARTHROPOD POPULATION DYNAMICS AS AFFECTED BY NITROGEN FERTILITY; HALFWAY, TEXAS

A long-term, ongoing study investigating the effects of differential nitrogen fertility on arthropod population dynamics in a typical drip-irrigation Texas High Plains cotton production system has been conducted for the last 11 years. Differential nitrogen fertility (0, 50, 100, 150, and 200 lbs N/acre) has been shown to significantly affect cotton plant physiological parameters, thereby influencing arthropod population dynamics.



Side-dressing variable rates of nitrogen fertilizer

#### INVESTIGATION OF GENETICALLY MODIFIED COTTON CONFERRING LYGUS-TOLERANCE; LUBBOCK, TEXAS

(IN COOPERATION WITH MONSANTO COMPANY)

As part of an ongoing Monsanto program to develop commercially available *Lygus*-tolerant cotton germplasm, numerous cotton lines, genetically modified to confer *Lygus* tolerance via protein expression (similar to *Bt* technology), are being evaluated for effectiveness under whole-plant cage field conditions, as well as for agronomic properties. Initial findings have been encouraging, and some exciting agronomic properties have been observed in gene-of-interest (GOI)-positive plants.



Field evaluation of transgenic cotton events

#### LYGUS SOURCE-SINK DYNAMICS IN COTTON-ALFALFA AGROECOSYSTEMS AND POPULATION GENETIC DIVERSITY

*Lygus* movement in a cotton-alfalfa agroecosystem was monitored in the Texas High Plains using protein markers. An enzyme-linked immunosorbent assay (ELISA) was used to precisely detect spray-applied protein markers adhering to the bodies of *Lygus* collected from cotton and alfalfa. The genomic DNA of eight *Lygus* populations from across the Texas High Plains was genotyped using microsatellite markers developed in our laboratory, revealing three genetically distinct regional populations. Phylogeographic study of *Lygus hesperus* from six western states is currently underway. Our investigations of pest movement and genetic structure will help in developing regional-level pest management strategies for Texas High Plains producers.

#### 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 growers, according to which, insecticide intervention for *Lygus* control may not be warranted when harvestable bolls accumulate  $\geq 350$  heat units or the boll is  $\geq 3$  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 Organic Research and Extension Initiative and Cotton Incorporated, are investigating ecological attributes of and management recommendations for thrips 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, both organic and conventional, should help growers in making informed and economically sound thrips management decisions.



In-field investigation of thrips densities on cotton seedling



## 2012 Beltwide Cotton Conference Research and Extension Report for the State of Texas

Compiled by

**Megha N. Parajulee**

**Professor, Faculty Fellow, and Texas A&M Regents Fellow**

**Texas.** The Texas cotton crop was much improved in 2012 compared to a devastating drought-induced loss of 58% in 2011. Still, approximately 25% of the Texas cotton crop failed in 2012, mostly owing to drought and blowing sands in the Texas High Plains region. The 2012 Texas cotton crop was expected to total 5.50 million bales, with an average yield of 539 pounds per acre. Texas planted 6.9 million acres of cotton with estimated 4.90 million acres harvested.

*Bt* technology, Bollgard II<sup>®</sup> and WideStrike<sup>™</sup>, represented approximately 70% of the harvested acres. Although the *Bt* trait is not an absolute necessity for West Texas because of >50% dryland acres and low caterpillar pest issues, West Texas region plants about 65% Bollgard<sup>®</sup> cotton due to superior agronomic attributes in these varieties. Southern and Far West Texas regions plant >90 *Bt* trait cultivars. Major seed sources for 2012 Texas cotton included PhytoGen 9%, All-Tex 9%, Deltapine 19%, Americot 19%, and FiberMax 40%.

Overall, insect pressure was extremely low in 2012. Throughout the state, insects resulted in only 0.46% of cotton yield losses, whereas other yield deterrents are attributed to have caused a 25% yield reduction on standing acres. Cotton fleahoppers caused the most damage to Texas cotton (0.23%) in 2012 with 3.5 million acres infested, 1.1 million acres treated, and 25,098 bales lost. Thrips were not as severe as most years with 3.8 million acres infested and a yield loss of only 9,199 bales (0.09%). Cotton aphids infested about 2 million acres and about 117,000 acres were treated, but no measurable yield loss from cotton aphids was detected. Bollworms caused about a 0.06% (6,238 bales) yield loss. *Lygus* bugs were almost non-existent while stinkbug caused about a 0.02% loss, which was primarily concentrated along the Gulf Coast. Spider mites are becoming more of a problem in Texas and 2012 saw spider mite issues statewide, which were attributed to a 0.03% yield loss. Approximately 280,000 acres were treated for mites in 2012. Kurtomathrips have become an emerging issue for Texas cotton, particularly in the High Plains, which infested 430,000 acres, but it did not cause a significant yield loss in 2012.

Boll weevil eradication in Texas has progressed well. Only two weevils were caught during 2012 in the Southern Blacklands, and 38 were caught in the Winter Garden area. Traps in the Lower Rio Grande Valley (LRGV) caught 207,833 weevils. The Rio Grande Valley is still an endemic region for cotton boll weevil due to absence of an effective program in the state of Tamaulipas, Mexico and due to the presence of volunteer cotton in some corn and sugarcane fields in the Rio Grande Valley.

**Texas Coast and Blacklands.** Soil moisture at planting was limited in most locations of the Texas Coastal Plains, with somewhat better conditions on the Upper Coast and Blacklands regions. Drought conditions persisted throughout the growing season along the Lower Gulf Coast with a high percentage of that cotton being destroyed before harvest. Rainfall in the other regions was generally good enough to harvest an outstanding crop with record high yields in some communities.

Generally speaking, thrips, cotton aphid, bandedwinged whitefly, beet armyworm, fall armyworm, saltmarsh caterpillar, southern armyworm, and cutworms were found in generally low numbers in the region. There were some exceptions where thrips and aphid numbers were substantial. Spider mites were higher than normal in the LRGV and Blackland regions. Fleahopper numbers were moderately high with many fields fruiting for a period of time before their populations rose sharply. Under dry conditions, fleahopper treatments did not result in yield or earliness improvement. Bollworm and tobacco budworm infestations were generally low even in non-*Bt* cotton fields. Plant feeding bugs such as verde plant bug, leafhopper, and stink bugs were high enough in number to result in some insecticide treatments, but once again, the problems were not widespread and in most cases treatments did not result in improved yield. Bollworm/tobacco budworm, cotton fleahopper, spider mites, and stink bugs were the only pests where yield loss was attributed (0.81% loss). The average number of foliar insecticide treatments was 3.2 with a high percentage of these treatments applied for cotton fleahopper.

Boll weevil eradication continued to progress toward elimination of the insect from the state. Trap captures amounted to 2 in the Southern Blacklands Zone, 38 in the Coastal Bend/Winter Garden Zone, and 207,833 in the

Lower Rio Grande Valley Zone (LRGV). It was estimated that boll weevil reproduction occurred on less than 0.5% of the state's acreage all of which was in the LRGV. Eradication in the LRGV has been hampered by the mild climate allowing cotton to grow throughout the year, cotton growing in other crops, and higher weevil populations in nearby fields in Mexico. An International Boll Weevil Technical Advisory Committee sanctioned by the National Cotton Council of America has been working to improve eradication programs on both sides of the border.

**Texas Far West and High Plains.** Texas Far West and South Plains region encompassed Texas Northern High Plains, Southern High Plains, and El Paso Valley. In this region, approximately 4.4 million acres of cotton were planted including 8,000 acres of pima cotton, which produced an estimated yield of 3.7 million bales. Conditions during early to mid-May were dry and cool, and cold soil temperatures deterred much of the planting, and the relentless winds quickly dried the soil. Continuous sandblasting suppressed early growth of cotton, whereas conditions turned extremely hot and windy during June and July, further exacerbating the droughty conditions. As cotton began to bloom, growers had difficulty meeting crop water demand. Yields were negatively impacted by the combination of high winds and drought, averaging 410 pounds/acre.

The High Plains region and El Paso Valley consisted of 75 and 95% Bollgard-technology cotton, respectively. Major varieties planted in this region included FM9170B2F, NG4012B2RF, EpicRF, FM2484B2RF, DP1044B2RF, PHY375WRF, PHY499WRF, DP164, FM1740B2F, DP1219B2RF, and ST4288B2F. Pima varieties included PHY802RF, DP357, PHY805RF, and DP340. Overall, insect pressure was minimal in 2012. Thrips and cotton aphids infested 2.2 and 1.4 million acres, respectively, but these pests required an average of one insecticide application, but their infestations did not result in significant yield loss. Cotton fleahoppers and *Lygus* bugs infested about 1.5 million acres, but the combined yield loss attributed to these two insect pests was estimated at 1,000 bales. The continuing drought caused significant expansion of Kurtomathrips throughout the region, infesting 430,000 acres, but the yield loss due to this late-season thrips was approximately 1,200 bales. Overall, insect-induced yield loss was estimated to be 0.014%.

**Rolling Plains.** Although somewhat improved from 2011, production issues in 2012 were dominated by the continuing drought. Winter and spring rains provided sufficient moisture for seed germination and early season cotton growth, but deep soil moisture was lacking. Approximately 1,698,787 acres were planted. Temperatures were not as hot as in 2012, but without summer rains, dryland crops failed to produce enough cotton to justify harvest across much of the Rolling Plains area. Irrigated crops fared better, but they required significant investments in energy to pump water. Some 60% of the planted acres were not harvested. On a planted acre basis, the cotton lint yield average in the Rolling Plains region was approximately 212 pounds/acre.

Thrips were not a major concern, but thresholds were exceeded and treatments were made on about 51,000 acres (3% of the planted acres). Cotton fleahoppers were also a concern on a few acres, but not a major concern across the region. Some 169,879 acres received insecticide treatments for cotton fleahoppers (10% of the planted acres). Bollworm and tobacco budworm populations were low with only an estimated 8,500 acres treated one time (0.5% of the planted acres). Grasshoppers were a concern on grasslands and coastal Bermuda pastures, but not a great concern in cotton fields. An estimated 3,400 acres were treated (0.2% of the planted acres). Spider mites, aphids and other pests were not a major concern. A local pink bollworm infestation south of Midland, TX was treated with sterile pink bollworm moths and is believed to have been eradicated. Limited pink bollworm trapping has not identified any other remaining pockets of pink bollworm in West Texas. **(Submitted by Megha Parajulee, Roy Parker, and Charles Allen).**

## Research Progress and Accomplishments

### Texas

*Sampling Strategies for Square and Boll-feeding Plant Bugs Occurring on Cotton along the Coastal Bend of South Texas and the Investigation of Landscape Metrics for Regional Pest Risk Assessment.* Cotton has experienced yield loss from boll and square damage from plant bugs (Hemiptera: Miridae) in South Texas during the last fifteen years, but the damage is variable across farms, years, and within seasons; and these insects are not readily controlled with region-wide techniques such as use of resistant cotton genotypes. In this situation, in-season field-specific decision-making for insecticide application is valuable. We previously identified insect sampling approaches for the main species in South Texas, verde plant bug, *Creontiades signatus*. We verified that its damage is similar to that caused by stink bugs reported in the southeast. For in-season decision-making, combining verde plant bug density

measurement taken at mid-bloom (scouting with a beat bucket) and measurement of proportion of bolls with internal injury (cracking green bolls < 1 inch in diameter) provided the best indication of at-harvest boll damage. In a new line of investigation, landscape analysis was applied to assess regional risk of verde plant bug. A spatially referenced crop data layer of South Texas, available from USDA, NASS, was input into a GIS. Landscape metrics in the area of 15 cotton fields were calculated and regressed on verde plant bug densities from these fields. A positive linear relationship between increasing pest density and increasing agricultural diversity was detected. As a basis for additional research, we hypothesize that this regional pest risk assessment approach can be used to allocate pest management resources (such as in-season insect monitoring) across a large cotton production region to locations that are more at risk. **(Brewer, Texas A&M AgriLife Research, Corpus Christi)**

*Cotton Fleahopper and its Damage to Cotton as affected by Plant Water Stress and Insect Seasonality.* Cotton fleahopper, *Pseudatomoscelis seriatus* (Hemiptera: Miridae) is the other major plant bug in South Texas and can cause excessive loss of cotton squares. In a second year field experiment in 2012, fleahopper populations were less sensitive to plant water stress and more sensitive to plant development stage. An early planting at mid-bloom (42 days after planting) had much higher populations when fleahopper first appeared in the field on June 1, compared to a later planting (31 days after planting). Insecticide sprays suppressed the population in the early planting. Two weeks later (June 14), cotton fleahopper populations increased as the later planting matured, and the sprays did not suppress this expanding population. Irrigation significantly increased yield, but the water regime did not affect fleahopper densities. Yield reduction attributable to fleahopper was not detected; even though the early planting had higher fleahopper populations (including ones above the economic threshold of 15 fleahoppers per 100 plants in our area). Overall, fleahopper populations were less sensitive to plant water stress and more sensitive to plant development stage, which partially explains field to field differences experienced by growers. Although more abundant during bloom, square sensitivity to cotton fleahopper damage and early season opportunity to suppress the population are the primary main considerations in insecticide use. Also, detection of fleahoppers during bloom in early planted cotton may serve as early warning of cotton fleahoppers in cotton planted later. **(Brewer, Texas A&M AgriLife Research, Corpus Christi)**

*Foliar insecticides for thrips control.* Two tests were conducted to evaluate actamiprid (Intruder<sup>®</sup>), acephate (Orthene<sup>®</sup>) and dicotophos (Bidrin<sup>®</sup>) to determine effectiveness of the various insecticides and application rates, to measure length of effective control, and specific to Intruder<sup>®</sup>, to compare application rates with and without MSO (methylated seed oil). Insecticides effectively reduced thrips numbers with the greatest effect on immature thrips. The low rate of Intruder<sup>®</sup> (0.5 oz/acre) in combination with methylated seed oil did not increase the effectiveness or length of control. For the entire report see <http://agrilife.org/coastalbend/program-areas/entomology/>, pages 26-37. **(Parker, Rankin and Biles, Texas A&M AgriLife Extension Service, Corpus Christi, George West, and Port Lavaca, respectively)**

*Cotton fleahopper control studies.* Two field studies were conducted to evaluate the effectiveness of insecticides on cotton fleahopper (*Pseudatomoscelis seriatus*). Chemicals evaluated included: acephate (Orthene<sup>®</sup>), acetamiprid (Intruder<sup>®</sup>), clothianidin (Belay<sup>®</sup>), CMT4586 (Bayer experimental), imidacloprid (Couraze<sup>®</sup>), sulfoxaflo (Transform<sup>®</sup>), and thiamethoxam (Centric<sup>®</sup>). All insecticides provided effective control of the fleahopper. In general Belay<sup>®</sup> and Centric<sup>®</sup> provided longer control of fleahopper nymphs, but all products performed well. The number of bolls produced in one of the two studies was numerically lower in the nontreated cotton compared with all tested insecticides. See the entire report at <http://agrilife.org/coastalbend/program-areas/entomology/>, pages 38-50. **(Parker and Rankin, Texas A&M AgriLife Extension Service, Corpus Christi and George West, respectively)**

*Cotton fleahopper response to plant density, insecticide treatment timing, and chemical treatments that alter normal fruiting pattern.* Three field studies were conducted to monitor effects of plant density, treatment timing and chemicals that altered normal fruiting with and without insecticide applied for the cotton fleahopper. Due to the drought conditions under which these studies were conducted, not as many differences were detected as would be expected had more water been available for plant development. As expected, more main stem nodes and higher numbers of bolls on vegetative branches were observed in plots with 3 plants per row foot compared to 6 plants per row foot. Percentage boll retention tended to be higher at 3 plants per foot and in insecticide treated cotton. Mite damage rating was higher where 4 insecticide treatments were applied for fleahopper at plant densities of 3 and 6 per row-foot. A numerical trend for higher yield in insecticide treated cotton was found in the plant density study. Another study was conducted to evaluate the effects of fleahopper control carried out at various weeks of squaring and to measure the impact of that timing on insect numbers, fruiting characteristics of plants, and lint production.



Fleahopper numbers in the test were moderately low, and only a few differences were found in fruiting characteristics. No differences were observed in lint production. In the third study, ethephon was applied at a very low rate to remove the first squares followed by either insecticide treated or not treated for fleahopper control. Yields were adversely affected by the ethephon treatment unlike that observed in previous studies where yield was not affected and the fruiting period was compacted over a shorter period of time. Thidazuron used at very low rates in the floral industry to retain buds had no major effects on cotton fruiting with or without fleahopper control. See the entire report at <http://agrilife.org/coastalbend/program-areas/entomology/>, pages 51-70. **(Parker, Texas A&M AgriLife Extension Service, Corpus Christi)**

*Evaluation of insecticide overspray on Bt cotton.* Caterpillars and damage to squares and bolls in Phytogen 367WRF cotton were almost nonexistent throughout a 7-week inspection period. Overspray insecticides included flubendiamide (Belt<sup>®</sup>), lambda-cyhalothrin + chlorantraniliprole (Besiege<sup>®</sup>), rynaxypyr (Prevathon<sup>®</sup>), and zeta-cypermethrin (Mustang Max<sup>®</sup>). Following application of the test insecticides, a spider mite infestation developed, but treatment was not needed as the population declined rapidly. Boll feeding true bug evidence of internal feeding was measured at 24% which required one treatment with Bidrin<sup>®</sup> over the entire test. No differences were found in cotton fiber characteristics or lint production. Lint yields were very high averaging 2,182 pounds/acre (4.55 bales/acre). Numerically, all insecticide treatments produced more lint than the untreated cotton. See the entire report at <http://agrilife.org/coastalbend/program-areas/entomology/>, pages 71-74. **(Parker, Texas A&M AgriLife Extension Service, Corpus Christi)**

*Monitoring of insects in commercial FiberMax<sup>®</sup> and TwinLink<sup>®</sup> cotton varieties.* The field study was conducted to determine if there were differences in development of caterpillar pest infestations in various FiberMax<sup>®</sup> Bt cotton varieties. The low infestation rate of caterpillar pests which were also observed in adjacent non-Bt cotton prevented screening of the cotton varieties for differences in susceptibility to these insects. Even though eggs of bollworm and cotton square borer were readily found, no larvae were detected in the study, and only two damaged squares were found. See the entire report at <http://agrilife.org/coastalbend/program-areas/entomology/>, pages 75-78. **(Parker, Texas A&M AgriLife Extension Service, Corpus Christi)**

*Bollworm and tobacco budworm pheromone trap survey.* Pheromone traps were monitored to measure relative abundance of bollworm (*Helicoverpa zea*) and tobacco budworm (*Heliothis virescens*) during the 2012 growing season. Both bollworm and tobacco budworm moth catch in traps were lower than in 2011, which were already lower than in years prior to 2011. The peak catch for bollworm numbers never exceeded 0.64/trap per night except for the first trapping week. The lower catch for the budworm probably related to fewer wild host plants and increased use of Bt cotton varieties. See the entire report at <http://agrilife.org/coastalbend/program-areas/entomology/>, pages 79-80. **(Parker, Texas A&M AgriLife Extension Service, Corpus Christi)**

*Evaluation of Bacillus thuringiensis Technology against Helicoverpa Population in Cotton Varieties Grown in the Upper Coastal Bend of Texas.* A study was conducted during the 2012 crop season to evaluate the *Bacillus thuringiensis* (Bt) genetically modified cotton in selected varieties grown in the Upper Coastal Bend of Texas and to record its effect on subsequent Heliothine populations. DeltaPine1048B2RF, FiberMax9058F, FiberMax1740B2F, and PhytoGen499WRF were selected for this evaluation. Data were randomly collected on the number of plants within a 10 ft. row section at 12 and 30 days after planting (DAP). Additional data were gathered on the number of beneficial insects, bollworm eggs, and bollworm larvae on 25 randomly selected plants, as well as observing 25 squares for bollworm damage, at 64, 71, and 78 DAP. Beneficial insects observed consisted of lacewing larvae, lady beetles, spiders, and minute pirate bugs. The plant stand data did not significantly vary between varieties until 30 DAP. Significant varietal differences were observed in the total number of damaged squares at 64 DAP only. There were no significant differences for the quantity of *Helicoverpa zea* eggs, larvae, and beneficial insects during the entire study. Only two larvae were seen in the variety FiberMax9058F, both were small and seen on separate days. However, it should be noted that the participating grower did overspray these plots between the 64 and 71 DAP data collection dates with a synthetic pyrethroid, which may have had a mitigating effect on subsequent *Helicoverpa zea* populations, egg lays and beneficials. **(Crumley, Texas A&M AgriLife Extension Service, Wharton)**

*Evaluation of Insecticide Overspray on Bt Cotton.* Test insecticide treatments included Prevathon<sup>®</sup>, Belt<sup>®</sup>, Besiege<sup>®</sup>, and Mustang Max<sup>®</sup>. These treatments were applied to plots of 4-row by 50-feet and the test was arranged in a randomized complete block experimental design with 4 replications of the treatments. The experimental treatments were applied with a Lee Spider Trac sprayer calibrated to deliver 12 GPA total volume. These treatments were made



on June 21 (3<sup>rd</sup> week of bloom). Up until that time the field had been inspected for the presence of caterpillar pests. Because infestations did not develop, the treatments were applied automatically without evidence of target pests. Caterpillars and damage to squares and bolls in FM 1944GLB2 cotton were almost nonexistent throughout the 7-week inspection period. Following application of the insecticide treatments, a stink bug infestation developed, but treatment was not needed as the population declined rapidly. Significant differences were found in lint yields regarding the Prevathon<sup>®</sup>, Belt<sup>®</sup> and Mustang Max<sup>®</sup> treatments. Insecticide treatments did not significantly increase the lint yield compared with that for untreated cotton. **(Crumley, Texas A&M AgriLife Extension Service, Wharton)**

*Evaluation of spider mite populations in south Texas.* The incidence of spider mites was evaluated in seven locations of south Texas from the Rio Grande Valley to the Coastal Bend from 4 April to 24 July of 2012. Investigation showed that spider mites were present throughout the entire growing season in all sites in greater number in 2012 compared with that in 2011. The increased presence of spider mites was the result of the severe drought in the state. Also, in addition to *Tetranychus urticae* and *T. tumidus* reported in 2011, *T. turkestanii* and *T. ludeni* were identified in samples from the Rio Grande Valley in 2012. **(Villanueva, Texas A&M AgriLife Extension Service, Weslaco)**

*Cotton Variety Tests and Evaluation of Flutriafol Applied at Planting for Control of Phymatotrichopsis Root Rot of Cotton.* Variety tests are utilized by growers of the Texas Blacklands to select varieties that will increase their net profits per acre. Varietal selection is an important component in keeping cotton a viable crop in the Blacklands. Evaluation of Flutriafol in the Blacklands combined with other studies in the state has had a significant impact on adoption of this material for control of cotton root rot. Reduction of cotton root rot provides producers with an easier harvest due to less dead stalks for cotton stripping and increases the net profit for cotton farmers. **(Jungman, Texas A&M AgriLife Extension Service, Hillsboro)**

*Cotton Entomology Research in the Sword Laboratory.* We have been working primarily in three areas this year: (1) the effects of fungal endophytes in cotton on insect and nematode-plant interactions, (2) environmental effects on *Helicoverpa zea* susceptibility to *Bt* toxins, and (3) ecological and geographic variation in microbial symbionts of the cotton fleahopper. Our fungal endophyte research entails the survey of cotton in Texas for potentially beneficial fungal endophytes and the screening of resulting candidate endophytes against target insect and nematode pests in lab, greenhouse and field assays. The goal of this project is to develop fungal endophytes as novel tools for both insect and nematode IPM. Research into the effects of environmental factors on the susceptibility of *H. zea* to *Bt* toxins involves rearing larvae on chemically-defined artificial diets varying in available protein and carbohydrate macronutrient content. Larvae on different optimal and suboptimal diets are then systematically challenged with different cry protein doses incorporated into the diet and measured for the effects of endogenous nutritional state on growth and development. The goal of this research is better characterize the mechanisms underlying variation in performance commonly observed in the field in *H. zea* susceptibility to *Bt* transgenic plants. Analyses of microbial symbiont communities associated with populations of cotton fleahopper are being conducted to test whether: 1) bacterial strains or communities associated with populations attacking cotton vary geographically across its distribution; 2) bacterial strains or communities vary according to host-plant species; and 3) bacteria strains influence the cotton fleahopper's fitness and reproductive isolation on different host-plant species. Results from this research aim at impacting cotton fleahopper monitoring through the inclusion of insect genotype and associated bacterial communities in the decision-making process prior to insecticide applications. **(Sword, Texas A&M University, College Station)**

*Effects of Water Stress on Cotton–Pest Interactions.* Over a 10-week field study at the Texas A&M Field Laboratory in Burleson Co., Texas, aphids and pulsed water stress were applied to 0.5 acres of cotton. Cotton plants were grown in 0.3 m<sup>3</sup> (6 ft<sup>3</sup>) cages in the field. Aphids were applied at a density of 25 aphids per plant at either the seedling or squaring stage, or were not present as a control. For water stress, furrow irrigation was applied when plants exhibited a turgor below -1.2 Mpa, determined by a pressure chamber. Control plants received water to keep them above a turgor of -1.2 Mpa. Aphids were counted on treatment plants throughout the study and removed from control plants by hand. COTMAN plant mapping and other plant measurements were taken. Plant samples for determining allelochemicals, ROS scavenging enzymes, amino acid, and carbohydrate concentrations were taken. We found major physiological differences between plants that had aphids applied at seedling compared to squaring plants. Seedlings with aphids were significantly smaller and had lower boll retention rates. Overall, aphid abundance did not differ between stressed and non-stressed plants, but plants with aphids introduced at the seedling stage had more aphids. During stress recovery, aphid abundance was greatest on plants that were water stressed and had

aphids during the seedling stage. With 1<sup>st</sup> fruiting position lint yield, stressed plants with aphids applied at the seedling stage produced the lowest amount of lint, while their non-stressed counterparts with aphids added at the seedling stage produced significantly more. There was a similar trend with the 2<sup>nd</sup> position as well. There were no significant differences between vegetative lint production across treatments. **(Eubanks, Texas A&M University, College Station)**

*Potential Benefits of Cotton Fleahopper Induced Defenses.* Plant mediated interspecific competition is an indirect interaction between herbivores in which the induction of defensive allelochemicals by one herbivore negatively affects the other. Here we investigate the potential for feeding by the cotton fleahopper, *Pseudatomoscelis seriatus*, (Hemiptera: Miridae) to initiate plant mediated interspecific competition which decreases the performance of Lepidopteron pests, beet armyworms, *Spodoptera exigua*. Moreover, we ask whether such interspecific herbivore competition can improve cotton yield by reducing the impact of the more damaging herbivore. In a preliminary field experiment, cotton plants were infested with cotton fleahoppers for 48 hours. Following fleahopper infestation, plants were infested with 10 neonate beet armyworm larvae. Larvae survival was tracked for 10 days and plant performance was tracked for the rest of the growing season. Preliminary results suggest that cotton plants may benefit from fleahopper feeding to induce defenses against lepidopteron herbivores and still maintain high yields. Beet armyworm retention was reduced on fleahopper infested cotton plants. At the end of the trial, only 25% of induced plants had caterpillars remaining, while 60% of control plants had at least 1 caterpillar remaining. This suggests that plants infested with fleahoppers are less palatable, or harbor induced allelochemical defenses which deter beet armyworms. Whether this effect can improve cotton yield remains to be seen. There were no differences in the total number of fruit produced as well as in the proportion of first position fruit retained among treatment groups. More study is needed to determine the potential benefits of fleahopper induced plant-mediated interspecific competition on cotton plants. **(Eubanks, Texas A&M University, College Station)**

*COTMAN monitoring of agronomic and entomological parameters in the evaluation of nitrogen fertility rate in drip irrigated cotton.* A multi-year comprehensive field study has been ongoing to examine the effect of soil nitrogen (residual nitrogen plus applied nitrogen) on cotton agronomic growth parameters and arthropod abundances under a drip irrigation production system. Fixed-rate nitrogen application experimental plots, established and fixed for 11 years, consisted of five augmented nitrogen fertility levels (0, 50, 100, 150, and 200 lb/acre) with five replications. Each year, soil in each experimental plot was sampled for residual nitrogen analysis immediately prior to planting or before treatment deployment. Rates of applied N exceeding 100 lb/acre resulted in higher residual nitrogen detection during the following season. However, variation in residual nitrogen did not significantly affect early plant growth (plant height, root length, or leaf area). Increased N levels corresponded to increased leaf chlorophyll content, but leaf chlorophyll content was generally similar across nitrogen levels exceeding 100 lb/acre. Aphid abundance was significantly lower in zero N plots versus other plots every year when cotton aphids were present. In 2010, aphid populations surpassed economic threshold in all N-augmented plots, whereas aphids remained below 50/per leaf, except for 1 week, in zero-N plots. Higher rates of applied N (>100 lbs/A) resulted in significantly higher leaf chlorophyll content compared to that in lower or zero N plots. No arthropod populations developed in 2011 due to extreme temperature and drought. A strong correlation was found between leaf chlorophyll content and lint yield. Nitrogen fertility level influenced fruiting profile and boll maturity. Plants ceased setting additional squares in zero and 50-lb N plots 2 wk into flowering while higher N plots were actively producing squares. Averaged over last four years (2008-2011), the zero-N treatment produced the lowest yield (912 lb/acre) and yield increased curvilinearly with each additional 50 lb N added, with highest average lint yield occurring in 150 lb N/acre treatment (1,288 lb/acre). Although two synthetic pyrethroid applications were applied to all treatment plots during August 2012 to encourage a cotton aphid buildup, observed aphid numbers remained low for 2012. This year's arthropod sampling then focused on community composition and relative numbers of other cotton pest and beneficial species among the five nitrogen treatments. Nitrogen augmentation rates significantly influenced all agronomic parameters evaluated in this study, most with curvilinear relationships. These relationships observed in 2012 are similar to what have been observed in previous years. **(Parajulee, Texas A&M AgriLife Research, Lubbock)**

*Development of Economic Threshold and Management Recommendations for Lygus in Texas High Plains 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. In 2012 study year, boll damage potential of *Lygus hesperus* was determined in a no-choice cup-cage study.

Ten cohorts of cup-caged single bolls (10-20 days old) were each exposed to a *Lygus* adult for 48 hours and the boll damages were quantified. After bolls reached 16 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 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 53% 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. There were no significant yield differences between control plants and *Lygus* infested plants when plants were first infested with *Lygus* bugs at 550 HU after first flower. 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. With these series of multi-year field studies, we hope to characterize the relationships between cotton boll maturity and *Lygus hesperus* infestations as well as to develop a *Lygus* economic threshold for Texas High Plains cotton. (Parajulee, Texas A&M AgriLife Research, Lubbock)

*Integrated production systems to enhance cotton quality for global markets.* Large plot, multi-disciplinary field experiments were established at three locations including AG-CARES (Lamesa), Helms Farm (Halfway), and the Chillicothe Research Station near Vernon, which represented three diverse production environments. Cotton yield, fiber quality, and profitability as affected by two irrigation methods, low energy precision application sprinkler (LEPA) and subsurface drip (SDI) and three irrigation levels, cropping system, cultivar, and methods of harvest (picker vs. stripper) were compared. Within the plot areas, plant disease, nematode, and arthropod pest management tactics as affected by treatment variables were investigated. Agronomic, entomological, and economic data will be used in formulating representative mathematical models to derive optimum decision guidelines. Specifically, entomological component of this project included the evaluation of two cotton cultivars (PHY 367WRF and ST 5458B2RF) for late season *Lygus* severity under low and high irrigation levels. There were a total of 24 experimental plots (2 insect release treatments x 2 water levels x 2 cultivars x 3 replications). Two 10 row-ft sections of cotton were randomly selected and flagged in each plot on August 6, 2012. *Lygus* bugs were released in one of the 10 ft sections in each plot, while the second 10 ft section per plot was maintained *Lygus*-free (control). The release treatment of 4-6 bugs per plant was designed to exert significant insect pressure on the fruiting cotton plants. After one week of *Lygus* infestation exposure, a total of six cotton plants (3 from *Lygus*-infested section; 3 from control) from each plot were cut and brought into the laboratory to evaluate the resulting *Lygus* external cotton boll injuries. The PHY 367WRF cultivar plants were significantly taller (21.7 cm) than ST 5458B2RF (18.4 cm). Cotton plants in high irrigation plots were significantly taller (21.1 cm) than low irrigation plots (19.0 cm). A significantly higher number of green bolls (8.5 bolls/plant) were observed in PHY 367WRF as compared to ST 5458B2RF (6.4 boll/plant) following one week of exposure to the *Lygus* infestation. Pre-harvest plant mapping data showed PHY 367WRF retained a significantly higher number of harvestable open bolls (7.9 per plant) compared to ST 5458B2RF (5.6 per plant). A significantly larger numbers of harvestable bolls were found on PHY 367WRF grown under high irrigation (8.9 bolls/plant) compared to those grown under low irrigation conditions (6.9 bolls/plant). The two irrigation levels did not significantly affect the lint yields within each of the two cultivars. The higher yielding cultivar, PHY 367WRF, produced significantly high lint yield in the *Lygus*-free control plots than in the *Lygus*-infested plots, while the late-season (74 days after planting) *Lygus* infestations did not significantly influence the lint yields of ST 5458B2RF as compared to the control treatment plot yields. (Parajulee, Texas A&M AgriLife Research, Lubbock)

*Characterization of Cotton Crop Response to Thrips Injury for Improved Thrips Management in Texas.* Thrips mass rearing protocols were evaluated for *Frankliniella occidentalis* and *Kurtomathrips morrilli*. Thrips were reared on four different types of diet including: 1) fresh green beans, 2) a diluted honey solution, 3) honey bee pollen, and 4) fresh cucumber. Green beans with honey and/or pollen supplement was found to be the most suitable and easily available food source for *Frankliniella occidentalis* rearing, but *Kurtomathrips morrilli* colony did not colonize successfully on green beans. Five types of thrips cages were designed to confine thrips to single cotton plants in the field. Thrips cages were optimized for higher thrips containment, survival and better plant quality. Ventilated 1-gallon plastic jar cages were selected for the field evaluations. The thrips cages were deployed to evaluate cotton seedling response to 5 selected thrips densities (0, 1, 2, 4 and 6 adult thrips per plant). Field cages failed to retain the targeted density of adult thrips on the caged plants. None of the cages evaluated were found to be satisfactory for the

planned thrips field-cage studies. Thrips were either killed due to overheating inside the cages or the thrips escaped out from the ventilation portions of the cage. The 2013 study will focus on refining the 2012 experimentation to develop thrips field-cages to retain thrips on caged plants with minimal effect on plant quality. Natural thrips infestations and cotton crop damage were evaluated in an open field experiment. Four selected cotton cultivars (SSG-HQ-212-CT, DP 357/360, FM 1740B2F and PhytoGen 367WRF) were planted as four treatments within each of four blocks (replicates) on May 18, 2012. Thrips densities were monitored in all 16 experimental plots via a plant washing technique. Plant response to thrips injury was monitored by measuring plant height, leaf area, root length and total biomass of cotton seedlings from each plot. A greenhouse study was conducted to evaluate the damage potential of *Frankliniella occidentalis*. Cotton seeds (FM 9180 B2F) were planted in 150 small plastic pots in a greenhouse. The cotton seedling response to various level of thrips injury was evaluated using clear plastic cup-cages covered with a paper towel. Five levels of thrips densities (0, 1, 2, 4, and 6 thrips adult per plant) were released onto caged cotton seedlings at the 2-3 true leaf stage and plants were exposed to the thrips for 6 days. The thrips injuries to cotton leaves were recorded using visual damage rating. The 2012 studies would serve the foundation for more refined studies that are expected to be repeated for the next two years. **(Parajulee, Texas A&M AgriLife Research, Lubbock)**

*High Plains Cotton Development of cultivars and IPM systems for organic cotton production.* Seed treatments and/or foliar applications of various insecticides are common practices for managing the western flower thrips [*Frankliniella occidentalis* (Pergande)] in seedling cotton in the Texas High Plains. However, available options for thrips management in organic cotton production systems are very limited. The efficacy of five organic products: Entrust<sup>®</sup> Naturalyte<sup>®</sup> Insect Control (spinosad microbial 2 oz/A), Aza-Direct<sup>®</sup> Biological Insecticide (Azadirachtin 1.2% 16 oz/A), Bugitol Concentrate (mustard oil 48 oz/A), Safe-T-Side<sup>™</sup> Spray Oil plus Ecotec (0.5 lb + 0.5 qt/A), and NoFly WP Mycoinsecticide (HT6-100; 4 lb/A rate) were evaluated and compared to an untreated control. Treatments were initiated soon after cotton seed germination and applied weekly for three consecutive weeks. In conjunction with treatment applications, cotton was sampled weekly via absolute sampling (plant washing) method. In addition to thrips densities, cotton seedling total (above and below soil surface) biomass, leaf area, and leaf chlorophyll indices were recorded. Due to very high pressure of immigrant adult thrips and subsequent reproduction on cotton seedlings, none of the treatments proved effective when cotton was at the 1 true-leaf stage. Nevertheless, Entrust<sup>®</sup> significantly suppressed thrips populations and kept it below ET when cotton was at the 2-5 true-leaf stage. Plants from Entrust<sup>®</sup>-treated plots had significantly higher leaf areas compared with that in other treatment plots, likely due to lower thrips numbers on Entrust<sup>®</sup>-treated cotton plants. Plant biomass was significantly higher in Entrust<sup>®</sup> and Azadirachtin<sup>®</sup> treatments compared with that in other treatments. While Entrust<sup>®</sup> provided suppression of thrips in this trial and residual activity appeared to be cumulative, no treatment provided any benefit in lint yield. **(Parajulee, Dever, Arnold, Texas A&M AgriLife Research, Lubbock; Vandiver, Texas A&M AgriLife Extension Service, Muleshoe)**

## EFFECT OF NITROGEN FERTILIZER ON COTTON HOST-PLANT QUALITY AND ITS IMPACT ON ARTHROPOD ACTIVITY

M.N. Parajulee, S.C. Carroll, R.B. Shrestha, J.P. Bordovsky

**Objective:** The objective was to evaluate the effect of nitrogen fertilizer application rates on the population dynamics of cotton arthropods, plant growth parameters, and lint yield.

**Methodology:** A high-yielding FiberMax cultivar, FM 9063B2R, was planted at a targeted rate of 56,000 seeds/acre on May 17, 2012. The experiment consisted of a randomized block design with five treatments and five replications. Pre-treatment soil samples (consisting of three soil cores; 0 to 24-inch depth), were collected from each of the 25 experiment plots on June 1, 2012. The five side-dress N fertilizer application treatments at rates of 0, 50, 100, 150, and 200 lb N/acre were applied on July 6, 2012. Crop growth and insect activity were monitored during the crop season. Weekly during most of July and August, numerous plant variables were measured to evaluate the influence of residual soil nitrogen on early plant growth patterns. Examples of collected plant data variables included: 1) plant biomass weight, 2) plant height, 3) total leaf area, 4) percent leaf nitrogen, 5) number of 1<sup>st</sup> position cotton squares/plant, and 6) percent fruit shed.

**Results:** Higher levels of available residual soil N and augmented N applications significantly affected plant biomass and height. Both plant biomass and height increased continuously from 0 lb/acre up to the 150 lb/acre N applied plots. Plant biomass was significantly highest in the 150 lb/acre N applied treatment, but it decreased significantly when an additional 50 lb/acre N was applied (200 lb/acre N plots). Leaf chlorophyll content (SPAD reading) increased linearly from zero-N to 100 lb N treatment plots (Fig. 1), but the plots which received the three highest N application rates (100, 150, and 200 lb N/acre) exhibited relatively consistent leaf chlorophyll readings. Arthropod densities were low across all N fertility treatments during 2012. As a result, treatment effect on overall arthropod abundance was not detected. Nitrogen fertility level influenced fruiting profile and boll maturity. Plants ceased setting additional squares in zero and 50-lb N plots 2 wk into flowering while higher N plots were actively producing squares.

Zero-N applied plots produced the lowest yield and yield increased curvilinearly, with highest average yield occurring in the 150 and 200 lb N/acre treatments (Fig. 2).

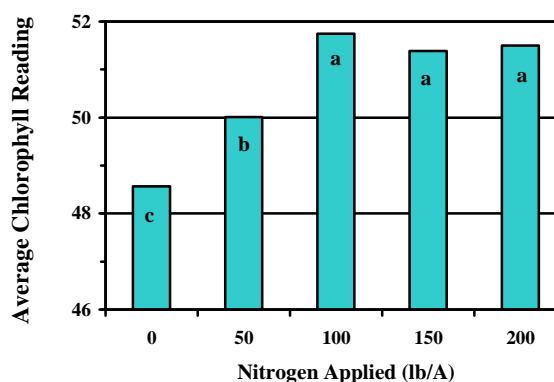


Fig. 1. Effect of nitrogen application rates on fifth mainstem leaf chlorophyll content, 2012.

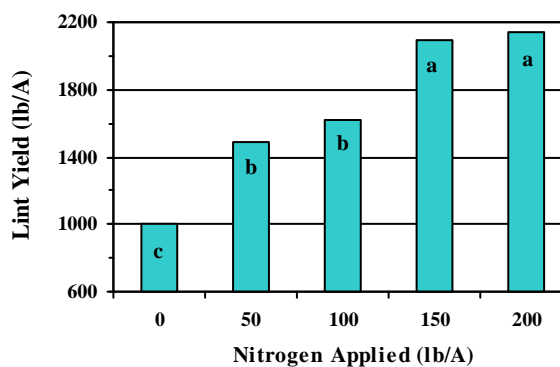


Fig. 2. Effect of N application rates on lint yield after 10 years of repetitive applications, 2012.

## Cotton yield response to late-season *Lygus* plant bug infestations as influenced by cultivar x irrigation level treatments, Lamesa, TX, 2012

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### MATERIALS AND METHODS:

Plot Size:	4 rows by 50 feet, 3 replications
Planting Date:	May 22, 2012
Varieties:	PHY 367WRF, ST 5458B2RF
Fertilizer:	130-40-0
In-season Irrigation:	Low = 4.6 inches; High = 9.0 inches
Insect Treatments:	Control (zero <i>Lygus</i> ); <i>Lygus</i> infested (4-6 nymphs per plant)
Insect Release Date:	August 6, 2012 (late-season boll developmental period)
Harvest Date:	October 18, 2012 (hand-harvested)

Two cotton cultivars (PHY 367WRF and ST 5458B2RF) were evaluated under low and high irrigation levels. There were a total of 24 experimental plots (2 insect release treatments x 2 water levels x 2 cultivars x 3 replications). Two 10 row-ft sections of cotton were randomly selected and flagged in each plot on August 6, 2012. *Lygus* bugs were released in one of the 10 ft sections in each plot, while the second 10 ft section per plot was maintained *Lygus*-free (control). The release treatment of 4-6 bugs per plant was designed to exert significant insect pressure on the fruiting cotton plants. After one week of *Lygus* infestation exposure, a total of six cotton plants (3 from *Lygus*-infested section; 3 from control) from each plot were cut and brought into the laboratory to evaluate the resulting *Lygus* external cotton boll injuries. Pre-harvest plant mapping was conducted on October 18, 2012 to monitor the harvestable boll retention profile as influenced by the bug augmentation treatment. Both flagged ten foot sections (control and *Lygus*-infested) from each plot were hand-harvested to determine the impact of a late-season *Lygus* infestation on lint yield and quality.

### RESULTS AND DISCUSSION:

The PHY 367WRF cultivar plants were significantly taller (21.7 cm) than ST 5458B2RF (18.4 cm). Cotton plants in high irrigation plots were significantly taller (21.1 cm) than low irrigation plots (19.0 cm). The late-season *Lygus* infestations did not significantly influence final plant height of the two evaluated cotton cultivars (Table 1). A significantly higher number of green bolls (8.5 bolls/plant) were observed in PHY 367WRF as compared to ST 5458B2RF (6.4 boll/plant) following one week of exposure to the *Lygus* infestation. A numerically higher numbers of green bolls were found on the high irrigation plots than those observed on plants of the low irrigation treatment, yet there were no statistical differences between the number of green bolls from control and *Lygus* infested plants (Table 2). Pre-harvest plant mapping data showed PHY 367WRF retained a significantly higher number of harvestable open bolls (7.9 per plant) compared to ST 5458B2RF (5.6 per plant). A significantly larger numbers of harvestable bolls were found on PHY 367WRF grown under high irrigation (8.9 bolls/plant) compared to those grown under low irrigation conditions (6.9 bolls/plant; Table 3). PHY 367WRF produced



a significantly higher lint yield (853 lb/A) compared to ST 5458B2RF (695 lb/acre). The two irrigation levels did not significantly affect the lint yields within each of the two cultivars. The higher yielding cultivar, PHY 367WRF, produced significantly high lint yield in the *Lygus*-free control plots than in the *Lygus*-infested plots (Table 4), while the late-season (74 days after planting) *Lygus* infestations did not significantly influence the lint yields of ST 5458B2RF as compared to the control treatment plot yields.

Table 1. Cotton plant height (cm) as influenced by a late-season *Lygus* infestation in an irrigation level x variety study, Lamesa, Texas, 2012.

Insect Treatment	Plant Height (cm)					
	PHY 367WRF			ST 5458B2RF		
	High Water	Low Water	Average	High Water	Low Water	Average
Control	22.6	20.3	21.5 a	18.8	17.7	18.3 a
Infested	24.6	19.3	22.0 a	18.5	18.8	18.7 a
Average	23.6 A	19.8 A		18.7 A	18.3 A	

Table 2. Average number of bolls retained per plant following 1-week of *Lygus* infestation exposure, Lamesa, Texas, 2012.

Insect Treatment	Number of Bolls/Plant					
	PHY 367WRF			ST 5458B2RF		
	High Water	Low Water	Average	High Water	Low Water	Average
Control	10.9	7.8	9.4 a	9.7	4.6	7.2 a
Infested	9.9	5.6	7.8 a	9.2	4.2	6.8 a
Average	10.4 A	6.7 A		9.5 A	4.4 A	

Table 3. Average number of harvestable cotton bolls per plant as influenced by a late-season *Lygus* infestation in an irrigation level x variety study, Lamesa, Texas, 2012.

Insect Treatment	Number of Harvestable Bolls/Plant					
	PHY 367WRF			ST 5458B2RF		
	High Water	Low Water	Average	High Water	Low Water	Average
Control	8.0	7.7	7.9 a	6.5	4.6	5.6 a
Infested	9.7	6.1	7.9 a	7.1	4.2	5.7 a
Average	8.9 A	6.9 B		6.8 A	4.4 A	

Table 4. Lint yield (lb/acre) as influenced by a late-season *Lygus* infestation in an irrigation level x variety study, Lamesa, Texas, 2012.

Insect Treatment	Lint Yields (lb/acre)					
	PHY 367WRF			ST 5458B2RF		
	High Water	Low Water	Average	High Water	Low Water	Average
Control	1088.0	711.2	899.6 a	762.5	577.7	670.1 a
Infested	1090.0	522.7	806.4 b	818.5	621.6	720.1 a
Average	1089.0 A	617.0 A		790.5 A	599.7 A	

## CHARACTERIZATION OF BOLL DAMAGE POTENTIAL OF *LYGUS HESPERUS* AND ITS RELEVANCE IN PEST MANAGEMENT

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Lubbock, TX

### Abstract

Cotton boll feeding biology and behavior of *Lygus hesperus* has not been fully understood. Boll damage potential of *Lygus hesperus* was determined in a no-choice cup-cage study. Ten cohorts of cup-caged single bolls (10-20 days old) were each exposed to a *Lygus* adult for 48 hours and the boll damages were quantified. After bolls reached 16 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 adults per cage) for one week when plants were at two selected boll development stages (350 and 550 HU after first flowering). 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 53% 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. There were no significant yield differences between control plants and *Lygus* infested plants when plants were first infested with *Lygus* bugs at 550 HU after first flower. 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 2011). *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 2010, a 3.9% reduction in U.S. cotton yields was attributable to arthropod pests – 0.68% due to *Lygus* species, which was ranked third among other yield-reducing pests (Williams 2011) 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 2010 (Williams 2011). *Lygus* can cause severe cotton square loss, anther damage, and seed damage depending upon which 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 a 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. Since 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, 4) quantify the yield loss caused by 4 different levels of *Lygus* infestations (0, 1, 2 and 4 *Lygus* adults per plant), and 5) determine the overall yield contribution of cotton bolls from different nodal positions. 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 located near Lubbock, Texas. On May 18, 2012, cotton cultivar ST 5458B2RF was planted on 40-inch spaced rows of a furrow-irrigated field. The targeted seeding rate was 56,000 seeds per acre. On June 2, 2012, the entire test was treated with Orthene<sup>®</sup> 97S for thrips at the rate of 3.0 oz per acre and with Cornerstone Plus<sup>®</sup> herbicide (41% glyphosate) at 32 oz per acre for weed management.

The experimental design was a split-plot randomized block with three replications. Ten cotton boll age cohorts (10 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 1<sup>st</sup> cup-caged bolls received about 450 HU, whereas the August 10<sup>th</sup> 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.



Figure 1. Deployment of cup-cages. Lubbock County, TX, 2012.

### **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. On May 18, 2012, cotton cultivar ST 5458B2RF was planted 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, 2012, this 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.

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 bugs/plant]). There were a total of 24 experimental units. Each experimental unit had 8 cotton plants as subsamples (4 used for damage assessment and 4 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, 2012. When the cotton plants reached the target maturity level (350 HU after first flower on August 7, and 550 HU after first flower on August 21), lab-reared *Lygus* were released into the whole-plant sleeve-cages at the rates of 0, 1, 2, and 4 bugs/plant. Again, the *Lygus* adults were initially reared on artificial diet, yet “trained” on fresh green beans and cotton squares for a week before using them for the boll feeding experiment. Cotton plants were exposed to the *Lygus* adults for 6-7 days, after which time, the insects were killed via a pesticide application. Four randomly selected cotton plants from each plot were cut and brought to the laboratory on August 13 and August 21 for the 350 HU and 550 HU plots, respectively. Boll positions, internal and external *Lygus* damage, boll weights, boll diameters, and boll hardness were recorded for all plants from Block 1. For plants from the other blocks, external boll damage, boll weight, and size were recorded. 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 on October 3, 2012. After the crop was ready to harvest, the remaining 4 caged plants from each plot, which had been maintained pest-free, were harvested manually to evaluate the lint yields and fiber quality.



Figure 2. Deployment of whole-plant cages. Lubbock County, TX, 2012.

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 ( $\alpha=0.1$ ) and treatment means were compared by LSMEAN procedure.

## **Results and Discussion**

### **Boll Development vs. *Lygus* Damage Potential**

During the 2012 active boll developmental stage of August 1-20, the Lubbock area cotton crop received, on average, 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). 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, bigger and tougher to puncture. When bolls reached an age of 16 days, *Lygus* caused very little seed damage (<2 seeds per boll) that did not result in significant lint yield reductions (Fig. 4). When cotton bolls received >350 HU after first flower (or approximately 16 days of age), 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 and 4).

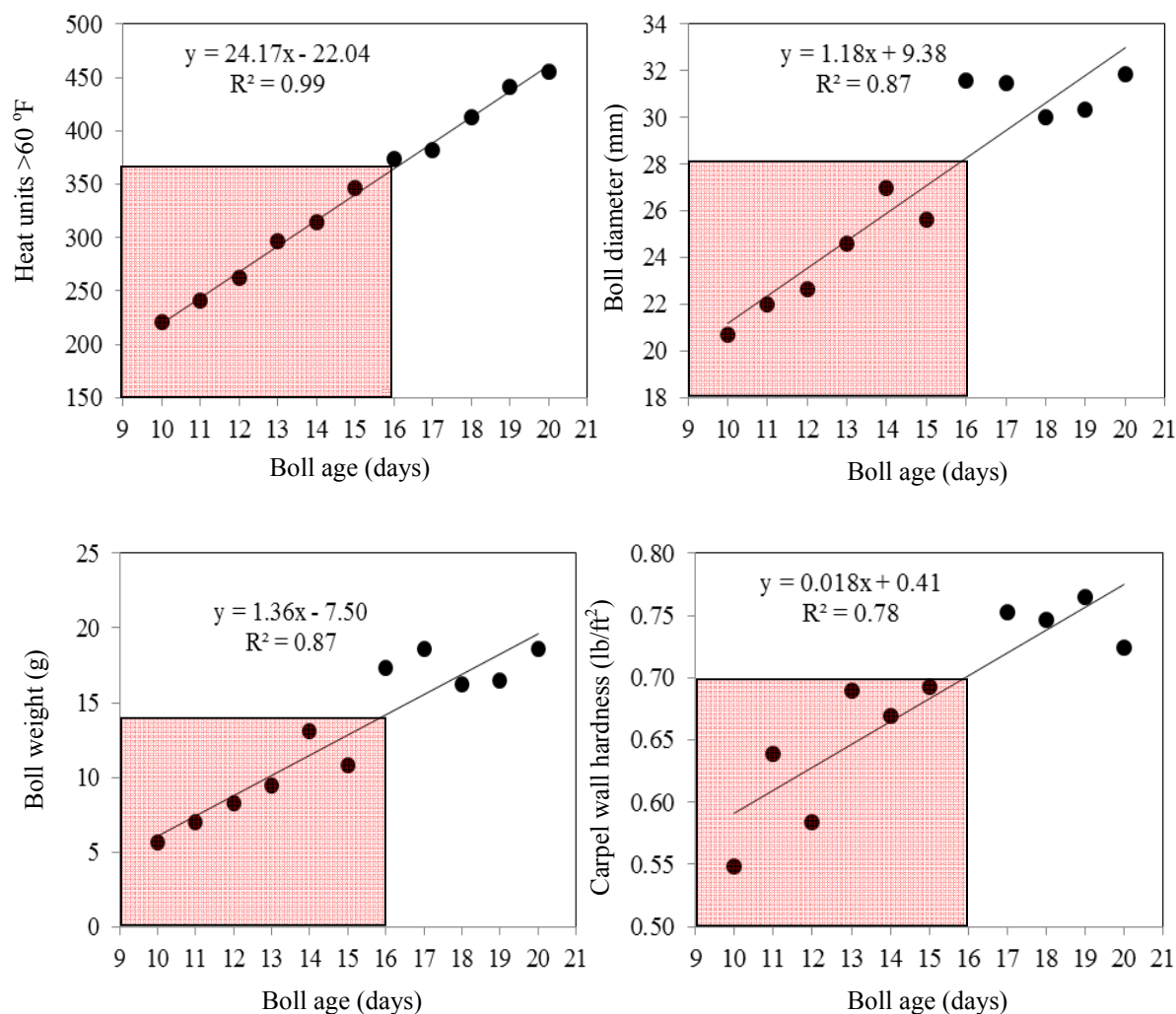


Figure 3. Cotton boll age relationships as associated to heat unit accumulations, boll size, boll weight, and carpel wall hardness. Lubbock County, Texas, 2012.

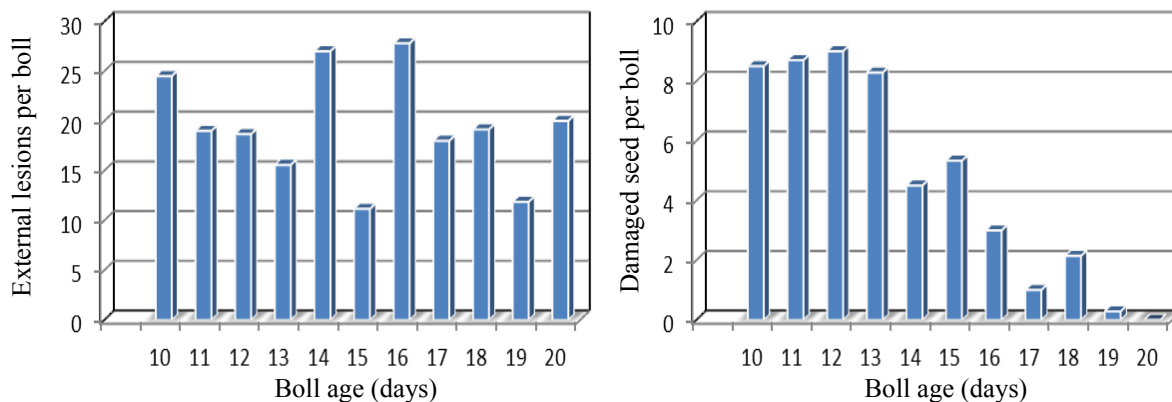


Figure 4. Following 48 hours of feeding by a single *Lygus* adult, boll injury (external lesions and damaged seeds) at various boll ages. Lubbock County, TX, 2012.

### Fruiting Profile

At 350 HU after first flower, average of 56.6% fruit retention was observed, but fruit retention was decreased to 36.7% 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. 5).

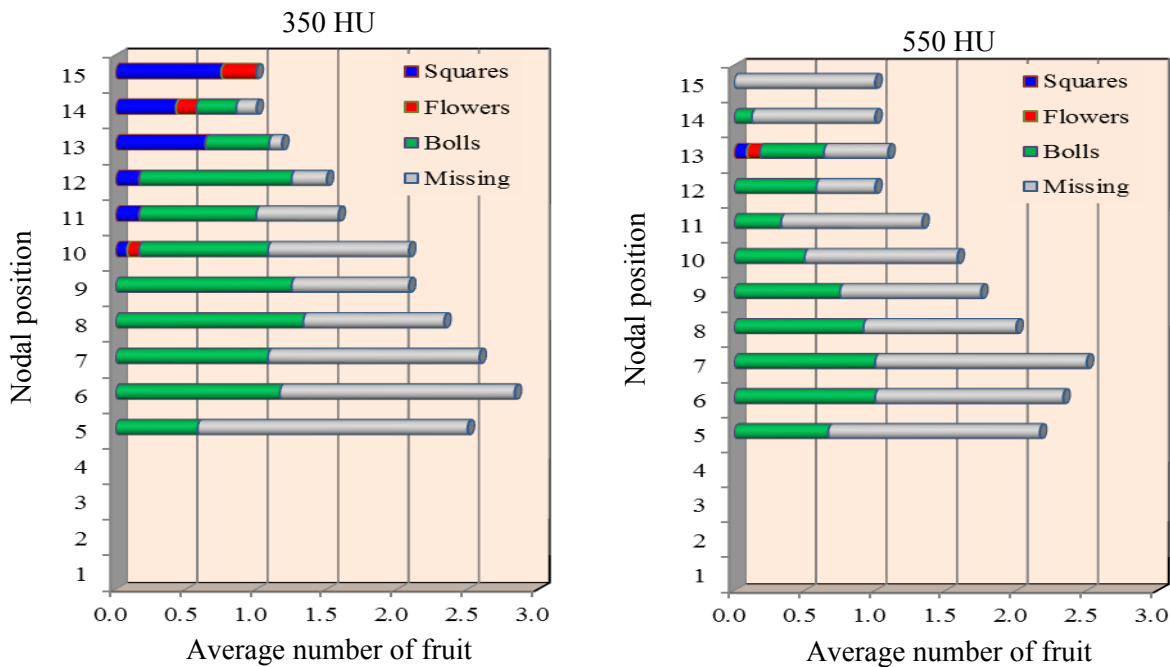


Figure 5. Cotton fruiting profile at 350 and 550 HU after first flower. Lubbock County, TX, 2012.

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. 6). When the cotton plants matured from 350 HU to 550 HU, they dropped all of the 4<sup>th</sup> fruiting position and most of the 3<sup>rd</sup> 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.

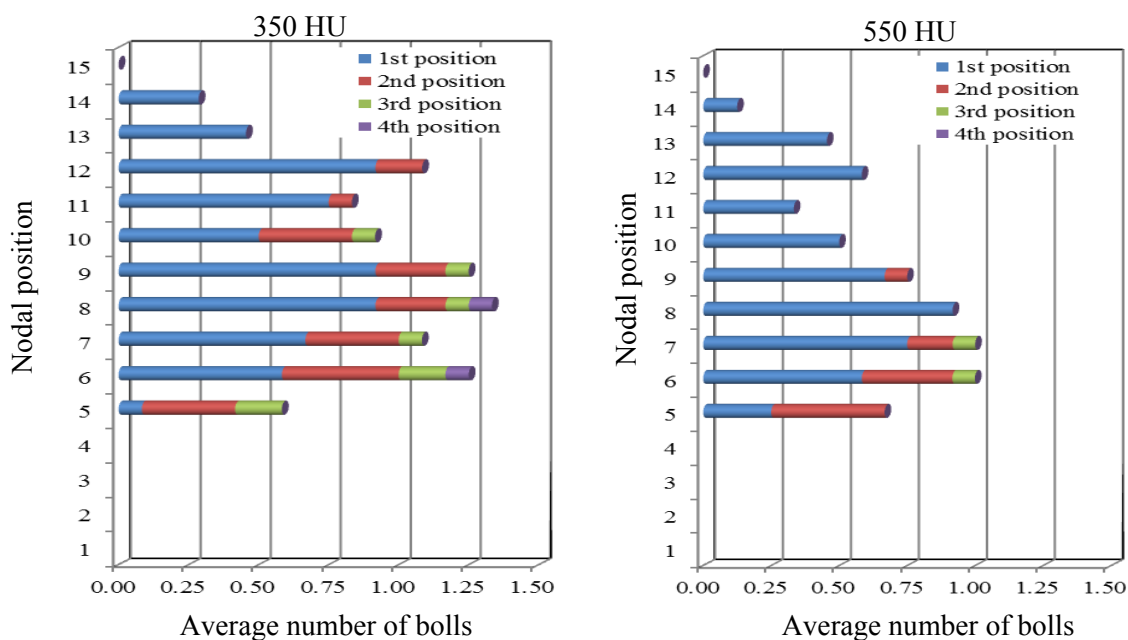


Figure 6. Cotton boll distribution on sympodial branches at 350 and 550 HU after first flower. Lubbock, TX, 2012.

#### **Boll Maturation Profile**

Thirty-two cotton plants were harvested (16 plants each from 350 HU plots and 550 HU plots) from which 643 cotton 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 plants in the same field had 70% of the bolls safe from *Lygus* damage (Fig. 7). 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 infested to a mid-season crop (350 HU) compared to that for a late season infestation (550 HU).

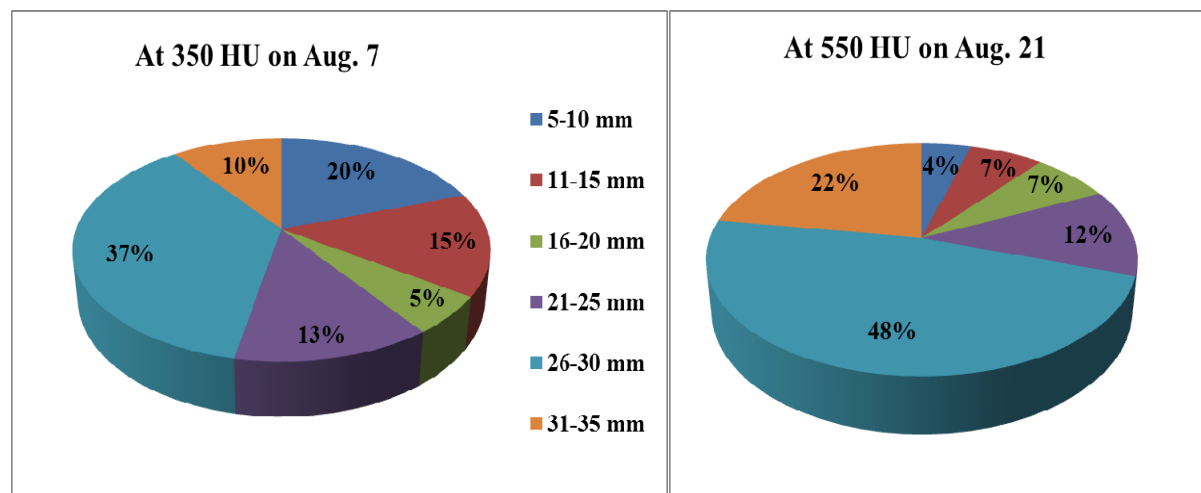


Figure 7. Within-plant boll maturity profile of cotton plants at 350 and 550 HU. Lubbock, TX, 2012.

For our 2012 cotton crop, within-plant cotton boll maturation profile shows that bolls distributed from the 5<sup>th</sup> to 13<sup>th</sup> nodes (Fig. 8). At the 350 HU stage, the top 4 bolls (from 10-13<sup>th</sup> 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 5<sup>th</sup> to 9<sup>th</sup> 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.

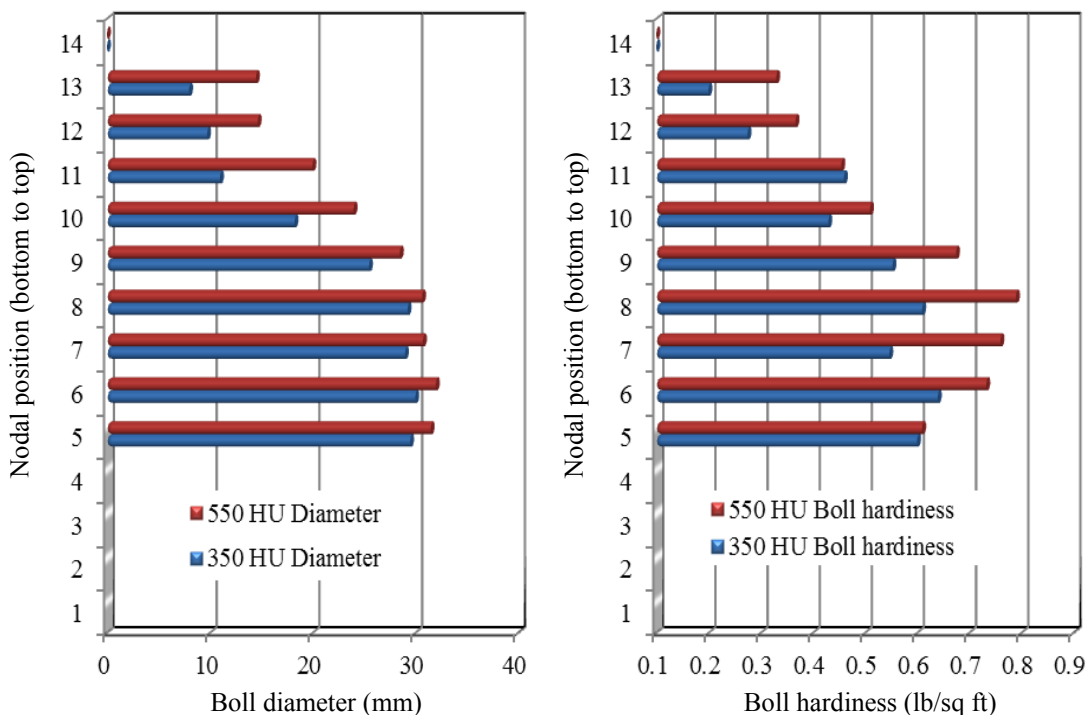


Figure 8. First position boll size profiles of 350 and 550 HU (after first flower) cotton. Lubbock County, TX, 2012.

### **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. 7). 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 difference 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.

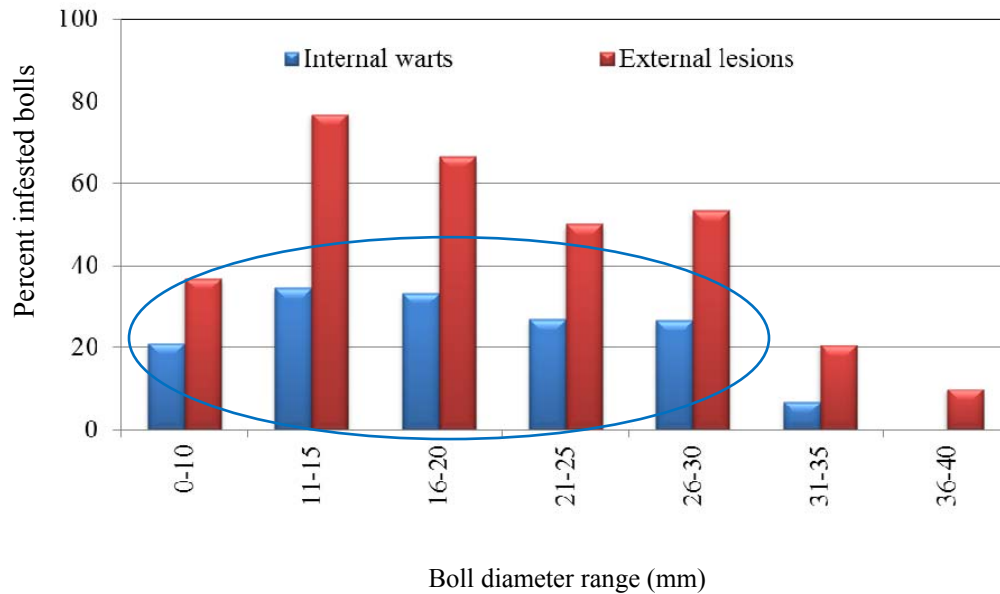


Figure 9. Boll feeding preference of *Lygus* in whole-plant cages based upon the proportion of external and internal boll damage. Lubbock County, TX, 2012.

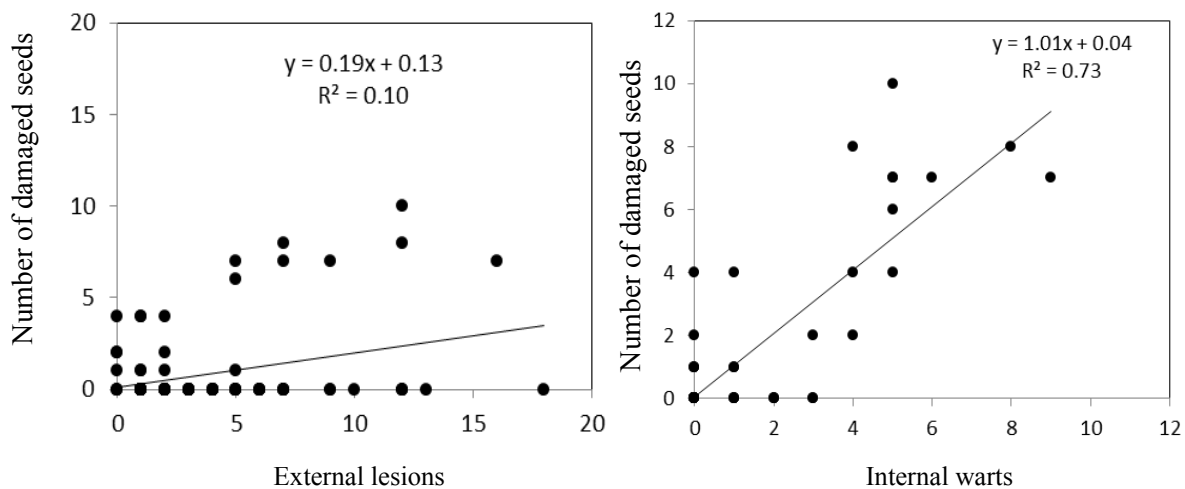


Figure 10. Relationships between the number of damaged seeds per boll and the number of external lesions or internal warts. Lubbock County, TX, 2012.

### Yield Loss

Artificial infestation 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). Although it is expected that the degree of yield loss, due to the same levels of *Lygus* infestation, varies with the crop stage the *Lygus* infestation occurred, it was rather a marked change in the crop tolerance to *Lygus* injury from 350 to 550 HU. 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. More detailed research is

needed to characterize the interaction between crop phenology and *Lygus* feeding-induced yield loss. Our continuing project is expected to address some of these issues.

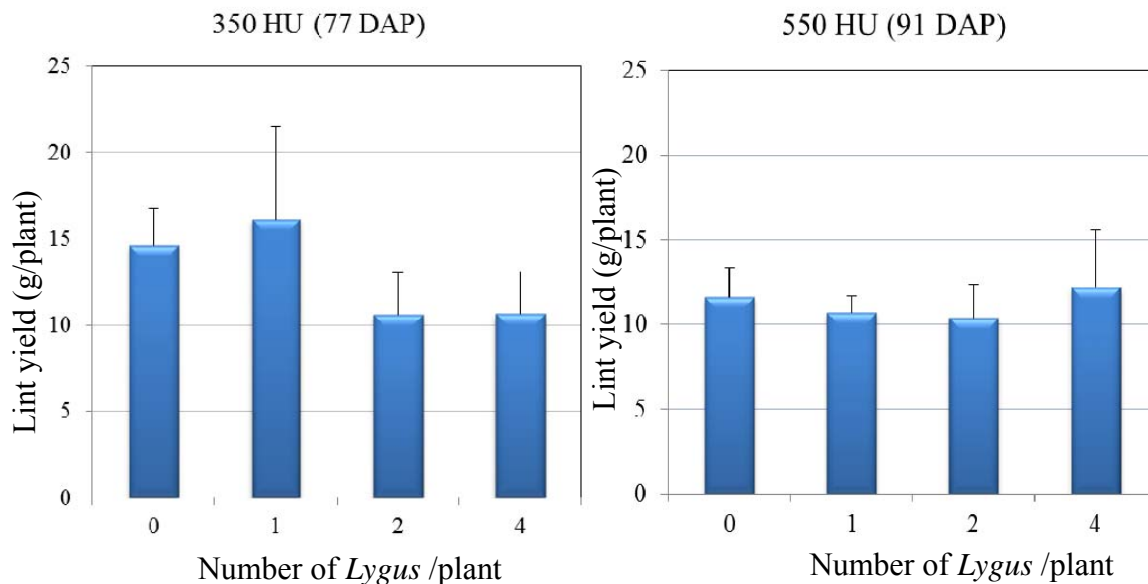


Figure 11. Influence of varying levels of *Lygus* infestations on lint yields at two crop phenological stages. Lubbock County, TX, 2012.

Our pre-harvest complete plant mapping (box mapping) data indicated that the cotton bolls from different nodal positions had different levels of contribution to the total final lint yield (Fig. 12). Nevertheless, bolls from the 5<sup>th</sup> to 14<sup>th</sup> nodes cumulatively contributed >95% of the lint yield. Of these 10 nodes, on average, 6-12 node bolls contributed significantly more lint per boll compared to the bolls from remaining nodes.

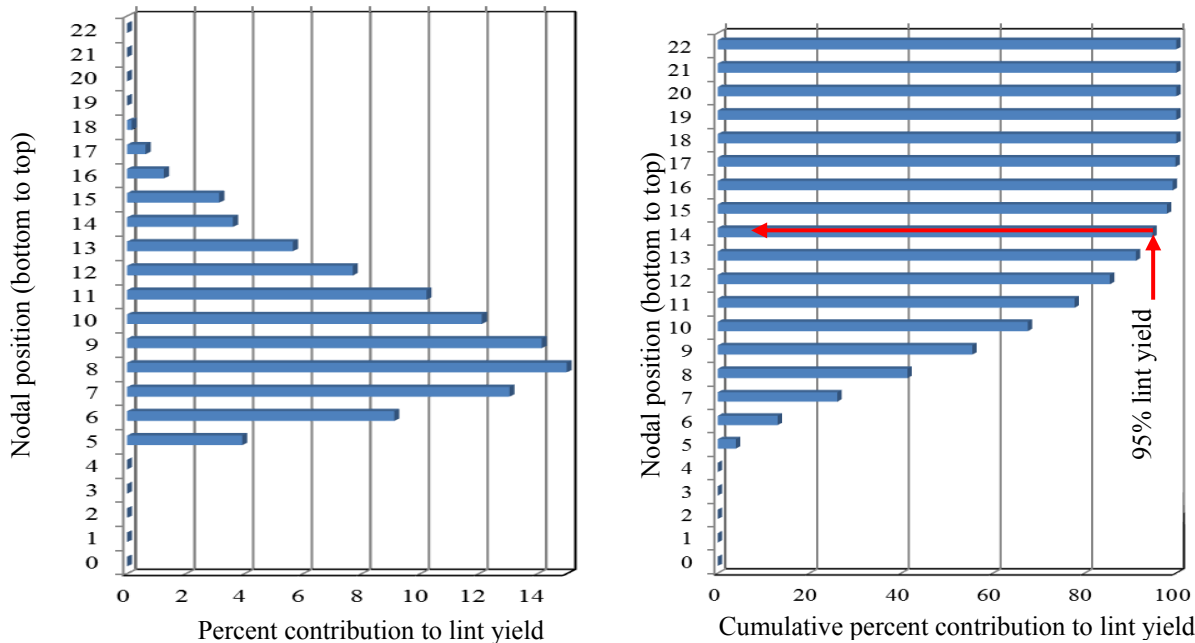


Figure 12. Percent final lint yield contributions of cotton bolls from different nodal positions. Lubbock, TX, 2012.

### 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 two weeks old bolls. Cotton boll developmental rates may vary depending on the crop cultivar and crop management system, therefore the interactions between *Lygus* damage potential and other cotton cultivars and various crop management systems need to be investigated to determine the *Lygus* safe boll developmental stages.

### Acknowledgements

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## **EFFECT OF NITROGEN FERTILIZER ON COTTON HOST-PLANT QUALITY AND ITS IMPACT ON ARTHROPOD COMMUNITY**

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### **Abstract**

The relationship between nitrogen fertilizer application in cotton and subsequent changes in lint and seed yield is well-understood. However, little research has been done to evaluate the role of nitrogen fertility in arthropod population abundance in cotton, particularly in a high yield potential subsurface drip irrigation production system. Previous work suggests that there exists a non-linear relationship between soil nitrogen availability and cotton aphid abundance in cotton. However, interactions between plant-available soil nitrogen and moisture ultimately determine arthropod population dynamics, at least for the cotton aphid. Also, there is a lack of information on plant parameter values with respect to varying rates of available soil nitrogen in cotton production. An ongoing multi-year comprehensive field study has examined the effect of soil nitrogen (residual nitrogen plus applied nitrogen) on cotton agronomic growth parameters and arthropod abundances under a drip irrigation production system for 10 years. This paper discusses the results of the Year 11 of this ongoing study. Fixed-rate nitrogen application experimental plots, previously established and fixed consisted of five augmented nitrogen fertility levels (0, 50, 100, 150, and 200 lb/acre) with five replications. Although two synthetic pyrethroid applications were applied to all treatment plots during August 2012 to encourage a cotton aphid buildup, observed aphid numbers remained low for 2012. This year's arthropod sampling then focused on community composition and relative numbers of other cotton pest and beneficial species among the five nitrogen treatments. Nitrogen augmentation rates significantly influenced all agronomic parameters evaluated in this study, most with curvilinear relationships. These relationships observed in 2012 are similar to what have been observed in previous years.

### **Introduction**

Second to water, nitrogen fertility limits cotton production yields in the Texas High Plains region. Variable-rate nitrogen management based on soil NO<sub>3</sub> tests may save farmers N fertilizer costs and protect groundwater quality. Nitrogen applications affect cotton plant growth and development that may ultimately affect the diversity and abundance of the arthropod community in cotton fields. A three-year study conducted near Lamesa, Texas, under a limited irrigation production system (Bronson et al. 2006) characterized the effect of nitrogen application on leaf moisture and leaf nitrogen content in cotton and the resulting influence on cotton aphid population dynamics (Matis et al. 2008). Leaf nitrogen content did not vary with nitrogen application method (variable N versus blanket N application of an optimal amount), but both the blanket application and variable-rate application resulted in significantly higher leaf nitrogen contents than were noted in zero-augmented nitrogen plots. As nitrogen application rates were increased from zero to an optimum rate, a significant decrease in both aphid birth and death rates occurred, translating to a decrease in crowding and an increase in aphid survival (Matis et al. 2008). While these data were useful in characterizing cotton aphid population dynamics between zero nitrogen fertility management and optimal nitrogen application rates, the population dynamics of cotton aphids and other cotton arthropods have not been examined under a full range of nitrogen fertility rates (Parajulee 2007; Parajulee et al. 2006, 2008). In particular, no known study has produced plant growth parameters or fruiting profile data pertaining to a spectrum of nitrogen application rates in cotton. The objectives of this study were: 1) to evaluate, in cotton growing under a subsurface drip irrigation production system, cotton crop growth parameters and arthropod population abundance, as influenced by varying N fertilizer application rates, and 2) quantify cotton arthropod abundance, diversity and community structure as a function of nitrogen application rates.

### **Materials and Methods**

The study was conducted on a five acre subsurface drip-irrigated cotton field at the Texas A&M AgriLife Helms Research Farm located two miles south of Halfway, Texas. The field was subdivided into 25 experimental plots, each 16 rows wide x 120 ft long. Five nitrogen application rates (0, 50, 100, 150, 200 lb/acre) had been deployed in a randomized block design to the same experimental units consistently for 10 consecutive years to induce maximum



discrimination among treatment plots through variation in soil residual nitrogen (Fig. 1). The data and discussion reported herein are based on the 2012 crop season study.

<b>0</b>	<b>50</b>	<b>200</b>	<b>50</b>	<b>200</b>
<b>100</b>	<b>100</b>	<b>0</b>	<b>100</b>	<b>50</b>
<b>200</b>	<b>150</b>	<b>50</b>	<b>150</b>	<b>0</b>
<b>50</b>	<b>200</b>	<b>100</b>	<b>200</b>	<b>100</b>
<b>150</b>	<b>0</b>	<b>150</b>	<b>0</b>	<b>150</b>

Figure 1. Helms Farm nitrogen study experimental plot layout following a five nitrogen rate treatment by five replications randomized block design. Annually, each of the 25 experimental plots received one of five nitrogen augmentation treatments including 0, 50, 100, 150, or 200 pounds N/acre. Hale County, TX.

A high-yielding FiberMax cultivar, FM 9063 B2R, was planted at a targeted rate of 56,000 seeds/acre on May 17, 2012. The experiment consisted of a randomized block design with five treatments and five replications. Pre-treatment soil samples (consisting of three soil cores; 0 to 24-inch depth), collected from each of the 25 experiment plots on June 1, 2012 (Fig. 2A). The five side-dress N fertilizer application treatments at rates of 0, 50, 100, 150, and 200 lb N/acre were applied on July 6, 2012 (Fig. 2B). The effect of variable rate of N on crop phenology was visually evident toward the end of the each growing season (Fig. 2C).



Figure 2. A) Annual pre-season soil sampling of 25 sub-surface drip irrigated cotton plots; B) Annually, near the time of first bloom, each plot received one of the long-term assigned side-dressed nitrogen application treatment rates; C) Differential cotton plant growth responses are often visually apparent between plots receiving high and low N application rates. Hale County, TX.

Crop growth and insect activity were monitored during the crop season (Fig. 3A-E). Weekly during most of July and August, numerous plant variables were measured to evaluate the influence of residual soil nitrogen on early plant growth patterns. Examples of collected plant data variables included: 1) plant biomass weight, 2) plant height, 3) total leaf area, 4) percent leaf nitrogen, 5) number of 1<sup>st</sup> position cotton squares/plant, and 6) percent fruit shed. COTMAN SQUAREMAN monitoring was used to monitor early plant growth, and was followed by measurement

of Nodes Above White Flower (NAWF). Foliage-dwelling mobile arthropods were monitored weekly using a ‘Keep It Simple Sampler’ (KISS; Beerwinkle et al. 1997; Fig. 3A) to collect the beneficial and pest arthropods from the upper-canopy foliage. Cotton aphid populations did not develop in 2012, despite two applications (August 16 and 23, 2012) of cyhalothrin intended to stimulate aphid population growth.

Two 10-ft sections were hand-harvested from each plot to obtain cotton lint and seed yields (Fig. 3F). The burr-cotton samples were processed through a research gin at the Texas A&M AgriLife Research and Extension Center, Lubbock, TX. Fiber samples from each experimental plot have been submitted to Cotton Incorporated’s Fiber Testing Laboratory (North Carolina) for HVI and AFIS lint quality parameter analyses.



Figure 3. A) ‘KISS’ modified leaf blower with net for arthropod collections, B) Processing of arthropod samples, C) Measuring leaf chlorophyll, D) Biomass plant collections, E) Leaf area, plant root and shoot biomass measurements, and F) Cotton harvesting (low nitrogen plot visible in the forefront).

### Results and Discussion

Higher levels of available residual soil nitrogen and augmented nitrogen applications significantly affected the cotton plant biomass and height during 2012. Both plant biomass and height increased continuously from 0 lb/acre up to the 150 lb/acre nitrogen applied plots (Fig. 4). Plant biomass was significantly highest in the 150 lb/acre nitrogen applied treatment, but it decreased significantly when an additional 50 lb/acre of nitrogen was applied to the 200 lb/acre N treatment plots. Likewise, the plant height increased constantly from the 0 lb/acre treatment up to the 150 lb/acre treatment, but then leveled off, resulting in similar plant heights to those in the 200 lb/acre plots. These data possibly indicate that the excess application of N fertilizer may negatively affect cotton plant growth parameters.

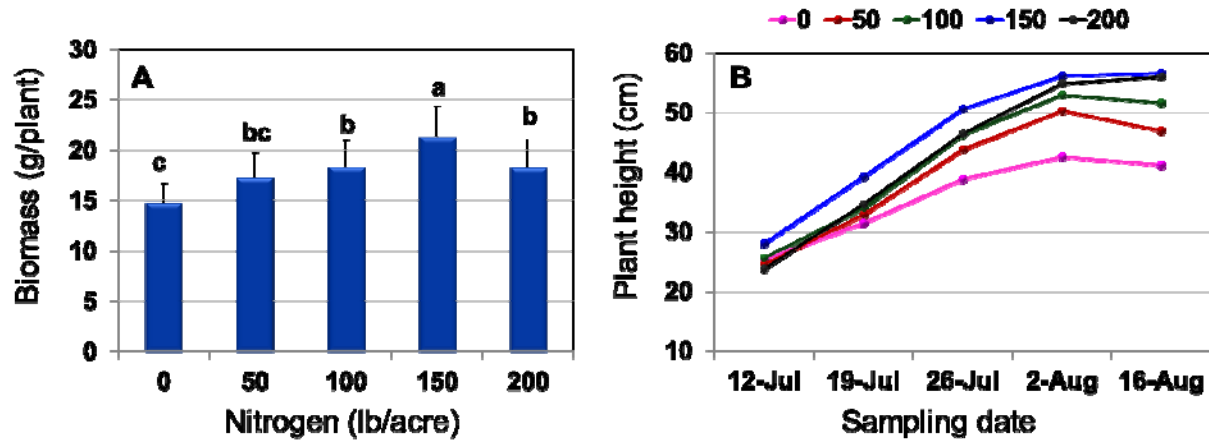


Figure 4. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on plant biomass (A) and plant height (B). Hale County, TX, 2012.

Relationship between N application rate and total leaf area per plant followed similar trends to what was observed with plant biomass and height. Higher N application rates (100, 150, and 200 lb/acre) all resulted in significantly higher leaf area compared to the 0 lb/acre N treatment (Fig. 5A). When compared to the 0 lb/acre treatment, soil N augmentation treatments significantly increased the leaf nitrogen content for all nitrogen rates evaluated (Fig. 5B). High leaf nitrogen content can enhance leaf feeding herbivore populations, especially cotton aphids.

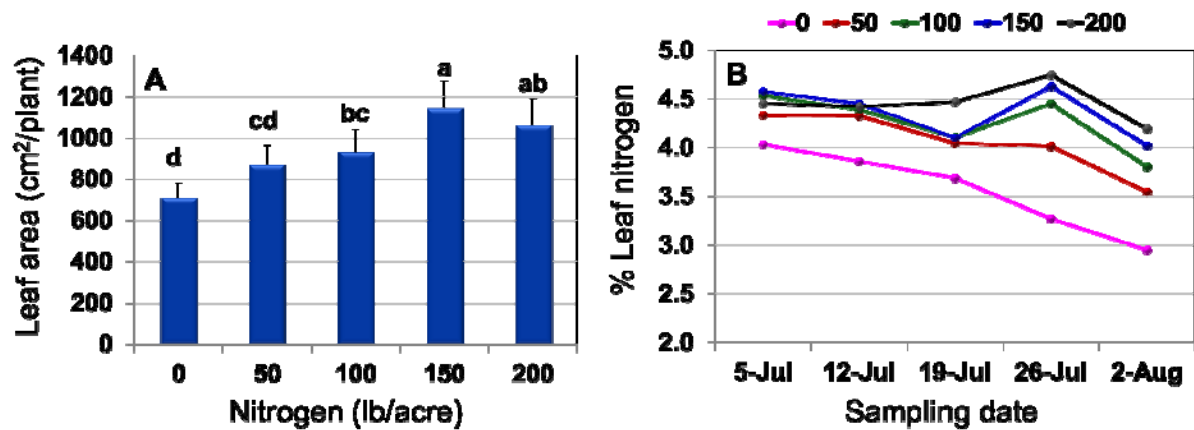


Figure 5. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on leaf area (A) and leaf nitrogen content (B), Hale County, TX, 2012.

Average number of first position squares progressively increased with increased rates of N fertility, with higher N rates (100, 150, and 200 lb/acre) resulting in significantly higher number of first position squares compared to the 0 lb/acre N treatment (Fig. 6A). In the absence of major insect pressure, physiologically-induced fruit shed was low, but higher N rates favored greater fruit retention (Fig. 6B). Although fruit retention was exceptionally higher in this study due to sufficient irrigation water supplied through the drip system coupled with the absence of quantifiable insect pressure, plants would normally shed some excess fruits during the boll maturation phase. Therefore, significantly improved fruit retention at much higher N rate (e.g., 150-200 lb/acre) may not be economically relevant if the irrigation water is not sufficient to support the increased fruit load.

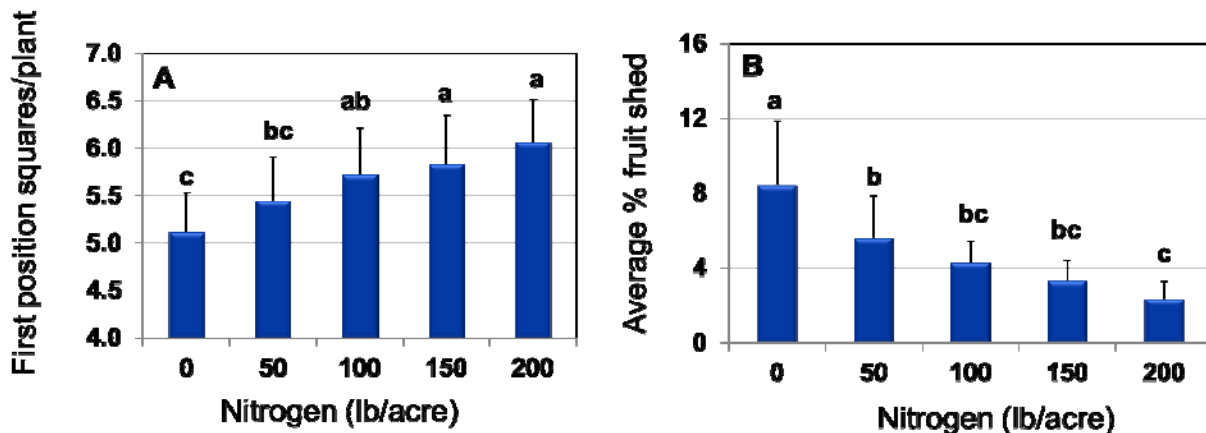


Figure 6. Effect of residual soil N levels and augmented N applications on average square retention (A) and percent fruit shed (B). Data were averaged over five sample dates (Fig. 5B) until crop cut-out. Hale County, TX, 2012.

Arthropod densities were low across all N fertility treatments during 2012. As a result, treatment effect on overall arthropod abundance was not detected (Fig. 7A). With the exception of late July, there were no significant differences in pest abundance across nitrogen treatments (Fig. 7B). In late July, the pest numbers appeared to trend higher in the lower N augmented treatments (0 and 50 lb/acre), yet we speculate that some of this difference may be due to the KISS method of sampling (Fig. 2A) possibly being less efficient on dislodging the pests from the larger and more dense high N treatment plant canopies into the sample device net. Figure 8 illustrates relative numbers of 17 cotton pest and beneficial insects monitored in each of the five N augmentation treatments. While the insect species compositions were similar across all treatments, hooded beetles were the most dominant upper canopy dwelling arthropods in all treatments (Fig. 8). This phenomenon has been the general trend over the last several years of this study. Hooded beetles are generally regarded as predatory arthropods, but we rarely observe significant abundance of prey arthropods to support such a large population of hooded beetles in our system. We speculate that these insects are more omnivorous than predatory in our cotton system.

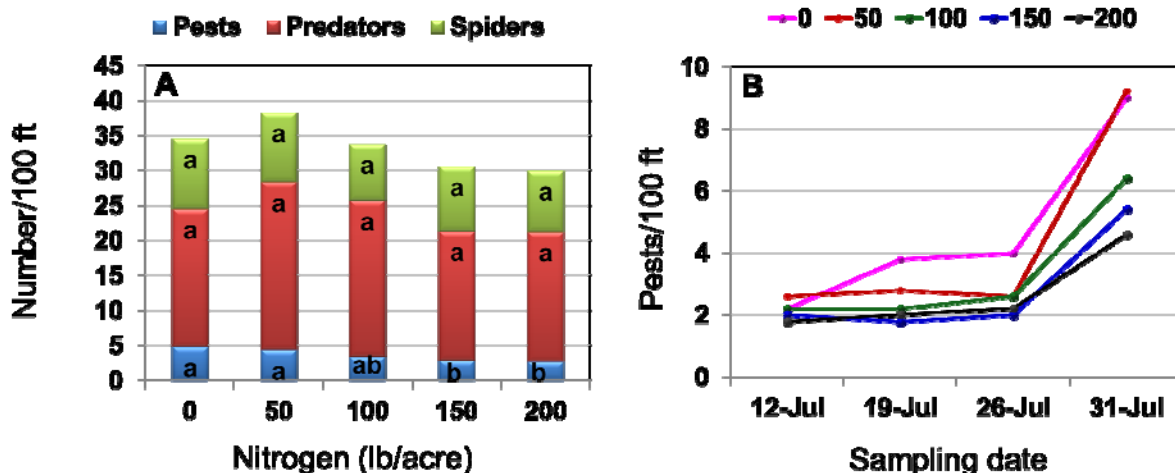


Figure 7. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on total arthropod abundance (A) and pest populations (B). Hale County, TX, 2012.



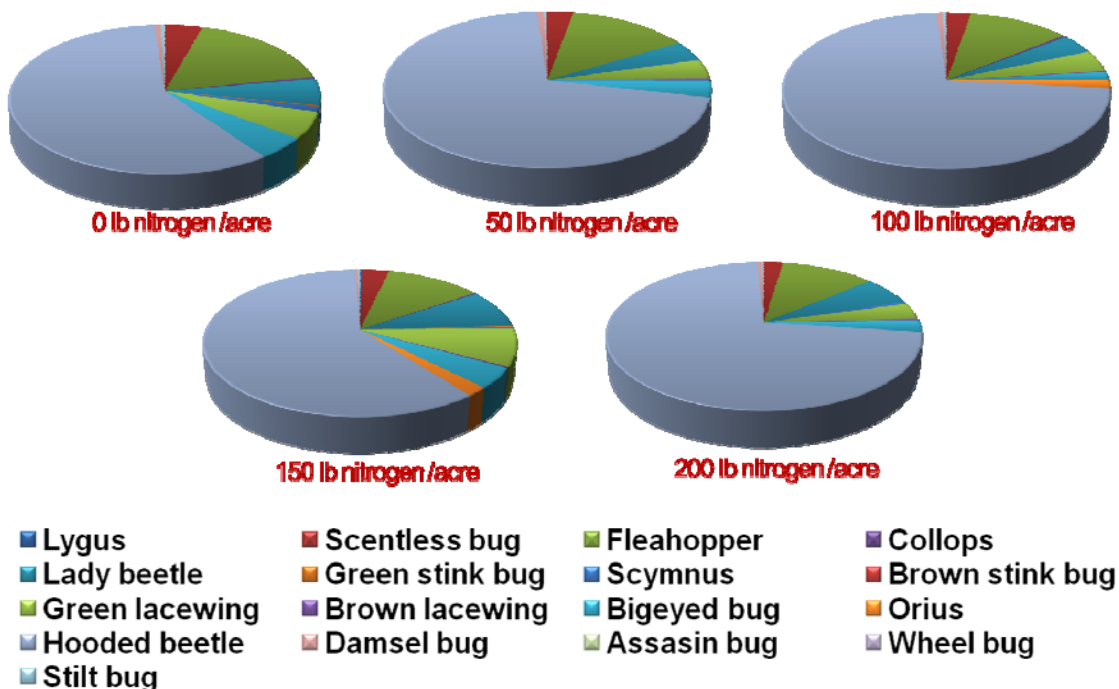


Figure 8. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on seasonal insect species composition in cotton during the crop growing season. Hale County, TX, 2012.

Both cotton lint (Fig. 9A) and seed yields (Fig. 9B) increased with higher rates of nitrogen augmentation. Statistically, lint yields separated into three tiers of similar yields; lowest average yield of 1,001 lb/acre was observed on the plots receiving zero nitrogen augmentation, followed by the 50 and 100 lb/acre mid-range N augmentation rates at 1,492 and 1,625 pounds of lint per acre, respectively. The highest tier lint yields per acre averaged 2,090 and 2,145 pounds harvested from the 150 and 200 lb/acre N treatments, respectively. As expected due to the inherent seed/fiber relationship, observed seed yields followed the same 3-tiered statistical treatment yield pattern as presented above for the lint.

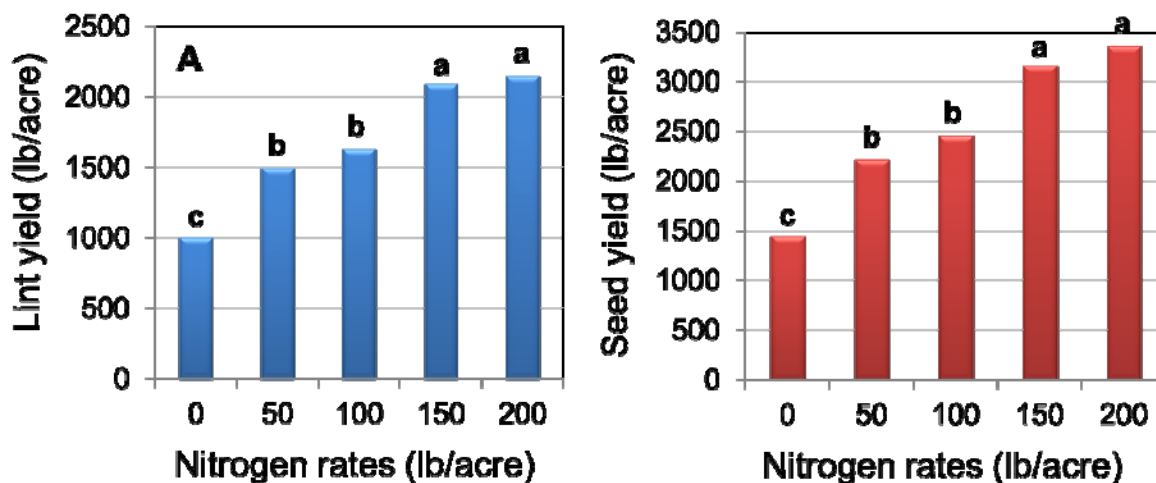


Figure 9. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on cotton lint yield (A) and seed (B) yield. Hale County, TX, 2012.

### Acknowledgments

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## EVALUATION OF ORGANIC PESTICIDES FOR WESTERN FLOWER THIRPS MANAGEMENT IN SEEDLING COTTON: EFFECT ON PLANT PARAMETERS

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### Abstract

Seed treatments and/or foliar applications of various insecticides are common practices for managing the western flower thrips [*Frankliniella occidentalis* (Pergande)] in seedling cotton in the Texas High Plains. However, available options for thrips management in organic cotton production systems are very limited. The efficacy of five organic products: Entrust<sup>®</sup> Naturalyte<sup>®</sup> Insect Control (spinosad microbial 2 oz/A), Aza-Direct<sup>®</sup> Biological Insecticide (Azadirachtin 1.2% 16 oz/A), Bugitol Concentrate (mustard oil 48 oz/A), Safe-T-Side<sup>™</sup> Spray Oil plus Ecotec (0.5 lb + 0.5 qt/A), and NoFly WP Mycoinsecticide (HT6-100; 4 lb/A rate) were evaluated and compared to an untreated control. Treatments were initiated soon after cotton seed germination and applied weekly for three consecutive weeks. In conjunction with treatment applications, cotton was sampled weekly via absolute sampling (plant washing) method. In addition to thrips densities, cotton seedling total (above and below soil surface) biomass, leaf area, and leaf chlorophyll indices were recorded. Due to very high pressure of immigrant adult thrips and subsequent reproduction on cotton seedlings, none of the treatments proved effective when cotton was at the 1 true-leaf stage. Nevertheless, Entrust<sup>®</sup> significantly suppressed thrips populations and kept it below ET when cotton was at the 2-5 true-leaf stage. Plants from Entrust<sup>®</sup>-treated plots had significantly higher leaf areas compared with that in other treatment plots, likely due to lower thrips numbers on Entrust<sup>®</sup>-treated cotton plants. Plant biomass was significantly higher in Entrust<sup>®</sup> and Azadirachtin<sup>®</sup> treatments compared with that in other treatments.

### Introduction

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 key pest in Texas cotton (Greenberg *et al.* 2009) and causes severe damage to cotton seedlings in infested fields, which are generally vulnerable to thrips damage during 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 (Reed *et al.* 2001, Sadras and Wilson 1998, Harp and Turner 1976). Williams (2012) reported that for 2011, Texas had an estimated total of 2,648,547 acres of cotton infested with thrips, which resulted in 28,230 cotton bale loss due to thrips damage.

There has been an increased demand of organic cotton during the recent years. The Organic Trade Association's 2010 Survey report indicated organic fiber sales in the United States grew by 10.4% in 2009 over the previous year, reaching a sales total of \$521 million. In response to rising consumer interest in organic cotton, organic production systems, though a specialized niche representing a fraction of the US cotton market, are garnering increasing cotton producer attention. Organic production poses new and unique problems for cotton producers in terms of arthropod pest and weed control issues as compared to conventional production systems where a variety of agricultural chemicals are available and allowed. This is due to pesticide use limitations imposed by organic, or, as is often debated, "sustainable" guidelines. Insect pest management practices under organic cotton production remain largely unexplored, and although numerous products approved and labeled for acceptable use in organic cotton production systems are available, a paucity of information directly comparing their efficacies against selected target key insect pests, such as thrips, represents an opportunity for beneficial, methodical scientific investigation. In addition, with the recent EPA/industry agreement to cancel all registered uses and remove aldicarb-containing products from the market, which for many years in the past conferred excellent thrips control in conventionally grown cotton, there exists a need for effective alternatives, including those which may have parallel uses in organic production systems. It has been announced that aldicarb will return to the market (new company, product trade name, labels and packaging) for an indefinite period of time with a restricted use classification, but the product has yet to appear on the market. This development will likely be beneficial to conventional cotton production systems, but uses of these



types of pesticides is not allowed under organic cotton production systems, thus other alternatives remain needed at the present time.

Our objective of this study was to evaluate available organic products to determine their efficacy in managing thrips in early-season cotton, with the intention of possibly supplying growers in both conventional and organic production systems with information which may facilitate thrips management decision-making in early-season irrigated, conventional and/or organically produced cotton. In 2011, from among numerous products available for insect control (and successfully applied) in organic crops with potential efficacy against thrips in organically grown cotton, Entrust<sup>®</sup> Naturalyte<sup>®</sup> Insect Control (spinosad microbial), PyGanic<sup>®</sup> Crop Protection EC 5.0 II (pyrethrum), and Surround<sup>®</sup> WP Crop Protectant (kaolin clay) were evaluated with some mixed results (Shrestha et al. 2012, Vandiver et al. 2012). In 2012, we evaluated Entrust<sup>®</sup>, Aza-Direct<sup>®</sup>, Bugitol, Safe-T-Side<sup>™</sup> + Ecotec, and HT6-100 compared to an untreated control.

Over-the-top organic or inorganic pesticide spray applications may affect cotton seedling growth and development in several ways. First, the pest population may be regulated, which may, in turn, alter the level of crop injury, the effects of which may be observed in terms of plant health, growth, and development. Second, the active ingredient of the product, or some other property of the product itself may directly alter plant photosynthesis, respiration, or transpiration, as examples. Third, these phenomena may manifest cumulatively or simultaneously. In addition to evaluating the thrips control efficacies of the products mentioned, a secondary objective of this study involved assessing, under an irrigated organic cotton production system, the effects of various levels of early-season thrips infestation, as actuated by the subject products, on cotton plant vegetative and reproductive development and yield.

In evaluating the two study objectives, it was hypothesized that the selected organic products would suppress thrips densities in irrigated organic cotton differentially, and assuming as much, that the resulting differential thrips densities would facilitate evaluation of subsequent seasonal cotton growth, development, and yield parameters.

### **Materials and Methods**

An irrigated organic cotton field was planted with cultivar FM 958 on 1 May 2012 near Muleshoe, Texas. The crop was cultivated using standard northern Texas High Plains organic cotton production practices. The experiment was deployed in a randomized complete block design with six treatments [five organic products - Entrust<sup>®</sup> Naturalyte<sup>®</sup> Insect Control (spinosad microbial 2 oz/A), Aza-Direct<sup>®</sup> Biological Insecticide (Azadirachtin 1.2% 16 oz/A), Bugitol Concentrate (mustard oil 48 oz/A), Safe-T-Side<sup>™</sup> Spray Oil + Ecotec (0.5 lb + 0.5 qt/A), and NoFly WP Mycoinsecticide (HT6-100; 4 lb/A rate) plus an untreated control] in order to achieve differential thrips densities in treated plots. Treatments were initiated during the week of full cotton seed germination and reasonable plant stand (May 19) and applied weekly for two weeks thereafter (May 25 and June 1). Simultaneously, adult and juvenile thrips were quantified for 7 weeks via “absolute” sampling, which involved whole-seedling immersion of ten plants per plot and subsequent, elaborate processes of sieve “washing” and vacuum filtration, followed by visual quantification under 10X or higher magnification. In addition, leaf chlorophyll, leaf area, plant height, and root length were measured on June 12 and 21. Plant biomass (root, leaf, branch, and reproductive plant parts) was also measured on June 12 and 21. Because of some herbicide drift issues, the influence of organic products on plant phenology and growth behavior was terminated only after two measurements on June 12 and 21. Lint yields were measured in each plot.

### **Results and Discussion**

The overall thrips abundance in our test study site was relatively higher in 2012 than in average years, but it was much higher than in 2011. Under ideal conditions, growth and development parameters would be carefully evaluated in reference to normal or “expected” background thrips densities, yet the 2011 growing season provided the unique opportunity to evaluate the effects of differential thrips densities, as actuated by the selected product treatments, on plant growth parameters under a very low thrips regime. Likewise, the 2012 season afforded us the opportunity to evaluate these products under a heavy thrips pressure situation.

#### **Effect on Thrips Density**

In untreated control plots, thrips densities were expected to increase seasonally due to the potential for thrips reproduction and, further, due to persistent immigration from nearby source habitats. It was certainly true for the

2012 growing season. We had intense thrips pressure during the cotyledon stage of cotton (Fig. 1). The densities were so high that the first spray application of organic products failed to suppress the thrips population and all the experimental plots had thrips densities above ET following the first application of the test products. However, the second application suppressed the thrips densities across all treatments. It appears that some of the adult thrips dispersed out of the field following the second application because the overall thrips densities were reduced including that in untreated control plots. Similarly, the thrips densities declined following the third application of these products. It is, however, noteworthy that only Entrust<sup>®</sup> significantly suppressed thrips populations and kept it below ET when cotton was at the 2-5 true-leaf stage. Due to very high pressure of immigrant adult thrips and subsequent reproduction on cotton seedlings, none of the treatments proved effective when cotton was at the 1 true-leaf stage. Also, average thrips abundance was significantly lower in Entrust<sup>®</sup> treated plots compared with that in the remaining treatments (Fig. 1). This suppression in Entrust<sup>®</sup> treated plots was largely due to reduction in juvenile thrips, clearly suggesting that Entrust<sup>®</sup> suppressed thrips reproduction in seedling cotton.

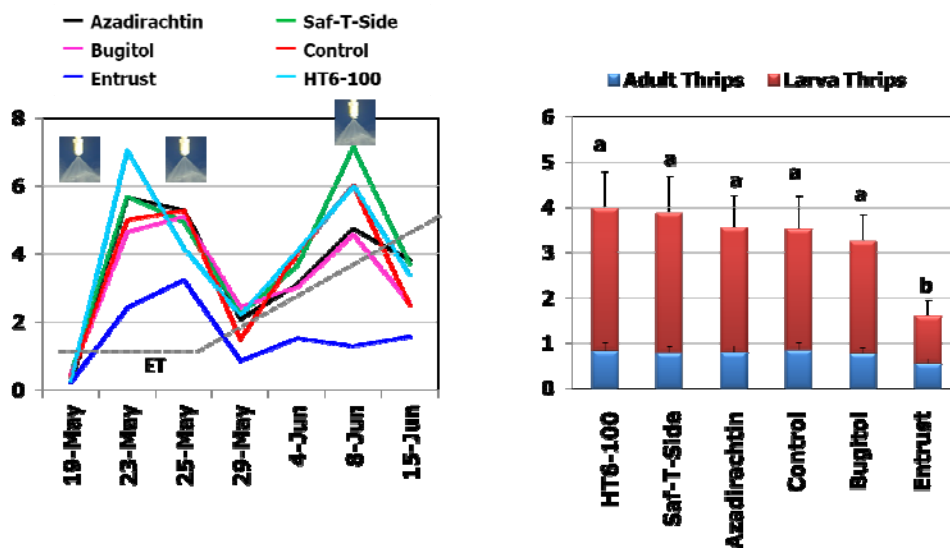


Figure 1. Effect of organic products on thrips abundances; a) Weekly thrips population dynamics, and b) Seven-week average densities of adult and larval thrips.

**Effect on Shoot and Root Lengths**

Two-week average data indicated no significant effect of test products on plant shoot or root growth behavior (Fig. 2). It is possible that the effect of product applications might not have manifested in root/shoot growth by the time the sampling was terminated due to herbicide drift from an outside source. In 2011 study, we had speculated some evidence of possible physiological effect of one of these organic products on plant growth.

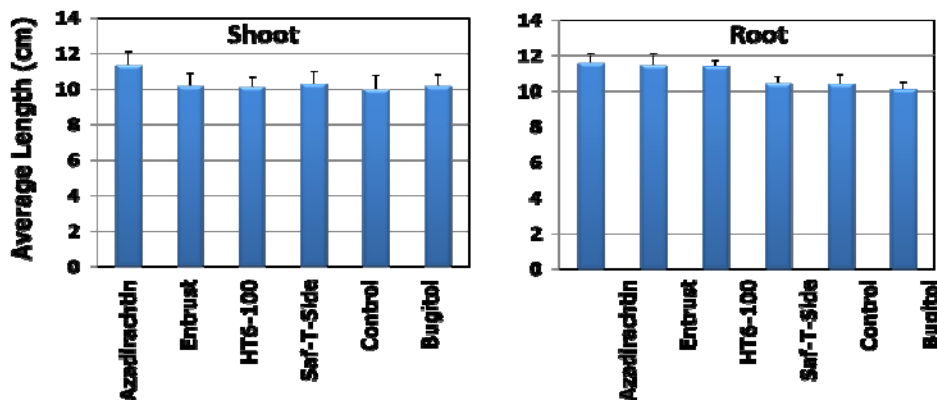
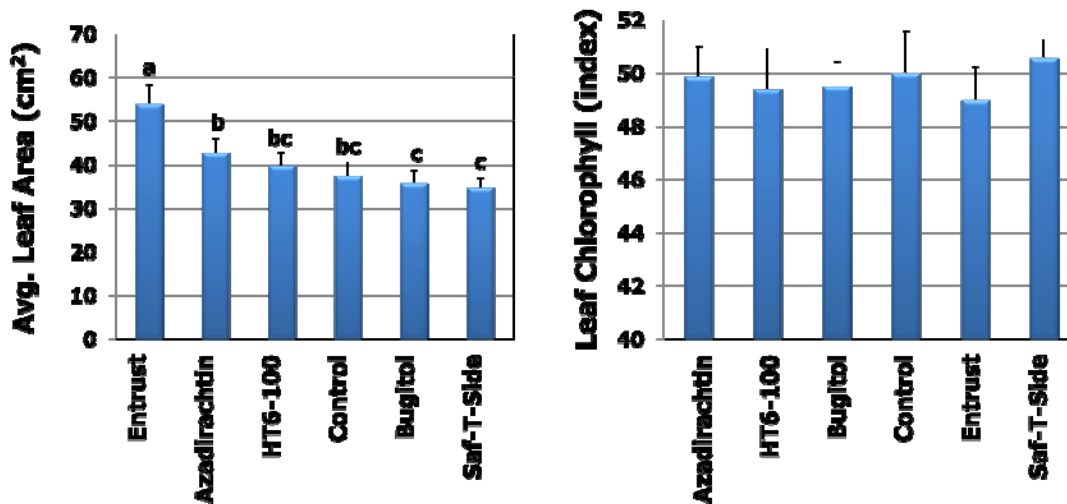


Figure 2. Average shoot and root lengths (n=2 weeks) as affected by selected treatments.

### Effect on Leaf Area and Leaf Chlorophyll

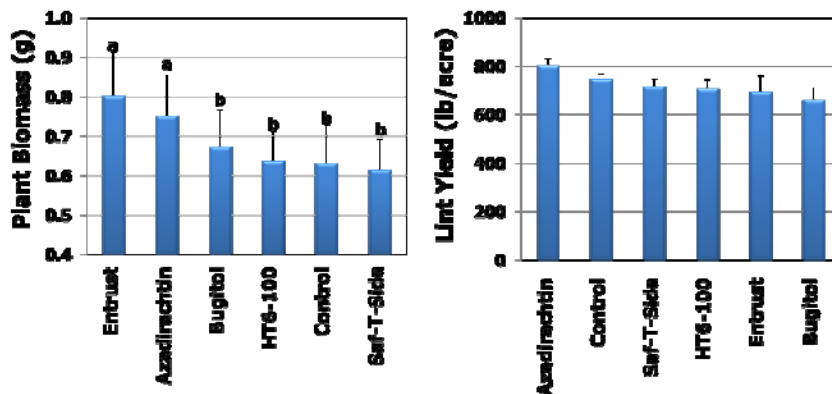
Average leaf area (per plant) varied with organic product treatments, with significantly higher leaf area on Entrust<sup>®</sup>-treated plots, followed by Azadirachtin, with lowest leaf area on Bugitol and Saf-T-Side plots (Fig. 3). The significantly enhanced leaf area on Entrust<sup>®</sup>-treated plots compared with that in other treatment plots may be attributed to lower thrips numbers on Entrust<sup>®</sup>-treated cotton plants (Fig. 1). This phenomenon was also clearly evident in 2011 (Shrestha et al. 2012). Nevertheless, treatments did not significantly influence the leaf chlorophyll content among the six treatments (Fig. 3).



**Figure 3.** Influence of organic product application treatments on average leaf area per plant and leaf chlorophyll content, June 12 and 19, 2012.

### Effect on Plant Biomass

Plant biomass measurements, as assessed, were observed to have followed a pattern generally similar to that of leaf area. That is, Entrust<sup>®</sup> and Azadirachtin treated plots had significantly higher whole-plant biomass compared to the other treatments (Fig. 4). This was congruent with expectations, particularly given the importance of foliage in plant growth and development. A significantly higher dry biomass (Fig. 4) in Entrust<sup>®</sup> plots could be related to lower thrips densities in that treatment (Fig. 1), but there may be other physiological reasons for Entrust<sup>®</sup> and Azadirachtin both to contribute to increased biomass. Lint yield did not significantly vary across six treatments, suggesting that plants must have compensated for any early season variations in plant growth parameters manifested by these products.



**Figure 4.** Influence of organic product treatments on cotton plant biomass (June 12 and 19, 2012) and lint yield.

### Summary

Overall, 2012 thrips pressure was higher than usual in our study. The first pesticide application did not result in thrips suppression, but the second and third applications showed significant reduction in thrips densities for about 4 days. Only Entrust<sup>®</sup> significantly suppressed thrips populations and kept densities below the ET when cotton was at the 2-5 true-leaf stage. Due to very high pressure of immigrant adult thrips and subsequent reproduction on cotton seedlings, none of the treatments proved effective when cotton was at the 1 true-leaf stage. Data analysis indicated that shoot and root length did not vary with organic product treatments. Plants from Entrust<sup>®</sup>-treated plots had significantly higher leaf areas compared with that in other treatment plots, likely due to lower thrips numbers on Entrust<sup>®</sup>-treated cotton plants. However, there was no significant treatment effect on leaf chlorophyll content. Plant biomass was significantly higher in Entrust<sup>®</sup> and Azadirachtin<sup>®</sup> treatments compared with that in other treatments. Even though Entrust<sup>®</sup> significantly reduced thrips densities, all tested organic products resulted in similar lint and seed yields. The 2012 data showed no yield benefit of application of the five organic pesticides evaluated. However, we plan to repeat this study in 2013 with possible addition of new available chemistries for organic production.

### Acknowledgements

We thank W. Owen McSpadden, Caleb Champion, Taylor Person, and Chris Moxon for technical assistance related to data collections for this study. Funding for this study was provided by a grant from the USDA, NIFA Organic Research and Extension Initiative.

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**2012 ANNUAL REPORT****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****Submitted by:**

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

### Project Summary

Thrips are serious seedling cotton pests in Texas. Cancellation of aldicarb, the most commonly used insecticide for thrips management, would directly affect cotton growers, who will subsequently be in critical need of alternative thrips management techniques. The ultimate goal of this project is to generate information which will assist Texas growers in timing thrips management actions for optimal yield potential and minimal economic loss. A pure and healthy laboratory colony of each thrips species is vital to efforts of conducting experiments related to their damage potential and characterization of the cotton plant's response to thrips injury in laboratory and field settings. Thrips mass rearing protocols were evaluated for *Frankliniella occidentalis* and *Kurtomathrips morrilli*. Thrips were reared on four different types of diet including: 1) fresh green beans, 2) a diluted honey solution, 3) honey bee pollen, and 4) fresh cucumber. Green beans with honey and/or pollen supplement was found to be the most suitable and easily available food source for *Frankliniella occidentalis* rearing, but *Kurtomathrips morrilli* colony did not colonize successfully on green beans. Thrips field-cage studies in cotton are challenging and have never been reported from Texas High Plains. Five types of thrips cages were designed to confine thrips to single cotton plants in the field. Thrips cages were optimized for higher thrips containment, survival and better plant quality. Ventilated 1-gallon plastic jar cages were selected for the field evaluations. The thrips cages were deployed to evaluate cotton seedling response to 5 selected thrips densities (0, 1, 2, 4 and 6 adult thrips per plant). Field cages failed to retain the targeted density of adult thrips on the caged plants. None of the cages evaluated were found to be satisfactory for the planned thrips field-cage studies. Thrips were either killed due to overheating inside the cages or the thrips escaped out from the ventilation portions of the cage. The 2013 study will focus on refining the 2012 experimentation to develop thrips field-cages to retain thrips on caged plants with minimal effect on plant quality.

Natural thrips infestations and cotton crop damage were evaluated in an open field experiment. Four selected cotton cultivars (SSG-HQ-212-CT, DP 357/360, FM 1740 B2F and PhytoGen 367 WRF) were planted as four treatments within each of four blocks (replicates) on May 18, 2012. Thrips densities were monitored in all 16 experimental plots via a plant washing technique. Plant response to thrips injury was monitored by measuring plant height, leaf area, root length and total biomass of cotton seedlings from each plot. A greenhouse study was conducted to evaluate the damage potential of *Frankliniella occidentalis*. Cotton seeds (FM 9180 B2F) were planted in 150 small plastic pots in a greenhouse. The cotton seedling response to various level of thrips injury was evaluated using clear plastic cup-cages covered with a paper towel. Five levels of thrips densities (0, 1, 2, 4, and 6 thrips adult per plant) were released onto caged cotton seedlings at the 2-3 true leaf stage and plants were exposed to the thrips for 6 days. The thrips injuries to cotton leaves were recorded using visual damage rating. The 2012 studies would serve the foundation for more refined studies that are expected to be repeated for the next two years.



## 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. Williams (2011) reported that for 2010, Texas had an estimated total of 5,343,620 acres of cotton infested with thrips, which resulted in 8,937 cotton bale loss due to thrips damage. 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 key pest in Texas cotton (Greenberg *et al.* 2009) 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 (Reed *et al.* 2001, 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<sup>®</sup>) discontinuation, their examination may be critical.

Lacking thrips-resistant cotton cultivars, cotton growers primarily use insecticides to control thrips. While several seed treatment options are available, soil-applied aldicarb has, thus far, been the most reliable and common method used for cotton seedling thrips control. Based upon a cumulative risk assessment of aldicarb dangers to humans and the environment, the United States Environmental Protection Agency (EPA) recently announced the receipt of the registrant's request for voluntary cancellation of aldicarb and an agreement with the manufacturer to phase out production and all uses of the compound. As a result, aldicarb is now unavailable for commercial use. Without aldicarb availability, 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 proposed objectives 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. We must use field cages and confine known number of thrips per caged plant to get desired thrips density. The specific objective for the first year of this project was to 1) develop thrips rearing protocols and maintain pure laboratory colonies as a continuous source of thrips for the field and laboratory studies, 2) design thrips-cages for potential field and greenhouse experiments, 3) quantify the level of natural thrips infestations and cotton cultivars growth responses to thrips injury, and 4) determine the damage potential of *Frankliniella occidentalis*. 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.

## **Materials and Methods**

### **Thrips Mass Rearing Protocol Development**

Thrips were collected from a blooming canola field at the Texas A&M AgriLife Research Farm located near Lubbock, Texas. Canola flowers with thrips were brought to the laboratory. The thrips were separated from the canola blooms, predators, and other insects. The collected thrips were sub-divided and reared on four different types of diet including: 1) fresh green beans, 2) diluted honey solution, 3) honey bee pollen, and 4) fresh cucumber. The thrips were reared in an incubator programmed to 77 °F, 12:12 (L:D) photoperiod, and approximately 40-50% RH. Kurtomathrips were collected from producer's cotton field near Seminole, Texas. Cotton leaves with Kurtomathrips adults and immatures was brought into the laboratory and reared on green beans and cotton seedlings.

### **Thrips Cage Design and Field Deployment**

Five types of thrips cages were designed to confine thrips to single cotton plants in the field. Cage types included: 1) transparent plastic cup cage, 2) wire mesh double sleeve cage, 3) opaque plastic cylinder, 4) transparent plastic jar without ventilation, and 5) transparent plastic jar with ventilation (Fig. 1). These cages were placed in a cotton field and then observations for each cage type included the inside cage temperature, plant growth, thrips survival and damage to the cotton leaves. FM 9180 B2F cultivar ("base" seed treatment only) was planted in Field 406 of the Texas A&M AgriLife Research Farm (Lubbock, TX) on May 18, 2012. Three hundred cotton plants were caged using newly designed thrips cages (Design #5 in Fig. 1) on May 25, 2012. This field experiment was deployed in a split plot design with 4 blocks. The main plot factors were 5 cotton growth stages (0, 1, 2, 3, and 4 true leaf stages), and subplot factors were 5 thrips densities (0, 1, 2, 4 and 6 adult thrips per plant). Each experimental plot consisted of a single 40 row-ft length of cotton. Thrips collected from an alfalfa field were released on caged

plant at the variable thrips densities (0, 1, 2, 4, and 6 adult thrips per plant). After 4 days, thrips survival was determined by *in situ* plant washing method (Fig. 2).



Figure 1. Five types of thrips cages evaluated: 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.



Figure 2. Deployment of thrips cages in cotton field for thrips survival pilot study (left) and quantifying thrips by plant washing (right).

### **Thrips Damage Potential Determination: A Greenhouse Study**

We were unable to stabilize or regulate thrips density in field cages; therefore, the cotton plant response to various densities of thrips under field conditions was not determined and the field cage study was abandoned. A greenhouse study was conducted to optimize the thrips cages and develop the method of regulating thrips density on cotton plants held under greenhouse conditions. Cotton seeds (FM 9180 B2F) were planted in 150 small plastic pots in a greenhouse. These cotton seedlings were used to evaluate the suitability of three cup-cage types for assessing the damage potential of *Frankliniella occidentalis* under greenhouse conditions. The primary reason for missing thrips observed in the field cages was speculated to be due to mortality or escape of adult thrips due to the very high heat (>140 °F) inside the cages. Therefore, the greenhouse study was conducted at 80 °F. Clear plastic cup-cages covered with Parafilm®, fine fabric, and paper towels were evaluated. The plastic cup-cages covered with Parafilm were

completely closed and was designed to contain the thrips but thrips could not survive in this cage due to the excessive condensation of water vapor inside the cage. A pilot study on the cotton seedling response to various level of thrips injury was evaluated using the clear plastic cup-cage covered with paper towel (Fig.3). Five levels of thrips densities (0, 1, 2, 4, and 6 thrips adult per plant) were released in caged cotton seedling at the 2-3 true leaf stage and plants were exposed to thrips for 6 days. The thrips injuries to cotton leaves were recorded using a visual damage rating system (Fig.4).



Figure 3. Cotton seedlings grown in greenhouse (left), cup-caged seedlings covered with Parafilm® (middle), and cup-caged seedlings covered with paper towel.

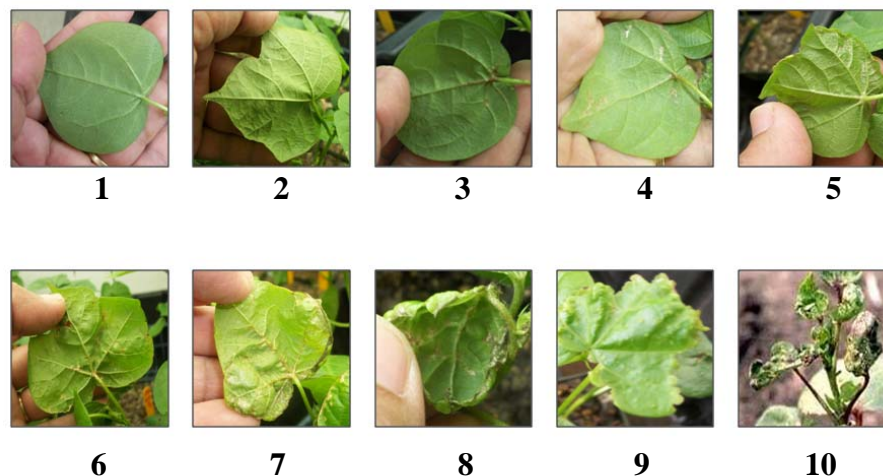


Figure 4. Damage rating system used to score the thrips injury to cotton seedlings.

#### **A Field Study on Cotton Cultivar Response to Thrips Injury**

A field study was deployed in a randomized block design with 4 replications and 4 treatments. Four cotton cultivars (SSG-HQ-212-CT, DP 357/360, FM 1740 B2F and PhytoGen 367 WRF) were planted as four treatments in each of 4 blocks on May 18, 2012. Thrips densities were monitored in all 16 plots using a 10-plant per plot washing technique. Plant response to thrips injury was monitored by measuring plant height, leaf area, root length and total biomass of cotton seedlings from each plot (Fig. 5). Both thrips and plant sampling were performed for two



weeks. The cotton crop was defoliated by spraying FOLEX 6EC at the rate of 12 oz per acre on October 15, 2012. Boll opener (Ethephon-6 at the rate of 32 oz per acre) was applied in the same tank mix with FOLEX 6 EC. Final plant mapping and hand-harvesting were both conducted after the crop was fully prepared for harvest.



Figure 5. Seedlings uprooted for biomass measurement (left). Leaf area and other plant parameters measured (right).

## Results and Discussion

### Thrips Mass Rearing

Although thrips survived on all four diets tested, green beans were found to be the best food source for mass rearing of western flower thrips. The green beans needed to be replaced every two days and eggs on the green beans hatched within 3-5 days. Although both western flower thrips and Kurtomathrips (Fig. 6) survived well on green beans, Kurtomathrips did not colonize on cotton seedling in the greenhouse. Once the development of a pure colony and mass rearing method is developed, the biology and behavior of Kurtomathrips will be investigated. The detail biology and ecology of Kurtomathrips is not available in the published literature and this species is an emerging pest of Texas High Plains cotton (Kerns and Anderson 2012).



Figure 6. Larva, pupa, and adults of the Kurtomathrips (left) and western flower thrips (right).

### **Thrips Cage Design and Field Deployment**

During the daytime hours, very high air temperatures were recorded inside the non-ventilated plastic jar cages compared to the outside ambient temperatures (Fig. 7). In the ventilated cages, inside air temperatures were much lower than in the cages without ventilation; therefore, we decided to add ventilation using finely woven fabrics. Various fabrics were tested in the laboratory for their mesh size and potential thrips retention. Most of the fabrics could not retain thrips inside the cages. The party taffeta fabric, which was available locally, was able to hold most of the thrips in a laboratory test so we decided to use this fabric for making ventilated field cages. Two types of party taffeta (Fabric #1 bright white color, and Fabric #2 yellowish-white) were evaluated in the field. The cages ventilated with Fabric#1 had slightly lower air temperatures compared to the cages ventilated with the off-white Fabric #2 (Fig. 8). Therefore, we decided to use Fabric #1 in our thrips cage design. No thrips survived in the opaque plastic cylinder cages or the transparent plastic jar cages without ventilation, whereas about 50% of the thrips survived in the transparent plastic cages with ventilation when days were cloudy and cool. Thus, these ventilated cages were selected for the field study. High daytime temperatures during the actual experiment (~104 °F) resulted in high thrips mortality in the cages. Only few thrips survived at 4 days post-release. Thrips were released onto a total of 144 caged cotton seedlings and only 16% of the cages (23 caged plants) retained some thrips. Although thrips were recovered in some caged plants, the thrips survival was very low in those cages. On average, 1, 1.6, 1.2 and 1.4 thrips per plant were recorded from plants on which thrips were released at the rate of 1, 2, 4, and 6 thrips per plant, respectively (Table 1). Thrips were found in 12% of the control cages indicating cotton seedling might have been infested with thrips on the day of seedling emergence. Due to extremely high thrips mortality, we were unable to determine the period of time to which the cotton seedlings were actually exposed to thrips. In addition, if the cage conditions were poor for thrips survival, then likely the conditions to encourage cotton tissue feeding were likely also lacking. After the cages were removed, there were high wind/sand storms that completely sand-blasted the test seedlings and all new leaves were heavily damaged. We could not differentiate the thrips damage from sand damage; consequently, this field study was abandoned for the 2012 crop season. We expect to utilize the modified design for the 2013 crop season. Also, multiple releases (daily release of 4X densities for 3 days) of adult thrips were deployed into cages to examine any possible survivorship of adult thrips in these cages to inflict measurable damage to the cotton seedlings. Approximately 10% of the thrips survived for 2 days when shade was provided. When high numbers of adult thrips were released on cotton plants, the adult thrips laid eggs on the cotton seedlings prior to dying and the cotton seedlings had high numbers of thrips larva when observed at 4 days following the first thrips release. Regulating the thrips density in test cages has been a major challenge due to high temperatures inside the cages despite attempts to maximize the ventilation, while at the same time being able to retain the thrips inside the cages.



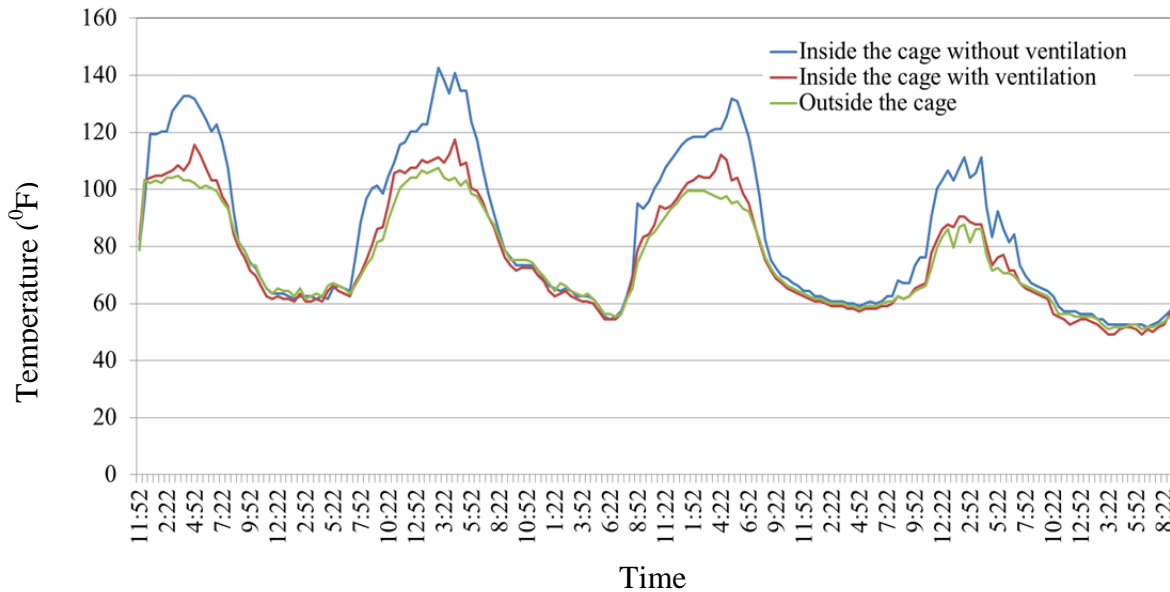


Figure 7. Fluctuation of air temperature inside and outside thrips cages.

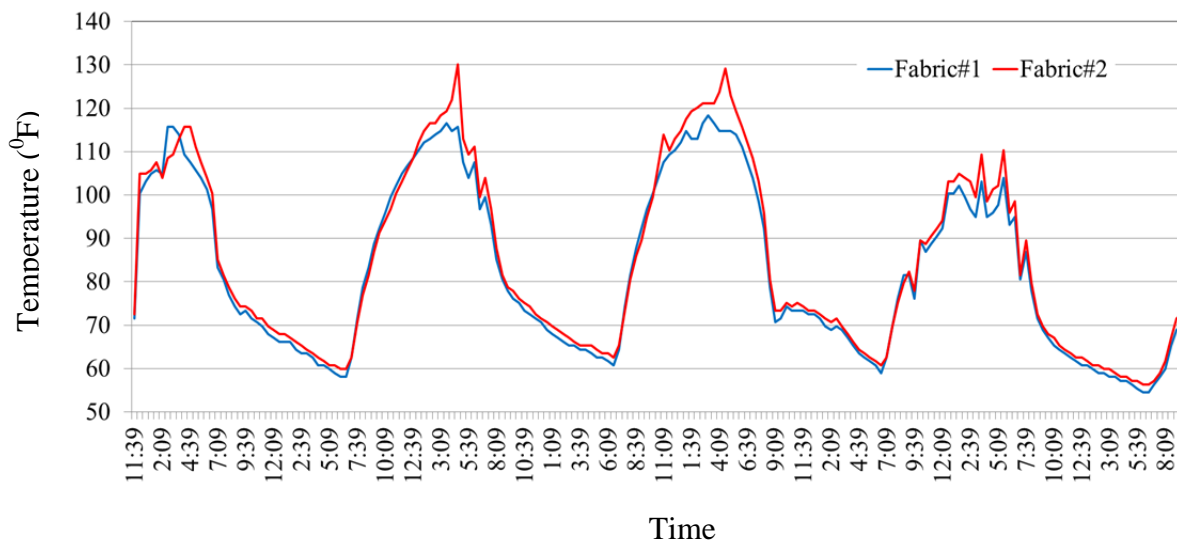


Figure 8. Inside-cage temperature fluctuation as influenced by two fabric types in designing thrips field cages.

Table 1. Survival of adult thrips on caged plants recorded 4 days after field release.

Treatment	No of caged plants	Percentage of plants with thrips	Percent survival*
0 thrips/plant	36	12	-
1 thrips/plant	36	6	100
2 thrips/plant	36	14	80
4 thrips/plant	36	25	29
6 thrips/plant	36	19	23

\*Note: Percent survival is calculated based on those cages which contained thrips excluding those did not contain any thrips.

### Thrips Damage Potential Determination

We were unable to regulate thrips densities in the field cages so the cotton plant response to various densities of thrips in field was not determined and the field cage study was abandoned. A greenhouse study was conducted to optimize the thrips cages and develop the method of regulating thrips densities on cotton plants grown under greenhouse conditions. The reason for missing thrips in field cages was speculated to be due to mortality or the escape of adult thrips as a result of the very high heat (>140 °F) inside the cages. Plastic cup-cages covered with Parafilm® were completely closed (designed to contain the thrips), but thrips could not survive in this cage due to the excessive condensation of water vapor inside the cage. There was partial but very low thrips retention in the plastic cup-caged plants that were covered with fine fabric. Thrips adult were able to bore a hole and escape out through the fine fabric. Whereas, plastic cup-cages covered with paper towels resulted in better retentions of thrips adults and minimal condensation. Higher humidity and accumulation of water vapor due to cotton plant transpiration and soil water evaporation is still a big problem in all three types of cup-cages. The thrips damage rating data showed that thrips infestation at densities of 1, 2, 3, 4, and 6 thrips per plant for 10 days during 3-4 true leaf stage, they caused plant damage scoring 1.4, 2.3, 3.1, and 3.4, respectively. Thrips damage in this experiment was low compared with field infestation. Low thrips damage to cotton seedling could be due to the different growing condition inside the greenhouse or high mortality of thrips in cages. Some thrips damage (score 2.3) was observed even with the control seedlings, suggesting that even in a greenhouse situation, cotton seedlings were not completely free from naturally occurring infestations of thrips. Extra care needs to be taken to grow thrips-free cotton seedlings in the greenhouse for future study.

Table 2. Thrips damage ratings on caged cotton seedlings following 10 days of exposure to varying thrips infestation levels.

Treatment	Cotyledon leaf 1	Cotyledon leaf 2	True leaf 1	True leaf 2	True leaf 3	True leaf 4	True leaf average
0 thrips/plant	1.0	1.0	2.0	2.8	2.5	1.7	2.2
1 thrips/plant	1.3	1.3	1.3	1.8	1.8	1.0	1.4
2 thrips/plant	1.5	2.5	1.8	2.3	3.5	1.5	2.3
4 thrips/plant	1.5	2.3	3.0	4.0	4.0	1.5	3.1
6 thrips/plant	1.5	3.0	3.0	4.0	4.8	1.7	3.4

### **Cotton Cultivar Response to Thrips Injury**

Based on two thrips sampling (May 31 and June 21, 2012) by plant washing method, there were small but significant differences in natural thrips infestation among four cotton cultivars evaluated. PHY 367 WRF had significantly higher level of thrips infestation compared to that in SSG HQ212CT, as evidenced by significant difference in larval thrips densities between these two lines (Fig. 9). There was no significant variation in adult thrips abundance across 4 cotton cultivars evaluated. Thus, the difference in total thrips number was largely due to difference in immature thrips. Significantly lower larval densities in SSG HQ212CT indicated that this cultivar may be relatively less favorable for thrips reproduction and/or immature survivorship compared to other cultivars. In 2013, efforts will be made to expand the list of commercial cultivars to investigate thrips severity variation across wider range of cotton cultivars.

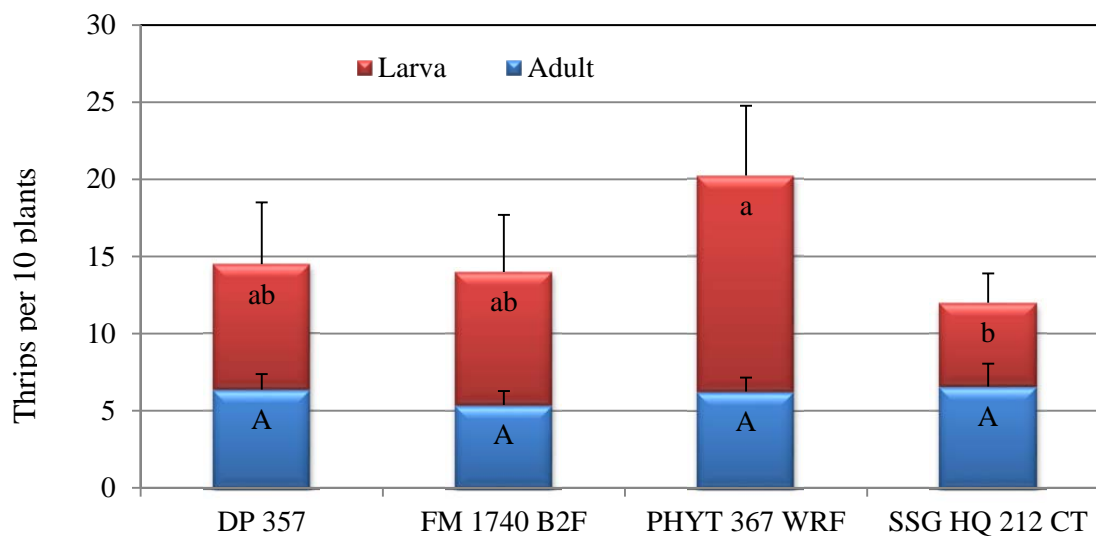


Figure 9. Effect of cotton cultivar on thrips abundance.

A significant variation was observed in plant vigor, especially on leaf growth, across the tested cultivars. Leaf area in DP 357 and FM 1740B2F were significantly higher than that for PHY 367WRF and SSG HQ212CT (Fig. 10). The difference in leaf area may be the inherent characteristics of these cultivars. The low leaf area in case of PHY 367WRF might be partially attributed to the higher level of thrips infestation. The detail response of cotton cultivars to various levels of thrips will be studied in field or laboratory condition by exposing them to various levels of thrips densities in 2013.

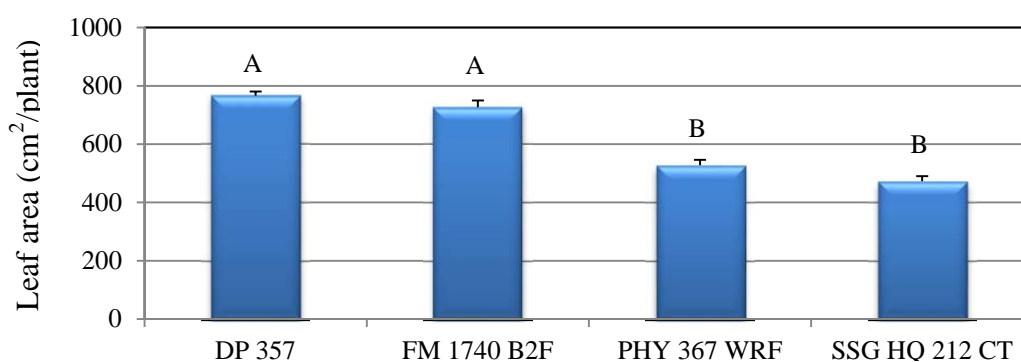


Figure 10. Leaf area measurements on four cotton cultivars at 34 days after planting.

Based on plant biomass measured 34 days after planting, cotton seedling growth patterns were similar in DP 357, FM 1740B2F and PHY 367WRF, but the seedling growth was significantly lower in SSG HQ212CT. Both leaf area and plant biomass data suggest that SSG HQ212CT was the lower vigor cultivar compared with other three cultivars tested (Fig. 11). Even though there was significantly higher level of thrips infestation in PHY 367WRF, it produced significantly

higher amount of lint compared to other 3 cultivars evaluated, owing to its high plant vigor and plant compensatory potential (Fig. 12).

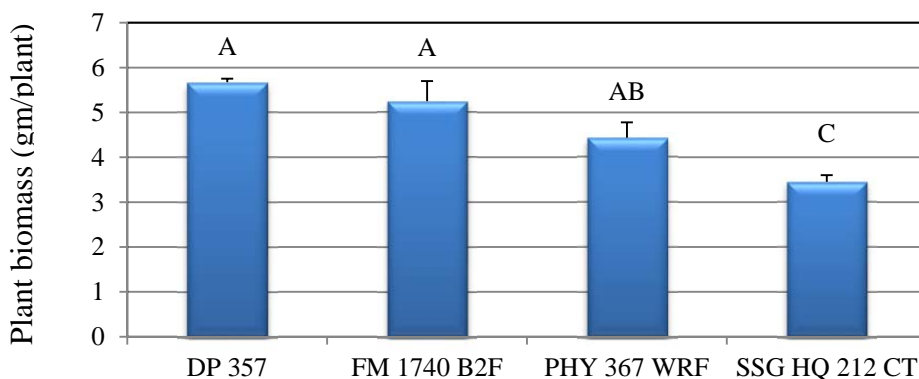


Figure 11. Plant biomass measurement on four selected cotton cultivars at 34 days after planting.

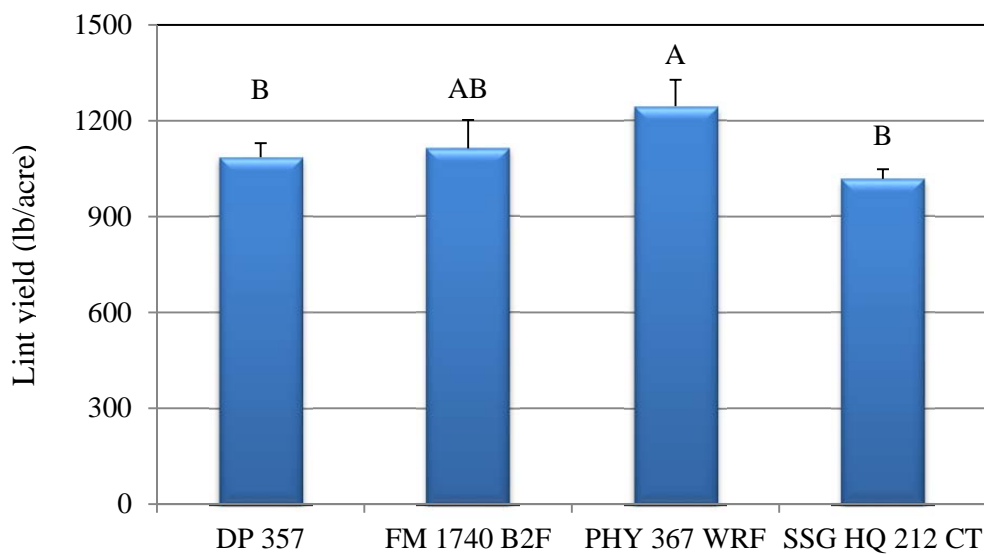


Figure 12. Variation in lint yield potential across four selected cotton cultivars.

### Summary

Mass rearing of western flower thrips can be achieved relatively easily on green beans with honey and pollen supplement and thrips eggs can be harvested using water egg packs. The major challenge of thrips rearing on green beans is the difficulty in avoiding contamination of thrips colony with other thrips species and also to some extent some predators that emerged from infested beans. Further investigation is planned to develop procedures for mass rearing of Kurtomathrips. Unpredictable weather condition, high thrips mortality and thrips escaping from

cages were major problems in conducting *in situ* cage trial. Further research is needed to understand biology and behavior of thrips species (especially *Kurtomathrips*). Further improvement of thrips field cage design and thrips damage quantification procedure is needed for development of dynamic economic thresholds.

### Acknowledgements

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**2012 ANNUAL REPORT****Cotton Incorporated Core Program****Project Number: 08-451****COTMAN Monitoring of Agronomic and Entomological Parameters in the  
Evaluation of Nitrogen Fertility Rate in Drip Irrigated Cotton****Submitted by:**

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## **COTMAN Monitoring of Agronomic and Entomological Parameters in the Evaluation of Nitrogen Fertility Rate in Drip Irrigated Cotton**

### **Project Summary**

The relationship between nitrogen fertilizer application in cotton and subsequent changes in lint and seed yield is well-understood. However, little research has been done to evaluate the role of nitrogen fertility in arthropod population abundance in cotton, particularly in a high yield potential subsurface drip irrigation production system. Previous work suggests that there exists a non-linear relationship between soil nitrogen availability and cotton aphid abundance in cotton. However, interaction between plant-available soil nitrogen and moisture ultimately determines arthropod population dynamics, at least for the cotton aphid. Also, there is a lack of information on plant parameter values with respect to varying rates of available soil nitrogen in cotton production. A multi-year comprehensive field study has been ongoing to examine the effect of soil nitrogen (residual nitrogen plus applied nitrogen) on cotton agronomic growth parameters and arthropod abundances under a drip irrigation production system. Fixed-rate nitrogen application experimental plots, previously established and fixed for five years prior to the initiation of this project in 2008, consisted of five augmented nitrogen fertility levels (0, 50, 100, 150, and 200 lb/acre) with five replications. Each year, soil in each experimental plot was sampled for residual nitrogen analysis immediately prior to planting or before treatment deployment. Rates of applied N exceeding 100 lb/acre resulted in higher residual nitrogen detection during the following season. However, variation in residual nitrogen did not significantly affect early plant growth (plant height, root length, or leaf area). Increased N levels corresponded to increased leaf chlorophyll content, but leaf chlorophyll content was generally consistent across nitrogen levels exceeding 100 lb/acre. Aphid abundance was significantly lower in zero N plots versus other plots every year when cotton aphids were present. In 2010, aphid populations surpassed economic threshold in all N-augmented plots, whereas aphids remained below 50/per leaf, except for 1 week, in zero-N plots. Higher rates of applied N (>100 lbs/A) resulted in significantly higher leaf chlorophyll content compared to that in lower or zero N plots. No arthropod populations develop in 2011 due to extreme temperature and drought. A strong correlation was found between leaf chlorophyll content and lint yield. Nitrogen fertility level influenced fruiting profile and boll maturity. Plants ceased setting additional squares in zero and 50-lb N plots 2 wk into flowering while higher N plots were actively producing squares. Averaged over four years (2008-2011), the zero-N treatment produced the lowest yield (912 lb/acre) and yield increased curvilinearly with each additional 50 lb N added, with highest average lint yield occurring in 150 lb N/acre treatment (1,288 lb/acre). Although two synthetic pyrethroid applications were applied to all treatment plots during August 2012 to encourage a cotton aphid buildup, observed aphid numbers remained low for 2012. This year's arthropod sampling then focused on community composition and relative numbers of other cotton pest and beneficial species among the five nitrogen treatments. Nitrogen augmentation rates significantly influenced all agronomic parameters evaluated in this study, most with curvilinear relationships. These relationships observed in 2012 are similar to what have been observed in previous years.

## Introduction

Second to water, nitrogen fertility limits cotton production yields in the Texas High Plains region. Variable-rate nitrogen management based on soil  $\text{NO}_3$  tests may save farmers N fertilizer costs and protect groundwater quality. Nitrogen applications affect cotton plant growth and development that may ultimately affect the diversity and abundance of the arthropod community in cotton fields. A three-year study conducted near Lamesa, Texas, under a limited irrigation production system (Bronson et al. 2006) characterized the effect of nitrogen application on leaf moisture and leaf nitrogen content in cotton and the resulting influence on cotton aphid population dynamics (Matis et al. 2008). Leaf nitrogen content did not vary with nitrogen application method (variable N versus blanket N application of an optimal amount), but both the blanket application and variable-rate application resulted in significantly higher leaf nitrogen contents than were noted in zero-augmented nitrogen plots. As nitrogen application rates were increased from zero to an optimum rate, a significant decrease in both aphid birth and death rates occurred, translating to a decrease in crowding and an increase in aphid survival (Matis et al. 2008). While these data were useful in characterizing cotton aphid population dynamics between zero nitrogen fertility management and optimal nitrogen application rates, the population dynamics of cotton aphids and other cotton arthropods have not been examined under a full range of nitrogen fertility rates (Parajulee 2007; Parajulee et al. 2006, 2008). In particular, no known study has produced plant growth parameters or fruiting profile data pertaining to a spectrum of nitrogen application rates in cotton. The objectives of this study were: 1) to evaluate, in cotton growing under a subsurface drip irrigation production system, cotton crop growth parameters and arthropod population abundance, as influenced by varying N fertilizer application rates, and 2) quantify cotton arthropod abundance, diversity and community structure as a function of nitrogen application rates.

## Materials and Methods

The study was conducted on a five acre subsurface drip-irrigated cotton field at the Texas A&M AgriLife Helms Research Farm located two miles south of Halfway, Texas. The field was subdivided into 25 experimental plots, each 16 rows wide x 120 ft long. Five nitrogen application rates (0, 50, 100, 150, 200 lb/acre) had been deployed in a randomized block design to the same experimental units consistently for 10 consecutive years to induce maximum discrimination among treatment plots through variation in soil residual nitrogen (Fig. 1). The data and discussion reported herein are based on the 2012 crop season study.

A high-yielding FiberMax cultivar, FM 9063 B2R, was planted at a targeted rate of 56,000 seeds/acre on May 17, 2012. The experiment consisted of a randomized block design with five treatments and five replications. Pre-treatment soil samples (consisting of three soil cores; 0 to 24-inch depth), collected from each of the 25 experiment plots on June 1, 2012 (Fig. 2A). The five side-dress N fertilizer application treatments at rates of 0, 50, 100, 150, and 200 lb N/acre were applied on July 6, 2012 (Fig. 2B). The effect of variable rate of N on crop phenology was evident toward the end of the growing season (Fig. 2C).

Crop growth and insect activity were monitored during the crop season (Fig. 3A-E). Weekly during most of July and August, numerous plant variables were measured to evaluate the

influence of residual soil nitrogen on early plant growth patterns. Examples of collected plant data variables included: 1) plant biomass weight, 2) plant height, 3) total leaf area, 4) percent leaf nitrogen, 5) number of 1<sup>st</sup> position cotton squares/plant, and 6) percent fruit shed. COTMAN SQUAREMAN monitoring was used to monitor early plant growth, and was followed by measurement of Nodes Above White Flower (NAWF). Foliage-dwelling mobile arthropods were monitored weekly using a 'Keep It Simple Sampler' (KISS; Beerwinkle et al. 1997; Fig. 3A) to collect the beneficial and pest arthropods from the upper-canopy foliage. Cotton aphid populations did not develop in 2012, despite two applications (August 16 and 23, 2012) of cyhalothrin intended to stimulate aphid population growth.

Two 10-ft sections were hand-harvested from each plot to obtain cotton lint and seed yields (Fig. 3F). The burr-cotton samples were processed through a research gin at the Texas A&M AgriLife Research and Extension Center, Lubbock, TX. Fiber samples from each experimental plot have been submitted to Cotton Incorporated's Fiber Testing Laboratory (North Carolina) for HVI and AFIS lint quality parameter analyses.

## Results and Discussion

Higher levels of available residual soil nitrogen and augmented nitrogen applications significantly affected the cotton plant biomass and height during 2012. Both plant biomass and height increased continuously from 0 lb/acre up to the 150 lb/acre nitrogen applied plots (Fig. 4). Plant biomass was significantly highest in the 150 lb/acre nitrogen applied treatment, but it decreased significantly when an additional 50 lb/acre of nitrogen was applied to the 200 lb/acre N treatment plots. Likewise, the plant height increased constantly from the 0 lb/acre treatment up to the 150 lb/acre treatment, but then leveled off, resulting in similar plant heights to those in the 200 lb/acre plots. These data indicate that the excess application of N fertilizer may negatively affect cotton plant growth parameters.

Relationship between N application rate and total leaf area per plant followed similar trends to what was observed with plant biomass and height. Higher N application rates (100, 150, and 200 lb/acre) all resulted in significantly higher leaf area compared to the 0 lb/acre N treatment (Fig. 5A). When compared to the 0 lb/acre treatment, soil N augmentation treatments significantly increased the leaf nitrogen content for all nitrogen rates evaluated (Fig. 5B). High leaf nitrogen content can enhance leaf feeding herbivore populations, especially cotton aphids.

Average number of first position squares progressively increased with increased rates of N fertility, with higher N rates (100, 150, and 200 lb/acre) resulting in significantly higher number of first position squares compared to the 0 lb/acre N treatment (Fig. 6A). In the absence of major insect pressure, physiologically-induced fruit shed was low, but higher N rates favored greater fruit retention (Fig. 6B). Although fruit retention was exceptionally higher in this study due to sufficient irrigation water supplied through the drip system coupled with the absence of quantifiable insect pressure, plants would normally shed some excess fruits during the boll maturation phase. Therefore, significantly improved fruit retention at much higher N rate (e.g., 150-200 lb/acre) may not be economically relevant if the irrigation water is not sufficient to support the increased fruit load.

Arthropod densities were low across all N fertility treatments during 2012. As a result, treatment effect on overall arthropod abundance was not detected (Fig. 7A). With the exception of late July, there were no significant differences in pest abundance across nitrogen treatments (Fig. 7B). In late July, the pest numbers appeared to trend higher in the lower N augmented treatments (0 and 50 lb/acre), yet we speculate that some of this difference may be due to the KISS method of sampling (Fig. 2A) possibly being less efficient on dislodging the pests from the larger and more dense high N treatment plant canopies into the sample device. Figure 8 illustrates relative numbers of 17 cotton pest and beneficial insects monitored in each of the five N augmentation treatments. While the insect species compositions were similar across all treatments, hooded beetles were the most dominant upper canopy dwelling arthropods in all treatments (Fig. 8). This phenomenon has been the general trend over the last several years of this study. Hooded beetles are generally regarded as predatory arthropods, but we rarely observe significant abundance of prey arthropods to support such a large population of hooded beetles in our system. We speculate that these insects are more omnivorous than predatory in our cotton system.

Both cotton lint (Fig. 9A) and seed yields (Fig. 9B) increased with higher rates of nitrogen augmentation. Statistically, lint yields separated into three tiers of similar yields; lowest average yield of 1,001 lb/acre was observed on the plots receiving zero nitrogen augmentation, followed by the 50 and 100 lb/acre mid-range N augmentation rates at 1,492 and 1,625 pounds of lint per acre, respectively. The highest tier lint yields per acre averaged 2,090 and 2,145 pounds harvested from the 150 and 200 lb/acre N treatments, respectively. As expected due to the inherent seed/fiber relationship, observed seed yields followed the same 3-tiered statistical treatment yield pattern as presented above for the lint.

This on-going N fertility versus arthropod community structure study will be repeated during the 2013 cotton crop season. In 2013, the previous years of research will be evaluated to determine if the study will be continued beyond 2013.

### **Acknowledgments**

Cotton Incorporated Core Program and Plains Cotton Growers, Inc. have provided funding for this on-going study. During 2012, W. Owen McSpadden, Caleb Champion, Taylor Person, and Chris Moxon provided technical assistance related to data collections for this study.

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<b>0</b>	<b>50</b>	<b>200</b>	<b>50</b>	<b>200</b>
<b>100</b>	<b>100</b>	<b>0</b>	<b>100</b>	<b>50</b>
<b>200</b>	<b>150</b>	<b>50</b>	<b>150</b>	<b>0</b>
<b>50</b>	<b>200</b>	<b>100</b>	<b>200</b>	<b>100</b>
<b>150</b>	<b>0</b>	<b>150</b>	<b>0</b>	<b>150</b>

Figure 1. Helms Farm nitrogen study experimental plot layout following a five nitrogen rate treatment by five replications randomized block design. Annually, each of the 25 experimental plots received one of five nitrogen augmentation treatments including 0, 50, 100, 150, or 200 pounds N/acre. Hale County, TX.



Figure 2. A) Annual pre-fertilizer soil sampling of 25 sub-surface drip irrigated cotton plots; B) Annually, near the time of first bloom, each plot received one of the long-term assigned side-dressed nitrogen application treatment rates; C) Differential cotton plant growth responses are often visually apparent between plots receiving high and low N rates. Hale County, TX.



Figure 3. A) ‘KISS’ modified leaf blower with net for arthropod collections, B) Processing of arthropod samples, C) Measuring leaf chlorophyll, D) Biomass plant collections, E) Leaf area, plant root and shoot biomass measurements, and F) Cotton harvesting (low nitrogen plot visible in the forefront).



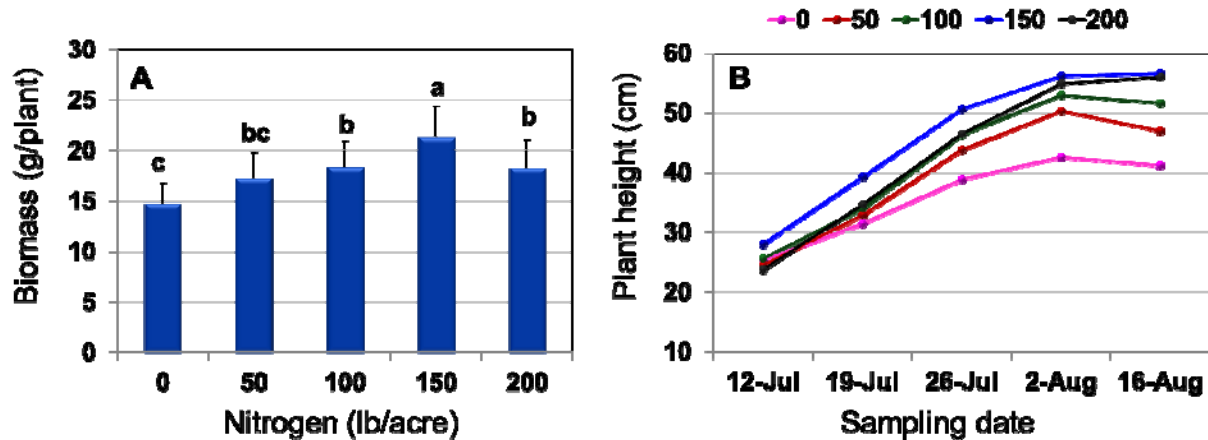


Figure 4. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on plant biomass (A) and plant height (B), 2012. Hale County, TX.

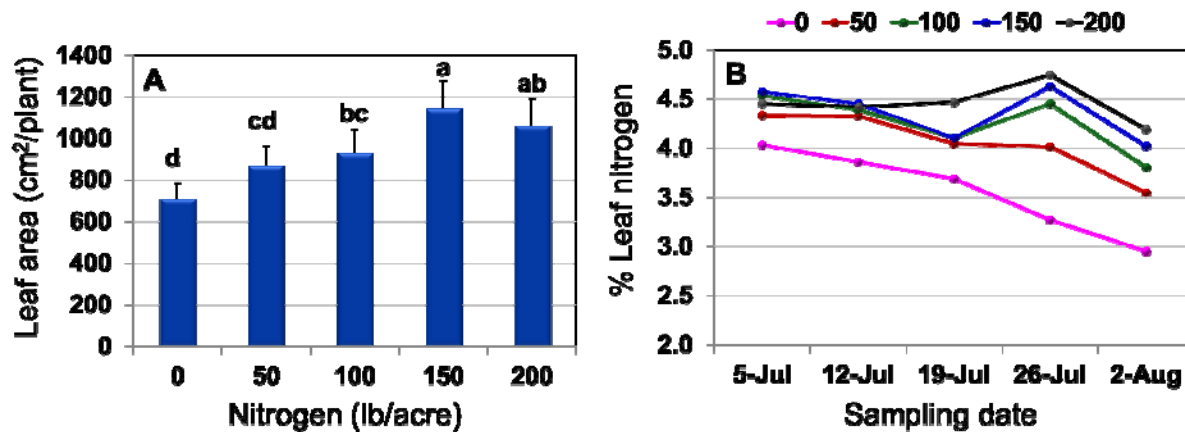


Figure 5. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on leaf area (A) and leaf nitrogen content (B), 2012. Hale County, TX.

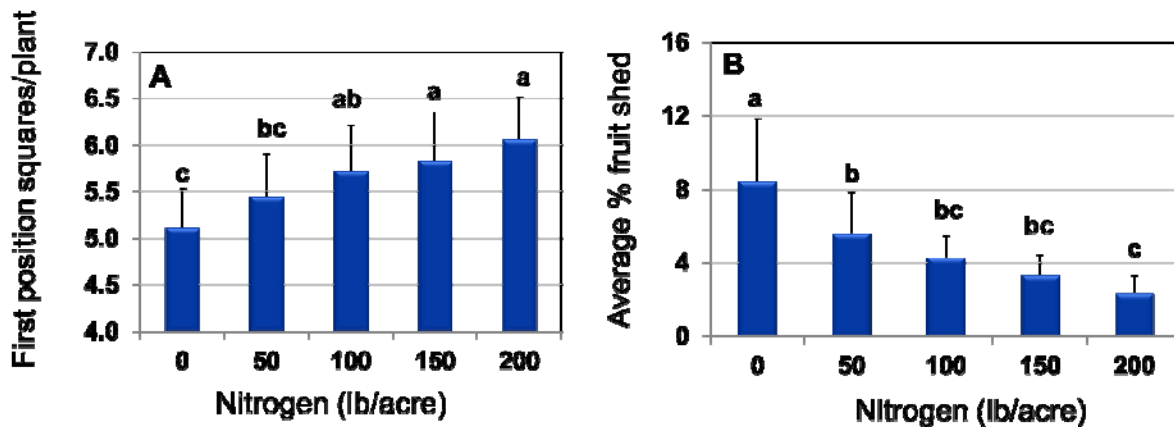


Figure 6. Effect of residual soil N levels and augmented N applications on average square retention (A) and percent fruit shed (B). Data were averaged over five sample dates (Fig. 5B) until crop cut-out, 2012. Hale County, TX.

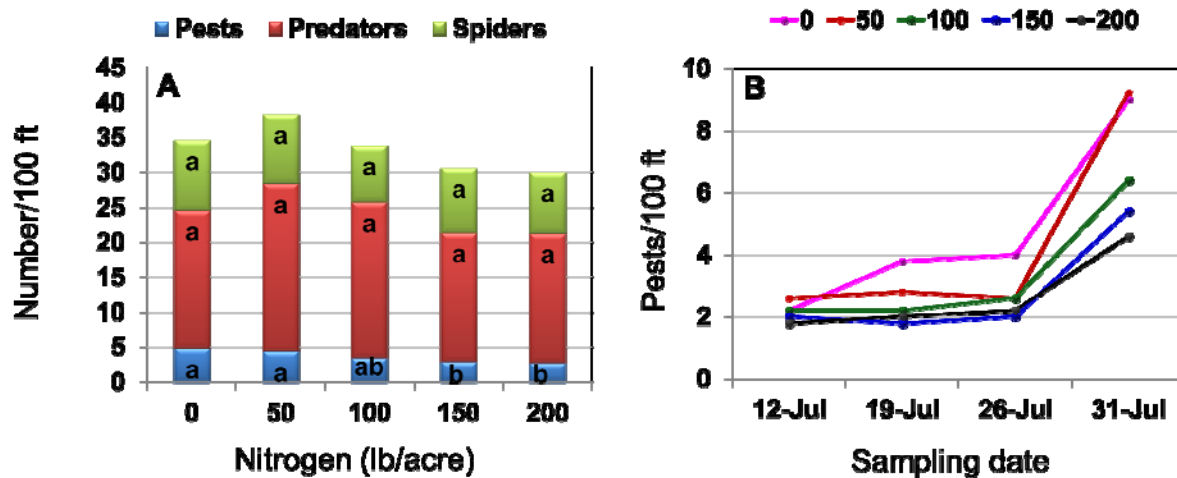


Figure 7. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on total arthropod abundance (A) and pest populations (B), 2012. Hale County, TX.

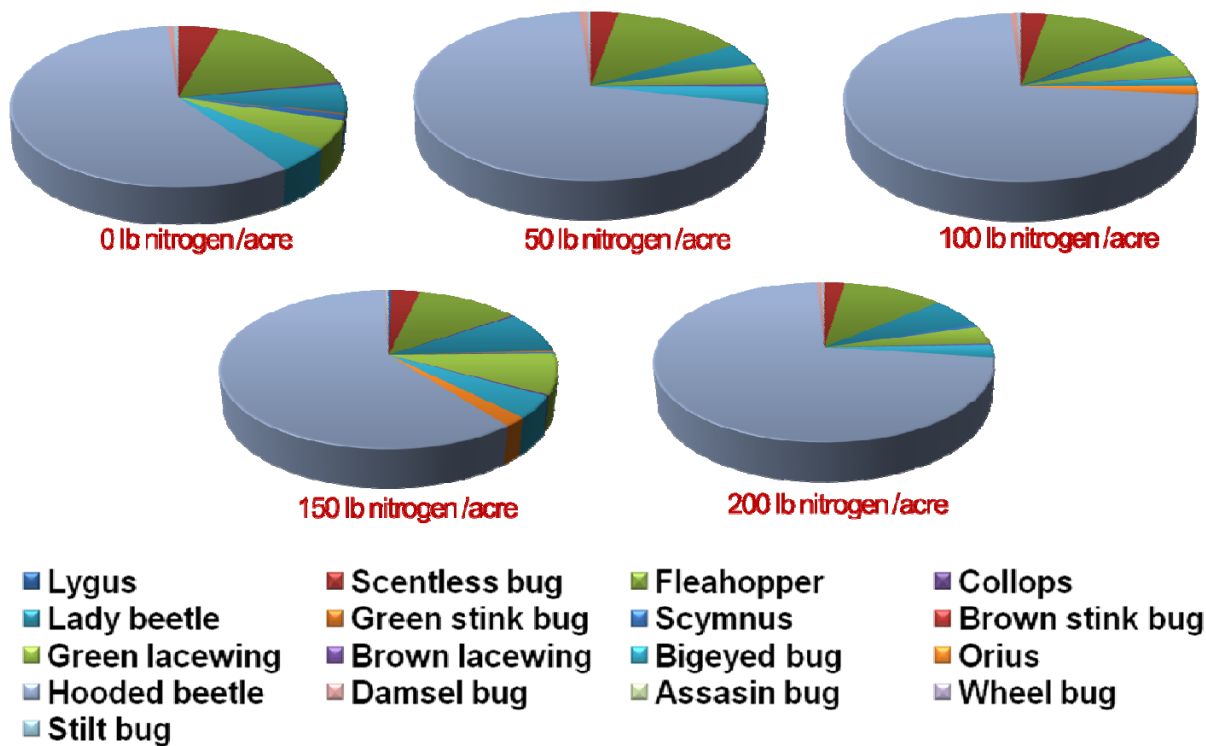


Figure 8. Effect of varied residual soil N levels and augmented nitrogen applications on seasonal insect species composition in cotton during the crop growing season, 2012. Hale County, TX.

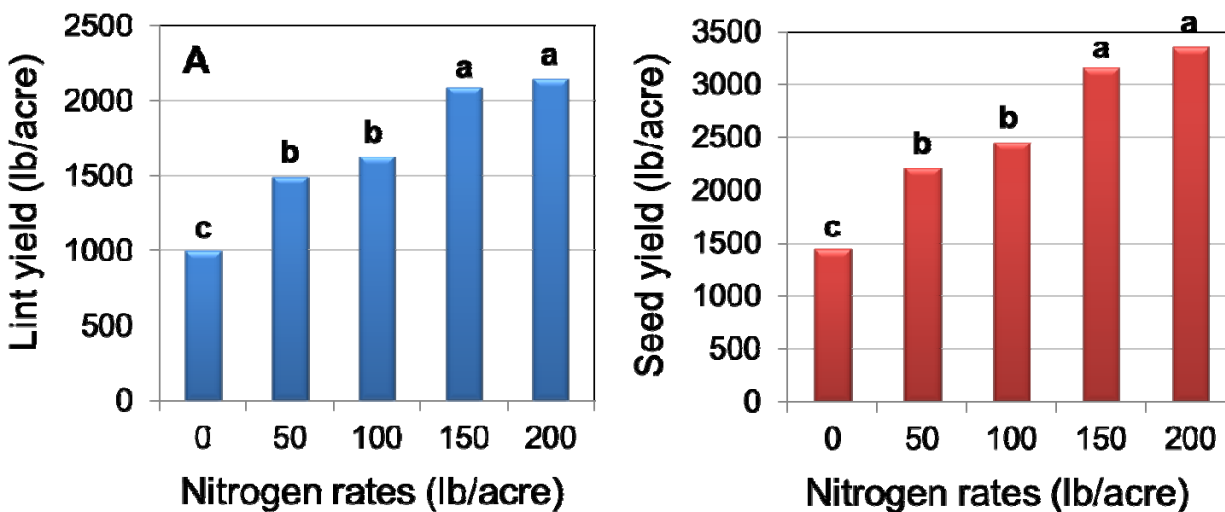


Figure 9. Effect of varied residual soil nitrogen levels and augmented nitrogen applications on cotton lint yield (A) and seed (B) yield, 2012. Hale County, TX.

**2012 ANNUAL REPORT (YEAR 1)**

**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 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 also 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* management has been developed for Texas High Plains cotton and our current management is based on thresholds from other states. This project aimed to conduct a comprehensive threshold study for *Lygus* bugs.

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. In 2012 study year, boll damage potential of *Lygus hesperus* was determined in a no-choice cup-cage study. Ten cohorts of cup-caged single bolls (10-20 days old) were each exposed to a *Lygus* adult for 48 hours and the boll damages were quantified. After bolls reached 16 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 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 53% 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. There were no significant yield differences between control plants and *Lygus* infested plants when plants were first infested with *Lygus* bugs at 550 HU after first flower. 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. With these series of multi-year field studies, we hope to characterize the relationships between cotton boll maturity and *Lygus hesperus* infestations as well as to develop a *Lygus* economic threshold for Texas High Plains cotton. This will improve *Lygus* management recommendations and the project outputs will be directly useful in making *Lygus* management decisions by cotton growers in Texas.

## 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 2011). *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 2010, a 3.9% reduction in U.S. cotton yields was attributable to arthropod pests – 0.68% due to *Lygus* species, which was ranked third among other yield-reducing pests (Williams 2011) 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 2010 (Williams 2011). *Lygus* can cause severe cotton square loss, anther damage, and seed damage depending upon which 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 a 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. Since 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, 4) quantify the yield loss caused by 4 different levels of *Lygus* infestations (0, 1, 2 and 4 *Lygus* adults per plant), and 5) determine the overall yield contribution of cotton bolls from different nodal positions. 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 located near Lubbock, Texas. On May 18, 2012, cotton cultivar ST 5458B2RF was planted on 40-inch spaced rows of a furrow-irrigated field. The targeted seeding rate was 56,000 seeds per acre. On June 2, 2012, the entire test was treated with Orthene<sup>®</sup> 97S for thrips at the rate of 3.0 oz per acre and with Cornerstone Plus<sup>®</sup> herbicide (41% glyphosate) at 32 oz per acre for weed management.

The experimental design was a split-plot randomized block with three replications. Ten cotton boll age cohorts (10 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 1<sup>st</sup> cup-caged bolls received about 450 HU, whereas the August 10<sup>th</sup> 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.





Figure 1. Deployment of cup-cages. Lubbock County, TX, 2012.

### **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. On May 18, 2012, cotton cultivar ST 5458B2RF was planted 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, 2012, this study was treated with Orthene<sup>®</sup> 97S for thrips at a rate of 3.0 oz per acre and with Cornerstone Plus<sup>®</sup> herbicide (41% glyphosate) at 32 oz per acre for weed management.

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 bugs/plant]). There were a total of 24 experimental units. Each experimental unit had 8 cotton plants as subsamples (4 used for damage assessment and 4 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, 2012. When the cotton plants reached the target maturity level (350 HU after first flower on August 7, and 550 HU after first flower on August 21), lab-reared *Lygus* were released into the whole-plant sleeve-cages at the rates of 0, 1, 2, and 4 bugs/plant. Again, the *Lygus* adults were initially reared on artificial diet, yet “trained” on fresh green beans and cotton squares for a week before using them for the boll feeding experiment. Cotton plants were exposed to the *Lygus* adults for 6-7 days, after which time, the insects were killed via a pesticide application. Four randomly selected cotton plants from each plot were cut and brought to the laboratory on August 13 and August 21 for the 350 HU and 550 HU plots, respectively. Boll positions, internal and external *Lygus* damage, boll weights, boll diameters, and boll hardness were recorded for all plants from Block 1. For plants from the other blocks, external boll damage, boll weight, and size were recorded. The cotton crop was defoliated by spraying FOLEX<sup>®</sup> 6EC (12 oz per acre) and a boll opener (Ethephon<sup>®</sup> 6; 32 oz per acre) in a tank mix on October 3, 2012. After the crop was ready to harvest, the remaining 4 caged plants from each plot, which had been maintained pest-free, were harvested manually to evaluate the lint yields and fiber quality. 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 ( $\alpha=0.1$ ) and treatment means were compared by LSMEAN procedure.



Figure 2. Deployment of whole-plant cages. Lubbock County, TX, 2012.

## Results and Discussion

### Boll Development vs. *Lygus* Damage Potential

During the 2012 active boll developmental stage of August 1-20, the Lubbock area cotton crop received, on average, 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). 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, bigger and tougher to puncture. When bolls reached an age of 16 days, *Lygus* caused very little seed damage (<2 seeds per boll) that did not result in significant lint yield reductions (Fig. 4). When cotton bolls received >350 HU after first flower (or approximately 16 days of age), 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 and 4).

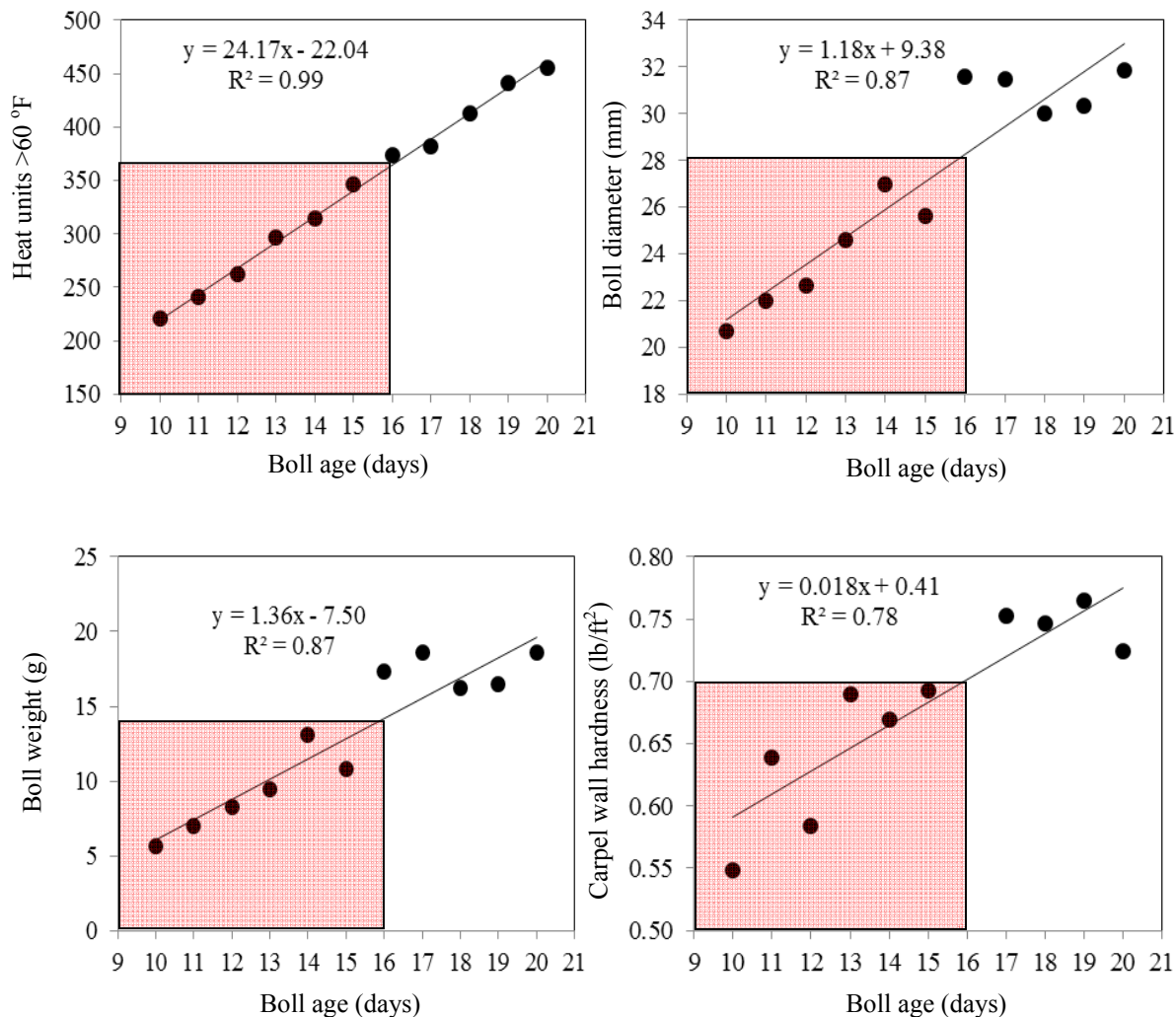


Figure 3. Cotton boll age relationships as associated to heat unit accumulations, boll size, boll weight, and carpel wall hardness. Lubbock County, Texas, 2012.

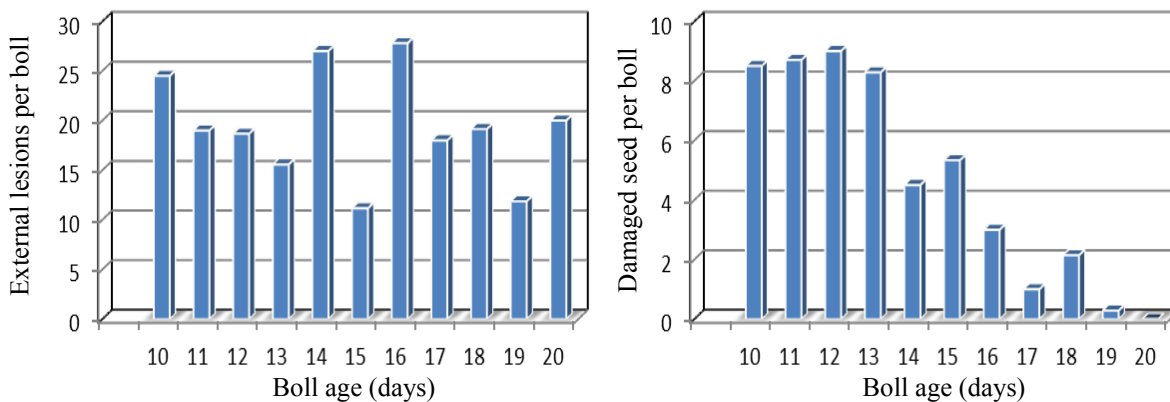


Figure 4. Following 48 hours of feeding by a single *Lygus* adult, boll injury (external lesions and damaged seeds) at various boll ages. Lubbock County, TX, 2012.

### Fruiting Profile

At 350 HU after first flower, average of 56.6% fruit retention was observed, but fruit retention was decreased to 36.7% 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. 5).

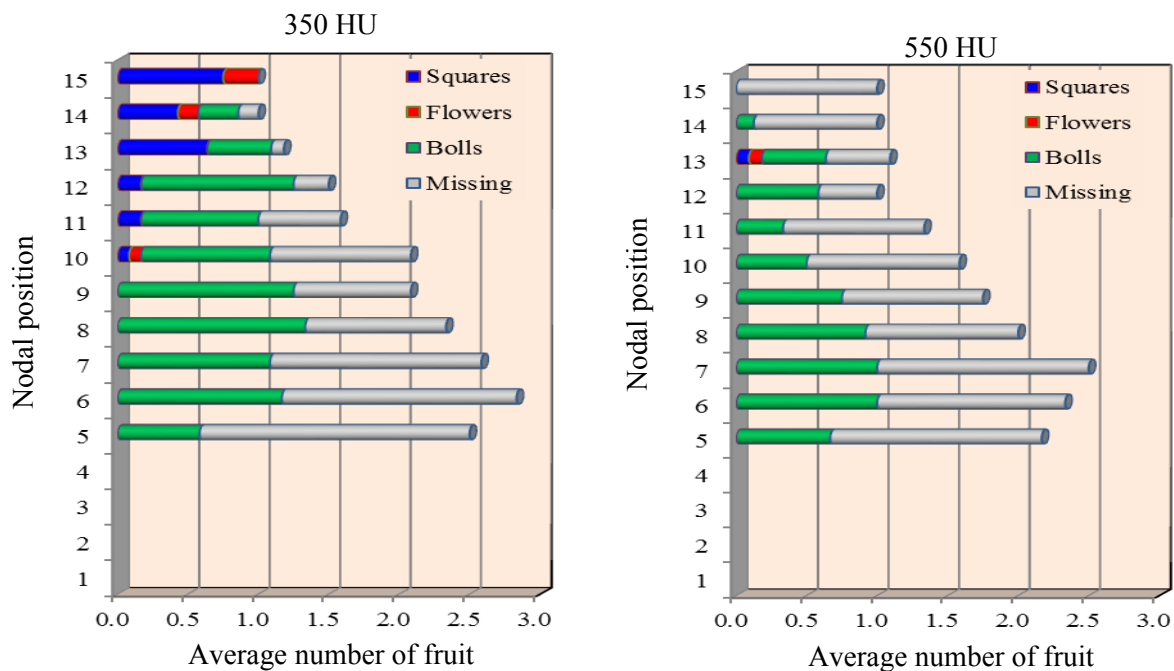


Figure 5. Cotton fruiting profile at 350 and 550 HU after first flower. Lubbock, TX, 2012.

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. 6). When the cotton plants matured from 350 HU to 550 HU, they dropped all of the 4<sup>th</sup> fruiting position and most of the 3<sup>rd</sup> 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.

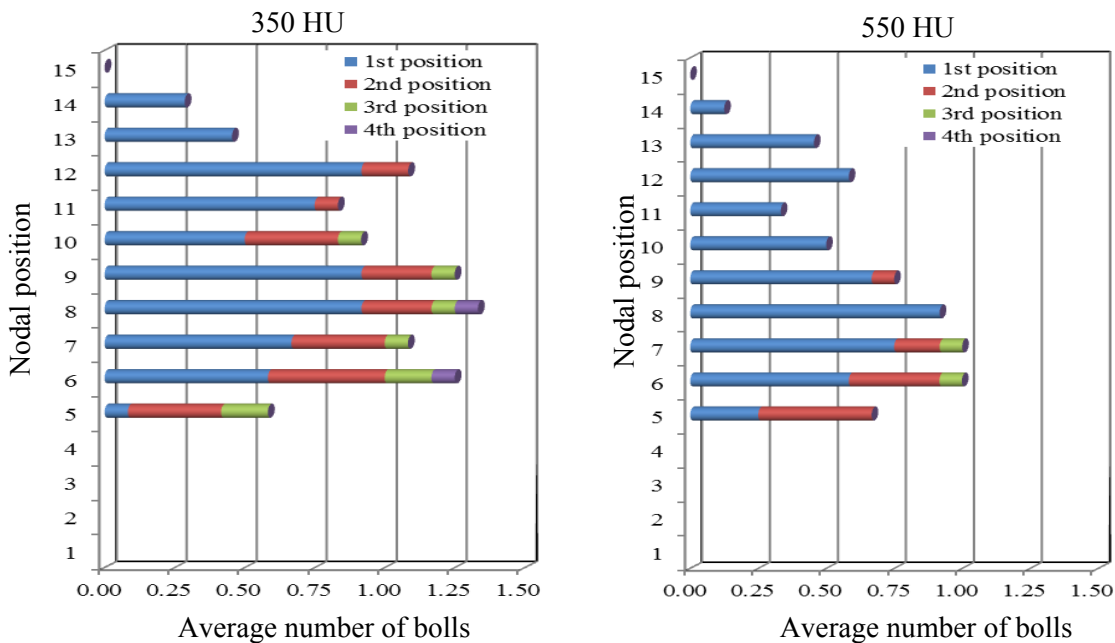


Figure 6. Boll distribution on sympodial branches at 350 and 550 HU AFF. Lubbock, TX, 2012.

**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 (Fig. 7). 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 infested to a mid-season crop (350 HU) compared to that for a late season infestation (550 HU).

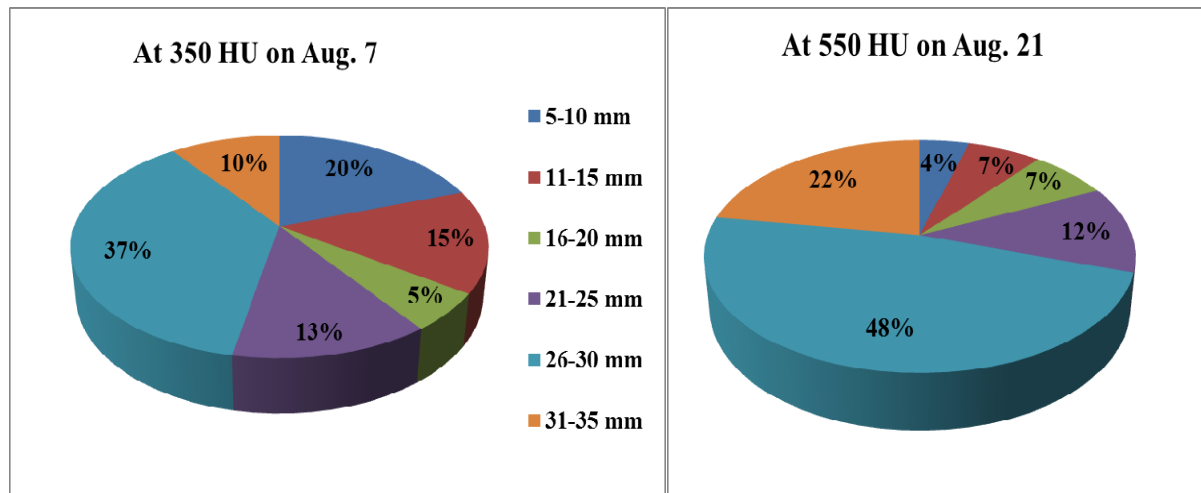


Figure 7. Within-plant boll maturity profile of cotton at 350 and 550 HU. Lubbock, TX, 2012.

For our 2012 cotton crop, within-plant cotton boll maturation profile shows that bolls distributed from the 5<sup>th</sup> to 13<sup>th</sup> nodes (Fig. 8). At the 350 HU stage, the top 4 bolls (from 10-13<sup>th</sup> 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 5<sup>th</sup> to 9<sup>th</sup> 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.

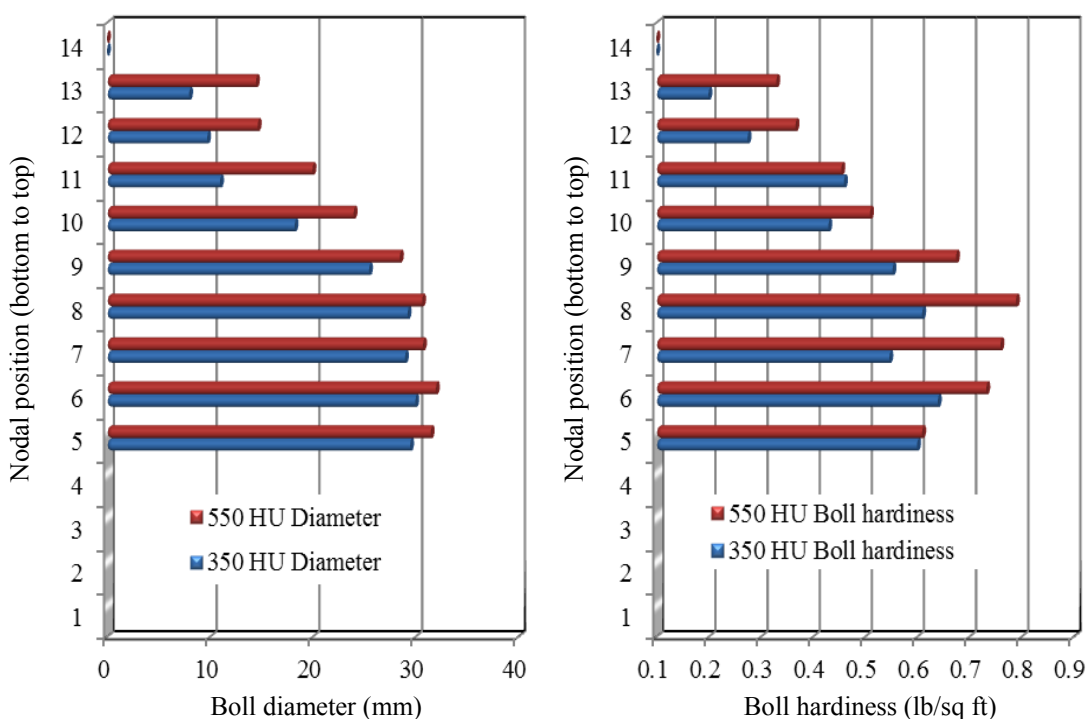


Figure 8. First position boll size profiles of 350 and 550 HU (after first flower) cotton. Lubbock County, TX, 2012.

### ***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.



7). 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 difference 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.

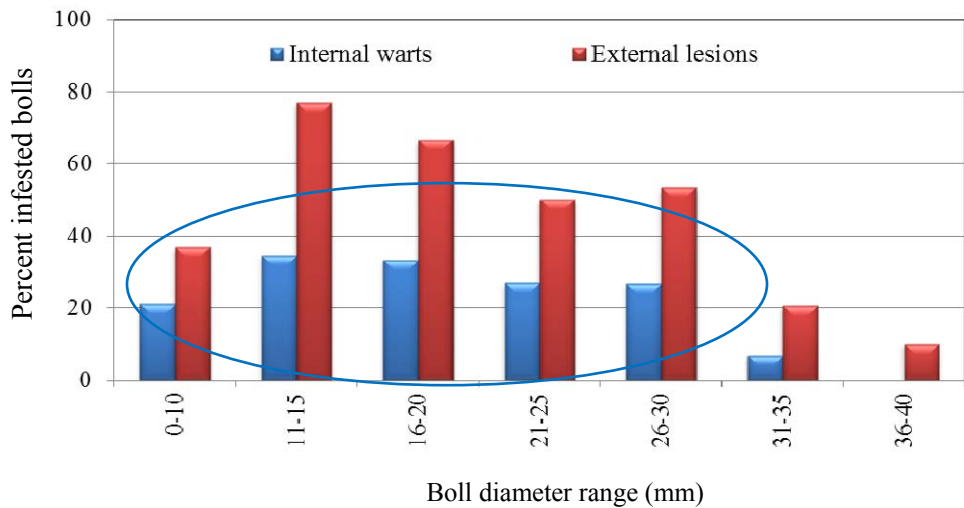


Figure 9. Boll feeding preference of *Lygus* in whole-plant cages based upon the proportion of external and internal boll damage. Lubbock County, TX, 2012.

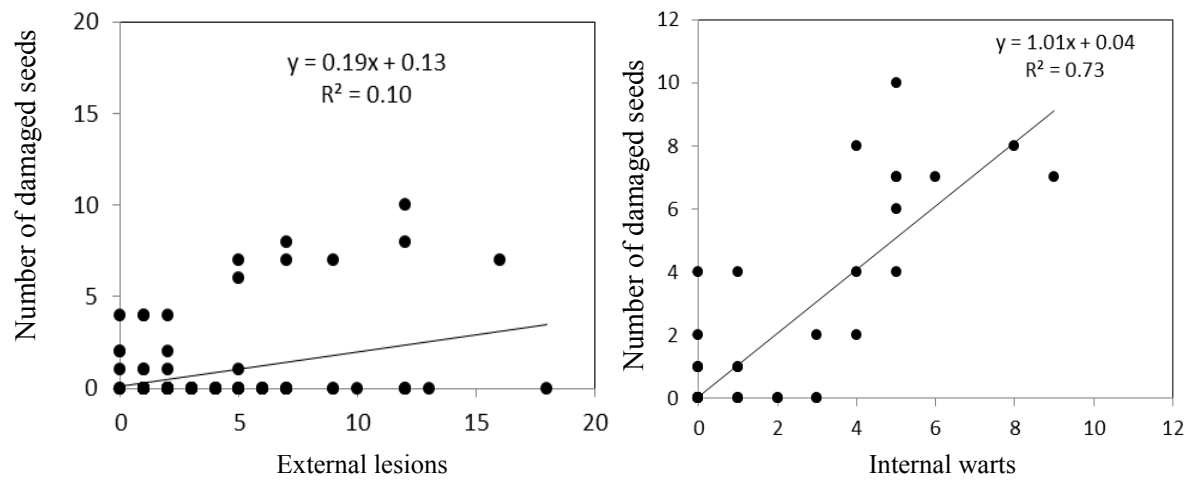


Figure 10. Relationships between the number of damaged seeds per boll and the number of external lesions or internal warts. Lubbock County, TX, 2012.



### Yield Loss

Artificial infestation 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). Although it is expected that the degree of yield loss, due to the same levels of *Lygus* infestation, varies with the crop stage the *Lygus* infestation occurred, it was rather a marked change in the crop tolerance to *Lygus* injury from 350 to 550 HU. 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. More detailed research is needed to characterize the interaction between crop phenology and *Lygus* feeding-induced yield loss. Our continuing project is expected to address some of these issues.

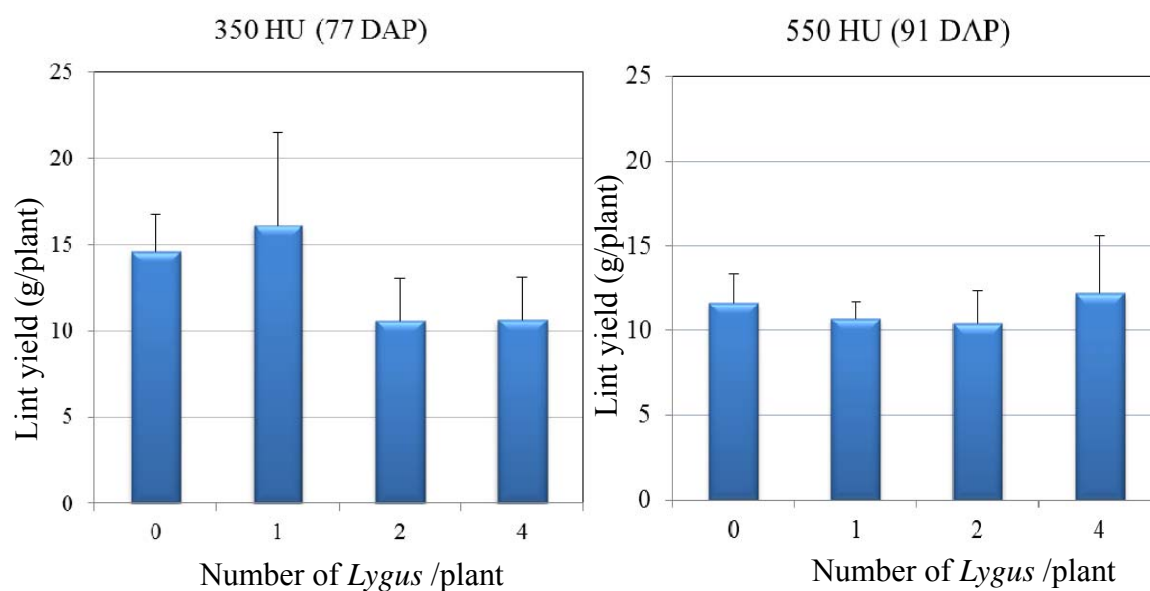


Figure 11. Influence of varying levels of *Lygus* infestations on lint yields at two crop phenological stages. Lubbock County, TX, 2012.

Our pre-harvest complete plant mapping (box mapping) data indicated that the cotton bolls from different nodal positions had different levels of contribution to the total final lint yield (Fig. 12). Nevertheless, bolls from the 5<sup>th</sup> to 14<sup>th</sup> nodes cumulatively contributed >95% of the lint yield. Of these 10 nodes, on average, 6-12 node bolls contributed significantly more lint per boll compared to the bolls from remaining nodes.

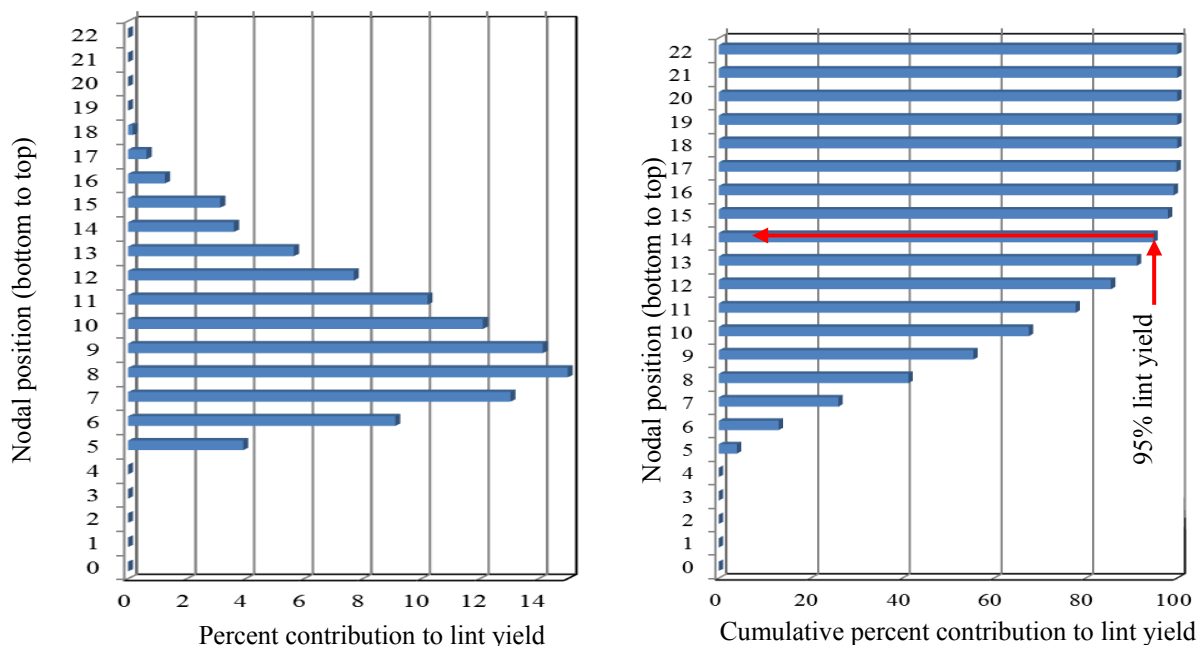


Figure 12. Percent final lint yield contributions of cotton bolls from different nodal positions. Lubbock, TX, 2012.

### 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 two weeks old bolls. Cotton boll developmental rates may vary depending on the crop cultivar and crop management system, therefore the interactions between *Lygus* damage potential and other cotton cultivars and various crop management systems need to be investigated to determining the *Lygus* safe boll developmental stages.

### Acknowledgements

We thank our funding agencies including Cotton Incorporated Texas State Support Committee and Plains Cotton Growers, Inc. The authors also are indebted to Owen McSpadden, Chris Moxon, Caleb Champion and Taylor Person, whose technical assistance in the field was invaluable.

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**Cotton fleahopper and its damage to cotton as affected by plant water stress and insect seasonality**  
 [Cotton Incorporated CORE Program grant: Final Report, 2012 (year two)]

**Project Title:** Cotton fleahopper and its damage to cotton as affected by plant water stress and insect seasonality

**Project Number:** 11-952

**Investigator:** Michael Brewer, Corpus Christi Research & Extension Center.

**Co-Investigator:** Megha Parajulee, Lubbock Research & Extension Center.

**Collaborator:** Charles Suh, Areawide Pest Management Unit, USDA ARS, College Station.

### Summary

We conducted a second year field experiment in 2012 at Corpus Christi and Lubbock, TX to test whether plant water stress, insect seasonality, and plant sensitivity are interacting factors that result in damage differences attributable to cotton fleahopper feeding. Fleahopper populations were less sensitive to plant water stress and more sensitive to plant development stage, which partly explain field to field differences experienced by growers. Although more abundant during bloom, square sensitivity to cotton fleahopper damage and early season opportunity to suppress the population are the primary main considerations in insecticide use. Plant/boll vigor in good soil moisture conditions likely benefits cotton in tolerating cotton fleahopper when cotton fleahoppers occur after squaring. Detection of fleahoppers during bloom in early planted cotton may serve as early warning of cotton fleahoppers in cotton planted later.

### Introduction

Cotton fleahopper, *Pseudatomoscelis seriatus* (Reuter) (Hemiptera: Miridae), can cause excessive loss of cotton squares, resulting in reduced yield and harvest delays (Fig. 1). Cotton fleahopper is a key insect pest of cotton in Texas and Oklahoma, and 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% in Texas (Williams 2000). Damage to individual fields vary from none to extremely high square loss even under similar population pressure.

How is this variability in cotton fleahopper damage explained? This variability is partly associated with cultivar differences and other host plant factors (Holtzer and Sterling 1980, Knutson *et al.* 2009, Barman *et al.* 2011), with timing and magnitude of cotton fleahopper movement from non-cultivated weed hosts to cotton and the stage of cotton development when migration occurs (Parajulee *et al.* 2006), and with physical stressors in particular soil moisture (Stewart and Sterling 1989).

Understanding how these factors contribute to cotton fleahopper fluctuations may allow better estimation of cotton risk from cotton fleahopper damage. Our ultimate goal is to discern when in-season management (i.e., insecticides, irrigation) is most useful to reduce risk to cotton fleahopper damage.



Fig. 1. From left to right, cotton fleahopper adult, nymph, square damage, and a healthy square. Photos provided by authors and Texas AgriLife Research Lubbock and Corpus Christi.

### **Experimental Question and Approach**

We propose that plant water stress, insect seasonality, and plant sensitivity are interacting factors that result in damage differences attributable to cotton fleahopper feeding which are currently difficult to predict. In field testing in 2011 and 2012 at Corpus Christi and Lubbock, drought conditions provided opportunity to assess insect activity in a high contrast of dryland and irrigated conditions (irrigation targeting a range of % ET replacements).

We report here cotton fleahopper and harvest results from Corpus Christi in 2012 (revealing fleahopper/water stress relationship), and plant measurement results in Lubbock in 2011 (very similar in 2012 per data analysis, revealing plant vigor increases under irrigation, graphics in preparation).

### **Corpus Christi**

This location had a split-split plot design with 5 replications. The three water regimes of the main plot were dryland, medium irrigation (scheduled at 75% ET replacement), and high irrigation (90%). Water regimes were applied by surface irrigation through drip tubes. The first split plot was four combinations of 2 planting dates (April 12 and April 30, 2012) and 2 cultivars (Phytogen 367 WRF and Stoneville 5458 B2RF). The last split represented insecticide treatment on one third of the plot (orthene sprayed weekly four times beginning at early squaring) where in-season insect data were collected. The remaining plot was left unsprayed, equally divided into use for in-season data collection and undisturbed for harvest. The plot size was 150 ft by four rows (38 in.), with data taken from the inner two rows. Fleahopper counts (adults and nymphs) were made weekly over a period of 5 weeks after the population exceeded 0.1 fleahopper per plant using the beat bucket technique (20 plants samples per plot) (Fig. 2). Plant measurements included yield, COTMAN (squareman and bollman), and complete plant mapping using PMAP.

### **Lubbock**

Extremely dry conditions resulting in very low cotton fleahopper populations limited the experiment at Lubbock. The plot design was a randomized complete block with 3 replications. Water treatment regimes were dryland, low irrigation (30%), medium irrigation (60%), and high irrigation (90%) applied via subsurface drip tubes. The cultivar planted in the test was Deltapline 1032 B2RF, and the plot size was 4 rows (38 in.) by 100 ft. KISS sampling (a very sensitive sampling protocol) was done in Lubbock which confirmed very low populations of fleahoppers. Plant measurements were total fruit set, percent fruit retention, and boll size by weight taken at 250 DD 60's.



Fig. 2. Beat bucket sampling for cotton fleahopper; visual observations were done previously and correlated well with beat bucket sampling at Corpus Christi, Texas.

## Results

**Corpus Christi: Insect Measurements.** Fleahoppers were detected late with good numbers first occurring June 1, corresponding to mid-bloom for the relatively early planting (42 days after planting) and early bloom for the later planting (31 days after planting). The early planting had much higher populations when fleahopper first appeared in the field, and sprays suppressed the population in the early planting (Fig. 3). As in the previous year, water regime did not affect initial fleahopper densities (Fig. 3). Two weeks later (June 14), cotton fleahopper populations increased as the later planting matured, and the sprays did not suppress this expanding population (Fig. 4).

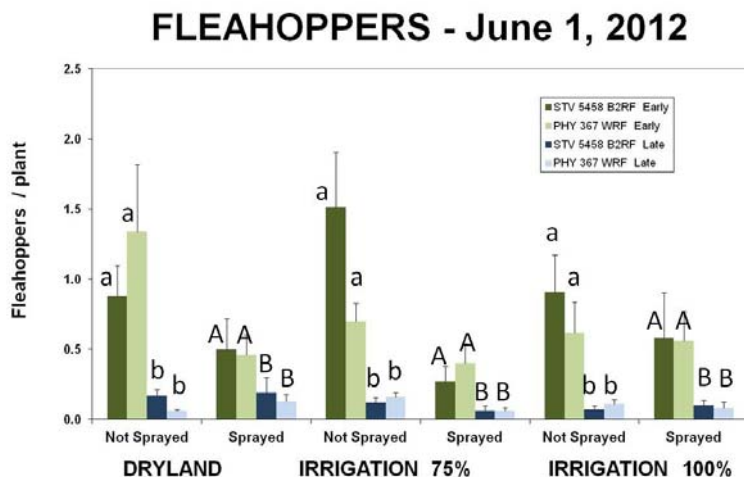


Fig. 3. Cotton fleahoppers per plant on June 1, 2012 on Stoneville 5458 B2RF and Phytogen 367 WRF, sprayed and not sprayed and planted relatively early (April 12, 2012) and late (April 30, 2012) across 3 water regimes (dryland, 75% ET irrigation, and 100% ET irrigation) at Corpus Christi, Texas. For this date, sprays significantly suppressed populations ( $P < 0.05$ ); therefore means comparisons across the four cultivar—plant date treatments were done separately in sprayed (capital letter) and unsprayed (lower case letter) plots. Means with different letters represent significant differences ( $P < 0.05$ ), Tukey's means separation test.

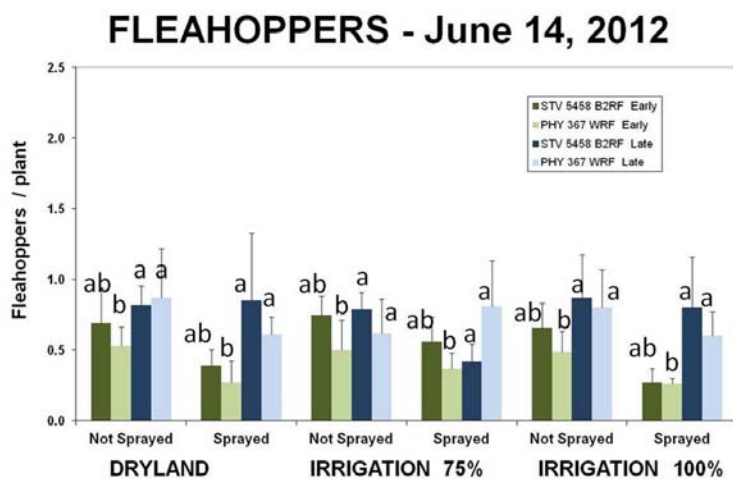


Fig. 4. Cotton fleahoppers per plant on June 14, 2012 on Stoneville 5458 B2RF and Phytogen 367 WRF, sprayed and not sprayed and planted relatively early (April 12, 2012) and late (April 30, 2012) across 3 water regimes (dryland, 75% ET irrigation, and 100% ET irrigation) at Corpus Christi, Texas. For this date, sprays did not suppress

the expanding population ( $P > 0.05$ ); therefore means were compared across all treatments. Means with different letters represent significant differences ( $P < 0.05$ ), Tukey's means separation test.

**Plant Measurements.** Irrigation significantly increased yield, and the relatively early planting had higher yields under irrigation. Yield reduction attributable to fleahopper was not detected; even though the early planting had higher fleahopper populations (including ones above the economic threshold of 15 fleahoppers per 100 plants in our area) (Fig. 5).

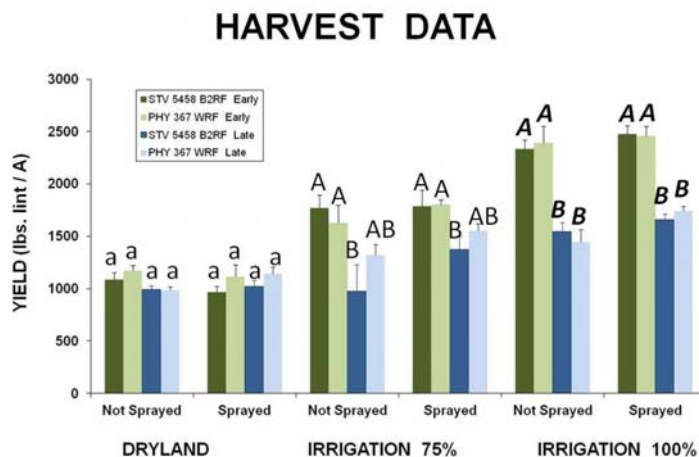


Fig. 5. Lint yield (lbs./A) of Stoneville 5458 B2RF and PhytoGen 367 WRF, sprayed and not sprayed and planted relatively early (April 12, 2012) and late (April 30, 2012) across 3 water regimes (dryland, 75% ET irrigation, and 100% ET irrigation) at Corpus Christi, Texas. Irrigation significantly increased lint yield ( $P < 0.05$ ); therefore means comparisons across the four cultivar—plant date treatments were done separately by insecticide and irrigation treatment. Means with different lower case, capital, and bold capital letters represent significant differences ( $P < 0.05$ ), Tukey's means separation test.

A late season occurrence of leaf-footed bug occurred in the test. We observed movement of leaf-footed bugs when the cotton was at cut-out (5 NAWF). No insecticide was applied and the population increased, including substantial egg-laying and nymphs occurring on the crop. Field observations indicated greater occurrence of leaf-footed bug on 1) relatively early planted plots compared to the late planted plots, 2) irrigated plots compared to the dryland plots, and 3) top crop bolls compared to the middle crop bolls. Boll damage was significantly higher in bolls near the top of the plant (top crop bolls) of the relatively late planted cotton when compared to the early planting. Despite this top crop damage, no differences in lint yield could be attributed to the leaf-footed bug damage.

**Lubbock: Plant Measurements.** The total number of fruit set per plant increased with increasing irrigation, but fruit retention suffered only when irrigation was reduced (low irrigation and dryland) (data taken from a complete plant mapping on August 3, 2011) (Fig. 6). The irrigation level significantly influenced cotton fruit physiology, with larger and heavier bolls with harder carpel walls produced at high irrigation regimes compared to those at the low irrigation and dryland (Figs. 7 and 8).



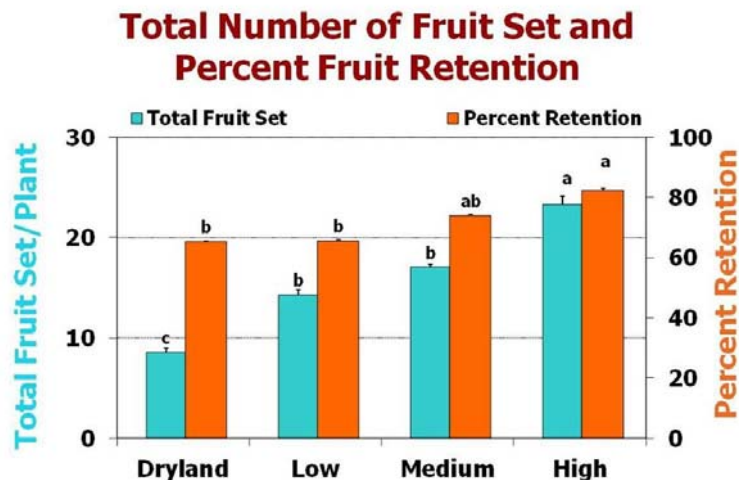


Fig. 6. Total number of fruit set per plant and percent fruit retention but fruit retention under 4 water regimes (dryland, low, medium, and high) on August 3, 2011 at Lubbock, Texas. Means with different letters represent significant differences ( $P < 0.05$ ).

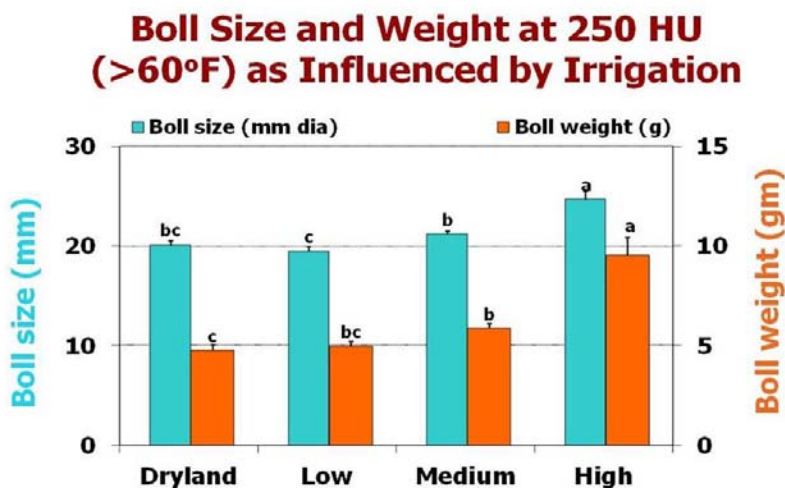


Fig. 7. Boll size(mm) and boll weight(gm) at 250 heat units (>60 °F) post bloom under 4 water regimes (dryland, low, medium, and high) at Lubbock, Texas. Means with different letters represent significant differences ( $P < 0.05$ ).

### Pressure Required to Puncture the Carpel Wall of Bolls (250 HU >60°F) as Influenced by Irrigation

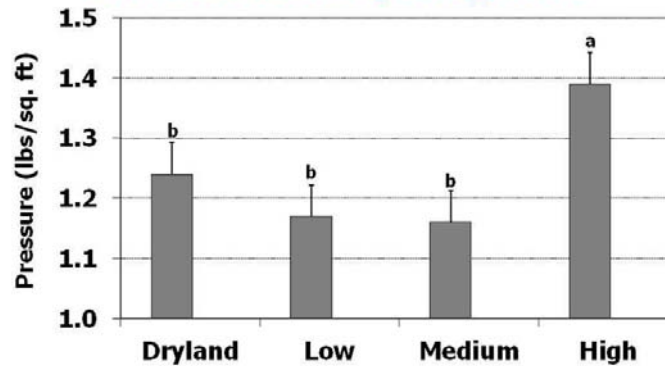


Fig. 8. Pressure required to puncture the carpel wall of bolls (250 heat units post bloom >60°F) under 4 water regimes (dryland, low, medium, and high irrigation). Means with different letters represent significant differences ( $P < 0.05$ ).

Overall, fleahopper populations were less sensitive to plant water stress and more sensitive to plant development stage, which partly explains field to field differences experienced by growers. Although more abundant during bloom, square sensitivity to cotton fleahopper damage and early season suppression of the population are the primary main considerations in insecticide use when fleahoppers occur during squaring. Plant/boll vigor in good soil moisture conditions likely benefits cotton in tolerating cotton fleahopper when cotton fleahoppers occur after squaring. Detection of fleahoppers during bloom in early planted cotton may serve as early warning of cotton fleahoppers in cotton planted later.

#### Acknowledgements

We thank R. Kurtz and P. O'Leary (Cotton Inc.) and Charles Suh (USDA ARS) for discussions as we developed this study. Cotton Inc. Core Program funds (project 11-952) and existing collaborations were critical in launching this project. Thank you.

## MANAGING THRIPS USING ORGANICALLY APPROVED INSECTICIDES

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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 5 treatments, 4 OMRI approved foliar insecticides and an untreated check, was conducted near Muleshoe, TX. Thrips pressure was moderate and lower than normally experienced. One or more treatments may have provided some repellency which may extend past treatment boundaries. Entrust did provide suppression of thrips in this trial and residual activity seems to be cumulative. No treatment provided any benefit in lint yield.

### Introduction

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. 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 13 Organic Materials Review Institute (OMRI) approved insecticides were investigated for suppression of thrips in cotton. The study was continued in 2012 but the treatment list was reduced to only those products which showed potential to provide significant thrips suppression in 2011. 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. The objectives of this trial were to evaluate the efficacy of numerous OMRI approved insecticides for thrips suppression in cotton and verify any possible yield benefits.

### Materials and Methods

The trial was conducted in 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. 'FiberMax 958' was planted 1 May, 2012 on 30-inch rows and irrigated using low elevation spray application (LESA) center pivot irrigation system. Plots were 4-rows wide × 45 ft long and were arranged in a randomized complete block design with 4 replicates. Treatments included 4 OMRI approved insecticides and an untreated check (UTC) (Table 1). All insecticides were applied in accordance with their respective label recommendations at 30 gallons/acre (GPA) total volume. Insecticide applications were made weekly, beginning at 85% emergence 19 May. Treatments were applied in a 10 inch band directly over the top of the crop row with a CO<sub>2</sub> pressurized backpack sprayer and hand held boom equipped with hollow cone nozzles. Thrips were counted before treatment as well as 3-4 and 6-7 days after each insecticide application. Ten plants/plot were collected and washed in an alcohol solution; adult and immature thrips collected in solution were filtered out and counted under a dissecting stereo scope. Samples collected were also separated by life stage. Plant damage ratings, from 1 to 5, were assessed when most plants had reached the 6 true leaf stage. One of the two middle rows, in which no plants had been sampled from, was hand harvested in its entirety November 1. Bur cotton grab samples were taken from each plot. The samples were ginned at the Texas A&M AgriLife Research and Extension Center in Lubbock, Texas. 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 methodology described by Robert F. Ruppel (JEE, Vol. 76, No. 2, April 1983).

**Table 1. Treatments and application detail from an organic thrips management trial, Muleshoe, TX, 2012.**

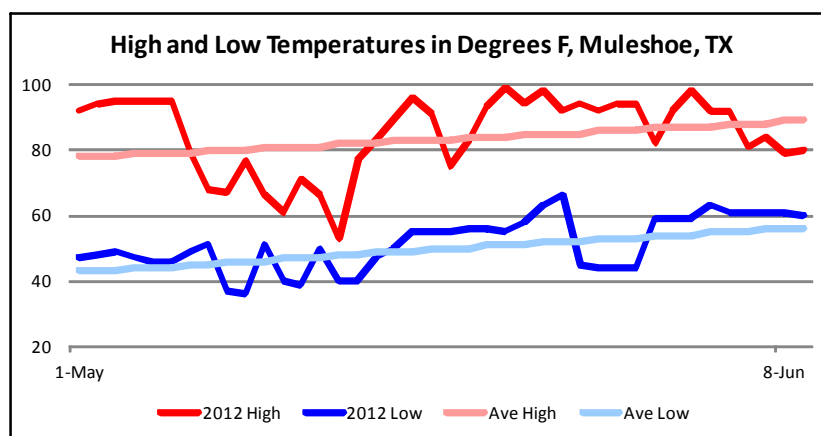
Trade name	Common name	Rate	GPA
Untreated	---	---	---
Aza-Direct <sup>1,2</sup>	Azadirachtin	16 fl-oz/ac	30
Entrust <sup>1</sup>	Spinosad	2 oz/ac	30
Bugitol	Capsicum /Mustard oils	96 fl-oz/100 gal	30
Saf-T-Side + Ecotec	Petroleum oil + Rosemary/Peppermint oil	1 gal + 1 qt/100 gal	50

<sup>1</sup>Ag-Aide added to spray mix at 8 fl-oz/100 gal (adjuvant)

<sup>2</sup>Constant BU<sub>P</sub>H-er added to the spray mix at 0.125% v/v (pH = 6)

### Results and Discussion

Environmental conditions at the trial site were harsh; extremely dry, very windy, and temperatures were erratic (Figure 1). Thrips pressure, in general, was moderate and lower compared to historical observations likely due to harsh conditions and lack of alternative hosts to support and bridge thrips populations until cotton emergence.



**Figure 1. High and low temperatures from 2012 vs. the 30 year long term averages (1980-2010).**

The cotton was very slow to develop, 18 days were required to attain 85% emergence 19 May and an additional 6 days from emergence until the 1st true leaf stage 25 May. Mean thrips numbers of untreated plots were less than 50% of action threshold when the initial insecticide application was applied (19 May, 85% emergence) but was near 5X the established action threshold of one thrips per true leaf by 23 May and remained near or above action threshold through the 5 true leaf stage 8 June (Table 2).

**Table 2. Thrips numbers and action threshold.**

Date	Thrips/ True Leaf <sup>1</sup>	Threshold <sup>2</sup>
5/19	.40	1
5/23	5.0	1
5/25	5.3	1
5/28	1.5	2
6/1	8.6	3
6/4	4.0	4
6/8	6.0	5

<sup>1</sup> Mean thrips per true leaf of UTC

<sup>2</sup>Established action threshold is 1 thrips/true leaf.

A significant difference was only observed between insecticide treatments and the untreated check at the 5 true leaf stage 8 June (Figure 2).

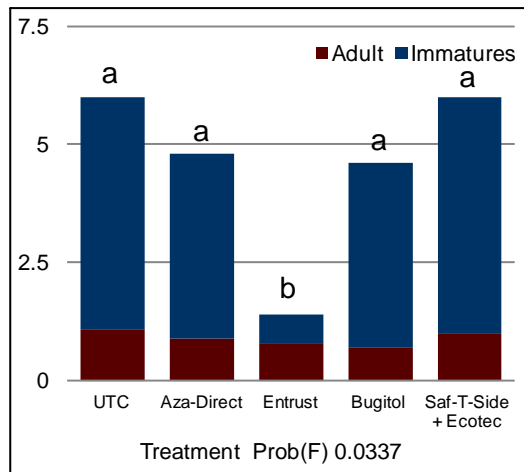
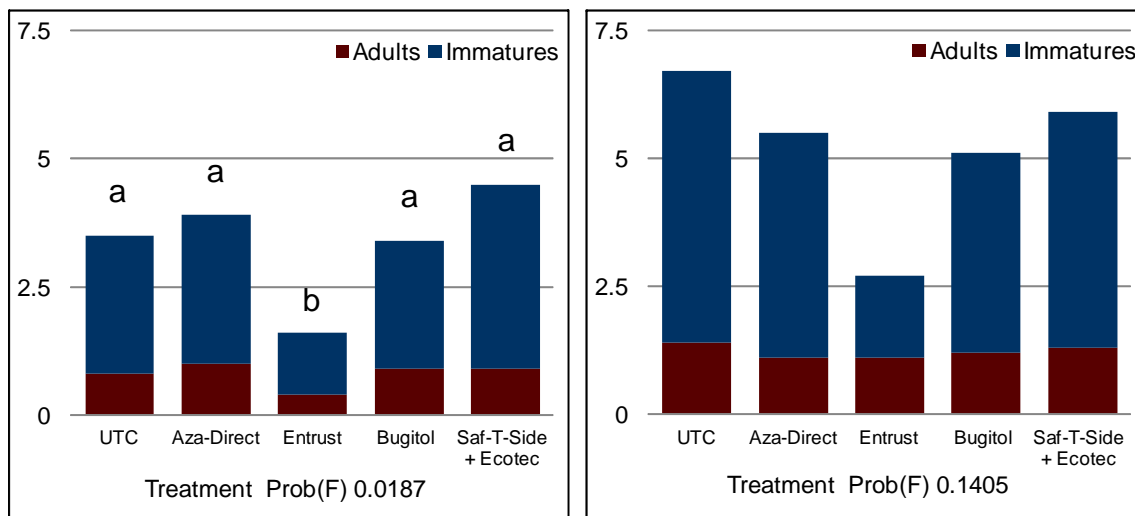


Figure 2. Mean thrips per plant at the 5 true leaf stage, 8 June.

Data were further analyzed by calculating seasonal means by treatment and days after treatment (DAT) (Figures 3 and 4). The Entrust treatment had significantly fewer thrips/plant compared to all other treatments and the UTC but no other treatment significantly differed from the UTC 3 DAT. The same analysis showed no significant differences 7 DAT.



Figures 3 and 4. Seasonal mean thrips per plant 3 and 7 DAT respectively.

When comparing thrips numbers/plant in all treatments and the UTC across all sampling dates, it appears that one or more of the treatments may be repelling thrips from the test area resulting in reduced pressure shortly after application followed by a population rebound (Figure 5). This phenomenon was observed following 2 of 3 insecticide applications. This and a comparison of seasonal means 3 and 7 DAT also suggests a very short residual

activity of suspected repellency.

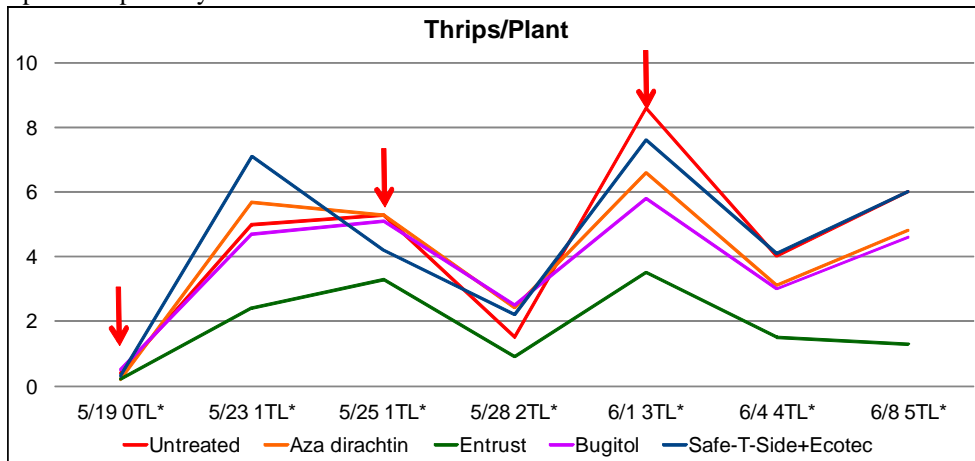


Figure 5. Mean thrips per plant and insecticide applications (↓) 19 May – 8 June (\*true leaves/plant).

Damage ratings, where 1 was least damage and 5 was greatest damage, taken at the 6 true leaf stage on 18 June showed Entrust with least damage with a rating of 2.3; Aza-Direct, Bugitol, Safe-T-Side+Ecotec, and the untreated had statistically similar damage ratings (Figure 6). Typically, damage ratings must exceed 3 to elicit a yield response.

The percent of a thrips population which is immature is a good indicator of that population’s ability to colonize; a higher percentage of immatures suggests a higher degree of colonization. When data from all post treatment sampling dates were merged and analyzed, the Entrust treatment had a significantly lower percentage of immature thrips compared to all other treatments (Figure 7). Based on this data, Entrust appears to suppress colonization to a greater degree compared to the other treatments.

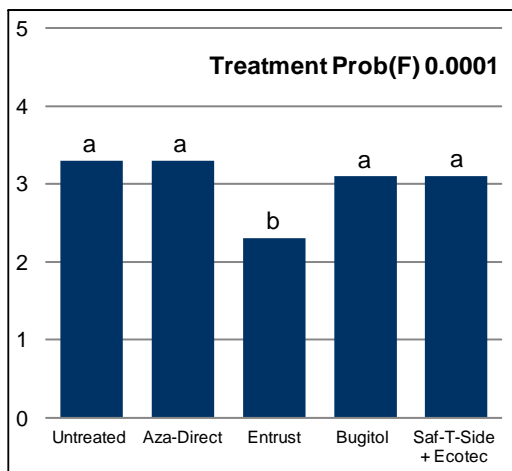


Figure 6. Plant damage rating at the 6 true leaf stage.

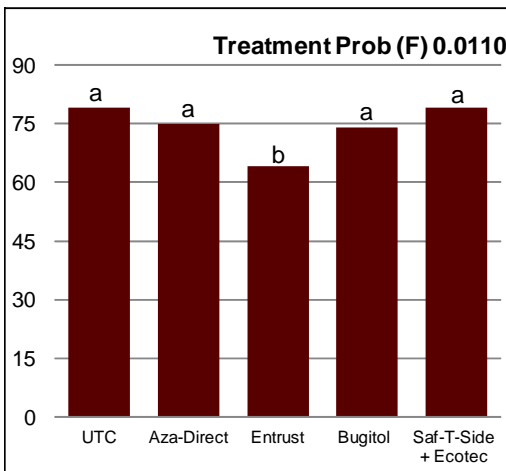
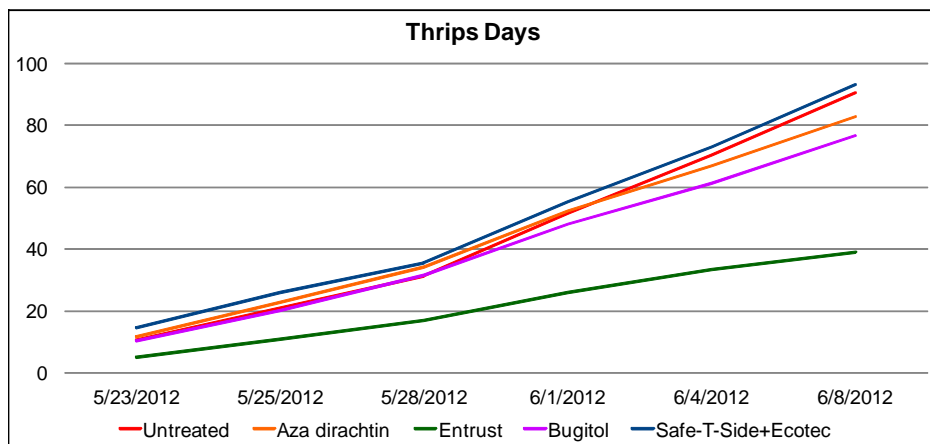


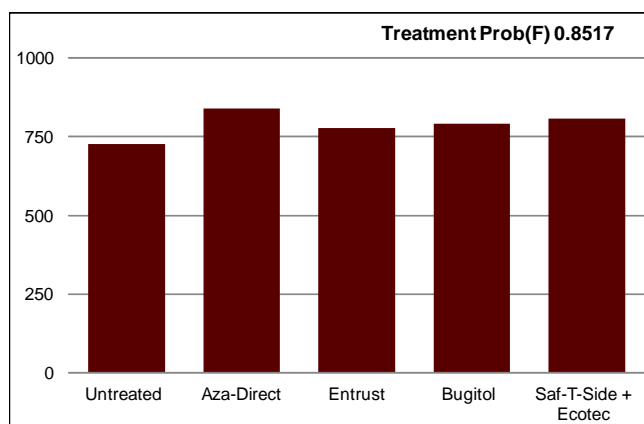
Figure 7. Seasonal means of the percent immature thrips.

Cumulative thrips days can give an overall indication of crop protection provided by insecticide treatments. Entrust reduced thrips days by over 50% when compared to all other treatments and the untreated check (Figure 8). This decrease is an indication of a reduction in overall thrips pressure and feeding duration.



**Figure 8. Cumulative thrips days (Ruppel, 1983, JEE)**

The trial mean lint yield across all treatments was 788 lbs/acre which is fair for the area; no differences between treatments were observed (Figure 9). Visual symptoms of phenoxy herbicide drift appeared across the trial shortly after thrips and plant damage data collection was completed. This plant injury may or may not have impacted yield.



**Figure 9. Lint yield in lbs/acre.**

### Conclusions

Thrips pressure was moderate but still less than normally experienced. One or more treatments may be providing repellency which may extend past treatment boundaries. Entrust did provide suppression of thrips in this trial and residual activity seems to be cumulative. Entrust clearly reduced thrips days, appeared to curb colonization to a greater degree, as well as reduce visual plant damage. No treatment provided any benefit in lint yield.

### 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, Austin Mason, Haden Hadley, Adrienne Precure, Owen McSpadden, Chris Moxon, Caleb Champion and Taylor Person for their contribution in processing thrips samples.



## SEASONAL FLIGHT PATTERNS OF BOLLWORM, TOBACCO BUDWORM AND BEET ARMYWORM MOTHS IN THE TEXAS HIGH PLAINS

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### Abstract

From 2002-2012, adult (moth) flight patterns of the cotton bollworm, *Helicoverpa zea* (Boddie), tobacco budworm, *Heliothis virescens* (F.), and beet armyworm (*Spodoptera exigua* (Hübner)) were monitored by using pheromone traps. During the first four years, moth captures were monitored approximately weekly during all months in three counties which represent the northern (Hale), central (Lubbock), and southern (Gaines/Dawson) regions of the Texas High Plains. Weekly monitoring has continued in Lubbock County since 2006. Yearly and historical flight profiles are provided and discussed for each of the three counties.

### Introduction

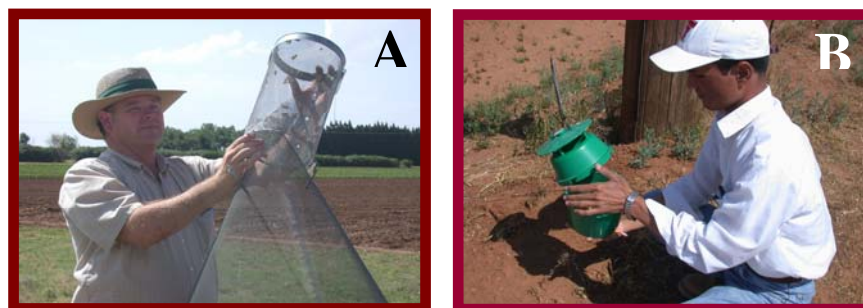
In 2002, an ongoing trapping study was initiated to investigate the weekly and seasonal flight activity patterns of the cotton bollworm, *Helicoverpa zea* (Boddie), tobacco budworm, *Heliothis virescens* (F.), and beet armyworm, *Spodoptera exigua* (Hübner) moths in the southern Texas High Plains (THP). Insect pheromone traps were used to measure the seasonal abundance of these pests. Previous research on two of these species includes Parajulee et al. (2004) report on a 14-year (1982-1995) study of monitoring THP bollworm and tobacco budworm populations. In the neighboring Texas Rolling Plains, Parajulee et al. (1998) report on a similar 15-year trapping study which included both weekly and daily trap service intervals.

These three species are significant cotton pests in the THP, which is widely recognized as the most intensive cotton growing area in the world. In the THP, the cotton bollworm is classified as an important economic pest of cotton and other regional crops, while the tobacco budworm and beet armyworm are classified as occasional pests. Seed from genetically modified cotton is available with Bollgard (*Bt*) technology which provides excellent crop protection from these lepidopteran pests. It is important to continue monitoring these pest populations due to the significant amount of cotton acreage that is not planted with this technology, particularly lower input dryland acres which account for approximately 40-50% of the THP cotton acreage. The Bollgard technology was adopted in less than 5% of the THP cotton acreage by 2004, but its adoption since then has increased significantly to the current level of nearly 80% in irrigated acreage due to superior agronomic characteristics in most cultivars with *Bt*-technology. There is also an interest in determining whether the widespread adoption of *Bt*-technology in crops such as cotton and corn will bring about an overall decrease in lepidopteran pest populations across local and neighboring regions. Continued long term monitoring of these pest populations will hopefully help address questions of this type.

## Materials and Methods

During the first four years (2002-2005), nine (3 monitored species x 3 replications) pheromone traps were placed in each of three selected counties representing northern (Hale), central (Lubbock) and southern (Gaines/Dawson) regions of the southern Texas High Plains. Monitored species included the cotton bollworm, tobacco budworm and beet armyworm. In each county, three sites (replications) were selected and one trap for each pest species was placed at each site, then baited and monitored approximately weekly throughout the year (2002-2005). Traps originally located in Gaines County (southern county) were moved to neighboring Dawson County after the second year of the study to facilitate more frequent monitoring. Beginning in 2006 and continuing to date, traps with the same protocols and sites were serviced only in Lubbock County.

Trap types used to capture the adult moths included the Texas pheromone trap (Fig. 1A, Hartstack et al. 1979) for bollworm and budworm moths and green bucket traps (Fig. 1B) for beet armyworm moths. Pheromone for all three species was secured from a single source (Trece™, Inc., Adair, OK). The cotton bollworm and tobacco budworm traps were re-baited approximately twice monthly, while the beet armyworm pheromone was changed monthly. The bucket (capture container) on beet armyworm traps also contained a 1-inch x 1-inch toxicant strip to kill the moths soon after capture. Exact locations of all trapping sites were determined using a hand-held Garmin® GPS device.



**Figure 1.** Texas pheromone traps (A) are commonly used to monitor moth populations such as the cotton bollworm and tobacco budworm, while the green bucket trap (B) is recommended for beet armyworms.

## Results and Discussion

### Cotton Bollworm

Figure 2A illustrates the calculated historical bollworm flight profiles (based upon pheromone trap captures) across years for the three counties. Bollworm flight activity was low or non-existent during the period of 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 fruit is vulnerable to damage. Within this extended period of activity, the highest numbers of moths responded to traps from early August to mid-September.

Four (2002-2005) individual yearly bollworm moth flight patterns for each county are shown in Fig. 3. For study years 2002-2005, the within-year county trap response patterns for the three counties were relatively similar to each other, and between years the patterns were also similar except for differences in cotton bollworm abundance. Overall population levels detected in Lubbock County were highest in 2002 with peaks of approximately 2,000 moths/trap/10-day period and lowest in 2004. Yearly flight profiles and moth abundance were relatively similar to each other in other years of the survey.

### **Tobacco Budworm**

Figure 2B shows the historical flight activity for tobacco budworms by county across year. Small numbers of budworms started responding to traps in late April while peak numbers were observed from early June to early October. Hale County (northern area) peaked one month later than the more southern areas and also had an abbreviated period of flight activity. After each county's peak activity period, numbers fell quickly with essentially no moth activity detected by late October. Lubbock County had the highest number of tobacco budworms responding to pheromone traps (Fig 2B) but this was likely skewed by the much higher Lubbock County counts in 2002 and 2003 (Fig. 4).

Within the four individual years (2002, 2003, 2004, 2005), tobacco budworm trap responses had similar patterns in all counties although moth numbers were notably higher in Lubbock County during the active periods of 2002 and 2003 (Fig. 4). With the exception of 2004, the period of highest flight activity for Hale County typically reached its peak approximately one month later than the more southern counties (Fig. 4).

### **Beet Armyworm**

The averaged county historical trap response profiles for 2002-2005 (Fig. 2C) indicate that beet armyworm populations on the Texas Southern High Plains displayed two peak periods of flight activity during the study. The first peak typically occurred in mid-April followed by an extended peak of moth activity during the period of late August to late November.

Based upon the individual yearly data for 2002-2005 (Fig. 5), the two peak periods of annual beet armyworm flight activity reflected in the historical profile (Fig. 2C) can be easily seen in most of the individual annual flight profiles. Although beet armyworms can be captured during all months of the year, they are primarily active during the period of early March to early December. Figure 5 illustrates the similarities of the county moth activity patterns within years and at the same time shows how vastly different overall moth abundance can be between individual years.

### **Moth Flight Dynamics during the Past 11 Years**

Figures 6-8 highlight the changing moth flight dynamics that have occurred the past 11 years (2002-2012) for these three significantly important lepidopteran species in the Texas High Plains region. The last 11 years of moth survey in the Lubbock County encompass: 1) the early timeframe with boll weevil free THP cotton production enterprise, 2) time spans of increasing adoption of Bollgard technology that confers resistance to these caterpillar species, and 3) several recent years characterized as a record-setting severe drought.

Figure 6 depicts the average number of bollworm moths/trap/week in Lubbock County during grouped years of significant THP cotton system changes including: 1) the years immediately following boll weevil eradication and beginning of Bollgard adoption (2002-2004), 2) increased Bollgard adoption years (2005-2007), 3) Bollgard adoption peak years (2008-2010), and 4) the two most recent years (2011-2012) which are characterized as extremely low rainfall years which were also coalesced with an obvious trend towards more limited THP irrigation pumping capacities across most of the region. First, it is evident that bollworm moth abundances have generally declined as the adoption of Bollgard technology increased. This is evidenced by the reduced peak numbers and the overall duration of the season moth flight activity. In addition, the peak moth activity has shifted earlier during the more recent 2008-2010 and 2011-2012 periods that coincided with regional Bollgard adoption rates of >50%. While closer scrutiny is warranted to ascertain this observation, it is plausible that the overall bollworm pressure could have been lower in THP during the recent years compared with that in 5-6 years when the adoption of Bollgard technology was <30% due partly to increased adoption of Bollgard technology. The bollworm flight profile for the severe drought years of 2011-2012 tended to have a similar profile to the higher rainfall 2008-2010 timeframe except that overall number of moths were greatly reduced (lower peaks) during the drought years. The 2011-2012 moth profile not only had reduced densities during the drought, but also did not display a distinct peak as is evident during the other three periods (2002-2004, 2005-2007, 2008-2010).

From 2002-2004 during the early stages of Bollgard technology adoption, tobacco budworm moth populations were observed to be extremely high plus their presence started earlier and ended late as compared to the flight profiles of 2005-2012 timespan of higher *Bt*-technology adoption (Figure 7). The flight profiles of the later time periods (2005-2007, 2008-2010, and 2011-2012) appeared very similar to each other in respect to the active period, overall population densities and timing of peak flight numbers.

Clear trends of peak moth activity and seasonal activity durations were not evident for beet armyworm moths over the four timespans (Figure 8). The highest peak activity (400/trap/week) of beet armyworms was observed in early September of the 2002-2004 period, yet the highest overall beet armyworm populations were observed during the drought stricken years of 2011-2012. The unusual drought and record temperatures could be attributed for the increased beet armyworm moth abundance in 2011-2012. Although the underlying reasons are unknown, lowest peak and overall population densities were observed during 2008-2010, two years of which annual rainfall totals for Lubbock, Texas were abnormally high (2008 @ 26.5 inches; 2010 @ 28.0 inches).

If possible, we intend to continue this survey work in 2013.

### **Acknowledgements**

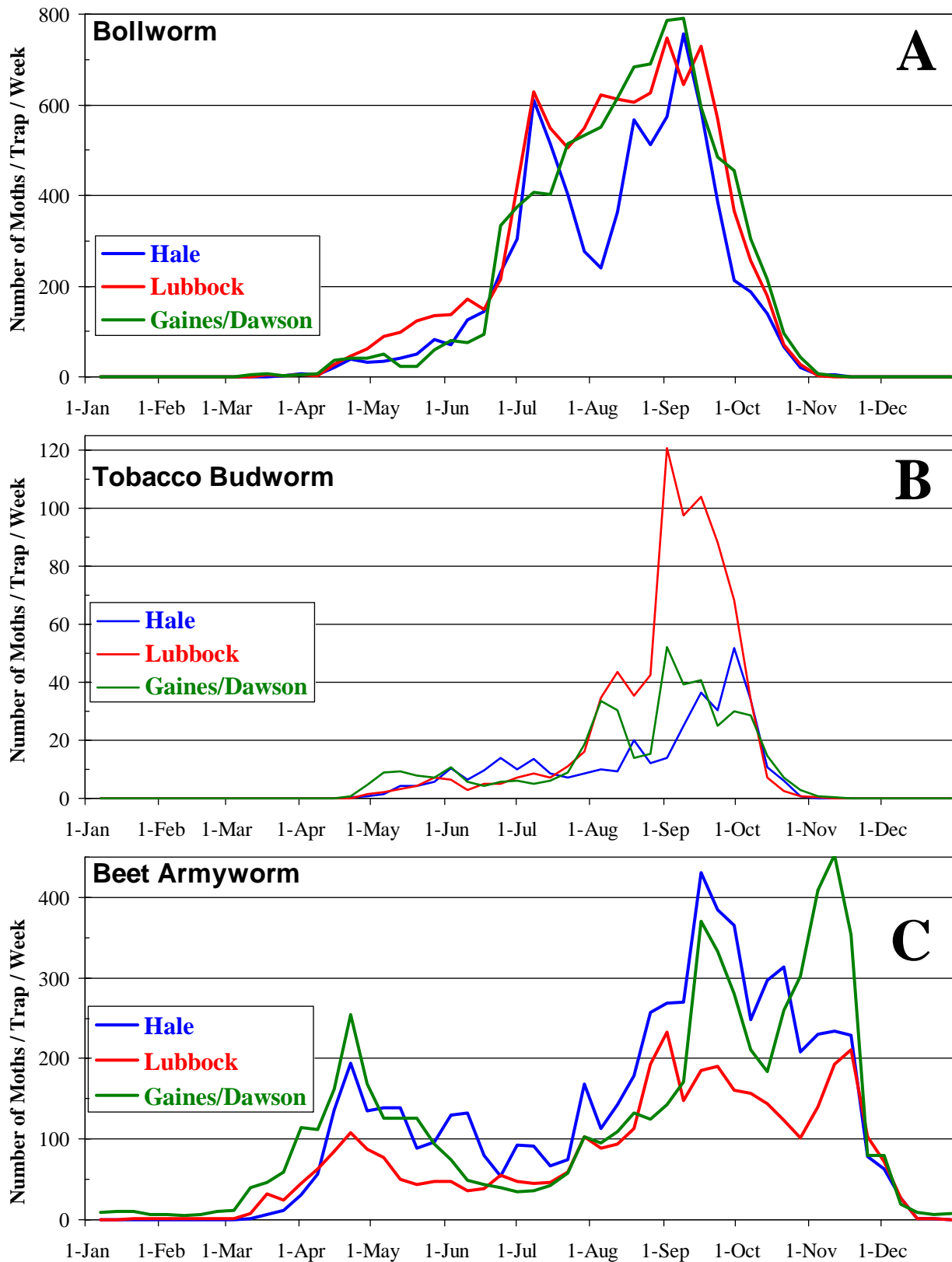
Funding for this study was provided by Texas A&M AgriLife Research Hatch Project 8810 and Plains Cotton Growers, Inc.

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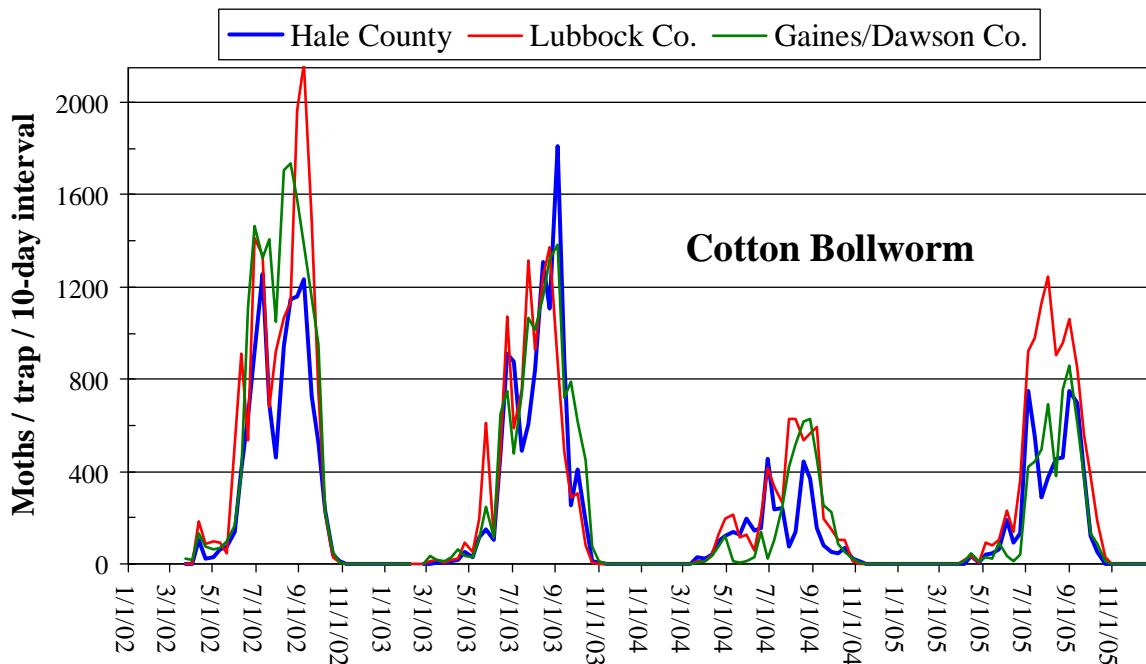
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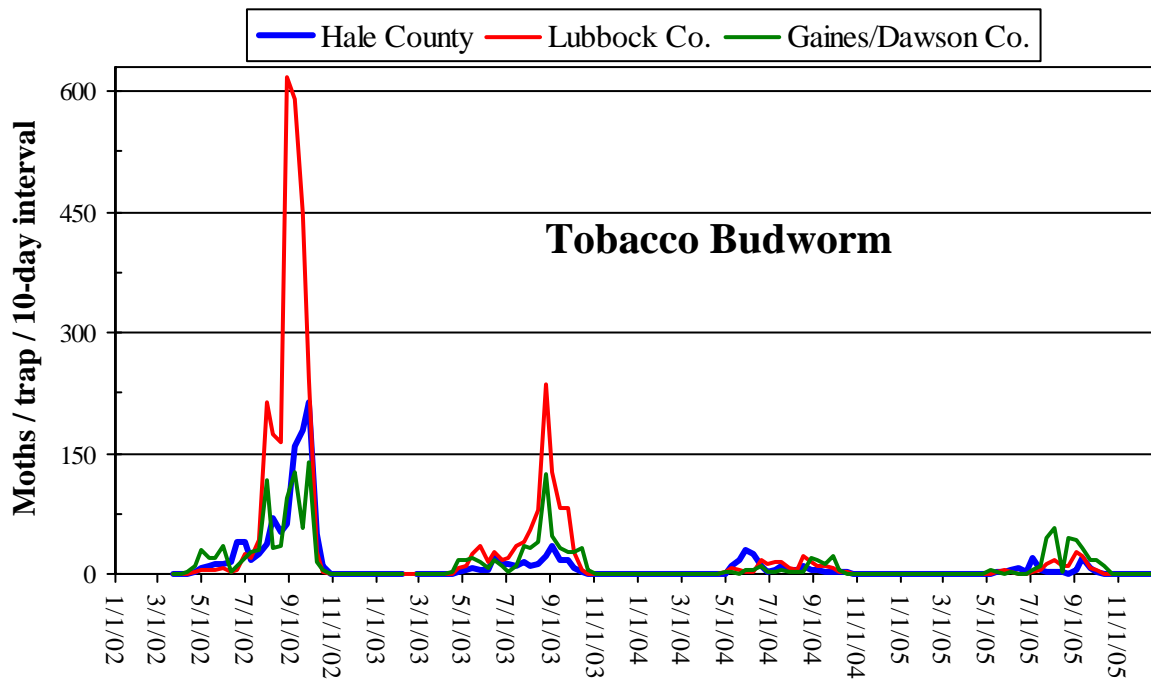
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**Figure 2.** Historical flight profiles (weekly trap captures averaged across all four years) for the cotton bollworm (A), tobacco budworm (B), and beet armyworm (C). For each of the three cotton pest species, county flight profiles are given so that comparisons can be made for areas roughly representing the northern (Hale), central (Lubbock) and southern (Gaines/Dawson) regions of the southern Texas High Plains region, 2002-2005.

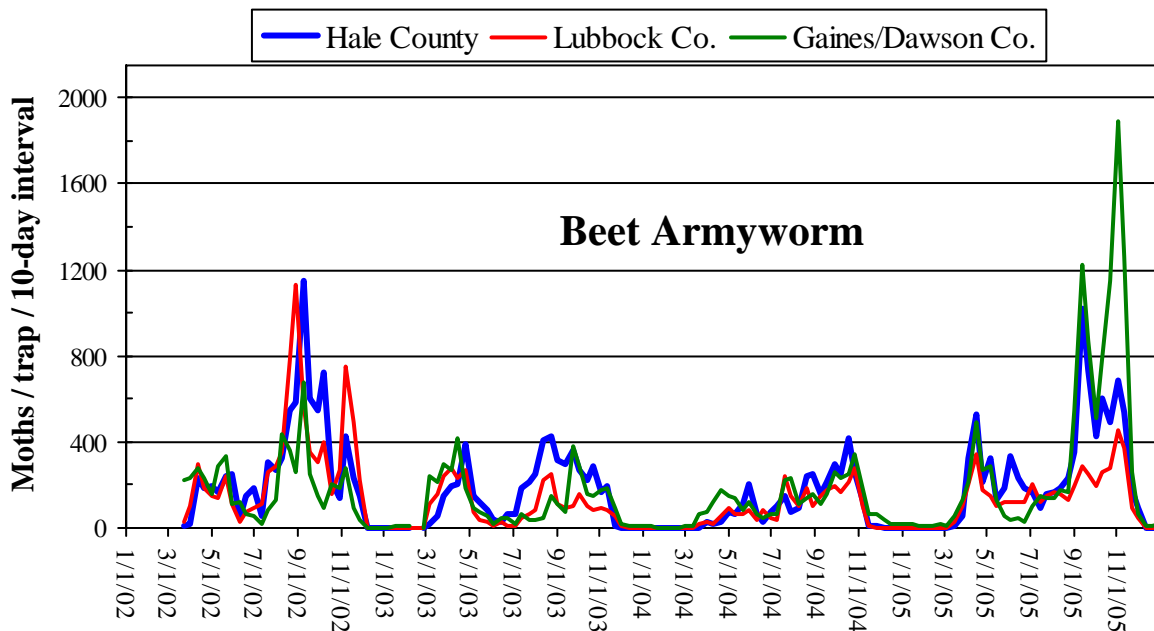


**Figure 3.** Average number of cotton bollworm moths/trap/10-day period in selected southern Texas High Plains counties, including Hale, Gaines/Dawson, and Lubbock County, 2002-2005.

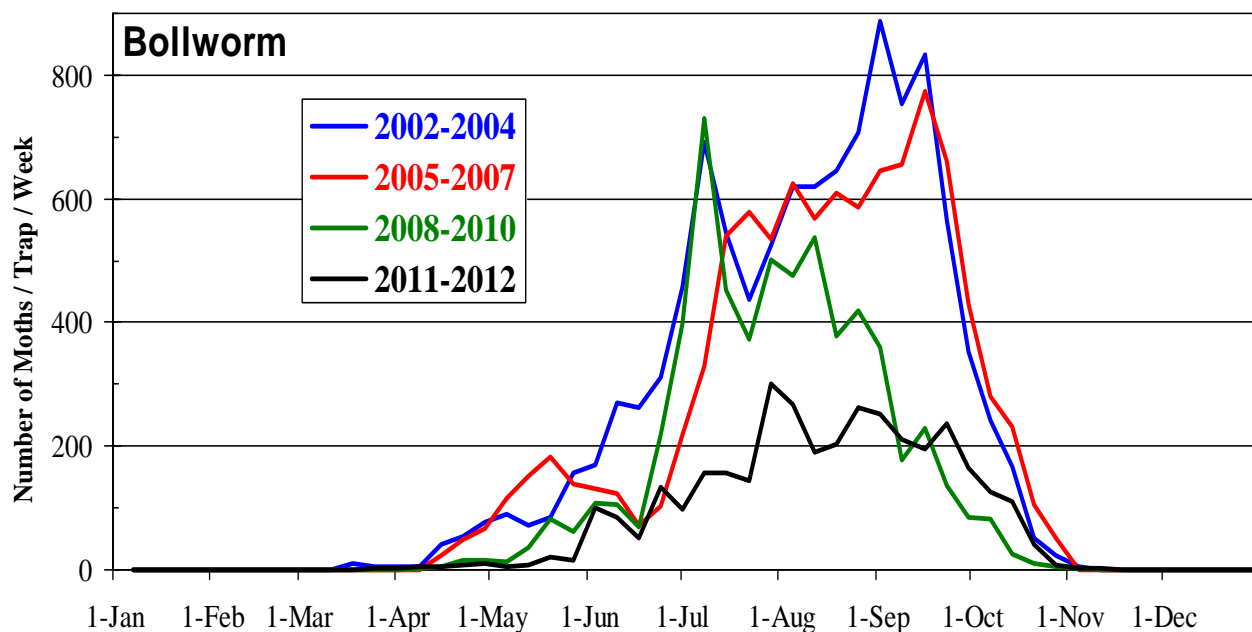


**Figure 4.** Average number of tobacco budworm moths/trap/10-day period in selected southern Texas High Plains counties, including Hale, Gaines/Dawson, and Lubbock County, 2002-2005.

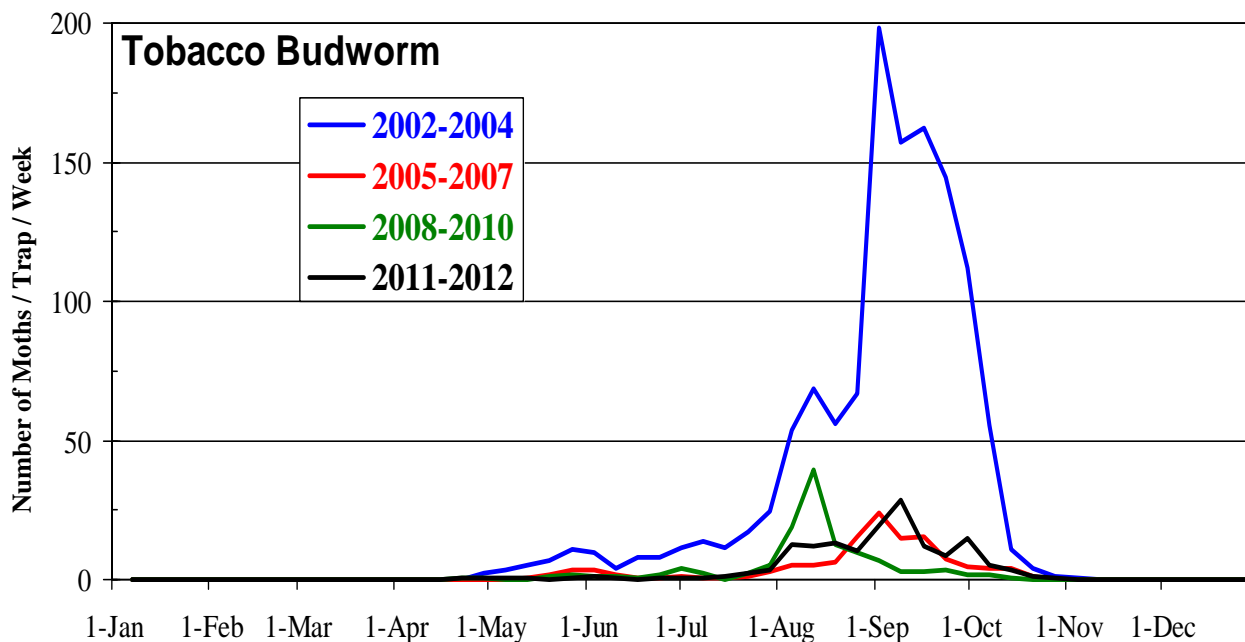




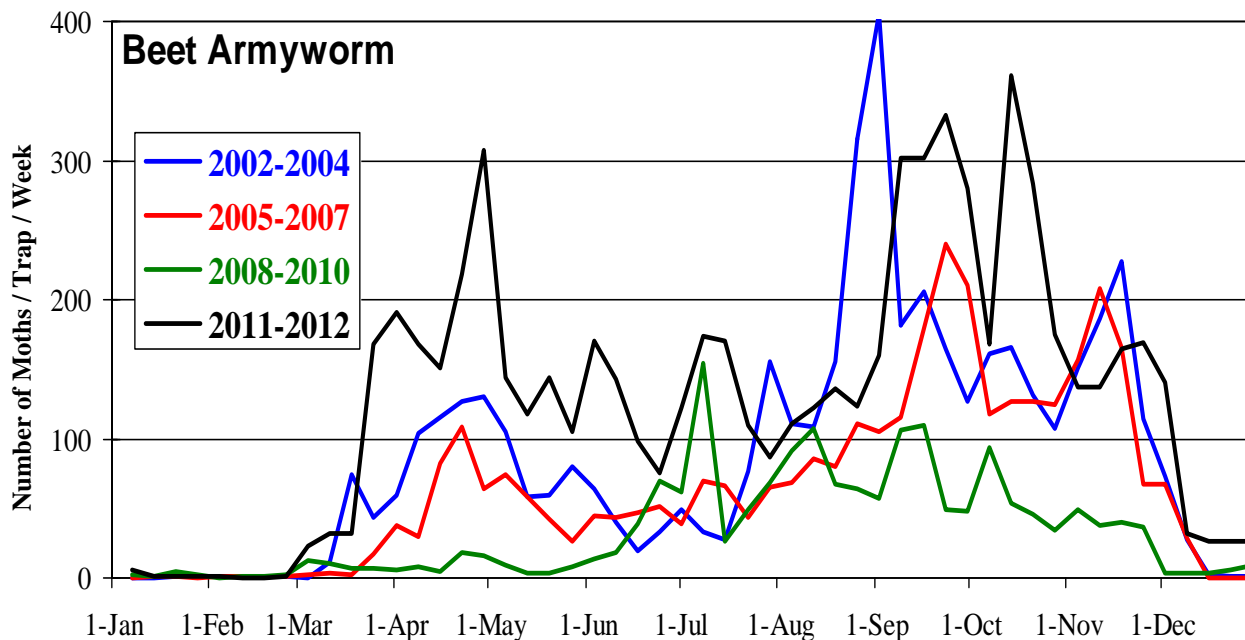
**Figure 5.** Average number of beet armyworm moths/trap/10-day period in selected southern Texas High Plains counties, including Hale, Gaines/Dawson, and Lubbock County, 2002-2005.



**Figure 6.** Average number of bollworm moths/trap/week in Lubbock County, depicting the years immediately following boll weevil eradication and beginning of Bollgard adoption (2002-2004), increased Bollgard adoption years (2005-2007), and Bollgard adoption peak years (2008-2010), compared with the recent two year (2011-2012).



**Figure 7.** Average number of tobacco budworm moths/trap/week in Lubbock County, depicting the years immediately following boll weevil eradication and beginning of Bollgard adoption (2002-2004), increased Bollgard adoption years (2005-2007), and Bollgard adoption peak years (2008-2010), compared with the recent two years.



**Figure 8.** Average number of beet armyworm moths/trap/week in Lubbock County, depicting the years immediately following boll weevil eradication and beginning of Bollgard adoption (2002-2004), increased Bollgard adoption years (2005-2007), and Bollgard adoption peak years (2008-2010), compared with the recent two years (2011-2012).

# Intra-species Mixture Alters Pest and Disease Severity in Cotton

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Environ. Entomol. 41(4): 1029–1036 (2012); DOI: <http://dx.doi.org/10.1603/EN11182>

**ABSTRACT** Widespread planting of crops genetically modified to express *Bacillus thuringiensis* (*Bt*) toxins for pest control may potentially affect nontarget pests and soil-borne disease. In this study, a field trial was conducted to explore the effects of habitat diversification, specifically an intraspecies mixture of genetically distinct cotton lines, on nontarget pests and soil-borne disease. It was hypothesized that the mixture would suppress disease severity and would alter pest and predator abundances. Results confirmed that a row-mixture of 75% insect-resistant cotton and 25% disease-resistant cotton suppressed *Fusarium* wilt and controlled cotton aphids. However, intercropping at the genotypic level increased mirid bug and sweetpotato whitefly densities in cotton. Moreover, the effect of the mixture on predator abundance ranged from neutral to positive and was highly variable among species and years. Species-specific pest responses to the crop mixture provide new insights for optimally sized and configured refuge construction in the future.

**KEY WORDS** intraspecies mixture, *Fusarium*, pest, predator, refuge

Cotton is a high market-value cash crop, but exposure to insect pests and plant diseases is imposing major limitations upon its production (Luttrell et al. 1994). Transgenic cotton engineered to continuously express a  $\delta$ -endotoxin from *Bacillus thuringiensis* Berliner (*Bt*) has effectively controlled *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) (Wu et al. 2008), and other lepidopterans. Meanwhile, *Bt* cotton adoption has significantly reduced insecticide usage (Huang et al. 2003, Men et al. 2005), contributing to protection of the environment and human health. These benefits are clearly acknowledged, as the cropping area of *Bt* cotton has expanded rapidly in northern China since 1998. Transgenic insect-resistant cotton has predominated in China since 2008 (Stone 2008, Zhang and Pang 2009).

Despite the predominance and success of transgenic cotton in China, there are some unexpected effects associated with it. For example, large-scale planting of *Bt* cotton has been observed to trigger arthropod guild rearrangement and outbreaks of secondary pests such as cotton aphids (Hemiptera: Aphididae), mirid bugs (Hemiptera: Miridae) (Wu et al. 2002, Lu et al. 2010), and sweetpotato whiteflies (Hemiptera: Aleyrodidae) (Naranjo 2005) in transgenic cotton fields. Furthermore, transgenic insect-resistant cotton lines exhibited reduced disease resis-

tance in comparison with conventional lines (Li et al. 2009), especially for *Fusarium* wilt, *Verticillium* wilt, and combination of these two diseases (Zhu and Feng 2005). As new, high-yielding cultivars conferring enhanced resistance are released and adopted, the naturally diverse agricultural landscape may be simplified by the resulting broad monoculture, especially in small-farm areas in northern China.

The nontarget effect associated with widespread adoption of transgenic insect-resistant cotton may respond to cultural practices such as intercropping. Previous studies have indicated the contribution of monoculture to condensed arthropod diversity and increased pest severity in cotton ecosystems versus that of intercropping (Altieri and Letourneau 1982). Furthermore, a broad monoculture may increase the risk of developing target pest resistance to *Bt* toxins and secondary pest outbreaks, both of which have the potential to compromise continued *Bt* cotton efficacy. Strategies favoring pest population suppression and delayed resistance development, including consideration of cultural control methodologies such as intercropping and refuge establishment, may ameliorate the risk of such compromise.

Habitat diversification, achieved by planting pattern alteration, is increasingly highlighted as an effective approach for crop pest control, disease suppression, and enhancement of farmland stability. Multiline or cultivar mixtures have been reported as promising habitat diversification tactic in terms of disease suppression (Browning and Frey 1969, Leonard 1969, Wolfe 1985, McDonald et al. 1988, Mahmood et al. 1991, Garrett and Mundt 1999, Zhu et al. 2000, Cowger and Mundt 2002, Mundt 2002, Leung et al. 2003, Pilet et al. 2006, Newton 2009, Newton and Guy 2009).

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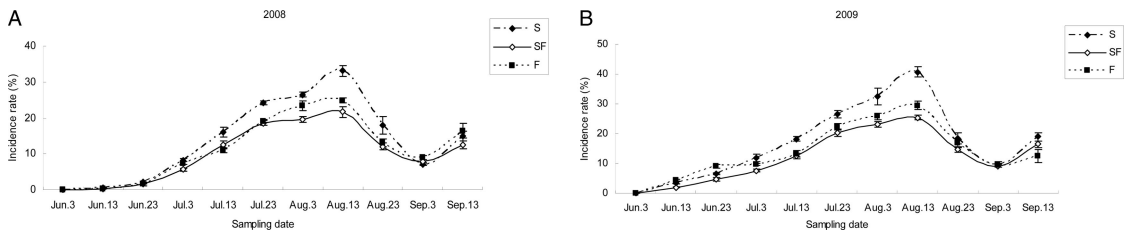


Fig. 1. Temporal dynamics of *Fusarium* wilt incidence rates (percentage) in two monoculture pure stands (transgenic Bt SZNC20B and non-Bt 93Fu56) and 3:1 mixture of both genotypes, Langfang, China, 2008–2009. S, monoculture of SZNC20B; SF, mixture of SZNC20B and 93Fu56; F, monoculture of 93Fu56.

Meanwhile, crop diversity has been viewed as an environmentally judicious pest control method. For instance, intercropping may increase predator densities and reduce cotton aphid abundances in cotton (Parajulee et al. 1997, Parajulee and Slosser 1999, Men et al. 2004). However, most intercropping studies address the effects of crop diversity enhancement on only disease suppression or pest control, but not both. This is contrary to the natural ecosystem, where diseases and pests manifest concurrently with complex interactions. In the natural ecosystem, predicting the effect of plant diversity on biocontrol of pests, diseases, and their complex interactions is virtually impossible, and in fact, the roles of crop mixture in biocontrol are poorly understood. Currently, no studies have examined the effects of genotype-level plant diversity on pests, diseases, and their interactions.

In this study, the responses of arthropod abundances and disease severity to crop mixture were examined in northern China in 2008 and 2009. It was hypothesized that intraspecific mixture would exert positive effects on disease suppression and pest control. Two questions were addressed: 1) Will an intraspecific mixture of disease-resistant and insect-resistant cotton lines suppress disease frequency and severity? 2) What impacts will intercropping exert on the population levels of target pests and their predators?

### Materials and Methods

**Experimental Setup.** Transgenic *Bt* cotton (SiZhuang NC20B) expressing Cry1Ac (Monsanto Company, St. Louis, MO), designed to control lepidopteran pests such as *H. armigera*, and a genotype (93Fu56) resistant to indigenous *Fusarium* wilt (*Fusarium oxysporum* f. sp.) and *Verticillium* wilt (*Verticillium dahliae* Kleb.) were used in this study. These varieties, both of which mature in  $\approx 130$  d, are planted commonly in northern China. Both are of medium height with a dark, medium green leaf color. In addition, the two genotypes share many agronomic characteristics.

Field trials were conducted at the Langfang Experiment Station (39.538° N, 116.708° E) in the Hebei Province, China, employing a randomized complete block design of two pure stands and a mixture of both varieties at a ratio of 3:1, with four replications. Before 2008, corn was grown at the selected field site. Plot layouts were identical between study years. The selected 3:1 ratio was based upon the current *Bt* cotton

adoption rate in the Hebei Province and recommended refuge size for *Bt* target pest resistance management (Vacher et al. 2003, Sisterson et al. 2004). Three treatments were deployed, including 1) monoculture planting of SiZhuang NC20B (treatment S), 2) intercrop planting of 75% SiZhuang NC20B and 25% 93Fu56 by row-mixing (treatment SF), and 3) monoculture planting of 93Fu56 (treatment F). SF plots were planted in a repeated pattern: one row of non-*Bt*, then three rows of *Bt*. The pattern continued until all rows within a plot were occupied. Seeds were carefully, manually sown, row by row. Each plot consisted of 0.33 ha, which is a typical cotton field size in Hebei Province. The seeding rate was targeted to produce 40,000 plants per ha. A 3-m fallow alley was left between plots to minimize insect dispersion among treatments. Standard agronomic practices common to northern China were followed in maintaining the test plots, and no insecticide or fungicide applications were applied.

**Data Collection.** Throughout the growing seasons of 2008 and 2009, disease severity assessment and arthropod sampling were conducted every 10 d. Sampling was initiated three (for disease) or 4 wk (for arthropods) after plant emergence (early June) and continued until crop defoliation (mid-September), resulting in 11 and 10 sampling dates, respectively. On each sampling date, disease severity monitoring and arthropod sampling were conducted simultaneously. For the disease severity assessment, five sites were selected uniformly within each plot to capture the within-plot field variability and then 10 plants were inspected randomly within each site, with 50 plants in total inspected per plot. *Fusarium* foliar symptoms on each plant were rated from zero to four by using the procedure of Colson-Hankse et al. (2000). Ratings included: 0 = no visible symptoms, 1 = 1/4 of total leaves showing symptoms (chlorosis or necrosis), 2 = 1/2 of total leaves showing symptoms, 3 = 3/4 of total leaves showing symptoms, and 4 = almost all leaves showing symptoms or a deceased plant. *Fusarium* incidence rate was expressed as the ratio of the total number of chlorotic or necrotic leaves to the total number of observed leaves. Disease severity was summarized for each plot as  $\{[(n_1 \times 1) + (n_2 \times 2) + (n_3 \times 3) + (n_4 \times 4)]/4 \times (n_1 + n_2 + n_3 + n_4)\} \times 100$ , where  $n_1$ ,  $n_4$  are the numbers of leaves in each of the respective disease categories, and 1, 2, 3, 4, is the disease symptom score.

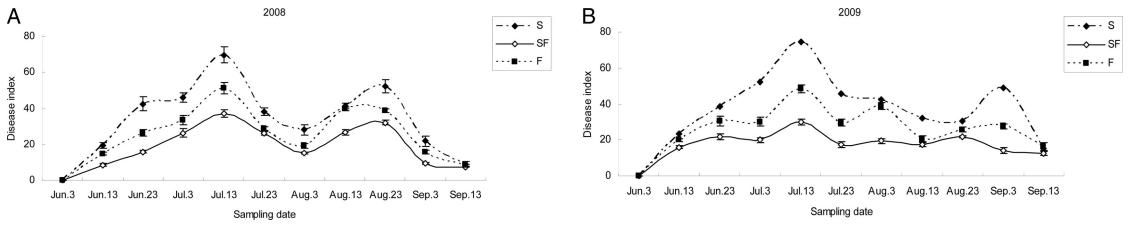


Fig. 2. Temporal dynamics of *Fusarium* wilt disease index (percentage) in two monoculture pure stands (transgenic Bt SZNC20B and non-Bt 93Fu56) and 3:1 mixture of both genotypes, Langfang, China, 2008–2009. S, monoculture of SZNC20B; SF, mixture of SZNC20B and 93Fu56; F, monoculture of 93Fu56.

Arthropod species sampled included three pest species [cotton aphids, *Aphis gossypii* Glover; mirid bug complex, *Lygocoris lucorum* Meyer-Dur, *Adelphocoris suturalis* Jackson, and *Adelphocoris fasciaticollis* Reuter; and sweetpotato whitefly, *Bemisia tabaci* (Gen-nadius) biotype B], and four predator species (lady-birds, lacewings, spider complex, and *Orius similis* Zheng). Arthropod species were sampled by visually inspecting 20 cotton plants at five randomly chosen sampling sites in each plot (100 plants per plot), in situ. Thus, on any sampling date, the total number of nymphs and adults inhabiting 100 cotton plants in each plot was quantified. Because of practical concerns owing to high densities, for each sampled plant, cotton aphid and sweetpotato whitefly abundances were quantified by visually inspecting three leaves each from upper, middle, and lower main stem portion of the plant. For other arthropods, entire plants were visually inspected in the morning (8:00–10:00) or afternoon (16:00–18:00), with particular attention paid to flowers and squares—likely hiding places for feeding insects. Unidentified sample specimens were collected and saved in 70% alcohol for subsequent laboratory identification.

**Data Analysis.** All analyses were performed using the linear mixed model procedure (SAS Institute 2010). Data from the five sample sites within a plot were averaged before analysis, and each plot was considered as an experimental unit. The responses of disease severity and arthropod abundances to treatment were processed in two steps. First, the effects of planting pattern, sample date, and their interaction on the disease severity, herbivore abundances, and natural enemies in each growing season were analyzed

separately with mixed model, repeated measures analysis of variance by using the PROC MIXED procedure, considering mixture pattern and dates as fixed variables. Then, the overall effects of these factors on disease severity, pest abundances, and predator abundances during the two study years were tested via linear mixed model by using year as a random factor. Data were  $\log(x + 1)$  transformed to satisfy analysis of variance assumptions of normality and homogeneity of variance before analysis. Treatment means were compared with the least significant difference test at  $\alpha = 0.05$ .

**Results**

***Fusarium* Wilt Disease Severity.** The tested cotton variety mixture pattern treatments exerted clear effects on both disease incidence rate ( $F = 464.84$ ;  $df = 2, 15$ ;  $P < 0.0001$ ) and disease index ( $F = 834.7$ ;  $df = 2, 15$ ;  $P < 0.0001$ ). Furthermore, pronounced differences in the incidence rate ( $F = 691.3$ ;  $df = 1, 15$ ;  $P < 0.0001$ ) and disease index ( $F = 459.5$ ;  $df = 1, 15$ ;  $P < 0.0001$ ) were observed between the two growing seasons. However, the interaction between year and variety mixture pattern was significant only for disease severity ( $F = 79.9$ ;  $df = 1, 15$ ;  $P < 0.0001$ ).

In each growing season, similar incidence rate and disease index trends were observed in plots of different measured cotton variety mix patterns (Figs. 1–2). In both years, the effects of variety mix pattern were manifested clearly on both indices, with significantly reduced disease incidence rate and disease index in crop mixture treatment (SF) compared with that in monoculture (S and F) treatments (Table 1; Figs. 1, 2).

Table 1. Mixed effect model statistics for disease incidence parameters and arthropod pest densities in cotton as affected by crop mixture pattern, block, and sample date, Langfang, China

Year	Effect	df	Incidence rate	Disease index	Cotton aphid	Mirid bug	Sweetpotato Whitefly
2008	Pattern	2, 6	109.25 <sup>d</sup>	107.06 <sup>d</sup>	157.91 <sup>d</sup>	15.12 <sup>c</sup>	25.09 <sup>d</sup>
	Block	3, 6	0.93 <sup>a</sup>	1.18 <sup>a</sup>	1.26 <sup>a</sup>	0.34 <sup>a</sup>	2.09 <sup>a</sup>
	Date	9, 99	92.90 <sup>d</sup>	89.12 <sup>d</sup>	600.74 <sup>d</sup>	100.43 <sup>d</sup>	87.12 <sup>d</sup>
2009	Pattern	2, 6	89.63 <sup>d</sup>	121.70 <sup>d</sup>	98.17 <sup>d</sup>	5.06 <sup>a</sup>	5.69 <sup>b</sup>
	Block	3, 6	1.51 <sup>a</sup>	3.84 <sup>a</sup>	1.67 <sup>a</sup>	0.58 <sup>a</sup>	3.81 <sup>a</sup>
	Date	9, 99	19.84 <sup>d</sup>	47.52 <sup>d</sup>	295.32 <sup>d</sup>	83.16 <sup>d</sup>	63.41 <sup>d</sup>

<sup>a</sup>  $P > 0.05$ .  
<sup>b</sup>  $0.05 < P < 0.01$ .  
<sup>c</sup>  $P < 0.01$ .  
<sup>d</sup>  $P < 0.001$ .

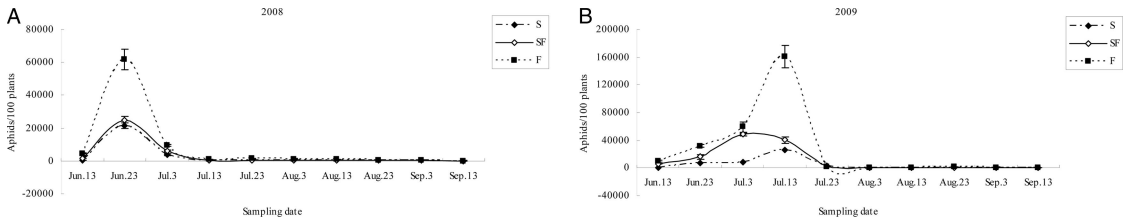


Fig. 3. Temporal dynamics of cotton aphids in two monoculture pure stands (transgenic Bt SZNC20B and non-Bt 93Fu56) and 3:1 mixture of both genotypes, Langfang, China, 2008–2009. S, monoculture of SZNC20B; SF, mixture of SZNC20B and 93Fu56; F, monoculture of 93Fu56.

In addition, temporal dynamics and the nature of the peak occurrence varied between disease incidence rate (Fig. 1) and disease index (Fig. 2). Although the incidence rate peaked in mid-August in both years (Fig. 1), the highest disease indices were observed in mid-July with a second peak in late August (2008) or early September (2009) (Fig. 2).

**Arthropod Pests.** Of the cotton insect pests measured, cotton aphids occurred in the highest densities, followed by mirid bugs, and whitefly biotype B.

**Cotton Aphids.** Although cotton variety mixture pattern exerted a marked effect on cotton aphid population dynamics ( $F = 458.8$ ;  $df = 2, 15$ ;  $P < 0.0001$ ), significant variation between the 2 yr ( $F = 248.8$ ;  $df = 1, 15$ ;  $P < 0.0001$ ) was observed (Table 1). The interaction between year and crop variety mixture pattern also influenced aphid abundances significantly ( $F = 25.7$ ;  $df = 2, 15$ ;  $P < 0.0001$ ). In both years, transgenic Bt monoculture plots (S) had significantly higher cotton aphid abundance compared with that in the *Fusarium*-resistant genotype monoculture (F) or variety mixture treatment (SF) (Fig. 3). Although similar aphid population dynamics trends were observed in plots of different mixture patterns (Fig. 3A, B), peak cotton aphid abundances differed significantly between years. In 2008, peak densities were observed on 23 June (Fig. 3A), whereas in 2009, the observed peak occurred on 13 July (Fig. 3B). Furthermore, observed peak cotton aphid abundances in plots of different variety mixture patterns (two monoculture treatments and one crop mixture) were more significantly separated in 2009 than in 2008 (Fig. 3).

**Mirid Bugs.** During the two consecutive growing seasons, mixture pattern significantly affected mirid bug population dynamics ( $F = 10.7$ ;  $df = 2, 15$ ;  $P = 0.001$ ), and although significant variation was ob-

served between the two seasons ( $F = 13.6$ ;  $df = 1, 15$ ;  $P = 0.002$ ), no significant interaction between year and mixture pattern was noticed ( $F = 1.3$ ;  $df = 2, 15$ ;  $P = 0.31$ ). In contrast to cotton aphid dynamics, mirid bug densities were significantly higher in the cotton variety mixture treatment (SF) compared with that in monoculture plots (S and F), especially during the peak population buildup (Fig. 4). Overall, mirid bug densities in plots of different mixture patterns followed a similar trend, with early population buildup, primarily owing to colonization via influx from the surrounding habitats, followed by population decline later in the season as the cotton crop matured. Although variety mixture pattern significantly affected mirid bug seasonal dynamics in both years (Table 1), the dates at which population peaks were observed differed significantly between years (Fig. 4). In 2008, the maximum densities occurred on 23 August (Fig. 4A), whereas the maximum densities in 2009 occurred on 3 August (Fig. 4B).

**Sweetpotato Whiteflies.** Crop genotype mixture ( $F = 220.1$ ;  $df = 2, 15$ ;  $P < 0.0001$ ) and year ( $F = 134.7$ ;  $df = 1, 15$ ;  $P < 0.0001$ ) significantly affected sweetpotato whitefly population dynamics, and the interaction between year and mixture pattern also was significant ( $F = 66.6$ ;  $df = 2, 15$ ;  $P < 0.0001$ ) (Table 1). Plots of different cotton variety mixture treatments exhibited a similar trend, with an initial increase after the arrival of immigrants from surrounding habitats and a decline later in the season (Fig. 5). Populations peaked 13 August in both years (Fig. 5A, B). However, population peaks in plots of different mixture patterns varied slightly in 2009 (Fig. 5B). It is also noteworthy that the peak sweetpotato whitefly densities in the transgenic Bt monoculture treatment were significantly dampened in both years compared with that in

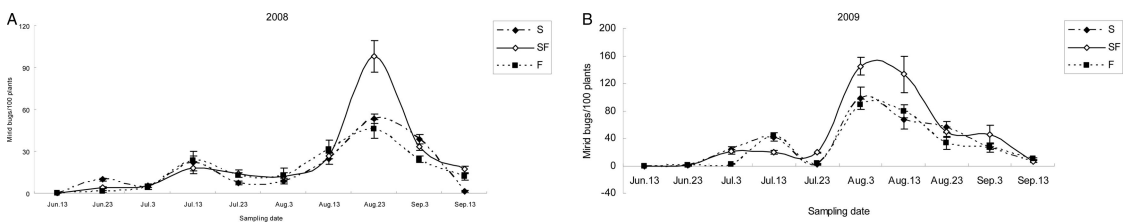
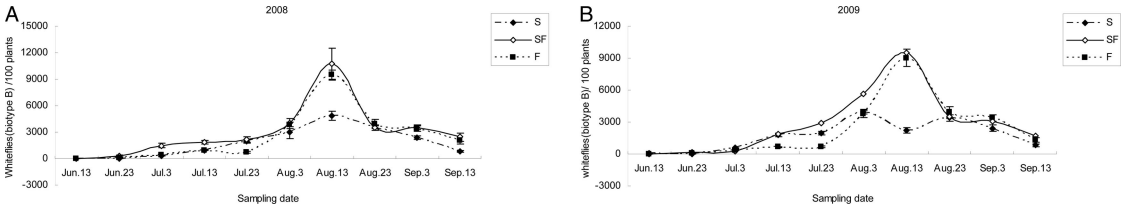


Fig. 4. Temporal dynamics of mirid bugs in two monoculture pure stands (transgenic Bt SZNC20B and non-Bt 93Fu56) and 3:1 mixture of both genotypes, Langfang, China, 2008–2009. S, monoculture of SZNC20B; SF, mixture of SZNC20B and 93Fu56; F, monoculture of 93Fu56.





**Fig. 5.** Temporal dynamics of whitefly biotype B in two monoculture pure stands (transgenic Bt SZNC20B and non-Bt 93Fu56) and 3:1 mixture of both genotypes, Langfang, China, 2008–2009. S, monoculture of SZNC20B; SF, mixture of SZNC20B and 93Fu56; F, monoculture of 93Fu56.

*Fusarium*-resistant genotype monoculture or the mixture.

**Predator Complex.** Arthropod predators frequently observed in cotton included ladybirds (Coleoptera: Coccinellinae), lacewings (Neuroptera: Chrysopidae), spider complex, and *Orius similis* (Hemiptera: Anthocoridae). For both years, genotype mixture pattern had no significant effect on seasonal temporal abundances of adult ladybird or *O. similis* (Table 2), but it significantly altered the relative abundance, or the ratio of prey and corresponding predator abundances (Table 3). Genotype mixture pattern greatly impacted larval ladybird, lacewing, and spider densities (Table 2), but their dominances were uninfluenced (Table 3). In addition, considerable temporal variation was observed in the number of predator species in 2008 (Table 2). In 2009, varietal mixture pattern treatments changed the abundances of larval ladybirds and spiders and the relative abundance of adult ladybirds (Tables 2 and 3). Averaged over 2 yr, cotton genotype mixture pattern exerted marked variable effects on the population dynamics of predators of various species and also within-species developmental stages, however there were exceptions, such as larval ladybirds. Furthermore, predator abundances differed significantly between years, and there was significant variation in the interactions between year and mixture pattern (Table 4).

**Discussion**

The widespread planting of crops genetically modified to express *Bacillus thuringiensis* (*Bt*) toxins for pest control may affect nontarget pests and soil-borne

disease incidence. This study examined the impacts of an intraspecies crop mixture on disease suppression and pest control, and the results demonstrate that a mixture of insect-resistant and disease-resistant cotton at the ratio 75 to 25% effectively suppressed the incidence rate and disease severity of *Fusarium* wilt. Results of this study support previous conclusions that intercropped genotype crop plantings can provide a promising strategy for disease control (Browning and Frey 1969, Natarajan et al. 1985, Newton 2009, Newton and Guy 2009, Newton et al. 2009, Zhu et al. 2000, Ganz and Ebert 2010).

The findings also show that row-mixtures of disease-resistant and insect-resistant cotton lines are equally effective in controlling cotton aphids. Feeny (1976) argued that plants in monocropped ecosystems are more visible, and thus more vulnerable, than those in natural ecosystems or in intercropped systems. In addition, Asman et al. (2001) predicted that phytophagous insects are more likely to find and remain on host plants growing in pure stands. As observed in this study, cotton aphids usually caused localized damage because of weak emigration of apterous individuals or alates (Wu and Guo 2003). Owing to area-restricted movement in monoculture setting, cotton aphid seasonal abundance is largely dependent upon early-stage colonization. Contrary to expectations, mixtures enhanced the abundances of mirid bugs and sweetpotato whitefly. This is consistent with Utsumi et al. (2011), wherein plant genotypic diversity increased the population size of an herbivorous insect.

Overall, the various effects of genotype mixtures on these pests indicate that the responses of pests to

**Table 2.** Mixed effects model statistics for temporal changes in predator abundances in cotton as affected by sample year and crop mixture pattern, Langfang, China

Predator species	Source of variation		
	Year	Mixture pattern	Year mixture pattern <sup>b</sup>
Adult ladybirds	F(1,15) = 552.66 <sup>d</sup>	F(2,15) = 5.50 <sup>b</sup>	F(2,15) = 5.94 <sup>b</sup>
Larval ladybirds	F(1,15) = 78.15 <sup>d</sup>	F(2,15) = 1.10 <sup>a</sup>	F(2,15) = 1.81 <sup>a</sup>
Adult lacewings	F(1,15) = 193.37 <sup>d</sup>	F(2,15) = 80.22 <sup>d</sup>	F(2,15) = 10.52 <sup>c</sup>
Larval lacewings	F(1,15) = 32.69 <sup>d</sup>	F(2,15) = 13.30 <sup>c</sup>	F(2,15) = 2.23 <sup>a</sup>
Spiders	F(1,15) = 141.03 <sup>d</sup>	F(2,15) = 25.94 <sup>d</sup>	F(2,15) = 0.42 <sup>a</sup>
<i>O. similis</i>	F(1,15) = 2147.05 <sup>d</sup>	F(2,15) = 4.34 <sup>c</sup>	F(2,15) = 31.92 <sup>d</sup>

<sup>a</sup> P > 0.05.

<sup>b</sup> 0.05 < P < 0.01.

<sup>c</sup> P < 0.01.

<sup>d</sup> P < 0.001.



**Table 3.** Mixed effect model statistics for temporal changes in predator abundances in cotton as affected by crop mixture pattern, block, and sample date, Langfang, China

Year	Factor	DF	Adult ladybirds	Larval ladybirds	Adult lacewings	Larval lacewings	Spiders	<i>O. similis</i>
2008	Pattern	2, 6	1.13 <sup>a</sup>	6.42 <sup>b</sup>	69.61 <sup>d</sup>	8.00 <sup>b</sup>	13.61 <sup>c</sup>	2.06 <sup>a</sup>
	Block	3, 6	2.92 <sup>a</sup>	0.07 <sup>a</sup>	2.26 <sup>a</sup>	0.09 <sup>a</sup>	0.25 <sup>a</sup>	0.05 <sup>a</sup>
	Date	9, 99	178.63 <sup>d</sup>	283.92 <sup>d</sup>	31.46 <sup>d</sup>	24.88 <sup>d</sup>	221.70 <sup>d</sup>	54.17 <sup>d</sup>
2009	Pattern	2, 6	0.96 <sup>a</sup>	15.01 <sup>c</sup>	0.76 <sup>a</sup>	4.85 <sup>a</sup>	9.14 <sup>b</sup>	0.41 <sup>a</sup>
	Block	3, 6	1.45 <sup>a</sup>	0.90 <sup>a</sup>	1.52 <sup>a</sup>	0.24 <sup>a</sup>	0.04 <sup>a</sup>	1.28 <sup>a</sup>
	Date	9, 99	34.64 <sup>d</sup>	429.63 <sup>d</sup>	24.71 <sup>d</sup>	14.14 <sup>d</sup>	132.81 <sup>d</sup>	31.22 <sup>d</sup>

<sup>a</sup> P > 0.05.<sup>b</sup> 0.05 < P < 0.01.<sup>c</sup> P < 0.01.<sup>d</sup> P < 0.001.

plant genotypic diversity are species-specific. This casts new light on how crop diversity is useful in pest control. Early studies of induced responses to herbivores noted that pests sometimes moved away from locally damaged plant parts and fed preferentially on undamaged tissues (Edwards and Wratten 1983). Thus, factors leading to the resurgence of mirid bugs and sweetpotato whiteflies were likely reduced competition and lower induced resistance from cotton aphids and pests of other species in mixture plots. Although cotton aphids are largely sedentary, mirid bugs and sweetpotato whiteflies feed more freely throughout the crop mixture. In addition, Bernays (2001) proposed that the ability to make suitable foraging choices is likely to vary among insect species resulting from the variability in their sensory and information-processing capacities. Therefore, the distinct responses of aphids, mirid bugs, and sweetpotato whiteflies may be directly related to the difference in foraging capability.

Field observations showed higher or comparable predator abundances in mixed plots versus monoculture fields. Enhanced predator numbers might be linked to correlation between prey abundance, predator diversity, and plant diversity. However, correlation analyses of our data suggested that prey abundance was a poor predictor for predator abundance. However, studies have demonstrated positive correlation between plant diversity and predator diversity (Haddad et al. 2009). In addition, Takizawa and Snyder (2011) hypothesized that predator biodiversity increased juvenile survivorship. Given this notion,

higher predator abundance may be partly explained by weak intraguild predation. Obtained data also revealed differential natural enemy responses to plant diversity (Tables 1, 2). Snyder et al. (2006) suggested that diverse predator communities produced more juvenile predators than did single-predator-species communities, which is supported by enhanced abundances of larval ladybirds and larval lacewings in this study.

One limitation of this study was failure to reject the possibility that the observed effects of intraspecific crop mixture on pest and disease incidence may have been compounded by microclimate alteration. Further exploration is warranted to elucidate the underlying mechanisms of the observed effects. Nevertheless, as far as is known, this report is the first to explore the feasibility of using habitat diversification, mediated by intraspecific crop mixture, for simultaneous control of nontarget arthropod pests and soil-borne diseases. The efficacy of refuge in delaying or mitigating resistance is common knowledge, and until now, it has been a primary factor in recommendations for avoiding secondary pest outbreaks. Refuge established via intraspecific intercropping in this study decreased the cotton aphid abundance and significantly reduced the incidence rate and disease index of *Fusarium* wilt. These findings demonstrate that plant genotypic diversity is an effective pest management approach. The negative effects of mixture on mirid bug and whitefly biocontrol warrants further study of the species-specific response.

**Table 4.** Mixed effects model statistics for temporal change in relative predator abundance in cotton as affected by crop mixture pattern, block, and sample date, Langfang, China

Year	Factor	df	Adult ladybirds	Larval ladybirds	Adult lacewings	Larval lacewings	Spiders	<i>O. similis</i>
2008	Pattern	2, 6	6.78 <sup>b</sup>	4.55 <sup>a</sup>	5.01 <sup>a</sup>	3.28 <sup>a</sup>	3.95 <sup>a</sup>	9.02 <sup>b</sup>
	Block	3, 6	0.15 <sup>a</sup>	0.07 <sup>a</sup>	3.27 <sup>a</sup>	0.44 <sup>a</sup>	0.48 <sup>a</sup>	0.07 <sup>a</sup>
	Date	9, 99	28.12 <sup>d</sup>	142.40 <sup>d</sup>	18.19 <sup>d</sup>	10.95 <sup>d</sup>	582.12 <sup>d</sup>	30.71 <sup>d</sup>
2009	Pattern	2, 6	6.34 <sup>b</sup>	2.10 <sup>a</sup>	1.43 <sup>a</sup>	1.54 <sup>a</sup>	0.02 <sup>a</sup>	0.90 <sup>a</sup>
	Block	3, 6	0.79 <sup>a</sup>	0.83 <sup>a</sup>	4.36 <sup>a</sup>	0.65 <sup>a</sup>	5.16 <sup>b</sup>	1.30 <sup>a</sup>
	Date	9, 99	26.61 <sup>d</sup>	74.73 <sup>d</sup>	12.32 <sup>d</sup>	7.40 <sup>d</sup>	263.71 <sup>d</sup>	25.72 <sup>d</sup>

<sup>a</sup> P > 0.05.<sup>b</sup> 0.05 < P < 0.01.<sup>c</sup> P < 0.01.<sup>d</sup> P < 0.001.

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## ORIGINAL ARTICLE

# Potential cotton aphid, *Aphis gossypii*, population suppression by arthropod predators in upland cotton

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**Abstract** The cotton aphid, *Aphis gossypii* Glover, predation rate of convergent lady beetle, *Hippodamia convergens* Guerin-Meneville, was determined by assigning a single predator randomly to each of four prey density treatments in the laboratory. Prey densities included 25, 50, 100, and 200 aphids per Petri dish arena. Predation response was recorded at 1, 4, 8, 16, 24, and 48 h after assigning predators to their prey treatments. Rate of consumption increased through time, with all 25 aphids consumed during the first 4 h of the experiment. At the highest density, adult lady beetle consumed on average 49, 99, 131, 163, 183, and 200 aphids within 1, 4, 8, 16, 24 and 48 h, respectively. Predators showed a curvilinear feeding response in relation to total available time, indicating that convergent lady beetles have the potential to suppress larger populations of aphids through continuous feeding by regulating their predation efficiency during feeding. The analysis of age-specific mortality in absence of prey revealed that lady beetles could survive for an extended period of time (more than 2 weeks) without prey. The ability of a predator to survive without prey delays or prevents the rebound of pest populations that is a significant factor in natural biological control. A two-year field sampling of 10 cotton arthropod predator species showed that spiders (27%) were the most dominant foliage dwelling predators in the Texas High Plains cotton followed by convergent lady beetles (23.5%), hooded beetles (13.5%), minute pirate bugs (11%), green lacewings (9.5%), bigeyed bugs (7.5%), scymnus beetles (3%), soft-winged flower beetles (2%), damsel bugs (1.5%), and assassin bugs (1.5%). A field cage study showed that one *H. convergens* adult per plant released at prey density of one aphid per leaf kept the aphid population below economic threshold for the entire growing season.

**Key words** *Aphis gossypii*, biological control, cotton IPM, functional response, *Hippodamia convergens*, natural suppression

## Introduction

The cotton aphid, *Aphis gossypii* Glover (Hemiptera: Aphididae), is one of the important pests of cotton in the Texas High Plains as well as in most United States cotton producing regions, causing an overall 4% of the insect-

induced lint yield reduction annually (Williams, 2001–2011). Several insect management practices have been developed to suppress cotton aphids, but cotton growers usually apply insecticides for aphid control in cotton. Applications of pyrethroid insecticides for the control of other cotton pests such as *Lygus* and cotton fleahoppers have the potential to cause rapid outbreaks of cotton aphids (Godfrey *et al.*, 2000). Most aphid outbreaks are associated with the application of insecticides, which kill naturally occurring predators and parasitoids and pyrethroids also physiologically interacts with cotton leaves enhancing cotton aphid reproduction (Kidd &

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Rummel, 1997). Arthropod predators can have a significant role in cotton aphid suppression; therefore, pest management programs should be designed to conserve and enhance natural enemies (Parajulee *et al.*, 1997).

The lady beetles have been considered the most important group of arthropod predators in cotton agroecosystems in Texas (Parajulee *et al.*, 1994; Slosser *et al.*, 1998; Parajulee & Slosser, 1999). Parajulee *et al.* (1994) documented that the lady beetle complex comprised 75% of the total predators in irrigated cotton during a three-year study in the Rolling Plains region of Texas, and Slosser *et al.* (1998) observed that lady beetles comprised 62% of the total predators in dryland cotton during a five-year study in the Rolling Plains region. However, the proportion of lady beetles in the total predator complex in cotton can be influenced by vegetation diversity and weather patterns. Previous studies have demonstrated up to 81% increase in lady beetles in the total predator complex when the cotton system was diversified with noncotton strip crops (Parajulee & Slosser, 1999). These authors also reported a significant decline in lady beetle abundance during extreme environments; lady beetles comprised only 25% and 18% of the total predators during the hot, dry summer of 1996 and milder, wet summer of 1997, respectively. These reports suggest that the value of lady beetles as natural control agents depend on climatic variation and habitat diversity. Of seven species of lady beetles reported from the Texas Rolling Plains and High Plains, the convergent lady beetle, *Hippodamia convergens* Guerin-Meneville, is perhaps the most important predator of cotton aphids in Texas cotton. Slosser *et al.* (1998) reported that *H. convergens* was the dominant lady beetle species and lady beetles had been considered a significant natural control agent influencing the rate of cotton aphid decline in late August in the northern Rolling Plains. However, data on consumption rate of cotton aphids by lady beetles are not available for use in the development of a decision rule system that relates abundance of lady beetles to expected rate of cotton aphid suppression. The lack of a decision rule system has limited our ability to quantify the contribution of biological and ecological approaches in pest suppression. However, the development of a decision rule system requires data on predator consumption rate (Parajulee *et al.*, 2006b), predator–prey behavior with respect to crop phenology (Berryman, 1992), and predator–prey ratios for effective pest suppression (Gutierrez, 1992).

This study was designed to generate information toward developing a decision rule system, specifically targeting the lady beetle–cotton aphid system. Laboratory feeding studies and field cage studies were conducted to quantify the predation potential of convergent lady beetles on cotton aphids. In addition, a field survey study was

conducted to quantify the population dynamics of cotton aphids and other related arthropod predators in the Texas High Plains cotton system. This paper provides the baseline data related to the predator–prey relationship between convergent lady beetle and cotton aphids. Development of a decision rule system for a given predator or predator guild will be the necessary first step toward further development of a biologically and ecologically based cotton IPM system.

## Materials and methods

### *Laboratory consumption rate and survivorship of lady beetle in relation to prey density*

**Cotton aphid consumption study** Before conducting a lady beetle adult functional response (prey aphid density dependent feeding rate) study, a laboratory study was conducted to evaluate the rate of aphid consumption by lady beetle larva and adults. Lady beetle eggs were collected from the cotton field and kept at room temperature (27°C) until they hatched. Lady beetle larvae were fed on cotton aphids collected from an upland cotton field. Small lady beetle larva (2nd instar), medium larva (3rd instar), and adult (1 week after adult emergence) specimens were used in this aphid consumption study. Large larvae were not evaluated because most of them pupated by the end of the experiment. Six small larva, six medium larva, and six adult lady beetles were individually confined in Petri dishes (10 cm diameter). They were starved (deprived of prey and water) for 48 h and cotton aphids were provided *ad libitum* (100 per insect). The experiment was conducted in a growth chamber maintained at 26.6°C. Lady beetles were allowed to feed on cotton aphids for 12 h and the number of consumed or missing cotton aphids were recorded. The average number of cotton aphids consumed during the 12 h period was calculated and cotton aphid consumption rate of different stages of lady beetles were analyzed with ANOVA (analysis of variance) and means were separated with least significant difference (PROC GLM; SAS Institute, 2010).

**Adult lady beetle functional response study** Lady beetle versus cotton aphid functional response study was designed based on the consumption rate study (above). Convergent lady beetle pupae were collected from a cotton field near Lamesa, Texas on August 6, 2002, brought to the laboratory, and reared to adulthood. Adult beetles were reared on cotton aphids. After the beetles were fed on cotton aphids for four days, they were individually confined in Petri dishes (10 cm diameter) and were starved



(deprived of prey and water) for 48 h. The experiment was conducted in a growth chamber maintained at 26.6°C and 12 h photoperiod. Single lady beetles were randomly assigned to one of four prey density treatments in a Petri dish with a single cotton leaf to measure its rate of cotton aphid predation. Prey density treatments included 25, 50, 100, and 200 aphids per Petri dish arena, with 15 replications.

Cotton aphids were collected from the cotton field, and within 1 h following collections aphids were offered to predators on the same detached cotton leaves from which the prey were collected. The total number of aphids consumed, hereinafter referred to as *response*, was recorded 1, 4, 8, 16, 24, and 48 h after releasing predators to a treatment. After 48 h, predators were again deprived of prey/water and monitored daily for their age-specific survivorship without access to prey and water.

Response data were fitted to Holling's curvilinear Type II model (Holling, 1959) using TableCurve 2D<sup>®</sup> software (Jandel Scientific, 2002). In this model, the number of aphids consumed ( $N_c$ ) is a hyperbolic function of aphid density ( $N_0$ ), as described by the equation  $N_c = a'T_a N_0 / (1 + a'T_h N_0)$ , in which  $a'$  is the predator's rate of prey discovery,  $T_a$  is the total time available (1–48 h in our case), and  $T_h$  is the prey handling/feeding time.

**Lady beetle survivorship study** Convergent lady beetle pupae were collected from cotton fields and reared in the laboratory until adult emergence. Adult lady beetles were reared on cotton aphids collected from the same cotton field as were lady beetle pupae. Thirty lady beetles (7-day old adults) were starved for 24 h and then were fed with three different quantities of cotton aphids (50, 100, and 200 aphids). Ten adult lady beetles were randomly assigned to each level of prey supply. Once they consumed all cotton aphids, beetles were kept in an incubator at 26°C without prey and water for 20 days (until the last individual from the cohort died). The survivorship and mortality of lady beetle adults were recorded every day for 20 days by visual observation. The age-specific survivorship rate data of lady beetles fed with different densities of aphids were modeled with a third degree polynomial equation:  $Y = aX^3 + bX^2 + cX + d$ , where 'Y' is the age-specific percent survivorship and 'X' is the predator age in days.

#### *Cotton aphid suppression by convergent lady beetle in cotton field cages*

A field cage study was conducted in a cotton field near Lubbock in 2003. Cotton cultivar "PM 2379 RR" was planted on 1.02-m spaced rows during the second week of May, a typical optimum planting window for the Texas

High Plains cotton. The field cage study was deployed in a completely randomized design with three levels of predator–prey ratio treatments and three replications. The three predator–prey treatment ratios were (i) 1 : 1 or one convergent ladybeetle larva per cotton plant when there was 1 aphid per leaf, (ii) 6 : 50 or six lady beetle larvae per plant when cotton aphid density increased to approximately 50 aphids per leaf, and (iii) 0 : 50 control or no lady beetle larva introduced and aphid populations were allowed to increase unchecked. These ratios were selected to represent grower field observations of low and high predator–prey ratios during the cotton growing season.

Twelve plants (two rows and six plants per row) were enclosed in each of the nine Lumite<sup>®</sup> cages (1.8 m × 1.8 m × 1.8 m; 32 mesh per inch) at presquaring stage of cotton. Plants in each cage were manually cleaned by removing all insect larva and eggs three times at 3–4 day intervals to ensure no resident predators were inside the cages. Cotton aphid colony was maintained on cotton plants grown in laboratory growth chambers. Cotton plants in all cages were artificially infested with cotton aphids at the rate of one aphid per leaf. Out of nine cages, three randomly selected aphid infested cages were kept without any predators as control cages, whereas the remaining six cages received two predator prey-ratio treatments.

Approximately, third-instar convergent lady beetle larvae were collected from a cotton field one day prior to release in field cages. Control cages did not receive any predators and 1:1 predator–prey treatment cages received one predator per plant 24 h after the introduction of cotton aphid prey into the cage (August 21, 2003). The remaining three cages were monitored at 2–3 day intervals for cotton aphids to determine the density of aphids (50 per leaf) for predator treatment deployment for the high density treatment. Once the aphid density increased to approximately 50 aphids per leaf, six lady beetle larvae per plant were released (September 2, 2003). After the deployment of predator–prey ratio treatments, cotton aphid populations were monitored by counting aphids on all 12 plants in each treatment cage at weekly intervals for the next 5 weeks. When the average cotton aphid population exceeded 100 aphids per leaf, sampling was modified to count aphids on only six leaves per plant (1 main-stem leaf and 1 branch leaf from each strata (upper, middle, and lower) of the cotton plant and only six plants per cage were sampled. Aphid observations were performed *in situ* without removing any plant leaves or aphids from the cages. Due care was taken not to allow the entry of other insect predators and pests into the cages. Cotton cultivation and management inside the cage was similar to the field immediately outside the cage. The cotton aphid data were analyzed with analysis of variance (ANOVA) using PROC GLM, SAS 9.2 (SAS

institute, 2010) and means were separated by using LSD procedure at the 0.05 significance level.

#### *Arthropod predator diversity and cotton aphid–predator population dynamics in the field*

A 2-year field survey study was conducted at the Agricultural Complex for Advanced Research and Extension Systems (AG-CARES) farm, near Lamesa, Texas. The Roundup Ready cotton cultivar PM 2326 RR, similar to that in the Lubbock field cage study, was planted in three plots (16.3 m × 30.5 m) during the second week of May (May 8, 2002 and May 9, 2003). Cotton was planted on rows with a 1.02-m row spacing with a targeted plant density of 153 000 plants per ha in sandy loam soil without an application of in-furrow insecticide. Cotton seed was also not treated with insecticides. The crop was irrigated (35 cm in 2002 and 21 cm in 2003) utilizing a center pivot system equipped with low energy precision application (LEPA) irrigation nozzles and drag socks. Herbicides were used when needed. Herbicide use included one application of trifluralin (Treflan®) at 0.6 kg AI/ha. Each study year, the cotton received fertilizer applications at the rate of 112–38–0 kg N–P–K per ha.

Arthropod predators were sampled weekly for 17 weeks from cotton emergence until 90% open-boll stage (mid-May to early October). Cotton aphids were sampled only for 11 weeks beginning from mid-July to early October. Randomly selected 10 plants per location were thoroughly observed visually in the field and pest and predator numbers were recorded. Ten predator species were sampled that included predaceous bugs (minute pirate bug *Orius* spp., damsel bug *Nabis* spp., big-eyed bug *Geocoris* spp., wheel bug *Arilis cristatus*, and assassin bug *Zelus renardii*), and predaceous beetles (lady beetle *Hippodamia convergens*, collops or soft-winged flower beetle *Collops* spp., scymnus beetle *Scymnus loewii*, and hooded beetle *Notoxus* spp., green lacewing *Chrysoperla* spp.), and spiders (predominantly *Misumenops* spp.). Only cotton aphids (*Aphis gossypii*) were sampled among the pest species.

Aphids and predators were sampled via a visual sampling method, counting aphids *in situ* on randomly picked 20 cotton leaves per plot, one leaf per plant and 20 plants per plot representing all leaf ages throughout the plant canopy, and observing predators on 5–10 whole plants per plot. Both mature and immature stages of each species were observed and recorded separately and combined to get the total number of arthropods per plant for further statistical analysis. Data were analyzed to characterize

**Table 1** Cotton aphid consumption rate (number of cotton aphids consumed per 12 h) of lady beetle larva and adults (mean ± SE) in the laboratory.

Lady beetle stage	Aphid consumption rate	
	Mean ± SE	Range
Adult	91.33 (4.98) a	67–100
Medium larvae	56.20 (8.39) b	26–75
Small larvae	12.40 (2.01) c	5–17

the species composition (%) and diversity (Shannon & Weaver, 1949) of arthropod predator complex and to establish the predator–prey functional relationship in the field.

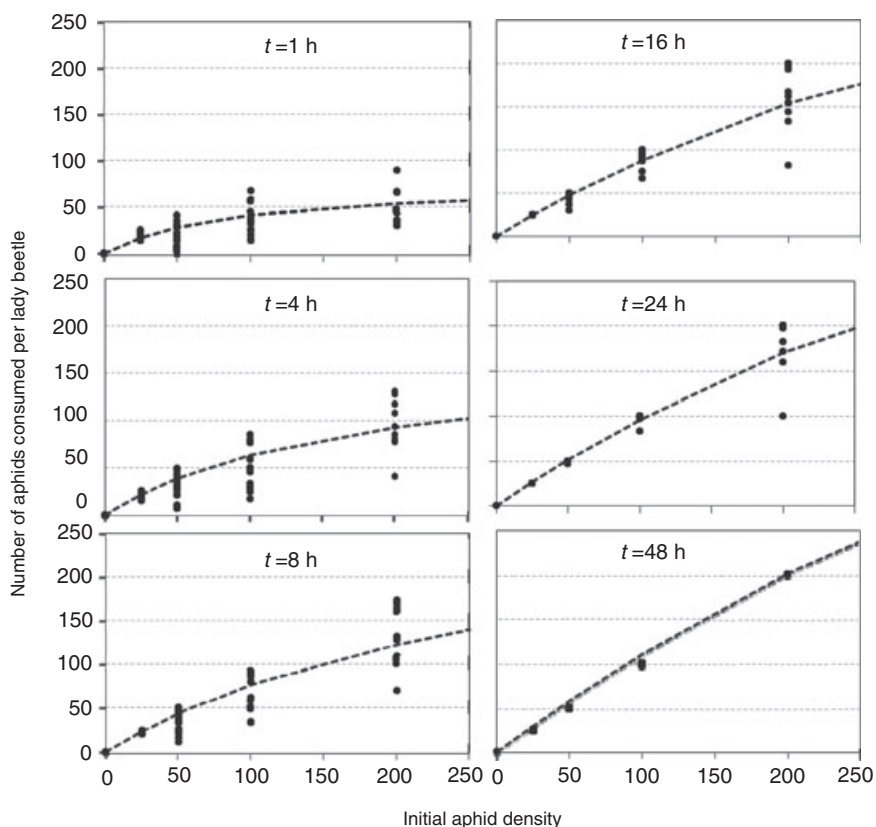
## Results and discussion

### *Laboratory consumption rate, functional response, and survivorship studies*

**Consumption rate** Predation rates of *Hippodamia convergens* adults showed the highest rate of cotton aphid consumption (91 aphids/12 h), followed by *H. convergens* medium sized larva (56 aphids/12 h), and *H. convergens* small larva (12 aphids/12 h) (Table 1). As lady beetle larva matured, they increased in size and also consumed a larger number of cotton aphids as they needed more energy for their growth and development. Cotton aphid predation rates of lady beetle adults and immatures were significantly different ( $df = 2, 16; f = 49.5$ , and  $P < 0.001$ ) (Table 1). The cotton aphid consumption rate of lady beetle adults was sevenfold greater than that of small larva. A similar phenomenon was observed in another lady beetle species, *Harmonia axyridis* (Pallas), in which Lee and Kang (2004) reported 10-fold greater cotton aphid consumption rate of adult lady beetles as compared with that for young second instar larva. Thus, age structure of the lady beetle population is an important factor in cotton aphid population suppression and it must be considered during quantification of the cotton aphid suppression by lady beetles in cotton.

**Functional response** Predation by convergent lady beetle adult on different densities of cotton aphids showed curvilinear Type II responses for all tested predation time periods (Fig. 1). As the prey density (cotton aphid availability) increased from 25 to 200 aphids per leaf, the aphid predation efficiency of the lady beetle adults decreased significantly in all six predation times (Fig. 1).

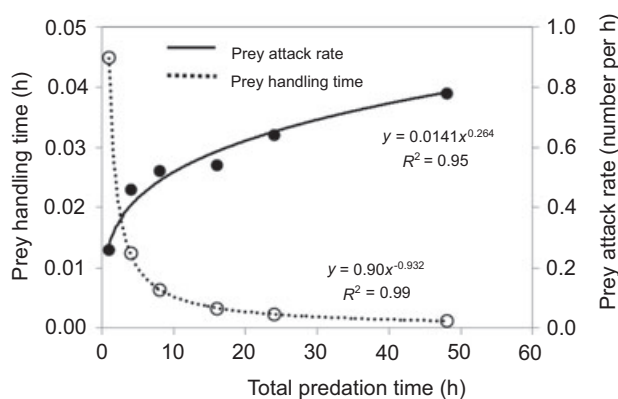




**Fig. 1** Cotton aphid consumption rate of lady beetle adults as a function of aphid density and time in a laboratory study;  $t$  = total predation time.

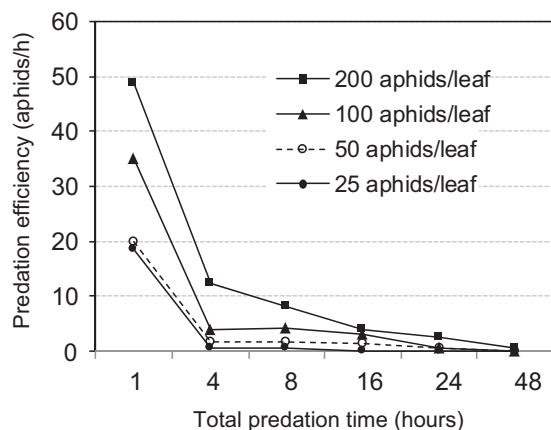
Lady beetle adults consumed 200 cotton aphids within 48 h of total predation time available. Total number of aphids consumed per hour by a single lady beetle increased as prey density increased. When the aphid density is increased, aphids become more readily available so the time needed to search and find an aphid prey is reduced. In other words, under higher prey densities, predators require less time searching for the prey, thus it increases the number of prey encountered and consumed within a given time (Hollings, 1959). However, the rate of increase in predation efficiency was not proportionate to the rate of increase in prey density. The lower rate of increase in predation rate with increased prey density might be due to two reasons: (i) the satiation effect and (ii) increasing prey handling time as the prey density increased. Lady beetle adults in this study displayed an inverse relationship between prey handling time and prey attack rate or searching time (Fig. 2).

The cumulative number of cotton aphids consumed by a lady beetle increased as total predation time increased up to 48 h. In other words, lady beetles continued consuming cotton aphids, with no complete satiation, for at least 48 h.



**Fig. 2** Cotton aphid handling time and attack rate of lady beetle adults as a function of total available predation time in the laboratory.

Nevertheless, the predation rate was significantly reduced as cumulative consumption of cotton aphids increased (Fig. 3), indicating the influence of satiation on predation efficiency.



**Fig. 3** Effect of satiation in predation efficiency of a lady beetle as a function of prey density and available predation time in the laboratory.

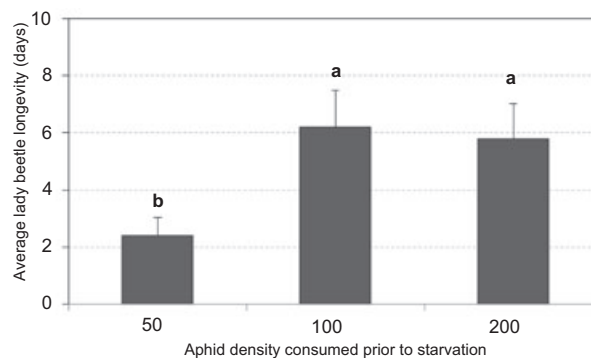
At the highest prey density (200 aphids/leaf), lady beetles consumed an average of 49 aphids in 1 h, 99 aphids in 4 h, 131 aphids in 8 h, 163 aphids in 16 h, 183 aphids in 24 h, and all 200 aphids in 48 h (Fig. 1). These data clearly demonstrate that the convergent lady beetle is a very potent predator that can suppress a large aphid population in a short period of time. Most other cotton insect predator species do not feed for an extended period of time after they satiate from their initial prey consumption. However, the convergent lady beetle fed continuously for 48 h until all the aphids in the highest prey density were consumed. These results are in general agreement with that reported by Dreistadt and Flint (1996) who found a density-dependent functional response of convergent lady beetles when fed cotton aphids infesting potted chrysanthemum.

The prey handling time determines the predation efficiency (Hollings, 1959; Wiedenmann & O'Neil, 1991). In our study, total number of aphids consumed increased continuously as total predation time increased. Lady beetle adults displayed longer prey handling time as the predation duration increased, which could largely be attributed to the onset of satiation as feeding duration increased, as depicted by a curvilinear response for prey handling time versus total available predation time (Fig. 2). Predators handled their prey more slowly with increased total available time ( $T_a$ ) up to 16 h, indicating a gradual decline in their feeding efficiency per unit time and moving toward satiation. However, lady beetles resumed accelerated rate of consumption without reaching complete satiation, which may increase their feeding efficiency to the level they had at initial hours of predation. This assertion has

not been evaluated and the amount of lag time it takes to increase its predation efficiency back to the original level has not been quantified. Higher densities and longer predation times need to be investigated to address this issue.

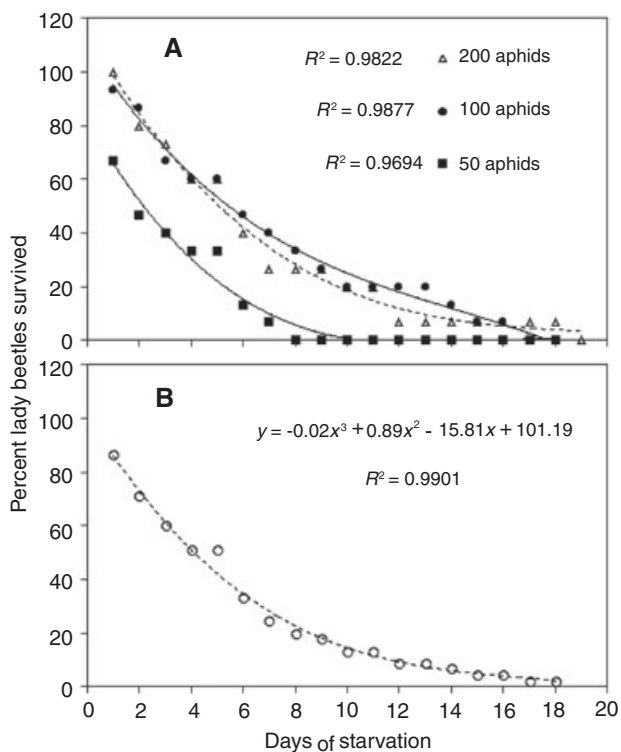
These data clearly indicate that convergent lady beetles possess potential to suppress larger populations of cotton aphids through continuous feeding by adjusting their predation efficiencies during feeding. However, the laboratory consumption rate can only serve as the maximum potential of lady beetles suppressing the cotton aphid population in an artificially ideal environment; the actual functional response and predation rates in field settings would invariably be much different (Wiedenmann & O'Neil, 1991; Parajulee *et al.*, 1994). Several factors including weather parameters, plant architecture, prey quantity or quality, and prey age structure can influence the predation behavior of natural enemies *in situ*. The birth and death rates of both prey and predators and their interactions with other arthropods in the community would determine the cotton aphid population suppression by lady beetles in the cotton field.

**Predator survivorship in absence of prey** Adult lady beetles survived an extended period of time following a total starvation (no prey or water). Although the average survivorship was 5 days, few lady beetles survived up to 19 days of starvation. The longevity of the beetles that consumed 100 and 200 aphids before starvation was significantly higher (6 days,  $df = 2,15$ ;  $f = 3.66$ ,  $P = 0.03$ ) than the beetles that had consumed only 50 aphids (2.4 days) (Fig. 4). The mortality of lady beetles fed with 50, 100, and 200 aphids reached 100% on day 9, 18, and 19, respectively (Fig. 5). The average survivorship curve

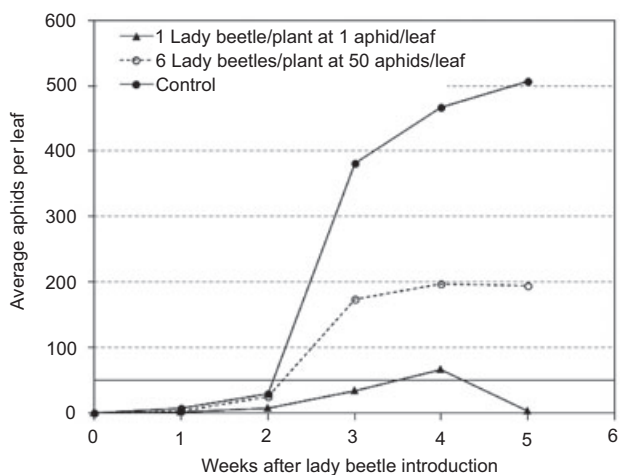


**Fig. 4** Average (+SE) longevity of adult lady beetles without prey or water after consuming 50, 100, or 200 aphids per beetle in the laboratory.

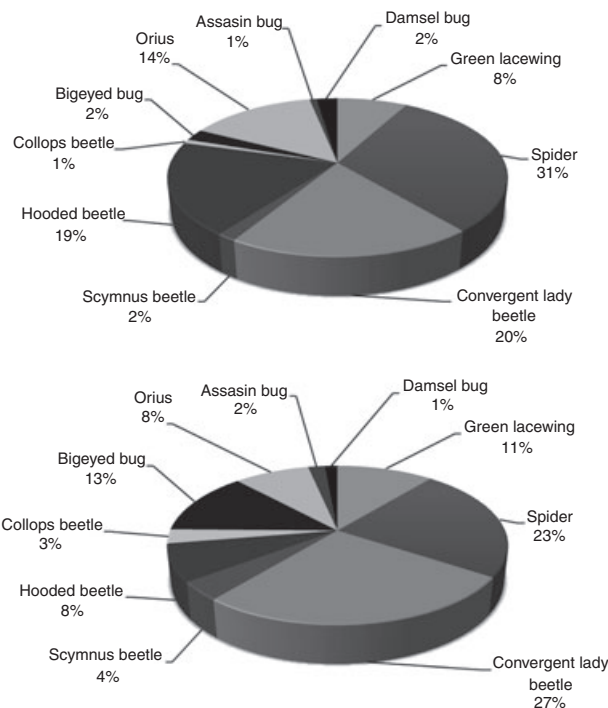
Predator suppression of cotton aphids in upland cotton



**Fig. 5** Lady beetle age-specific survivorship affected by number of prey available before the onset of starvation. (A) Convergent lady beetle survivorships after predation on various densities of cotton aphids. (B) Convergent lady beetle survivorship averaged over all feeding densities. Lines represent lady beetle percent survivorship predicted by a third-order polynomial equation.



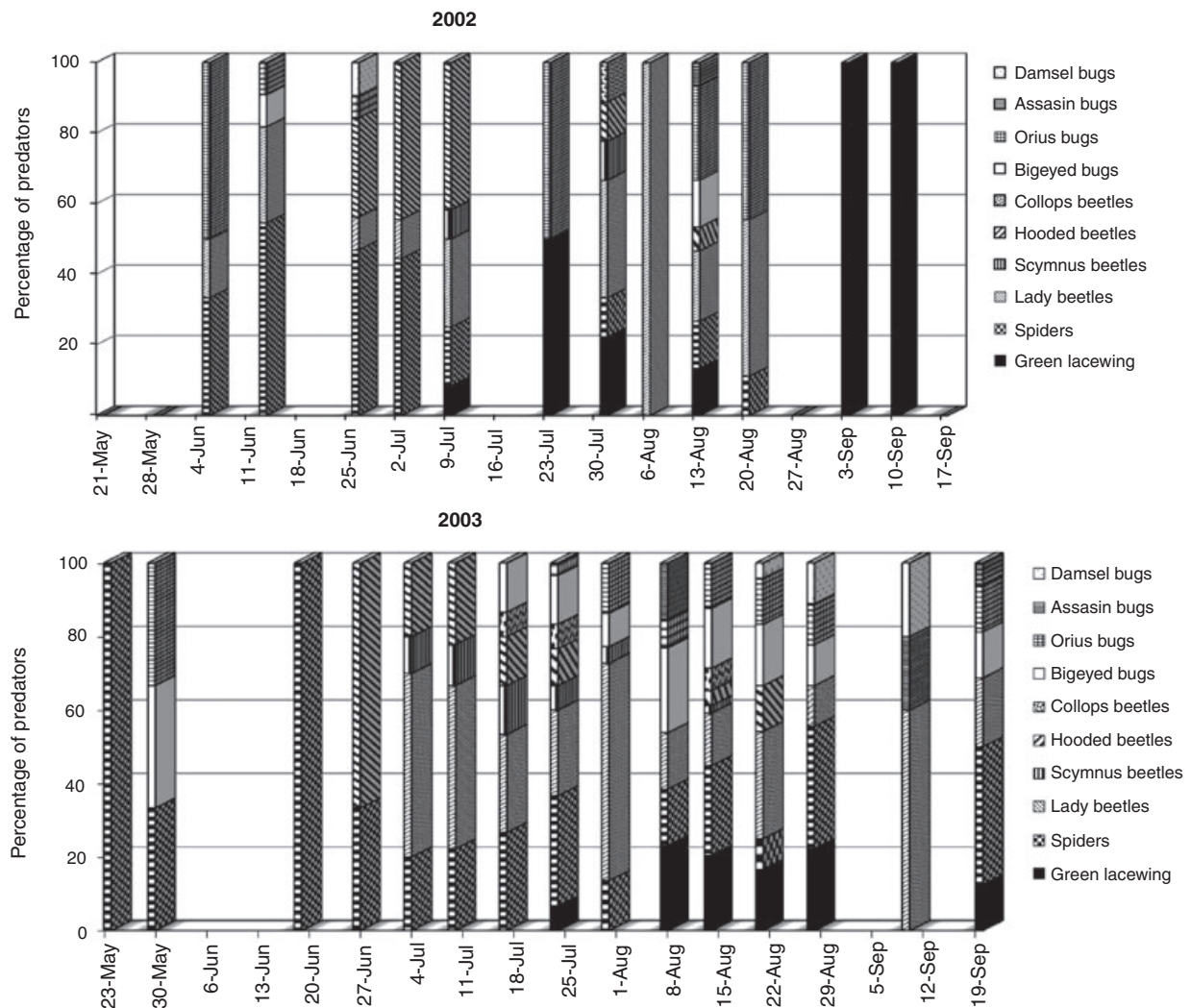
**Fig. 6** Cotton aphid population suppression by lady beetles in a field cage study, Lubbock, Texas.



**Fig. 7** Seasonal species composition of arthropod predators in an upland cotton field, Lamesa, Texas, 2002–2003.

(Fig. 5) showed that >24% of the predators survived past one week with no prey or water, whereas 7% survived 14 days. A third-degree polynomial equation modeled the predator survivorship in absence of prey, with high degree of predictability for all feeding densities before the onset of starvation ( $r^2$  ranged from 0.96–0.98).

Because adult convergent lady beetles disperse quickly to new locations in response to fluctuating prey densities and the density-dependent mortality is not a significant population regulatory factor in nature (Schellhorn & Andow, 1999), the ability to survive without prey becomes critical. Also, the ability of adult insects to tolerate a long period of starvation may reduce the degree of cannibalism to conspecific immatures (Parajulee & Phillips, 1995). Our study suggests that the amount of prey consumed before the onset of starvation affects the duration of survivorship in the absence of prey. The ability of a predator to survive without prey for an extended period of time is a significant factor in natural biological control as it delays or prevents the rebound of pest populations. In the field, convergent lady beetles usually disperse after aphid population vanished. However, the extended survivorship allows the dispersing predators sufficient time to locate another prey source.



**Fig. 8** Temporal dynamics of species composition of predatory arthropods in an upland cotton field, Lamesa, Texas, 2002–2003.

#### *Aphid population suppression by convergent lady beetle in field cages*

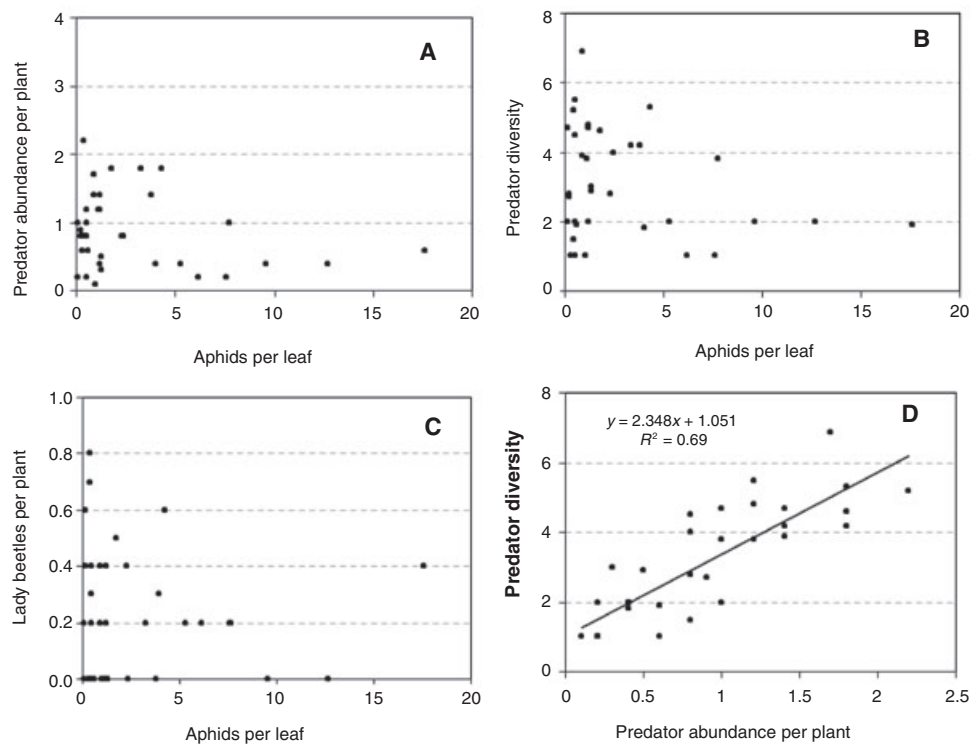
Cotton aphid density was significantly ( $df = 2,6$ ;  $f = 7.95$ ,  $P = 0.001$ ) lower in lady beetle introduced cages as compared to that in control cages (no lady beetles introduced). Lady beetle larva suppressed cotton aphid populations more effectively when predators were introduced at lower prey density (1 aphid per leaf) compared to when the prey densities were around the current regional treatment threshold (50 aphids per leaf) (Fig. 6). One lady beetle larva per plant released at the prey density of one aphid per leaf kept the aphid population below economic threshold (i.e., <50 aphids per leaf) for the entire growing season, except for the fourth week after the lady beetle release. Aphid population suppression efficiency of lady

beetle larva decreased when aphid populations increased to 50 aphids per leaf before the introduction of predator larvae, resulting in aphid population of above threshold level during most of the sampling period. Reduced cotton aphid suppression rate of lady beetle was partly due to the large number of rapidly reproducing mature aphids overwhelming aphid predation ability of lady beetles in the cages.

#### *Predator species diversity and cotton aphid population abundance in cotton field*

**Predator species composition, abundance, and diversity** Arthropod predator activity in cotton was low in both years (2002 and 2003). A total of 730 cotton plants





**Fig. 9** Predator–prey field population dynamics (A and C) and relationships between predator diversity (Shannon's diversity indices) and cotton aphid abundance (B) and predator abundance (D) in upland cotton, Lamesa, Texas, 2002–2003 ( $n = 35$  sample points, each consisting of data from 5–10 cotton plants).

(335 in 2002 and 395 in 2003) were visually inspected for arthropods and a seasonal total of 336 arthropod predators (126 in 2002 and 210 in 2003) were found inhabiting those plants. Figure 7 shows the yearly percentages of predator species for 2002 and 2003. The two-year combined data of ten arthropod predator species showed that spiders (27%) were the most dominant foliage dwelling predators followed by convergent lady beetles (23.5%), hooded beetles (13.5%), minute pirate bugs (11%), green lacewings (9.5%), bigeyed bugs (7.5%), scymnus beetles (3%), collops or soft-winged flower beetles (2%), damsel bugs (1.5%), and assassin bugs (1.5%). Predator activity began soon after cotton germination during cotyledon or 1 true leaf cotton stage. Crab spiders, minute pirate bugs and lady beetles were the first to colonize the cotton field, which is a common phenomenon in the Texas High Plains (Shrestha, 2004). Spiders and lady beetles remained active in the cotton field throughout the growing season, whereas green lacewings were observed as late season predators (Fig. 8). A strong linear relationship between total predator abundance and predator diversity was observed (Fig. 9). As the total number of predators increased, the diversity of predators also increased. Predator abundance and associated predator diversity were low

at the beginning of the season, whereas both abundance and diversity increased as the cotton season progressed. Maximum peak predator abundance and diversity were recorded in mid-August (100 days after planting) when cotton was at early boll development stage (Fig. 10). Predator abundance and diversity are generally influenced by prey availability in the field. The increased thrips and cotton aphids during cotton blooming and early boll development period served as good source of prey that might have attracted and retained large and varied number of predators in cotton (Parajulee *et al.*, 2006a).

**Cotton aphid population dynamics in relation to predator abundance in the field** In 2002, aphid activity began in late July at cotton blooming stage (76 DAP) and the number increased rapidly to reach the peak population (5.8 per leaf) in early August at boll development stage (92 DAP) (Fig. 11). After this peak, the aphid numbers declined rapidly. In 2003, however, aphid activity began in early July (55 DAP) and densities increased to reach the peak (11.1 per leaf) in early September with greater densities and extended peak than in 2002. As a result, average seasonal abundance of cotton aphids was significantly higher in 2003 (3.9/leaf) compared with that in

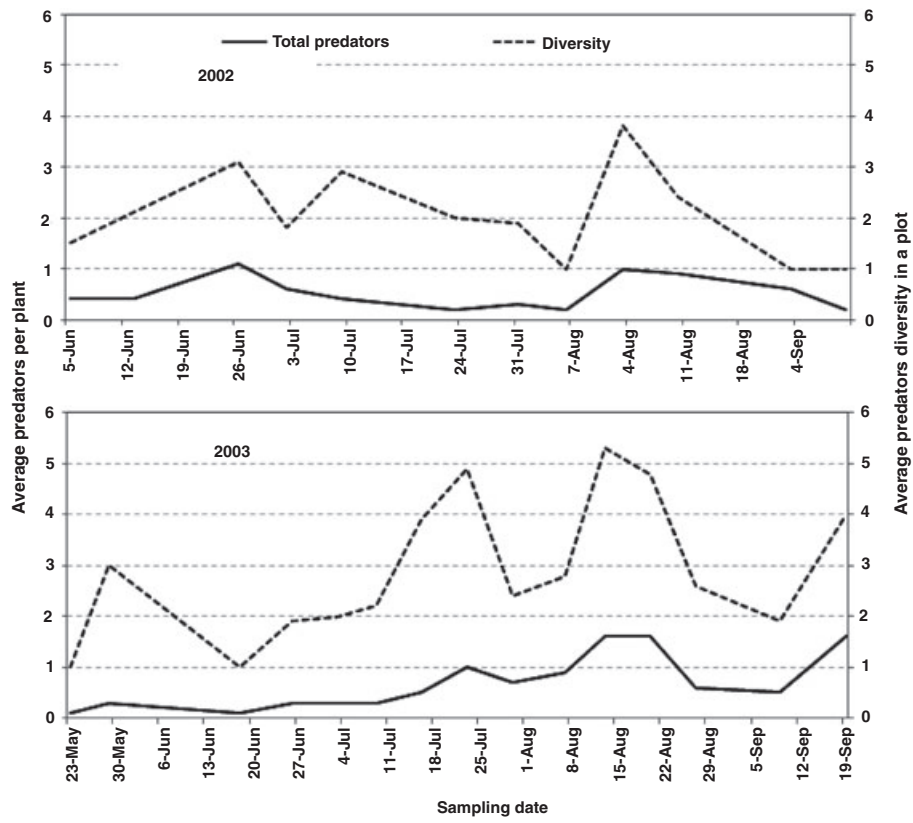


Fig. 10 Temporal dynamics of predator abundance and diversity (Shannon's indices) in cotton, Lamesa, Texas, 2002–2003.

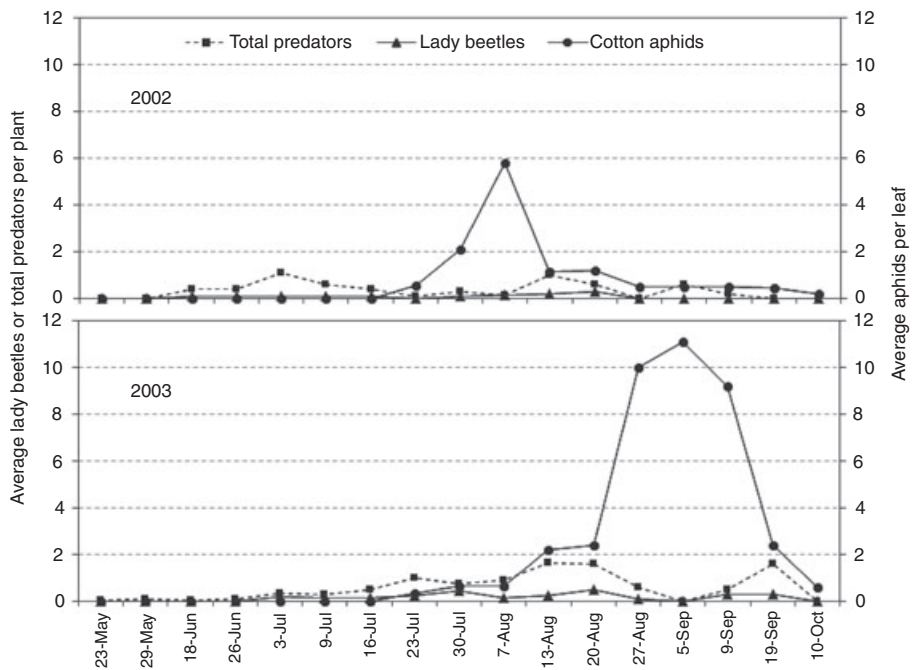


Fig. 11 Seasonal population dynamics of cotton aphids, convergent lady beetles, and total predatory arthropods in cotton, Lamesa, Texas, 2002–2003.

2002 (1.3/leaf). The total predator abundance increased with plant age when the number of cotton aphids also increased. However, the total predator population exhibited two population peaks, the first peak (1.07 predators/plant) in late June (2002) or late July (2003) and the second peak (1.15 and 1.63 predators/plant in 2002 and 2003, respectively) was observed on August 13, immediately after the aphid population peak (Fig. 11). A third predator peak was also observed in mid-September in 2003, which coincided with the decline of the extended 3-week long cotton aphid peak. Because there were clear lags between cotton aphid population dynamics and predator dynamics, no significant correlation was observed between total predator abundance and cotton aphid abundance based on 2-year combined data. Parajulee *et al.* (1994) also found a similar relationship between cotton aphid densities and predator densities in the neighboring Texas Rolling Plains. They reported a 3-week lag between cotton aphid and predator density peaks in an irrigated cotton study, wherein naturally occurring predator population, via predator conservation approach, was attributed to delayed cotton aphid population build-up in cotton. In our study, predaceous beetles were the most dominant foliage-dwelling predators followed by spiders, predatory bugs, and lacewings, which is a typical phenomenon in most Texas High Plains cotton agroecosystems.

## Disclosure

The authors declare that they have no conflicts of interest.

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## Simple and Effective Method for Evaluating Cotton Seedlings for Resistance to Thrips in a Greenhouse, and a Thrips Species Composition on the Texas High Plains

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**Abstract.** The method described in this paper uses wheat, *Triticum aestivum* L., grown in a greenhouse as the rearing medium for thrips forced to move to cotton, *Gossypium hirsutum* L., seedlings after the wheat was killed with herbicide. As measured by numbers of thrips and reduction of leaf surface area, the method produced abundant thrips pressure and was useful for initial evaluation of cotton for resistance. The method was used to evaluate cotton seedlings for six years and had a 0% failure rate. Western flower thrips, *Frankliniella occidentalis* Pergande, followed by onion thrips, *Thrips tabaci* Lindeman, were dominant during the study. Numbers of thrips of other species were scarce.

### Introduction

Pest thrips are a perennial problem, causing damage from cotyledon to fifth true-leaf stages of cotton, *Gossypium hirsutum* L., across most of the United States cotton belt. The amount of damage varies by year and location, but serious plant injury is common -- often with more than 50% reduction in true-leaf biomass, and in extreme cases stand reduction. Reduction in leaf biomass delays development of the seedlings, resulting in yield loss as great as 21% when thrips are not controlled (Leser and Vandiver 2003). In 2008, 2009, and 2010, thrips infesting United States cotton early in the season were ranked as the fourth (50,455 bales lost), first (138,207 bales lost) and sixth (45,964 bales lost) most damaging insect pests, respectively (Williams 2009, 2010, 2011).

With the uncertainty of re-registration of aldicarb insecticide, interest in the development of cotton cultivars with natural resistance to thrips has increased. For the purpose of this paper, plant resistance is defined as antixenosis, antibiosis, or tolerance. The genetic base of modern cotton cultivars has become narrow, with many sharing the same parent and grandparent lines. An ongoing evaluation program by Texas AgriLife Research at Lubbock is attempting to broaden the genetic base of cotton. The various obsolete race stock and wild cotton collections around the world contain a wealth of germplasm that could provide germplasm for development of thrips-resistant cotton cultivars. This type of effort begins with evaluation of the germplasm, which requires large numbers of thrips. The method reported herein was developed for this purpose.

Researchers have used a variety of methods that have been improved over time to rear thrips in the laboratory. Andrewartha (1934) collected *Thrips inaginis* Bagnall from roses, then reared the insects in beakers on rose buds with the petals removed. Citrus thrips, *Scirtothrips citri* (Moulton), were first reared on lemon leaves attached with paraffin to a glass plate and watered daily (Munger 1942). Methods were improved by development of an acrylic cage (Tashiro 1967) and use of an aspirator to collect thrips, which resulted in reduction of mortality and improved efficiency during insect transfers (Beavers and Ewart 1971). Last in this progression and most relevant to this paper, citrus thrips were reared on live California sumac plants in pots in a greenhouse (Tanigoshi and Nishio-Wong 1981).

The western flower thrips, *Frankliniella occidentalis* Pergande, has been reared using methods similar to those for citrus thrips. Murai and Ishii (1982) reared "flower thrips" on pollen and honey solution, and reported that pollen in the diet improved fecundity. Mollema et al. (1990) reared western flower thrips on flowering cucumber plants, then used an aspirator to collect the thrips. Teulon (1992) developed a laboratory method to rear western flower thrips larvae of a known, even age on disks of sweet pepper leaves. An apparatus was constructed that used a parafilm membrane for oviposition, improving egg handling and increasing hatch. Doane et al. (1995) increased the output of thrips and decreased the amount of labor required for rearing by using bean leaves as the oviposition medium. Murai and Loomans (2001) described improved methods for rearing western flower thrips on tea and pine pollen and also reported that pollen-baited food traps improved collection of thrips. More recently, DeGraaf and Wood (2009) used simplified cages and a whole Persian cucumber as a host to rear western flower thrips. In their book chapter of 1997, Loomans and Murai presented an excellent, comprehensive overview for rearing thrips that included western flower thrips.

All but two of the papers reported development or refinement of laboratory methods that are to varying degrees labor and equipment intensive, and can be complicated by such problems as medium degradation, loss of colony vigor, and invasion by pathogens. The method described herein takes the strategy of Tanigoshi and Nishio-Wong (1981) and Mollema et al. (1990) by rearing thrips on a live host in a greenhouse. The host is inexpensive and easy to grow. The method causes the thrips to move directly to the test plants, which reduces labor and injury to thrips caused by handling.

In the west Texas plains, western flower thrips can develop large populations in winter wheat, *Triticum aestivum* L. (Slosser et al. 2005). It is purported that these thrips move to seedling cotton in large numbers as the wheat begins to senesce and dry in late spring. Although not scientifically documented, strong circumstantial evidence of this movement is reported in the literature (Slosser et al. 2005). Orange-colored clouds of western flower thrips moving from wheat into seedling cotton in early to mid-May have been observed by many working in the cropping part of the cotton industry.

Based on patterns observed in the Texas High Plains agroecosystem, it was decided to attempt to develop a method to mimic the natural system. Simply rearing thrips-infested wheat in flats and using herbicide to terminate the initial host, thus forcing the thrips to move to the adjacent test cotton seedlings might be much simpler and less labor intensive than maintaining a thrips colony in the laboratory. Because the method was effective the first time it was used, results from the plant resistance studies will be used to illustrate its effectiveness and resulting

usefulness. The method was also used successfully to examine degradation of aldicarb insecticide in soil (Wheeler et al. 2008), but was not described in detail.

### Materials and Methods

This study was conducted in 25.9 x 12.2 m greenhouses at the Texas AgriLife Research and Extension Center at Lubbock. Two rearing cages were constructed from pressure-treated lumber, PVC pipe, and organdy cloth. For each cage, two 2.44 x 1.37 m rectangular frames were constructed from 5.1 x 10.2 cm pine boards and hinged on one long side. The frame to be used as the lower was sunk into the greenhouse gravel and spiked to the ground with 45.7 cm nails made from #4 (12.7 mm diameter) concrete reinforcement bar. This design was used to allow the cage to open like a clam shell. A raised, arched cover frame was constructed from 19.1 mm PVC pipe and fastened together with screws rather than glue to allow for adjustment or future design change. Organdy material was stretched over the PVC frame and attached to the top wood frame using screen molding and nails. The cage was sealed using silicone caulk. Each cage used 3.34 square meters of greenhouse space and was designed to accommodate 40 flats. Cages were also equipped with a sprinkler system for automatic watering.

Wheat at a rate of 100 g of untreated seed per flat was planted in potting soil in 15 x 35 x 9 cm (4.7 liter) flats. The greenhouse had a natural infestation of thrips at the time of the first experiment, so it was not necessary to artificially infest the wheat. The wheat was allowed to grow for 20 days to allow thrips to complete at least one life cycle before use. With automatic watering, the entire process required approximately four man-hours of labor. Thrips were maintained by leaving four flats of wheat inside the cages to reinfest wheat planted for the next series of experiments.

Free-choice tests were used in a randomized complete block design with five blocks. In every test, the commercial cotton cultivar All-Tex<sup>®</sup> Atlas was included as a susceptible check and starting with Test 17, accession TX110 found to be resistant in early testing was included as a positive check. With the exceptions of Tests 46-49, 10 accessions were evaluated per test and each experimental unit consisted of six plants. Test cottons were planted at the rate of four seeds per 0.47-liter cup, then thinned after germination to one plant per cup. Flats of wheat were placed next to test plants, then sprayed with a 2% solution of glyphosate, forcing the thrips to move to the only available living plant tissue, that of the target cotton. Eight flats of wheat per block (12 experimental units) were used in Tests 6-25, and then reduced to six thereafter to moderate the amount of damage by thrips. Three methods were used to evaluate cotton plants at the fifth true-leaf stage. Plants were given a subjective rating of zero to nine (by experimental unit), with zero equaling complete damage and nine no discernible damage by thrips. The rating was based on silvering of cotyledons, crinkling of true leaves, and necrosis. A method described by Burris et al. (1990) was used to wash plants for thrips and thrips were collected, counted, and identified. Adult thrips were mounted in PVA on microscope slides and identified to species using the CSIRO interactive software key (Moritz et al. 2001). A LI-COR<sup>®</sup> Model 3100C surface area meter (LI-COR Environmental, Lincoln, NE) was used to measure the surface area of all true leaves, and percentage of leaf surface area reduction was calculated using plants of the same genotype kept thrips-free with insecticide by a method described by Quisenberry and Rummel (1979).

To be certain that wheat was the source of thrips infesting tested cotton plants, two additional studies were done. The first compared plants of the cultivar All-Tex Atlas grown in the original test greenhouse and 3.05 meters away in an adjacent greenhouse of the same complex. The two environments were identical, but any thrips moving out of the wheat in the original test greenhouse were obstructed from entering the second greenhouse by the 10 mm Lexan<sup>®</sup> walls and glass roofs of the greenhouses. Chemical treatments included no insecticide (both greenhouses) and biweekly acephate sprays (original test greenhouse only). Nine units (six plants each) per treatment were grown. Because it was not possible to randomize the “greenhouse” effect, data were compared using simple two-sample *T*-tests. In the second study, a sample of 25 flats of wheat was selected randomly and each flat was turned upside down and tapped 10 times with moderate force on a 38 x 48 cm white plastic sheet. Adult thrips dislodged onto the sheet were counted.

### Results and Discussion

The plant resistance evaluations using the method described in this paper are summarized in Table 1. Tests 1-5 were preliminary and are not reported. Eight (13.3%) of the tests failed, but no failures were caused by lack of thrips; all were caused by human error or mechanical failure. The number of tests that could be done was limited by availability of seed and in one case by a shut-down while the greenhouse was repaired. The lack of testing seed was because only 150 accessions can be increased per year because of the short-day flowering habit of most of the accessions from the collection. With these limitations removed, it should be possible to evaluate approximately 30 accessions per month using the method and a five-block replicated design. If seed stocks of more cottons were available, for example the entire 10,000+ accessions in the USDA collection, and it was deemed that more rapid, but less precise evaluation was adequate to fulfill objectives, the testing rate could be increased by as much as five fold by reducing the number of blocks to less than five or eliminating the inferential statistical approach.

Numbers and species of thrips collected from tested cotton plants subjected to thrips pressure using the wheat method are reported in Table 2. Although presence of thrips in Tests 32-65 has been confirmed, the specimens await processing, so species identification data only from Tests 6-31 are presented. The number of plants sampled ranged from 244 to 359. The variation was caused by poor germination and stand establishment of some of the accessions tested. Thrips were found on test cotton plants in every experiment. With the exception of Tests 20-22 (conducted concurrently in October 2006), all tests using eight flats of wheat per block had counts that ranged from 145 to 1,035 adults and 245 to 9,506 larvae. Of this group, only two tests had fewer than 500 larvae. Tests 26-31 used six trays of wheat per block and had fewer thrips. These results indicated successful rearing of thrips on wheat, then transfer to and colonization of the experimental cotton seedlings. Although pollen was not included as part of the thrips diet, thrips fecundity was sufficient to cause enough damage for evaluating plant resistance. During the 21-month period (September 2005-May 2007) reported in Table 2, western flower thrips was by far the dominant species, comprising almost 100% of the thrips in 16 of 24 experiments. Onion thrips was next most abundant, comprising

Table 1. Summary of Thrips Host Plant Free-choice Experiments Showing Success (S) or Failure (F) of Test to Achieve Objectives

Test	Month/year	Success/failure	Test	Month/year	Success/failure
6	Sep. 2005	S	36	Apr. 2008	S
7	Oct. 2005	F	37	Apr. 2008	S
8	Nov. 2005	S	38	May 2008	S
9	Nov. 2005	S	39	May 2008	S
10	Mar. 2006	S	40	Dec. 2008	F
11	Mar. 2006	S	41	Dec. 2008	F
12	May 2006	S	42	Dec. 2008	F
13	May 2006	S	43	Feb. 2009	S
14	Jun. 2006	S	44	Feb. 2009	S
15	Jun. 2006	S	45	Feb. 2009	S
16	Jun. 2006	S	46	May 2009	S
17	Jul. 2006	S	47	May 2009	S
18	Jul. 2006	S	48	May 2009	S
19	Jul. 2006	S	49	Jun. 2009	S
20	Oct. 2006	S	50	Oct. 2009	S
21	Oct. 2006	S	51	Oct. 2009	S
22	Oct. 2006	S	52	Oct. 2009	S
23	Jan. 2007	S	53	Jan. 2009	S
24	Jan. 2007	S	54	Jan. 2009	S
25	Jan. 2007	S	55	Jan. 2009	S
26	Mar. 2007	S	56	Apr. 2010	S
27	Mar. 2007	S	57	Apr. 2010	S
28	Mar. 2007	S	58	Oct. 2010	F
29	May 2007	S	59	Oct. 2010	F
30	May 2007	S	60	Jan. 2011	S
31	May 2007	F	61	Jan. 2011	S
32	Aug. 2007	F	62	Mar. 2011	S
33	Aug. 2007	S	63	Mar. 2011	S
34	Nov. 2007	S	64	Sep. 2011	S
35	Nov. 2007	S	65	Sep. 2011	S

2.3 to 11.1% of the thrips in five experiments. Other species were found in very small numbers as reported in a Table 2 footnote.

Percentage of leaf surface area reductions calculated using untreated plants and plants of the same genotype treated with acephate to control thrips are presented in Table 3. Tests that failed are not included in the table. Hundreds of cottons were evaluated during the course of the study, but results from the cultivar All-Tex Atlas that was used as the susceptible check are presented to illustrate the effectiveness of the method. Mean leaf surface area reductions ranged from 19.23 to 99.30%. Nine of the successful tests had reductions <50% while 12 had reductions >90%. In Test 9, the cotton seedlings were nearly killed, and had no living leaf tissue to measure. Most damage rating means ranged from 0.6 to 3.8, low in the rating scale indicating moderate to heavy thrips damage symptoms. Tests 64 and 65, with mean ratings of 6.2 and 7.0, respectively, were not typical and probably were because younger (14 day old) wheat was used in an attempt to reduce the amount of time needed to complete a test. In all experiments, the

Table 2. Total Number of Thrips on Untreated Cotton Plants Approximately 21 Days after Exposure to Herbicide-treated, Dying Wheat

Test	Flats of wheat	No. plants sampled	Total thrips			Adult species composition <sup>ab</sup>		
			Adults	Larvae	Mounted <sup>c</sup>	<i>F. occidentalis</i>		<i>T. tabaci</i>
						Female	Male	Female
6	8	324	503	9,506	603	392	113	60
9	8	244	198	543	190	161	28	0
10	8	316	277	384	267	211	56	0
11	8	284	283	245	267	217	33	0
12	8	323	649	1,595	615	412	192	3
13	8	344	1,035	3,629	814	451	343	4
14	8	257	192	724	141	97	39	0
15	8	336	233	1,057	195	127	65	0
16	8	314	380	1,261	277	182	92	0
17	8	301	155	2,591	120	86	25	0
18	8	343	290	3,785	250	197	43	0
19	8	339	395	3,131	330	250	69	0
20	8	333	86	1,960	82	65	16	1
21	8	335	56	840	48	26	21	0
22	8	349	59	597	45	16	27	1
23	8	289	145	1,621	131	94	31	0
24	8	331	284	1,878	258	223	34	0
25	8	318	165	1,803	150	124	24	0
26	6	322	40	361	29	21	8	0
27	6	310	54	256	45	32	13	0
28	6	271	56	207	41	34	7	0
29	6	335	20	94	18	9	7	2
30	6	336	13	44	12	5	5	1
31	6	359	46	81	45	29	12	4

<sup>a</sup>Genera are: F: *Frankliniella*, T: *Thrips*.

<sup>b</sup>Other thrips were found in small numbers, specifics: *Anaphothrips obscurus* (Muller) (2), *Chirothrips manicatus* Haliday (1), *Frankliniella williamsi* (Hood) (3), and *Neohydatothrips variabilis* (Beach) (1).

<sup>c</sup>Some thrips that were mounted were damaged and could not be identified to species.

number of thrips on seedlings was enough to produce rating and leaf surface area reduction data that allowed successful evaluation of the cottons for resistance.

Results of the experiment that tested 'greenhouse' as an effect are presented in Table 4. Seedlings in the greenhouse with no wheat suffered no discernable leaf injury and had the most leaf surface area per plant (48.58 cm<sup>2</sup>). The plants subjected to thrips by using the wheat method had significantly less leaf surface area per plant at 21.8 cm<sup>2</sup>. The lack of injury in the greenhouse with no thrips-infested wheat verifies observations made during the entire evaluation. Although cotton seedlings in other greenhouses often suffered a small amount of damage by thrips, it was negligible when compared to that in the thrips evaluation experiments. The acephate-treated plants in the wheat greenhouse had an intermediate mean leaf surface area, which was not expected. Acephate may have only partially controlled the thrips. This will be the subject of future study.

Table 3. Mean Percentage of Leaf Surface Area (LSA) Reduction and Means of a Subjective Damage Rating of Cotton Plants Subjected to Heavy Thrips Pressure<sup>ab</sup>

Test	n	% LSA reduction (±SEM)	Damage <sup>c</sup> rating	Test	n	% LSA reduction (±SEM)	Damage <sup>c</sup> rating
6	5	30.95 ± 4.96	--	35	5	50.14 ± 28.87	2.3
9	5	100.0 ± 0.00	--	36	5	93.55 ± 4.49	1.4
10	5	88.76 ± 6.43	--	37	5	96.55 ± 3.45	0.6
11	5	82.13 ± 10.52	--	38	5	63.04 ± 17.21	2.4
12	5	68.55 ± 3.29	--	39	5	58.58 ± 14.43	2.6
13	5	55.27 ± 9.36	--	43	5	76.15 ± 4.05	2.4
14	4	63.44 ± 4.50	--	44	4	76.58 ± 5.79	2.0
15	5	43.01 ± 14.01	--	45	5	70.36 ± 9.35	2.2
16	5	30.15 ± 10.08	--	46	5	94.11 ± 2.07	3.2
17	5	75.50 ± 7.86	--	47	5	93.09 ± 1.51	3.6
18	5	49.81 ± 12.86	--	48	5	92.70 ± 1.10	3.8
19	5	39.82 ± 15.25	--	49	15	49.75 ± 8.04	5.1
20	5	27.16 ± 12.82	--	50	5	81.43 ± 7.38	3.8
21	5	53.75 ± 12.17	--	51	5	89.55 ± 4.18	3.4
22	5	52.87 ± 15.56	--	52	5	58.34 ± 14.56	3.2
23	5	90.08 ± 6.42	3.4	53	5	90.54 ± 2.58	2.8
24	5	97.25 ± 0.44	2.0	54	5	70.16 ± 7.03	2.8
25	5	98.87 ± 0.15	2.0	55	5	52.04 ± 11.10	3.0
26	5	97.86 ± 0.79	2.0	56	5	69.55 ± 5.58	3.6
27	5	97.84 ± 0.78	1.6	57	5	62.32 ± 7.34	3.0
28	5	99.13 ± 0.25	1.4	61	5	81.82 ± 7.32	1.4
29	5	89.22 ± 3.47	1.8	62	5	56.47 ± 6.44	3.8
30	5	78.65 ± 6.37	4.0	63	5	45.37 ± 6.85	3.6
31	5	85.03 ± 1.50	3.2	64	5	19.23 ± 8.64	6.2
34	5	80.29 ± 10.25	2.4	65	5	53.82 ± 7.75	7.0

<sup>a</sup>Surface area reduction was calculated using untreated plants and plants of the same genotype kept thrips free with acephate insecticide.

<sup>b</sup>Tested cultivar was All-Tex Atlas.

<sup>c</sup>Damage rating scale was 0 to 9, with 0 complete damage (without mortality) and 9, no visible damage.

Table 4. Mean True-leaf Surface Area per Plant of Cotton Exposed to a Wheat-based Thrips Rearing and Movement Method as Compared to Acephate-treated Plants and Plants Grown 3.05 Meters Away in an Adjacent Greenhouse Sharing a Common Wall in the Same Complex

Greenhouse <sup>a</sup>	Wheat-based thrips pressure	Acephate treated	Leaf surface <sup>b</sup> area (cm <sup>2</sup> )
1	N	N	48.58a
2	Y	Y	32.80b
2	Y	N	21.84c

<sup>a</sup>Different greenhouses were maintained under identical temperature, humidity, and shading regimens.

<sup>b</sup>Means followed by the same letter are not significantly different ( $P > 0.05$ ,  $T$ -test).



Tapping upended flats of wheat on a white plastic counting sheet dislodged thrips in every case. Numbers ranged from 17 to 54, with a mean of 29.7 per flat. The beating method had moderate efficiency as evidenced by the fact that further beating of a tested flat dislodged more thrips. Although not a precise quantification of the thrips living in the wheat, the counts do demonstrate the presence of thrips in numbers sufficient to meet the needs of the evaluation experiments. One serious problem occurred in about 10% of the colony wheat sets. The wheat became infested with aphids, most commonly corn leaf aphids, *Rhopalosiphum maidis* (Fitch). This situation was rectified by what was initially believed to be a simple inundative release of *Aphidius colemani* Viereck, a chalcid parasitoid commercially available for biological control in the greenhouse. This one release also proved to be inoculative, possibly maintained by small numbers of aphids in other greenhouses of the complex, and has effectively controlled aphids in the colony for four years. In the greenhouse system, aphids usually appear, begin to increase in abundance, then soon decrease because of parasitism.

### Conclusion

The method presented here allows for development of large numbers of thrips and their movement to plants to be evaluated. Compared to laboratory methods, it requires a minimum amount of labor and skills and can be perpetuated for years. While not as precise as other methods, the large number of thrips on test plants makes the method useful to quickly and effectively eliminate susceptible genotypes in the free-choice phase of large numbers of evaluations for plant resistance. The method was used to identify thrips-resistant genotypes and resulted in a cultivar development effort that has carried resistance to thrips to the F<sub>5</sub> generation. The method also might be useful for evaluating other crops for resistance to thrips. Further, by changing the rearing host plant species, this concept could be used to evaluate resistance to other multi-host arthropod pest species.

Identification of thrips collected during the experiments revealed that western flower thrips was dominant in the greenhouse setting, followed by onion thrips a pattern similar to what is generally encountered in Texas High Plains cotton. All other thrips species were found in very low numbers.

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