FARMER FRIENDLY SUMMARY

Cotton Insect Pest Management, Agreement 13-589 TN
Scott Stewart, The University of Tennessee

Funding is used to partially support general extension activities such as scout training, moth trapping, resistance monitoring, insecticide testing and on-farm evaluations of various insect control technologies and treatment thresholds. Funding is also used to help support regional projects, several of which are also partially supported by core grants from Cotton Incorporated. Pheromone moth trapping for bollworm and tobacco budworm are improving the decision making of crop managers. In 2016, moth catches for each trap location were reported weekly on the UTcrops News Blog (http://news.utcrops.com/). This information and other IPM news updates were distributed to agents, producers, consultants and other agricultural professionals. Statewide, average moth catches for bollworm (i.e., corn earworm) were similar to average trap catches for the previous decade. As typical, statewide catches of tobacco budworm in 2016 were lower than bollworm and somewhat lower than catches typically observed. Moth traps did well at predicting corn earworm (bollworm) pressure observed in flowering cotton. Assays using bollworm moths indicated substantially higher resistance to pyrethroid insecticides than in previous years, a phenomenon that also occurred in other states.

Many experiments were done in Tennessee to assess control options for thrips, tarnished plant bug, bollworms, and spider mites. The results of most experiments have been individually summarized and published at UTcrops.com (http://utcrops.com/MultiState/MultiState.htm). Examples presented in this report include 1) the influence of nozzle type on insecticidal control of tarnished plant bugs, 2) how retreatment interval affected insecticide control of tarnished plant bug, 3) the relative sensitivity of selected cotton varieties to thrips, and 4) the efficacy of a virus in controlling caterpillar pests in cotton.

Regional research efforts included are 1) an evaluation of insecticide and insecticide/nematicide seed treatments on thrips control, 2) the validation of a new treatment threshold for controlling caterpillar pests, 3) experiments to assess when insecticide applications for tarnished plant bug can be terminated, and 4) monitoring for bollworm resistance to Bt toxins. Comprehensive reports for these efforts have been submitted by the appropriate project leaders, but abbreviated reports for these projects are included here. As neonicotinoid seed treatments continue to diminish in their activity, finding alternative thrips treatments continues to be a challenge. No new insecticides appear to provide adequate plant protection. Bt cottons are providing substantial protection form caterpillar pests, but our data show than foliar insecticide applications are sometimes warranted when bollworm pest pressure in high. A new threshold based on fruit injury worked well in cotton our experiments, regardless of the Bt technology used. Regional monitoring efforts indicate bollworm is developing resistance to one of more toxins present in Bt cotton. Other data indicate that the first four weeks of bloom is the critical window to protect cotton for threshold infestations of tarnished plant bug, and applications after this time have not significantly increased yield.

The data generated from these above experiments are used to validate and modify extension insect control recommendations in Tennessee. Over 1,500 copies of the UT insect control recommendations for cotton were distributed to stakeholders.

ANNUAL REPORT

Cotton Insect Pest Management, Agreement 13-589 TN
Scott Stewart, The University of Tennessee

Funding for this project was used to partially support general extension activities such as scout training, moth trapping, evaluation of existing and new Bt cotton technologies, resistance monitoring, and insecticide testing and validation of treatment thresholds. In addition, funds were also used to help support several regional projects as described herein.

1) Moth Trapping. Despite the use of Bt-transgenic cotton on over 98% of the acreage in Tennessee, bollworm and tobacco budworm still compose an important pest complex. Bollworms may cause significant economic damage to Bt cotton fields, and the bollworm/budworm can be even more damaging to non-Bt cotton. More importantly, the threat of tobacco budworm infestations results in high adoption of Bt cotton.

Area-wide monitoring remains a valuable tool in predicting the occurrence and size of pest populations. Pheromone trapping programs for bollworm or tobacco budworm provide insight into the timing and intensity of moth flights. For example, unusually high trap catches for a particular species can alert consultants and producers to the potential for impending outbreaks. When performed on a regional level and over a number of years, moth trapping can indicate historical and geographical patterns in the distribution of pest populations. They might even indicate resistance to Bt cotton, should, for example, tobacco budworm moth catches begin increasing over time. Moth monitoring improves the decision making process, helping crop managers in the selection of insecticides and to indicate the need for intensified sampling efforts. This ultimately helps to minimize control costs and/or yield losses incurred by producers. Traps are used to collect moths used in assays for resistance to pyrethroid insecticides.

Pheromone moth traps for corn earworm (bollworm) and tobacco budworm were run on a weekly basis from early May through mid-August. Pheromone lures were obtained from Great Lakes IPM (Vestaburg, MI) and were changed at two week intervals. At least one, and usually two, sets of bollworm and tobacco budworm traps were run in each of the following 12 counties in West Tennessee: Carroll, Crockett, Dyer, Fayette, Gibson, Hardeman, Haywood, Shelby, Tipton, Lake, Lauderdale, and Madison. At each location, Hartstack traps baited with Hercon® pheromone lure was used to trap for corn earworm (bollworm) and tobacco budworm. The trapping period in 2016 began in early-May and extended through mid-August. Traps were checked weekly, and the number of moths in each trap was recorded. Pheromone lures were replaced every second week.

Outcomes: Moth catches for each trap were reported weekly on the UTcrops News Blog (http://news.utcrops.com/) which was distributed to agents, cotton producers, consultants and other agricultural professionals.

Few if any cotton fields were treated for tobacco budworm. This is not surprising considering the very low acreage of non-Bt cotton, and moth catches were relatively low in 2016. The highest single-trap capture was recorded in Dyer County where 25 moths were caught the week preceding August 11.

As usual, catches of corn earworm moths in pheromone traps were substantially greater than tobacco budworm. As is also commonly observed over the years, the highest moth catches were observed in Lake County (Fig. 2). The highest single trap catch also occurred in Lake County where 255 moths were caught the week preceding August 4. Corn earworm moth catches were low for most of the season in 2016 compared with previous years, but average moth catches in early August peaked substantially higher than average catches in previous years (Fig. 3).

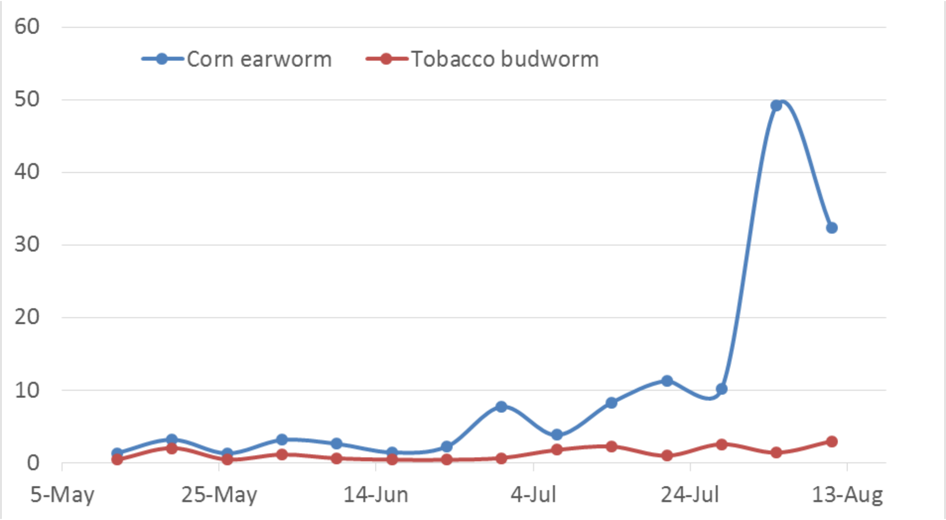


Figure 1. Average number of tobacco budworm and corn earworm (bollworm) moths caught per trap in pheromone traps across West Tennessee, 2015.

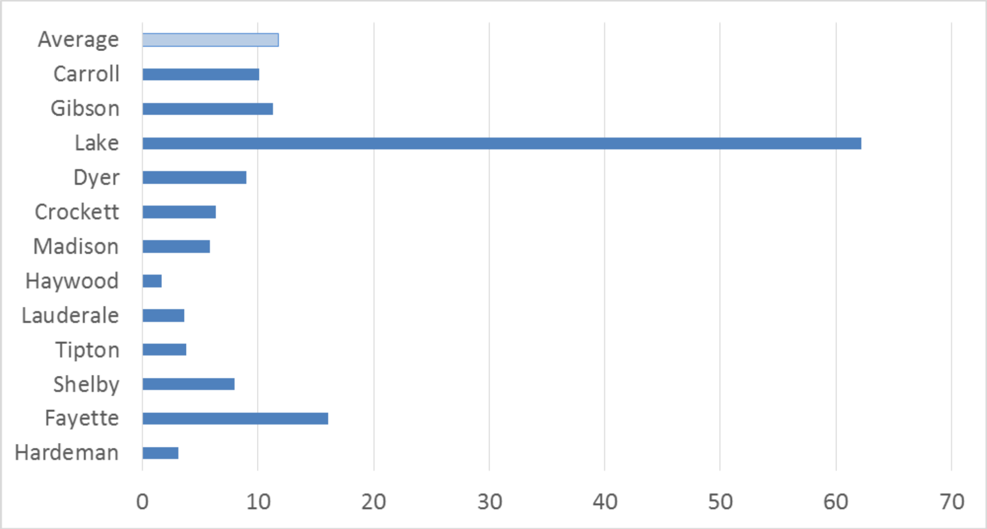


Figure 2. Seasonal average number of corn earworm (bollworm) moths caught per trap per week in each county, 2015.

Moth trapping did not necessarily reflect all local variations in pest densities observed in cotton fields, in part because trap density was not high and because other factors influence oviposition and survival of these pests in cotton. The bollworm is Tennessee's most significant caterpillar pest in cotton because this species is able to cause economic injury to Bt cotton. As predicted by trap catches, early and mid-season infestations in cotton were generally low. Pheromone trapping predicted the generally low infestations of bollworms observed in cotton until early-August, at which time there were several reports of late-season bollworm injury in Bt cotton.

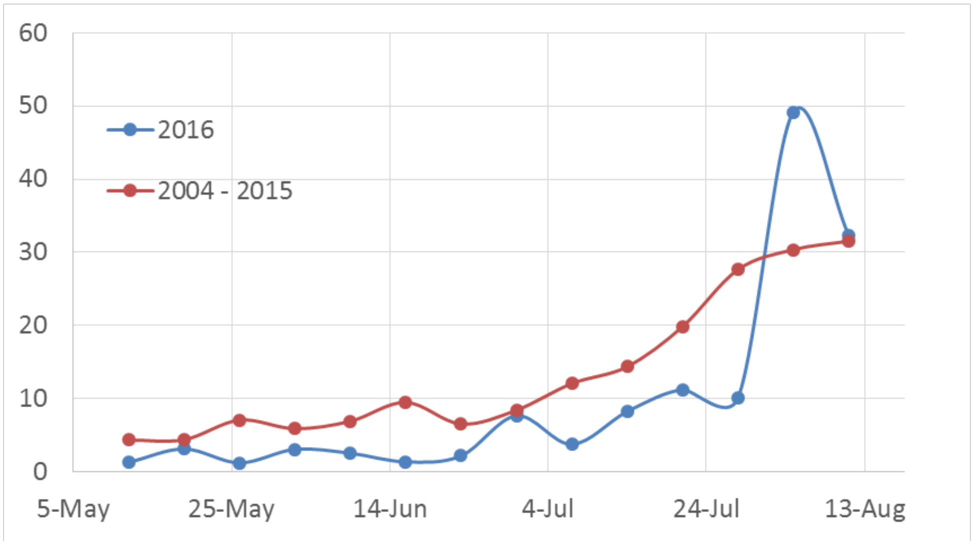


Figure 3. Average statewide catches of corn earworm (bollworm) moths, comparing 2016 to the average catches for 2004 - 2015.

2) Insecticide Efficacy, Bt Cotton, and Threshold Verification Trials. Multiple replicated experiments are done annually to evaluate insecticide efficacy, new or alternative treatments, and validate IPM recommendations. Small-plot experiments are done as randomized and replicated trials on UT experiment stations or in grower fields. Individual plot size varies from 4-8 rows and 35-50 feet in length. The data generated from these activities are used to validate and modify extension insect control recommendations for Tennessee. These evaluations included insecticide efficacy trials for thrips, plant bugs, stink bugs, and bollworm. A few examples are presented below. The results of most experiments are published on UTcrops.com at the following link: http://www.utcrops.com/MultiState/MultiState.htm.

2.1) Impact of nozzle selection on insect control. The objective of this research was to evaluate the level of insect control offered by various nozzle technologies, several of which required for use with certain herbicides on new herbicide-tolerant traits in cotton (e.g., dicamba). These nozzles mitigate off-target drift and therefore have an increased droplet size compared with the flat fan tips traditionally used for insect control. Additionally, many of these nozzles are designed specifically for use with pulse width modulation (PWM) systems found on newer commercial sprayers. Selected nozzles were evaluated for the control of thrips in seedling cotton or tarnished and clouded plant bugs in flowering cotton.

Tests were done at the West Tennessee Research and Education Center in Jackson, TN. Phytogen® 333 WRF (WideStrike) cotton was planted no-till for a thrips trial May 9, 2016. Individual plots were 2 rows wide with a buffer row between plots (38 inch centers) x 35 feet. Phytogen® 333 WRF (WideStrike) cotton was planted no-till May 22, 2015 and May 19, 2016 for the plant bug trials. Plots were 8 rows (38 inch centers) x 35 feet. Treatments were replicated in a randomized complete block design.

A foliar application of 0.25 lb ai/acre acephate 90SP was made to seedling cotton to evaluate thrips control June 1, 2016. Acephate 97SP at 0.75 lb ai/acre was sprayed for plant bugs July 31, 2015 and July 22, 2016. All nozzles were operated under manufacturers suggested parameters for optimal performance. An application rate of 15 gallons per acre (GPA) was targeted for the thrips trial whereas the targeted rate for the plant bug trial was 15 GPA in 2015 and 10 GPA in 2016. Thrips samples were taken by cutting five seedling (2 lf stage) plants per plot at the soil level and immediately placing into jars containing a 70% ethanol solution. Samples were collected two days after treatment (2 DAT) in the thrips trial. Samples were processed in an ethanol wash and counted in the lab using a dissecting microscope. Plots in the plant bug trials were sampled with a standard 2.5 x 2.5 ft black cloth shake sheet. Two shake sheet samples were taken on the center two rows (10 row feet) of all plots. Ratings for the plant bug trials were done 5 DAT and 4 DAT in 2015 and 2016, respectively.

Nozzles used in the thrips and the plant bug trial in 2016 were flat fan, AIXR, TTI, Wilger Y and Wilger UR (Table 1). The nozzle types evaluated in the 2015 plant bug trial were flat fan, TTI, TADF, and Wilger MR.

Table 1. Nozzles types evaluated for insect control in 2015 and 2016.

Nozzle	Type	Droplet size (microns)
Spraying Systems TeeJet XR11002FF	Flat Fan	200, Fine
Wilger MR110-02	Mid-Range	250, Fine
Greenleaf Technology TADF02	TurboDrop Assymetrical Dual Fan	350, Coarse
Spraying Systems AIXR11002	Air Induction Extended Range	350, Coarse
Spraying Systems TTI11004	Turbo TeeJet Induction (TTI)	800, Ultra coarse
Wilger UR04	Prototype	800, Ultra coarse
Wilger Y on Plant Bug Trial: MR110-015 facing forward at 1/3 spray volume DR110-025 facing rear at 2/3 spray volume	Multi-tip for use with PWM system	MR 250, Fine; DR 300, Coarse

Wilger Y on Thrips Trial MR110-02 facing forward at 1/3 spray volume DR110-06 facing rear at 2/3 spray volume	Multi-tip for use with PWM system	MR 250, Fine; DR 500, Very coarse

<u>Outcomes</u>: Thrips or tarnished plant bug populations greatly exceeded the recommended treatment thresholds in these tests. Insecticide applications with several new nozzle designs, including those designed for use with a pulse width modulating spray system, provided control of thrips and plant bugs similar to that of traditional applications with a flat fan nozzle (Figs. 4 - 6). These data suggest that insect control will not be substantially affected by the use of these specific nozzles intended for the application of herbicides. However, additional testing is needed to assess the impact of nozzle/application technologies on the control of these and other insects with different insecticides.

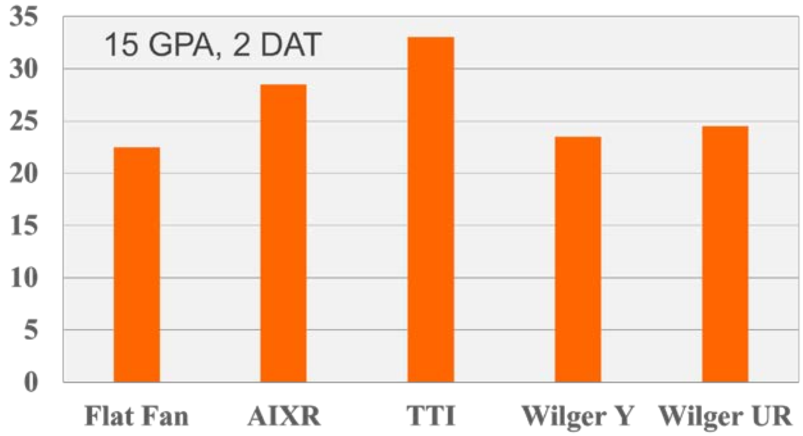


Figure 4. Average total numbers of thrips per five plants. Nozzle effects were not significant (P = 0.8056). Note that and non-treated control was not included in this study.

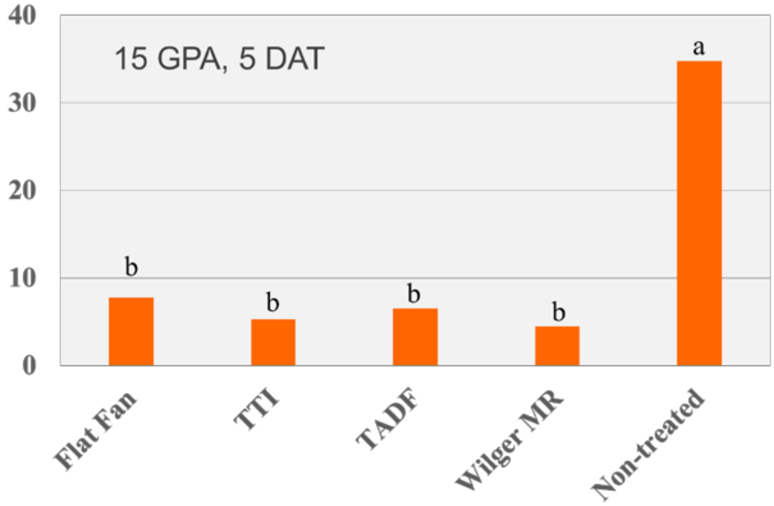


Figure 5. Average total plant bugs per 10 row feet in flowering cotton, 2015 (P = 0.0004).

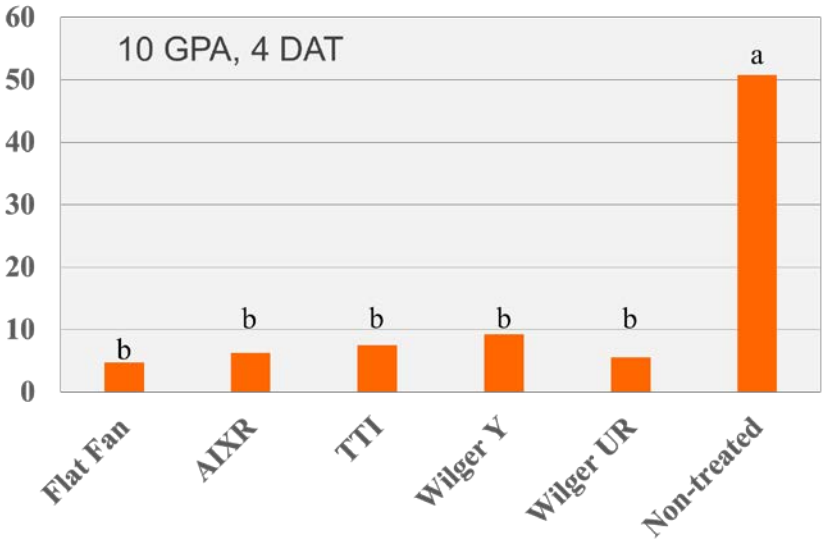


Figure 6. Average total plant bugs per 10 row feet in flowering cotton, 2016 (P = 0.0001).

2.2) Impact of retreatment interval on tarnished plant bug control and yield. A small-plot replicated experiment was done to evaluate how application intervals affected control of tarnished plant bugs and yield on Bt cotton. Orthene 97SP was applied at 0.75 lb/acre to cotton during the second week of bloom. A subsequent application of the same insecticide and rate was applied 4, 5, 7 or 10 days later. A comparison treatment was included that consisted of a single application of Orthene at the same rate tank mixed with the maximum rate of Transform (2.25 oz/acre). No other insecticide applications were made. The objective of this test was to demonstrate that shortened retreatment intervals provided similar or superior crop protection in situations of intense plant bug pressure.

Outcomes: All insecticide treatments substantially increased yield (Fig. 7). Yield was numerically but not statistically higher when retreatment was made 4 or 5 days after the original application. After the final retreatment and regardless of the retreatment interval, plant bug numbers were similar and much lower in insecticide-treated plots versus untreated plots. However, tarnished plant bug numbers were significantly higher where only a single, ‘going on vacation’ treatment was made. These data indicate that the original application was most important in protecting yield. Shortening retreatment intervals did not result in the need for third insecticide application, and in fact, tended to increase yield compared with longer-retreatment intervals or making a single ‘high-rate’ application. This is probably because cotton is relatively sensitive to tarnished plant bug injury is during the first few weeks of bloom become relatively tolerant after this time (see section 4.4 below).

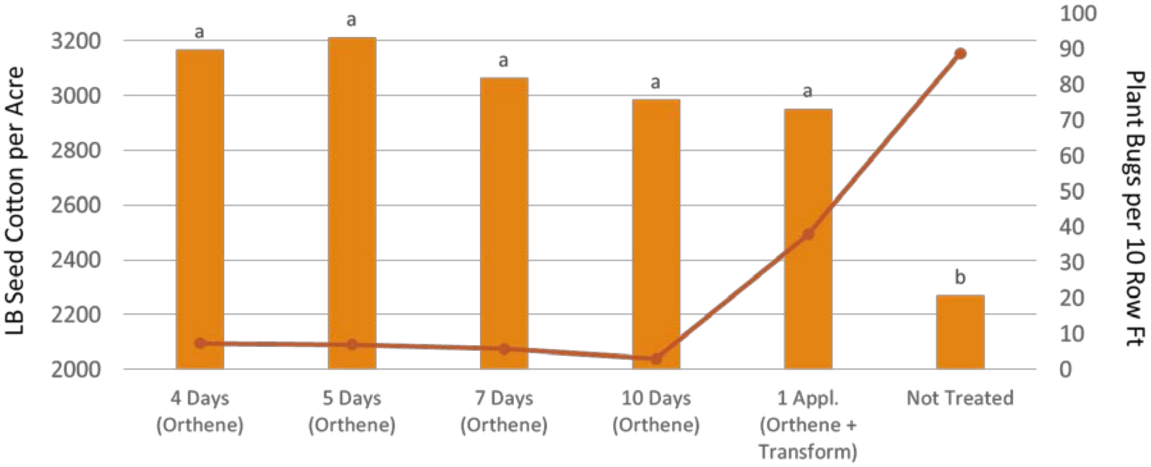


Figure 7. Seed cotton (lb/acre) and tarnished plant bug numbers (per 10 row ft) for two insecticide treatments made at different application intervals (4, 5, 7, and 10 days apart) vs. a single applications. Plant bug numbers are shown for ratings made 14 days after the initial application.

2.3) Variety sensitivity to thrips. This experiment was done at several locations throughout the Mid-South to evaluate varietal susceptibility to thrips injury. Only a fungicide seed treatment was used. The test, including 7 Bt cotton varieties, was a small-plot replicated trial planted on May 9, 2016 at the West Tennessee Research and Education Center. Thrips counts, thrips injury ratings, and vigor ratings were made on several dates.

Outcomes: Data for Tennessee are shown in Table 2. Thrips injury ratings and thrips numbers were not significantly affected by variety. There were differences among varieties in vigor ratings, and vigor was highest for ST 4946 (= 3.8) and lowest for DP 1646 (= 3.2). There were also differences in yield among varieties, but these differences were likely the result of inherent variety characteristics rather than differences in susceptibility to thrips.

Table 2. Summarized results of an experiment to evaluate the relative sensitivity of different cotton varieties to thrips injury (selected data shown).

Description	Thrips Injury 0 - 5 scale		Vigor 0 - 5 scale		Thrips/5 plts. Adult + Imm.		Seed cotton lbs/acre	
Days after planting	25		25		28			
Variety								
1 ST4747	3.0	a	3.4	bc	81.3	a	3647.2	a
2 ST4946	3.0	a	3.8	a	54.0	a	3825.1	a
3 DP1410	3.0	a	3.4	bc	74.0	a	3422.1	a
4 DP1538	3.3	a	3.3	c	61.5	a	2605.6	b
5 DP1518	3.3	a	3.6	ab	59.5	a	3482.5	a
6 DP1646	3.6	a	3.2	c	55.0	a	2891.2	b
7 PHY339	3.0	a	3.6	ab	64.5	a	3819.7	a

Means followed by same letter or symbol do not significantly differ (P=0.05, LSD).

2.4) Evaluation of a virus for control of caterpillar pests. An experiment was done on non-Bt cotton to evaluate the effectiveness of a nuclear polyhedrosis virus (Helicovex, Andermatt Biocontrol) insecticide sprayed twice for the control of bollworm and tobacco budworm larvae. Comparison treatments included a single applications the virus tank mixed with Karate Z (lambda-cyhalothrin, Syngenta), Karate Z applied alone, or Besiege (a premix of lambda-cyhalothrin and chlorantraniliprole, Syngenta). DP 1441 RF was planted on May 31, 2016. This small-plot test was arranged in a randomized complete block with four replicates, and insecticide application were made in early August. Bollworm and fall armyworm were the predominant caterpillar pests observed, and larvae counts and square and boll damage ratings were made several times after application.

Outcomes: The Helicovex virus is specific to bollworm and tobacco budworm, and thus has no activity on fall armyworm. Data collected from this experiment indicated this virus had so significant effect on boll injury or bollworm or fall armyworm larval numbers (Table 3). Indeed, Besiege was the only treatment which statistically reduced boll injury compared with non-treated plots. Yields were highest in plots treated with Besiege but not significantly higher than those treated with Karate. The yield of non-treated plots and those treated twice with Helicovex were not different.

Table 3. Summarized results of an experiment to evaluate the efficacy of a virus (Helicovex) for the control of caterpillar pests in cotton (selected data shown).

Description			Bollworm larvae	FAW larvae	Damaged bolls	FAW larvae	Seed Cotton lbs/acre
Sample size			20 Plant	20 Plant	50 Boll	50 Boll	
Days after treatment			4 DA-B	4 DA-B	14 DA-B	14 DA-B	
Treatment	Rate (oz/a)	Appl					
1 Helicovex	2.5	AB	5.8 a	3.0 a	13.5 a	3.5 a	2639 bc
2 Helicovex	2.5	A	6.8 a	2.0 a	6.3 b	1.5 b	2857 ab
Karate Z	1.96	A					
3 Besiege	8	A	1.0 b	0.0 a	1.0 c	0.0 c	3015 a
4 Karate Z	1.96	A	1.5 b	1.0 a	5.3 b	0.8 bc	2781 ab
5 Untreated			2.8 ab	1.0 a	8.8 b	1.3 b	2503 c

Means followed by same letter or symbol do not significantly differ (P=0.05, LSD). FAW = fall armyworm.

2.5) Evaluation of a new Bt technology for the control of thrips and tarnished plant bug. Monsanto has developed a new Bt trait that has activity on both thrips and tarnished plant bug. A PhD student, Scott Graham, at the University of Tennessee is evaluating the efficacy and potential value of this technology. This research is partially funded by Monsanto (≅ 50%). Thus, support from Cotton Incorporated is used to help support these efforts.

In 2016, trials were conducted at Research and Education Centers in Jackson and Milan, TN. The trial was designed as a spilt-plot design with three main factors. Factor A was level of tarnished plant bug control: untreated control, insecticide applications made on TPB populations, and weekly automatic applications. Factor B was the trait: Bt vs. non-Bt. Factor C was the level of thrips control: insecticide seed treatment + foliar application vs. untreated control.

Outcomes: For reasons of confidentiality, complete results will not be presented. However, the Bt trait provided protection from thrips similar or better than a non-Bt commercial seed treatment (Aeris, Bayer CropScience) treated with a foliar application of Acephate made at the first leaf stage. For tarnished plant bugs, the Bt trait significantly increased square retention in plots not treated with insecticide. The trait also significantly reduced the total number of tarnished plant bugs and especially large nymphs. Compared with the non-Bt cotton and based on current treatment thresholds, the Bt trait required three less insecticide applications for tarnished plant bug at one location (Jackson) and one less insecticide application at the other location (Milan). The trait also increased yield compared with non-Bt cotton not treated with an insecticides for tarnished plant bug. However, insecticide applications for tarnished plant bug increased yields in both Bt and non-Bt cotton, indicating foliar insecticides for tarnished plant bug will still be needed for this new technology.

3) Support of General Extension Activities. Funding for this project is used to support general IPM Extension activities in Tennessee. This includes the delivery of the annual Cotton Scout School held at the West Tennessee Research and Education Center. Scouts are delivered classroom-style and in-field training related to cotton plant development, insect management (identification, sampling, etc.) and disease and weed identification. A scouting notebook was prepared for each attendee. This project also supports the preparation and publication of *Insect Control Recommendations for Field Crops* (UT Publication, PB1768) which contains IPM information for cotton and other crops. This publication is also available at www.UTcrops.com.

Outcomes: Approximately 70 scouts participated in the Cotton Scout School during 2016. A scouting notebook was prepared for each attendee. 1,500 copies of the *Insect Control Recommendations for Field Crops* (PB1768) were distributed to clientele groups.

4) Regional Projects. In 2016, a limited amount of core Cotton Incorporated funding was received to support regional research efforts (Project 08-454 *Helicoverpa zea* Pyrethroid Resistance Monitoring; Project 15-994 Insect Management in Mid-South Cotton). Additionally, unsupported regional efforts are also performed. Shortened summaries are included here because state-support funding is substantially used to complete these efforts.

4.1) Bollworm - Pyrethroid Resistance Monitoring (edited from report prepared by Fred Musser, Mississippi State University). Bollworm is a pest in numerous crops where it may be exposed to pyrethroid insecticides. Since it can have 5 or more generations per year in the southern U.S., it has the potential to develop large populations. One to two of these generations occur in cotton, sometimes causing substantial economic loss. Because pyrethroid insecticides are relatively inexpensive, they were traditionally the first choice of growers for foliar control of bollworms. However, control with pyrethroids has become erratic in some regions, so knowledge of the susceptibility of bollworms to pyrethroid insecticides is critical for effective management of this pest.

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

<u>Outcomes</u>: A total of 8,047 moths were assayed during 2016. The number of assayed moths per state range from 120 in Missouri to 1815 in Virginia. Average survival to the 5 µg cypermethrin / vial concentration was 36.1% in 2016 (Table 4), which was the highest rate of survival since monitoring began in 2007 and more than twice as high as most years (Fig. 8). The states that had been having the highest survival didn't change much in 2016, but the states that previously had susceptible populations had large increases in survival (Fig. 2), with South Carolina going from 6.4% to 43.3% survival in one year. Arkansas and Texas also saw >15% increases in survival from 2015 to 2016. For the first time since monitoring began, all states had >20% survival. The earliest tests with available data conducted during 1998-2001 did not have more than 20% survival in any state, and even as recently as 2012, this level of survival only occurred in Louisiana and Virginia.

Table 4. Bollworm survival (%) to 5 µg cypermethrin per vial in 24-h vial tests during 2016.

State	May	June	July	Aug	Sep	Overall	Bollworms tested
AR	13.3	38.1	52.1	41.0		40.6	499
GA		53.1	38.2	55.6	37.3	47.8	1126
LA	30.1	29.2	51.2	28.1	25.4	32.0	1512
MS	9.8	38.6	51.4	34.1	20.6	36.6	1081
MO			20.8			20.8	120
NC				40.1	25.0	31.4	176
SC		37.5	32.3	50.5	45.2	43.3	920
TN				34.4		34.4	438
TX			38.9	25.0	28.0	30.6	360
VA		36.5	39.3	46.2	43.3	43.2	1815
Average	17.7	38.8	40.5	39.4	32.1	36.1	8047

The rapid spread of resistance during the last two years suggests that the genetics of resistance have become well established, and any fitness costs associated with resistance are minor enough that these genes have spread throughout the southern United States. Even though pyrethroids are not applied to control bollworms as frequently as in the past, there are still many pyrethroid applications made in the agricultural landscape for various pests. This sustained selection for resistance genes continues to decrease pyrethroid susceptibility, making the choice of this class of chemistry for managing bollworms a risky decision that will often result in unsatisfactory control.

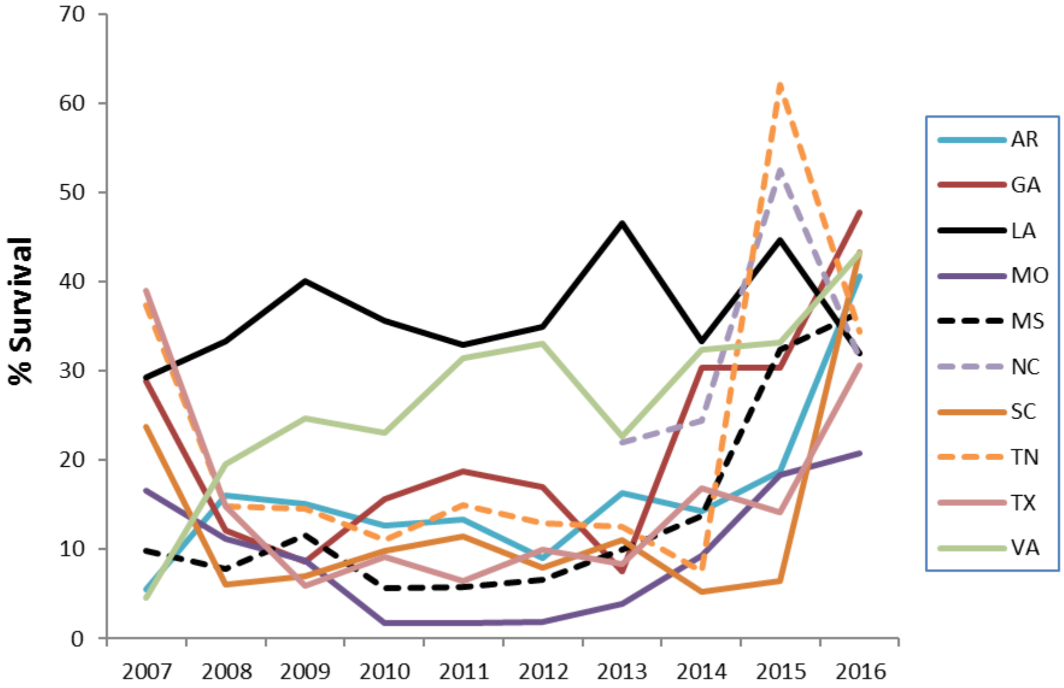


Figure 8. Average bollworm survival (%) at 5µg cypermethrin per vial from 2007 – 2016.

4.2) Evaluation of insecticide, fungicide and nematicide seed treatments (edited from report prepared by Don Cook, Mississippi State University). Thrips are one of the first insect pests to infest cotton following seedling emergence, with tobacco thrips, *Frankliniella fusca* Hinds, being the predominate species in the Mid-South. Following the introduction of the neonicotinoid seed treatments, imidacloprid and thiamethoxam, thrips management has been almost exclusively accomplished with seed treatments and supplemental foliar treatments. However, diminished performance of neonicotinoid seed treatments in recent years has been linked with tobacco thrips resistance to neonicotinoid insecticides. During 2016, two identical studies were conducted in Arkansas, Louisiana, Missouri, Mississippi, and Tennessee to evaluate the performance of standard insecticide seed treatments and alternative insecticide treatments targeting thrips. Cotton seed were treated by Dr. Gus Lorenz, and Phytogen 333 WRF cotton seed was used in all trials. Treatments were arranged in a randomized complete block with four replicates. In both studies, thrips densities were determined by sampling 5 plants per plot at the 1, 2, 3, and 4 leaf stage using a modified whole plant washing procedure. Also, plant damage was also estimated at these timing using a 0 - 5 scale, with a rating of 0 = no damage and 5 = severe damage. Seed cotton yields were collected in all trials.

For the regional seed treatment trial, insecticide treatments consisted on untreated seed, Gaucho, Aeris, Cruiser, Avicta Duo, and Orthene seed treated at rates shown in Table 5. Aldicarb (AgLogic 15G) has been reintroduced in some areas and was also included in this experiment. Treatments of Cruiser or Avicta Duo received Dynasty CST (3.1 oz/cwt) fungicide treatment while seed for all other treatments received Trilex Advanced 300FS (1.6 oz/cwt) fungicide. Planting dates ranged from 5 May to 6 Jun.

For the "alternatives test" treatments included acephate and cyantraniliprole (Verimark) evaluated as a seed treatment and an in-furrow spray, flupyradifurone (Sivanto) as an in-furrow spray, and aldicarb (AgLogic 15G) as an in-furrow granule (Table 6). Aeris seed treatment was included as a neonicotinoid comparison. Seed for all treatments received Trilex Advanced 300FS (1.6 oz/cwt) fungicide. Planting dates ranged from 4 May to 9 Jun.

Outcomes: On average across the regional seed treatment trial, both Cruiser and Avicta Duo demonstrated less than satisfactory efficacy against thrips, which was similar to observations from previous years. Also in these studies Gaucho (imidacloprid) did not perform as well as Aeris (imidacloprid plus thiodicarb), which has become the standard insecticide seed treatment that includes a neonicotinoid component. Aldicarb (AgLogic 15G) has been reintroduced in some areas and was also included in these studies. Of the products tested, only AgLogic and Acephate applied as a seed treatment demonstrated performance similar to Aeris when averaged across these regional trials. However, all insecticides increased yield compared with the control (fungicide only treatment).

On average across the regional trial done to evaluate potential alternatives to neonicotinoid seed treatments, only Acephate applied as a seed treatment and AgLogic provided efficacy against thrips similar to the standard Aeris. Verimark (cyantraniliprole) as a seed treatment and Sivanto applied as an in-furrow spray did not perform well. All of the insecticide treatments, except Sivanto and the Verimark seed treatment, resulted in significantly higher yields than the control (data not shown).

Table 5. Thrips numbers and injury ratings resulting from the use of insecticide seed treatments and AgLogic (Tennessee, 2016).

Description				Thrips Injury		Thrips Counts		Thrips Injury		Thrips Counts	
Pest Stage Majority				0-5 scale		Ad + Imm.		0-5 scale		Ad + Imm.	
Sample Unit				1 Plot		5 Plants		1 Plot		5 Plants	
Days after planting				22 DP-1		22 DP-1		28 DP-1		28 DP-1	
Treatment		Rate									
1	Untreated			3.5	a	97.5	a	3.1	a	35.5	a
2	Orthene 97	6.4	oz ai/cwt	3.2	a	81.3	ab	2.9	ab	51.3	a
3	Orthene 97	15	oz ai/cwt	2.8	b	54.8	bc	2.7	b	31.3	a
4	Orthene 97	24	oz ai/cwt	2.6	b	46.8	cd	2.6	b	33.0	a
5	Cruiser	0.375	mg ai/seed	3.3	a	80.3	ab	3.0	a	45.8	a
6	Gaucho 600 FS	0.375	mg ai/seed	1.6	c	61.3	bc	2.1	c	44.0	a
7	Aeris	0.75	mg ai/seed	0.9	e	35.0	cd	1.3	e	26.0	a
8	Avicta Duo	0.525	mg ai/seed	1.7	c	41.8	cd	1.6	d	39.8	a
9	AgLogic	5	lb/a	1.3	d	22.8	d	1.5	de	23.5	a

Means followed by same letter or symbol do not significantly differ (P=0.05, LSD).

Results in Tennessee were somewhat unique compared with other locations. Aeris performed better than other treatments in reducing thrips injury (e.g. Tables 5 and 6). Both Cruiser and Orthene seed treatments performed relatively poorly, whereas Avicta Duo provided protection similar to AgLogic (Table 5). Substantial rainfall shortly after planting likely reduced the efficacy of AgLogic and Orthene treatments, which are relatively water soluble. As seen regionally, Sivanto did not provide adequate plant protection.

Table 6. Thrips numbers and injury ratings resulting from the use of insecticide seed treatments and in-furrow application of insecticides (Tennessee, 2016).

Description				Thrips counts		Thrips Injury		Thrips Injury	
Pest Stage Majority				Adult + Imm.		0 - 5 scale		0 - 5 scale	
Rating Date				5/31/2016		6/1/2016		6/6/2016	
Sample Size, Unit				5 plants		1 Plot		1 Plot	
Trt-Eval Interval				20 DP-1		21 DP-1		26 DP-1	
Treatment		Rate							
1	Untreated			43.0	a	2.8	a	2.7	a
2	Orthene 97*	15	oz ai/cwt	37.5	abc	1.9	b	2.3	ab
3	Orthene 97**	1	lb/a	28.0	bcd	1.7	bc	2.0	bcd
4	Verimark*	13	oz ai/cwt	28.5	bcd	1.9	b	1.9	cd
5	Verimark**	13	fl oz/a	23.3	cd	1.7	bc	2.0	bc
6	AgLogic**	3.5	lb/a	22.5	d	1.7	bc	1.6	de
7	AgLogic**	5	lb/a	22.5	d	1.5	c	1.4	e
8	Sivanto**	7	fl oz/a	41.0	ab	1.9	b	2.6	a
9	Aeris*	0.75	mg ai/seed	7.0	e	1.1	d	0.7	f

Means followed by same letter or symbol do not significantly differ (P=0.05, LSD).
* Seed treatment
** In-furrow application

4.3) Validation of a new threshold for controlling caterpillar pests in non-Bt and Bt cotton (edited from report prepared by Dr. David Kerns, LSU AgCenter). The introduction, in 1996, of transgenic cotton containing genes expressing *Bacillus thuringiensis* (Bt) proteins ushered in a new era in cotton insect pest management. The first Bt cotton introduced in the U.S. was Bollgard I which expressed the Cry1Ac endo-toxin. This toxin was highly effective towards tobacco budworm but only moderately toxic towards bollworm. Insecticide applications targeting tobacco budworm were completely eliminated, while those targeting bollworm were greatly reduced. To increase efficacy and for resistance management, dual and multi-Bt gene cotton varieties have since been introduced including Bollgard II, WideStrike and WideStrike 3. Although these introductions have increased the efficacy of transgenic cotton targeting lepidopteran pests, including bollworm, there are still incidents where unacceptable fruit injury is experienced and insecticidal over sprays are required to preserve yield. The objective of this project was to validate a new treatment threshold for Bt cotton based on square and boll injury.

A threshold of 6% injured fruit (average of equal numbers of square and bolls) was suggested based on data collected in 2014 and 2015. In 2016, threshold validation tests were conducted at seven locations across four states in the Mid-South. Plots were 4 rows wide x 40-50 ft in length in a 3 × 3 factorial with four replications. Factor A consisted of cotton technologies: non-Bt, WideStrike or Bollgard 2. Factor B consisted of either a non-treated, sprayed at a threshold of 6% injured fruit (squares and bolls), or a preventative threshold sprayed at first occurrence of bollworm egg lay and/or small larvae. The insecticide application at all locations was Prevathon at 19 fl-oz per acre. Square and boll injury were determined prior to foliar treatment and weekly thereafter by arbitrarily sampling 25 squares and 25 bolls per plot. All plots were harvested and yields were determined. Profitability (P) was determine for each location base on the equation: $P (\$^{-acre}) = V - ((C_I \times A) + C_T))$. Where V is the crop value (lbs lint/acre × \$0.70), C_I is the cost of the insecticide and insecticide application (set at \$22.50 per acre), A is the number of applications, and C_T is the cost of the Bt technology set at \$27.00 per acre for both WideStrike and Bollgard 2.

Profitability data was pooled across each location by variety for analysis. Data were analyzed using PROC GLIMMIX ($P < 0.05$) with random effects of site, rep(site) and variety*rep(site). Where significant ($P < 0.05$) LSMEANS were separated using Tukey's HSD ($P < 0.05$).

Outcomes: In Tennessee, application of Prevathon significantly increased yield in a non-Bt and WideStrike cotton variety. A threshold of 6% fruit injury was not met in the BG2 variety, and yield were not significantly higher in the BG2 variety when Prevathon was applied (data not shown). Across all locations, significant yield increases were observed for all technologies when Prevathon was applied. Profitability was similarly increased by the application of Prevathon either preventatively or according to the 6% threshold (Fig. 9). These results suggest that percent fruit injury can be used as a viable treatment threshold for cotton. This threshold may vary depending on crop value, crop yield potential and control cost. However, it appeared to work equally well on a non-Bt and Bt cotton varieties. Field validation demonstrated that using the threshold resulted in fewer insecticide applications and profitability equivalent to preventative sprays.

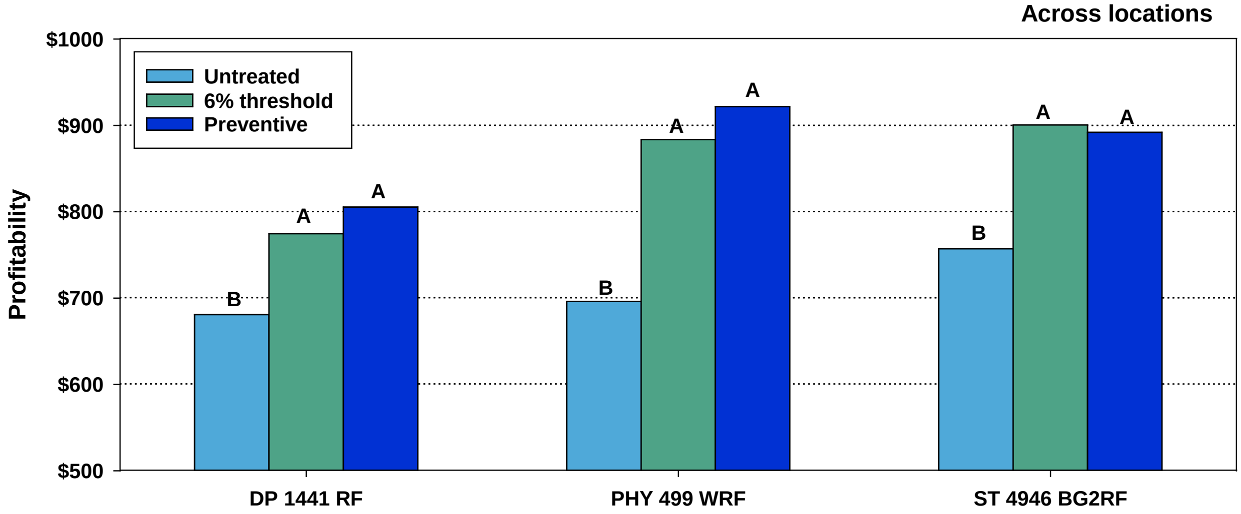


Figure 9. Profitability ($/acre) of non-Bt (DP 1441 RF), WideStrike (PHY 499 WRF) and Bollgard 2 (ST 4946 BG2RF) technologies either non-treated, sprayed at a threshold of 6% injured fruit or sprayed preventatively, across 7 locations in the Mid-South.

4.4) Termination of Insecticide Applications for Tarnished Plant Bug in Cotton (edited from report prepared by Dr. Jeff Gore, MSU). Multiple experiments were conducted during 2015 and 2016 in Arkansas, Louisiana, Mississippi, Missouri, and Tennessee to determine the optimum time to terminate insecticide sprays for tarnished plant bugs without sacrificing yields. All experiments were in a randomized complete block design with four replications. Plot size was 4-8 rows by 40-50 ft. at each location. The treatments included terminating insecticide applications for tarnished plant bug at specified weeks of flowering. The weeks when insecticides were terminated included weeks 2, 3, 4, 5, and 6 of flowering. For all of the termination treatments, plots were sprayed weekly or twice per week beginning at first flower. In addition to the termination treatments, an untreated control was included at each location in each year. A season long control was included in 2015 and a threshold treatment was included in 2016. For all sprays, insecticides were selected to maximize control and were specific to each location. Plots designated for termination treatments were sprayed through the designated weeks for termination. After insecticide sprays were terminated for specific plots, those plots were not sprayed with insecticides with tarnished plant bug activity throughout the remainder of the season.

<u>Outcomes</u>: In Tennessee, tarnished plant bug numbers in untreated plots exceeded 90 tarnished plant bugs per 10 row feet by the third week of bloom. All insecticide application made up to the third week of flowering increased yield, whereas applications made after the third week of bloom did not significantly increase yield (data not shown). On average across all locations in 2015 and 2015, insecticide applications for tarnished plant bug made after the fourth week of bloom did not significantly increase yield compared with season long control or applications made on a threshold basis (Fig. 10). In general, most consultants and growers do not track nodes above white flower and heat unit accumulation during the season. As a result, termination of insecticide applications targeting tarnished plant bug is not consistent and sprays are likely made much later in the flowering period that do not provide an economic return. These results may reduce up to 2 late season insecticide applications in many areas.

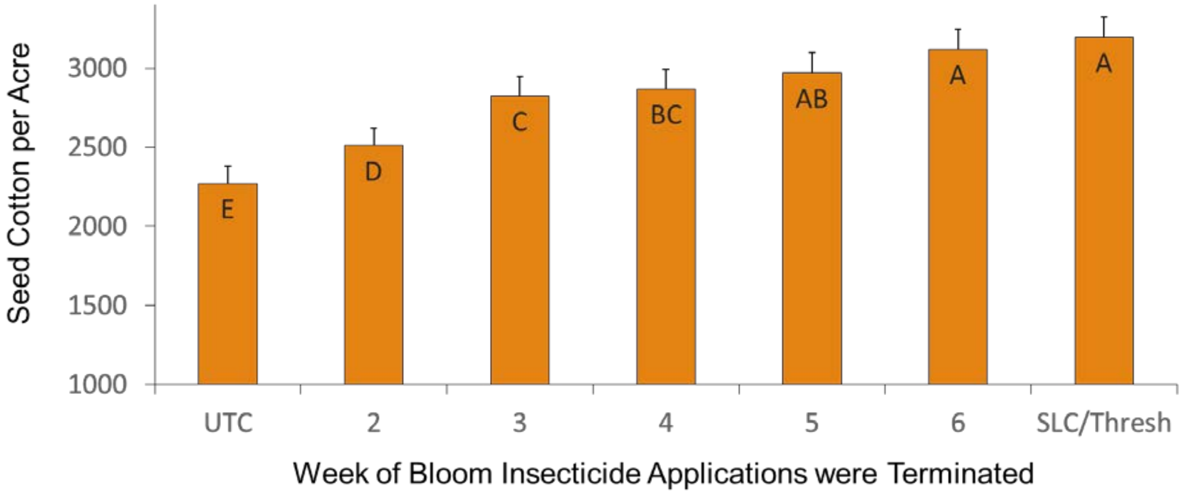


Figure 10. Average cotton yields across all trials (N = 16) in 2015 and 2016 where insecticide applications targeting tarnished plant bug were terminated at different weeks of flowering. SLC = season-long control; UTC = Untreated control.

4.5) Monitoring for bollworm resistance to Bt toxins (edited from report prepared by Dr. David Kerns, Texas A&M University). The risk of bollworm developing resistance to Bt toxins expressed in cotton and corn is of concern. Cry1A and Cry 2A proteins are present in both Bt corn and Bt cotton. Cry1F is present in some Bt corn hybrids and WideStrike cotton varieties. WideStrike 3, Bollgard 3, and TwinLink Plus all also express the Vip3a protein. The objective of this study was to conduct bioassays on field-collected populations of bollworm submitted from academic cooperators. A total of five field populations of bollworm were collected from across the central and eastern Cotton Belt in 2016. F1 or F2 generations of these field-collected populations were used for the bioassays described below. Collection information is listed in Table 7 (below). In addition, two susceptible colonies were also used in the current study, including a susceptible colony obtained from USDA-ARS, Stoneville, MS and a colony from a commercial source Benzon Research Inc., Carlisle, PA.

Susceptibility of bollworm was determined against four Bt proteins: Cry1Ac, Cry1F, Cry2Ab2, and Vip3a. Cry1Ac protein was provided by Monsanto Company as lyophilized MVPII powders with 20.0% AI. Cry1F protein was provided by Dow AgroSciences as lyophilized powders with 53.0% AI. Cry2Ab2 protein was provided by Monsanto in the form of lyophilized (freeze-dried) Bt-corn leaf powder expressing ~4 mg of Cry2Ab2 protein/g. Syngenta Biotechnology Inc. provided the Vip3Aa19 protein with a purity of 77.8%. Resistance ratios, calculated from the LC50 value of test populations versus a known susceptible populations, are used for the discussion below. Complete assay details have been described elsewhere by Dr. David Kerns.

Outcomes: Two populations of bollworms were submitted by Tennessee, one from grain sorghum at the West Tennessee Research and Extension Center and one from a field of BG2 cotton in Lauderdale County. The population from sorghum was collected in mid-August and most likely originated from moths emerging from nearby Bt field corn. The population from cotton was collected in late-August after a control failure was reported in a grower's field. Boll damage in this field was approximately 20% at the time of the collection.

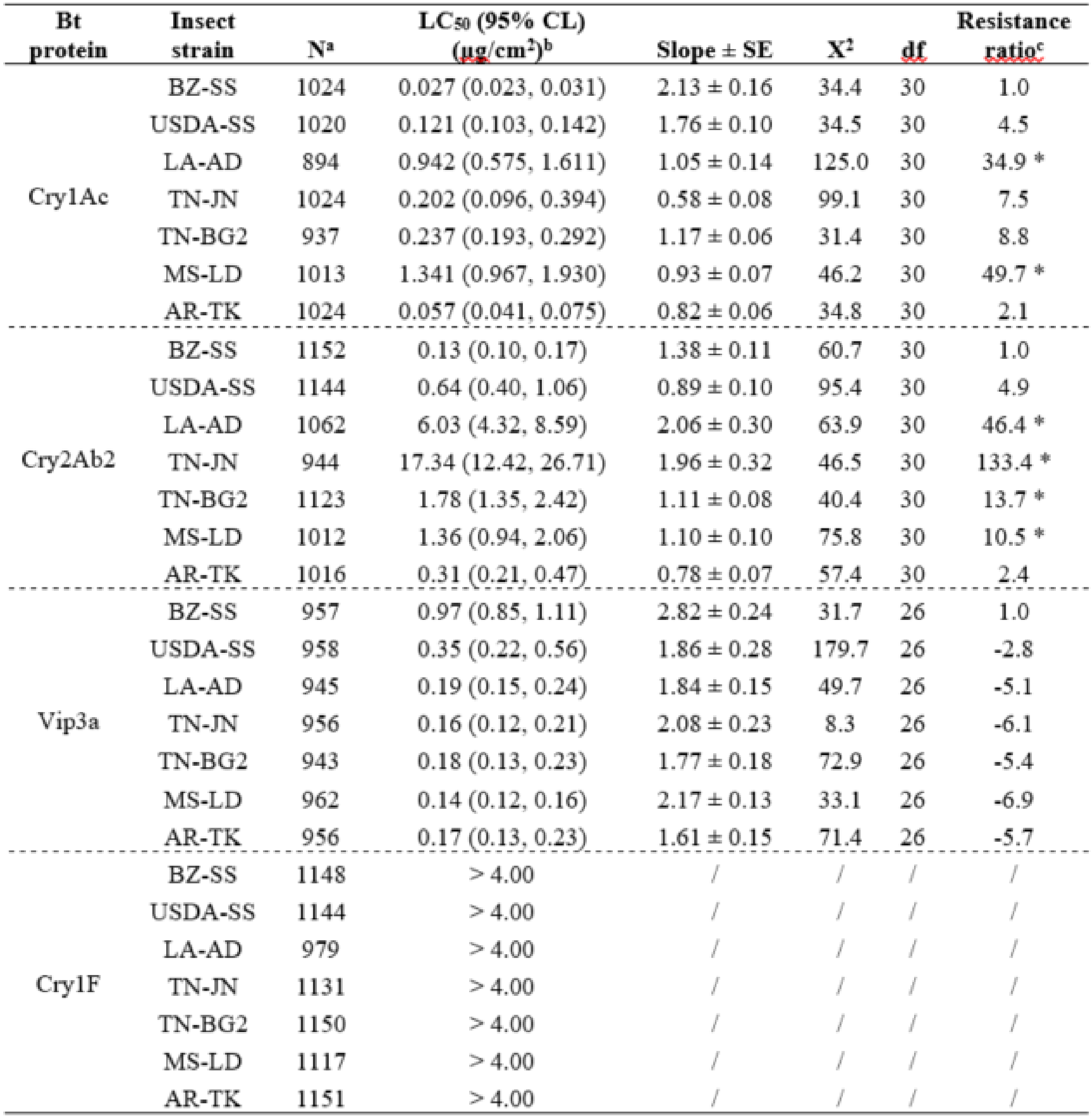
Table 7. Insect sources used in the study.

Insect colony	Collected Site	Collected date	Host	No. Pupae
BZ-SS	Benzon	/	/	/
USDA-SS	USDA, MS	/	/	/
LA-AD	Alexandria, LA	07/27/2016	BG2 cotton	37
TN-JN	Jackson, TN	08/15/2016	Grain sorghum	118
TN-BG2	Jackson, TN	08/26/2016	BG2 cotton	92
MS-LD	Leland, MS	06/29/2016	VT2P corn	182
AR-TK	Toadsuck, AR	07/07/2016	Non-Bt corn	114

None of the populations tested showed appreciable mortality to Cry1F, regardless of the rate tested. In contrast, all populations were highly susceptible to Vip3a. Using a resistance ratio >10 as a criteria for resistance, the population collected from BG2 cotton in Tennessee did not show high levels of tolerance to Cry1Ac (Table 8). However this population did have some tolerance to Cry2Ab2 (RR = 13.7). Interestingly, the population collected from grain sorghum had a very high resistance ratio for Cry2Ab2. This resistance ratio (133.4) is perhaps higher than ever recorded for a field-collected population of bollworm.

Populations assays from other states showed variable levels of tolerance or resistant to Cry1A or Cry2A proteins. Collectively, these data suggest that bollworms are becoming decreasingly susceptibility of one of more Bt toxins present in Bt cotton and Bt corn. No populations showed tolerance to the Vip3a protein which is a Bt trait being included now in some newer Bt cotton and corn hybrids. These populations are being maintained by Dr. Kerns to further assess the level and mechanisms of resistance.

Table 8. LC$_{50}$ and 95% confidence limits (CL) based on larval mortality of bollworm to four Bt proteins.



Bt protein	Insect strain	N[a]	LC$_{50}$ (95% CL) (µg/cm^2)[b]	Slope ± SE	X^2	df	Resistance ratio[c]
Cry1Ac	BZ-SS	1024	0.027 (0.023, 0.031)	2.13 ± 0.16	34.4	30	1.0
	USDA-SS	1020	0.121 (0.103, 0.142)	1.76 ± 0.10	34.5	30	4.5
	LA-AD	894	0.942 (0.575, 1.611)	1.05 ± 0.14	125.0	30	34.9 *
	TN-JN	1024	0.202 (0.096, 0.394)	0.58 ± 0.08	99.1	30	7.5
	TN-BG2	937	0.237 (0.193, 0.292)	1.17 ± 0.06	31.4	30	8.8
	MS-LD	1013	1.341 (0.967, 1.930)	0.93 ± 0.07	46.2	30	49.7 *
	AR-TK	1024	0.057 (0.041, 0.075)	0.82 ± 0.06	34.8	30	2.1
Cry2Ab2	BZ-SS	1152	0.13 (0.10, 0.17)	1.38 ± 0.11	60.7	30	1.0
	USDA-SS	1144	0.64 (0.40, 1.06)	0.89 ± 0.10	95.4	30	4.9
	LA-AD	1062	6.03 (4.32, 8.59)	2.06 ± 0.30	63.9	30	46.4 *
	TN-JN	944	17.34 (12.42, 26.71)	1.96 ± 0.32	46.5	30	133.4 *
	TN-BG2	1123	1.78 (1.35, 2.42)	1.11 ± 0.08	40.4	30	13.7 *
	MS-LD	1012	1.36 (0.94, 2.06)	1.10 ± 0.10	75.8	30	10.5 *
	AR-TK	1016	0.31 (0.21, 0.47)	0.78 ± 0.07	57.4	30	2.4
Vip3a	BZ-SS	957	0.97 (0.85, 1.11)	2.82 ± 0.24	31.7	26	1.0
	USDA-SS	958	0.35 (0.22, 0.56)	1.86 ± 0.28	179.7	26	-2.8
	LA-AD	945	0.19 (0.15, 0.24)	1.84 ± 0.15	49.7	26	-5.1
	TN-JN	956	0.16 (0.12, 0.21)	2.08 ± 0.23	8.3	26	-6.1
	TN-BG2	943	0.18 (0.13, 0.23)	1.77 ± 0.18	72.9	26	-5.4
	MS-LD	962	0.14 (0.12, 0.16)	2.17 ± 0.13	33.1	26	-6.9
	AR-TK	956	0.17 (0.13, 0.23)	1.61 ± 0.15	71.4	26	-5.7
Cry1F	BZ-SS	1148	> 4.00	/	/	/	/
	USDA-SS	1144	> 4.00	/	/	/	/
	LA-AD	979	> 4.00	/	/	/	/
	TN-JN	1131	> 4.00	/	/	/	/
	TN-BG2	1150	> 4.00	/	/	/	/
	MS-LD	1117	> 4.00	/	/	/	/
	AR-TK	1151	> 4.00	/	/	/	/

[a] Total number of neonates assayed.

[b] The LC$_{50}$ value of an insect strain was considered to be greater than the highest Bt protein concentration used in the bioassay if its larval mortality was <50% at the highest concentration. Larval mortality were calculated based on the number of dead larvae plus survivors that were still in the first instar (mortality = dead+L1) divided by the total number of insects assayed.

[c] Resistance ratio for Bt protein were calculated by dividing the LC$_{50}$ value of an insect population by that of the susceptible strain (BZ-SS). If the LC$_{50}$ an insect population was smaller than that of the BZ-SS, a negative sign was assigned to the resistance ratio.

* Indicates significant resistance ratios that were ≥ 10-fold.