## USE OF ALTERNATE HOSTS AS A TRAP FOR TARNISHED PLANT BUGS AND A REFUGE FOR BT-SUSCEPTIBLE TOBACCO BUDWORMS

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#### **Abstract**

In 1998, we continued testing the potential for a group of non-crop plant species to act as a trap crop for the tarnished plant bug and a refuge for heliothines (tobacco budworm and cotton bollworm) susceptible to endotoxin proteins found in Bt cotton. Field studies were conducted, as in 1997, on commercial farms at several locations in the Mississippi Delta to determine the effectiveness of the trap crop/refuge system. Four alternate hosts (kenaf, pigweed, velvetleaf and sesame) were planted in a strip in the middle of three Bt cotton fields in 1998. As in 1997, velvetleaf appeared to be an excellent refuge host for heliothines. Pigweed, at least during part of the season, was highly preferred over cotton by plant bugs, and plant bug numbers remained low in cotton adjacent to the weed strip throughout June. However, populations of tarnished plant bugs exceeded treatment thresholds in all fields during July. Treatment of the weed strip with insecticide active against plant bugs significantly reduced their numbers in the adjacent cotton.

#### **Introduction**

Midsouth cotton producers are facing serious insect control problems, especially insecticide resistant populations of tarnished plant bug (Snodgrass and Scott 1988; Snodgrass 1996a,b) and tobacco budworm (Plapp 1987, Plapp et al. 1990, Graves et al. 1991, Elzen et al. 1992, Luttrell and Layton 1995). An alternative method of controlling these insecticide resistant pests is needed. The tarnished plant bug is frequently the key pest of cotton in the Midsouth, because it is often the first pest that requires control action during the growing season (Scott et al. 1985). Sprays targeted at tarnished plant bugs reduce natural enemies and often cause outbreaks of other pests, especially the tobacco budworm and the cotton bollworm (Luttrell 1994).

Traditionally, tarnished plant bugs have been controlled with insecticides initiated as the plant reaches first square and continued on an as-needed basis later in the season (Layton 1998). Plant bugs are often suppressed later in the season by insecticides targeting boll weevil and heliothines. In recent years, resistance to organophosphate and pyrethroid insecticides in tarnished plant bug populations in the Midsouth has been reported (Snodgrass and Scott 1988; Snodgrass and Elzen 1995; Snodgrass 1996a,b). Successful eradication of the boll weevil (Brazzel 1996) and widespread use of Bt cotton (Umbeck et al. 1987, Layton 1996) will reduce the need to control boll weevil and heliothines, but the tarnished plant bug remains as a key pest in the system.

In the early 1960's, Stern et al. (1964, 1969) proposed alternative means for controlling *Lygus hesperus* (Knight) in cotton and designed an alfalfa harvest strategy for the San Joaquin Valley of California that reduced the movement of *L. hesperus* into cotton. This technique, although effective, was abandoned because the staggered harvest intervals proved to be too difficult to manage and the economic cost still favored continued use of insecticides. The value of interplanting alfalfa with cotton was not limited to control of *L. hesperus* in the western United States (Stern et al. 1964, Sevacherian and Stern 1973, and Godfrey and Leigh 1994). Schuster demonstrated the effectiveness of this same technique for management of tarnished plant bugs in Mississippi (Schuster 1980).

Manipulation of preferred hosts either by mowing or insecticide application has also been studied as a management approach for Lygus control. Mueller and Stern (1973) found that accurate timing of insecticide treatments to safflower before L. hesperus dispersal to cotton significantly reduced densities of L. hesperus in cotton. Fleischer et al. (1988) and Snodgrass and Stadelbacher (1994) showed that management of natural host plants either by mowing or insecticide treatment could be effective in managing tarnished plant bug movement into cotton. Timing of treatment or mowing is critical, however, because destroying the host at the wrong time can flush the insects into a susceptible crop (Fleischer et al. 1988). As a result, trap crops must be carefully managed to prevent the production and dispersal of large densities of plant bugs into the cotton system. Recent work by Fleischer and Gaylor (1987, 1988) in Alabama demonstrated the value of using wild host plants to avoid tarnished plant bug problems in cotton.

Bt cotton promises to be an effective means of controlling tobacco budworm and cotton bollworm (Jenkins et al. 1991). Cotton bollworms are less susceptible than tobacco budworm to Bt cotton, but control is acceptable under most

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population densities (Lambert et al. 1996, 1997; Benedict et al. 1996; Mahaffey et al. 1995). Development of resistance to the insecticidal protein in pest populations is a major concern because continuous exposure to the insecticidal protein will result in season-long selection for resistance (Tabashnik 1994). With a resistant gene frequency as high as 1 in 1000, severe resistance problems could develop in only a few generations (Gould et al. 1995, Gould 1998). Simulation studies of resistance evolution in the pest-crop system predict that when refuges of only 5-10% are incorporated into a management system, dramatic increases in the effective life of Bt cotton are observed (Gould et al. 1995, Liu and Tabashnik 1997, Gould 1998). Current EPA guidelines require growers utilizing Bt cotton to plant a percentage of their crop as a refuge to produce susceptible adults (EPA 1997). Wild host plants may also serve as refuge for heliothines throughout the growing season (Stadelbacher et al. 1986), but quantitative information on population structure is generally lacking.

With documented tarnished plant bug and heliothine resistance to insecticides, anticipated boll weevil eradication and expanded use of Bt cotton, the utilization of a trap crop system for the tarnished plant bug has an increased economic appeal. Because Bt cotton needs some type of refuge, the trap crop has an additional utility if it could be used to maintain populations of heliothines which are susceptible to insecticidal plants. The purpose of this study was to develop a trap crop system that could be used to manage tarnished plant bugs and jointly provide a suitable refuge for tobacco budworm and cotton bollworm genotypes susceptible to Bt cotton.

## **Materials and Methods**

#### **Selection of Alternate Hosts**

Because tarnished plant bug, tobacco budworm and cotton bollworm are highly polyphagous insects and attracted to many non-crop hosts (Snow and Brazzel 1965, Snodgrass et al. 1984, Stadelbacher et al. 1986, Young 1986, Fleischer and Gaylor 1987), a large number of potential plant species were candidates for the trap crop/refuge system. We chose four species of alternate hosts to serve as our trap/refuge crop based on previous screening studies (Craig 1998). Redroot pigweed, *Amaranthus retroflexus* (L.), and kenaf, *Hibiscus cannabinus* (L.), were to serve as alternate hosts for the tarnished plant bug. Velvetleaf, *Abutilon theophrasti* (L.), and sesame, *Sesamum indicum* (L.), were selected as refuge hosts for tobacco budworm and cotton bollworm.

#### Plot Design

Large-field plots were established on three commercial farms in the Mississippi Delta to test the effectiveness of the trap crop/refuge system in 1998. Plot sites were chosen based on the availability of cooperators, the absence of preemergence herbicides and location within the Mississippi Delta. The three locations chosen were Morgan City (Leflore County), Sumner (Tallahatchie County) and Perthshire (Bolivar County).

Each experimental location (replicate) consisted of a 20-60 ha field of cotton. In the center of the field, eight rows were left untreated with herbicide. These eight rows (300-400 m long) were planted with the trap crop/refuge species. Two rows of each plant species were planted on raised beds spaced 97 cm apart. In the eight-row strip, velvetleaf was planted on rows 1 and 8, sesame on rows 2 and 7, pigweed on rows 3 and 6, and kenaf was planted in the center two rows. The trap crop/refuge was planted between (April 16-23) using an International 900 four row planter. Velvetleaf and kenaf were planted using sorghum plates. Due to the small size of sesame and redroot pigweed seed, these plant species were planted using the insecticide hoppers. At both the Perthshire and Sumner locations, spot planting of the various alternate hosts was done in early May with a onerow push planter to fill in stand gaps. The adjacent cotton was planted by the cooperator within 30 d of the trap crop/refuge planting. Cotton cultivars used were 'D&PL NuCotn 33', 'Paymaster 1220 BG' and 'Paymaster 1220 RR BG'. All cotton cultivars planted expressed the Cry1Ac delta endotoxin protein of B. thuringiensis.

At each location, the host strip was separated into a sprayed and unsprayed halves. Approximately 150-200 m of each strip were treated each week with imidacloprid (Provado<sup>®</sup> 0.05 kg (AI)/ha) until mid July. This was done to determine if treating the strip would increase heliothine larval survival and decrease tarnished plant bug movement into the adjacent cotton.

## **Sampling**

Sampling for insects was initiated about the time the cotton began squaring (June 1) and continued at about weekly intervals until late July. We established four sampling transects extending from the center of the trap crop/refuge strip into the cotton. Two transects were associated with the insecticide-treated half of the strip, and the other two transects were associated with the untreated half. Insect samples were taken for each plant species within the strip, along each transect, and also at increasing distances from the strip in the cotton (5-10, 45-50, 95-100, and 145-150 rows) to determine if there was a distance effect on insect numbers caused by the trap crop/refuge strip.

The primary method of sampling was visual observation. Generally, the top 15 cm (terminal) of each of the plant species was visually searched to observe heliothine eggs, larvae and tarnished plant bugs. On several occasions, whole plant samples were taken rather than just terminal counts. Samples of 10-20 consecutive plants were taken at each sampling site along each of the four transects. Heliothine larvae were categorized as small (1<sup>st</sup> and 2<sup>nd</sup> instar), medium (3<sup>rd</sup> and 4<sup>th</sup> instar) or large (5<sup>th</sup> instar and larger). Heliothine larvae from visually sampled plants were collected each week and placed in 29 ml plastic cups

containing a wheat germ diet (King et al. 1985). Larvae were reared and moths were identified as they emerged.

Two drop cloth samples were also taken in the cotton at each sampling site, for each transect, usually on a weekly basis. In both 1997 and 1998, other methods of sampling (e.g., suction samples and square retention) were done, but these data will not be addressed here. Data on beneficial insects were also collected, but comments in this paper are confined to effects on tarnished plant bug and heliothine populations.

### Data Analysis

Data were analyzed with split-plot ANOVA procedures (SAS 1988). Main plots were the presence or absence of insecticides. Subplots were sample sites within the field at each experimental location. Analyses were also done separately for the months of June and July so that temporal changes in treatment effects might be identified.

### **Results and Discussion**

## Summary of Results in 1997

During the first year of this study, in four test fields, the results were circumstantially encouraging in that no significant tarnished plant bug infestations occurred in any test field; whereas, relatively large populations of plant bugs were found on pigweed and kenaf. Also, velvetleaf proved an excellent refuge host for cotton. Nevertheless, the results from the 1997 study were not incontrovertible, particularly for the plant bug component, because distance from the trap crop did not affect plant bug populations in cotton. Also, treating the trap crop/refuge with insecticide had no significant effects on plant bug populations in cotton or heliothine populations within the refuge. Thus, it could not be conclusively demonstrated that the trap crop was responsible for reducing plant bug populations in cotton. The complete results of the research were published by Craig (1998).

Low plant bug populations in 1997 may explain why definitive effects of the trap crop on their populations in cotton could not be determined. One contributing factor explaining the low insect populations in the cotton was that the cooperating growers made several insecticide applications to each test field with materials that had activity on tarnished plant bug. In 1998, relatively few insecticide applications were made to the test fields, and when they were made, we avoided the use of insecticides with high activity on tarnished plant bugs.

### Tarnished Plant Bug (1998)

Seasonal averages of visual samples showed that tarnished plant bug adults preferred pigweed to all other plant species sampled (Table 1). In June, tarnished plant bug populations in cotton were almost indiscernible despite high populations on pigweed and kenaf. In July however, more plant bugs were found in cotton that in pigweed or the other species

within the trap/refuge strip. Populations of plant bugs exceeded Mississippi's treatment threshold (15 bug /100 plants, Layton 1998) in all test fields during mid July based on visual examination of the plants. As in 1997, distance away from the trap crop had no statistical impact on plant bug populations in the cotton, although a 20% decrease in plant bug numbers was observed at a distance of 145-150 rows from the trap during July (Table 1). A numerical reduction in plant bugs was observed in the insecticide treated portion of the trap crop and also in cotton adjacent to this treated area compared with the untreated portion (Table 2), but this difference was not statistically significant. Drop cloth samples in cotton also did not detect an effect of distance away from the trap crop on tarnished plant bug populations, but a significant (53%) reduction in plant bug populations was observed in cotton adjacent to the treated trap crop compared to cotton adjacent to the untreated trap (Table 3).

The observation that plant bug populations were highest in the trap crop/refuge strip in June, and then highest in cotton in July, indicates that the trap crop may have served as a plant bug nursery for the cotton. Based on the drop cloth data, treatment of the trap crop reduced plant bug numbers in the adjacent cotton, further indicating the trap crop was affecting the distribution of this pest in cotton and acting as a nursery when left untreated. Because the distance in cotton away from the trap crop had little effect on tarnished plant bug numbers, it is not clear what ultimate effect the trap crop had on populations. Indeed, it is possible the trap crop actually increased plant bug populations in the adjacent cotton. Regardless, it is evident that at least under some circumstances it would be necessary to spray the trap crop with insecticide to avoid its acting as a nursery for tarnished plant bugs.

# Heliothines (1998)

Results in 1998 were similar to 1997 in that velvetleaf proved to be a superior host for tobacco budworm and cotton bollworm compared to other alternate hosts, including cotton (Table 4). As expected very low numbers of heliothines were found in the Bt cotton, kenaf and pigweed; whereas, velvetleaf and sesame had relatively large populations of heliothines. Most of the larvae collected in sesame were small, and a much greater number of medium and large sized larvae were found in velvetleaf than in sesame (Table 4). These data agree with data collected in 1997. Sampling efficiency could be partly responsible for differences observed between sesame and velvetleaf, but low numbers of large larvae have been observed in sesame in previous studies (Laster and Furr 1972). Sesame is an attractive host for heliothines, but larval survival may be low, and it may not be an ideal refuge host. Velvetleaf was the best-suited plant for development of heliothine populations in this study.

Because heliothine populations were so low in cotton, we did not expect, nor could we detect, differences in

heliothine populations in cotton as a result of sampling distance away from the refuge (Table 4). Insecticide treatments may be needed to eliminate predators and achieve higher heliothine survival in the refuge strip. In both 1997 and 1998, heliothine numbers were higher in treated portions of the refuge, but these differences were not significant (Table 5).

In 1997, 74 larvae (24%) of the larvae collected from velvetleaf survived to the adult stage. Of the 74 emerging as moths, 95% were tobacco budworms and 2.5% were cotton bollworms (Craig 1998). Overall, fewer larvae were collected from velvetleaf in 1998 than in 1997, in part due to a reduced level of sampling and because one less test location was involved. Of the 30 larvae collected from velvetleaf that survived to adulthood in 1998, 80% were cotton bollworms and 20% were tobacco budworms. Thus, velvetleaf appears to be a good host for both tobacco budworm (1997) and cotton bollworm (1998). Only 2 of 10 worms collected in Bt cotton survived to adulthood in 1998, and both were cotton bollworms. Regardless of species, overall heliothine numbers were higher in the trap crop/refuge strip than in the adjacent cotton, and production of large numbers of both species of heliothines is a major goal of the proposed trap crop/refuge system.

### Summary

This research demonstrated that higher numbers of heliothines and tarnished plant bugs were found in the trap crop/refuge strips than in the adjacent cotton. Redroot pigweed was identified as the most preferred host of tarnished plant bug while velvetleaf proved to be excellent hosts for heliothines. However, our data indicate that treating the trap crop/refuga may be necessary to avoid movement of plant bugs into adjacent cotton may also increase the value of the refuge in producing Bt susceptible heliothines. Velvetleaf, although a noxious weed, may have real potential to increase numbers of heliothines in a refuge. Although consistently higher numbers of insects were found to inhabit the trap crop/refuge strips, data from the adjacent cotton did not fully define the effects of the trap crop/refuge system. Further research is needed to adequately measure the effective size of this trap crop/refuge system in the Mississippi Delta.

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Table 1. Mean numbers of tarnished plant bugs per 100 plants based on visual sampling.

Host	June	July	Season
Cotton (all rows)	0.05 B	17.41 A	8.54 BC
Rows 5-10	0.19 x	18.10 x	8.97 x
Rows 45-50	0.00 x	18.63 x	9.13 x
Rows 95-100	0.00 x	17.27 x	8.46 x
Rows 145-150	0.00 x	14.44 x	6.99 x
Velvetleaf	0.21 B	1.94 B	0.96 C
Sesame	8.39 B	6.39 AB	7.31 BC
Kenaf	13.47 B	6.11 AB	10.35 B
Pigweed	35.56 A	10.56 AB	23.06 A

Means, within columns, followed by the same letter are not significantly different (P < 0.05, LSD).

Table 2. Mean numbers of tarnished plant bugs per 100 plants, based on visual sampling, for samples associated with insecticide treated and untreated portions of the trap crop/refuge.

Host	Cotton	Other hosts	
Treated transects	7.83 a	8.56 a	
Untreated transects	9.29 a	11.69 a	

Means, within columns, followed by the same letter are not significantly different (P  $< 0.05,\,LSD$ ).

Table 3. Mean numbers of tarnished plant bugs per drop cloth in cotton by sampling location and for samples associated with insecticide treated and untreated portions of the trap crop/refuge.

Rows	June	July	Season
5-10	0.19 a	0.88 a	0.35 a
45-50	0.21 a	1.13 a	0.42 a
95-100	0.30 a	1.04 a	0.47 a
145-150	0.21 a	0.75 a	0.34 a
Treated transects	0.13 x	0.68 x	0.26 x
Untreated transects	0.33 y	1.25 x	0.55 y

Means, within columns, followed by the same letter are not significantly different (P < 0.05, LSD).

Table 4. Mean numbers of small, medium and large heliothine larvae per 100 plants based on visual sampling.

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Host	Small	Meduim	Large	Total
Cotton (all rows)	0.42 B	0.18 B	0.03 B	0.53 B
Rows 5-10				0.32 x
Rows 45-50				0.43 x
Rows 95-100				0.85 x
Rows 145-150				0.52 x
Velvetleaf	5.01 A	4.70 A	2.65 A	12.41 A
Sesame	3.24 A	0.45 B	0.30 B	4.03 B
Kenaf	0.12 B	0.58 B	0.12 B	0.82 B
Pigweed	0.00 B	0.28 B	0.00 B	0.28 B

Means, within columns, followed by the same letter are not significantly different (P < 0.05, LSD).

Table 5. Mean numbers of total heliothines larvae per 100 plants, based on visual sampling, for samples associated with insecticide treated and untreated portions of the trap crop/refuge.

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Host	Cotton	Other hosts	
Treated transects	0.52 a	4.77 a	
Untreated transects	0.54 a	4.29 a	

Means, within columns, followed by the same letter are not significantly different (P < 0.05, LSD).