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WHEN DOES BROWN STINK BUG, *EUSCHISTUS SERVUS* (SAY), BEGIN TO INJURE COTTON?

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Abstract

Field studies were conducted to determine the impact of brown stink bug, *Euschistus servus* (Say), infestations on pre-flowering and flowering cotton plants. Cotton seedlings (pre-squaring), cotton with a small (match-head) square, large (pre-candle) squares, and bolls were infested with one adult brown stink bug in a no-choice test. There were no significant differences in plant height, height to node ratio, square retention, and flower initiation for seedling cotton plants or cotton plants with a match-head square between infested and non-infested treatments for pre-flowering studies. There were no significant differences in abscission of large squares or bolls for infested and non-infested treatments. In boll infestation studies, the relationship between boll maturity, as determined by heat units beyond anthesis, and boll growth, abscission, and seedcotton yield was measured. Boll growth was significantly lower for bolls infested with brown stink bug through 266.5 heat units beyond anthesis. Brown stink bugs induced boll abscission through ca. 350 heat units beyond anthesis. Seedcotton yield was significantly reduced for infested bolls as compared to non-infested bolls through ca. 550 heat units beyond anthesis.

Introduction

The management of insect pests in mid-south and southeastern cotton, *Gossypium hirsutum* L., producing states has changed over the last decade. New insect management technologies include target-selective insecticides and Bollgard cotton. Both technologies have influenced scouting protocols, and have been contributed to the change in the insect pest spectrum because of a reduction in the use of broad-spectrum insecticides. The ability of crop managers in cotton to adapt to production practices and pests greatly influences the implementation, success, and longevity of these new technologies, and the management of secondary pests.

Fewer broad spectrum insecticides are applied against cotton pests as a result of the use of selective insecticides, adoption of Bollgard cotton, and boll weevil, *Anthonomous grandis grandis* Boheman, eradication programs. This has led to increased abundance of Hemipteran insect pests, including brown stink bug, *Euschistus servus* (Say), southern green stink bug, *Nezara viridula* (L.), and tarnished plant bug, *Lygus lineolaris* (Palisot de Beauvois) (Greene and Herzog 1999, Leonard et al. 1999, Roberts 1999). Additionally, the agricultural landscape in most cotton producing regions provides suitable hosts year-round. Wide-spread adoption of conservation tillage, crop rotation, and Conservation/Wetland Reserve Programs provide a broad range of host plants for stink bugs that subsequently infest cotton. Stink bugs infested 6,180,966 acres among the seventeen states of the cotton belt in 2001, ranking fourth among all cotton insect pests (Williams 2002).

Much of the data collected on stink bug (particularly southern green and green stink bugs) problems in cotton is related to their occurrence during boll development stages. Stink bugs can cause small bolls to abscise, and reduce lint quality, seed germination, and overall yield (Wene and Sheets 1964, Barbour et al. 1990, Greene et al. 1999). Little data has been published on the impact of stink bugs on cotton seedlings, or on flower buds (squares).

Cotton seedlings and squares are particularly sensitive to injury from several bugs, including clouded plant bug, *Neurocolpus nubilus* (Say), and tarnished plant bug (Tugwell et al. 1976). Tarnished plant bugs can feed in the plant terminal of cotton seedlings and subsequently cause the terminal to abort (Tugwell et al. 1976, Burris et al. 1997). Feeding damage by tarnished plant bugs also results in the abscission of pin-head squares, flower malformation, and boll abscission (Burris et al. 1997, Russell 1999). In Louisiana, tarnished plant bug infestations must be controlled in pre-flowering cotton when first position square retention falls below 70% and at infestation levels of insects ≥ 10% (Bagwell et al. 2002).

Stink bugs may be found in cotton fields from seedling emergence until harvest. Crop managers are concerned about the effects of stink bugs on pre-flowering cotton, if treatments should be initiated at these growth stages, and at what age is a boll safe from damage. Therefore, the following studies were conducted to determine when brown stink bugs begins to cause injury, through infestations on cotton plant seedlings, and reproductive structures of various ages.

Materials and Methods

Study Site and Plant Material

These studies were conducted at the Macon Ridge location of the Northeast Research Station near Winnsboro, Louisiana (Franklin Parish) during 2001 and 2002. The soil at the site was a Gigger-Gilbert silt loam complex. Plots of cotton used for infestations on seedlings (pre-squaring), cotton with small (match-head) squares, large (pre-candle) squares, and bolls in 2001 and 2002 was 'DP458 BRR' and 'Stoneville 4892 BR', respectively. Aldicarb was applied at-planting (Temik 15G, 0.5 lb AI/acre, Aventis Crop Science, Research Triangle Park, NC), and cultural practices and integrated pest management strategies were followed as recommended by the Louisiana State University Agricultural Center. No supplemental irrigation was applied to plots in 2001 or 2002.

Insects

Brown stink bug adults were collected during May and June of 2001 and 2002 from mustard, *Brassica* spp., and corn*, Zea mays* L. Late-season (July and August 2001, 2002) collections were made from soybean in northeast Louisiana. Insects were collected using a standard 38.1 cm diameter sweep net or hand-removed from the plant. Insects were held in a polypropylene cage (30.0 x 30.0 x 30.0 cm, BugDorm, Megaview Science Education Services CO. Ltd., Taichung, Taiwan) to reduce mortality from physical injury and to eliminate parasitized insects. Stink bugs were fed washed green beans, *Phaseolus vulgaris* (L.), and shelled peanuts, *Arachis hypogae* L. Collections were made ca. 24-h prior to infestations on cotton. Individual adult brown stink bugs were placed in nylon #280 mesh cages and transported to the field in a chilled cooler to eliminate mortality from heat stress.

Infestation on Pre-Flowering Cotton Plants

The experimental design for infestation studies on cotton seedlings (2 to 3 main stem nodes above the cotyledon), cotton with a match-head square (7 to 8 main stem nodes above the cotyledon), and large squares (>0.8 cm diameter, 8 to 9 d old squares on the first node of a fruiting branch from the main stem of the plant) was a randomized block, with infestation dates as blocks. Similar plants within each block (date) were paired and were randomly infested or non-infested. Non-infested plants also had mesh cages placed over the structure infested. All variables measured were analyzed using a paired t-test by comparing infested and non-infested plants (α= 0.05) (PROC TTEST, SAS Institute 1998). Data for 2001 and 2002 were pooled for analysis.

On cotton seedlings, one adult brown stink bug per mesh cage (15 cm x 15 cm) was placed on a plant terminal. The opening of the cage was tightly closed around the main stem of the plant with a drawstring. Brown stink bugs were caged on each plant for 7 days after which time the cages and insects were removed. At 7, 14, and 21 d after infestation, plants were observed for aborted terminals and main stem height (cm) was recorded. Height on an individual plant was measured from the soil surface to the top of the terminal. At 21 d after infestation, square retention (total number of squares/total number of fruiting sites) and the total number of main stem nodes per plant was recorded. Individual plant heights and number of nodes were used to calculate height to node ratios (HNR) [plant height (cm) / number of nodes)].

On cotton with a small (match-head) square, infestations were similar to cotton seedling infestations, except that mesh cages measured 17.5 cm x 16 cm and the duration of infestations was 5 d. Each plant terminal that was caged contained one match-head square. Square retention for each plant was measured at 5, 12, and 19 DAI. The number of days from planting for each plant to form a flower and the growth stage corresponding to that date (total number of main stem nodes above the cotyledon) was determined.

Individual large squares on cotton plants (9 to 15 nodes above the cotyledon) were infested with one brown stink bug per mesh cage (15 cm x 15 cm). The opening of the cage was tightly closed around the peduncle of the square with a drawstring. The duration of infestations was 5 d. At 9 d after mesh cages were removed (14 DAI) from infested and non-infested plants, the following was recorded: 1) square abscission, 2) occurrence of a white flower with necrotic anthers, 3) occurrence of a boll, and 4) boll abscission.

Infestations on Flowering Cotton Plants

Cotton plants were monitored bi-weekly until the first week of flowering. First position white flowers (flower located on the first fruiting node of a fruiting branch from the main stem of the plant) were marked with a yellow "snap-on-tag" (A.M. Leonard, Inc. Piqua, Ohio) placed on the fruiting branch between the peduncle of the flower and the main stem of the plant. The date of anthesis was recorded on the tag in permanent ink to ascertain the age of the boll at the time of infestation. Boll age was calculated using heat unit (HU) accumulation beginning at anthesis, as described by Bagwell and Tugwell (1992). HU were calculated as:

$$\frac{\text{(Maximum daily temperature + Minimum daily temperature)}}{2} (-)\ 60 = \text{HU}$$

Infestation procedures were similar to that used by Adamczyk et al. (1998) and Russell (1999) for caging lepidopterous larvae and tarnished plant bugs, respectively, on cotton bolls. One brown stink bug adult per nylon mesh cage was placed on an individual boll using the same procedures for large square infestations. For each boll infested, a blue “snap-on-tag” was also placed in the same position as the yellow “snap-on-tag” and labeled with the date of infestation and the date of white flower. Non-infested treatments consisted of cages placed on bolls, with blue “snap-on-tags” placed on the fruiting branch labeled as the control with the date of infestation and date of white flower included. For each date infested, equal number of infested and non-infested bolls were used. Stink bugs were caged on each boll for 72 h, at which time the cages and insects were removed.

In 2002 only, at 72 HAI, the diameter of each boll was recorded using a dial caliper (Forestry Suppliers, Inc., Jackson, MS). Individual boll measurements were taken at the widest diameter using two diametrically opposite points. Boll diameter was also measured for bolls without mesh cages (controls) using the same procedure as described above. A total of 90 bolls were measured for approximately 25 days, each at intervals of two to three days.

The number of abscised bolls was recorded at 72 hours after infestation (HAI) and at harvest. Stink bug infestations were initiated at white flower (0 HU) and continued through 892 HU beyond anthesis. All harvestable bolls were individually collected and seedcotton weights were recorded. Cumulative abscission data and weights of individual bolls within the same HU were then grouped into 17 classes of 50 HU, ranging from 0 - 50 to 851-900.

The experimental design for the boll infestation studies was a completely randomized design. Boll size and seedcotton yield were analyzed using a paired t-test by comparing diameter and weight, respectively, of infested bolls to those of non-infested bolls within the same age class (PROC TTEST, SAS Institute 1998). Within infested, non-infested, and control treatments, boll diameter data was analyzed using regression analysis (PROC REG, SAS Institute 1998) by plotting diameter (dependent variable) against HU accumulated (independent variable) of that same boll (HU accumulated for infested and non-infested bolls was that on the day mesh cages were removed). Boll abscission data for infested bolls was corrected for natural abscission in the non-infested bolls using Abbott’s formula (Abbott 1925) and analyzed using regression analysis (PROC REG, SAS Institute 1998). Corrected abscission (dependent variable) data was plotted against accumulated heat units (independent variable) of infested bolls to determine regression equations describing the relationship. The analytical model included only those HU in which abscission occurred. For all variables measured, data for 2001 and 2002 were pooled for analysis.

Results

Cotton Seedlings (Pre-Squaring)

There were no significant differences in plant height on 7 [df = 146, t =0.09, P = 0.9287], 14 [df = 146, t = 0.74, P = 0.458], and 21 [df = 146, t = 1.13, P = 0.2597] DAI between cotton seedlings that were infested with brown stink bugs or non-infested (Table 1). Additionally, there were no differences in HNR [df = 146, t = 0.4, P = 0.6925] and square retention [df = 146, t = 1.26, P = 0.2081] between infested and non-infested plants at 21 DAI (Table 1).

Cotton With a Small (Match-Head) Square

Square retention on cotton with match-head squares on 5 [df = 138, t = 0.73, P = 0.4691], 12 [df = 138, t = 0.78, P = 0.435], and 19 [df = 138, t = 1.13, P = 0.2621] DAI was not significantly different between infested and non-infested plants [$F(2,406)$ = 0.03, P = 0.9663] (Table 2). The number of days after planting in which a flower first appeared was not significantly different between infested and non-infested plants [df = 138, t = 0.68, P = 0.5003] (Table 2). Additionally, the plant growth stage (number of main stem nodes above the cotyledon) during which that flower appeared was not significantly different between treatments [df = 138, t = 0.07, P = 0.9434] (Table 2).

Large (Pre-Candle) Squares on Cotton Plants

Square abscission between infested and non-infested plants was not significantly different [df = 18, t = 0.97, P = 0.3457] (Table 3). Squares that did not abscise within each treatment became a white flower. On infested and non-infested plants, 70.9 and 73.5%, respectively, of the white flowers were produced from squares on the day mesh cages were removed or on subsequent days. In contrast, 29.0% and 27.5% of white flowers on infested and non-infested plants, respectively, were produced from squares on 2, 3, and 4 DAI. Flowers produced on these days were bolls on the day mesh cages were removed. White flowers were examined for necrotic anthers and no significant differences [df = 18, t = 1.86, P = 0.0917] between infested and non-infested plants were observed (Table 3). Fruiting forms were examined until 14 DAI. There was no significant difference in boll abscission between infested and non-infested plants [df = 18, t = 1.40, P = 0.1771] (Table 4). Boll abscission in infested and non-infested plants was 13.6 and 6.4%, respectively. Although not significantly different, the larger proportion of boll abscission in infested plants was due to the larger proportion of bolls that were present on the day mesh cages were removed (Table 4). Therefore, stink bugs were given the opportunity to feed on small bolls and induce abscission.

Bolls on Cotton Plants

There was a negative linear relationship describing cumulative abscission of infested bolls ($F = 7.09$; df= 1,5; $P < 0.0448$) as a function of heat unit accumulation (Figure 1). Boll abscission ranged from 50.9% for bolls infested at 51 to 100 HU to 0% for bolls infested at ≥ 351 HU (14.0 d) beyond the date of anthesis.

Seedcotton yields for bolls infested with brown stink bugs were significantly reduced ($P < 0.05$) for age classes infested from 0 through 550 HU (22.0 d) beyond anthesis (Figure 2). No significant reductions in yield were observed for bolls that had been infested when they had accumulated > 551 HU (22.0 d) beyond anthesis. Although no stink bug-induced abscission occurred on bolls that accumulated ≥ 351 HU (14.0 d) beyond the date of anthesis, seedcotton yield was reduced. Significant yield losses were observed for bolls infested after they had accumulated 351 to 550 HU ($P < 0.05$).

In 2002, 32 heat units, ranging from 0 to 823.5, were infested with adult brown stink bug. There was a significant quadratic relationship between the age (heat unit) of an individual boll on the day mesh bags were removed and the corresponding diameter (cm) for that same boll when infested [diameter = $0.529 + 9.49\text{x}10^{-3}\text{HU} - 7.5\text{x}10^{-6}\text{HU}^2$; $P<0.0001$, $r^2=0.87$] or non-infested [diameter = $0.816 + 8.79\text{x}10^{-3}\text{HU} - 7.06\text{x}10^{-6}\text{HU}^2$; $P<0.0001$, $r^2=0.87$] (Figure 3). There was also a significant quadratic relationship between age and diameter for control bolls [diameter = $0.06 + 1.33\text{x}10^{-2}\text{ HU} - 1.34\text{x}10^{-5}\text{HU}^2$, $P<0.0001$, $r^2 = 0.94$]. Diameter of bolls infested with brown stink bug was significantly lower compared to non-infested bolls for 12 of the 14 boll age classes infested from 0 to 266.5 HU ($P<0.05$) (Figure 4).

Discussion

Results from field studies demonstrated that infestations of brown stink bugs on cotton seedlings (pre-squaring), cotton with a small (match-head) square, and large (pre-candle) squares do not negatively affect growth and development of cotton, and production of bolls from squares. Brown stink bugs begin to injure cotton when bolls are present. Brown stink bugs induced abscission in bolls that accumulated ≤ 350 HU (ca. 14 d) beyond anthesis. Seedcotton yields, in bolls ranging in age from 0 to 550 HU beyond anthesis, were significantly reduced as compared to non-infested bolls. Boll growth, as measured by diameter, was significantly reduced for bolls infested with brown stink bugs through 266.5 HU (ca. 10 to 11 d) beyond anthesis. Based on results from these studies, using brown stink bugs at an infestation density of one bug per structure, it can be concluded that control measures should be initiated at the time plants begin to set bolls. No yield loss from brown stink bug feeding should occur when a boll has accumulated ≥ 550 HU (ca. 22 d) beyond anthesis.

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Table 1. Response of cotton seedlings (pre-squaring) to infestations of adult brown stink bug for 7 days.

		Height (cm)			21 DAI	
	n	**7**[1]	**14**[1]	**21**[1]	**HNR**[2]	**Square Retention (%)**
Infested	74	23.4	32.4	46.1	4.89	73.8
Non-infested	74	23.5	31.6	45.0	4.85	68.2
P-Value		0.9287	0.4580	0.2597	0.6925	0.2081

[1]Days After Infestation (DAI).
[2]Height to node ratio [plant height (cm)/number of main-stem nodes].

Table 2. Response of cotton seedlings with a match-head square to infestations of adult brown stink bug for 5 days.

		Square Retention (%)			Presence of First Flower	
	n	**5**[1]	**12**[1]	**19**[1]	**Days After Planting**	**Growth Stage (Node)**
Infested	70	88.6	69.7	62.4	63.6	11.13
Non-infested	70	90.7	71.8	65.3	64.0	11.11
P-Value		0.4691	0.435	0.2621	0.5003	0.9434

[1]Days After Infestation.

Table 3. Response of large squares to infestations of adult brown stink bug for 5 days.

			Presence of White Flowers (%)[1]						
			Days After Infestation						
	n	**Square Abscission (%)**	**2**[2]	**3**[2]	**4**[2]	**5**	**After Cage Removal**	**Total**	**Presence of necrotic anthers (%)**
Infested	154	3.0	2.0	8.1	18.9	27.7	43.2	100.0	4.2
Non-infested	154	1.5	1.3	5.3	19.9	24.5	49.0	100.0	0.5
P-Value		0.3457							0.0917

[1]Presence of those squares not shed.
[2]Boll on the day of cage removal.

Table 4. Response of bolls to infestations of adult brown stink bug on large squares for 5 days.

	Total Boll Abscission (%)[1]	Abscission (%) By Fruiting Form on 5 DAI[2]	
		Square or White Flower	Boll
Infested	13.6	24.3	45.7
Non-infested	6.4	26.0	24.0
P-Value	0.1771	0.9259	0.2764

[1]Percentage of infested squares that abscised by 14 DAI.
[2]Days after infestation.

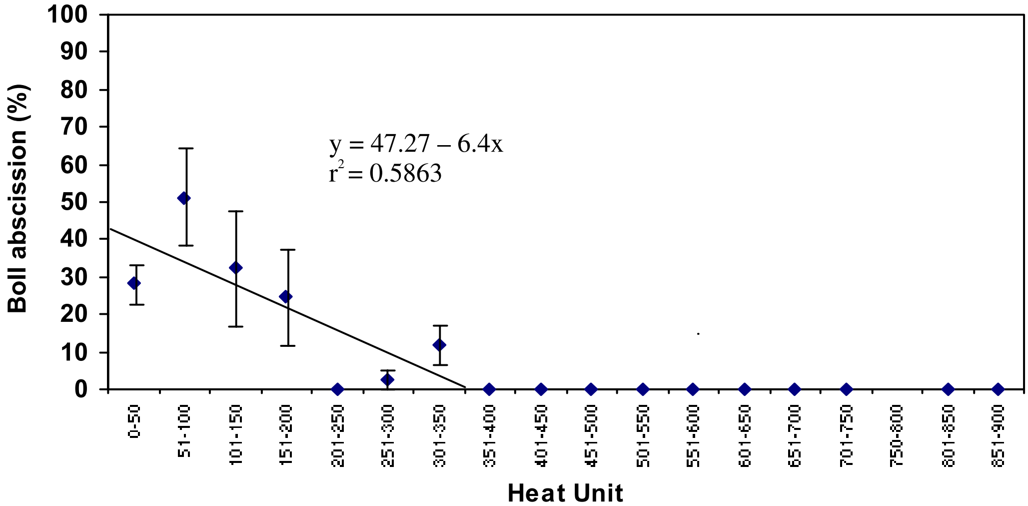

Figure 1. Relationship between boll abscission by brown stink bug and accumulated heat units after anthesis, 2001 and 2002.

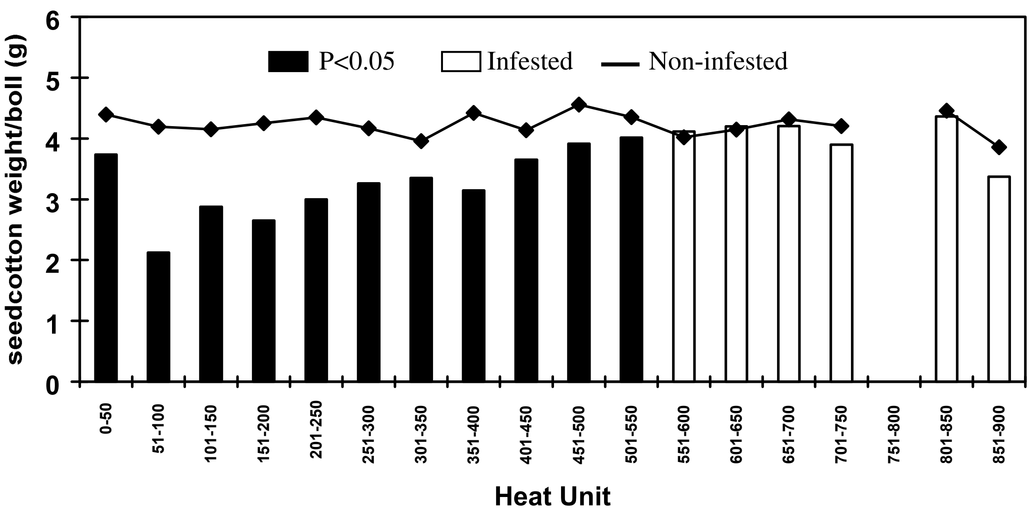

Figure 2. Relationship of heat unit accumulation and brown stink bug injury on seedcotton yield of individual bolls (bars represent mean weights of infested bolls and the line represents mean weights of non-infested bolls; dark bars represent weights of infested bolls significantly [α=0.05] different from non-infested boll weights [P<0.05]), 2001 and 2002.

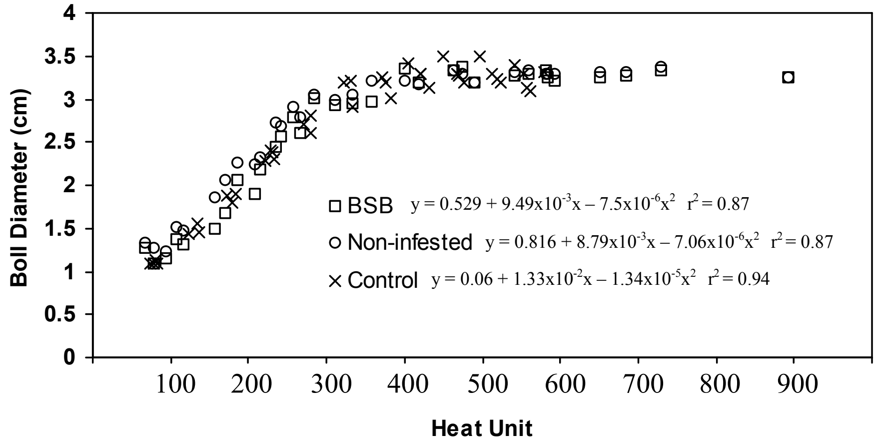

Figure 3. Quadratic relationship of heat unit accumulation and boll growth of individual bolls infested with brown stink bug, non-infested, and control (without cages) (P < 0.0001), 2002.

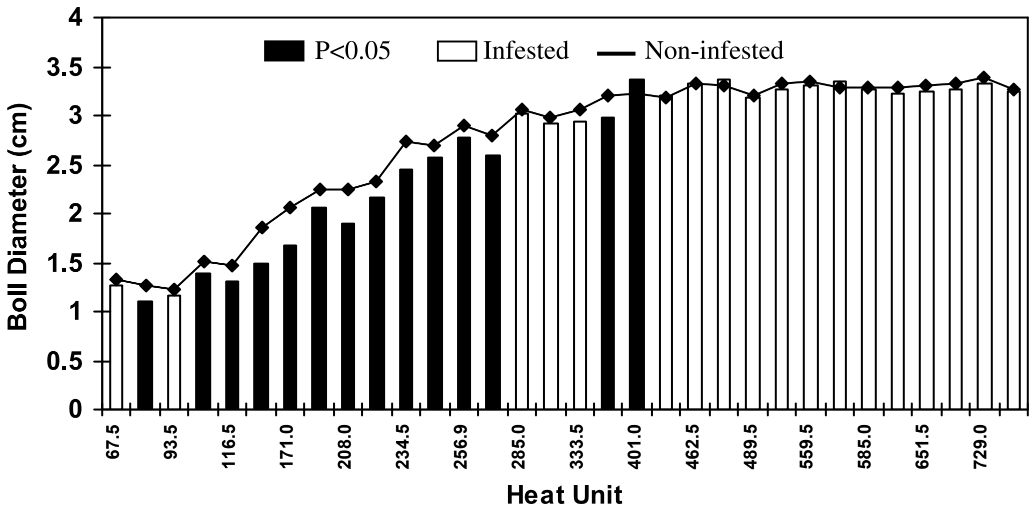

Figure 4. Relationship of heat unit accumulation and brown stink bug injury on boll diameter of individual bolls (bars represent mean diameters of infested bolls and the line represents mean diameters of non-infested bolls; dark bars represent diameters of infested bolls significantly [α=0.05] different from non-infested boll diameters [P<0.05]), 2002.