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Original article (Orijinal araştırma)

Investigation of the damage of Miridae species on cotton in Çukurova Region of Turkey

Çukurova Bölgesi (Türkiye)'nde Miridae türlerinin pamukta meydana getirdikleri zararın araştırılması

Okan ÖZGÜR[1*] **Ekrem ATAKAN[2]** **Serkan PEHLİVAN[2]**

Abstract

Damage of the Miridae species on cotton was determined in cotton fields in two locations of Adana Province (Çukurova Region), Turkey during the years, 2013-2014. Experiments were conducted over 2 years with sprayed and unsprayed plots. *Creontiades pallidus* (Rambur, 1839) and *Lygus italicus* Wagner, 1950 were detected at both experimental areas. Also, the correlations between the mirid population and stained bolls have been investigated. The damage caused by the mirids on squares and bolls was measured in caged branch experiments. Mirid population increased from mid-July reaching the highest population density in late July or early August. Where mirid numbers were high, stained bolls and shed bolls were also high. As the number of *C. pallidus* nymphs released into the cages increased, the damage rates increased on the squares after 7 and 14 d. In cages with zero, one, two and four individuals released, damage rates in squares were 0, 0.9, 5.9 and 33.6%, respectively, after 7 d, and 0, 22.2, 41.4 and 58.0%, respectively, after 14 d. In addition, all bolls with a diameter of 0.5-0.9 cm after 7 d in cages with nymphs released were damaged.

Keywords: Boll, cotton, damage, Miridae*,* square

Öz

Adana İli (Çukurova Bölgesi), Türkiye'de pamuk tarlalarında 2013 ve 2014 yıllarında iki farklı bölgede yürütülen çalışmada Miridae türlerinin pamukta meydana getirdikleri zararlar tespit edilmiştir. Denemeler iki farklı lokasyonda ilaçlı ve ilaçsız parseller şeklinde 2 yıl üst üste kurulmuştur. Deneme yapılan 2 tarlada hem *Creontiades pallidus* (Rambur, 1839) hem de *Lygus italicus* Wagner, 1950 türü bulunmuştur. Ayrıca, mirid popülasyonu ile lekeli kozalar arasındaki ilişki incelenmiştir. Miridlerin tarak ve kozalarda oluşturduğu zarar dal-kafes denemeleriyle ortaya konulmuştur. Mirid popülasyonu temmuz ayı ortasından sonra artmış, temmuz sonu veya ağustos başında en yüksek noktaya ulaşmıştır. Mirid popülasyonu yüksek olduğunda, lekeli koza sayısı ve yere dökülen kozaların sayısı da yüksekti. *C. pallidus*'un kafeslere salınan nimf sayıları arttıkça 7 ve 14 gün sonunda taraklarda verdikleri zarar oranlarının da arttığı görülmüştür. Sıfır, bir, iki ve dört birey salınan kafeslerde taraklarda zarar oranları 7 gün sonra sırasıyla %0, 0.9, 5.9 ve 33,6 bulunmuşken, 14 gün sonra %0, 22,2, 41,4 ve 58,0 bulunmuştur. Ayrıca kozalardaki zarar oranı, 0.5-0.9 cm çaplı kozalarda 7 gün sonra kafeslerde tüm bireyler için %100 bulunmuştur.

Anahtar sözcükler: Koza, pamuk, zarar, Miridae*,* tarak

[1] Biological Control Research Institute, 01321, Yüreğir, Adana, Turkey
[2] Çukurova University, Faculty of Agriculture, Department of Plant Protection, 01330, Sarıçam, Adana, Turkey
* Corresponding author (Sorumlu yazar) e-mail: okanozgur80@gmail.com
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Introduction

The Miridae family is the largest family of insects (Hemiptera: Heteroptera) with about 1400 genera containing over 10,000 described species (Schuh, 1995). Miridae species are both pests and predators of some important pests depending on environmental conditions (zoophytophagous species). Miridae species globally cause significant damage in a wide array of hosts such as cotton, clover, soybean, mung bean, strawberry, sorghum, cocoa, apple and tea (Wheeler, 2001). In some parts of the world where transgenic cotton cultivars are grown, the mirids were dominant species due to a reduced number of insecticide sprays against *Helicoverpa armigera* Hübner, 1827 and the increased use of targeted selective insecticides in recent years. Most chemical control is against cotton pests including the mirids in China and the USA. (Wu et al., 2002; Musser et al., 2009). Mirids become important especially for the damage which they give to generative organs in cotton. They suck leaves, shoots, squares (floral buds), flowers and young bolls and cause damage of drying and shedding. *Lygus hesperus* Knight, 1917 (Hemiptera: Miridae) adults and nymphs feeding on small and medium sized squares cause shedding after 3-4 d (Hake et al., 1996). Some studies conducted in the USA found that *L. hesperus* negatively affects yield mostly by feeding on fingers (Jubb & Carruth, 1971; Mauney & Henneberry, 1979; Bailey, 1982; Leight et al., 1988; Zink & Rosenheim, 2005). Karman & Akşit (1961) identified as *Lygus pratensis* Leston, 1957, *Creontiades* spp. and *Creontiades pallidus* (Rambur, 1839) (Hemiptera: Miridae) in cotton fields in Aydın (Turkey). In addition, Efil & İlkan (2003) and Efil & Bayram (2009) reported that *C. pallidus* causes shedding of young bolls of the cotton in the southeastern Anatolia (Turkey). While there are many reasons for shedding of squares and bolls, this shedding can reach significant levels at times when the mirids are high density. Efil & İlkan (2003) monitored the population development of *C. pallidus* in cotton fields on the Harran Plain (Şanlıurfa, Turkey). During a 3-year study, the pest population reached the peak density during the last 3 weeks of August and the first week of September.

Çukurova is one of the important production areas for cotton. In the region, farmer concerns have increased in recent years due to damage caused by mirids, as well as the main pests thrips, cotton aphids, leafhoppers and whiteflies. Despite concerns about the increase of mirid damage in the cultivated area of cotton in Turkey, studies on this pest have been quite limited. This study aimed to reveal some important features of pest control. For this purpose, experiments were conducted in Çukurova in order to determine the population fluctuations of pest mirids in cotton fields in two locations. In addition to the development of mirid populations, stains on bolls and mirid damage in generative organs were investigated.

Material and Methods

Sampling of harmful mirids

In order to investigate the damage status of the mirids on cotton, sprayed plots (20 rows x 10 m, 142.5 m^2) which were kept at the lowest level of the pest and a unsprayed plot (20 rows x 10 m, 142.5 m^2) which allow a certain population of mirids were laid out with four replicates. Plots were sprayed at intervals of 2 weeks during the season to keep the mirid population at the lowest level (Table 1). Experiments were established in Çukurova University Research and Implementation Area Balcalı, and Research Area of the Eastern Mediterranean Agricultural Research Institute, Hacıali, Adana, Turkey in 2013 and 2014.

Two sampling methods were used to determine the chemical control of the mirids according to the Integrated Management Technical Instructions; (i) by direct counting of generative organs (economic threshold: four mirids per 100 generative organs after square formation, 20 mirids after 80% of bolls begin to mature), and (ii) by sweep net (economic threshold: seven mirids per 50 sweep net after square formation, 30 mirids after 80% of bolls beginning to be mature) (TOB, 2017). The population fluctuations of harmful mirids were recorded weekly in the mornings (8-10 am) by both methods from the 2-3 leaf stages until harvest (in Hacıali from 2 July to 17 September 2013 and 18 June to 10 September 2014, and in Balcalı from 3 July to 18 September 2013 and 12 to 4 September 2014). For sweep netting, the net was swept 25 times in four subplots to a total of 100 times for each insecticide-sprayed and unsprayed plot. Every 25 sweep samples were transferred to fabric bags (30 x 40 cm) and labeled.

Table 1. Insecticides which were applied for the mirids in experiment areas

Hacıali		Balcalı	
2013	2014	2013	2014
Dimethoate 1 l/ha (9 July)	Thiacloprid & Deltamethrin 0.8 l/ha(3 July)	Dimethoate 1 l/ha (4 July)	Thiacloprid & Deltamethrin 0.5 l/ha (19 June)
Dimethoate 1 l/ha (23 July)	Thiacloprid & Deltamethrin 0.8 l/ha (18 July)	Dimethoate 1 l/ha (19 July)	Thiacloprid & Deltamethrin 0.8 l/ha (5 July)
Thiacloprid & Deltamethrin 0.5 l/ha (6 August)	Thiacloprid & Deltamethrin (0.8 l/ha) Cypermethrin (0.3 l/ha)	Thiacloprid & Deltamethrin 0.5 l/ha (5 August)	Thiacloprid & Deltamethrin (0.8 l/ha) Cypermethrin (0.6 l/ha)
Thiacloprid & Deltamethrin 0.5 l/ha (21 August)	Thiacloprid & Deltamethrin (0.8 l/ha) Cypermethrin (0.3 l/ha)	Thiacloprid & Deltamethrin 0.5 l/ha (16 August)	Dimethoate 1 l/ha (22 July)
Thiacloprid & Deltamethrin 0.5 l/ha (12 September)	Thiacloprid & Deltamethrin 0.8 l/ha (21 August)	Thiacloprid & Deltamethrin 0.5 l/ha (6 September)	Thiacloprid & Deltamethrin (0.8 l/ha) Cypermethrin (0.3 l/ha)
			Thiacloprid & Deltamethrin (0.8 l/ha) Cypermethrin (0.3 l/ha)

For direct counting, 25 squares, 25 flowers (10 flowers during the periods when the numbers are decreasing; in Hacıali from 23 July to 10 September 2013 and 31 July to 20 August 2014, and in Balcalı from 24 July to 11 September 2013 and 24 July to 14 August 2014), 25 bolls (mostly young green bolls) and 25 opened bolls were randomly sampled in each insecticide-sprayed and unsprayed plot. Nymphs and adults of the two Miridae species were easily distinguished. Nymphs and adults of *Lygus italicus* Wagner, 1950 have dark spots on the pronotum and scutellum.

Detection of stains on bolls

At the beginning of boll formation, 25 bolls from four subplots of each insecticide-sprayed and unsprayed plot were examined at weekly intervals. The stains caused by the mirid feeding were counted and recorded.

Determination of boll shedding

In each unsprayed subplot, 3 rows of cotton 3-m long were selected for sampling and marked. On each sampling date, shed bolls were collected from the ground below the plants. Shed bolls were cut to record color changes in anthers, damage to anther sac, color changes on external surfaces of squares, color changes in fruit tissue, and stain density on the boll surface. If there was no insect damage evident, the shedding of these bolls was assumed to have been physiological.

Determination of *Creontiades pallidus* damage in caged branches

Rearing of insects

Creontiades pallidus was collected by sweep net in a clover field (cultivar Elçi) in Balcalı (37º02'16.7" N, 35º22'12.5" E). *Creontiades pallidus* was the common species and regularly found in alfalfa fields. Under laboratory conditions (climatic chamber at 25±1ºC, 60% RH, 16:8 h L:D photoperiod), they were fed on green fresh beans and laid eggs on these beans. Hatched nymphs were fed on the fresh green beans obtained from the local market.

Determination of the damage on squares

Two cage experiments were conducted in 2014 in Balcalı. In the first experiment, two healthy squares selected from the upper half of the plant, were placed in a cage with dimensions of 47 x 27.5 cm (49 holes/cm^2, i.e., 10 mesh). The branches were cleared of other insects before Miridae insects were added to cages. The plant and the cages were labeled. Squares larger than 0.3 cm in diameter were selected for these cages. Fruiting branches were selected on the same position of the plants. Then, fourth stage nymphs of *C. pallidus* were released as one, two and four individuals into each cage on 25 June 2014. Also, the control cages were established with no nymphs released. Seven d after the releasing of the nymphs, fruit branches were cut and taken to laboratory. Squares were examined under the stereomicroscope with X45 magnifications (1 July 2014). The experiment was replicated 11 times.

A second experiment was established on 26 June 2014 as above but with 10 replicates. Fourteen d after nymph release, fruit branches were taken to laboratory and examined (10 July 2014). Assessments were made as in the first experiment; the damage symptom was investigated. The squares were examined individually. Damage indications in squares such as discoloration at the anther, softening and deformation in the tissues, shrinkage, dryness or shedding were examined and evaluated as observable percentage loss.

Determination of the damage on bolls

Damage indications in the squares and the bolls such as discoloration at the anther, softening and deformation in the tissues, shrinkage, dryness or shedding were examined and evaluated as observable percentage loss (Figure 1).

Figure 1. Some damage views caused by the Miridae species in bolls and squares: a) stains on the boll, b) rots in the boll tissue, c) desiccation of the boll, d) rots in the square tissue, and e) desiccation of the square.

Cage experiments for boll damage were conducted on two dates in Balcalı and Hacıali locations (Adana Province) in 2014. First experiment was established in Balcalı on 14 July 2014. One healthy boll larger than 1 cm in diameter on the upper half of the plant, were placed into cages, and the experiment conducted as for the caged squares as described above. Seven d after releasing the nymphs, fruit branches were taken to laboratory and they were examined (21 June 2014) for Miridae damage. Diameters of the bolls were measured by caliper. The experiment had 20 replicates. A second experiment was established in Hacıali on 17 July, healthy bolls larger than 0.5 cm in diameter caged and again treated as above. Seven d after releasing the nymphs, fruit branches were taken to laboratory and examined (24 June 2014). The second experiment had five replicates. The bolls were examined individually under the stereomicroscope with X45 magnifications and damage assessed as described above.

Statistical analysis

Relationships between the Miridae population, and stained and shed bolls for both sampling methods were analyzed. The average number of stains on bolls in unsprayed and sprayed plots were compared by simple t-test. Differences in damage rates of *C. pallidus* on the squares and bolls in the cage experiments were evaluated by analysis of variance (one-way ANOVA). Differences between means were grouped by Duncan's test ($P < 0.05$).

Results and Discussion

Relationship between mirid populations and number of stains on bolls

Both *C. pallidus* and *L. italicus* species were present in the experimental fields. The apparent damage of the Miridae species on the bolls was often dark round, slightly precipitated stains, which resulted from sucking.

In 2013 in Hacıali, Miridae populations reached the highest number on generative organs on 6 August (14.5 mirids/100 generative organs) in unsprayed plots. When mirid population peaked, the number of stains due to mirid feeding increased. There was a significant and positive relationship between Miridae population density and number of stains on bolls (r^2 = 0.72, y = 0.522x + 1.32) (Figure 2).

On the generative organs in sprayed plots, the mirid population reached the highest density on 23 July (8.17 mirids/100 generative organs). However, the number of stains was highest on 6 August. So, there was no close relationship between the mirid population and stain density on the bolls (r^2 = 0.27, y = 0.350x + 0.863) (Figure 2).

In unsprayed plots, mirid population reached the highest number on 30 July (15.5 mirids/sweep net) as determined by sweep net sampling (Figure 2). The number of stains on the bolls also increased following the pest population density. The mirid population generally decreased after 6 August, but had a small increase on 2 September. Due to the short-term increase in the mirid population in September, the number of stains increased a little. However, there was no relationship between mirid population densities and stain densities on the bolls (r^2 = 0.086, y = 0.18x + 2.23).

In the sprayed plots, mirid populations were low during 16 to 30 July, but then reached the highest level with a mean of 17 mirids/sweep net on 6 August (Figure 2). The number of stains on the bolls increased following the increase of the pest population and decreased following the decrease of the pest population. As a result, there was a significant positive relationship (r^2 = 0.51, y = 0.290x + 0.650). The densities of stains on bolls in the sprayed plot were lower than found in unsprayed plots.

The mirid population exceeded the economic threshold for the two sampling methods in the early period for unsprayed and sprayed plots.

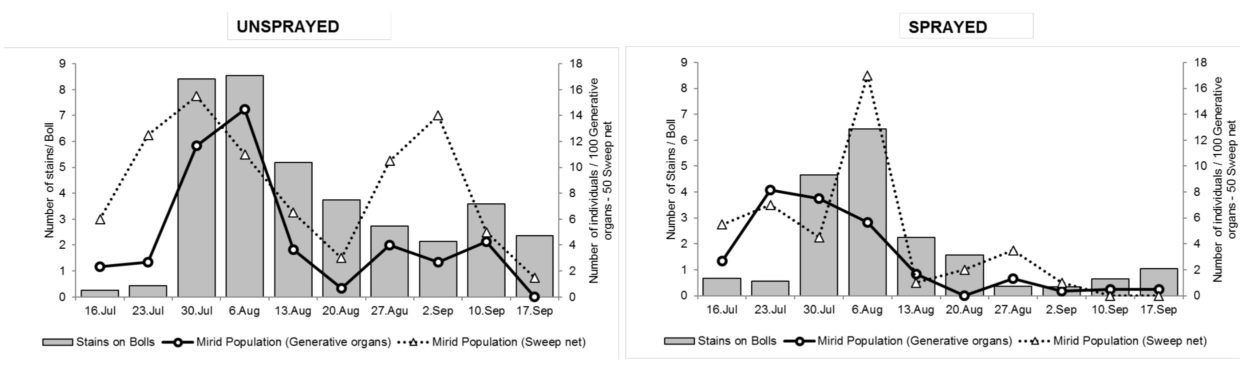

Figure 2. The relationship between numbers of mirids (counted directly and by sweep net) and densities of stains on the bolls in unsprayed and sprayed plots in Hacıali in 2013.

In 2014 in Hacıali, stained bolls appeared after the decrease in the mirid population in unsprayed plots (Figure 3). On 27 August, the number of stains on the bolls increased relatively on the other sampling dates. In other words, the numbers of mirids on generative organs were lower on sampling dates when the dense stains were observed on the bolls. There was no correlation between the number of stains and the number of pests on the bolls (r^2 = 0.04, y = -0.156x + 0.791) (Figure 3).

In the generative organs of sprayed plots, following a slightly increase in the mirid population, number of stained bolls increased on that date. Although the mirid population decreased after 13 August, there was a slight increase in the number of stains on the bolls. No correlation was detected ($r^2 = 0.1$, $y = 0.149x + 0.281$) (Figure 3).

In the unsprayed plots, the number of stains increased in early August and early September following mirid feeding (Figure 3). There was no relationship between the number of stains and the number of individuals ($r^2 = 0.02$, $y = -0.056x + 0.792$).

Mirid population determined by sweep net was low throughout the season in the sprayed pots. The number of stains on bolls had increased by a low rate by 13 August and the following dates. The numbers of mirids counted by both methods were slightly lower than those found in unsprayed plots. The densities of stains were also found to be lower than that of unsprayed plots (Figure 3).

Economic threshold was not exceeded in two sampling methods during the season for unsprayed and sprayed plots.

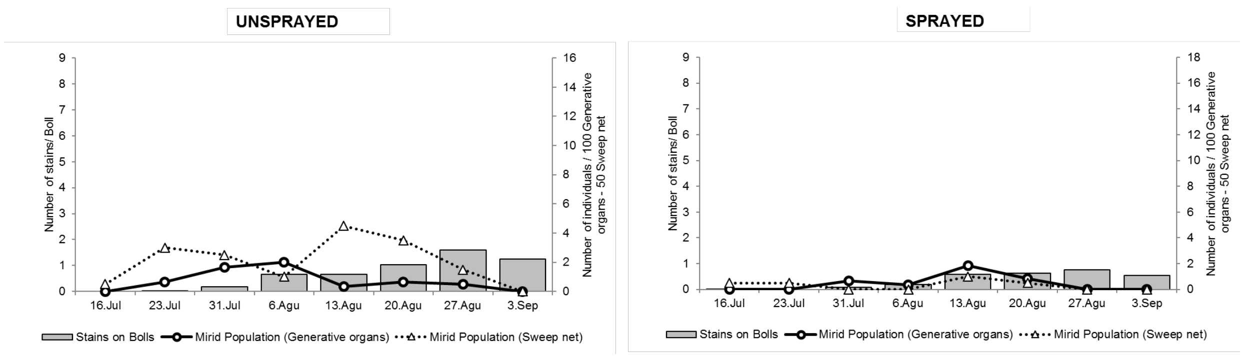

Figure 3. The relationship between numbers of mirids (counted directly and by sweep net) and densities of stains on the bolls in unsprayed and sprayed plots in Hacıali in 2014.

In 2013 in Balcalı, in the unsprayed plots, the number of stains on bolls increased from 31 July to 21 August. During this period, the population density of mirids on the generative organs increased for a short time, but then remained stable. After that date, although the numbers of the pest increased for a short time, the density of stains on bolls decreased. There was no relationship between the number of stains and the number of mirids ($r^2 = 0.13$, $y = -0.354x + 2.34$) (Figure 4).

In sprayed plots, the density of the mirid population on the generative organs remained low throughout the cotton growing season (Figure 4). No correlation was found between the number of mirids and number of stains on bolls during the season ($r^2 = 0.002$, $y = -0.0472x + 1.03$).

For sweep net sampling, the number of stains increased together with the mirid population, which had stable densities throughout the season in the unsprayed plots until 21 August. Although numbers of stains apparently increased according to population increase of pest mirids, no significant relationship was found between the number of stains and the number of pests ($r^2 = 0.16$, $y = 0.262x + 0.913$).

Population density of mirids reached 4 individuals/50 sweep net on 31 July in sprayed plots. Although number of mirids decreased after this date, the number of stains did not decrease (Figure 4). There was no correlation between them ($r^2 = 0.07$, $y = 0.108 + 0.848$) (Figure 4). During the season, relatively higher mirid populations in unsprayed plots than sprayed plots caused higher stain density.

Mirid population did not exceed the economic damage threshold in both sampling methods during the season for unsprayed and sprayed plots.

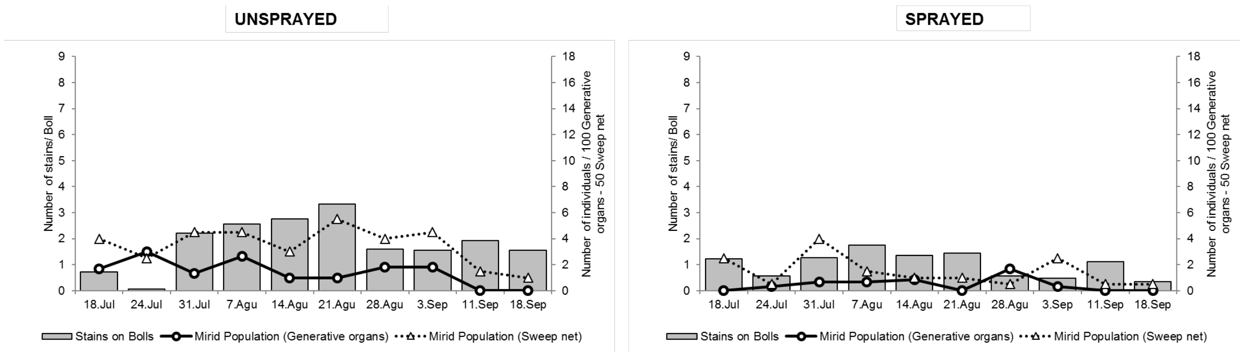

Figure 4. The relationship between numbers of mirids (counted directly and by sweep net) and densities of stains on the bolls in unsprayed and sprayed plots in Balcalı in 2013.

In 2014 in Balcalı, mirid populations on the generative organs in unsprayed plots increased in the last week of July and the first week of August. When the mirid population reached the highest level, on 31 July, the density of stains on the bolls was found to be relatively low. There was no relationship between number of individuals and number of stains ($r^2 = 0.04$, $y = -0.0686x + 1.68$) (Figure 5).

The mirid density on generative organs in sprayed plots peaked on 31July. Similar to the other sampling, numbers of the stains increased after the high mirid population occurred (Figure 5). However, there was no relationship between the number of stains and the number of pest ($r^2 = 0.17$, $y = -0.0672x + 0.782$).

In unsprayed plots, population densities of mirids were similar for direct and sweep net counting as well as by stain sampling of bolls. The pest population increased on 31 July, while the density of stains increased towards the end of August (Figure 5). There is no correlation between number of individuals and number of stains as a result of the analysis ($r^2 = 0.02$, $y = -0.124x + 1.77$).

In the sweep net sampling in sprayed plots, the mirid population remained low throughout the sampling period. There was no relationship between pest and stain density ($r^2 = 0.11$, $y = -0.175x + 0.814$) (Figure 5).

Mirid population reached the economic threshold 31 July on generative organs for the unsprayed plots.

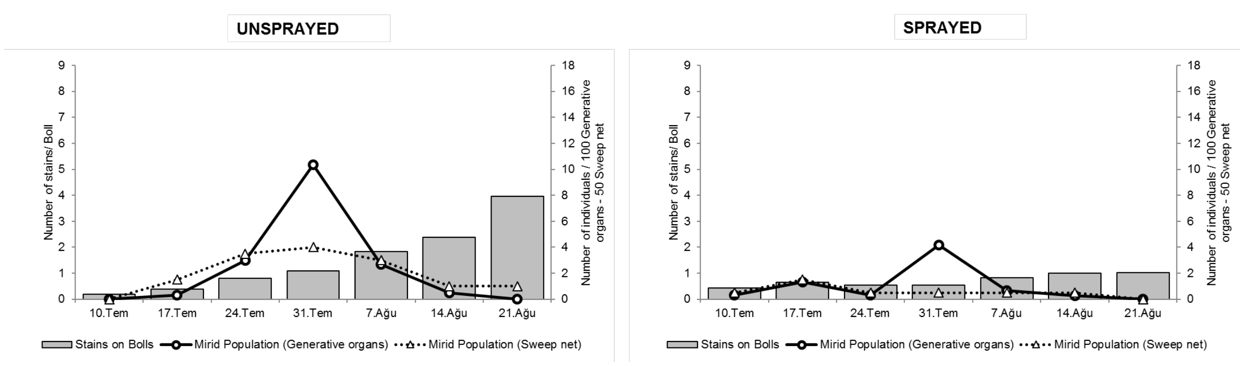

Figure 5. The relationship between numbers of mirids (counted directly and by sweep net) and densities of stains on the bolls in unsprayed and sprayed plots in Balcalı in 2014.

Numbers of stains in unsprayed and sprayed plots

The number of stains on bolls in unsprayed and sprayed plots are given Figure 6. The difference between the number of stains in sprayed and unsprayed plots was statistically significant on 30 July ($t = -3.61$, $P = 0.01$), 6 August ($t = -3.22$, $P = 0.02$), 13 August ($t = -3.25$, $P = 0.02$), 20 August ($t = -7.59$, $P = 0.0002$), 27 August (t = -5.66, P = 0.001), 10 September (t = -4.53, P = 0.004) and 17 September (t = -2.81, P = 0.03). The number of stains was higher in the unsprayed plots (Figure 6).

In unsprayed plots in Hacıali (Figure 6), no stained bolls were found between 16 and 23 July 2014. The differences in the number of stains in unsprayed and sprayed plots were found to be significant on 6 August (t = -4.73, P = 0.003), 27 August (t = -3.24, P = 0.02) and 3 September (t = -3.21, P = 0.02). Both number of stains on bolls and mirid population were lower in 2014 than in 2013.

In Balcalı in 2013, the stain densities on the bolls in unsprayed plots remained quite low level on 18 to 24 July (Figure 6). The numbers of stains in sprayed plots were statistically significant on 14 August (t = -3.61, P = 0.01), 21 August (t = -4.03, P = 0.007), 28 August ($F_{1.6}$ = 12.675, t = -3.56, P = 0.01), 3 September (t = -3.20, P = 0.02), 18 September (t = -4.35, P = 0.01) than unsprayed plots.

In Balcalı in 2014, the stain density on the bolls in unsprayed plots remained quite low level on 10 July. The number of stains increased towards the end of the season and reached the highest level on 21 August. Differences in stain numbers were found to be higher and statistically significant on 31 July (t = -3.01, P = 0.02), 14 August (t =-2.49, P = 0.05) and 21 August (t = -5.30, P = 0.002) for the unsprayed plot. The number of stains on boll in sprayed plot was significancy higher than that of unsprayed plot on 17 July (t = 4.42, P = 0.004).

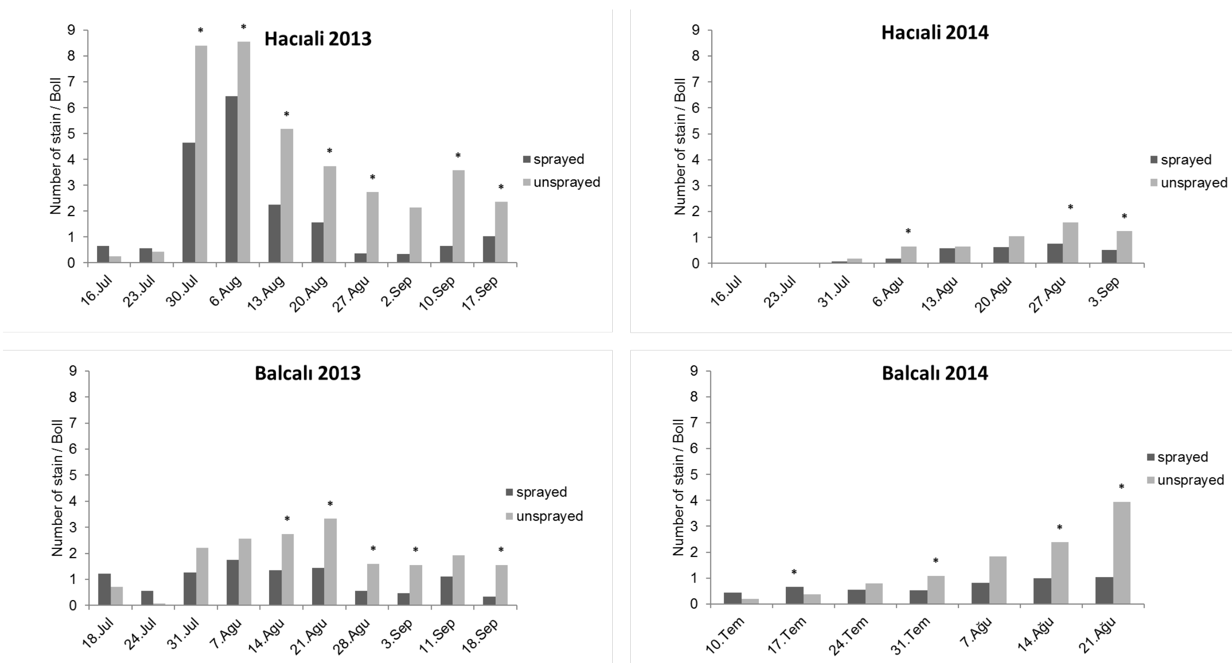

Figure 6. The number of stains in unsprayed and sprayed plots in Hacıali and Balcalı in 2013-2014
*Means on the bars are significant according to t-test (P < 0.05).

Findings in both Hacıali and Balcalı in 2013 and 2014 indicated that harmful mirid populations reached the highest density at the end of July or at the beginning of August. Stains on the bolls were detected after occurrence of mirid populations on plants in both sampling locations. Nakash et al. (1990) reported that puncture of *C. pallidus* is easily detected as a small black shiny spot on small bolls. Efil & İlkan (2003) revealed that population of *C. pallidus* in cotton fields reached to peak point in last week of August and first week of September in Şanlıurfa, Turkey. Some researchers reported that the population of mirids increased in the cotton fields after July in the USA. (Gore et al., 2012; Asiimwe et al., 2014). Khan et al. (2006) stated that during December-January, many of the cotton fields in Australia were in the early boll period and that in this period *Creontiades dilutus* (Stål, 1859) (Hemiptera: Miridae) could lead to significant damage. He reported that feeding generally causes shedding of squares and yield losses.

On a weekly basis, there was no significant relationship between the dynamics of the number of mirids and the stains on the bolls. This may be related to the physiology of the bolls, stage of the nymphs and adults of mirids, the time required for stain formation, the feeding behavior on bolls and the boll preference. However, when the number of mirids and stains were taken into consideration, the number of stains were higher in the years, locations and the plots (sprayed and unsprayed) where the number of mirids was higher. In both locations and years, spraying kept the number of pest low, but it was not able to prevent the occurrence of the mirid damage (stained bolls). Stained bolls were also seen at low levels of the mirid densities.

Relationship between boll shedding and mirid population

Mirid damage was examined in bolls collected from the soil and the bolls were cut to determine the mirid sucking damage. Also, stains on the bolls caused by mirids were noted. In Hacıali, in 2013, the number of the bolls shed on soil was quite low at the beginning of sampling (in July). While number of mirids was the highest on generative organs and by sweep net sampling (30 July and 6 August), number of the bolls shed on soil was the highest. At the end of August and at the beginning of September, the numbers of shed bolls did not increase, although the population of the mirid increased for a short time. The reason for this could be that mature bolls have hardened tissues which may not be suitable for mirid feeding. Although there was a significant relationship between the number of mirids on generative organs and the numbers of shed bolls ($r^2 = 0.84$, $y = 0.294x - 0.352$), the relationship was not high with sweep net sampling ($r^2 = 0.28$, $y = 0.155x - 0.344$) (Figure 7).

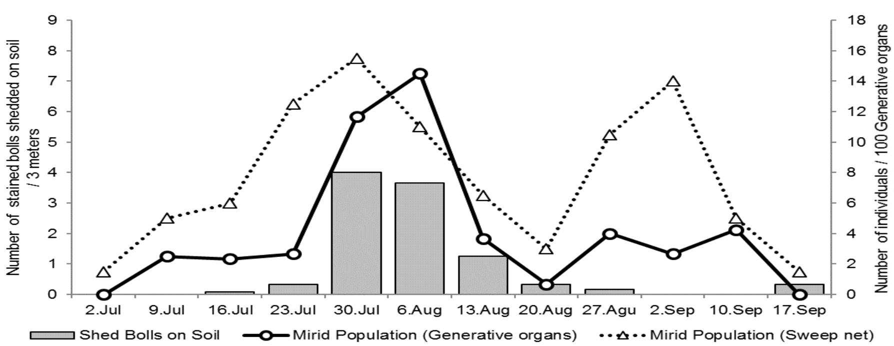

Figure 7. The relationship between the number of mirid (counted directly and by sweep net) and shed bolls on soil in unsprayed plots in Hacıali in 2013.

In 2014, there was an increase at the mirid population on generative organs and sweep net at the end of July and the beginning of August in unsprayed plots in Hacıali (Figure 8). After mirid population densities on bolls slightly increased, shed boll numbers also increased, Similar case was seen at the sweep net sampling. However, there was no relationship to between the number of shed bolls and the number of pests on generative organs ($r^2 = 0.35$, $y = 0.627x + 0.161$) and by sweep net sampling ($r^2 = 0.19$, $y = 0.212x + 0.125$) (Figure 8).

The numbers of stained and shed bolls in 2014 was lower than in 2013. This may be related to the fact that mirid numbers in 2014 were lower than in 2013.

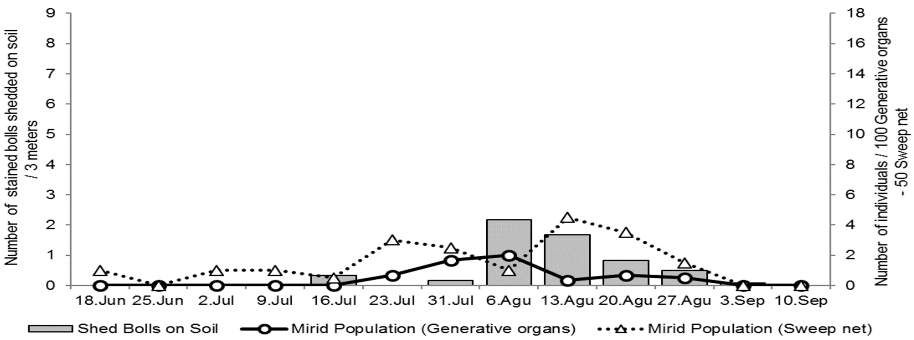

Figure 8. The relationship between the number of mirid (counted directly and by sweep net) and shed bolls on soil in unsprayed plots in Hacıali in 2014.

In Balcalı at the beginning of sampling in 2013, the number of shed bolls was quite low in the unsprayed plots (Figure 9). According to direct and sweep net counting, the mirid population generally showed similar and low population densities on most sampling dates. The number of shed bolls had increased by 31 July and 7 August. At the end of the season, the number of mirids increased for a short time, but no shed bolls that were found. There was no significant relationship between the number of shed bolls and the number of mirids on generative organs (r^2 = 0.13, y = 0.348x + 0.116) and by sweep net sampling (r^2 = 0.12, y = 0.215x - 0.092) (Figure 9).

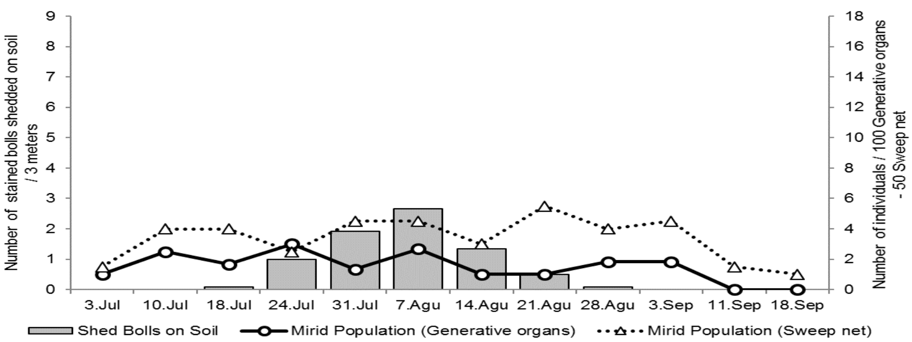

Figure 9. The relationship between the number of mirid (counted directly and by sweep net) and shed bolls on soil in unsprayed plots in Balcalı in 2013.

In Balcalı, in 2014, the mirid population on generative organs and sweep net in unsprayed plots peaked on 31 July (Figure 10). For sweep net sampling, the mirid population was detected in the middle of June and early July, in this period no shed bolls were recorded. The number of bolls was higher in the period when the mirid population started to increase (24 July). After 31 July, the mirid population declined rapidly, and numbers of shed bolls was low during that period (14 August). There was no significant relationship between number of shed bolls and the number of pests on the generative organs (r^2 = 0.24, y = 0.061x + 0.172) and by sweep net sampling (r^2 = 0.22, y = 0.117x + 0.079) (Figure 10).

In both locations, mainly young bolls were shed due to mirid feeding. Shedding of stained bolls were detected mostly in mid-July and mid-August. In this period, the number of young bolls on plants were relatively higher and the number of mirids were also higher. After early or mid-August, even though mirids were present, ratios of shed bolls were low. In other words, mature bolls were not shed even if they were exposed to mirid feeding.

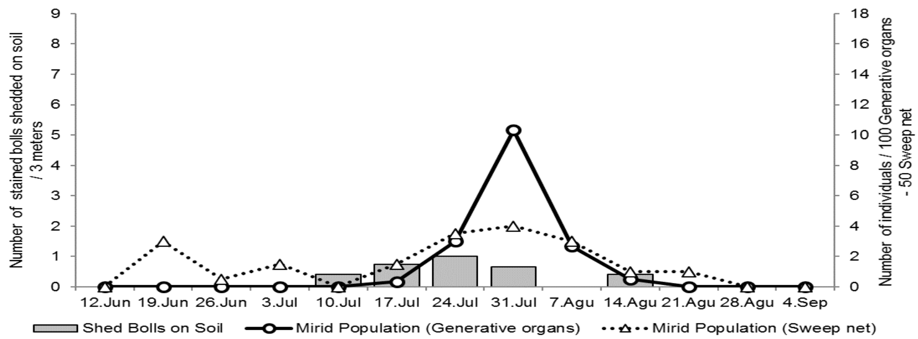

Figure 10. The relationship between the number of mirid (counted directly and by sweep net) and shed bolls on soil in unsprayed plots in Balcalı in 2014.

Pack & Tugwell (1976) reported that *L. lineolaris* does not always cause shedding of older squares, but leads to damage in developing flower anthers. Researchers noted that feeding damage, which did not lead to shedding of larger squares, usually resulted in abnormal flower formation. If the damage to the anthers is severe, pollination does not occur and this leads to the development of small abnormal bolls. These bolls are usually shed a few days after pollination. Nakash et al. (1990) reported that the puncturing of squares by *C. pallidus* caused a rapid deterioration of the flowers and caused them to shed later. They also reported that small bolls were sensitive to shedding as quickly as flowers. Mature and larger bolls were not shed due to *C. pallidus* infestations. Layton (2000) noted that feeding on the young bolls generally causes shedding and yield loss. Rosenheim et al. (2006) noted that *Lygus* feeds on squares and bolls. Mirids damage anthers and seeds then causes shedding of squares and bolls. Researchers noted that with *Lygus* feeding, the damage that occurs in cotton can vary considerably. Square and boll losses are often inconsistent with *Lygus* population estimates. Armstrong et al. (2010) noted that smaller bolls are more susceptible to feeding of *Creontiades signatus* (Distant, 1884) but damaged more than larger bolls. According to researchers, *C. signatus* seeks developing embryos and feeds actively on them. In the present study, nymphs and adults were recorded on the squares but few squares were found to have shed. It was thought that squares could have been shed for physiological reasons given that no significant feeding damage was observed on the squares. The reason for this may be the increase in the number of bolls during the periods of high mirid population and the fact that these mirid species prefer fresh bolls rather than squares. There was no mirid damage to flowers on which most adults were encountered. No linear relationships were recorded between weekly dynamics of the mirid population and the number of shed bolls during the season. However, it is clear that the number of shed bolls were higher in the years, locations and the plots (sprayed and unsprayed) where the number of mirids was higher.

Damage to squares

Seven d after the release of *C. pallidus* nymphs into the cages, less feeding damage was seen in cage with one individual (0.9%) and two individuals (5.9%) than in cage with four individuals (33.6%) on squares. There was no statistically significant difference between cages with no nymphs (control), and one and two individuals in the occurrence of damage. Whereas, feeding damage is greater in cages with four individuals and damage rate was statistically significant ($F_{3.84}$ = 9.53, P = 0.0001). There was no damage to squares in cages with no nymphs released. Figure 11 shows the damage to squares 14 d after release of *C. pallidus* nymphs into the cages. Accordingly, no damage was observed in the cages with no nymphs released. When the number of individuals that released into the cages increased, damage ratios of squares also increased. The highest losses (58.0%) were seen in cage with four individuals ($F_{3.72}$ = 7.66, P = 0.0001). As the number of nymphs which released into the cages increased, the damage rates also increased after 7 and 14 d.

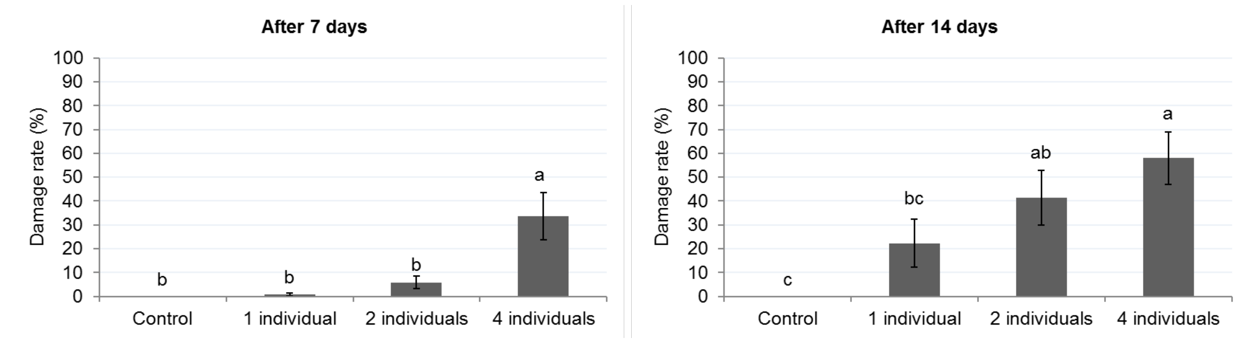

Figure 11. Damage (%) caused by *Creontiades pallidus* nymphs in squares (0.3-0.5 cm in diameter) in branch cages after 7 and 14 d in Balcalı in 20141

*The mean numbers with same letter on bars are not significant according to the Duncan multiple range test (P > 0.05).

Stam (1987) noted, the developing anthers in the squares were dark brown and narrowed in Syrian cotton fields. He indicated that damaged squares and bolls were shed and there was a significant relationship between the *C. pallidus* population and the damaged squares. Leight et al. (1988) reported a positive relationship between the density of *L. hesperus* and the number of shed squares in their study in San Joaquin Valley, CA, USA.

Damage to bolls

Seven d after the release of the *C. pallidus* nymphs to the bolls with 0.5-0.9 cm in diameter, no damage was seen in the control cages. However, all of the bolls in the cages with one, two, and four individuals all bolls were damaged. Seven d after the release of the *C. pallidus* nymphs to the bolls with a 1-1.5 cm in diameter, damage rates in the cages with two individuals (68.4%) and four individuals (70.0%) were similar but more than the bolls infested with 0 (5.26%) and one individual (36.8%). The difference for boll damage among the treatments was statistically significant ($F_{3.71} = 9.83$, $P = 0.0001$) (Figure 12).

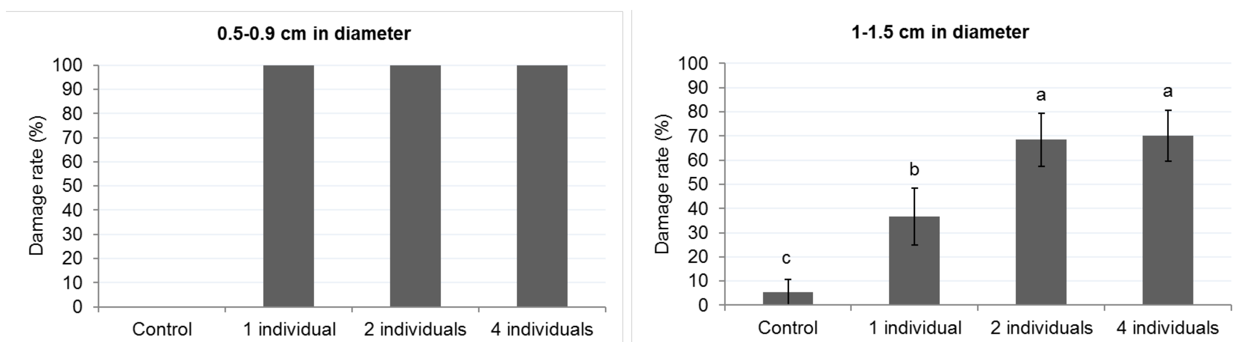

Figure 12. Damage (%) caused by *Creontiades pallidus* nymphs in bolls (0.5-0.9 cm) in Hacıali and (1-1.5 cm) in Balcalı in branch cages after 7 d in 2014.

*The mean numbers with same letter on bars are not significant according to the Duncan multiple range test (P > 0.05).

Creontiades pallidus nymphs negatively affected boll development, with 7-day feeding. The smaller bolls were more damaged in comparison to the matured bolls 7 d after releasing nymphs to the bolls with a diameter of 0.5-0.9 cm. No damage was seen in control cages but with one, two and four individuals release all bolls were damaged. The damage rate on bolls with 1-1.5 cm in diameter increased with increased density of nymphs. Damage rates of the bolls were 70% in the cages with two and four individuals. The differences between the damage rates of bolls were also found to be statistically significant (Figure 12). Stam (1987) reported that *C. pallidus* was found frequently on squares and bolls in cotton, and that small bolls had more black stains caused by mirids sucking. Small bolls are very sensitive to mirid feeding compared to the larger bolls. In other words, small green boll stage is highly susceptible to mirid damage. Greene et al. (1999) reported that *Lygus lineolaris* (Palisot de Beauvois, 1818) caused significant damage to small bolls (8 d from white flowers) in the cage experiments. Layton (2000) reported that *L. lineolaris* usually leads to the loss of small bolls and thus to yield losses. Rosenheim et al. (2006) reported that *Lygus* fed on squares and bolls in California. The pest caused shedding of squares and bolls due to feeding damage to anthers and seeds. Armstrong et al. (2010), investigated *C. signatus* over a 10-year period in Texas. They found that younger bolls were more susceptible to feeding of mirids and thus, they were more damaged. Some of the digestive enzymes are released through the insertion of the stigmata of the mirid and slowing the growth of the bolls by preventing accumulation of assimilates. The developing fiber and seed are damaged, or boll shedding occurs.

Conclusions

The main population development of the pest mirids in Çukurova starts in July and continues until the beginning of August. This period is a critical for development of cotton fruiting bodies as the young square and boll ratio is high on the plants. This period would be suitable for control mirid pests. Young bolls should be taken into consideration when considering insecticide applications to prevent mirid damage to cotton in the region. Cultivation of early cotton cultivars in the region may be as important as a cultural practice. Furthermore, early planting cotton may be also be important in other cotton growing areas where the mirids are regarded as a problem. There was significant relationship between mirid densities and stain numbers on bolls sampled in some experimental years and locations. It was clear that low or high populations of mirids caused significant damage. In fact, stains occur several days after mirid feeding on young and mature bolls. The stage of the pest, the age of bolls and the amount of secreted digestive enzyme are the important factors that affect the occurrence of stains on bolls. It is therefore difficult to correlate a weekly relationship between the number of mirids and the number of stains. However, it is clear that the total number of stains was higher in the year and plots when the total number of mirids was high. It was also found that mirid populations in Adana can exceed the economic threshold. The mirids should be sampled using the recommended methods and chemical control should be applied if needed.

Acknowledgments

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