Research Article

Side Effects of Lambda Cyhalothrin and Thiamethoxam on Insect Pests and Natural Enemies Associated with Cotton

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Abstract | Cotton (*Gossypium hirsutum*) is cultivated mainly for the production of fiber for the textile industry and is attacked by different insect pests, therefore it is produced with frequent pesticide applications. Research was conducted to evaluate the effect of lambda cyhalothrin + thiamethoxam on the incidence of insect pests and its associated natural enemies. Results revealed that populations of *B. tabaci* were higher in plots treated with lambda cyhalothrin + thiamethoxam associated with low levels of parasitism. Further, *A. gossypii* were high and their parasitism low, in both treated and untreated plots. Individuals of *T. palmi*, *B. thurberiella*, *H. virescens* and *A. vestitus* were low in the treatments; while individuals of *Dysdercus* spp. were lower in the plots treated with the chemical insecticide. However, the predators, *Coleomegilla maculata*, *Cycloneda sanguinea*, *Cheilomenes sexmaculata* and *Zelus* sp., were inferior in the treated plots. Yield ranged from 1.2 to 1.4 tons. *Thrips palmi* is first reported feeding on cotton in Ecuador. The results showed high populations of *B. tabaci* in the chemical treatment, and its suppressive effect on the beneficial entomofauna. Future research should focus on evaluating the selective applications of some insecticides to control *A. gossypii* and *Dysdercus* spp. infestations that impact as little as possible on non-target organisms such as parasitoids and predators.

Received | April 17, 2021; **Accepted** | June 21, 2021; **Published** | August 15, 2021
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Citation | Zambrano, N.D., W. Arteaga, J.Velasquez and D.T. Chirinos. 2021. Side effects of lambda cyhalothrin and thiamethoxam on insect pests and natural enemies associated with cotton. *Sarhad Journal of Agriculture,* 37(4): 1098-1106.
DOI | https://dx.doi.org/10.17582/journal.sja/2021/37.4.1098.1106
Keywords | Insecticides, Outbreak of pests, Parasitoids, Predators, Sustainability

Introduction

The genus *Gossypium* (Malvaceae) comprises of 40 species, among which four are commercially cultivated. One of the latter, the upland cotton, *Gossypium hirsutum* L., predominates with 95% of the total production area (FAO, 2015; Trapero *et al*., 2016). It is grown mainly to obtain fiber as a raw material for the textile industry (FAO, 2015). In 2019, 82,589,031 tons of cotton were produced from approximately 38,640,608 hectares (FAOSTAT, 2019) cultivated in more than 78 countries (Razaq *et al*., 2020). This represents 2.3% of all cultivated land in the world and indicates its economic and social importance (FAO, 2015).

Until the end of the 19th century, there were few cases of pests associated with cotton (Razaq *et al*., 2020). However, the large number of hectares planted in different biogeographic regions, resulted in the insertion of a variety of arthropod species within the trophic networks of the agroecosystem, which expanded the diversity of these organisms, compared to what was originally found in center of origin (Naranjo, 2011). There are currently 1,300 species of phytophagous arthropods associated with cotton,

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among which nearly 40 are considered primary pests (Naranjo, 2011; Razaq *et al*., 2020).

Given the importance of the crop, as well as the existing number of arthropods, the application of synthetic insecticides is the main method used to control these pests (Razaq *et al*., 2020), which negatively impacts human health and the productivity of the plant cultivation are not based on an agroecosystem analysis (FAO, 2015). In addition, laboratory and field assays report that applications of organosynthetic insecticides used for the control of important pests in cotton also have side effects for non-target organisms such as predators and parasitoids (Ruberson and Roberts, 2004; Prabhaker *et al*., 2007; El-Wakeil *et al*., 2013).

In Ecuador, cotton played an important role in the agricultural sector as part of the edible oil industry from the 1970s to the 1990s (FAO, 2017). It is currently grown for the textile industry, mainly in the provinces of Guayas and Manabí, with a sowing area of approximately 1,800 ha (Cañarte-Bermudez *et al*., 2020) and an estimated fiber production of 1,200 tons (FAOSTAT, 2019). As in other latitudes, a wide variety of organosynthetic pesticides are used in this country for pest control (FAO, 2017).

Pest management based on the continuous use of chemical pesticides is unsustainable and does not necessarily guarantee the objective of production. In fact, the devastating attack of pests on some crops has reported despite frequent spraying of chemical insecticides, as well as the collateral effects on health and the environment (Chirinos *et al*., 2020). This makes it necessary to reconsider the approaches to agricultural production, especially with regard to the functioning of the agroecosystem and the socioeconomic criteria related to benefits and losses, in order to return to ancestral forms of pest management and make them evolve within the framework of new scientific and technological knowledge (Chirinos *et al*., 2020).

Recent projects propose the reactivation of cotton planting for Ecuador, within the framework of the strengthening of sustainable agriculture for South America (FAO, 2017). As part of these objectives, this work aimed to evaluate the impact of applications of an insecticide based on a mixture of lambda cyhalothrin and thiamethoxam on pest populations and some natural enemies.

Materials and Methods

This research was conducted at the experimental campus "La Teodomira", located in Santa Ana canton (coordinates 01° 09' 51" S and 80° 23' 24" W), Manabí province, Ecuador. Its life zone corresponds to a Tropical Dry Forest.

To start the trial, a 750 m^2 plot of cotton was established, variety Alcalá DP 90. Two plants per point were planted at a distance of 0.4 m x 1.0 m (planting point x row), designed in blocks completely randomized with three replicates and two treatments. Each experimental plot consisted of 125 m^2 (14 rows of 9 m long) and a total of 616 plants (44 plants per row). The evaluated treatments were: T1. Applications of a commercial insecticide consisting of a mixture of lambda cyhalothrin (106 g L^{-1} of active ingredient (i.a.)) + thiamethoxam (141 g.L^{-1} i.a.) +, at a dose of 1.5 cc.L^{-1}. T2. Control. The insecticide was applied three weeks after germination at weekly intervals, totaling ten sprays. The samplings were carried out in each experimental plot, omitting the rows of the ends, in order to avoid the effect of the borders between the treatments. In the 12 internal rows, 30 leaves per plot were randomly selected, for a total of 90 for each treatment.

Once a week during four months, the numbers of: nymphs of the whitefly, *Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae), individuals of the cotton aphid, *Aphis gossypii* Glover, larvae of the leaf borer, *Bucculatrix thurberiella* Busck (Lepidoptera: Bucculatricidae) and thrips species present were counted using a Carl-Zeiss® stereoscope (magnification: 10 - 40X). The number of nymphs and parasitized individuals of *B. tabaci* and *A. gossypii* was also counted, estimating their respective percentage of parasitism [(nymphs or parasitized individuals / total individuals counted) x 100].

In bottles with ethyl alcohol (70%) individuals of predators and the cotton strainers, *Dysdercus* spp. (Hemiptera: Pyrrhocoridae) were collected for 20 minutes in each experimental plot. The bolls from ten plants were sampled to detect the tobacco worm, *Heliothis virescens* F. (Lepidoptera: Noctuidae) and the Peruvian boll weevil, *Anthonomus vestitus* Germar (Coleoptera: Curculionidae). These samplings were conducted from August to October 2019.

The identification of the thrips species was achieved following the taxonomic key proposed by Moritz *et al*. (2004). The coccinellid species were determined by the characters indicated by González (2015). The parasitoid species associated with *B. tabaci* was diagnosed as indicated by Polaszek *et al*. (1992). The predatory bug and the parasitoid of *A. gossypii* were identified by comparing with specimens preserved in the entomological collection of the Crop Protection Laboratory of the Agency for the Regulation of Phyto and Zoosanitary Control (Agrocalidad), Manta, Ecuador. Voucher specimens were stored in this laboratory.

To estimate the yield, three harvests were realized on plants from the two central rows per experimental plot per treatment. The fiber with seed was weighed (grams) with a weight scale and the seed subsequently removed, to obtain the weight of the fiber. The yield was calculated in kilograms per plot [(grams of fiber plant^{-1}/1000) x 616), per plot of 125 m^2 (kg plant^{-1} x 616 plants) and per hectare (kg plant^{-1} x 50000 plants ha^{-1}). The ginning percentage was determined with the differences between the weight of the fibers, with and without seed.

All variables related to number and percentages were analyzed with the Mann-Whitney test (P <0.05), while the performance variables were compared with the student's "t" test (P <0.05). Simple regression models were performed between the number of *B. tabaci* nymphs and the number of *A. gossypii* individuals versus the percentage of parasitism for each species (P <0.05).

Results and Discussion

Whitefly, Bemisia tabaci

The nymphs reached the highest levels in the plants treated weekly with the mixture of lambda cyhalothrin + thiamethoxam. These levels increased from 7 to 34 nymphs leaf^{-1} while for untreated plots varied from 0.2 to 2.0 nymphs leaf^{-1}. The highest number of *B. tabaci* nymphs in plants treated with lambda cyhalothrin + thiamethoxam was related to low parasitism, that did not exceed 12% (Figure 1), while in untreated plots there was a lower number of nymphs associated with higher percentages of parasitism (ranging from 34 to 100%).

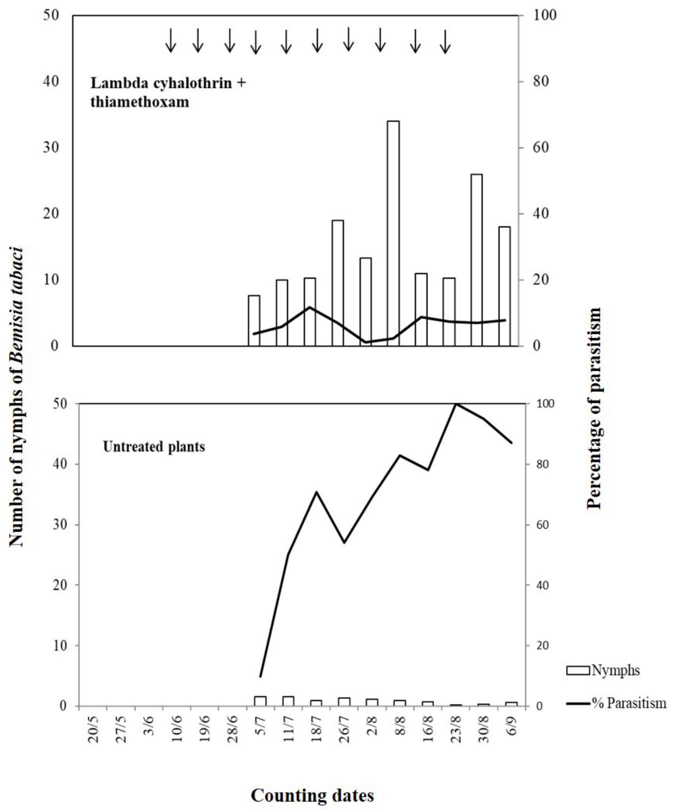

Figure 1: *Number of nymphs of Bemisia tabaci and percentage of parasitism exerted by Encarsia pergandiella on cotton leaves for the treatments. Arrows indicate spray dates. Year: 2019.*

The high dispersion detected in the regression analysis between *B. tabaci* nymphs versus the percentage of parasitism suggests a low or no response of this biological control agent when the plot was treated with lambda cyhalothrin + thiamethoxam (R^2: 0.087) (Figure 2). This would indicate the adverse effects of weekly applications of the chemical pesticide on parasitism. In contrast, the model calculated for untreated plots showed a high coefficient of determination (R^2: 0.83) that would attribute a parasitism response to variations in population densities of *B. tabaci* when there was no pesticide interference. Additionally, the statistical analysis corroborates that on treated plots the populations were higher, together with the lower percentages of parasitism (Table 1, P<0.05).

Encarsia pergandiella Howard (Hymenoptera: Aphelinidae) was the parasitoid species associated with *B. tabaci*, identified in this research. Schuster *et al*. (1998) had reported this species for Ecuador in a survey of *Bemisia* spp. parasitoids, carried out in Florida, Central and South America. The results suggest the adverse effects of the chemical treatment on the parasitism of *B. tabaci*. Dutcher (2007) pointed out that pest resurgence occurs when a pesticide treatment destroys their population and kills or negatively affects their natural enemies. Thus, the residual activity of the pesticide expires and the populations of the pests can increase more quickly and to a greater abundance when the natural enemies are in low proportion. In cotton, insecticide applications for the control of this species have historically resulted in conspicuous population increases attributed in part to the development of resistance, but the main cause is the suppression of its parasitoids (Oliveira *et al*., 2001).

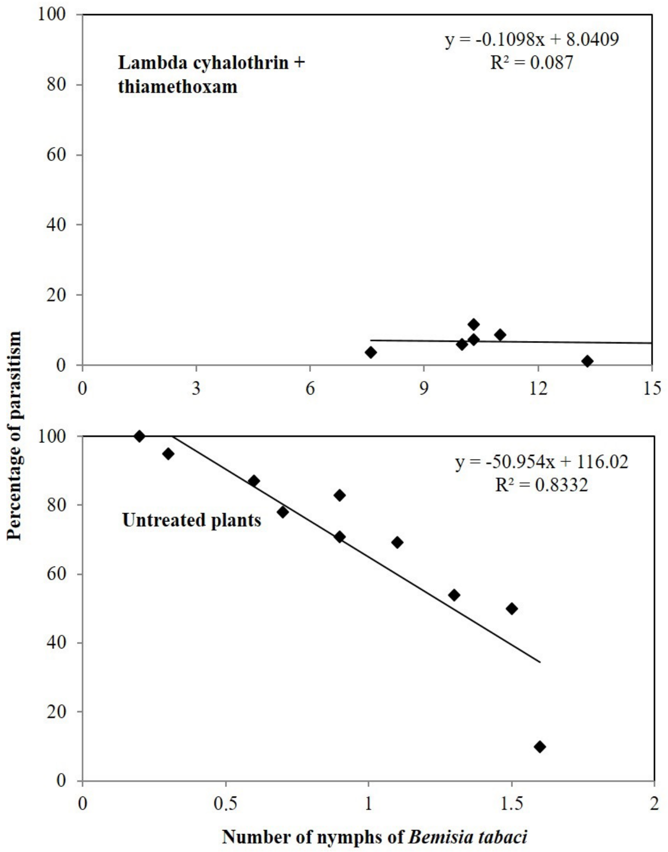

Figure 2: *Linear regression equations between the number of Bemisia tabaci nymphs (X) and the percentage of parasitism (Y) exerted by Encarsia pergandiella on cotton leaves, for the treatments.*

The cotton aphid, Aphis gossypii

Aphis gossypii increased in plots treated with lambda cyhalothrin + thiamethoxan from the eleventh week, reaching the highest levels of 63 individuals leaf^{-1} (thirteenth week) while in the control the increase began in the sixth week showing the maximum number of 17 individuals leaf^{-1} (fourteenth week) (Figure 3). Although the population peaks of the aphids were higher in treated plants, there were no differences between treatments (Table 1).

On *A. gossypii* parasitism by *Lysiphlebus testaceipes* (Cresson) (Hymenoptera: Braconidae) was observed, but in the treated plots with lambda cyhalothrin + thiamethoxam it was significantly lower (Table 1). This endoparasitoidis referred to as an important biological control agent for *A. gossypii*, in several crops (Tomanović *et al*., 2014; Albittar *et al*., 2016). However, in this assay, parasitism remained low, showed maximum levels of 44% (Figure 3). The absence of a functional response of parasitism to variations in the population density of aphids would be indicated by the low coefficients of determination of the calculated regression models both in treated and untreated plots (R^2: 0.0002 - 0.04) (Figure 4).

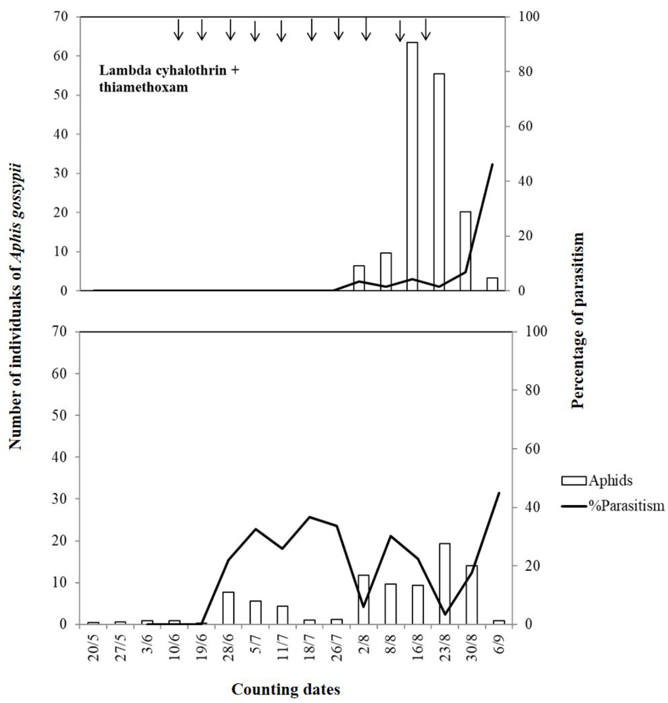

Figure 3: *Number of Aphis gossypii individuals and percentage of parasitism exerted by Lysiphlebus testaceipes in cotton leaves, for the treatments. Arrows indicate spray dates. Year: 2019.*

Same results were obtained by Romero *et al*. (2019), who in a field study conducted in cucumber plots without insecticides, found populations of 19 individuals leaf^{-1} of *A. gossypii*, with percentages of parasitism that ranged from 35 to 70%. Kaleem *et al*. (2014) reported that natural biological control in aphids is effective when there are low population densities per plant. This aphid is considered an important pest that can infest cotton from the beginning of the cycle until harvest, causing delays in the development of the plant (Pinto *et al*., 2013).

The thrips, Thrips palmi

The species of thrips present was identified as *Thrips palmi* Karny (Thysanoptera: Thripidae) whose main characteristics are summarized in Figure 5. This species had not been mentioned feeding on cotton plants in Ecuador. In fact, a previous investigation, only reported another genus of thrips (*Frankliniella*) but without determining the species (Cañarte-Bermudez *et al*., 2020). A low number of *T. palmi* individuals was found on leaves, which showed no significant differences between treated and untreated plants (Table 2). These abundances are in contrast to those obtained in previous field investigations in other regions. Janu *et al*. (2017) observed population levels of *Thrips tabaci* Lindenmann that reached 28 and 24 individuals leaf^{-1} in a fieldwork conducted during two production cycles (years 2014 and 2015) to evaluate the response of some transgenic cotton genotypes in India. Jaramillo-Barrios *et al*. (2018) indicated maximum peaks of approximately 85 individuals of *T. palmi* on cotton leaves in a fieldwork performed in Colombia to determine the preference of plant organs by species of thrips.

The leaf borer, Bucculatrix thurberiella

Bucculatrix thurberiella was another species found feeding on cotton leaves at very low levels. There were no significant differences between treatments (Table 2). The population levels detected (ranging from 0.02 to 0.2 larvae leaf^{-1}, Table 2) were very similar to those obtained by research performed in the same area that used chemical insecticide to managing. Cañarte-Bermudez *et al*. (2020) pointed out general averages of 0.48 individuals leaf^{-1} spraying lambda cyhalothrin + thiamethoxam, azadirachtin, abacmectin and cypermethrin. However, this species represents an

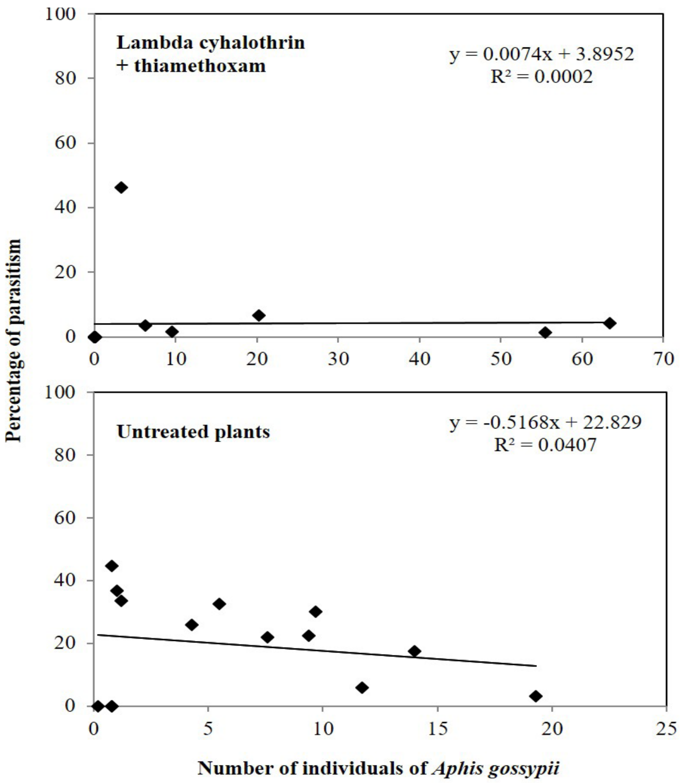

Figure 4: *Linear regression equations between the number of individuals of Aphis gossypii (X) and percentage of parasitism (Y) exerted by Lysiphlebus testaceipes in cotton leaves, for the treatments.*

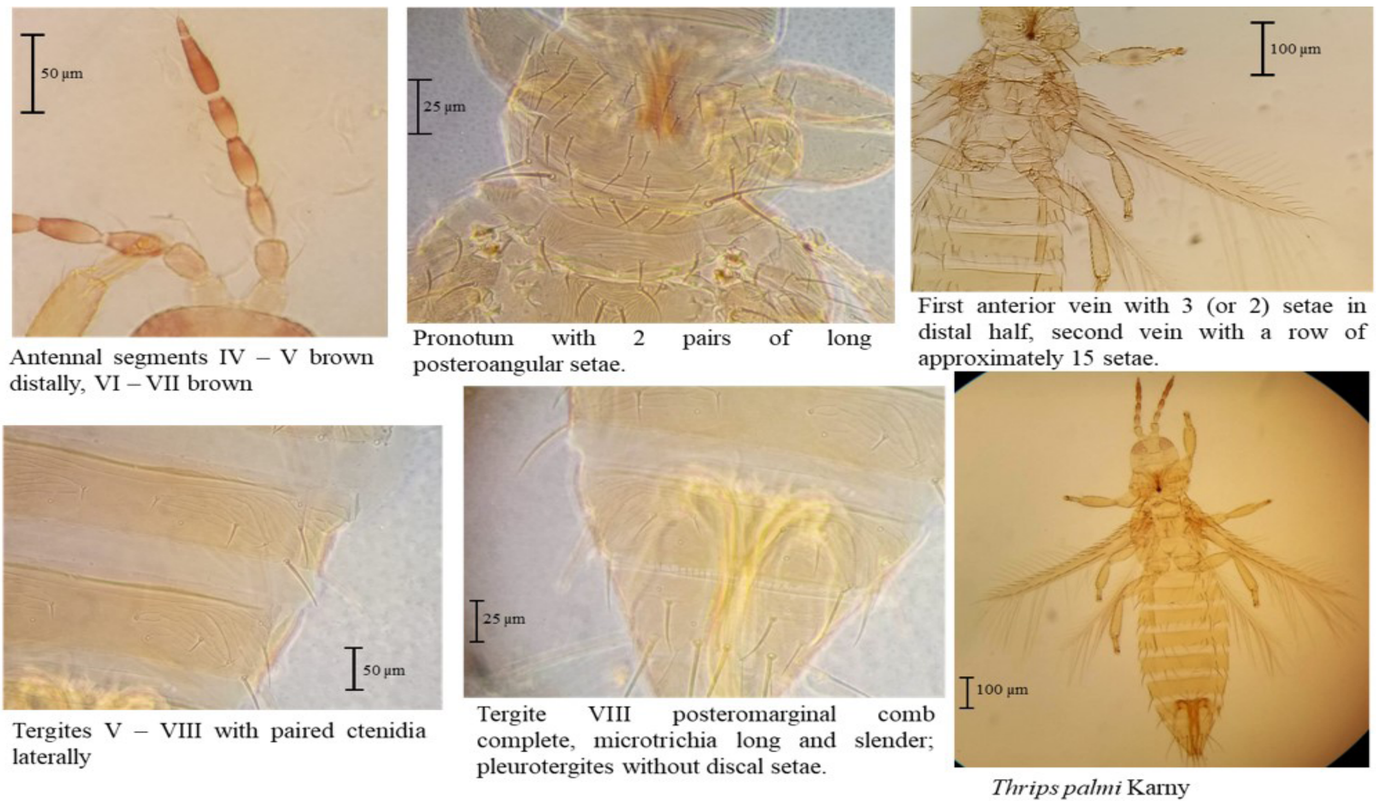

Figure 5: *Morphological characteristics of the species Thrips palmi Karny.*

Table 1: *General average number of nymphs of Bemisia tabaci, and of individuals of Aphis gossypii, and percentage of parasitism on cotton leaves.*

	Bemisia tabaci		*Aphis gossypii*	
Treatment	Nymphs	Parasitism (%)	Individual	Parasitism (%)
Lambda cyhalothrin + thiamethoxam	15.9 ± 2.8 [a]	6.5 ± 1.0 [b]	9.9 ± 2.8 [a]	3.9 ± 1.8 [b]
Untreated plants	1.1 ± 0.1 [b]	69.4 ± 2.1 [a]	5.3 ± 1.5 [a]	17.2 ± 2.1 [a]

Means±standard error. Mean followed by different letters in each column is significantly different at 0.05 level of significance using Mann-Whitney test.

Table 2: *General average of the number of individuals of the species Thrips palmi, Bucculatrix thurberiella, Dysdercus spp., Heliothis virescens and Anthonomus vestitus on cotton plants.*

Treatment	*Thrips palmi*	*Bucculatrix thurberiella*	*Dysdercus spp.*	*Heliothis virescens*	*Anthonomus vestitus*
Lambda cyhalothrin+ thiamethoxam	0.1±0.1 [a]	0.02±0.02 [a]	1.7 ± 0.3 [a]	0.0 [b]	0.0 [b]
Untreated plants	0.6 ± 0.1 [a]	0.2 ± 0.1 [a]	7.7 ± 1.0 [b]	0.5±0.2 [a]	0.3 ± 0.1 [b]

Means±standard error. Mean followed by different letters in each column is significantly different at 0.05 level of significance using Mann-Whitney test.

important pest on cotton crops in other regions (Gil and Lopez, 2017). Herrera and García (1978) indicated that natural enemies play an important role in the population regulation of *B. thurberiella*, but the applications of chemical insecticides for other important pests could generate ecological imbalances and increase their populations in the crop.

The cotton strainers, Dysdercus spp.

The number of individuals of *Dysdercus* spp. showed differences between treatments. While in plots treated with lambda cyhalothrin + thiamethoxam, 1 to 2 individuals plant^{-1} were observed, under untreated plot, the number of individuals increased from 5 to 11 (Figure 6), resulting significantly higher than this last treatment (Table 2). The results are similar to those reported by Cañarte-Bermudez *et al.* (2020) who for the same area and months of evaluation reported a high incidence of *Dysdercus* spp. associated with the fiber formation stage. Regarding the chemical control of this pest, tests conducted by Rafiq *et al.* (2014) show the effectiveness of pyrethroids, carbamates and organophosphates. These researchers conclude that, to determine the number of minimum sprays necessary to control species of this genus, it is essential to study the biological cycle and estimate the population development parameters.

The tobacco worm, Heliothis virescens and the cotton weevil, Anthonomus vestitus

During this research, 0.5 and 0.3 individual boll^{-1} of *H. virescens* and *A. vestitus*, respectively were observed

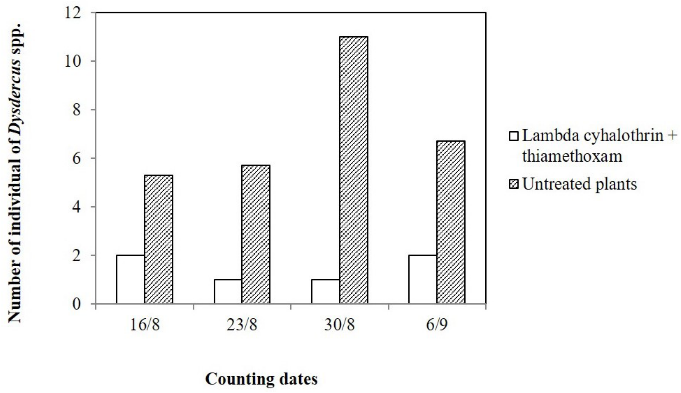

Figure 6: *Number of individuals of Dysdercus spp. on cotton leaves, for the treatments. Year: 2019.*

on untreated plot (Table 2). Despite the fact that in other regions of South America, both species are relevant pests in cotton crops (FAO, 2017; Gil and López, 2017), the damage was no significant in this fieldwork. In Ecuador, *H. virescens* has been reported attacking in the flowering stage and it has been indicated that *A. vestitus* does not represent a devastating pest in that country due to its low incidence (FAO, 2017).

Predators

A total of 298 individuals were observed belonging to four species of predators, three from the Coccinellidae (Coleoptera) and one from the Reduviidae (Hemiptera). Of these, *Coleomegilla maculata* De Geer together with *Zelus* sp., were the most abundant species (Table 3). The latter species of predators had previously been reported in the area (Cañarte-Bermudez *et al.*, 2020). The results showed a significantly lower number of predators in treated plants with lambda cyhalothrin + thiamethoxam (Table 3). Field investigations conducted on cotton in Egypt have reported high mortality rates of predators, *Coccinella undecimpunctata* L. (Coleoptera: Coccinellidae) and *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae) with frequent sprays of insecticides belonging to various chemical groups (El-Heneidy *et al*., 2015; Eldesouky, 2019).

Table 3: *Number of individuals per species of predator found in cotton plants.*

Treatment	*Coleomegilla maculata*	*Cycloneda sanguínea*	*Cheilomenes sexmaculata*	*Zelus* sp.
Lambda cyhalothrin+thiamethoxam	14 [b]	7 [b]	8 [b]	12 [b]
Untreated plants	49 [a]	55 [a]	36 [a]	70 [a]
Total	110	62	44	82

Means±standard error. Mean followed by different letters in each column is significantly different at 0.05 level of significance using Mann-Whitney test.

Yield cotton

The fiber yield (with and without seed) estimated for the plot and per hectare showed no significant differences between treatments (Table 4). The ginning percentage was approximately 40%.

Table 4: *Yield of cotton fiber per plot of 125 m^2 and estimated ha^{-1} (kg).*

Treatment	Fiber with seed (kg 125 m^2)	Fiber without seed (kg ha^{-1})	Fibra (kg ha^{-1})
Lambda cyhalothrin + thiamethoxam	45.98 [a]	3679.16 [a]	1471.67 [a]
Untreated plants	41.15 [a]	3291.67 [a]	1298.23 [a]

Means±standard error. Mean followed by different letters in each column is significantly different at 0.05 level of significance using the student's "t test.

The fiber yields estimated ha^{-1} varied from 1.4 (treated plot) to 1.2 ton ha^{-1} (untreated plot), and were higher. These yields were higher than those of Campuzano *et al*. (2015) determined in a trial carried out in Colombia and similar to those obtained in another study conducted in the area (Cañarte *et al*., 2020), using the DP Alcalá 90 variety in both cases. Although there were no significant differences between treatments, this would represent that the yield in treated plots was 11.6% higher than that estimated in untreated plots. However, the higher percentage in yield in plots treated with the insecticide must be analyzed in the context of the ecological and economic costs involved.

Conclusions and Recommendations

In the present research, the high population levels of *B. tabaci* were associated with low levels of parasitism by *E. perganidiella* due to interference from weekly sprays with lambda cyhalothrin + thiamethoxam that also decreased the number of predators. *Thrips palmi*, *B. thurberiella*, *H. virescens* and *A. vestitis* showed low levels, despite their importance as pests in other regions. *Thrips palmi* is reported for the first time feeding on cotton in Ecuador. Future research should focus on evaluating the selective applications of some insecticides to control *A. gossypii* and *Dysdercus* spp. infestations that impacts as little as possible on non-target organisms such as parasitoids and predators.

Acknowledgements

To Dr. Angel Luis Viloria (IVIC, Venezuela) for kindly reviewing the manuscript and the English text. This research was partially funded by The Grants No. PYTAUTO1639-2019-FIAG0002 (Universidad Técnica de Manabí (UTM)), Ecuador and South-South Cooperation for Strengthening the Cotton Sector, Food and Agriculture Organization of the United Nations (FAO).

Novelty Statement

General average of the number of individuals of the species *Thrips palmi*, *Bucculatrix thurberiella*, *Dysdercus* spp., *Heliothis virescens* and *Anthonomus vestitus* on cotton plants.

Author's Contribution

Nelson D. Zambrano and Wilber Arteaga: field evaluations lab counts; tabulation of results, interpretation of data.
José Velasquez: mounting and preservation of insects; identification of pests and natural enemies.
Dorys T. Chirinos: trial design; data analysis; wrote the manuscript.

Conflict of interest

The authors have declared no conflict of interest.

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