Evaluation of Three Commercial Biopesticides for the Reduction of *Diaphorina citri* (Hemiptera: Liviidae) Populations

Augusto Ramírez-Godoy, María del Pilar Vera-Hoyos, Natalia Jiménez-Beltrán, and Hermann Restrepo-Díaz¹

Departamento de Agronomía, Facultad de Ciencias Agrarias, Universidad Nacional de Colombia, Bogotá, Colombia

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Abstract. Asian citrus psyllid (ACP), *Diaphorina citri* Kuwayama, is the most serious threat to the global citrus industry, and its management has mainly depended on the application of chemical insecticides. The use of biopesticides can play an important role in regulating this pest. In a first test, two separate experiments were conducted in two different municipalities (Apulo and Jerusalem, Cundinamarca, Colombia) to evaluate the effect of foliar *Beauveria bassiana* and imidacloprid sprays on ACP populations in 2015 and 2016, respectively. In a second test, two separate tests were carried out in commercial Tahiti lime orchards to evaluate the efficacy of three different commercial biopesticides (*Beauveria bassiana* and extracts of *Sophora* sp. and garlic-chili pepper). In test 1, imidacloprid-treated trees showed a reduction (60% and 80%) in cumulative ACP adults in 2016. ACP cumulative nymphs were also diminished by foliar imidacloprid and *B. bassiana* application, between 40% and 65% in 2015 and 2016, respectively. ACP cumulative eggs showed lower individuals in imidacloprid-treated flushes at 3 and 4 weeks after treatment 9 (WAT) in 2016. In test 2, the results obtained showed ACP adults and eggs unaffected by biopesticide treatments; nymphs were reduced 50% to 75% in trees treated with the three biopesticides in comparison with control trees. All three biopesticides tested can be considered useful tools in integrated pest management (IPM) programs for ACP, since these products reduced immature ACP individuals between 50% and 75% under field conditions.

Asian citrus psyllid (ACP), *Diaphorina citri* Kuwayama (Hemiptera: Liviidae), is currently the most important insect pest in the global citrus industry because it is the vector responsible for transmission of the bacteria *Candidatus Liberibacter asiaticus* and *Candidatus Liberibacter americanus*, the putative causative agents of Huanglongbing disease (HLB) (Halbert et al., 2000). ACP was officially reported in Colombia in 2007 (Santivañez et al., 2013). Recently, the Colombian Agricultural Institute (ICA) declared a state of phytosanitary emergency in 2015 due to the presence of ACP adults carrying the bacteria that causes HLB (ICA, 2015). In Colombia, citrus crops occupy an area of 75,000 ha, with production of 1.75 million tons per year. Acid limes are one of the most cultivated species, with production of 360,836 tons in 2014 (Agronet, 2017).

The frequent application of insecticides has been an important tool used by growers to control ACP populations (Chen and Stelinski,

2017; Tiwari et al., 2011). Repeated application of insecticides, however, has caused strong selection pressure, resulting in resistance and reducing the effectiveness of insecticides (Boina and Bloomquist, 2015). Likewise, the excessive use of insecticides may be economically unfeasible for growers and may also affect natural enemies that contribute to the control of ACP (Lezama-Gutiérrez et al., 2012). For this reason, biopesticides have been considered as substitutes or in rotation with current synthetic insecticides in integrated pest management (IPM) programs for ACP (Khan et al., 2015; Santos et al., 2015; Weathersbee and McKenzie, 2005).

Entomopathogenic fungi have shown promising results as eco-friendly biopesticides in the control of the ACP populations because they manage to infect the insect in all stages of its development (Gandarilla-Pacheco et al., 2013; Kumar et al., 2017; Saldarriaga-Ausique et al., 2017). Additionally, these biocontrol agents are commercially available and represent an effective control alternative, since they help to reduce the problem of insecticide resistance and negative effects on populations of natural enemies (Saldarriaga-Ausique et al., 2017). Some strains of the entomopathogenic fungus

Beauveria bassiana (Bals.-Criv.) (Hypocreales: Clavicipitaceae) have been reported to be a potential regulator of ACP populations. Several studies under controlled conditions have shown high efficacy against the immature stages (nymphs) of this insect (Ferreira Pinto et al., 2012; Ibarra-Cortés et al., 2017). However, efficacy tests under field conditions are still scarce (Orduño-Cruz et al., 2015; Saldarriaga-Ausique et al., 2017).

Insecticides obtained from natural sources such as plant extracts can have pesticidal effects on arthropods and have been used in IPM programs because they help to reduce environmental pollution, conserve natural enemies of pests, and ward off pest resurgence (Khan et al., 2014, 2015). Phytochemicals extracted from the leaves, stems or roots of *Sophora* sp. (L.) (Fabales: Fabaceae) or *Azadirachta indica* (A. Juss) (Sapindales: Meliaceae), such as matrine or azadirachtin, have pesticide activity (Xin et al., 2016). Recently, matrine extracts have been studied alone or combined with other chemicals to control different pests, such as *Coptotermes formosanus* (Shiraki) (Mao and Henderson, 2007), *Bemisia tabaci* (Gennadius) (Ali et al., 2017), *Mythimna separata* (Walker), and *Plutella xylostella* (Linnaeus) (Huang et al., 2017). Additionally, the use of matrine-based biopesticides has also become important in controlling ACP as an IPM tool in recent years (Fang et al., 2018; Zanardi et al., 2015).

Allium sativum (L.) extracts have shown positive effects in controlling insect pests in commercial crops (Zehnder et al., 2007). ACP adults showed 85% repellence when citrus trees were treated with foliar garlic extract sprays at a concentration of 50 ppm (Cazares-Alonso et al., 2014). Bioassays developed by Mann et al. (2011) also showed higher repellency of adults in response to exposure to sulfur volatiles obtained from garlic bulbs. The application of *Capsicum annum* (L.) extracts has also shown positive results in the management of insect pests because they contain capsaicin, which has lethal and antifeeding effects on invertebrates (Baidoo and Mochiah, 2016; Kazem and El-Shereif, 2010).

Although ACP was reported in Colombia about a decade ago, control of this pest with biopesticides is still scarce (Mejía-Vásquez, 2016). Additionally, most information available on the effectiveness of biopesticides on ACP has been found mainly through research carried out under laboratory or greenhouse conditions. The effectiveness of these biopesticides under field conditions has been poorly documented, especially in tropical dry forest conditions, and ACP mainly develops in these habitats in Colombia (Ebratt-Ravelo et al., 2011). For this reason, the objectives of this study were to evaluate the reduction of ACP populations (adults, nymphs, and eggs) through the application of three different commercial biopesticides [*B. bassiana*, extracts of *Sophora* sp., and a mixture of extracts of *Allium sativum* (L.) and *Capsicum annum* (L.)] under field conditions.

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¹Corresponding author. E-mail: hrestrepod@unal.edu.co.

Materials and Methods

Test 1: Evaluation of a commercial biopesticide vs. a synthesis insecticide on ACP population control. In this test, the objective was to evaluate the efficacy of a commercial biopesticide (*B. bassiana*) vs. a synthetic insecticide on ACP control. Two separate short-term evaluations (five weeks) were conducted in two commercial orchards of Tahiti lime located on two different farms in the Department of Cundinamarca in Colombia. The first evaluation was carried out on a farm with a drip irrigation system located in the municipality of Jerusalén (lat. 4°33'33.91"N, long. 74°41'39.09"W, altitude: elevation 316 m) from Oct. to Nov. 2015. The second evaluation was also performed on a farm with drip irrigation located in the municipality of Apulo (lat. 4°30'44.73"N, long. 74°34'57.58"W, altitude: elevation 420 m) from Oct. to Nov. 2016. The climatic conditions (rainfall and average temperature) during two experiments in both locations are presented in Fig. 1. The two evaluations were carried out on 6- and 8-year-old 'Tahiti lime' (*Citrus latifolia* Tanaka) grafted onto 'Citrumelo CPB 4475' (*Poncirus trifoliata* L. × *Citrus paradise*

Macfad) trees in both locations, respectively. Additionally, all trees were spaced 8 × 5 m.

The treatments studied were 1) control, no foliar treatment; 2) trees treated with imidacloprid (Confidor® 350 SC; Bayer, Bogotá, Colombia) at 0.10 L a.i. per ha; and 3) trees treated with a commercial strain of *B. bassiana* (spores of the GHA strain) (Mycotrol®; Laverlam S.A., Cali, Colombia). A conidial germination viability of 90% for *B. bassiana* was obtained using the method described by Oliveira et al. (2015).

Foliar imidacloprid and *B. bassiana* sprays were applied early morning at 0 weeks after treatment (WAT). In both treatments, spray applications (2 L per tree) were performed with a pesticide backpack sprayer (Royal Condor; Soacha, Colombia), wetting the upper and lower surfaces until dripping. Treatments were arranged in randomized complete block design (four blocks). For each experimental unit (treatment), three trees were used per treatment, for a total of 12 trees per treatment. In all treatments, guard trees were used to avoid edge effects due to drift by foliar sprays.

Test 2: Evaluation of three commercial biopesticides on ACP population control. Taking into account the previous observations

in test 1, the aim of these experiments was to compare the use of *B. bassiana* vs. plant extract biopesticides (*Sophora* sp. and a mixture of extracts of *A. sativum* and *C. annuum*). Two separate experiments were carried out between Aug. and Sept. 2017 at the farms 'La Rochela' (lat. 4°33'22.1"N, long. 74°43'36.8"W, altitude: elevation 305 m) and 'Rochelita' (lat. 4°33'07.6"N, long. 74°44'45.6"W, altitude: elevation 263 m) located in the municipality of Jerusalén in the Department of Cundinamarca in Colombia. The climatic conditions (precipitation and maximum and minimum temperatures) during the experiments at both farms are summarized in Fig. 2. The two experiments were conducted on 2- and 5-year-old 'Tahiti lime' (*C. latifolia* Tanaka) grafted onto 'Citrumelo CPB 4475' (*P. trifoliata* L. × *C. paradisi* Macfad) trees at the two farms under drip irrigation, respectively. All trees were spaced 8 × 5 m.

The treatments established in both farms were 1) trees without foliar biopesticide sprays (control); 2) trees treated with foliar *B. bassiana* (spores of the GHA strain) applications (Mycotrol®; Laverlam S.A.) at a commercial dose of 1×10^9 spores/L H₂O; 3) trees treated with foliar *Sophora* spp. extract (with a content of 0.03% matrine)

Fig. 1. Rainfall records and average temperatures in the municipalities of Jerusalén (2015) (A and B) and Apulo (2016) (C and D), respectively.

Fig. 2. Rainfall records (A) and average temperature (B) for the evaluation trials of the effects of three biopesticides on the control of ACP populations in the municipality of Jerusalén. Data were obtained from a nearby weather station [Jerusalén station (lat. 4°56'18.06"N, long. 74°70'23.99"W, altitude: elevation 297 m) of the Institute of Hydrology, Meteorology and Environmental Studies (IDEAM)].

(ADNGREEN®; A.D.N Adelante SAS, Rio-negro, Colombia) at a commercial dose of 3.2 mL·L⁻¹ H₂O; and 4) trees treated with foliar garlic-chili pepper applications (with a concentration of 54.2% garlic + 43.4% chili pepper extract) (Capsialil SL®; Ecoflora Agro, Medellín, Colombia) at a commercial dose of 2.8 mL·L⁻¹ H₂O. A conidial germination viability of 93% for *B. bassiana* was obtained using the method described by Oliveira et al. (2015).

Foliar biopesticide applications at both farms were carried out at 0 and 2WAT) between 7:00 and 9:00 AM using a back pump with an application volume of 2 L H₂O per tree, wetting both the upper and lower leaf surfaces. Foliar applications were performed without any adjuvants. For each treatment, three trees were also used in each experiment.

Treatments were arranged in a randomized complete block design (four blocks). In all treatments, guard trees were also used to avoid edge effects due to drift by foliar sprays.

Population dynamics of ACP individuals. The method described by Sétamou et al. (2008) was used to estimate ACP population variability in both tests. Relative infestation levels of ACP adults, eggs, and nymphs were estimated separately by randomly reviewing five young flush shoots (0.5–1.0 cm) per tree at each of the four cardinal points. In addition, eggs and nymphs were counted in situ and registered per flush shoot using a 15X handheld lens. In general, samples for both trials were collected between zero and four WAT at intervals of seven days per sampling.

Efficacy percentage. The method described by Henderson and Tilton (1955) was used to assess treatment efficacy on ACP populations at the end of the experiments (4 WAT) in both tests. ACP population (adults, nymphs, and eggs) levels recorded separately before and after treatments were used to calculate efficacy percentage. In general, the efficacy percentage for each treatment was determined using the following formula:

$$\% \text{ Efficacy} = 100 \times \left(1 - \frac{(Ta \times Cb)}{(Tb \times Ca)} \right)$$

where *T* is the treated population, *C* is the control population, *a* is the sampled population after treatment, and *b* is the sampled population before treatment.

Experimental design and statistical analysis. A randomized block design was used to study the effects of foliar *B. bassiana* and imidacloprid treatments on ACP populations in test 1. A multiobservational *F*-protected analysis (a series of experiments) was used for test 2. To analyze ACP populations, data were transformed (Log *X* + 1), since data were not normally distributed. Percentage data were transformed using the arcsine formula. When differences were observed (*P* ≤ 0.05) in an ANOVA, the Tukey post hoc test was used to compare means. The data were analyzed using the statistical program Statistix (Version 9, Tallahassee, FL).

Results and Discussion

Test 1: Cumulative individuals and efficacy percentage of a commercial biopesticide vs. a synthesis insecticide. Significant differences were observed in the number of ACP eggs, nymphs, and adults in Tahiti lime trees due to treatments in the two different experiments (Fig. 3). Foliar treatment applications affected cumulative adults in both experiments. Imidacloprid-treated trees showed a reduction (between 60% and 80%) on cumulative ACP adults, obtaining differences among treatments mainly at 1 (*F* = 3.09; *df* = 2,35; *P* = 0.040) and 2 (*F* = 3.48; *df* = 2,35; *P* = 0.043) WAT in 2015, and 3 (*F* = 4.36; *df* = 2,47; *P* = 0.019) and 4 (*F* = 6.10; *df* = 2,47; *P* = 0.004) WAT in 2016 (Fig. 3A and B). Regarding ACP cumulative nymphs, foliar imidacloprid application also caused a reduction in this developmental stage. In 2015, differences (*F* = 2.15; *df* = 2,35; *P* = 0.05) were only recorded at 4 WAT, where imidacloprid-treated flush shoots had 4.2 individuals compared with 8.5 individuals in untreated flushes (Fig. 3C) and cumulative nymphs were affected by treatments starting at 2 (*F* = 2.41; *df* = 2,47; *P* = 0.049) WAT in 2016 (Fig. 3D). Control trees showed higher individual numbers (10 cumulative individuals per flush shoot) in comparison with *B. bassiana* and imidacloprid treatments (4 cumulative individuals). In relation to cumulative eggs, differences were not observed in 2015 (Fig. 3E). However, differences were recorded in 2016, when imidacloprid-treated flushes showed lower cumulative individuals at 3 (*F* = 3.31;

Fig. 3. Average cumulative ACP individuals (adults, nymphs, and eggs) per flush shoot in Tahiti lime trees treated with imidacloprid and *Beauveria bassiana* in experiment 1 (Jerusalem, 2015) (A, C, and E) and experiment 2 (Apulo, 2016) (B, D, and F). Points represent the mean of four values $\pm$ SE. NS: No significance; ***significant difference ($P \leq 0.001$); **significant difference ($P \leq 0.01$); *significant difference ($P \leq 0.05$) compared with the control according to the Tukey test. Foliar treatment sprays were carried out at 0 weeks after treatments.

df = 2,47; $P = 0.046$) and 4 ($F = 3.82$; df = 2,47; $P = 0.03$) WAT (Fig. 3F).

The percent efficacy of treatments (*B. bassiana* and imidacloprid) was evaluated on ACP adults, nymphs, and eggs at 2 and 4 WAT in both experiments (2015 and 2016) (Fig. 4). In general, differences were not found on the percent efficacy in all developmental stages (adults, nymphs, and eggs)

between imidacloprid and *B. bassiana* at 2 WAT in both years. However, flush shoots treated with imidacloprid showed a percent efficacy of 58% and 62% on adult population compared with 33% and 35% in trees treated with *B. bassiana* in 2015 and 2016 at 4 WAT, respectively (Fig. 4A and B). Imidacloprid-treated flushes also showed a higher efficacy percentage (72%) on

nymph control than *B. bassiana* (43%) in 2015 (Fig. 4C). Nevertheless, opposite results were observed for this variable in 2016, in which *B. bassiana* showed a better efficacy (62%) in comparison with imidacloprid (40%) (Fig. 4D). Differences were not obtained in percentage efficacy (45%) on ACP eggs among treatments in both years (Fig. 4F).

Fig. 4. Summary of the efficiency percentage per flush shoot in Tahiti lime trees treated with imidacloprid and *Beauveria bassiana* in experiment 1 (Jerusalem, 2015) [adults (A), nymphs (C), and eggs (E)] and experiment 2 (Apulo, 2016) [adults (B), nymphs (D), and eggs (F)]. Bar charts represent the mean of four values $\pm$ SE. ^{NS}No significance; *significant difference ($P \leq 0.05$) according to the Tukey test. Foliar treatment sprays were carried out at 0 weeks after treatments.

Test 2: Cumulative individuals and efficacy percentage of three commercial biopesticides. ACP adults were unaffected by the use of the three different commercial biopesticides in this test (Table 1; Fig. 5A). However, the nymph population was reduced by the use of *B. bassiana* and commercial extracts of *Sophora* spp. and garlic-chili pepper, with differences ($F = 3.23$; $df = 3,95$; $P = 0.026$) obtained in comparison with untreated trees from 1 WAT. Tahiti lime trees treated with foliar biopesticide sprays presented $\approx 50\%$ to 75% fewer nymphs compared with control trees at 1 (1.33 vs. 2.42 nymphs per flush shoot), 2 (0.43 vs. 1.33 nymphs per flush shoot), 3 (0.40 vs. 1.67

Table 1. Probability of F statistic ($P > F$) of ACP individuals per flush shoot (adults, nymphs, and eggs) on Tahiti lime trees treated with three commercial biopesticides.

	Weeks after treatments (WAT)				
	0 ^a	1	2	3	4
<i>Adults</i>					
Farms (F)	$P = 0.739$	$P = 0.079$	$P = 1.00$	$P = 0.513$	$P = 0.565$
Treatments (T)	$P = 0.924$	$P = 0.285$	$P = 0.119$	$P = 0.281$	$P = 0.801$
F $\times$ T	$P = 0.517$	$P = 0.062$	$P = 1.00$	$P = 0.933$	$P = 0.306$
<i>Nymphs</i>					
Farms (F)	$P = 0.336$	$P = 0.008$	$P = 0.751$	$P = 0.003$	$P = 0.074$
Treatments (T)	$P = 0.135$	$P = 0.026$	$P = 0.002$	$P = 0.000$	$P = 0.000$
F $\times$ T	$P = 0.396$	$P = 0.018$	$P = 0.991$	$P = 0.452$	$P = 0.191$
<i>Eggs</i>					
Farms (F)	$P = 0.000$	$P = 0.001$	$P = 0.592$	$P = 0.045$	$P = 0.373$
Treatments (T)	$P = 0.051$	$P = 0.101$	$P = 0.001$	$P = 0.183$	$P = 0.000$
F $\times$ T	$P = 0.089$	$P = 0.896$	$P = 0.774$	$P = 0.652$	$P = 0.902$

^aFoliar applications of biopesticides were conducted at 0 and 2 weeks after initiating the treatments.

nymphs per flush shoot) and 4 (0.35 vs. 1.45 nymphs per flush shoot) WAT (Fig. 5B). Regarding egg population dynamics in flush shoots, differences were only observed at 2 (F = 5.49; df = 3,95; P = 0.001) and 4 (F = 11.31; df = 3,95; P = 0.000) WAT. The three commercial biopesticides caused a reduction of 54% and 62% compared with untreated trees at 2 and 4 WAT, respectively (Fig. 5C).

The analysis of the population dynamics expressed as individuals that had accumulated throughout the test showed that the use of biopesticides had no effect on adults, but it did affect the immature stages (nymphs and eggs) (Table 2; Fig. 6). Differences were observed for nymphs and eggs at 1 (F = 3.53; df = 3,95; P = 0.018) and 2 (F = 3.63; df = 3,95; P = 0.016) WAT, respectively. In general, the three biopesticides caused an average decrease of 60% and 40% in nymph and egg cumulative individuals at the end of the experiment (4 WAT), respectively.

In test 2, the percent efficacy of the treatments (*B. bassiana* and commercial extracts of *Sophora* spp. and garlic-chili pepper) was also evaluated at 2 and 4 WAT. Foliar biopesticides sprays were equally effective on suppressing ACP adults, nymphs, and eggs, showing a percent efficacy of around 40% in the management of ACP individuals (adults, nymphs, and eggs) for all treatments (Fig. 7).

The use of *B. bassiana* showed its potential as a biological control agent of *D. citri*, with a nymph control percentage between 50% and 60% in both tests. Several authors have reported similar results under laboratory conditions (Ferreira Pinto et al., 2012; Lezama-Gutiérrez et al., 2012). Padulla and Alves (2009) found a control percentage of ≈80% after 7 d of exposure under laboratory conditions. Saldarriaga-Ausique et al. (2017) estimated mortality levels of ≈50% under commercial conditions in the dry season of the year due to the influence of temperature and relative humidity. Padulla and Alves (2009) and Ferreira Pinto et al. (2012) reported that the mycotoxin oosporin is related to nymph mortality, affecting mainly third-instar nymphs. Our results did not show any differences on ACP adult control. Saldarriaga-Ausique et al. (2017) found 80% mortality of adults, and reported that the high mortality rate is mainly related to environmental (rainfall, temperature, ultraviolet radiation) and technical (application number, rate) conditions. A plausible explanation for the lack of ACP adult control by foliar *B. bassiana* sprays could be that weather conditions (low rainfall and high daytime temperatures) during trials may have conditioned proper ACP colonization by oospores (Figs. 1 and 2).

Use of botanical insecticides could reduce ACP populations, insecticide resistance, and/or negative impact on ACP natural predators (Khan et al., 2014). The application of commercial extracts of *Sophora* spp. and garlic-chili pepper also had noteworthy effects in keeping the populations of immature ACP low (Table 1). Previous studies by

Fig. 5. Mean of ACP adults (A), nymphs (B), and eggs (C) per flush shoot in Tahiti lime trees treated with three commercial biopesticides (*Sophora* spp., garlic-chili pepper concentrated emulsion, or *B. bassiana*). Points represent the mean of four values $\pm$ se. NS: No significance; ***: significant difference ($P \leq 0.001$); **: significant difference ($P \leq 0.01$); *: significant difference ($P \leq 0.05$) compared with the control according to the Tukey test. Arrows indicate foliar commercial biopesticide applications.

Table 2. Probability of F statistic ($P > F$) of cumulative ACP individuals (adults, nymphs, and eggs) on Tahiti lime trees treated with three commercial biopesticides.

		Weeks after treatments (WAT)				
		0 ²	1	2	3	4
		<i>Adults</i>				
Farms (F)	<i>P</i> = 0.739	<i>P</i> = 0.254	<i>P</i> = 0.302	<i>P</i> = 0.164	<i>P</i> = 0.210	
Treatments (T)	<i>P</i> = 0.924	<i>P</i> = 0.558	<i>P</i> = 0.635	<i>P</i> = 0.616	<i>P</i> = 0.717	
F × T	<i>P</i> = 0.517	<i>P</i> = 0.072	<i>P</i> = 0.562	<i>P</i> = 0.413	<i>P</i> = 0.217	
		<i>Nymphs</i>				
Farms (F)	<i>P</i> = 0.336	<i>P</i> = 0.212	<i>P</i> = 0.692	<i>P</i> = 0.327	<i>P</i> = 0.217	
Treatments (T)	<i>P</i> = 0.135	<i>P</i> = 0.018	<i>P</i> = 0.000	<i>P</i> = 0.000	<i>P</i> = 0.000	
F × T	<i>P</i> = 0.396	<i>P</i> = 0.203	<i>P</i> = 0.262	<i>P</i> = 0.409	<i>P</i> = 0.311	
		<i>Eggs</i>				
Farms (F)	<i>P</i> = 0.003	<i>P</i> = 0.000	<i>P</i> = 0.000	<i>P</i> = 0.000	<i>P</i> = 0.000	
Treatments (T)	<i>P</i> = 0.051	<i>P</i> = 0.242	<i>P</i> = 0.016	<i>P</i> = 0.006	<i>P</i> = 0.000	
F × T	<i>P</i> = 0.089	<i>P</i> = 0.177	<i>P</i> = 0.464	<i>P</i> = 0.339	<i>P</i> = 0.180	

^aFoliar applications of biopesticides were conducted at 0 and 2 weeks after initiating the treatments.

Fig. 6. Cumulative average of ACP adults (A), nymphs (B), and eggs (C) per flush shoot in Tahiti lime trees treated with three commercial biopesticides (*Sophora* spp., garlic-chili pepper concentrated emulsion, or *B. bassiana*). Points represent the mean of four values $\pm$ SE. NS No significance; ***significant difference ($P \leq 0.001$); **significant difference ($P \leq 0.01$); *significant difference ($P \leq 0.05$) compared with the control according to the Tukey test. Foliar treatment sprays were carried out at 0 weeks after treatments.

Fig. 7. Summary of the efficiency percentage per flush shoot in Tahiti lime trees treated with three commercial biopesticides (*Sophora* spp., garlic-chili pepper concentrated emulsion, or *B. bassiana*) at 2 [adults (A), nymphs (C), and eggs (E)] and 4 [adults (B), nymphs (D), and eggs (F)] week after treatments (WAT). Bar charts represent the mean of four values $\pm$ SE. NS No significance at $P \leq 0.05$. Foliar treatment sprays were carried out at 0 and 2 weeks after treatments.

Mann et al. (2011) and Cazares-Alonso et al. (2014) also indicated that garlic extracts show repellent activity and inhibition of ACP. Garlic extracts contain compounds such as trisulfites, which are known as insect-repellent chemicals (Mann et al., 2011). On the other hand, chili pepper contains capsaicin, which has lethal, antifeeding, and/or repellent effects on invertebrates (Baidoo and Mochiah, 2016; Kazem and El-Shereif, 2010). Regarding the use of the commercial extracts based on alkaloids, studies by Khan et al. (2013) and (2014) showed that the use of plant alkaloids such as tropine caused nymph mortality higher than 50% in laboratory and field conditions. Also, Zanardi et al. (2015) reported the positive effects on ACP control with the use of another plant alkaloid, matrine. These conclusions are in accordance with our findings in which *Sophora* (matrine) sprays caused a reduction of over 50% in ACP nymphs. A plausible explanation for ACP nymph reduction in our study could be that plant extract alkaloids affect the production of acetylcholine by the insect and cause an antifeeding effect since they are applied on the plant surface (Ali et al., 2017; Khan et al., 2013).

The development of sustainable strategies in commercial citrus orchards is of vital importance for ACP management and spread in Colombia. The application of commercial biopesticides based mainly on plant extracts or entomopathogenic fungi is a strategy that can be used to minimize the use of chemical synthesis insecticides within an IPM program, especially in tropical dry forest habitats, since these products reduced mainly ACP immatures (nymphs and eggs). Long-term studies are necessary to determine the potential use of *B. bassiana* and plant extracts in different habitats.

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