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ORIGINAL ARTICLE

Botanicals, selective insecticides, and predators to control *Diaphorina citri* (Hemiptera: Liviidae) in citrus orchards

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Abstract The Asian citrus psyllid (ACP) *Diaphorina citri* Kuwayama vectors pathogens that cause huanglongbing (HLB) or citrus greening devastating and economically important disease present in most citrus growing regions. Young citrus shoots are required for psyllid reproduction and development. During winter citrus trees produce little or no new growth. Overwintering adults reproduce in spring on newly emerging shoots also attractive to other pests and beneficial insects. Botanicals and relatively selective insecticides could help to conserve beneficial insects and reduce pest resistance to insecticides. Sprays of Azadirachtin (Neem), Tropane (Datura), Spirotetramat, Spinetoram, and broad-spectrum Imidacloprid were evaluated to control ACP in spring and summer on 10-year-old "Kinow" *Citrus reticulata* Blanco trees producing new growth. Psyllid populations were high averaging 5–9 nymphs or adults per sample before treatment application. Nymphs or adults were significantly reduced to 0.5–1.5 per sample in all treatments for 3 weeks, average 61%–83% reduction. No significant reduction in ladybeetles *Adalia bipunctata*, *Anegleі scardoni*, *Cheilomenes sexmaculata*, and *Coccinella septempunctata* was observed. Syrphids, spiders and green lacewings were reduced in treated trees except with Tropane. Studies are warranted to assess impact of these predators on ACP and interaction with insecticides. Observed reduction in ACP populations may not be enough considering its reproductive potential and role in the spread of HLB. Follow-up sprays may be required to achieve additional suppression using rotations of different insecticides.

Key words Asian citrus psyllid; biopesticides; Coccinellids; huanglongbing; predators

Introduction

The Asian citrus psyllid (ACP), *Diaphorina citri* Kuwayama (Hemiptera: Liviidae), is a serious pest of citrus and vector of a fatal disease called "huanglongbing" (HLB) or citrus greening (Halbert & Manjunath, 2004; Bove, 2006). An extensive survey carried out in India revealed that 83%–95% of 25% yield losses in citrus were associated with *D. citri* through direct damage or sooty mold fungus (Shivankar & Singh, 2006). It has been reported that up to 20% of trees in poorly managed orchards in China were lost to HLB within few years of planting, and due to rapid spread of the disease most orchards lost their commercial value within 7–8 years of planting (Aubert, 1990). Both nymphs and adults of ACP feed voraciously on young shoots of citrus causing death of highly infested terminals.

The presence of ACP and HLB in Pakistan and elsewhere pose a serious threat to citrus production (Ahmad *et al.*, 2004). Therefore, development of sustainable pest and disease management strategies is critical. *D. citri*

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require newly opening buds and young shoots to develop and reproduce. Soil applied systemic insecticides are more common in young trees and provide good control of ACP (Sétamou *et al*., 2010; Stansly & Kostyk, 2012, 2013). Foliar sprays of broad-spectrum insecticides applied during tree dormancy kill the majority of overwintering adults leaving few to infest new shoots in spring (Qureshi & Stansly, 2010). During the growing season foliar sprays of synthetic and organic insecticides are used for additional suppression of ACP (Qureshi *et al.*, 2010, 2011, 2012a, 2013a; Rogers *et al*., 2012) and there are reports of their negative impact on beneficial insects which contribute to biological control of ACP and other pests (Qureshi & Stansly, 2007; Qureshi *et al*., 2009a,b; Qureshi & Stansly, 2009). There is also evidence of psyllid resistance against some commonly used insecticides (Tiwari *et al*., 2011), suggesting that lower use of hard chemistry insecticides and similar modes of action is important for sustainable citrus pest and disease management. Use of biological control, botanicals, and selective insecticides could contribute to reduce ACP and other pests and risk of insecticide resistance.

Insecticides obtained from natural sources help to reduce environmental pollution, conserve natural enemies of pests, and ward off pest resurgence. For the same reason use of biopesticides with selectivity against phytophagous insects has increased recently (Tengerdy & Szakacs, 1998; Rausell *et al*., 2000). Biodegradable and insecticidal liminoid azadirachtin formulated from neem tree *Azadirachta indica* A. juss, is effective against a wide range of insect pests including brown citrus aphid, *Toxoptera citricida* (Kirkaldy), root weevil, *Diaprepes abbreviates* (L.), and western flower thrips, *Frankliniella occidentalis* (Isman, 1999; Tang *et al*., 2002; Thoeming *et al*., 2003; Weathersbee & McKenzie, 2005). Azadirachtin at 4.5% provided a repellent effect against ACP adults in choice tests although no preference for oviposition was observed (Weathersbee & McKenzie, 2005). At a concentration of 22.5 ppm azadirachtin, ecdysis was not observed past 4 d after treatment (DAT) and all nymphs were dead within 7 d. When treated plants were exposed to a greenhouse population of ACP nymphs were significantly reduced by concentrations as low as 10 ppm azadirachtin (Weathersbee & McKenzie, 2005).

Species specific parasitoid *Tamarixi aradiata* Waterston which attack psyllid nymphs is an important contributor to psyllid mortality in different regions, including France (Réunion), Puerto Rico, USA (Florida), China, and Pakistan (Pluke *et al*., 2008; Qureshi *et al*., 2009b, 2012b; Barr *et al*., 2009; Qureshi & Stansly, 2010). In addition, ladybeetles and other predators such as spiders, cockroaches, and lacewings have been observed as major contributors to reduction of psyllid populations in different regions. Michaud (2004) reported *Harmonia axyridis* Pallas, *Olla v-nigrum* Mulsant, *Cycloneda sanguinea* L., and *Exochomus children* Mulsant as key predators of *D. citri* in Florida. Later, Qureshi and Stansly (2009) observed more than 90% mortality of ACP in Florida citrus attributed mainly to the ladybeetles *O. v-nigrum*, *Curinus coeruleus*, *H. axyridis*, and *C. sanguinea*. Pluke *et al.* (2005) reported ladybeetles *Coelophora inaequalis* F., *C. sanguinea*, *Cladis nitidula* F., *Chilocorus cacti* L., *Coleomegilla innonata* Mulsant, *Scymnus* sp. *Hippodamia convergens* Guerin, and *Cryptolaemus montrouzieri* Mulsant from citrus in Puerto Rico, however, only *C. inaequalis* and *C. sanguinea* were common. These findings suggest that natural enemies particularly ladybeetles could be an important component of ACP and HLB management.

Studies conducted at Faisalabad, Pakistan, identified 22 species of predaceous ladybeetles from crops and forest habitat of Pakistan (Zahoor *et al*., 2003) some of which could be feeding on ACP. Other generalist predators such as lacewings, spiders, syrphid flies are also common in citrus and other habitats and could contribute to control ACP and other pests of citrus (Michaud, 2004; Qureshi & Stansly, 2008, 2009). Therefore, use of biological control compatible insecticides effective against ACP could improve citrus pest management. We evaluated the impact of insecticidal sprays of botanical and relatively selective insecticides on *D. citri* and generalist predators on bearing citrus trees.

Materials and methods

Site selection

Field experiments were conducted in 3 citrus orchards of progressive farmers one each at 38-SB, 53-NB, and Risala No.5 in Sargodha (32.1506°N. 72.6454°E), District of Pakistan during 2010–2011. The orchards were planted in years 2002–2004 at 247–494 plants per hectare using the "Kinow" *Citrus reticulata* Blanco plants from nurseries maintained by the Citrus Research Institute Sargodha, Agriculture Department of Punjab. Orchard in 36-SB is at east, 53-NB at north–west, and Risla No.5 at west of main city of Sargodha and were 20–30 km apart.

Preparation of spray solutions

To prepare extracts of neem, *Azadirachta indica* A. juss, and Datura, *Datura alba* Nees, 100 g powder of each plant obtained using dry leaves was mixed in 1 L of water

Table 1 List of insecticides and their application rates used in the experiments.

Active ingredient	Plant/product name	Application rate (product/1 L water)	Company name
Azadiractin 100 g	Neem extract	20 mL	Self-Made (Sridhar & Vijayalakshami, 2002)
Tropane 100 g	Datura extract	20 mL	Self-Made (Dawar *et al*., 2010)
Spinetoram 11.7%	Radiant® 120SC	0.40 mL	Dow AgroSciences
Spirotetramat 240 g	Movento® 240SC	1.25 mL	Bayer CropScience
Imidacloprid 20%	Confidor® 20% SC	0.40 mL	Bayer CropScience

to prepare 100% solution. These solutions were kept in cocked bottles at 40 °C (Sridhar & Vijayalakshmi, 2002; Dawar *et al*., 2010). Other insecticides were obtained from registered dealers of respective companies listed in Table 1. All spray solutions were prepared in tap water. Treatments and application rates are provided in Table 1.

Experimental design and treatment application

A Randomized Complete Block Design (RCBD) with orchards as blocks was used. Five treatments and an untreated control were randomly distributed across 18 plants in each of 3 orchards. Eight- to ten-year-old and 6–8 feet tall trees were used and 20% of the canopy was pruned to stimulate new growth and psyllid infestation. Treatments of Azadirachtin (Neem extract), Tropane (Datura extract), Spinetoram (Radiant®, Dow AgroSciences, 9330 Zionsville Road, Indianapolis, IN 46268, USA), Spirotetramat (Movento®, Bayer CropScience, Plot No. 23, Sector 22, Korangi Industrial Area, Karachi, PAKISTAN), and broad-spectrum Imidacloprid (Confidor®, Bayer CropScience, Plot No. 23, Sector 22, Korangi Industrial Area, Karachi, PAKISTAN) were evaluated twice in summer and spring. Applications for 2010 summer experiments 1 and 2 were made on July 17 and August 26, respectively. For 2011 spring experiments 1 and 2 applications were made on February 12 and March 19, respectively. In all experiments, a backpack power sprayer China made model No. TF-70 with Turbo T-jet wide angle spray tip nozzle delivering 2 L/tree was used.

Data collection and analysis

Four randomly selected young shoots viable to psyllid infestation were selected on each tree 1 on each side and number of ACP nymphs and predators counted from top 100 mm of each shoot (Ahmed *et al*., 2004). For nymphal counts selected shoots were collected and examined under stereomicroscope in the laboratory. *D. citri* adults were sampled using stem tap method (Qureshi & Stansly 2007, 2010). Density of psyllid adults was estimated from each tree by counting adults falling on a clipboard covered with a 22 × 28 cm laminated white sheet held horizontally under randomly chosen branches, which were then struck 3 times with a PVC pipe to make a count for 1 "tap" sample. Four tap samples were conducted per tree 1 on each side. Averages of 4 shoots or tap samples per tree were used for analysis. Pretreatment data was recorded 24 h prior to application of insecticides, while posttreatment data recorded at 2, 12, and 22 DAT. Same procedures were used in all experiments.

Univariate procedure was used to analyze data for the assumptions of parametric analysis (SAS Institute, 2004). Data with normal distributions were subjected to ANOVA using the GLM procedure to evaluate treatment effects on ACP, and treatment means were separated using LSD, contingent on a significant treatment effect ($P < 0.05$) (SAS Institute, 2004). Nonnormal data were analyzed by using the nonparametric Kruskal–Wallis test.

Results

Effects on ACP

Summer 2010 Experiment 1: Before application of treatments on July 17 there were no significant differences in the distribution of nymphs or adults on the trees. Means of 5.9 ± 0.3 nymphs per shoot and 6.1 ± 0.4 adults per tap sample were observed 24 h prior to treatment application. All treatments provided significant reduction in ACP nymphs compared to control through 22 DAT (2 DAT: $F = 6.22$, df = 5, $P = 0.0002$; 12 DAT: $H = 17.66$, df = 5, $P = 0.0034$; 22 DAT: $H = 11.72$, df = 5, $P = 0.0389$) except Tropane at 12 and 22 DAT and Azadirachtin at 22 DAT (Table 2). More nymphal reduction was observed with Spinetoram than Azadirachtin at 2 DAT and with Azadirachtin, Spinetoram, Spirotetramat, and Imidacloprid than Tropane at 12 DAT ($P < 0.05$). Significant reduction in adults was observed only at 12 DAT ($H = 22.40$, df = 5, $P = 0.0004$) in all treatments except Tropane and most with Azadirachtin than others which were statistically similar (Table 2).

Table 2 Mean (± SEM) number of Asian citrus psyllid nymphs/shoot and adults/tap sample in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on July 17, 2010 and sampled at 2, 12, and 22 d after treatment (DAT).

Treatment	Nymphs per shoot†			Adults per tap sample‡		
	2 DAT	12 DAT	22 DAT	2 DAT	12 DAT	22 DAT
Imidacloprid (Confidor®)	1.67 ± 0.33 bc	1.11 ± 0.26 c	0.67 ± 0.33 b	1.78 ± 0.32 a	1.22 ± 0.15 b	1.56 ± 0.29 a
Spirotetramat (Movento®)	1.67 ± 0.24 bc	0.89 ± 0.26 c	0.78 ± 0.36 b	2.33 ± 0.37 a	1.22 ± 0.15 b	1.33 ± 0.44 a
Spinetoram (Radiant®)	1.33 ± 0.24 c	1.44 ± 0.41 bc	0.78 ± 0.36 b	2.67 ± 0.33 a	1.22 ± 0.15 b	0.89 ± 0.31 a
Azadirachtin (Neem)	2.56 ± 0.67 b	1.00 ± 0.29 c	1.22 ± 0.15 ab	1.44 ± 0.41 a	0.33 ± 0.17 c	1.56 ± 0.18 a
Tropane (Datura)	1.56 ± 0.29 bc	2.89 ± 0.45 a	1.22 ± 0.15 ab	1.44 ± 0.38 a	1.77 ± 0.28 ab	1.67 ± 0.24 a
Control	4.00 ± 0.47 a	2.22 ± 0.43 ab	1.78 ± 0.22 a	2.11 ± 0.51 a	2.00 ± 0.37 a	2.11 ± 0.31 a

†Four shoots examined per tree and average per shoot analyzed.
‡Four tap samples conducted per tree and average per tap sample analyzed.
Means in a column sharing a common letter are not significantly different ($P > 0.05$).

Table 3 Mean (± SEM) number of Asian citrus psyllid nymphs/shoot and adults/tap sample in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on August 26, 2010 and sampled at 2, 12, and 22 d after treatment (DAT).

Treatment	Nymphs per shoot†			Adults per tap sample‡		
	2 DAT	12 DAT	22 DAT	2 DAT	12 DAT	22 DAT
Imidacloprid (Confidor®)	1.67 ± 0.17 b	1.33 ± 0.33 b	1.00 ± 0.24 b	1.89 ± 0.31 b	1.11 ± 0.11 c	1.44 ± 0.29 b
Spirotetramat (Movento®)	1.44 ± 0.38 b	0.89 ± 0.20 b	1.22 ± 0.43 b	1.56 ± 0.41 b	0.78 ± 0.32 c	0.44 ± 0.18 c
Spinetoram (Radiant®)	1.78 ± 0.22 b	1.22 ± 0.15 b	1.11 ± 0.39 b	1.44 ± 0.18 b	1.33 ± 0.24 bc	0.89 ± 0.26 bc
Azadirachtin (Neem)	2.00 ± 0.29 b	1.11 ± 0.26 b	1.78 ± 0.52 b	1.44 ± 0.18 b	2.22 ± 0.32 b	0.78 ± 0.22 bc
Tropane (Datura)	1.56 ± 0.24 b	1.67 ± 0.33 b	1.33 ± 0.41 b	1.44 ± 0.24 b	1.78 ± 0.28 bc	0.67 ± 0.17 bc
Control	3.89 ± 0.56 a	3.00 ± 0.47 a	3.00± 0.41 a	7.33 ± 2.52 a	3.78 ± 0.74 a	4.00 ± 0.65 a

†Four shoots examined per tree and average per shoot analyzed.
‡Four tap samples conducted per tree and average per tap sample analyzed.
Means in a column sharing a common letter are not significantly different ($P > 0.05$).

Experiment 2: At 24 h prior to treatment application on August 26 means of 5.7 ± 0.4 nymphs per shoot and 9.1 ± 0.7 adults per tap sample were observed on experimental trees without any significant difference. Nymphal reduction compared to control was significant with all treatments through 22 DAT (2 DAT: $F = 7.45$, df = 5, $P = < 0.0001$; 12 DAT: $F = 6.06$, df = 5, $P = 0.0002$; 22 DAT: $H = 12.40$, df = 5, $P = 0.0297$) without any statistical differences among treatments (Table 3). All treatments reduced adults significantly through 22 DAT (2 DAT: $H = 19.59$, df = 5, $P = 0.0015$; 12 DAT: $H = 25.53$, df = 5, $P = 0.0001$; 22 DAT: $H = 29.37$, df = 5, $P \leq 0.0001$, Table 3). There were no differences among treatments at 2 DAT. More reduction was observed with Spirotetramat and Imidacloprid than Azadirachtin at 12 DAT and with Spirotetramat than Imidacloprid at 22 DAT.

Spring 2011 Experiment 1: At 24 h before treatment application on February 12 means of 5.8 ± 0.3 nymphs per shoot and 5.9 ± 0.3 adults per tap sample were observed on experimental trees. All treatments provided significant reduction in ACP nymphs compared to control through 22 DAT (2 DAT: $H = 24.15$, df = 5, $P = 0.0002$; 12 DAT: $H = 27.91$, df = 5, $P \leq 0.0001$; 22 DAT: $H = 16.50$, df = 5, $P = 0.0055$) with somewhat similar effectiveness except at 12 DAT when more reduction in Imidacloprid, Spirotetramat, and Spinetoram treatments than Tropane was observed but not the Azadirachtin which was not different in effectiveness from Tropane (Table 4). Significant reduction in adults compared to control was also observed through 22 DAT in all treatments without any statistical difference among treatments (2 DAT: $F = 18.29$, df = 5, $P \leq 0.0001$; 12 DAT: $H = 26.49$, df = 5, $P \leq 0.0001$; 22 DAT: $H = 19.03$, df = 5, $P = 0.0019$, Table 4).

Experiment 2: Means of 6.0 ± 0.3 nymphs per shoot and 8.9 ± 0.7 adults per tap sample were observed 24 h prior to treatment application on March 19. Compared to control significant reduction in nymphs

Table 4 Mean (± SEM) number of Asian citrus psyllid nymphs/shoot and adults/tap sample in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on February 12, 2011 and sampled at 2, 12, and 22 d after treatment (DAT).

Treatment	Nymphs per shoot†			Adults per tap sample‡		
	2 DAT	12 DAT	22 DAT	2 DAT	12 DAT	22 DAT
Imidacloprid (Confidor®)	0.89 ± 0.26 b	0.89 ± 0.26 c	1.33 ± 0.33 b	1.22 ± 0.40 b	0.67 ± 0.24 b	1.33 ± 0.24 b
Spirotetramat (Movento®)	1.22 ± 0.40 b	0.89 ± 0.26 c	0.89 ± 0.26 b	1.44 ± 0.29 b	1.22 ± 0.32 b	1.33 ± 0.24 b
Spinetoram (Radiant®)	1.56 ± 0.34 b	0.78 ± 0.28 c	1.00 ± 0.24 b	1.78 ± 0.28 b	0.89 ± 0.20 b	0.89 ± 0.26 b
Azadirachtin (Neem)	1.44 ± 0.29 b	1.22 ± 0.28 bc	1.11 ± 0.26 b	0.89 ± 0.31 b	0.78 ± 0.15 b	1.33 ± 0.17 b
Tropane (Datura)	1.44 ± 0.29 b	1.89 ± 0.35 b	1.33 ± 0.33 b	1.22 ± 0.28 b	1.67 ± 0.47 b	1.33 ± 0.41 b
Control	4.22 ± 0.32 a	4.33 ± 0.41 a	4.33 ± 0.65 a	4.78 ± 0.43 a	5.00 ± 0.80 a	3.67 ± 0.44 a

†Four shoots examined per tree and average per shoot analyzed.
‡Four tap samples conducted per tree and average per tap sample analyzed.
Means in a column sharing a common letter are not significantly different ($P > 0.05$).

Table 5 Mean (± SEM) number of Asian citrus psyllid nymphs/shoot and adults/tap sample in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on March 19, 2011 and sampled at 2, 12, and 22 d after treatment (DAT).

Treatment	Nymphs per shoot†			Adults per tap sample‡		
	2 DAT	12 DAT	22 DAT	2 DAT	12 DAT	22 DAT
Imidacloprid (Confidor®)	1.56 ± 0.24 b	1.22 ± 0.36 b	1.00 ± 0.40 b	1.22 ± 0.32 b	0.89 ± 0.20 b	1.22 ± 0.28 b
Spirotetramat (Movento®)	1.33 ± 0.33 b	1.00 ± 0.17 b	0.67 ± 0.24 b	0.89 ± 0.20 b	0.56 ± 0.18 b	1.11 ± 0.39 b
Spinetoram (Radiant®)	1.33 ± 0.29 b	1.11 ± 0.26 b	1.11 ± 0.11 b	1.33 ± 0.24 b	1.11 ± 0.20 b	1.11 ± 0.20 b
Azadirachtin (Neem)	1.89 ± 0.61 b	0.89 ± 0.20 b	1.44 ± 0.44 b	0.67 ± 0.17 b	1.33 ± 0.37 b	0.67 ± 0.24 b
Tropane (Datura)	1.56 ± 0.60 b	0.78 ± 0.28 b	1.67 ± 0.58 b	1.67 ± 0.29 b	1.67 ± 0.44 b	1.00 ± 0.24 b
Control	4.56 ± 0.53 a	3.78 ± 0.36 a	4.22 ± 0.40 a	5.67 ± 0.67 a	3.56 ± 0.71 a	4.11 ± 0.26 a

†Four shoots examined per tree and average per shoot analyzed.
‡Four tap samples conducted per tree and average per tap sample analyzed.
Means in a column sharing a common letter are not significantly different ($P > 0.05$).

(2 DAT: $H = 18.67$, df = 5, $P = 0.0022$; 12 DAT: $H = 24.43$, df = 5, $P = 0.0002$; 22 DAT: $F = 11.87$, df = 5, $P \leq 0.0001$) and adults (2 DAT: $F = 27.88$, df = 5, $P \leq 0.0001$; 12 DAT: $F = 7.21$, df = 5, $P \leq 0.0001$; 22 DAT: $H = 25.87$, df = 5, $P \leq 0.0001$) was observed through 22 DAT in all treatments without statistical differences among treatments (Table 5). No phytotoxicity was observed in any of the 4 experiments.

Effects on predators Four ladybeetle species observed in the experiments included *Coccinella septempunctata*, *Adalia bipunctata*, *Cheilomenes sexmaculata*, and *Anegleis cardoni* and were grouped together under Coccinellid adults or larvae. Syrphids, spiders, and green lacewing were also observed and grouped as other predators. No significant negative effect of the treatments on the number of ladybeetles was observed when compared between treated and untreated trees or between trees treated with different insecticides (Figs. 1–4, $P > 0.05$). Some negative effects of the treatments applied on July 17 were observed at 22 DAT on the predatory group which included syrphids, spiders, and green lacewing ($F = 5.88$, df = 5, $P = 0.0003$). Fewer of these 3 predators were observed in the treatments of Imidacloprid and Spirotetramat than control or in Imidacloprid, Spinetoram, Spirotetramat, and Azadirachtin than Tropane (Fig. 1). There was no significant difference among these later 4 treatments or between Spinetoram, Azadirachtin and control in the numbers of predators observed. In another experiment conducted on February 12 numbers of syrphids, spiders, and green lacewing were again reduced in the treatments of Imidacloprid and Spirotetramat and also in Azadirachtin compared to control at 22 DAT ($F = 2.57$, df = 5, $P = 0.0384$, Fig. 3). Reduction with Spirotetramat was also significantly more compared to Tropane at the same time.

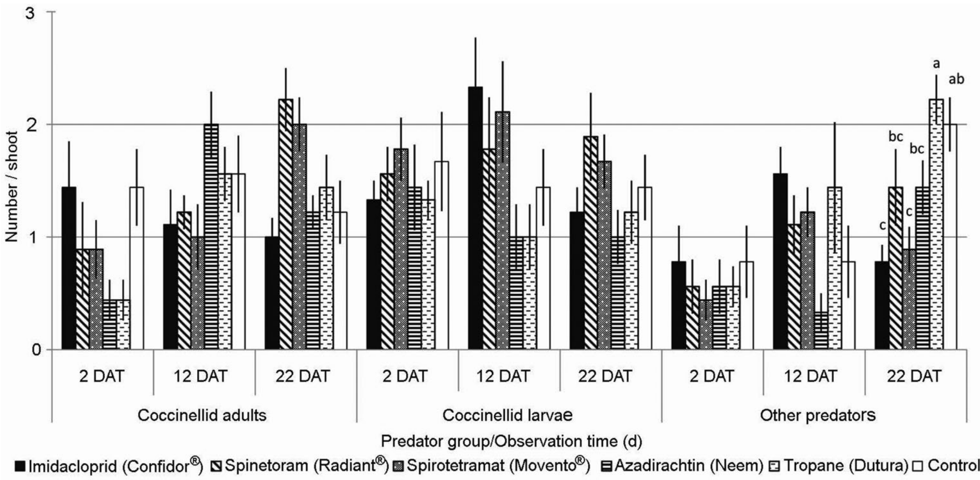

Fig. 1 Mean (± SEM) number per shoot of Coccinellids (larvae, adults) and other predators in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on July 17, 2010 and sampled at 2, 12, and 22 d after treatment (DAT). The shoots observed were same that were selected for the nymphs of *D. citri*. Coccinellids included *Coccinella septempunctata*, *Adalia bipunctata*, *Cheilomenes sexmaculata*, and *Anegleis cardoni*. Other predators included syrphids, spiders, and green lacewings. Treatment means for a predator group were not significantly different at any observation time ($P > 0.05$) except for other predators group at 22 DAT ($P < 0.05$). Bars sharing a common letter represent means that were not significantly different ($P > 0.05$).

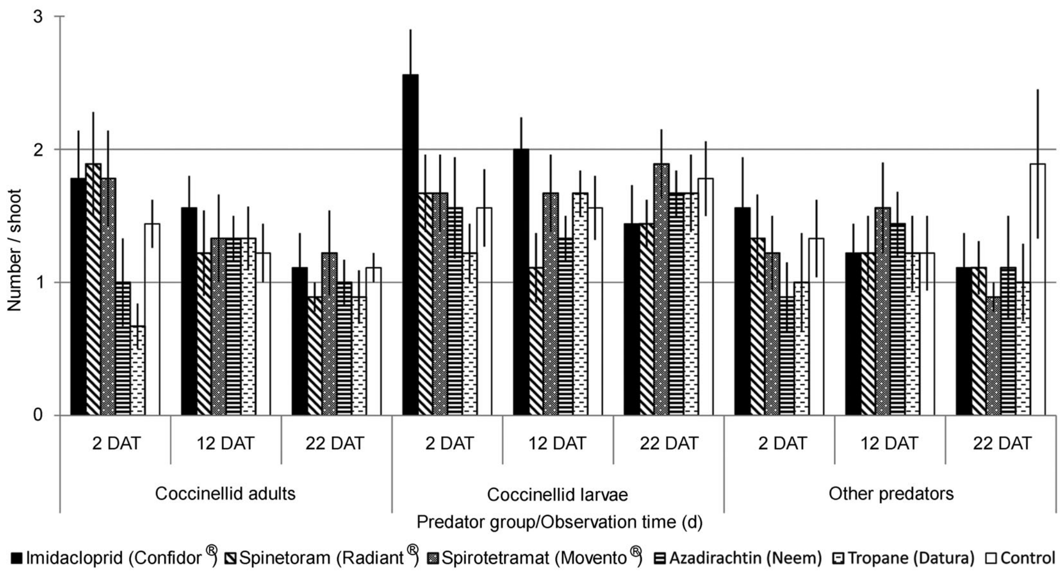

Fig. 2 Mean (± SEM) number per shoot of Coccinellids (larvae, adults) and other predators in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on August 26, 2010 and sampled at 2, 12, and 22 d after treatment (DAT). The shoots observed were same that were selected for the nymphs of *D. citri*. Coccinellids included *Coccinella septempunctata*, *Adalia bipunctata*, *Cheilomenes sexmaculata*, and *Anegleis cardoni*. Other predators included syrphids, spiders, and green lacewings. Treatment means for a predator group were not significantly different at any observation time ($P > 0.05$).

Discussion

Sprays of Imidacloprid, Spirotetramat, Spinetoram, Azadirachtin, and Tropane targeted at flushing "Kinow" citrus trees in spring or summer provided 61%–83% reduction in nymphs and adults of ACP lasting for 3 weeks compared to untreated control trees indicating that they all were very useful in reducing ACP populations. The only

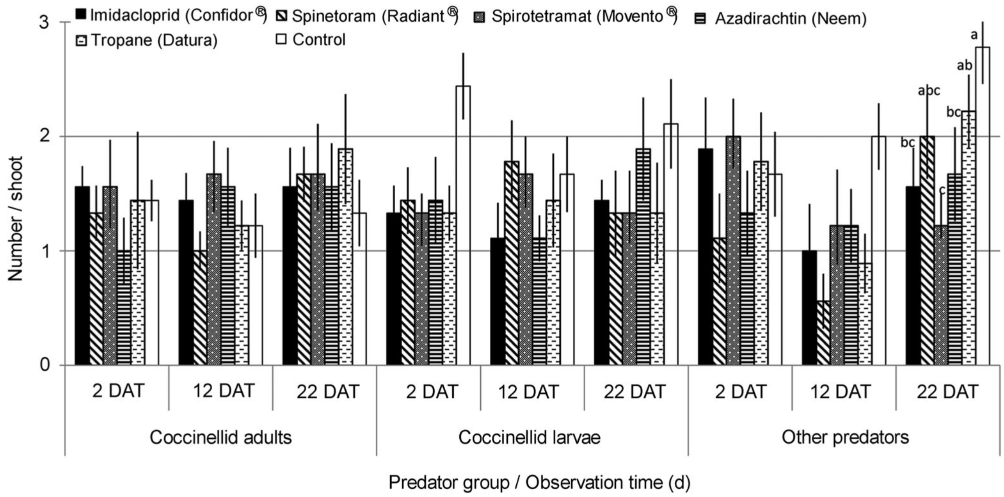

Fig. 3 Mean (± SEM) number per shoot of Coccinellids (larvae, adults) and other predators in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on February 12, 2011 and sampled at 2, 12, and 22 d after treatment (DAT). The shoots observed were same that were selected for the nymphs of *D. citri*. Coccinellids included *Coccinella septempunctata*, *Adalia bipunctata*, *Cheilomenes sexmaculata*, and *Anegleis cardoni*. Other predators included syrphids, spiders, and green lacewings. Treatment means for a predator group were not significantly different at any observation time ($P > 0.05$) except for other predators group at 22 DAT ($P < 0.05$). Bars sharing a common letter represent means that were not significantly different ($P > 0.05$).

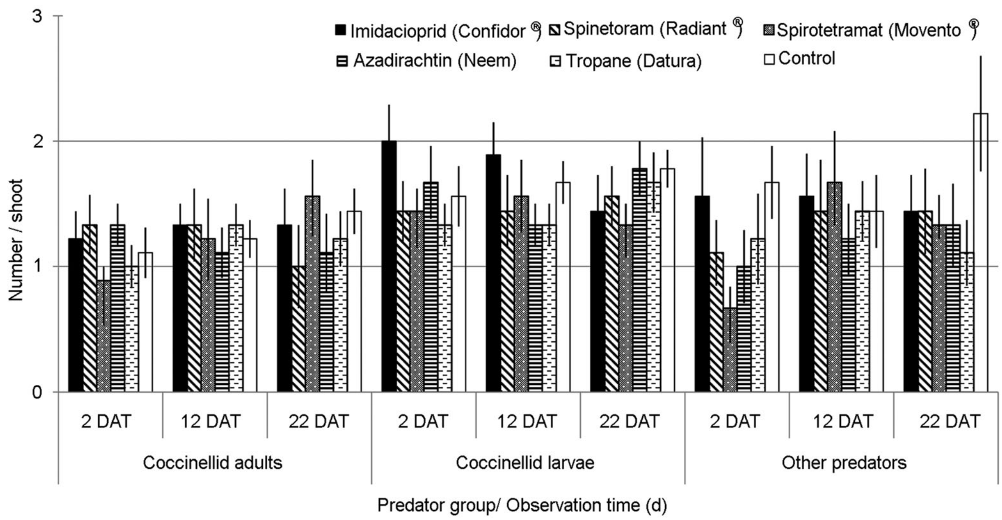

Fig. 4 Mean (± SEM) number per shoot of Coccinellids (larvae, adults) and other predators in 10-year old "Kinow" orange trees that were untreated or treated with foliar sprays of insecticides on March 19, 2011 and sampled at 2, 12, and 22 d after treatment (DAT). The shoots observed were same that were selected for the nymphs of *D. citri*. Coccinellids included *Coccinella septempunctata*, *Adalia bipunctata*, *Cheilomenes sexmaculata*, and *Anegleis cardoni*. Other predators included syrphids, spiders, and green lacewings. Treatment means for a predator group were not significantly different at any observation time ($P > 0.05$).

exception of reduced effectiveness of the treatments was observed in the experiment conducted on July 17, when nymphal reduction was not significant with Tropane at 12 and 22 DAT and with Azadirachtin at 22 DAT. Adult reduction with Tropane was not significant and lasted only through 12 DAT with other treatments. Number of adult ACP dropped unexpectedly in the untreated trees soon after the start of the experiment resulting in mean difference of one or less psyllids with treated trees at 2 DAT compared to other 3 experiments where difference was

3–6 psyllids. More activity of biological control in the untreated trees could be associated with psyllid reduction but same was not observed in the other experiments.

Effects of Azadirachtin on ACP were also comparable to those of Imidacloprid, Spirotetramat, and Spinetoram in the laboratory study (Khan *et al*., 2013). Weathersbee and McKenzie (2005) observed significant effect of Azadirachtin on ACP at low concentration under controlled conditions. However, Azadirachtin 1.2% provided only 25% mortality in adults although effects on the nymphs were more pronounced resulting 100% mortality in 72 h under controlled conditions (Qureshi *et al*., 2013b). A similar trend was observed in the field where nymphs were reduced for about 2 weeks but effect on adults was not statistically significant (Stansly *et al*., 2009). Effectiveness of neem-based products is also demonstrated in controlling some other phloem-feeding insect pests including aphids such as brown citrus aphid which is vector of citrus tristeza virus (Lowery *et al*., 1993; Tang *et al*., 2002) and the pistachio psyllid, *Agonoscena targionii* (Lisht.) (Lababidi, 2002). Tropane another botanical insecticide tested in this study was also effective against ACP although little less compared to Azadirachtin. Kuganathan and Ganeshalingam (2011) evaluated the acute toxicity of varying concentrations of Datura metel on grasshoppers and red ants and found EC_{50} values of 12 000 and 11 600 ppm, respectively. Percentage mortality increased from 20% to 60% with increasing concentrations confirming the insecticidal potential of Datura.

Effectiveness of Spinetoram and Spirotetramat that we observed against ACP was also demonstrated in other studies (Qureshi *et al*., 2009a, 2010, 2011, 2012a). Similarly effective control of ACP with broad-spectrum Imidacloprid is also shown by others, however, rates allowed are better suited for drench application in young trees to provide even more and extended suppression (Qureshi & Stansly, 2007; Qureshi *et al*., 2009a; Stansly & Kostyk, 2012, 2013). Application of an insecticide both as spray and soil application makes it more viable to development of resistance in pests. Therefore, rotation with other insecticides is important to conserve such chemistries. Currently, the insecticides that provide extended suppression of ACP in young trees are all neonicotinoids including imidacloprid with same 4A mode of action (MoA) and therefore psyllid resistance against this class is a serious concern. Cyantraniliprole (MoA 28) was found to be very effective against ACP and now available to rotate with imidacloprid (Stansly & Kostyk, 2012, 2013; Tiwari & Stelinski, 2013). In regions where drench applications are not common Imidacloprid MoA 4A is a good choice to rotate with sprays of other insecticides such as Spinetoram and Spirotetramat which represent MoA 5 and 23, respectively. Psyllid reduction that we observed with these 3 insecticides was not as high as expected considering work done in the other regions. This could be due to the differences in application rates and very high populations of psyllid observed in this study. It is also possible that there were some Imidacloprid resistant populations. Growers commonly use Imidacloprid and some other insecticides such as Spinosad and Bifenthrin against sucking pests.

The observed suppression of ACP is due to the combined effect of insecticides and predators. The 4 species of ladybeetles *C. septempunctata*, *A. bipunctata*, *C. sexmaculata*, and *A. cardoni* that we observed were common in treated and untreated trees indicating no apparent effect of insecticides on their populations thus their suitability for integrated pest management. Syrphids, green lacewings, and spiders are also important predators although their numbers were reduced in the treated trees toward 22 DAT. Nymphal and adult reduction of 61%–83% that we observed in ACP populations may not be enough considering oviposition potential of this pest and its primary role as vector of HLB pathogens particularly in regions where disease is present. Availability of one nymph or adult or even half per sample at 22 DAT indicate tremendous potential for increase in ACP populations if not suppressed with follow-up sprays of different MoA insecticide. The natural enemies observed in these studies warrant detailed investigations. Despite no apparent effect of treatments on their numbers feeding behavior or other demographic parameters could be impacted resulting in reduced performance. The suitability of ACP as prey to these species and their interaction with insecticides need to be investigated in detail. *Adalia bipunctata* developed and reproduced successfully on diet of ACP and also showed potential to provide significant reduction in its populations (Qureshi *et al*., 2013c). The high level of natural mortality of ACP observed in some regions was attributed to other species of ladybeetles and some were negatively impacted by insecticidal sprays of imidacloprid and spirotetramat (Michaud, 2004; Pluke *et al*., 2005; Qureshi & Stansly, 2007, 2010; Qureshi *et al*., 2009a). The botanical insecticides were found to be less toxic to some beneficial insects including coccinellids and spiders (Mansour *et al*., 1986; Hoelmer *et al*., 1990; Lowery & Isman, 1995; Naumann & Isman, 1996; Walter, 1999; Tang *et al*., 2002) and may be more suitable for integrated pest management programs and for habitats where conventional insecticides are not allowed or appropriate such as organic citrus and urban areas. The evidence of ACP resistance to some commonly used broad spectrum insecticides including Imidacloprid (Tiwari *et al*., 2011) also indicate the need for reduced use of hard chemistry insecticides and their integration with botanicals and other softer chemistries.

Disclosure

The authors declare that they have no conflict of interest.

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