

## EFFECT OF CONTACT AND SYSTEMIC INSECTICIDES ON THE SHARPSHOOTER *BUCEPHALOGONIA XANTHOPHIS* (HEMIPTERA: CICADELLIDAE), A VECTOR OF *XYLELLA FASTIDIOSA* IN CITRUS

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### ABSTRACT

The knockdown and toxic effects of insecticides of different chemical groups and modes of action registered for citrus in Brazil were investigated for effective control of *Bucephalagonia xanthophis*, a sharpshooter vector of *Xylella fastidiosa* in citrus. The active ingredients dimethoate (1.2 mL/1.2L), imidacloprid (0.24 mL/1.2L) and lambda-cyhalothrin (0.24 mL/1.2L), as well as a control (water), were sprayed onto branches of potted citrus nursery trees to evaluate the effect of residual contact. The insects were confined on sprayed branches by using sleeve cages, in groups of 10 per branch (5 branches/treatment). Lambda-cyhalothrin showed a knockdown effect on *B. xanthophis* (>70% mortality within 2 h of exposure), and the residues were effective for approximately one wk. Imidacloprid, lambda-cyhalothrin and dimethoate suppressed the vector populations for up to 3 wk after application, when the insects were exposed to sprayed plants for at least 24 h. In another experiment, 2 neonicotinoid insecticides (thiamethoxam and imidacloprid) were applied by soil drench to potted nursery trees, in order to study their systemic effect, i.e., mortality by ingestion on sharpshooter adults. Thiamethoxam and imidacloprid effectively controlled the vectors at all concentrations tested, when the insects were exposed to treated plants for 24 h (>80% mortality) or 48 h (near 100% mortality). The knockdown effect of thiamethoxam and lambda-cyhalothrin might be particularly important to prevent vector transmission of *X. fastidiosa* in citrus groves.

Key Words: Citrus variegated chlorosis, phytopathogenic bacterium, leafhopper vector, chemical control, knockdown effect

### RESUMO

O efeito de choque e tóxico de inseticidas de diferentes grupos químicos e modos de ação registrados para a cultura de citros no Brasil foram investigados para o controle de *Bucephalagonia xanthophis*, uma cigarrinha vetora de *Xylella fastidiosa* em citros. Os princípios ativos dimethoate (1,2 mL/1,2L), imidacloprid (0,24 mL/1,2L) e lambda-cyhalothrin (0,24 mL/1,2L), bem como um controle (água), foram pulverizados sobre ramos de mudas de citros para avaliação do efeito de contato residual. Os insetos foram confinados sobre ramos pulverizados em gaiolas de tule, em grupos de 10 indivíduos por ramo (5 ramos/tratamento). Lambda-cyhalothrin mostrou efeito de choque (mortalidade > 70% dentro 2 h de exposição) sobre *B. xanthophis*, perdurando por cerca de uma semana. Imidacloprid, lambda-cyhalothrin e dimethoate suprimiu a população do vetor por até 3 semanas após aplicação, quando os insetos foram expostos a plantas tratadas por pelo menos 24 h. Em outro experimento, dois inseticidas (thiamethoxam e imidacloprid) foram aplicados via “drench”, para estudar o efeito sistêmico, ou seja, mortalidade por ingestão, sobre adultos da cigarrinha. Thiamethoxam e imidacloprid controlaram efetivamente o vetor em todas as concentrações, quando os insetos foram expostos a plantas tratadas por 24 h (>80% de mortalidade) ou 48 h (cerca de 100% de mortalidade). O efeito de choque de thiamethoxam e lambda-cyhalothrin podem ser particularmente importantes para prevenir transmissão de *X. fastidiosa* pelo vetor em pomares de citros.

Palavras Chave: Clorose variegada dos citros, bactérias fitopatogênicas, cigarrinhas, controle químico, efeito de choque

Brazilian citriculture has been affected by diseases associated with insect vectors, such as Citrus variegated chlorosis (CVC), whose causal agent is a vector-borne bacterium, *Xylella fastidiosa* Welles et al., 1987; Xanthomonadales: Xanthomonadaceae. The vectors are leafhoppers from the subfamily Cicadellinae (Hemiptera: Cicadellidae), also called sharpshooters. Around 40 species of sharpshooters are registered as vectors of *X. fastidiosa*, but less than 1/3 of them are associated with plant disease epidemics (Redak et al. 2004). In the case of CVC, 12 sharpshooter species have been identified as vectors (Redak et al. 2004; Yamamoto et al. 2007), but only *Acrogonia citrina* Marucci & Cavichioli, *Bucephalagonia xanthophis* (Berg), *Dilobopterus costalimai* Young and *Oncometopia facialis* (Signoret) are commonly found on citrus trees (Paiva et al. 1996). Among them, *B. xanthophis* is the predominant species in open nurseries and young citrus orchards up to 2 yr old (Roberto et al. 2000; Yamamoto et al. 2001), the period when citrus plants are most vulnerable to *X. fastidiosa* infection. Disease management during this critical period involves roguing of diseased plants (reduction of inoculum sources) and vector control (Lopes, 1999).

In the absence of other effective vector control methods, insecticide applications in citrus orchards are important to suppress sharpshooter populations and reduce CVC spread. The application of systemic insecticides in the soil or on the plant trunk is a strategy recommended to control the sharpshooter *O. facialis* on young citrus trees (Yamamoto et al. 2000, 2001, 2002a, 2002b, 2003). Older citrus orchards need aerial sprays for vector control because the uptake and distribution in the tree canopy of soil-applied systemic insecticides is inefficient (Yamamoto et al. 2002a).

There are no insecticides registered for control of *B. xanthophis* on citrus in Brazil and the effects of insecticides on this sharpshooter species are undocumented to date. Ideally any contact or systemic insecticide should act rapidly on the insect vector, if possible preventing stylet penetration or feeding on the plant, in order to avoid pathogen transmission. Therefore, the knockdown and toxic effects of different chemical groups and modes of action of insecticides registered for citrus were investigated for the control of *B. xanthophis*.

## MATERIAL AND METHODS

### Insects and Plants

A healthy colony of *B. xanthophis* was established on plants of *Vernonia condensata* Baker (Asteraceae), as described by Marucci et al. (2003), with adults collected on young branches of the ornamental shrub, *Duranta repens* L., in Piracicaba, SP, Brazil. The colony was maintained

in a greenhouse without exposure to insecticides, and oviposition cages were set up every 2 wk in order to obtain adults of known age (1-2 wk old) for the experiments.

The citrus plants used in the experiment were healthy sweet orange [*Citrus sinensis* (L.) Osbeck, cv. Pera] nursery trees, grafted on rangpur lime [*Citrus x limonia* Osbeck], kept in a vector-proof greenhouse. The nursery trees were about 1 m high, grown in 5-L plastic bags containing potting soil mix (Tropstrato V8®), and fertilized weekly with macronutrients (ammonium nitrate and calcium nitrate) and micronutrients (magnesium sulfate, potassium nitrate, monoammonium phosphate, zinc, copper and iron) diluted in the irrigation water and applied on the plants. The plants were pruned 3-4 wk before the experiment, and treated with insecticides when the young shoots showed totally expanded leaves. The experiments were carried out in a greenhouse with temperatures in the range of 20-32 °C.

### Insecticides

The commercial formulations of 4 insecticides, from different chemical groups and modes of action, were evaluated (Table 1). All the insecticides are registered for citrus pest management in Brazil. Provado® (imidacloprid) and Actara® (thiamethoxam) are registered for control of the sharpshooter, *O. facialis*. Karate® (lambda-cyhalothrin) is registered to control the sharpshooter, *D. costalimai*. Finally, Perfekthion® (dimethoate) is not registered for sharpshooters, but is registered for the control of other piercing-sucking insects, e.g. mealybugs and aphids (Agrofit 2012).

### Contact Effect

The active ingredients, dimethoate (1 mL/L), imidacloprid (0.2 mL/L), lambda-cyhalothrin (0.2 mL/L) and a control (water), were sprayed onto citrus nursery trees using a compression sprayer (Guarany®), covering the whole plant until product runoff, as in field applications. After the spraying and drying of the insecticides, groups of 10 adult sharpshooters per replicate were confined to a single branch of the treated plants using sleeve cages. Insect mortality was evaluated at 2 h (knockdown), 24 h and 48 h after confinement. The experimental design was completely randomized with 4 treatments (insecticides) and 5 repetitions.

### Residual Contact Effect

The active ingredients, dimethoate (1 mL/L), imidacloprid (0.2 mL/L), lambda-cyhalothrin (0.2 mL/L) and a control (water), were sprayed onto new nursery trees using a compression sprayer (Guarany®), as described in the previous experi-

TABLE 1. INSECTICIDES TESTED AGAINST *BUCEPHALOGONIA XANTHOPHIS* IN THE ASSAYS.

Active ingredient	Trade name	Class	Mode of action	Manufacturer	<sup>1</sup> Dosage on citrus (citadellid species)
Dimethoate	Perfekthion	Organophosphate	Acetylcholinesterase inhibitor	BASF	— <sup>2</sup>
Imidacloprid	Provado 200 SC	Neonicotinoid	Nicotinic acetylcholine receptor agonist / antagonist	BAYER S. A.	15 to 20 mL/100L ( <i>Oncometopia fasciatis</i> )
Lambda-cyhalothrin	Karate Zeon 50 CS	Synthetic Pyrethroid	Sodium channel modulator	SYNGENTA	200 to 400 mL/ha ( <i>Dilobopterus costalimai</i> )
Thiamethoxam	Actara 250 WG	Neonicotinoid	Nicotinic acetylcholine receptor agonist / antagonist	SYNGENTA	3g/plant or 600g/ha ( <i>Oncometopia fasciatis</i> )

<sup>1</sup>Dose of commercial product registered for sharpshooter control (Hemiptera: Cicadellidae) in citrus (Agrofit, 2012).

<sup>2</sup>Insecticide without registration for sharpshooter control in citrus (Agrofit, 2012).

ment. At intervals of 1, 2, 3, 6 and 8 wk after insecticide spraying, groups of 10 adult sharpshooters were confined on a single branch of the treated plants with sleeve cages. Insect mortality was evaluated at 2 h (knockdown), 24 h and 48 h after confinement. Different groups of plants were used for assessing insect mortality at each weekly interval after insecticide application. The experimental design was completely randomized with 4 treatments and 5 repetitions per weekly interval.

#### Systemic Effect

The insecticides thiamethoxam (Actara 250 WG) and imidacloprid (Provado 200 SC) were diluted into 5 concentrations and 50 mL of each dilution were applied per plant in the soil (via “drench”). Ten days after application, groups of 10 sharpshooters were confined to a single branch of the treated plants with sleeve cages. Insect mortality was evaluated at 2 (knockdown), 24 and 48 h after sharpshooter confinement. The experimental design was entirely randomized with 11 treatments (2 insecticides × 5 dilutions and a control) and 5 repetitions.

#### Data Analysis

The mortality data of the contact effect experiment were submitted to a factorial (4 × 3) analysis of variance using the F and Tukey's test, with factors represented by the ‘insecticide treatments’ (dimethoate, imidacloprid, lambda-cyhalothrin or water) and the ‘contact time’ (2 h, 24 h or 48 h). For the residual contact experiment, the mortality data of each time interval after insecticide application (1, 2, 3, 6 and 8 wk) were submitted to a factorial (4 × 3) analysis of variance using the F and the Tukey tests, with ‘insecticide treatments’ (dimethoate, imidacloprid, lambda-cyhalothrin or water) and the ‘contact time’ (2 h, 24 h or 48 h) as factors. In the systemic effect experiment, the mortality data were submitted to an analysis of variance using the F test and the Tukey tests, with a 11 × 3 factorial design, in which the ‘insecticide treatments’ and the ‘exposure time’ (2 h, 24 h or 48 h) were considered as factors. The tests of Bartlett (Bartlett 1937) and Shapiro-Wilk (Shapiro & Wilk 1965) were applied to evaluate the assumptions of homoscedasticity for treatment variances and residual normality, respectively. The analyses in this study were done using the SAS statistical software (SAS Institute, 2003).

## RESULTS

#### Contact Effect

The active ingredients imidacloprid and lambda-cyhalothrin caused a knockdown effect on the

sharpshooter (Table 2), with more than 80% mortality to confined insects immediately after spraying and evaluated after 2 hr of confinement ( $F = 21.69$ ;  $P < 0.0001$ ). After 24 hr, the 3 insecticides (imidacloprid; lambda-cyhalothrin and dimethoate) caused a high mortality to *B. xanthophis* ( $F = 379.72$ ;  $P < 0.0001$ ), and after 48 hr, practically 100% of the adults were killed by the insecticides ( $F = 200.81$ ;  $P < 0.0001$ ). There was an interaction between the insecticides and the contact time ( $F = 11.40$ ;  $P < 0.0001$ ).

#### Residual Contact Effect

There was an interaction between the treatments and the contact time for wk 1 ( $F = 4.27$ ;  $P = 0.0016$ ) and wk 2 ( $F = 2.74$ ;  $P = 0.0226$ ) for the residual analysis of the first 2 wk. However, this interaction was not found after the third wk (Table 3). Considering only the first 2 hr of contact, it was observed that the insecticide lambda-cyhalothrin caused 78% and 48% mortality in the first and second wk after spraying, respectively, differing from the control [wk 1 ( $F = 7.14$ ;  $P = 0.0029$ ); wk 2 ( $F = 4.41$ ;  $P = 0.0193$ )] (Table 3).

After 24 hr of contact, it was found that the 3 insecticides caused significant mortality to confined insects for up to 3 wk after spraying [wk 1 ( $F = 35.58$ ;  $P < 0.0001$ ); wk 2 ( $F = 19.80$ ;  $P < 0.0001$ ); wk 3 ( $F = 6.96$ ;  $P = 0.0033$ )]. The residual activity of the 3 insecticides after 48 hr of contact was consistent until 21 d after spraying (wk 3). During these 3 wk, the active ingredients differed significantly from the control [wk 1 ( $F = 111.49$ ;  $P < 0.0001$ ); wk 2 ( $F = 24.78$ ;  $P < 0.0001$ ); wk 3 ( $F = 11.55$ ;  $P = 0.0003$ )]. Insecticide efficiency for this period was  $\geq 70\%$  (Table 3). The sixth week showed no significance between treatments [wk 6 ( $F = 2.98$ ;  $P = 0.0626$ )]. Finally, in the eighth week, treatments were significantly different [wk 8 ( $F = 5.98$ ;  $P = 0.0062$ )], but insecticide efficacy was  $< 70\%$  (Table 3).

#### Systemic Effect

A significant interaction was found between the each of the 2 insecticide treatments (thiamethoxam and imidacloprid) and insect exposure time on treated plants ( $F = 8.33$ ;  $P < 0.0001$ ) (Table 4). There were no mortality differences between treatments when the exposure time was  $\geq 24$  h, with the exception of imidacloprid at 1.40 ppm. On the other hand, the mortality rates caused by both insecticides during the first 2 h were lower than those observed at 24 and 48 h of exposure to treated plants.

The knockdown effect on the insect vector by ingestion was observed when the insecticides were applied systemically at the highest concentrations ( $F = 8.92$ ;  $P < 0.0001$ ) (Table 4). Thiamethoxam caused a knockdown effect on *B. xanthophis* at a concentration of 20 ppm, and mortality was higher than that caused by the other treatments, with the exception of thiamethoxam at 2 ppm and imidacloprid at 35 ppm. After 24 h of exposure, mortality rates  $\geq 80\%$  were observed even for the smallest concentration of the imidacloprid and thiamethoxam (Table 4); the mortality rates caused by all the concentrations differed numerically from the control, although there was no significant statistical difference between them ( $F = 8.92$ ;  $P < 0.0001$ ). After 48 h, nearly 100% of the insects were killed by the insecticides (Table 4), and the mortality caused by different insecticide concentrations were similar, only differing from the control ( $F = 415.31$ ;  $P < 0.0001$ ). Ninety-six percent of sharpshooters in the control were still alive after 48 h (Table 4).

The insecticides Provado® (imidacloprid) and Actara® (thiamethoxam) were highly toxic to *B. xanthophis* after 48 h of exposure (Table 4). Thiamethoxam caused  $\geq 96\%$  mortality in the first 24 h and all concentrations of this insecticide reached 100% efficacy after 48 h (Table 4). For imidacloprid, most concentrations reached 100%

TABLE 2. CONTACT INSECTICIDE KNOCKDOWN AND TOXIC EFFECTS ON *BUCEPHALOGONIA XANTHOPHIS*.

Treatments	Knockdown <sup>1</sup>		24 h		48 h	
	Mortality <sup>2</sup>	E <sup>3</sup>	Mortality <sup>2</sup>	E <sup>3</sup>	Mortality <sup>2</sup>	E <sup>3</sup>
Water	0 ± 0.0 bA	—	2 ± 2.0 bA	—	12 ± 2.4 bA	—
Dimethoate	20 ± 15.5 bB	20.0	98 ± 2.0 aA	98.0	100 ± 0.0 aA	100.0
Imidacloprid	84 ± 5.1 aA	84.0	94 ± 4.0 aA	93.9	98 ± 2.0 aA	97.7
Lambda-cyhalothrin	82 ± 8.6 aA	82.0	100 ± 0.0 aA	100.0	100 ± 0.0 aA	100.0
Factor	DF		F		P	
Treatments (T)	3		157.20		<0.0001	
Time of contact (Tc)	2		33.95		<0.0001	
T × Tc	6		11.40		<0.0001	

<sup>1</sup>Evaluation made 2 hr after confinement.

<sup>2</sup>Means in a column followed by different small letter, or in a row by different capital letter are significantly different by Tukey's test ( $P < 0.05$ ).

<sup>3</sup>E: Efficacy calculated from Abbott's formula, 1925.

TABLE 3. INSECTICIDE RESIDUAL EFFECTS BY CONTACT ON *BUCEPHALOGONIA XANTHOPHIS*.

Sampling interval	Treatment	Mortality <sup>2</sup>			Efficacy <sup>3</sup>
		Knockdown <sup>1</sup>	24 h	48 h	
Week 1	Water	0 ± 0.0 bA	2 ± 2.0 cA	2 ± 2.0 bA	—
	Dimethoate	32 ± 19.9 abB	72 ± 8.6 abAB	100 ± 0.0 aA	100.0
	Imidacloprid	16 ± 8.1 bC	50 ± 8.9 bB	86 ± 7.5 aA	85.7
	Lambda-cyhalothrin	78 ± 13.2 aA	94 ± 4.0 aA	94 ± 4.0 aA	93.9
	Factor	DF	F	P	
	Treatments (T)	3	56.60	<0.0001	
	Time of contact (Tc)	2	20.87	<0.0001	
	T × Tc	6	4.27	0.0016	
Week 2	Water	0 ± 0.0 bA	10 ± 5.5 bA	22 ± 13.2 bA	—
	Dimethoate	20 ± 11.4 abB	82 ± 5.4 aA	96 ± 4.0 aA	94.9
	Imidacloprid	28 ± 8.6 abB	78 ± 10.2 aA	100 ± 0.0 aA	100.0
	Lambda-cyhalothrin	48 ± 12.4 aB	86 ± 9.8 aA	94 ± 6.0 aA	92.3
	Factor	DF	F	P	
	Treatments (T)	3	38.40	<0.0001	
	Time of contact (Tc)	2	44.45	<0.0001	
	T × Tc	6	2.74	0.0226	
Week 3	Water	0 ± 0.0 aB	12 ± 5.4 bAB	32 ± 8.6 bA	—
	Dimethoate	28 ± 12.0 aB	74 ± 12.5 aA	90 ± 6.3 aA	85.3
	Imidacloprid	10 ± 4.5 aB	68 ± 11.1 aA	92 ± 8.0 aA	88.2
	Lambda-cyhalothrin	30 ± 18.4 aA	68 ± 13.2 aA	78 ± 9.7 aA	67.6
	Factor	DF	F	P	
	Treatments (T)	3	14.76	<0.0001	
	Time of contact (Tc)	2	31.17	<0.0001	
	T × Tc	6	1.37	0.2458	
Week 6	Water	14 ± 14.0 aB	34 ± 19.1 aA	50 ± 18.2 aA	—
	Dimethoate	14 ± 14.0 aB	68 ± 10.7 aA	94 ± 4.0 aA	88.0
	Imidacloprid	6 ± 4.0 aB	74 ± 19.4 aA	82 ± 15.6 aA	64.0
	Lambda-cyhalothrin	22 ± 9.7 aA	90 ± 5.5 aA	96 ± 4.0 aA	92.0
	Factor	DF	F	P	
	Treatments (T)	3	4.28	<0.0001	
	Time of contact (Tc)	2	29.59	0.0093	
	T × Tc	6	1.02	0.4248	
Week 8	Water	0 ± 0.0 aA	0 ± 0.0 aA	2 ± 2.0 bA	—
	Dimethoate	0 ± 0.0 aA	2 ± 2.0 aA	14 ± 9.3 bA	12.2
	Imidacloprid	10 ± 10.0 aA	32 ± 17.4 aAB	68 ± 11.6 aB	67.3
	Lambda-cyhalothrin	0 ± 0.0 aA	16 ± 6.8 aA	38 ± 18.6 abA	36.7
	Factor	DF	F	P	
	Treatments (T)	3	9.13	<0.0001	
	Time of contact (Tc)	2	9.46	0.0003	
	T × Tc	6	1.86	0.1077	

<sup>1</sup>Evaluation made 2 hr after confinement.

<sup>2</sup>Means in a column followed by different small letter, or in a row by different capital letter are significantly different by Tukey's test ( $P < 0.05$ ).

<sup>3</sup>E: Efficacy at 48 h after exposure calculated from Abbott's formula, 1925.

TABLE 4. SYSTEMIC INSECTICIDE KNOCKDOWN AND TOXIC EFFECTS ON *BUCEPHALAGONIA XANTHOPHIS*.

Treatment (ppm)		Mortality <sup>2</sup>			Efficacy <sup>3</sup>
		Knockdown <sup>1</sup>	24 h	48 h	
Water	0.00	0 ± 0.0 dA	2 ± 2.0 bA	4 ± 2.4 bA	—
Thiamethoxam	0.08	22 ± 8.0 bcdB	98 ± 2.0 aA	100 ± 0.0 aA	100.0
	1.00	24 ± 12.1 bcdB	100 ± 0.0 aA	100 ± 0.0 aA	100.0
	1.34	40 ± 15.2 bcdB	96 ± 2.4 aA	100 ± 0.0 aA	100.0
	2.00	60 ± 16.4 abB	98 ± 2.0 aA	100 ± 0.0 aA	100.0
	20.00	94 ± 2.4 aB	100 ± 0.0 aA	100 ± 0.0 aA	100.0
Imidacloprid	1.40	12 ± 5.8 cdC	80 ± 7.1 aB	100 ± 0.0 aA	100.0
	1.74	2 ± 2.0 dB	82 ± 9.2 aA	94 ± 4.0 aA	93.7
	2.34	10 ± 3.2 cdB	92 ± 5.8 aA	100 ± 0.0 aA	100.0
	3.50	20 ± 5.5 bcdB	92 ± 4.9 aA	100 ± 0.0 aA	100.0
	35.00	54 ± 15.0 abcB	98 ± 2.0 aA	100 ± 0.0 aA	100.0
Factor		DF	F	P	
Treatments (T)		10	48.18	<0.0001	
Time (Ti)		2	318.15	<0.0001	
T × Ti		20	8.33	<0.0001	

<sup>1</sup>Evaluation made 2 hr after confinement.

<sup>2</sup>Means in a column followed by different small letters, or in a line by different capital letters are significantly different by Tukey's test ( $P < 0.05$ ).

<sup>3</sup>E: Efficacy at 48 h after exposure calculated from Abbott's formula, 1925.

efficacy after 48 h; the only exception was 1.74 ppm, with an efficacy of 93.75%.

## DISCUSSION

Chemical control has been commonly used to manage diseases caused by *X. fastidiosa* in various crops, aiming to reduce vector population numbers and their frequency of visits to susceptible crops and inoculum sources. Insecticides may also affect vector behavior and competence for acquisition and inoculation of *X. fastidiosa*. In the case of oleander leaf scorch, for example, Bethke et al. (2001) observed that insecticides inhibit feeding of the sharpshooter vector, *Homalodisca coagulata* (Say), resulting in a substantial decrease in transmission efficiency of *X. fastidiosa* to oleander (*Nerium oleander* L.; Gentianales: Apocynaceae).

*X. fastidiosa* has a wide host plant range, which includes species from more than 30 families of mono and dicotyledons (Hopkins 1989; Purcell & Hopkins 1996), and some of them may serve as inoculum sources for infection of susceptible crops (Hopkins 1989; Leite et al. 1997). In Brazilian citrus groves, however, infected trees appear to be the most important (if not the only) sources of inoculum for CVC spread by sharpshooter vectors (Laranjeira, 1997; Lopes 1999). In this case, a knockdown effect of insecticides on sharpshooters could drastically reduce vector efficiency in acquiring and/or inoculating bacterial cells when

feeding on treated citrus trees. Based on this hypothesis, we conducted a series of experiments to verify the contact and systemic toxicity as well as possible knockdown effects of some insecticides commonly used in Brazilian citriculture.

We showed that imidacloprid and lambda-cyhalothrin cause a contact knockdown effect on the sharpshooter, *B. xanthophis*, which is an important vector of *X. fastidiosa* in young citrus groves (Yamamoto et al., 2001). These 2 insecticides are registered for citrus in Brazil to control the sharpshooters, *O. facialis* and *D. costalimai*, respectively (Agrofit 2012). Both cause mortality by provoking hyperexcitability, but with distinct modes of action. Imidacloprid is a neonicotinoid and acts as an acetylcholine agonist on the post-synaptic nicotinic receptors (Nauen et al. 2001). As a pyrethroid, lambda-cyhalothrin is a sodium channel modulator, causing a potential repetitive action and consequent mortality (Soderlund & Knipple 2003). The knockdown effect of pyrethroids on insects are well known. Imidacloprid is widely used as a systemic insecticide to control piercing-sucking insects (Nauen et al. 1998; 1999), including sharpshooters in citrus orchards and vineyards (Yamamoto et al. 2000; Castle et al. 2005; Byrne & Toscano 2006; Byrne & Toscano 2007). The contact effect of imidacloprid, causing high sharpshooter mortality within a short period of time, is described here for the first time.

It is also important to determine the duration of the crop protection period by using these insecticides.

ticides. Our data shows that effective knockdown activity by lambda-cyhalothrin is maintained for about 1 wk after spraying, decreasing thereafter. Only 48% and 30% insect mortalities within 2 h of exposure to treated plants were recorded in the second and third weeks after application, respectively. In the case of imidacloprid, no residual knockdown effect was detected.

The contact knockdown effect of insecticides to control sharpshooter vectors of *X. fastidiosa* has been little studied. However, this effect is very desirable to avoid disease spread. Up to now, only acetamiprid (6 g of active ingredient/100 L) had been described as having a knockdown effect against *O. facialis* on citrus (Yamamoto et al. 2003), as well as fenpropathrin, fenpropathrin + acephate and carbaryl on *H. coagulata* on oleander (Bethke et al. 2001).

With longer exposure periods on sprayed plants, *B. xanthophis* is effectively controlled by imidacloprid, lambda-cyhalothrin and dimethoate through their contact effects. When the insects were confined immediately after the spraying of the active ingredients, about 100% mortality was observed for the 3 insecticides during the first 24 h. The residual contact activity of these compounds lasted for 3 wk after spraying. Other insecticides are also considered effective for controlling sharpshooters by contact action. Pyrethroids such as bifenthrin, esfenvalerate and fenpropathrin were highly toxic to adults and nymphs of *H. coagulata* under laboratory conditions (Prabhaker et al. 2006a; Prabhaker et al. 2006b). The neonicotinoids imidacloprid and acetamiprid caused mortality to *O. facialis* adults in a greenhouse study (Yamamoto et al. 2003).

In Brazil, the control of sharpshooter and other vectors of plant pathogens in young citrus groves (up to 3 yr old) is done mainly with systemic insecticides applied in the soil or on the plant trunk. Thus, we already expected an efficient control of *B. xanthophis* by adopting this strategy. The toxic effect of the 2 systemic insecticides, thiamethoxam and imidacloprid, at all concentrations tested, was indeed very clear after 24 h of sharpshooter exposure to treated plants. Interestingly, we also noted a strong knockdown effect (94% mortality in only 2 h of exposure) caused by thiamethoxam at the highest concentration (20 ppm). This knockdown effect is often less evident for a systemic insecticide applied in the soil than for a contact insecticide sprayed directly on the leaves. In adult citrus trees under field conditions, thiamethoxam needs less time for translocation in the plant compared to imidacloprid (Castle et al. 2005), and this characteristic may explain the greater knockdown effect of thiamethoxam observed on *B. xanthophis* when applied via soil in citrus nursery trees in the present study. On the other hand, imidacloprid has a greater residual effect on citrus in the field (Castle et al. 2005).

Systemic insecticides are used for sharpshooter control due to their longer residual action compared to contact insecticides. Therefore, they provide a longer protection period to the plant, with a lower number of applications and, consequently, with lower production cost. Apart from this, systemic applications have the advantage of being selective to natural enemies, whose preservation can contribute to an increase in the biological control of citrus pests (Gravena et al. 1997).

Because lambda-cyhalothrin and thiamethoxam have a knockdown effect on *B. xanthophis* for at least one wk after application, both insecticides might be effective to avoid *X. fastidiosa* transmission by this sharpshooter. For up to 3 wk, imidacloprid, lambda-cyhalothrin and dimethoate showed an effective contact action that can be helpful to manage CVC by suppressing vector populations and perhaps affecting vector feeding behavior and competence to transmit the pathogen in citrus groves. More detailed studies are needed to confirm the efficacy of these insecticides in preventing bacterial acquisition and/or inoculation by sharpshooters.

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#### REFERENCES CITED

- AGROFIT. 2012. Ministério de Agricultura. Disponível em: <[http://extranet.agricultura.gov.br/agrofit\\_cons/principal\\_agrofit\\_cons](http://extranet.agricultura.gov.br/agrofit_cons/principal_agrofit_cons)>. Acesso em: 02 Jan. 2012.
- BARTLETT, M. S. 1937. Properties of sufficiency and statistical tests. Proc. R. Soc. London Series A. 160: 268-282.
- BETHKE, J. A., BLUA, M. J., AND REDAK, R. A. 2001. Effect of selected insecticides on *Homalodisca coagulata* (Homoptera: Cicadellidae) and transmission of oleander leaf scorch in a greenhouse study. J. Econ. Entomol. 94: 1031-1036.
- BYRNE F. J., AND TOSCANO, N. C. 2006. Uptake and persistence of imidacloprid in grapevines treated by chemigation. Crop Prot. 25: 831-834.
- BYRNE F. J., AND TOSCANO, N. C. 2007. Lethal toxicity of systemic residues of imidacloprid against *Homalodisca vitripennis* (Homoptera: Cicadellidae) eggs and its parasitoid *Gonatocerus ashmeadi* (Hymenoptera: Mymaridae). Biol. Control. 43: 130-135.
- CASTLE S. J., BYRNE, F. J., BI, J. L., AND TOSCANO, N. C. 2005. Spatial and temporal distribution of imidacloprid and thiamethoxam in citrus and impact on *Homalodisca coagulata* populations. Pest Manag. Sci. 61: 75-84.
- GRAVENA S., LOPES, J. R. S., PAIVA, P. E. B., YAMAMOTO, P. T., AND ROBERTO, S. R. 1997. Os vetores da *Xylella fastidiosa*, pp. 37-53 In L. C. Donadio and C. S. Moreira [eds.], Clorose variegada dos citros. Bebedouro: Estação Experimental de Citricultura.
- HOPKINS, D. L. 1989. *Xylella fastidiosa*: Xylem-limited bacterial pathogen of plants. Annu. Rev. Phytopathol. 27: 271-290.

- LARANJEIRA, F. F. 1997. Dez anos de clorose variegada dos citros: O que sabemos? Laranja 18: 123-141.
- LEITE, R. M. V. B. C., LEITE JR, R. P., AND CEREZINE, P. C. 1997. Flutuação populacional de *Xylella fastidiosa* em ameixeiras suscetíveis e resistentes à escaldadura da folha. Fitopatol. Brasileira 22: 58-63.
- LOPES, J. R. S. 1999. Estudo com vetores de *Xylella fastidiosa* e implicações no manejo da clorose variegada dos citros. Laranja 20: 329-344.
- MARUCCI, R. C., GIUSTOLIN, T. A., MIRANDA, M. P., MIQUELOTE, M., ALMEIDA, R. P. P., AND LOPES, J. R. S. 2003. Identification of a non-host plant of *Xylella fastidiosa* to rear healthy sharpshooter vectors. Sci. Agric. 60: 669-675.
- NAUEN, R., HUNGENBERG, H., TOLLO, B., TIETJEN, K., AND ELBERT, A. 1998. Antifeedant effect, biological efficacy and high affinity binding of imidacloprid to acetylcholine receptors in *Myzus persicae* and *Myzus nicotianae*. Pestic. Sci. 53: 133-140.
- NAUEN, R., KOOB, B., AND ELBERT, A. 1999. Antifeedant effects of sublethal dosages of imidacloprid on *Bemisia tabaci*. Entomol. Exp. Appl. 88: 287-293.
- NAUEN, R., EBBINGHAUS-KINTSCHER, U., ELBERT, A., JESCHKE, P., AND TIETJEN, K. 2001. Acetylcholine receptors as sites for developing neonicotinoid insecticides, pp. 77-105 In I. Ishaaya [ed.], Biochemical sites in insecticide action and resistance. Springer. New York.
- PAIVA, P. E. B., SILVA, J. L., GRAVENA, S., AND YAMAMOTO, P. T. 1996. Cigarrinhas de xilema em pomares de laranja no Estado de São Paulo. Laranja 17: 41-54.
- PRABHAKER, N., CASTLE, S. J., AND TOSCANO, N. C. 2006a. Susceptibility of immature stages of *Homalodisca coagulata* (Hemiptera: Cicadellidae) to selected insecticides. J. Econ. Entomol. 99: 1805-1812.
- PRABHAKER, N., CASTLE, S., BYRNE, F., HENNEBERRY, T. J., AND TOSCANO, N. C. 2006b. Establishment of baseline susceptibility data to various insecticides for *Homalodisca coagulata* (Homoptera: Cicadellidae) by comparative bioassay techniques. J. Econ. Entomol. 99: 141-154.
- PURCELL, A., AND HOPKINS, D. L. 1996. Fastidious xylem-limited bacterial plant pathogens. Annu. Rev. Phytopathol. 34: 131-151.
- REDAK, R. A., PURCELL, A. H., LOPES, J. R. S., BLUA, M. J., MIZELL III, R. F., AND ANDERSEN, P. C. 2004. The biology of xylem fluid – feeding insect vectors of *Xylella fastidiosa* and their relation to disease epidemiology. Annu. Rev. Entomol. 49: 243-270.
- ROBERTO, S. R., DALLA PRIA JÚNIOR, W., YAMAMOTO, P. T., FELLIPE, M. R., AND FREITAS, E. P. 2000. Espécies e flutuação populacional de cigarrinhas em viveiros de citros, em Gavião Peixoto (SP). Laranja 21: 65-79.
- SAS, INSTITUTE. 2003. SAS User guide: versão 9.1. Cary, NC.
- SHAPIRO, S. S., AND WILK, M. B. 1965. An analysis of variance test for normality. Biometrika 52: 591-611.
- SODERLUND, D. M., AND KNIPPLE, D. C. 2003. The molecular biology of knockdown resistance to pyrethroid insecticides. Insect Biochem. Mol. Biol. 33: 563-577.
- YAMAMOTO, P. T., ROBERTO, S. R., AND DALLA PRIA JÚNIOR, W. 2000. Inseticidas sistêmicos aplicados via tronco para controle de *Oncometopia facialis*, *Phyllocnistis citrella* e *Toxoptera citricida* em citros. Sci. Agric. 57: 415-420.
- YAMAMOTO, P. T., ROBERTO, S. R., DALLA PRIA JÚNIOR, W., FELIPPE, M. R., FREITAS, E. P., CAETANO, A. C., AND SANCHES, A. L. 2001. Inseticidas sistêmicos aplicados via tronco no controle da cigarrinha *Oncometopia* em citros. Laranja 22: 49-63.
- YAMAMOTO, P. T., FELIPPE, M. R., CAETANO, A. C., SANCHES, A. L., ALMEIDA, E. J., AND NOCITI, L. A. S. 2002a. Eficiência de inseticidas neonicotinóides aplicados via tronco no controle de *Oncometopia Facialis* (Signoret) (Hemiptera, Cicadellidae) em mudas de laranja 'pêra'. Laranja 23: 101-114.
- YAMAMOTO P. T., DALLA PRIA JÚNIOR, W., ROBERTO, S. R., FELIPPE, M. R., ALMEIDA, E. J., AND FREITAS, E. P. 2002b. Controle químico da cigarrinha em citros. Laranja 23: 141-154.
- YAMAMOTO, P. T., FELIPPE, M. R., NOCITI, L. A. S., MONTESINO, L. H., AND COELHO, J. H. C. 2003. Uso de Acetamiprid no controle da cigarrinha em citros. Laranja 24: 337-351.
- YAMAMOTO, P. T., FELIPPE, M. R., CAETANO, A. C., SANCHES, A. L., AND LOPES, J. R. S. 2007. First report of *Fingeriana dubia* Cavichioli transmitting *Xylella fastidiosa* to citrus. Fitopatol. Brasileira 32: 266-267.