

Toxicity Effect of Three Insecticides on Important Pests and Predators in Tomato Plants

**J. E. R. Gorri¹, R. C. Pereira², F. M. Alves¹, F. L. Fernandes^{3*},
Í. W. da Silva¹, and M. E. S. Fernandes³**

¹Universidade Federal de Viçosa – *Campus* Rio Paranaíba, Rio Paranaíba, Brasil

²Universidade Federal de Viçosa, Viçosa, Brasil

³Universidade Federal de Viçosa – *Campus* Rio Paranaíba, Instituto Ciências Agrárias, Rio Paranaíba, Brasil

*Correspondence: Flávio Lemes Fernandes, Universidade Federal de Viçosa-Campus Rio Paranaíba, Rio Paranaíba, CEP 38810-000, Brasil. Tel: +55 34 3855-2267. E-mail: flaviofernandes@ufv.br

DOI: 10.12735/as.v3i1p01

URL: <http://dx.doi.org/10.12735/as.v3i1p01>

Abstract

The objective of this study was to evaluate insecticide toxicity on *Bemisia tabaci*, *Tetranychus evansi*, *Orius insidiosus*, *Cycloneda sanguinea* and *Chauliognathus flavipes* in tomato plants. The following toxicity treatments were applied: T1: control, T2: chlorpyrifos (Pitcher) 450 EC (1.25 L.ha⁻¹), T3: chlorpyrifos 450 EC (Pitcher) (0.62 L.ha⁻¹), T4: thiamethoxam 100 WG (1.00 L.ha⁻¹), T5: thiamethoxam 250 WG (0.50 L.ha⁻¹), T6: teflubenzuron 150 CS (0.025 L.ha⁻¹) and T7: teflubenzuron 150 CS (0.0125 L.ha⁻¹). For the sub-lethal effect, a tenth of the recommended concentration was used. The insecticide teflubenzuron was effective against whitefly nymphs, while chlorpyrifos and thiamethoxam were efficient against adult whiteflies. Insecticides at the dose and lower dose were not efficient to control the adults and nymphs of mite *T. evansi*. Teflubenzuron was selective at the two doses tested against *C. sanguinea* and *C. flavipes*, while thiamethoxam was selective (lower dose) against these two predators. Chlorpyrifos was selective against *C. flavipes* (lower dose). All insecticides were selective or moderately selective at 21 days after application. Insecticides were not efficient to control the adults and nymphs of *T. evansi*. The growth regulating insecticide teflubenzuron was effective against whitefly nymphs. Chlorpyrifos and thiamethoxam were efficient against adult whiteflies. In general, the insecticides neonicotinoid and pyrethroid were selective to three predatory species and had lower sublethal impact compared with the organophosphate.

Keywords: effectiveness, predators, red spider mite, selectivity, whitefly

1. Introduction

The whitefly *Bemisia tabaci*, (Gennadius) (Hemiptera: Aleyrodidae), and the red spider mite *Tetranychus evansi* (Acari: Tetranychidae) are important pests in tomato plants (*Solanum lycopersicum*) (Pedigo & Rice, 2005). *B. tabaci* of the whitefly is responsible for major losses in agriculture (Lourenção, Yuki, & Alves, 1999). This species can cause direct damage by sucking sap and injecting toxins or indirect damage by transmitting phytoviruses, as well as favor the occurrence of sooty molds (*Capnodium* sp.) (Hendrix & Wei, 1992). Sap sucking and toxin

injection cause changes in vegetative and reproductive development and cause physiological changes in plants (Toscano, Boiça Júnior, & Maruyama, 2004). Moreover, the pest can transmit 17 different types of virus. In contrast, the red spider mite attacks the older leaves and scrapes the tissues, causing small red spots between the veins of leaves, which coalesce, and later dry up and fall (Pedigo & Rice, 2005).

Control of these pests is accomplished by the application of insecticides, which results in low effectiveness, as well as losses to farmers and to the environment. For effective integrated management of these pests in tomato plants, products should have over 80% of effectiveness, be selective to their natural enemies, and allow use with other products alternately. Moreover, it is of utmost importance that insecticides do not affect the behavior of natural enemies at sublethal doses (Fernandes *et al.*, 2008).

Natural enemies include predators *Orius insidiosus* (Hemiptera: Anthocoridae), *Cycloneda sanguinea* (Linnaeus) (Coleoptera: Coccinellidae) and *Chauliognathus flavipes* (Coleoptera: Cantharidae) which feed from the pests in tomato plants (Oliveira, Oliveira, Sarmento, Fadini, & Moreira, 2005; Pedigo & Rice, 2005).

Despite the importance of the whitefly and the red spider mite and the biological control of these pests by *O. insidiosus*, *C. sanguinea* and *C. flavipes*, there are a few studies assessing the effectiveness of insecticides against them, selectivity and sublethal effect on these natural enemies in tomato plants. Thus, the aim of this study was to evaluate the effectiveness of insecticides against the *B. tabaci* and *T. evansi*, physiological selectivity and sublethal effect on predators *O. insidiosus*, *C. sanguinea* and *C. flavipes* for management of insecticides in tomato plants.

2. Method

The experiment was conducted in a greenhouse at the Integrated Pest Management Laboratory - Federal University of Viçosa. For this purpose, the experiment used tomato plants (*S. lycopersicum*), cultivar Santa Clara. The seeds were sown on 128-cell styrofoam trays, and commercial vegetal substrate Bioplant was used. 3 or 4 seeds were placed in each tray cell. After 35 days, the seedlings were transplanted to 3 L pots. The substrate used for this planting was composed of earth and cattle manure (2:1).

2.1. Insects for Bioassays

T. evansi adults were brought from infested leaves of tomato plants cultivated in the garden of UFV, free from insecticide application. The population of *T. evansi* was kept in tomato plants inside cages covered with organza (80 cm long, 30 cm wide and 95 cm high) until obtaining nymphs and adults.

Adults of *B. tabaci* were collected in a greenhouse at UFV, with the aid of a small vacuum cleaner and plastic pot (Pedigo & Rice, 2005). The adults were released in a 4 × 6 m greenhouse with 25 cabbage plants, *Brassica oleracea capitata* L., which were free of diseases and other insects. Weekly, new cabbage plants were added to the greenhouse to maintain the population of this insect pest, in order to provide adults and nymphs for the bioassays.

Adults of *O. insidiosus*, *C. sanguinea* and *C. flavipes* were captured in citrus (*Citrus sinensis*) and maize (*Zea mays*) crops in the experimental station of UFV, with a vacuum cleaner. All specimens of insects were stored in 4 mL vials with 70% ethanol and sent to taxonomists for identification. To the bioassays, these predators were capture method used soda bottles with rectangular openings (15 × 20 cm) closed with a thin organza tissue. On the inside of the bottle a paper towel was added to facilitate insect movement. The insects were sucked up with a simple sucker hose, blown to the inside of the bottle and transported to the laboratory to initiate bioassays.

2.2. Bioassays

The experiment with pests was conducted in a completely randomized design (CRD) in factorial arrangement (seven treatments, six evaluation dates and two pest species). The treatments with the dose (100%) and lower dose (50%) were: T1: control, T2: chlorpyrifos 450 EC (1.25 L.ha⁻¹), T3: chlorpyrifos 450 EC (0.62 L.ha⁻¹), T4: thiamethoxam 250 WG (1.00 L.ha⁻¹), T5: thiamethoxam 250 WG (0.50 L.ha⁻¹), T6: teflubenzuron 150 CS (0.025 L.ha⁻¹) and T7: teflubenzuron 150 CS (0.0125 L.ha⁻¹). Likewise, the same treatments in CRD were applied to natural enemies in the (7 × 6 × 3) arrangement for the study of physiological selectivity. The choice of insecticides considered the products registered for the control of whitefly *B. tabaci* in tomato plants in Brazil which are also used to control red spider mites. Spreader-sticker N-dodecyl benzene sulfonate sodium 320 EC was used at a concentration of 30 mL.100 L⁻¹ of solution in all treatments (Andrei, 2005). Only water and Spreader-sticker were used in the control. The use of the lower dose aimed to observe the impact of these insecticides on the insects when the former are broken down to half of their original concentration (Gusmão, Picanço, Gonring, & Moura, 2000).

When tomato plants were 35 days old (about 8 fully expanded leaves), a single spray of insecticides and control was performed, using a CO₂ pressurized sprayer with flat fan nozzles (80°) at 35 pounds.pol⁻² and calibrated to 150 L.ha⁻¹. Applications were made outdoors by spraying each insecticide on tomato plants. After insecticide application, the pots with plants were spaced at 30 × 30 cm on wooden benches (1.2 m wide × 6.0 m long × 1.3 high) inside a greenhouse.

Treated plants were left to dry in the greenhouse for two hours. After that, leaflets were removed from the treatments with neurotoxic insecticides (chlorpyrifos and thiamethoxam) and the control (four repetitions). They were placed on Petri dishes (9.0 cm wide and 2.0 cm tall) in order to cover the entire bottom of the dishes, which were covered with organza attached with elastic bands. Ten adults of *B. tabaci* were released in each treatment. Ten adult mites were transferred to another dish in the same treatment. The Petri dishes were taken to an incubator at an air temperature of 25 ± 0.5 °C and relative humidity of 75 ± 5%. Twenty-four hours later, the number of dead insects per experimental unit was assessed. Insects that were not able to move were considered to be dead.

The leaves of the plants treated with the growth regulating insecticide (teflubenzuron) were removed and the petioles were immersed in 25 ml transparent vials and introduced into polyethylene bottles through an opening 15 cm wide × 15 cm high on the side of the bottle, which was then sealed with organza (four repetitions). Ten whitefly nymphs were introduced inside a bottle with the aid of a fine brush. Ten nymphs of the red spider mite were introduced in another bottle, in the same treatment. The bottles were in a lit environment in the laboratory for six days, enough for the growth regulating insecticide to produce an effect. Nymphs that did not move on to the adult stage were considered to be dead.

As for the natural enemies, 10 adults of *O. insidiosus*, *C. sanguinea* and *C. flavipes* were released in each plate properly identified with the treatment and four repetitions. A 10% honey solution was added in each experimental unit. The Petri dishes were taken to an incubator at a temperature of 25 ± 0.5°C and relative humidity of 75 ± 5%. Twenty-four hours later, estimates of the dead insects per sample unit were performed. Insects were considered to be dead when they were not able to crawl or fly. Every seven days, leaves were taken off the plants and the process above-mentioned was repeated with the pests and natural enemies, with evaluation of mortality at up to 35 days after application.

2.3 Sublethal Effect

To evaluate the sublethal effect of insecticides on predators, behavioral tests were performed. The same insecticides used in toxicity tests were used to determine the sub-lethal effect. The experiment

used a completely randomized design (CRD) with three insecticides (dose = 1/10 the recommended dose) and the control, with 20 repetitions (each insect was considered as a repetition).

Tomato leaflets uncontaminated by insecticides were immersed in insecticide solutions for five seconds. The leaves were dried for two hours and then put in test tubes 2 cm wide and 15 cm high, whose inlet was sealed with hydrophobic cotton. One adult of *O. insidiosus*, *C. sanguinea* and *C. flavipes* was placed in each tube, thus forming an experimental unit. The tubes were taken to an incubator at a temperature of 25 ± 0.5 °C and relative humidity $75 \pm 5\%$. The insects were exposed to treatments without food for six hours. Each treatment was performed at one hour intervals to reduce exposure time error.

A styrofoam ring 2.5 cm thick, 40 cm wide was made, surrounded with a white cardstock strip, 10 cm wide, vertically positioned to prevent the escape of insects by the sides of this arena. This ring was divided into four quadrants of equal size. Five red spider mites and five whitefly nymphs were released in the center of each quadrant. They had been recently killed in a refrigerator at a temperature of -2 °C. The natural enemy was released in the center of the ring, and finally, a behavioral ethogram was made to analyze the sequence of individual behaviors in order to determine which behavioral activities would be evaluated.

2.4 Data Analysis

Mortality results obtained for the two pest species and three species of predators were adjusted for mortality that occurred in the control using the formula by Abbott (1925). The results were transformed into $\arcsin(x/100) * 0.5$ for the analysis of variance and the Scott-Knott test at $p < 0.05$.

The evaluation of insecticide effectiveness used a criterion of 80% mortality for *B. tabaci* and *T. evansi*. This criterion is required for registration of insecticides by the Ministry of Agriculture in Brazil. Thus, the insecticide was considered effective if it caused mortality equal to or greater than 80%. Also, the insecticides were classified as non-selective and highly toxic (mortalities between 100-70%), moderately selective or moderately toxic (69-30% of mortality) or selective and having low toxicity (mortality between 29-0%).

The criterion for assessing selectivity was based on the concept of physiological selectivity, in which the insecticide, at the same dose, is classified as selective if it causes higher mortality to the pest than to its natural enemy. It is considered as non-selective if causes the same mortality to both of them. If the insecticide causes higher mortality to the natural enemy than to the pest, it is considered to be harmful (Pedigo & Rice, 2005).

To evaluate the sublethal effect of insecticides on the three predators, the frequencies of their main behavioral activities was calculated, and those with frequency above 20% were selected for comparison of behavior between species in each treatment. Comparisons between the times of each selected behavior were based on a 95% confidence interval of each species in each treatment.

3. Results and Discussion

3.1 Effectiveness and Selectivity

There were statistically significant differences in mortality of pests *B. tabaci* and *T. evansi* for treatments ($F = 3.27$, $df = 6, 28$, $P = 0.01367$), days after application ($F = 27.05$, $df = 5, 28$, $P < 0.00001$), interactions between treatments and days after application ($F = 2.45$, $df = 30, 28$, $P = 0.00826$), interactions between species and treatments ($F = 3.91$, $df = 12, 28$, $P = 0.0381$) and interactions between species and days after application ($F = 4.43$, $df = 10, 180$, $P < 0.0001$).

Insecticides at the dose and lower dose were not efficient to control the adults and nymphs of mite *T. evansi*. On the other hand, after 24 hours (day 1), the growth regulating insecticide

teflubenzuron was effective against whitefly nymphs, with a mortality of 99.1% when using the dose (100%). In contrast, neurotoxic insecticides chlorpyrifos and thiamethoxam were efficient against adult whiteflies. However, no insecticide was efficient against *T. evansi*. The efficiency of all tested insecticides decreased when lower dose was used (50%) and over days after application. All insecticides tested (dose and lower dose) had lost their effectiveness at seven days after application (Table 1).

Table 1. Mean Mortality \pm standard error (%) of *Bemisia tabaci* and *Tetranychus evansi* based on two doses (50 and 100%) of three insecticides over days after application

Insecticide	Mortality (%) ¹					
	Days after application					
	1 st	7 th	14 th	21 st	28 th	35 th
<i>Bemisia tabaci</i> adults						
-Chlorpyr.						
100%	68.5 \pm 11.3 AaA	2.5 \pm 1.6 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
50%	12.0 \pm 2.6 AbA	2.5 \pm 1.6 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
-Thiamet.						
100%	81.2 \pm 5.3 AcA	10.0 \pm 6.3 BaA	10.0 \pm 7.8 BaA	0.0 \pm 0.0 CaA	0.0 \pm 0.0 CaA	0.0 \pm 0.0 CaA
50%	40.9 \pm 7.5 AdA	1.2 \pm 1.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
-Control	2.5 \pm 1.6 AbA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA
<i>Tetranychus evansi</i> adults						
-Chlorpyr.						
100%	21.0 \pm 0.0 AaB	77.4 \pm 4.1 BaB	10.0 \pm 6.2 CaA	0.0 \pm 0.0 DaA	0.0 \pm 0.0 DaA	0.0 \pm 0.0 DaA
50%	24.7 \pm 11.2 AbA	38.7 \pm 15.0 AbB	5.0 \pm 3.1 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
-Thiamet.						
100%	33.7 \pm 8.0 AbB	33.7 \pm 8.4 AbB	16.2 \pm 6.8 AaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
50%	28.7 \pm 11.7 AbB	11.2 \pm 4.4 BcA	11.1 \pm 5.5 BaA	0.0 \pm 0.0 CaA	0.0 \pm 0.0 CaA	0.0 \pm 0.0 CaA
-Control	0.0 \pm 0.0 AcA	0.0 \pm 0.0 AdA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA
<i>Bemisia tabaci</i> nymphs						
-Teflub.						
100%	99.1 \pm 0.6 AaA	3.7 \pm 1.1 BaA	1.2 \pm 0.4 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
50%	70.6 \pm 8.9 AbA	3.7 \pm 1.8 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
-Control	0.0 \pm 0.0 AcA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA
<i>Tetranychus evansi</i> nymphs						
-Teflub.						
100%	53.7 \pm 11.0 AaB	5.0 \pm 1.7 BaA	2.5 \pm 0.4 BaA	1.2 \pm 0.1 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
50%	42.5 \pm 16.4 AaB	3.7 \pm 0.8 BaA	3.6 \pm 1.8 BaA	0.2 \pm 0.0 BaA	0.0 \pm 0.0 BaA	0.0 \pm 0.0 BaA
-Control	0.0 \pm 0.0 AbA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA	0.0 \pm 0.0 AaA

¹Means followed by the same uppercase letter on the line (comparison of dose and lower dose of each insecticide with days after application), lowercase letter in the column (comparison of insecticide treatments) or italics uppercase letter in the column (comparison of tolerance between species to the same dose of each insecticide at the same stage of development [adults with adults, nymphs with nymphs]), do not differ by the Scott-Knott test ($P < 0.05$). -Chlorpyr. = chlorpyrifos; -Thiamet = Thiamethoxam.; -Teflub = Teflubenzuron.

Table 2. Mean mortality ± standard error (%) of adults of *Cycloneda sanguinea*, *Orius insidiosus* and *Chauliognathus flavipes* based on two doses of three insecticides used to control whitefly *Bemisia tabaci* and red spider mite *Tetranychus evansi* over days after application

Insecticide	Mortality (%) ¹					
	Days after application					
	1 st	7 th	14 th	21 st	28 th	35 th
<i>Cycloneda sanguinea</i>						
-Clorpyr.						
100%	100.0±12.4 AaA	75.0±12.0 BaA	60.0±10.7 BbA	47.5±8.2 CaA	22.5±12.4 DaA	7.5±1.2 EaA
50%	35.0±3.3 AbA	5.0±3.2 BbB	5.0±1.5 BcA	5.0±0.0 BbA	5.0±1.0 BbA	2.5±0.3 BaA
-Thiamet.						
100%	100.0±0.0 AaA	97.5±1.6 AcA	100.0±0.0 AaA	35.0±8.2 BcA	7.5±5.8 CbA	2.5±1.0 CaA
50%	27.5±15.9 AbA	0.0±0.0 BbA	0.0±0.0 BcB	0.0±0.0 BbA	0.0±0.0 BbA	0.0±0.0 BaA
-Teflub.						
100%	10.0±3.8 AbB	0.0±0.0 AbB	0.0±0.0 AcA	0.0±0.0 AbA	0.0±0.0 AbA	0.0±0.0 AaA
50%	10.0±3.8 AcB	0.0±0.0 AbA	0.0±0.0 AcA	0.0±0.0 AbA	0.0±0.0 AbA	0.0±0.0 AaA
-Control	2.5±1.6 AdA	0.0±0.0 AbA	0.0±0.0 AcA	0.0±0.0 AbA	0.0±0.0 AbA	0.0±0.0 AaA
<i>Orius insidiosus</i>						
-Clorpyr.						
100%	85.0±7.5 AaA	77.5±4.9 AaB	30.0±6.0 BaB	18.7±5.5 CaB	8.7±4.4DaA	0.0±0.0 DaA
50%	57.5±14.2AbB	47.5±6.2 AbA	0.0±0.0 BbA	0.0±0.0 BbA	0.0±0.0BbA	0.0±0.0 BaA
-Thiamet.						
100%	68.7±11.7 AbB	20.0±3.8 BcC	25.0±4.2 BaB	0.0±0.0 CbB	0.0±0.0CbA	0.0±0.0 CaA
50%	56.2±8.2 AbB	2.5±1.6 BdA	3.7±1.8 BbB	0.0±0.0 BbA	0.0±0.0BbA	0.0±0.0 BaA
-Teflub.						
100%	85.0±4.2AaA	30.0±6.0 BcA	10.0±3.8 CbA	1.2±0.1 CbA	1.2±1.0CbA	0.0±0.0 CaA
50%	32.0±8.4AcA	0.0±0.0 BdA	3.6±1.8 BbA	0.2±0.0 BbA	0.1±0.1BbA	0.0±0.0 BaA
- Control	3.7±1.0AdA	0.0±0.0 AdA	0.0±0.0 AbA	0.0±0.0 AbA	0.0±0.0AbA	0.0±0.0 AaA
<i>Chauliognathus flavipes</i>						
-Clorpyr.						
100%	35.0±10.1 AaB	37.5±4.9 AaC	26.2±3.7 AaB	7.5±2.5 BaB	1.3±0.0 BaA	0.0±0.0 BaA
50%	7.5±4.9 AbC	10.0±6.2 AbB	10.0±4.6 AbA	6.2±3.7 AaA	0.0±0.0 AaA	0.0±0.0 AaA
-Thiamet.						
100%	47.5±4.9 AaB	40.0±7.0 AaB	36.2±6.8 AaB	5.0±1.9 BaB	0.0±0.0 BaA	0.0±0.0 BaA
50%	7.5±4.4 AbC	7.5±4.9 AbA	12.5±4.1 AbB	3.7±1.8 AaA	0.0±0.0 AaA	0.0±0.0 AaA
-Teflub.						
100%	0.0±0.0 AcB	0.0±0.0 AbB	2.5±1.6 AbA	1.2±0.3 AaA	0.0±0.0 AaA	0.0±0.0 AaA
50%	0.0±0.0 AcB	0.0±0.0 AbA	0.5±0.3 AbA	0.3±0.3 AaA	0.0±0.0 AaA	0.0±0.0 AaA
- Control	0.0±0.0 AcA	0.0±0.0 AbA	0.0±0.0 AbA	0.0±0.0 AaA	0.0±0.0 AaA	0.0±0.0 AaA

¹Means followed by the same uppercase letter on the line (comparison of doses of each insecticide with days after application), lowercase letter in the column (comparison of insecticide treatments) italics uppercase letter in the column (comparison of tolerance between species to the same dose of each insecticide), do not differ by the Scott-Knott test (P <0.05). Chlorpyr. = chlorpyrifos; -Thiamet = Thiamethoxam.; -Teflub = Teflubenzuron.

Lack of insecticide effectiveness for mites may be due to detoxifying ability of this pest to organophosphate insecticides, neonicotinoids and benzoylphenyl ureas. The presence of monooxygenases with affinity for organophosphates may be associated with tolerance of *T. avansi* to this class of insecticide. This detoxification of the enzyme system transforms lipophilic insecticides into polar metabolites that are excreted (Brattsten, Holyoke Jr., Leeper, & Raffa, 1986). The organophosphate insecticide endosulfan 350 EC at 2.0 L.ha⁻¹ provided a higher percentage of attacked plants than the control. The possible mechanisms for the lack of effectiveness of neonicotinoids and benzoylphenyl ureas are yet to be elucidated. Lee, Clarke, Jenner, and Williamson (1990) found that benzoylphenyl ureas caused no mortality to adults of mite *Tetranychus urticae* Koch (Acari: Tetranychidae).

The insecticide thiamethoxam (neonicotinoid) and teflubenzuron (benzoylphenyl urea) have been effective for controlling *B. tabaci*. Bacci *et al.* (2007) observed high effectiveness (81%) of the insecticide neonicotinoid for the control of *B. tabaci*. These findings could have practical implications for managing populations of *B. tabaci*, indicating that the use of the recommended dose is essential for reducing the population in the field. Furthermore, as the neurotoxic insecticide and growth regulator performed an effective control, they can be used alternately to avoid problems of pest resistance (Palumbo, Horowitz, & Prabhaker, 2001). Although these insecticides have shown to be effective against whiteflies, populations should be monitored, as the insecticides are less effective as of the 7th day.

Significant differences were found in mortality of predators *C. sanguinea*, *O. insidiosus* and *C. flavipes* as for treatments ($F = 4.92$, $df = 6$, 180 , $P = 0.0002$), species ($F = 1$, 62 ; $2 df = 180$, $p = 0.0214$) days after application ($F = 1.83$, $df = 5.180$, $P = 0.0329$), interactions between species and treatments ($F = 1.88$; $df = 12$, 180 , $P = 0.0293$), interactions between treatments and days after infestation ($F = 1.79$, $df = 30$, 180 , $P = 0.0416$) and interactions between species and days after application ($F = 4.43$, $df = 10$, 180 , $P < 0.0001$).

In general, all insecticides (dose) were selective as of the 28th day after application. Moreover, when using one half of the active ingredient, the insecticides showed average and high selectivity to natural enemies on all days after application (Table 2).

On the 1st day, the insecticide teflubenzuron was selective, at the two tested doses, to *C. sanguinea* and *C. flavipes*. Thiamethoxam was selective (lower dose) to *C. sanguinea* and *C. flavipes*. Chlorpyrifos was selective to *C. flavipes* and moderately selective to *C. sanguinea* and *O. insidiosus* in lower dose. However, chlorpyrifos was moderately selective to *C. flavipes* when applied at the recommended dose. Thiamethoxam was moderately selective to *O. insidiosus* (dose and lower dose) and *C. flavipes* (dose). Furthermore, teflubenzuron (at the lower dose) was moderately selective to *O. insidiosus*. The insecticide chlorpyrifos was not selective to *C. sanguinea* and *O. insidiosus* (dose), but when half the doses were used, mortalities decreased. In contrast, the insecticide thiamethoxam (dose) was not selective to *C. sanguinea* (Table 2).

All insecticides were selective or moderately selective as of 21 days after application. However, the insecticide thiamethoxam was not selective to *C. sanguinea* until the 14th day. The species *O. insidiosus* and *C. flavipes* were also more tolerant to the dose of thiamethoxam than *C. sanguinea*. *C. flavipes* was the most tolerant to the lower dose of this insecticide, and *O. insidiosus* was in an intermediate situation. *C. flavipes* was also more tolerant to the dose and lower dose of chlorpyrifos and teflubenzuron than *C. sanguinea* and *O. insidiosus* (Table 2).

The high toxicities (no selectivity) of the insecticides chlorpyrifos and thiamethoxam to predators *C. sanguinea* and *O. insidiosus*, and the average selectivity of these insecticides to *C. flavipes* may be due to physicochemical properties of the insecticides and tolerance differences between the three species of predators studied. The high toxicity of chlorpyrifos to natural enemies may lie in the pro-insecticide activity of these organophosphates - because it undergoes certain

reactions while penetrating the body and becomes more toxic - and also in the lipophilic nature of the molecule. The difference in tolerance between species may be associated with the thickness and lipid composition of the cuticle of insects. Thus, considering that lipophilicity is inversely proportional to solubility of the insecticide in water, lipophilic compounds generally enter the body of the insect at a higher dose, given the similarity with their cuticle (Leite, Picanço, Guedes, & Gusmão, 1998). These explanations are possible since the organophosphate chlorpyrifos has very low solubility in water (2 ppm), while the neonicotinoid thiamethoxam is more soluble (610 ppm) (Berg, Sine, Meister, & Poplyk, 2003).

In addition to the composition of the cuticle, the body size of the predator *C. flavipes*, bigger than that of the other predators, could be the reason for the improved tolerance of this natural enemy compared with *O. insidiosus* and *C. sanguinea*. Thus, the selectivity of thiamethoxam and teflubenzuron in favor of this insect may be associated with differences in the dose of penetration, target site and/or enzyme detoxification in the larger predator (Hornsby, Wauchope, & Herner, 1996).

3.2 Sublethal Effect

12 behavioral categories have been found for predators *C. sanguinea*, *O. insidiosus* and *C. flavipes*. Behaviors with frequencies above 20% were foraging (food search), stationary (motionless), moving (mobile), ambulatory (still with motion of any member of the body), licking (except for *O. insidiosus*) and feeding (capturing prey and eating) (Table 3). These behavioral categories varied between species for the different insecticides tested (Figure 1).

Table 3. Frequency [%] of the main behavioral activities by predators *Cycloneda sanguinea*, *Orius insidiosus* and *Chauliognathus flavipes* subjected to treatments with insecticide and without insecticide

Behavior	Species	Behavioral activities
1. Moving	<i>C. sanguinea</i> , <i>O. insidiosus</i> and <i>C. flavipes</i>	- Still (motionless) [85,00] - Moving (slow or fast) [92,00] - Ambulatory (still and moving its legs only) [32,00] - Search (food search) [38,00]
2. Licking itself	<i>C. sanguinea</i> and <i>C. flavipes</i>	- Licking (licks legs and wings) [47,00]
3. Rubbing	<i>C. sanguinea</i> and <i>C. flavipes</i>	Legs on wings [14,00] - Front legs with back legs [4,00] - Wings with wings [7,00]
4. Flight	<i>C. sanguinea</i> , <i>O. insidiosus</i> and <i>C. flavipes</i>	- Spreads wings but does not fly [15,00] - Spreads wings and flies [11,00]
5. Feeding	<i>C. sanguinea</i> , <i>O. insidiosus</i> and <i>C. flavipes</i>	- Does not feed (touches food but does not eat) [15,00] - Feeds (touches food and eats) [58,00]

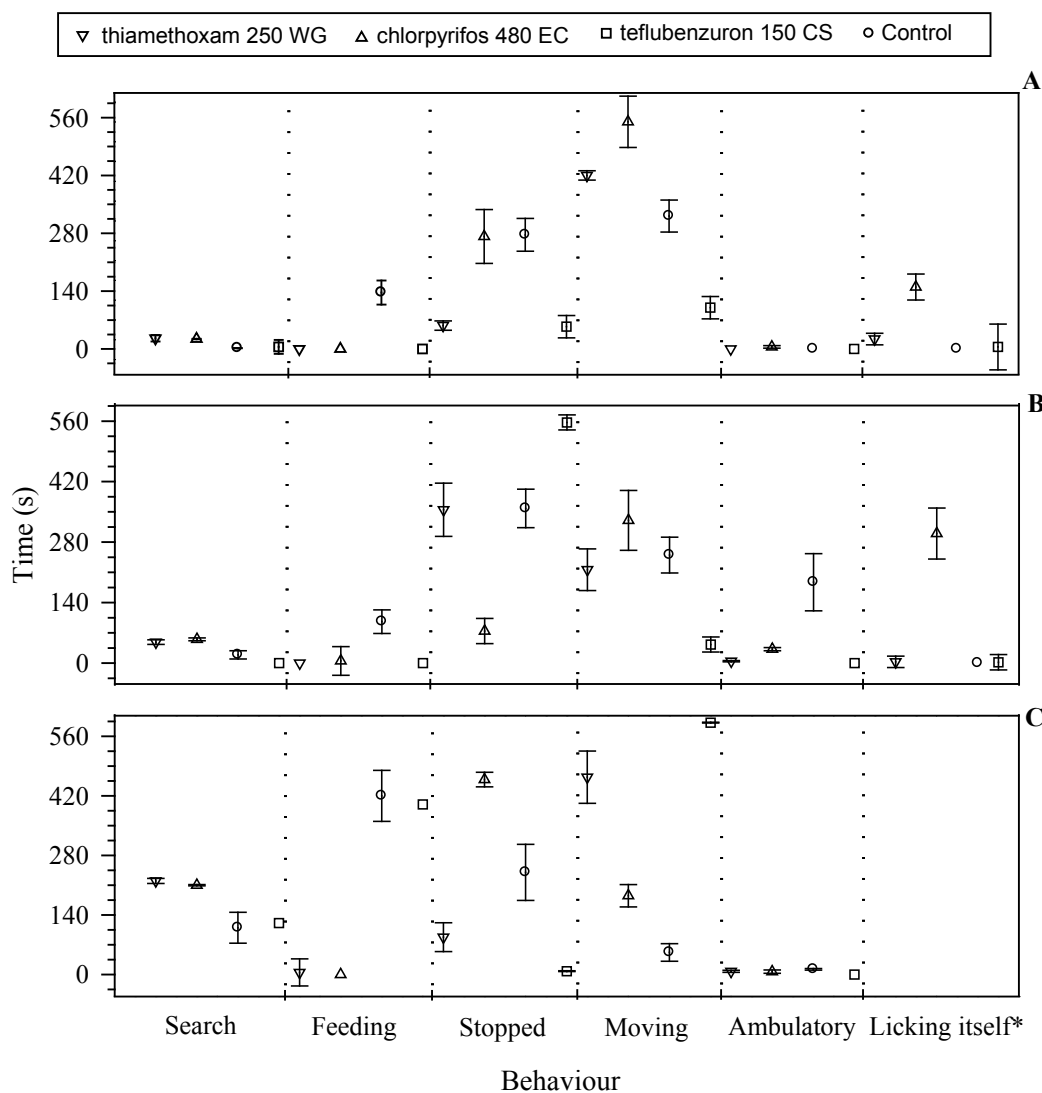


Figure 1. Effect of insecticides thiamethoxam 250 WG, chlorpyrifos 480 EC and teflubenzuron CS 150 at average time(s) for each behavioral activity undertaken by predators *Cycloneda sanguinea* (A), *Orius insidiosus* (B) and *Chauliognathus flavipes* (C). * asterisk on behavioral activity licking itself shows that this behavior was not evaluated for *O. insidiosus*. The symbols (\perp and \top) around the mean represent the confidence interval (95% CI)

All predators had more time to search for food when subjected to insecticides chlorpyrifos and thiamethoxam compared the the control and teflubenzuron (Figure 1 A, B and C). *C. flavipes* spent more time feeding when exposed to the insecticide teflubenzuron and the control than to the insecticides thiametoxam and chlorpyrifos (Figure 1 C). In contrast, predators *O. insidiosus* and *C. sanguinea* spent more time feeding when in contact with the control, than all other insecticides (Figure 1 A and B).

Regarding the behavioral category of moving, *C. flavipes* spent more time moving under the insecticide teflubenzuron, followed by thiamethoxam, chlorpyrifos and the control. The predator *O. insidiosus* moved more when in contact with the insecticide chlorpyrifos, followed by thiamethoxam and the control, and finally, teflubenzuron. *C. sanguinea* moved more when exposed to the treatment other insecticides (Figure 1 A, B and C).

In general, the greatest movement of the predators under the insecticides shows that these products have repellency to these natural enemies. Repellency is the removal of the food source without direction (Schaller, 2008). Although there are no previous studies on these insects, there are models of insecticide effect on behavior for other predators. Thornham, Blackwell, Evans, Wakefield, and Walters (2008) observed that the insecticide of the group of organophosphates increased the movement of the predator *Coccinella septempunctata* (Coleoptera: Coccinellidae). Foraging and feeding time may be inversely related, because *C. sanguinea* has low foraging time and high feeding time in the control and when in contact with teflubenzuron. All predators spent more time licking themselves after exposed to the insecticide chlorpyrifos than to the other treatments (Figure 1 A and B).

The effectiveness of predators is determined by their search strategy. The search behavior is usually done fast, with linear movements (Hodek, & Honek, 1996). Thus, mobility and orientation are essential to find the prey, kill it and feed (Schaller, 2008). The sensory system of insects is responsible for enabling the orientation in the environment through the assimilation of external stimuli. The faster an insect finds food, the more preys it can feed on and, therefore, the greater its reproductive success (Price, 1997). Thus, insecticides that reduced the foraging time of predators *C. sanguinea*, *O. insidiosus* and *C. flavipes* probably interfered in their sensory system. Singh, Walters, Port, and Northing (2004) observed low rate of aphid consumption by adults and larvae of the coccinellid *Coccinella septempunctata* (Coleoptera: Coccinellidae) whose substrate was treated with the organophosphate insecticide.

Once the need for control of *B. tabaci* and *T. evansi* was determined by sampling, product selection must take into consideration the effectiveness in control and selectivity to predators, because these predators are important in regulating populations of these pests. The differences in tolerance of wasp species to insecticides show the importance of correct identification of the species found in the tomato agroecosystem. In general, the insecticides neonicotinoid and pyrethroid were selective to three predatory species and had lower sublethal impact compared with the organophosphate. It is expected that the selectivity of these insecticides is greater in the field, given the fact that the exposure of these predators to insecticides is lower in natural conditions. Thus, in order to preserve the predators, after the need for control is detected, the insecticides thiamethoxam and teflubenzuron can be used alternately.

Acknowledgements

The funding and fellowships provided by the following Brazilian agencies are acknowledged: CAPES Foundation (Ministry of Education), National Council of Scientific and Technological Development (CNPq) and Minas Gerais State Foundation for Research Aid (FAPEMIG).

References

- [1] Abbott, W. S. (1925). A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology*, 18(2), 265-267.
- [2] Andrei, E. (2005). *Compêndio de defensivos agrícolas* (7th ed.). São Paulo, SP: Andrei Editora Ltda.
- [3] Bacci, L., Crespo, A. L. B., Galvan, T. L., Pereira, E. J. G., Picanço, M. C., Silva, G. A., & Chediak, M. (2007). Toxicity of insecticides to the sweetpotato whitefly (Hemiptera: Aleyrodidae) and its natural enemies. *Pest Management Science*, 63(7), 699-706.
- [4] Berg, G. L., Sine, C., Meister, R. T., & Poplyk, J. (Eds.). (2003). *Farm chemicals handbook*. Willoughby, OH: Meister.

- [5] Brattsten, L. B., Holyoke Jr., C. W., Leeper, J. R., & Raffa, K. F. (1986). Insecticide resistance: Challenge to pest management and basic research. *Science*, 231(4743), 1255-1260.
- [6] Fernandes, F. L., Picanço, M. C., Fernandes, M. E., Chediak, M., Tomé, H. V. V., & Gontijo, P. C. (2008). Impacto de inseticidas e acaricidas sobre organismos não-alvo. In L. Zambolim, M. C. Picanço, A. A. da Silva, L. R. Ferreira, F. A. Ferreira, & W. C. de Jesus Junior (Eds.), *Produtos fitossanitários* (pp. 575-606). Viçosa, MG: UFV/DFP.
- [7] Gusmão, M. R., Picanço M. C., Gonring, A. H. R., & Moura, M. F. (2000). Seletividade fisiológica de inseticidas a Vespidae predadores do bicho-mineiro-do-cafeeiro. *Pesquisa Agropecuária Brasileira*, 35(4), 681-686.
- [8] Hendrix, D. L., & Wei, Y. (1992). Detection and elimination of honeydew excreted by the sweetpotato whitefly feeding upon cotton. In J. M. Brown, & T. C. Nelson (Eds.), *Proceedings of the Beltwide cotton production conference* (vol. 2, p. 671-673). Memphis, TN.: National Cotton Council.
- [9] Hodek, I., & Honek, A. (1996). *Ecology of coccinellidae (series entomologica 54)*. Dordrecht, NL: Kluwer Academic.
- [10] Hornsby, A. G., Wauchope, R. D., & Herner, A. E. (1996). *Pesticide properties in the environment*. New York, NY: Springer.
- [11] Lee, A. S., Clarke, B. S., Jenner, D. W., & Williamson, W. (1990). Cytochemical demonstration of the effects of the acilureas flufenoxuron and diflubenzuron on the incorporation of chitin into insect cuticle. *Pesticide Science*, 28, 367-675.
- [12] Leite, G. L. D., Picanço, M., Guedes, R. N. C., & Gusmão, M. R. (1998). Selectivity of insecticides with and without mineral oil to *Brachygastra lecheguana* (Hymenoptera: Vespidae), a predator of *Tuta absoluta* (Lepidoptera: Gelechiidae). *Ceiba*, 39(1), 191-194.
- [13] Lourenção, A. L., Yuki, V. A., & Alves, S. B. (1999). Epizootia de *Aschersonia* cf. *goldiana* em *Bemisia tabaci* (Homoptera: Aleyrodidae) biótipo B no Estado de São Paulo. *Anais da Sociedade Entomológica do Brasil*, 28(2), 343-345.
- [14] Oliveira, E. E., Oliveira, C. L., Sarmiento, R. D. A., Fadini, M. A. M., & Moreira, L. R. (2005). Aspectos biológicos do predador *Cycloneda sanguinea* (Linnaeus, 1763) (Coleoptera: Coccinellidae) alimentado com *Tetranychus evansi* (Baker e Pritchard, 1960) (Acari: Tetranychidae) e *Macrosiphum euphorbiae* (Thomas, 1878) (Homoptera: Aphididae). *Bioscience Journal*, 21(2), 33-39.
- [15] Palumbo, J. C., Horowitz, A. R., & Prabhaker, N. (2001). Insecticidal control and resistance management for *Bemisia tabaci*. *Crop Protection*, 20(9), 739-765.
- [16] Pedigo, L. P., & Rice, M. E. (2005). *Entomology and pest management* (5th ed.). New Jersey, NJ: Prentice Hall.
- [17] Price, P. W. (1997). *Insect ecology* (3rd ed.). New York, NY: John Wiley & Sons.
- [18] Schaller, A. (Ed.). (2008). *Induced plant resistance to herbivory*. Berlim, BE: Springer.
- [19] Singh, S. R., Walters, K. F. A., Port, G. R., & Northing, P. (2004). Consumption rates and predatory activity of adult and fourth instar larvae of the seven spot ladybird, *Coccinella septempunctata* (L.), following contact with dimethoate residue and contaminated prey in laboratory arenas. *Biological Control*, 30(2), 127-133.
- [20] Thornham, D. G., Blackwell, A., Evans, K. A., Wakefield, M., & Walters, K. F. A. (2008). Locomotory behaviour of the seven-spotted ladybird, *Coccinella septempunctata*, in response to five commonly used insecticides. *Annals of Applied Biology*, 152(3), 349-359.

- [21] Toscano, L. C., Boiça Júnior, A. L., & Maruyama, W. I. (2004). Avaliação de aspectos fisiológicos de três genótipos de tomateiro infestados por *Bemisia tabaci* (Gennadius) biótipo B (Hemiptera: Aleyrodidae). *Neotropical Entomology*, 33(6), 777-782.

Copyrights



Copyright for this article is retained by the author(s), with first publication rights granted to the journal. This is an open-access article distributed under the terms and conditions of the [Creative Commons Attribution 4.0 International License](https://creativecommons.org/licenses/by/4.0/).

