

Research Article

## Evaluation of toxicity of some biocompatible insecticides on *Trichogramma brassicae* and *T. evanescens* under laboratory and semi-field conditions

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**Abstract:** The effects of four insecticides, abamectin (1500 and 750mg l<sup>-1</sup>), emamectin benzoate (1000 and 500mg l<sup>-1</sup>), acetamiprid (500 and 250mg l<sup>-1</sup>), and flubendiamide (500 and 250mg l<sup>-1</sup>), were studied on different preimaginal stages of *T. brassicae* and *T. evanescens*, the egg parasitoids of tomato leaf miner *Tuta absoluta* (Lepidoptera: Gelechiidae). Parasitized eggs of the Angoumois grain moth *Sitotroga cerealella* (Lepidoptera: Gelechiidae) were treated by the dipping method at the larval, prepupal, and pupal stages of the parasitoid. For persistence evaluation, the insecticides were applied at the recommended concentration on tomato plants by a hand sprayer till runoff point. Plants were maintained under a transparent polyethylene rain cover in the field. Leaves of the treated tomato plants were sampled and transferred to the laboratory at time intervals of 3, 5, 16, and 31 days after application. Based on our study, abamectin was the most harmful insecticide for immature stages of both parasitoids *T. brassicae* and *T. evanescens*. Treatment by abamectin at the pupal stage had more adverse effects compared to prepupal or larval stages. Acetamiprid with 30.5% and 31.6% mortality in less than five days was classified as the short-lived insecticide for *T. brassicae* and *T. evanescens*, respectively. The same result was obtained in flubendiamide treatment which caused 27.2% and 26.1% mortality to the parasitoids, respectively. Abamectin with 16.1% and 13.8% mortality in less than 16 days was slightly persistent. However, emamectin benzoate with 13.3% and 15.5% mortality in less than 30 days was classified as moderately persistent for those two species, respectively. Therefore, flubendiamide and acetamiprid were non-harmful to both *T. brassicae* and *T. evanescens* wasps and are good candidates to be incorporated into IPM programs in combination with biological agents for the control of tomato leaf miner *T. absoluta*. By contrast, emamectin and abamectin should be used with greater care as a part of an IPM procedure.

**Keywords:** *Tuta absoluta*, egg parasitoids, insecticides, persistency, IPM

### Introduction

The tomato leaf miner moth *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is one of

the most important pests of the tomato plant. The pest can produce between 10 and 12 generations in a year, and each female can lay 250-300 eggs in its lifetime (Barrientos *et al.*, 1998). Excess use of insecticides to control the tomato leaf miner has led to the appearance of resistant genotypes against different groups of insecticides, contamination of the environment, and increased pesticide residues on the produce (Siqueira *et al.*,

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2001). These issues have encouraged the use of biocompatible insecticides alongside biological agents for pest control. The pest is attacked by various biological control agents including egg parasitoids *Trichogramma* species which are the most prevalent parasitoids in the world (Knutson, 2005). According to Pratisoli and Parra (2001) family Trichogrammatidae has significantly contributed to reducing the populations of lepidopteran pests as well as the number of insecticide applications on tomato crops. However, nonselective insecticides can reduce the control potential of these biological agents (Desneux *et al.*, 2007; Vianna *et al.*, 2009; Croft, 1990; Bartlett, 1964). Many of these chemicals can maintain their deleterious effects for a long period of time after application (Zappala *et al.*, 2012). Due to the wide range effects of pesticides on natural enemies, it is necessary to study their side effects in order to adopt chemical and biological control for minimizing their negative impacts (Souza *et al.*, 2014). Insecticides have both lethal and sublethal effects on arthropods, thus in addition to death, they can adversely affect life parameters such as developmental rate, sex ratios, parasitism, and emergence rate (Poorjavand *et al.*, 2014). Acute toxicity is usually assessed after the organism is exposed to a chemical for a short period of time (e.g. a few hours to a few days), and the endpoint is the death of the organism (Stark and Banks, 2003). The estimated lethal concentration during acute toxicity tests may only be a partial measure of the deleterious effects. Sublethal effects are defined as effects, either physiological or behavioral, on individuals that survive exposure to a pesticide (Desneux *et al.*, 2007). Therefore, it is necessary to evaluate the lethal and sublethal effects of insecticides on the natural enemies. This evaluation provides a better understanding of the interaction of chemicals and the biological control agents in the system (Stark and Banks, 2003). Integrating biological control agents with pesticides should regulate arthropod pest populations without directly or indirectly affecting their natural enemies (Ruberson *et al.*, 1998). In some cases, the use of short-lived insecticides that only interfere slightly with biological control agents

should be a solution for effective pest control. Some studies have revealed the effects of many insecticides on different *Trichogramma* species (Hassan, 1998; Hewa-Kapuge *et al.* 2003; Jiu-Sheng *et al.*, 2009).

Abamectin is a chemical originally isolated from the soil bacterium *Streptomyces avermitilis* or generated through semisynthetic modifications. Abamectin stimulates the chloride channels that are regulated by the neurotransmitter glutamate causing paralysis in arthropods (Fisher and Mrozik, 1989). Emamectin benzoate has a strong effect against Lepidopteran pests. Similar to abamectin, emamectin belongs to the avermectin family of compounds, all of which exhibit toxicity to several pests. The benzoate salt of emamectin is widely used as an insecticide (Mistretta and Durkin, 2010). Acetamiprid, a neonicotinoid compound, is a synthetic “nicotine-like” chemical that binds tightly to the acetylcholine receptor site on the post-synapse nerve cell and causes nerve overstimulation. It is highly effective against Lepidoptera, and the insecticide is applicable for controlling pests of vegetables (Yamada *et al.* 1999). Flubendiamide acts as a stomach poison and oral intoxicant. It belongs to a new class, phthalic acid diamide. Flubendiamide has the novel biochemical action of affecting calcium ion balance, which is an effective control of lepidopteran pests (Das *et al.*, 2017).

The present study was designed to evaluate *T. brassicae* and *T. evanescence* for their susceptibility to four insecticides: abamectin, emamectin benzoate, acetamiprid, and flubendiamide when applied at the insects' immature stages.

In order to determine the most suitable insecticides for effective pest control, we also investigated the persistence rate of the insecticides at field conditions when applied at the recommended concentration according to IOBC procedure (Sterk *et al.*, 1999).

## Materials and Methods

### Rearing

The experiments were carried out in the Laboratory of Plant Protection at the Agricultural

and Natural Resources Research and Education Center of Markazi Province, Arak, Iran. The egg parasitoids *T. brassicae* and *T. evanescens* (Hymenoptera: Trichogrammatidae) were provided from the Biological Control Department of Plant Protection Research Institute (Tehran, Iran). Parasitoids were reared on the Angoumois grain moth, *Sitotroga cerealella* (Lepidoptera: Gelechiidae) in the laboratory under the controlled conditions of  $25 \pm 1$  °C,  $60 \pm 10\%$  RH, and a photoperiod of 16:8 h (L: D).

### Insecticides

The insecticides used were: abamectin (EC 1.8%Vertimec<sup>®</sup>, Golsam Co., Gorgan), emamectin benzoate (WG50% Proclimfit<sup>®</sup>, Syngenta Co., Swiss), acetamiprid (SP20% Mospilan<sup>®</sup>, Golsam Co., Gorgan), and flubendiamide (WG20%Takumi<sup>®</sup>, Syngenta Co., Swiss). Field recommended concentrations (RC) and half RC of the insecticides were used (Table 1).

### Immature stages bioassay

The effect of insecticides was studied on different preimaginal stages of *T. brassicae* and *T. evanescens*. Parasitized eggs of Angoumois grain moth *Sitotroga cerealella* (Lepidoptera: Gelechiidae) were treated by the dipping method at different intervals suggested by the IOBC working group (Costa et al., 2014; Sterk et al., 1999). The experiment was repeated six times. The control was treated with distilled water. Randomly selected egg cards, having 140 parasitized eggs were dipped into the solution of RC or half RC of each insecticide for 10 seconds on the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> day after the eggs were parasitized. These days corresponded to the developmental stages of

*Trichogramma* larvae (3 d), prepupae (6 d), and pupae (9 d). When the dipped eggs were dried, they were placed in glass tubes at the laboratory under the controlled conditions of  $25 \pm 1$  °C,  $60 \pm 10\%$  RH, and a photoperiod of 16:8 h (L: D). The parameter evaluated was adult emergence rate of parasitoids from each of the three stages and then divided by the control for calculation of emergence reduction. The insecticides were classified into the toxicity categories proposed by the IOBC working group for semi-field trials on beneficial arthropods: Class 1: harmless (< 30% mortality or effect), Class 2: slightly harmful (30%-79%), Class 3: moderately harmful (80%-99%), and Class 4: harmful (> 99%) (Costa et al., 2014; Sterk et al., 1999).

### Persistence rate evaluation

The insecticides were applied at RC on tomato plants by a hand sprayer till runoff point. Plants were maintained under a transparent polyethylene rain cover in the field. Leaves of the treated tomato plant were sampled and transferred to the laboratory at time intervals of 3, 5, 16 and 31 days after application. These intervals were proposed by the IOBC/WPRS Working Group (Costa et al., 2014; Sterk et al., 1999). The samples were placed in ventilated plastic Petri dishes for exposure to the adult parasitoids. The experiment was repeated six times on 30 adult individuals of each species. Assessment of mortality was made on the basis of 24h post-exposure time. Persistence rate of each insecticide was evaluated according to IOBC/WPRS Working Group. The categories under laboratory conditions included: A, short-lived (< 5days); B, slightly persistent (5-15days); C, moderately persistent (16-30days); D, Persistent (> 30 days) (Costa et al., 2014; Sterk et al., 1999).

**Table 1** Names, doses and manufacturers of the insecticides that were tested in the study.

Name of insecticides	Trade name	Recommended Concentration (ml/ha)	Manufacturer
Abamectin EC 1.8%	Vertimec	1500	Golsam, Gorgan
Emamectinbenzoate WG50 5%	Proclaim Fit	1000	Syngenta, Swiss
Acetamiprid SP20 20%	Mospilan	500	Golsam, Gorgan
Flubendiamide WG20 20%	Takumi	500	Syngenta, Swiss

### Statistical analysis

These experiments were carried out in a completely randomized design (CRD) with six replications. Data on percent efficacy were arcsine-square root transformed to normalize them and were subjected to one-way analyses of variance using SPSS software (SPSS Inc. 2007). Abbott's formula (Abbott, 1925) was used to correct mortality. The means were separated using least significant difference (LSD) test at 5% level of significance.

### Results

#### Emergence rate

Parasitoid emergence rate after treatment at larval, prepupal and pupal stages was reduced when treated by the recommended concentration of each insecticide. Maximum and minimum reduction of emergence for abamectin was 52.23%, 47.34% and 59.49%, and for flubendiamide was 25.44%, 13.27% and 20.25%, respectively (Table 2). The data for half RD of abamectin was 47.32%, 43.36% and 54.85%, and for flubendiamide was 20.98%, 7.96%, and 14.34%. There were significant differences among insecticides for their effects on emergence rate of the parasitoid at the larval ( $F = 259.40$ ;  $df = 4, 29$ ;  $P = 0.0001$ ), prepupal ( $F = 243.79$ ;  $df = 4, 29$ ;  $P = 0.0001$ ) and pupal stages ( $F = 624.72$ ;  $df = 4, 29$ ;  $P = 0.0001$ ) (Table 2).

#### *Trichogramma evanescens*

Reduction of *T. evanescens* emergence rate after treatment at the recommended concentration of abamectin was 50.68%, 45.49% and 53.78%, and for flubendiamide was 20.09%, 10.36% and 12.89% at the larval, prepupal and pupal stages, respectively (Table 3). These values for half RD of abamectin were 43.83, 40.99, 51.55%, and for flubendiamide were 16.43, 4.95 and 8.88%. There were significant differences among insecticides for their effects on emergence rate of the parasitoid at the larval ( $F = 202.50$ ;  $df = 4, 29$ ;  $P = 0.0001$ ), prepupal ( $F = 162.96$ ;  $df = 4, 29$ ;  $P = 0.0001$ ) and pupal stage ( $F = 256.18$ ;  $df = 4, 29$ ;  $P = 0.0001$ ) (Table 3).

### Persistency

#### *Trichogramma brassicae*

At three days post-treatment (= DPT), a significant difference was observed among various treatments for the rate of adult parasitoid mortality ( $F = 243.41$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Abamectin caused  $86.67 \pm 2.58\%$  mortality which was the most among all treatments, however, flubendiamide showed  $27.22 \pm 2.00\%$  mortality which was the least. At five DPT, abamectin caused  $63.33 \pm 2.11\%$  mortality which differed significantly from other treatments ( $F = 147.08$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Acetamiprid and flubendiamide with mean mortality of  $24.44 \pm 1.65\%$  and  $21.67 \pm 1.43\%$ , respectively, showed less lethal effect than abamectin but were not significantly different from one another. At 16 DPT emamectin caused  $41.11 \pm 1.64\%$  mortality which was the highest and significantly different from the other treatments ( $F = 63.83$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Acetamiprid and flubendiamide with mean mortality of  $11.11 \pm 1.40\%$  and  $10.56 \pm 1.34\%$  showed less lethal effects. At 31 DPT emamectin caused  $13.33 \pm 1.22\%$  mortality which still significantly differed from other treatments ( $F = 12.94$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Acetamiprid and flubendiamide with mean mortality of  $8.89 \pm 1.11\%$  and  $7.78 \pm 1.11\%$ , respectively, showed less lethal effect than emamectin with no statistical difference between the two (Table 4).

#### *Trichogramma evanescens*

At three DPT the rate of mortality differed significantly among various treatments ( $F = 151.564$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Abamectin caused  $85.56 \pm 2.94\%$  mortality which was the most among all treatments, however, flubendiamide showed  $26.11 \pm 1.59\%$  mortality which was the least. The mortality rates of emamectin, acetamiprid, and flubendiamide were  $61.67 \pm 2.54\%$ ,  $31.67 \pm 1.43\%$  and  $26.11 \pm 1.59\%$ , respectively. At five DPT, abamectin caused  $60.55 \pm 2.00\%$  mortality which differed significantly from other treatments ( $F = 94.323$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Acetamiprid and flubendiamide with mean mortality of  $26.11 \pm 1.59\%$  and  $20.56 \pm 1.59\%$ , respectively, showed less lethal effect than emamectin without any

statistical difference between them. At 16 DPT, emamectin was significantly different from other treatments ( $F = 43.601$ ;  $df = 4, 29$ ;  $P = 0.0001$ ). Acetamiprid and flubendiamide with mean mortality of  $12.22 \pm 1.11\%$  and  $10.56 \pm 1.59\%$  showed less effects than emamectin. At 31 DPT, emamectin significantly differed from

other treatments ( $F = 4.972$ ;  $df = 4, 29$ ;  $P = 0.004$ ). Acetamiprid and flubendiamide with mean mortality of  $9.44 \pm 1.59\%$  and  $7.22 \pm 1.34\%$ , respectively, showed less lethal effects than emamectin without any statistical difference between those two insecticides (Table 5).

**Table 2** Effects of insecticide treatments on emergence of *Trichogramma brassicae* when treated at larval, prepupal and pupal stages inside its host egg under laboratory conditions.

Treatment	Larvae (%emergence)	%Reduction in emergence of larvae (Category)	Prepupae (%emergence)	%Reduction in emergence of prepupae (Category)	Pupae (%emergence)	%Reduction in emergence of pupae (Category)
<b>Abamectin</b>						
RC	44.58 ± 1.34a	52.23 (2)	49.58 ± 1.19a	47.34 (2)	40.00 ± 3.02a	59.49 (2)
0.5RC	49.17 ± 2.24a	47.32 (2)	53.33 ± 2.26a	43.36 (2)	44.58 ± 1.98a	54.85 (2)
Control	93.33 ± 1.26e	0	94.17 ± 0.99d	0	98.75 ± 0.72e	0
<b>Emamectin</b>						
RC	50.00 ± 1.35b	46.43 (2)	51.67 ± 1.29a	45.13 (2)	61.25 ± 1.57b	37.97 (2)
0.5RC	54.17 ± 2.26b	41.96 (2)	56.67 ± 1.43a	39.81 (2)	67.08 ± 1.15b	29.07 (1)
Control	93.33 ± 1.26e	0	94.17 ± 0.99d	0	98.75 ± 0.72e	0
<b>Acetamiprid</b>						
RC	60.42 ± 0.87c	35.27 (2)	65.42 ± 1.36b	30.52 (2)	70.42 ± 1.88c	28.69 (1)
0.5RC	65.00 ± 1.89c	30.35 (2)	69.58 ± 1.42b	26.10 (1)	73.75 ± 2.02c	22.02 (1)
Control	93.33 ± 1.26e	0	94.17 ± 0.99d	0	98.75 ± 0.73e	0
<b>Flubendiamide</b>						
RC	69.58 ± 1.07d	25.44 (1)	81.67 ± 2.15c	13.27 (1)	78.75 ± 2.20d	20.25 (1)
0.5RC	73.75 ± 2.07d	20.98 (1)	86.67 ± 2.64c	7.96 (1)	84.58 ± 1.87d	14.34 (1)
Control	93.33 ± 1.26e	0	94.17 ± 0.99d	0	98.75 ± 0.72e	0

Means followed by the same letters are not significantly different (LSD test,  $p < 0.05$ ).

RC: Recommended concentration. Category 1: harmless, 2: slightly harmful.

**Table 3** Effects of insecticide treatment on the adult emergence of *Trichogramma evanescens* when treated at larval, prepupal and pupal stages inside its host egg under laboratory conditions.

Treatment	Larvae (%emergence)	%Reduction in emergence of larvae (Category)	Prepupae (%emergence)	%Reduction in emergence of prepupae (Category)	Pupae (%emergence)	%Reduction in emergence of pupae (Category)
<b>Abamectin</b>						
RC	45.00 ± 1.06a	50.68 (2)	50.42 ± 2.16a	45.49 (2)	43.33 ± 1.07a	53.78 (2)
0.5RC	51.25 ± 2.56a	43.83 (2)	54.58 ± 1.42a	40.99 (2)	45.42 ± 2.32a	51.55 (2)
Control	91.25 ± 1.48e	0	92.50 ± 1.18d	0	93.75 ± 1.19e	0
<b>Emamectin</b>						
RC	52.08 ± 1.67b	42.93 (2)	51.25 ± 1.38a	44.59 (2)	58.33 ± 2.33b	37.78 (2)
0.5RC	57.50 ± 1.44b	36.99 (2)	55.42 ± 3.49a	40.09 (2)	64.17 ± 1.31b	31.55 (2)
Control	91.25 ± 1.48e	0	92.50 ± 1.18d	0	93.75 ± 1.19e	0
<b>Acetamiprid</b>						
RC	60.00 ± 1.97c	34.24 (2)	63.75 ± 2.08b	31.08 (2)	67.50 ± 3.16c	8.00 (1)
0.5RC	63.33 ± 2.80c	30.59 (2)	67.92 ± 1.40b	26.57 (1)	72.08 ± 1.15c	23.11 (1)
Control	91.25 ± 1.48e	0	92.50 ± 1.18d	0	93.75 ± 1.19e	0
<b>Flubendiamide</b>						
RC	72.92 ± 2.17d	20.09 (1)	82.92 ± 2.29c	10.36 (1)	81.67 ± 2.31d	12.89 (1)
0.5RC	76.25 ± 1.18d	16.43 (1)	87.92 ± 1.40c	4.95 (1)	85.42 ± 1.22d	8.88 (1)
Control	91.25 ± 1.48e	0	92.50 ± 1.18d	0	93.75 ± 1.19e	0

Means followed by the same letters are not significantly different (LSD test,  $p < 0.05$ ).

RC: Recommended concentration. Category: 1: harmless, 2: slightly harmful.

**IOBC classification**

Results regarding the effects of the insecticides on different developmental stages of *T. brassicae* and *T. evanescens* revealed that flubendiamide was the most selective compound among the tested insecticides. However, abamectin, emamectin benzoate, and acetamiprid were slightly harmful to both parasitoids (Tables 2 and 3). Persistence tests based on IOBC classification for more than 30% mortality at each period of time revealed that both flubendiamide and acetamiprid had less than five days persistence. This result classified them as short-lived insecticides (class A) for *T. brassicae* wasps. Abamectin

with more than five days and less than 16 days effect was slightly persistent (class B). However, emamectin which persisted more than 16 days but less than 30 days was moderately persistent for the parasitoid (class C) (Table 4). For *T. evanescens* adult parasitoid, acetamiprid and flubendiamide with less than five days persistence were classified as short-lived insecticides (class A). Abamectin which persisted for more than five days but less than 16 days was slightly persistent (class B). Emamectin with persistence of more than 16 days and less than 30 days was moderately persistent for the parasitoid (class C) (Table 5).

**Table 4** Mortality of *Trichogramma brassicae* wasps after exposure to leaf residues of insecticides at 3, 5, 16 and 31 days post-treatment (DPT) at field conditions.

Insecticide	% Mortality (Mean $\pm$ SE)				Classification
	3 d	5 d	16 d	31 d	
Abamectin	86.67 $\pm$ 2.58a	63.33 $\pm$ 2.11a	16.11 $\pm$ 1.59a	12.22 $\pm$ 1.40a	B, Slightly persistent
Emamectin	60.56 $\pm$ 1.59b	51.11 $\pm$ 2.81b	41.11 $\pm$ 1.64b	13.33 $\pm$ 1.22a	C, Moderately persistent
Acetamiprid	30.56 $\pm$ 1.59c	24.44 $\pm$ 1.65c	11.11 $\pm$ 1.40b	8.89 $\pm$ 1.11ab	A, Short lived
Flubendiamide	27.22 $\pm$ 2.00c	21.67 $\pm$ 1.43c	10.56 $\pm$ 1.34b	7.78 $\pm$ 1.02b	A, Short lived

Means followed by the same letters are not significantly different (LSD test,  $p < 0.05$ ).

**Table 5** Mortality of *Trichogramma evanescens* wasps after exposure to leaf residues of insecticides at 3, 5, 16 and 31 days post-treatment (DPT) at the field conditions.

Insecticide	% Mortality (Mean $\pm$ SE)				Classification
	3 d	5 d	16 d	31 d	
Abamectin	85.56 $\pm$ 2.94a	60.55 $\pm$ 2.00a	13.89 $\pm$ 1.34a	10.55 $\pm$ 1.59a	B, Slightly persistent
Emamectin	61.67 $\pm$ 2.54b	52.78 $\pm$ 2.00a	43.89 $\pm$ 1.02b	15.56 $\pm$ 1.65ab	C, Moderately persistent
Acetamiprid	31.67 $\pm$ 1.43c	26.11 $\pm$ 1.59b	12.22 $\pm$ 1.11b	9.44 $\pm$ 1.59ab	A, Short lived
Flubendiamide	26.11 $\pm$ 1.59c	20.56 $\pm$ 1.59b	10.56 $\pm$ 1.59b	7.22 $\pm$ 1.34b	A, Short lived

Means followed by the same letters are not significantly different (LSD test,  $p < 0.05$ ).

**Discussion**

Our study revealed that abamectin was the most harmful insecticide for immature stages of both parasitoids. We also found that treatments with abamectin at the pupal stage had more adverse effects compared to prepupal or larval stages. In a similar study Jiu-Sheng *et al.* (2009) showed that field recommended concentration of

abamectin (7.2mg/L) severely reduced the emergence rate of *T. pretiosum* from treated eggs of *Corcyra cephalonica* (Stainton). The effect of abamectin was also evaluated by Hussain *et al.* (2015) on *T. chilonis* under laboratory conditions which had adverse effects on the emergence rate of adults. Those results were the same as our findings. In a similar study, by examining the effect of the field

recommended concentration of abamectin (480ml/100L) on larval and pupal stages of *T. pretiosum*, Khan and Ruberson (2017) found that abamectin significantly decreased the emergence percent of adult parasitoids. Similar to other insects, immature stages of *Trichogramma* have been considered less susceptible to pesticides than adults, because the eggshell of the host could provide protection. However, preimaginal mortality of *Trichogramma* parasitoids may be related to a phenomenon called partial emergence. C nsoli et al. (2001) who explained this phenomenon, reported that some chemicals might be unable to penetrate through the host eggshell, but the parasitoid might ingest the product during the opening of the emergence hole. When the parasitoid is cutting a small area of the host eggshell with its mandibles, a small quantity of the eggshell surface could be swallowed as well as the product residues that exist on the host surface. This phenomenon also explains why treatment at the pupal stage resulted in less emergence than the treatment at the larval stage because the first produces fresher residues at the time of parasitoid emergence. We also found RD of abamectin on tomato plants in the field was slightly persistent (5-15days) for both parasitoids when exposed to the residues. This result is the same as those of Hassan (1998) who classified abamectin as slightly persistent for adult *T. cacoeciae*, or Jiu-Sheng et al. (2009) who declared abamectin to be slightly persistent for *T. chilonis* at greenhouse conditions. Based on the results of Sabahi et al. (2009) regarding the effect of abamectin in semi-field conditions on *Lysiphlebus fabarum* (Marshall), abamectin was classified in short-lived category that differs with the results of this research due to differences in natural enemies. This research also clearly showed that flubendiamide caused less than 20% reduction of emergence rate of parasitoid and persisted less than five days (shortly-lived) at field conditions. In a similar study, the effects of flubendiamide were investigated under laboratory conditions on immature stages of *T. chilonis* within *Helicoverpa armigera* (H bner)

eggs (Hussain et al., 2015). Their results showed that flubendiamide had low effects on the emergence rate and was considered as safe for the parasitoid. In another study, Madhusudan (2015) reported that flubendiamide does not have any adverse effect on *T. chilonis* under laboratory conditions. Our results also concur with those obtained by Khan and Ruberson (2017). They studied the effects of flubendiamide on larval and pupal stages of *T. pretiosum* and those by Sattar et al. (2011). They also concluded that flubendiamide had low effects on the parasitoid *T. chilonis* in field conditions and ranked flubendiamide as a short-lived compound. We found acetamiprid is a slightly harmful (class 2) insecticide for the tested parasitoids. Some other researchers also emphasized on the harmlessness of acetamiprid. In a similar study Zhu et al. (2009) investigated the effects of several insecticides, including acetamiprid, on immature stages of *T. evanescens* under laboratory conditions. Their results showed that acetamiprid had a low toxicity to all developmental stages. Carvalho et al. (2010), had declared that acetamiprid is slightly harmful to developmental stages of *T. pretiosum*. Our result showed acetamiprid causes only a 20-30% reduction in parasitoid emergence, which is the same as their findings.

We also classified acetamiprid as a shortly-lived insecticide. In a similar study, Uma and Jacob (2013) placed flubendiamide as short-lived for *T. chilonis* at field conditions. Emamectin was slightly harmful (class 2) for both parasitoids. These results concur with those declared by Sattar et al. (2011) who reported a slight reduction of emergence rate for *T. chilonis* after immature stages were treated at the field recommended concentrations of emamectin benzoate. In a similar study, the effect of emamectin benzoate was tested on immature stages of *T. chilonis*. Hussain et al. (2015) found emamectin benzoate had a deleterious effect on the emergence rate of the parasitoid. In this research, emamectin also ranked as a moderately persistent insecticide. In other similar work, Hewa-Kapuge et al. (2003) found emamectin benzoate was moderately

persistent for *T. chilonis*. This study also revealed that both parasitoids responded at the same level of susceptibility after exposure to all the tested insecticides.

### Conclusion

This research focused on the impact of insecticide exposure on *Trichogramma* species present in Markazi Province. Pesticides that control pests without severe side effects on substantial natural enemies are always a necessity for integrated control programs. Without such insecticides, the use of short-lived insecticides could allow a rapid re-colonization of the parasitoids. We found flubendiamide and acetamiprid are non-harmful to both *T. brassicae* and *T. evanescence* wasps and good candidates to be incorporated into IPM programs in combination with the biological agents for the control of the tomato leaf miner *T. absoluta*. By contrast, emamectin and abamectin should be used with care as a part of IPM procedures.

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## ارزیابی سمیت برخی حشره‌کش‌های زیست سازگار روی دو گونه زنبور *Trichogramma brassicae* و *T. evanescens* در شرایط آزمایشگاهی و نیمه مزرعه‌ای

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**چکیده:** اثرات چهار حشره‌کش آبامکتین (۱۵۰۰ و ۷۵۰ میلی‌گرم بر لیتر)، امامکتین بنزوات (۱۰۰۰ و ۵۰۰ میلی‌گرم بر لیتر)، استامی پرید (۵۰۰ و ۲۵۰ میلی‌گرم بر لیتر) و فلوبندیامید (۵۰۰ و ۲۵۰ میلی‌گرم بر لیتر) روی مراحل مختلف رشدی دو گونه زنبور *Trichogramma brassicae* (Hymenoptera: Trichogrammatidae) و *T. evanescens* از پارازیتوئیدهای تخم شب‌پره مینوز گوجه‌فرنگی مطالعه شد. تخم‌های پارازیته شده بید غلات در مراحل لاروی، پیش‌شفیرگی و شفیرگی به‌روش غوطه‌وری با محلول‌های حشره‌کش‌ها تیمار شد. جهت ارزیابی پایداری حشره‌کش‌ها غلظت‌های توصیه شده مزرعه‌ای آنها به‌کمک یک سم‌پاش دستی روی گیاهان گوجه‌فرنگی تا جاری شدن محلول سمی پاشیده شد. گیاهان گوجه‌فرنگی زیر یک پوشش پلاستیکی به‌عنوان حفاظ باران نگهداری شدند. نمونه‌برداری از گیاهان گوجه‌فرنگی در روزهای ۳، ۵، ۱۶ و ۳۱ روز پس از تیمار انجام شد. نتایج نشان داد که استامی پرید در کم‌تر از پنج روز با ۳۰/۵ و ۳۱/۶ درصد مرگ‌ومیر به‌ترتیب برای گونه‌های *Trichogramma brassicae* و *T. evanescens* در گروه حشره‌کش‌های بی‌دوام قرار گرفت. نتیجه مشابهی برای فلوبندیامید با درصد مرگ‌ومیر ۲۷/۲ و ۲۶/۱ به‌ترتیب برای دو گونه مذکور حاصل شد. آبامکتین با ۱۶/۱ و ۱۳/۸ درصد مرگ‌ومیر به‌ترتیب برای دو گونه مذکور در کم‌تر از ۱۶ روز کم‌دوام ارزیابی شد، هر چند در تیمار مراحل نابالغ، زیان بارترین ترکیب مورد آزمایش برای هر دو گونه زنبور تریکوگراما بود. امامکتین بنزوات با ۱۳/۳ و ۱۵/۵ درصد مرگ‌ومیر در کم‌تر از ۳۰ روز برای دو گونه مذکور در گروه حشره‌کش‌های بادوام متوسط قرار گرفت. فلوبندیامید و استامیپرید برای دو گونه زنبور بی‌زیان ارزیابی شدند و می‌توان از آنها در کنار عوامل بیولوژیک جهت کنترل شب‌پره مینوز گوجه‌فرنگی استفاده نمود. در مقابل آبامکتین و امامکتین بنزوات بایستی با احتیاط بیش‌تری مورد استفاده قرار گیرند.

**واژگان کلیدی:** شب‌پره مینوز گوجه‌فرنگی، پارازیتوئیدهای تخم، حشره‌کش‌ها، پایداری، مدیریت تلفیقی آفات