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Side effects of insecticides on the adult longevity of *Cotesia vestalis*, a larval parasitoid of the diamondback moth, *Plutella xylostella*

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Abstract
The effect of several insecticides, including Chlorpyrifos, Indoxacarb, Cypermethrin and Matrin, on the longevity of *Cotesia vestalis* was investigated under laboratory conditions. Field recommended concentrations of insecticides were prepared. The experiment was conducted within tubes (1.5 cm diameter and 10 cm high) with six replications of each treatment. After 24 hours Chlorpyrifos with 100% mortality showed a significant difference in comparison with other insecticides. After 48 hours the highest and lowest mortality of *C. vestalis* occurred in Chlorpyrifos (100%) and Cypermethrin (35.8%) treatments, respectively. However, there was no significant difference between treatments 72 hours and 96 hours post-treatment. The findings of the present study thus suggest that in order to support natural enemies, the field application of dangerous insecticides such as Chlorpyrifos against diamondback moth must be limited.

Keywords: *Cotesia vestalis*, insecticide, longevity, mortality

1. Introduction
The diamondback moth (DBM), *Plutella xylostella* (L.) (Lepidoptera; Plutellidae), is the most important pest of crucifers in central parts of Iran [1]. This pest feeds on all above ground plant parts from germination till harvest and greatly reduces both quality and quantity of the produce [6, 14]. The overuse of pesticides has led to the development of resistance in target organism apart from causing harm to the natural enemies. *P. xylostella* feeds and pupates on the leaf surface and this exposure makes it vulnerable to attack by natural enemies such as predators, insect pathogens and especially parasitoids. More than 90 parasitoid species have been reported to attack *P. xylostella*, but only about 60 of them are important [12]. Larval parasitoids are the most effective in checking *P. xylostella* populations [1, 14]. *Cotesia vestalis* (Haliday) (Hymenoptera; Braconidae) is one of the most effective host specific larval parasitoids of *P. xylostella*. This parasitoid has sufficient population in the field of cruciferous plants and can provide adequate control of *P. xylostella* in most places, along with the support from other control methods. Conservation of this parasitoid is crucial for the control of *P. xylostella* [13]. *C. vestalis* is a solitary, koinobiont, larval endoparasitoid of *P. xylostella* with three larval instars before pupation. This parasitoid prefers second or early third instars of *P. xylostella* for parasitism [1, 2]. High temperature range (25 to 35 ºC) is ideal for activities of *C. vestalis* because this parasitoid is suitable for *P. xylostella* control in lowland areas [16, 17].
One of the side effects of pesticides is their impact on natural enemies. These effects in agricultural ecosystems, and in particular, for biological control are of prime importance. Different studies have shown that the effects of pesticides were prominently evaluated based on target pests. Compared with the target pest, parasitoids are more susceptible to broad-spectrum pesticides [9, 10, 15]. In this regard, the insecticides must be selected for their highest effects on the target pest and for their lowest effects on the parasitoids. The present study aimed to study the efficacy of a botanic insecticide, Matrin (Agro®), and three registered chemical insecticides, Chlorpyrifos, Indoxacarb and Cypermethrin, against *C. vestalis* in laboratory.

2. Materials and methods

2.1 Plant and insect rearing protocol

The period of investigation was from September 2013 until January 2014. Chinese cabbage (*Brassica pekinensis*) was grown organically in plastic pots (10 cm diameter) under greenhouse conditions (25±5 ºC, 70±5% RH and L:D 16:8 h) without the application of any pesticide or fertilizer. These plants were used to rear *P. xylostella*. To start a culture of *P. xylostella*, its pupa and larvae were collected from common cabbage and cauliflower fields in Isfahan province (central Iran). *C. vestalis* culture was started with the parasitized *P. xylostella* larvae collected from the same fields. Populations of *P. xylostella* were maintained on 5-week-old Chinese cabbage in ventilated cages (40×40×40 cm). Similarly, the cultures of *C. vestalis*, in turn, were maintained on *P. xylostella* larvae in ventilated oviposition cages (40×40×40 cm). Both cultures were reared at standard constant environment (25 ± 2 ºC, 70 ± 5% RH and L:D 16:8 h). Aqueous honey solutions (35%) were placed in each cage to feed the adults of *P. xylostella* and the wasp. The adult food supply (honey solution) was replaced every 48 h [8, 9, 13].

2.2 Toxicity test

To test contact effect of the mentioned insecticides, the test tubes (1.5 cm diameter and 10 cm high) were contaminated with the insecticides [Indoxacarb SC150 (250 ppm), Cypermethrin EC 10% (500 ppm), Chlorpyrifos EC 10% (1000 ppm), Matrin SL 0.6% (2000 ppm)] and control (water) for 10 seconds.

Five one to two days old adults of *C. vestalis* were released in each test tube, containing 35% honey solution for the wasp feeding, covered with a piece of netting cloth. The wasp mortality was recorded after 24, 48, 72 and 96 h. Treatments were replicated six times in a randomized complete block design, and the standard environmental conditions were maintained (25±2 ºC; 70±5% RH; L:D 16:8 h).

2.3 Statistical analysis

Percentage mortality was corrected with Abbott formula, and the data was then arcsine transformed and analyzed using one-way ANOVA. Pair comparisons were done using Tukey's honest significance test. All statistical analyses were completed in *R. 2.10.0* [3, 4].

3. Results

After 24 h, Chlorpyrifos showed 100% mortality, and indicated a significant (P_{value} < 0.001, F = 4.644, df = 3, 20) difference in comparison with other insecticides (Table 1). Percentage mortality among treatments varied from 3.3 (indoxacarb) to 100 (Chlorpyrifos). After 48 h, there was also a significant (P_{value} < 0.05, F = 4.864, df = 3, 20) difference between treatments (Table 1), where Cypermethrin showed the lowest (35.8%) percentage mortality. After 72 h, there was no significant (P_{value} = 0.355, F = 1.146, df = 3, 20) difference between the treatments (Table 1), where Indoxacarb and Chlorpyrifos showed 100% mortality on *C. vestalis*. Finally, after 96 h there was no significant (P_{value} = 0.305, F = 1.29, df = 3, 20) difference between treatments; percentage mortality among treatments varied from 79.2 (Cypermethrin) to 100 (Indoxacarb and Chlorpyrifos).

Table 1: Percentage mortality of *C. vestalis* adults subjected to different treatments

	Percentage mortality			
Treatments	**24h**	**48h**	**72h**	**96h**
Cypermethrin	13.3 ± 1.0 a*	35.8 ± 1.3 a	79.2 ± 1.6 a	79.2 ± 1.6 a
Indoxacarb	3.3 ± 0.3 a	53.3 ± 1.9 ab	100 ± 0.0 a	100 ± 0.0 a
Matrin	10.0 ± 0.7 a	50.0 ± 1.3 ab	87.5 ± 1.2 a	91.7 ± 0.8 a
Chlorpyrifos	100 ± 0.0 b	100 ± 0.0 b	100 ± 0.0 a	100 ± 0.0 a

* Means with same letter in each column are not significantly (P > 0.05) different

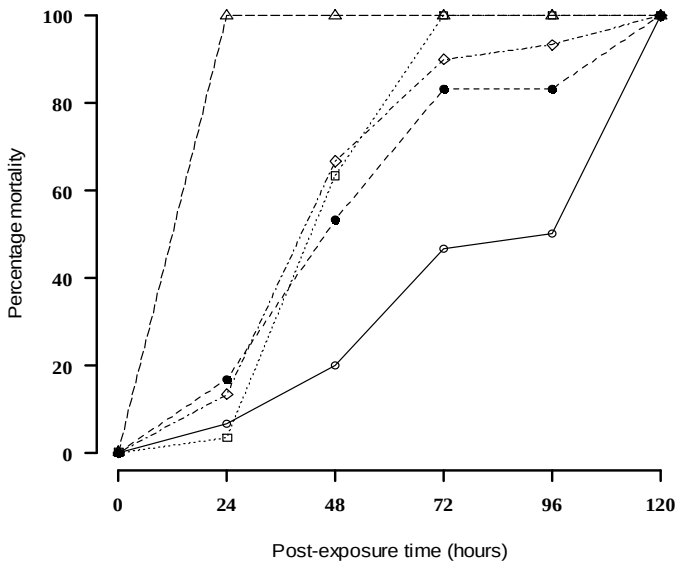

Fig 1: Percentage mortality of treatment in different post-exposure time (Δ Chlorpyrifos, □ Indoxacarb, ● Cypermethrin, ◊ Matrin and ○ Control)

4. Discussion

The results showed that, Chlorpyrifos contributed to 100% mortality of the parasitoid wasp and it can be considered as a harmful insecticide. Different studies also have reported such a harmful effects for many natural enemies [5, 11, 15].

Indoxacarb caused low percentage mortality on *C. vestalis* at 24 h and 48 h, but this insecticide led to 100% mortality earlier than Matrin and Cypermethrin over 72 h. Over the period of 96 h, all of the insecticides showed the highest mortality for *C. vestalis* (Figure 1). Karimzadeh *et al.* (2014) investigated the efficacy of toxicity of four insecticides (Chlorpyrifos, Indoxacarb, Cypermethrin and Matrin) on the diamondback moth in field. They have been put Matrin with having high percentage mortality and Chlorpyrifos with having lower percentage mortality in the first and last group, respectively [7]. According to the results of the present study, considering the toxicity of Matrin on the adults of *C. vestalis*, it should be taken into account that time of release of the parasitoids must not be in accordance with the application of this insecticide to support the populations of *C. vestalis*.

In general, the time of application of insecticides and the release of biocontrol agents in the crucifers fields should be regulated so as to avoid contact of these insecticides with *C. vestalis.*

5. Conclusion

In order to conserve and support the population of natural enemies of *P. xylostella*, especially larval parasitoids having the capacity of control diamondback moths in the field, it is necessary to limit the application of any kind of harmful insecticides like Chlorpyrifos.

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