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Bio efficacy of microbial, botanical and chemical pesticides on adult emergence of *Trichogramma chilonis* and contact toxicity on adult of larval parasitoid *Bracon hebetor*

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Abstract
Indiscriminate use of non-selective pesticides in agriculture has several detrimental effects, of which the harm caused to natural enemies. The present experiment was carried out to find out the toxicity of some ecofriendly pesticides on *Trichogramma chilonis* and contact toxicity of *Bracon hebetor* under laboratory conditions. Contact toxicity of *Bracon* adult, after four days of exposure, spinosad 45% SC @ 0.01% caused lowest mortality (12.50%). The adult emergence *T. chilonis* ranged from 29.50 to 91.50 per cent with highest emergence in NSKE @ 5%, *i.e* 90.50%. Pesticide treated Corcyra eggs were parasitized by *T. chilonis* in the tune of 44.50 to 92.25% and maximum parasitisation 92.25% was observed in NSKE at 5%. From the findings it can be concluded that *T. chilonis* and *B. hebetor* may be successfully utilized for IPM programme in filed condition along with microbial pesticides such as *B.t.* and spinosad 45% SC and botanicals NSKE.

Keywords: contact toxicity, natural enemies, *Bracon hebetor*, *Trichogramma chilonis*

Introduction

The insect pest problem in Indian agriculture is becoming more and more threatening and one of the factors affecting crop production by lowering the yield and impairing the quality of the produce by several ways. To manage these insect pests, farmer used different kinds of chemical insecticides. Intensive and successive applications of broad spectrum chemical insecticides for insect pest management may cause multitudes problems, such as insecticides resistance to different insect, environmental pollution and its results, increase in the costs of insect pest management and mainly, the death of natural enemies. Use of non-selective insecticides which are incompatible with natural enemies has caused serious problems for crops such as target pest resurgence, secondary pest outbreaks. In insect pest management naturally occurring natural enemies make important contributions to pest control. The use of natural enemy agents in combination with selected insecticides, which have no effect on them, is effective in depressing the population density of the pest. Therefore, it is immensely necessary to preserve natural enemies, so that they may present a good performance in pest biological control, which is a vital management practice used in the programs of integrated pest management (IPM), but producers need to have a clear idea about which insecticides allow natural enemies to persist to allow for informed product selection decisions. The egg parasitoid *Trichogramma chilonis* is found promising as a natural and augmentation control agent attacks more than 400 pest species, mostly Lepidopteran and Hetroptera insects. It has been successfully used in inundative and inoculative biological control programmes worldwide Pinto and Stouthamer (1994) [10]; Krishnamoorthy (2012) [7]. Among the ecto-parasitoid *Bracon hebetor* (Braconidae: Hymenoptera) is a highly gregarious polyphagous natural enemy, attacks a variety of important Lepidopterous species of field crops as well as stored products Landge *et al*. (2009) [8]. Unfavourable effects on natural enemies due to the harmful effects of insecticides on agriculture, it is momentous to find out which insecticides have the potential to disrupt natural enemies; therefore, the aim of this experiment was to find out the relative risk to natural enemies from different insecticides. This information can be combined with pest control efficacy data for making recommendations and allowing informed decisions regarding the use of insecticides. Our objectives were to find out the 1. Contact toxicity of pesticides on adult of larval parasitoid *B. hebetor*. 2. Adult emergence of *T. chilonis* by dipping the tricho-card in pesticide solution. 3. Effect of pesticides on *T. chilonis* by dipping UV sterilized egg cards in pesticides solution before parasitisation. Keeping these views in mind, the present experiment was conducted to evaluate the Bio efficacy of microbial, Botanical and chemical pesticides on adult emergence of *T. chilonis* and contact toxicity on adult of larval parasitoid *B. hebetor* under laboratory conditions.

Materials and Methods

The egg parsitoid *T. Chilonis* and ecto-parasitoid *B. hebetor* used in this study were obtained from the bio control laboratory, BCKV, Kalyani, W.B. The experiments were conducted on completely randomized block design and each treatment was replicated four times. The treatments comprised of microbials pesticides, *B. thuringiensis* 5% WP @ 0.2% *a.i.*, abamectin 400FS @ 0.04% *a.i.*, spinosad 45% SC @ 0.01% *a.i.*, botanicals as NSKE @ .0 % *a.i.*, synthetic pesticides like diflubenzuron + deltamethrin 22% SC @ 0.022% *a.i.*

i. Effect of eco friendly pesticides on *Bracon hebetor*

Safety of microbial, botanicals and chemical pesticides was conducted by contact toxicity test, when adult *Bracon* were exposed to pesticides. For contact toxicity study, 0.5 ml of pesticide solution or suspension was pipetted out into the glass tube of 15 x 2.5 cm size, which was later constantly rotated and rolled on a flat surface so as to form a thin and uniform coating inside wall of test tube. Each tube served as a replicate with four such replications per treatment. Freshly emerged 10 adults of *B. hebetor* were transferred into each treated tube. The tubes were then covered with a cotton cloth. Honey as food was provided with the help of cotton swab at the top of the cloth. Mortality of the parasitoids was recorded 24, 48, 72 and 96 hours after release.

ii. Effect of ecofriendly pesticides on *Trichogramma chilonis* Ishii

To study the effect of different microbial pesticides, botanicals and chemicals on *T. chilonis*, two different methods were followed.

In first method, the parasitized egg cards were cut into small pieces having different number of parasitized eggs. Numbers of parasitized eggs in each small piece was recorded separately. The three days old, hundred percent parasitized eggs (eggs appearing black and plumpy) were dipped in pesticide solution at different concentration as required for different treatment. For untreated check, cards were dipped in only distilled water. The treated egg cards were shade dried for 10 minutes and then kept inside a test tube 20 x 1.5 cm size. The number of parasitoids emerged from each treatment were recorded and per cent emergence was worked out.

In another set of experiment, UV light sterilized fresh corcyra egg cards (without parasitisation) of equal size having 100 eggs for each treatment with three replications were dipped in pesticide solution for few seconds. The treated fresh cards were allowed to dry in shade for half an hour and for parasitization each card was exposed to 3 days old parasitized tricho-card of equal size having same quantity of parasitized eggs in a glass vial of 15 x 1.5 cm. Parasitization was recorded by observing blackend eggs. Then the newly formed tricho-cards were left for adult emergence and the emerged adults were counted for per cent emergence. In untreated check fresh corcyra egg card was dipped in distilled water only before it was kept with tricho-card for parasitization. Both the experiments were conducted in a complete randomized block design and the data collected on adult emergence only from experiment one (*i.e.* where parasitized tricho-cards were dipped in pesticide solution) and on per cent parasitization & adult emergence from experiment two (*i.e.* where UV sterilized fresh corcyra cards without parasitization were dipped in pesticide solution before exposed to tricho-card) were subjected to statistical analyses after angular transformation and the means were separated by DMRT Gomez and Gomez (1984) [4]. The data were then categorized to know the level of safety of the products tested following the method of -

1. Reduction of Parasitism viability *i.e* Adult emergence (Manzoni *et al.*, 2007) [9]

$E (\%) = (1 - V_t/V_c) \times 100$

Where,

E = Adult emergence

V_t = parasitism on treatment.

V_c = Parasitism on control.

2. Reduction in Parasitism (Hassan *et al*., 2000) [5]

$RP (\%) = (1 - f/t) \times 100$

Where,

RP = Reduction in parasitism

f = No of parasitized eggs in treatment.

t = No of parasitized eggs in control.

3. IOBC class (Sterk *et al.*, 1999) [12]:

Category		E and RP (%)
Harm less (H-)	=	(<30%)
Slightly harmful (SH)	=	(30-79%)
Moderately harmful MH	=	(80-99%)
Harmful (H+)	=	(>99%)

Results and Discussions

1. Ecofriendly pesticides and their Contact toxicity on *Bracon hebetor*

The results achieved on the bio-efficacy of different test chemicals against adult of larval parasitoid *Bracon hebetor* have been presented in table 1.

Table 1: Contact toxicity of ecofriendly pesticides on *Bracon hebetor*

Treatments	Dose (% *a.i.*)	Adult mortality (%) of *Bracon hebetor*			
		1 DAT	2 DAT	3 DAT	4 DAT
Bacillus thuringiensis	0.2	2.75 (8.49)^{bc}	8.75 (16.77)^{bc}	12.50 (20.47)^{b}	16.25 (23.73)^{b}
Neem Seed Kernel Extract (NSKE)	5.0	5.13 (12.08)^{b}	6.25 (14.30)^{c}	11.25 (19.52)^{b}	16.25 (23.73)^{b}
Abamectin 400FS	0.04	5.13 (12.08)^{b}	11.25 (19.52)^{b}	13.75 (21.70)^{b}	16.25 (23.59)^{b}
Diflubenzuron + deltamethrin 22% SC	0.022	17.50 (24.68)^{a}	25.00 (29.94)^{a}	30.00 (33.17)^{a}	35.00 (36.25)^{a}
Spinosad 45% SC	0.01	2.75 (8.49)^{bc}	7.50 (15.68)^{bc}	10.00 (18.14)^{b}	12.50 (20.61)^{b}
Untreated control	0.00	0.00 (4.05)^{c}	0.00 (4.05)^{d}	0.00 (4.05)^{c}	0.00 (4.05)^{c}
SEm.±		2.31	1.48	1.77	1.20
CD at 0.05%		6.86	4.40	5.26	3.56

* DAT = Days after treatment.
* Figures within parentheses are angular transformed values.
* In a column, means followed by same alphabet are not significantly different (p=0.05) by DMRT.

Newly born *Bracon* adult were exposed to pesticides for contact upto 4 days. After four days of exposure, diflubenzuron + deltamethrin - 22%SC @ 0.022% caused relatively more mortality (35.00%), followed by 16.25% in *B.thuringiensis* @ 0.2%, abamectin 400FS @ 0.04% and NSKE @ 5%. Mortality of *Bracon* caused by *B.thuringiensis* varied from 2.75% to 16.25%, NSKE from 5.13% to 16.25%, Abamectin 400FS @ 0.04% from 5.13% to 16.25% where as in case of diflubenzuron + deltamethrin - 22%SC @ 0.022% it was 17.50% to 35.00% and spinosad 45%SC @ 0.01% from 2.75% to 12.50% at different days after treatment.

Above mentioned all the microbials, botanicals and IGRs were relatively safer pesticide to adults of larval parasitoids. Raghuraman and Singh (1998) [11] reported that the parasitoid larvae of *Bracon hebetor* were killed by feeding on contaminated host larvae and also through contact with neem extract at 2.5 and 5% concentration. Similar study was carried out by Ahmed and Ahmad (2006) [1]. They reported that some environmentally safe insecticide, like, lambda-cyhalothrin and spinosad for their toxicity against *B. hebetor*. Khan *et al*. (2009)[6]Observed that the higher dose rates (10% above the recommended dose rate) of abamectin, leufenuron and spinosad proved to be slightly harmful as the percentage mortality in the adults of *B. hebetor*, treated with these dose rates ranged between 50-79%, after 24 hours of interval. Same insecticides were harmless, after 24 hours of their application, at lower dose rates (10% below the recommended dose rate). However, minimum mortality was found in spinosad (47.500 ± 0.833) @ 110 ml/acre. The insecticide treatments of abamectin and spinosad were ranked slightly harmful after 48 hours of their application. According to them, spinosad was least toxic to the adults of *B.hebetor* and these results are in consistency with our results, narrating minimum mortality with spinosad.

2. Bio efficacy of microbial, botanical and chemical pesticides on adult emergence of *T. chilonis* by dipping the tricho-card in pesticide solution

Three days old parasitized tricho-card was dipped in microbial, botanicals and chemical pesticide solution to evaluate the adult emergence of *T. chilonis* and is data presented in table- 2.

Table 2: Bio efficacy of microbial, botanical and chemical pesticides on adult emergence of *T. chilonis* by dipping the tricho-card in pesticides solution

Treatments	Dose (% *a.i.*)	Parasitism	E (%)	Category
Bacillus thuringiensis	0.2	79.50 (63.09)^d	17.19	H-
Neem Seed Kernel Extract (NSKE)	5.0	91.50 (73.08)^b	4.69	H-
Abamectin 400 FS	0.04	44.50 (41.84)^e	53.65	SH
Diflubenzuron + deltamethrin 22% SC	0.022	29.50 (32.89)^f	69.27	SH
Spinosad 45% SC	0.01	83.50 (66.05)^c	13.02	H-
Untreated control	0.00	96.00 (78.70)^a		
SEm±		0.71		
CD at 0.05%		2.13		

* Figures within parentheses are angular transformed values.
* In a column, means followed by same alphabet are not significantly different (p=0.05) by DMRT.
* H- = Harmless, H+ = Harmful, MH = Moderately Harmful, SH = Slightly Harmful.

The adult emergence ranged from 29.50 to 91.50 per cent with highest emergence in NSKE @ 5%, *i.e* 90.50% followed by 83.50% in spinosad 45% SC @ 0.01%; 79.50% in *B. t.* @ 0.2%, 44.50% in abamectin 400FS @ 0.04%, and 29.50% in diflubenzuron + deltamethrin 22% SC @ 0.022% compared to 96.00% in untreated control. From the above mentioned results it is clear that NSKE, spinosad 45% SC and *B.t.* were safe to *T. chilonis*. Among the treatments significant difference were noticed with regards to adult emergence of *T.chilonis*. The lowest per cent (29.50%) adult emergence was noticed in diflubenzuron + deltamethrin 22% SC @ 0.022% followed by 44.5% in abamectin 400FS @ 0.04% which was significantly lower than other treatments. On the basis of the above study under laboratory condition it can be concluded that *T.chilonis* may be successfully utilized for insect pest management in filed condition along with microbial pesticides such as *B.t.* and spinosad 45% SC and botanicals NSKE.

3. Effect of ecofriendly pesticides on *Trichogramma chilonis* by dipping UV sterilized egg cards in pesticide solution before parasitisation

The results achieved on the bio efficacy of various ecofriendly pesticides like microbial, botanical and chemical pesticides under laboratory condition (temp. 26.53 -32.75^0 C and RH. 78.20 - 93.86%) against *T. chilonis* have been presented in table. 3.

Table 3: Effect of ecofriendly pesticides on *Trichogramma chilonis* by dipping UV sterilized egg cards in pesticide solution before parasitisation

Treatments	Dose (% *a.i.*)	parasitized eggs	(RP)%	Category	Parasitism	E (%)	Category
Bacillus thuringiensis	0.2	67.00 (54.94)^c	31.63	SH	79.50 (63.11)^d	17.19	H-
Neem Seed Kernel Extract (NSKE)	5.0	92.25 (74.12)^b	05.87	H-	91.50 (73.08)^b	04.69	H-
Abamectin 400 FS	0.04	52.00 (46.15)^d	46.94	SH	45.00 (42.13)^e	53.13	SH
Diflubenzuron +deltamethrin-22% SC	0.022	46.00 (42.70)^e	53.06	SH	30.00 (33.21)^f	68.75	SH
Spinosad 45% SC	0.01	44.50 (41.84)^e	54.59	SH	83.50 (66.05)^c	13.02	H-
Untreated control	0.00	98.00 (82.01)^a			96.00 (78.70)^a		
SEm±		0.93			0.79		
CD at 0.05%		2.77			2.35		

* Figures within parentheses are angular transformed values.
* In a column, means followed by same alphabet are not significantly different (p=0.05) by DMRT.
* H- = Harmless, H+ = Harmful, MH = Moderately Harmful, SH = Slightly Harmful.

UV sterilized Corcyra eggs treated with pesticide solution were parasitized by *T. chilonis* in the tune of 44.50 to 92.25% as compared to 98.00% in untreated check Maximum parasitisation 92.25% was observed in NSKE at 5% followed by 67.00% in *B. thuringiensis* at 0.2%. The lowest egg parasitisation 44.50% was recorded in case of spinosad 45% SC @ 0.01% followed by 46.00% in diflubenzuron + deltamethrin 22% SC @ 0.022%, but they did not differ significantly from each other but differed from the remaining treatments.

Adult emergence percentage of *T. chilonis* varied from 30.00 to 91.50% as compared to 96.00% in untreated checked. Lowest and highest adult emergence 30.00% and was 91.50% noticed in diflubenzuron + deltamethrin 22% SC @ 0.022% and NSKE @ 5% respectively. All the treatments significantly differed from each other with respect to adult emergence.

The results of the present studies are in conformity with the earlier findings of Dhal and Seth (2017) [2] reported that UV sterilization of host eggs markedly enhanced parasitisation capacity of *T. Chilonis* with respect to unsterilization host eggs. Dileep (2012) [3] who have observed that the highest per cent adult emergence was recorded in control (94.00%) and among the different insecticides tested azadirachtin recorded the maximum per cent adult emergence (83.67%).

Conclusion

From the findings it can be concluded that *B.thuringiesis*, NSKE and spinosad were harmless (H-) but abamectin 400FS and diflubenzuro+deltamethrin 22% SC were slightly harmful (SH) for natural enemies. The NEs. *T.chilonis* and *B. hebetor* may be successfully utilized for IPM programme in filed condition along with microbial pesticides such as *B.t.* and spinosad 45% SC and botanicals NSKE.

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