Journal of Entomology and Zoology Studies 2020; 8(5): 1037-1040

Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
www.entomoljournal.com
JEZS 2020; 8(5): 1037-1040

Received: 07-07-2020
Accepted: 09-08-2020

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Evaluation of selected insecticides on adults of Lepidopteron biocontrol egg parasitoid *Trichogramma chilonis* (Trichogrammatidae, Hymenoptera)

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Abstract

Egg parasitoids are known to be very effective against a number of crop pests. Parasitoids belonging to the genus *Trichogramma,* have worldwide distribution and play an important role as natural enemies of the lepidopteran pests on a wide range of agricultural crops. But these egg parasitoids are highly susceptible to insecticides. In order to study the insecticide tolerance, the effect of the insecticides avermectin (abamectin) and a few organophosphates (chlorpyriphos, malathion, quinolphos, triazophos), oxadiazine (indoxacarb), and spinosyn (spinosad) as well as with pyrethroids (cypermethrin) on laboratory and field adult populations of the egg parasitoid *Trichogramma chilonis* (Hyn: Trichogrammatidae) was evaluated under laboratory conditions, using the standard tests (residue test on glass tubes) described by IOBC. When tested on the adult populations of the parasitoids chlorpyriphos, malathion, quinolphos, triazophos proved to be most lethal insecticides on the survival of the adult parasitoid. Similarly, abamectin and cypermethrin were also found to be harmful on the survival of the adult parasitoid. Indoxacarb and spinosad were found to be least harmful on the adult survival of the egg parasitoid.

Keywords: Biological control, egg parasitoid, hymenoptera, insecticides, natural enemies, Trichogrammatidae

Introduction

Egg parasitoids are known to be very effective against a number of crop pests. Parasitoids belonging to the genus *Trichogramma* have worldwide distribution and play an important role as natural enemies of the lepidopteron pests on a wide range of agricultural crops [10]. During the last decades, *Trichogramma* wasps have been used as biological control agents for pest suppression in several countries [1]. The use of reared *Trichogramma sp*. in the biological control has gained widespread interest in many countries [11]. Due to the importance of the *Trichogramma sp*. as natural enemies of several pests, the study of insecticide selectivity to them is of major importance for their role as biological control agents. The species *Trichogramma chilonis* is an important natural enemy and one of the important biological control agent used to control Lepidopteron pests in several countries [5]. As the use of insecticides is the common practice among tomato producers, the use of selective insecticides is an important strategy for the maintenance of the Trichogramma populations within the crops. However, there are few published studies about the effect of insecticides used to control the tomato pests on this parasitoid. Therefore, the effect of insecticides used on this crop, on the adult survival of laboratory and field populations of *T. chilonis,* using the standard tests recommended by IOBC (International Organization for Biological Control) were evaluated.

Materials and Methods

Field collection of *T. chilonis*

About 100 eggs of lepidopteron pest *Helicoverpa armigera* were collected from tomato fields of Malur, Karnataka. Four to six collections were made during crop season. Eggs were brought to the laboratory and observed for parasitism and subsequently adult emergence. Identity of emerged *Trichogramma* was determined by Division of Biosystematics, National Bureau of Agriculturally Important Insect Resources, (NBAII), ICAR, Bangalore. During the survey insecticide usage pattern was also recorded.

Maintenance of laboratoryand field *T. Chilonis*

Stock populations of laboratory (obtained from Mass Production Unit of NBAII, Bangalore) and field collected *T. chilonis* were maintained on the eggs of the *Corcyra cephalonica* (Stainton) under the controlled conditions at 27 ± 1ºC, RH of 70 ± 10% with 14 h of photo phase. The host eggs about 0.2 cc (0.2 = 2000 eggs) were glued on a white paper cards (7 X 2 cm) impregnated with 50% diluted gum with water and UV killed before being offered to parasitism @ 50 eggs / female parasitoid. Approximately after 5 days of the parasitism period, parasitized eggs (indicated by black shiny appearance of the egg chorion) were transferred to glass tubes (8.5 cm height X 2.5 cm dia.) and kept in the incubator until adult emergence, at same controlled conditions previously described. Fine streaks of honey and water (1:1) was provided as food for adults (Protocols followed as per the procedures of NBAIIR, ICAR, Bangalore).

Insecticides selected for the study

In the present study, following eight commonly used insecticides were selected (Table- 1) in order to document insecticide resistance in laboratory and field populations of *T. chilonis*.

Preparation of insecticidal concentrations

Different dilutions of the selected insecticides such as abamectin (1.9% EC), chlorpyriphos (20% EC), cypermethrin (25% EC), indoxacarb (14% EC), malathion (50% EC), quinalphos (25% EC), spinosad (2.5% EC) and triazophos (40% EC) were prepared out of the respective formulated insecticides. The top dose was at respective field recommended concentration was used for preparing stock solution in 1 litre of water (Protocols followed as per the procedures of NBAIIR, ICAR, Bangalore).

Insecticide bioassay studies on laboratory and field populations of *T. chilonis*

The commercial insecticides were diluted in distilled water at concentrations recommended for selected insecticides. Seven serial dilutions were prepared starting from field recommended dose for laboratory population and the field populations (Table 1). Diluted insecticides according to its respective concentrations were sprayed with an atomizer to obtain a uniform layer of spray droplets on the inside of the glass tubes (20 x 4 cm) with both ends open. After spraying, the tubes were shade dried. One end of the sprayed and dried tube, was closed with double layered long black cloth of (32 µm pore size). About 100 *Trichogramma* adults were allowed to move into sprayed tubes (labeled with different concentrations of insecticides) through the open end and after 15 minutes, the other end of the tube was also closed withdouble layered long black muslin cloth. Adult morality/survival after1 h, 2 h, 4 h, 6 h and 24 h release was recorded. For calculating LC_{50}, mortality at 4 h was considered as differential response was noticed for different insecticides at 1 h and 2 h mortality pattern. Data from the replicates were pooled and dose-mortality regressions were computed by probit analysis using SPSS 18.0 (SAS Institute Inc, Cary, NC) to obtain LC_{50} and fiducial limits for each insecticide. Non- overlapping method of the fiducial limits was used to differentiate variation in insecticide resistance. Resistance factor (RF) was calculated based on LC_{50} of the resistant strain/ LC_{50} of the laboratory strain (Protocols followed as per the procedures of NBAIIR, ICAR, Bangalore).

Table 1: Details of different insecticides used for the calculation of the LC_{50} for *T. chilonis*

Sl. No	Trade Name	Formulation	Chemical Name	Group	Conc.		Range	
					S	F	Susceptible	Field
01.	Tagmec	1.9% EC	Abamectin	Avermectin	07	03	0.3-18.96	0.3-2.37
02.	Lethal TC	20% EC	Chlorpyriphos	Organophosphate	07	03	6.25-400	6.25-25
03.	Mokard	25% EC	Cypermethrin	Pyrethroid	07	03	0.625-40	0.625-2.5
04.	King Doxa	14% EC	Indoxacarb	Oxadiazine	07	03	4.37-280	70-280
05.	Malamal	50% EC	Malathion	Organophosphate	07	03	0.77-49.6	0.77-3.1
06.	Quinalphos	25% EC	Quinalphos	Organophosphate	07	03	7.81-500	125-500
07.	Success	2.5% EC	Spinosad	Spinosyn	07	03	0.152- 10	1.25-5
08.	Hostathionn	40% EC	Triazophos	Organophosphate	07	03	0.67-39.68	0.67-2.48

*Range: Calculated from ml/l (Percentage concentration X 1000/Emulsified concentration) as ppm of respective insecticides

Results and Discussions

Insecticide bioassay on laboratory and field (Tomato) populations of *T. chilonis*

The statistical comparison of toxicity and resistant factor of laboratory and field populations of *T. chilonis* is shown in Table 2. The data revealed very minute variations in the responses of laboratory and field populations of *T. chilonis* to the insecticides applied. In field populations, the median lethal concentration (LC_{50}) ranged from 1.03 to 271.13 ppm for the eight different insecticides, with the adults exhibiting the highest LC_{50} to quinalphos and the lowest to malathion (Table 2). In laboratory populations median lethal concentration (LC_{50}) ranged from 1.05 to 322.47 ppm for the eight different insecticides, with highest LC_{50} values obtained for quinalphos and the lowest for malathion (Table2).

The LC_{50} of field populations to organophosphate compounds was found to be highest for quinalphos (271.23) followed by chlorpyriphos (12.93), triazophos (1.33) and least in malathion (1.03) and in laboratory strain the LC_{50} organophosphate compounds was found to be more in quinalphos (322.47) followed by chlorpyriphos (11.34), triazophos (1.29) and least was in malathion (1.05). The LC_{50} of indoxacarb was 176.05 and 288.09respectively in field and laboratory strain populations; similarly, LC_{50} of abamectin was 1.62 and1.72 respectively in field and laboratory strain. The LC_{50} of parathyroid (cypermethrin) was 1.69 and 1.30 respectively infield and laboratory strain. The LC_{50} of spinosad was 2.75 and 2.86 respectively in field and laboratory strains. Comparing the LC_{50} values of all eight insecticides of field and laboratory populations, it shows that field populations of *T. chilonis* have no resistance to the insecticides tested.

This study with avermectin (abamectin) and some organophosphates (chlorpyriphos, malathion, quinalphos, triazophos) and oxadiazine (indoxacarb), spinosyn (spinosad) as well as with pyrethroids (cypermethrin), clearly demonstrated that there was no such observable resistance found in field populations of *T. chilonis* when compared to that of laboratory populations. From the current results, it can be seen that field and laboratory populations of adult *T. chilonis* are highly susceptible to the insecticides tested and this is due to compounds belonging to chemical groups of the lactones, carbamates, neonicotinoids and organophosphates and other chemicals are related in the scientific literature as highly harmful to parasitoids [6, 25, 9, 22, 21, 1, 4, 14, 15, 25] High mortality was observed for the organophosphate insecticides even at the lowest concentrations. Among organophosphates malathion was found to be highly toxic to the adults followed by triazophos, chlorpyriphos and quinalphos. The high mortality of adults observed in the emergence tubes caused by these insecticides is not clear until this moment. A similar effect was related by Abdelgader and Hassan, 2002 [1]. Similarly, cypermethrin was found to be highly toxic to both field and lab adult population of *T. chilonis*. Findings of Console *et al*. 1998 [5] and Hassan, 1998b [8] also suggested that organophosphates and pyrethroids adversely affected survivability of the adult populations of *T. chilonis*.

Table 2: Comparison of toxicity of eight commonly used insecticides on field (tomato) and susceptible populations of *T. chilonis* after 24 h of exposure

Test Population of adult *T. chilonis*	Insecticide	LC_{50} (ppm)	95% FL of LC_{50}		RF	Slope ± SE	Chi square
			Lower	Upper			
Field (Tomato)	Abamectin	1.62	-	-	0.92	0.0708 ± 0.1481	9.90
	Chlorpyriphos	12.93	-	-	1.14	0.0914 ± 0.0164	8.77
	Cypermethrin	1.69	-	-	1.31	0.6050 ± 0.1428	11.11
	Indoxacarb	176.05	-	-	0.61	0.0050 ± 0.0012	13.50
	Malathion	1.03	-	-	0.91	0.9386 ± 0.1567	20.55
	Quinalphos	271.23	-	-	0.94	0.5020 ± 0.0006	22.08
	Spinosad	2.75	-	-	0.96	0.4564 ± 0.0758	7.98
	Triazophos	1.33	-	-	1.03	0.7521 ± 0.1584	14.13
Susceptible strain	Abamectin	1.72	-	-	-	0.6454 ± 0.1442	10.92
	Chlorpyriphos	11.34	-	-	-	0.0845 ± 0.0158	12.52
	Cypermethrin	1.30	0.80	1.95	-	0.3719 ± 0.1414	3.90
	Indoxacarb	288.09	-	-	-	0.0049 ± 0.0013	8.47
	Malathion	1.05	-	-	-	0.7309 ± 0.1345	26.84
	Quinalphos	322.47	-	-	-	0.0017 ± 0.0005	25.41
	Spinosad	2.86	-	-	-	0.4522 ± 0.0825	4.44
	Triazophos	1.29	-	-	-	0.7245 ± 0.1604	13.14

LC_{50}: Concentration of insecticide that killed 50% of the adult population in the observation period of 4 h. FL: Fiducial Limit. SE: Pooled binomial standard error

Though abamectin had moderate effect on survivability of adult *T. chilonis*, complete mortality was observed after 24 h of exposure and this is in accordance with the studies conducted by Moura *et al*. 2006 [16] for the effect of chlorpyriphos, abamectin on *T. precious*. Findings of Smith *et al*. 1995 [19], Consoli *et al*. 1998 [5] and Hussain *et al*. 2010 [13] also showed that abamectin had detrimental effect on adult survival of *T. chilonis*.

At the field recommended doses spinosad had moderate effect on the survivability of adult *T. chilonis* but at higher concentration 100 per cent mortality was seen after 24 h of exposure. These results are in agreement with Williams *et al*. 2003 [24] who reported that spinosad residues degrade quickly in the field, with little residual toxicity to 25 species of hymenopteran parasitoids at 3–7 days after application. While on the other hand indoxacarb had very less effect on the survivability of adult *T. chilonis* even at higher insecticide dose. Indoxacarb has been tested having no or little effects on hymenopteran parasitoids (*Aphidius, Cotesia, Bracon, Mocroplitis, and Trichogramma*), spiders and predacious mites, and has no or moderate effects on lacewings and coccinellids [20, 3, 23, 7, 2]. Hewa-Kapuge *et al*. 2003 [12] bioassayed seven insecticides for effects on *T. brassicae*, and found that indoxacarb was not toxic to *T. brassicae* in any assay when applied at field rates, and they consider indoxacarb as potentially suitable for inclusion in integrated pest management strategies. Because, it does not influence adult survival or development of immature stages. When using indoxacarb against *H. armigera* on populations of *T.pretiosum* on sweet corn in Queensland, Australia, Scholz and Zalucki [18] found that indoxacarb appeared to be very safe for *T. pretiosum*, with only minor mortality due to residues on leaves. Therefore, indoxacarb should be applied with caution if the sensitive natural enemies are dominant at the time of application. In contrast, Williams *et al*. 2003 [24] reviewed the effects of spinosad on seven species of *Trichogramma*, and summarized that hymenopteran parasitoids are more laboratory to spinosad than predatory insects in laboratory and field studies. Nevertheless, spinosad has consistently been reported to be more harmful to parasitoids than indoxacarb [17].

Conclusions

Earlier studies have reported that insect group Trichogrammatidae is highly susceptible to most of the insecticide groups and other toxic chemicals. However, there are few published studies about the effect of insecticides used to control tomato pests and its parasitoids. The present results revealed, high susceptibility of wasp to insecticides particularly adults, but demonstrated that some insecticides may be more compatible or suitable for conserving natural or released populations of *Trichogramma* wasps also indicated that these insecticides could be used in conjunction with *T. chilonis* to control lepidopteran pests.

Acknowledgements

The authors thank, The Director and Principal Scientists of Molecular Entomology Laboratory, National Bureau of

Agriculturally Important Insect Resources (NBAIIR), Indian Council of Agricultural Research, Bangalore for providing necessary facilities and guidance and laboratory facilities to carry out insecticides bioassay studies.

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