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Relative toxicity of various insecticides against biological attributes of egg parasitoid, *Trichogramma japonicum* Ashmead

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Abstract
Trichogramma japonicum Ashmead is one of most important egg parasitoid found in rice and cotton ecosystem. But indiscriminate use of pesticide not only reduces its population but also limits parasitic potential of such potent parasitoid. Thereby, disrupt the ecological balance. Therefore, this study was conducted to check the relative toxicity of seven various insecticides viz., Flubendiamide 39.35 SC; Chlorantraniliprole 18.5 SC; fipronil 5 SC; Thiacloprid 21.7 SC; quinalphos 25 EC; lambda-cyhalothrin 5 EC and Thiamethoxam 25 WG against *T. japonicum*, and studied the egg parasitization and adult emergence of *T. japonicum*. The results of the present investigation revealed that quinalphos 25 EC exerted harmful effect on egg parasitization of *T. japonicum* as well as on adult emergence. Further, lambda-cyhalothrin 5 EC and fipronil 5 SC were found moderately harmful and slightly harmful to the egg parasitization, respectively while both were considered as harmless for adult emergence of *T. japonicum*. The remaining insecticides were found comparatively safer to both egg parasitization and adult emergence of *T. japonicum*. Basing upon the relative toxicity effect, Flubendiamide 39.35 SC; Chlorantraniliprole 18.5 SC; Thiacloprid 21.7 SC and Thiamethoxam 25 WG can be used in IPM programme of rice as a consolidated manner with *T. japonicum* as they were found comparatively safer than rest of the tested insecticide during the present investigation.

Keywords: *Trichogramma japonicum*, insecticides, relative toxicity, adult emergence, parasitization, etc

Introduction

The diversity and abundance of the *Trichogramma sp.* are well known for its potent effect on the biocontrol programme in many agro-ecosystems (Smith 1996; Hussain *et al*. 2010) [25, 13]. The easy mass production and maximum geographical distribution of *Trichogramma* spp. shows its raise in use in pest management programme (Preetha *et al*. 2009) [22]. The potent species of *Trichogramma* are mass multiplied and released annually in an estimated 80 million acres of agricultural crops as well as forests in 30 countries (Li 1994; Olkowski and Zhang 1990) [17, 20]. Hymenoptera is one of the important entomophagous order and widely used for biocontrol programme throughout the world. Among the trichogrammatids, the genus *Trichogramma* contains 130 species, out of which 20 species have been recorded from India *Trichogramma* spp. parasitize the eggs of over 400 species belonging to seven insect orders (Sattar *et al*. 2011) [23]. In spite the key role of the bio-control agents in agro-ecosystems, chemical control is required and is still being regarded as a vital component of Integrated Pest Management system and is used in consolidated manner with the biological control. On the other hand, the indiscriminate use of nonselective and broad-spectrum insecticides greatly limits the parasitic efficacy of these potent biocontrol agents and have detrimental effects on biological agents, particularly on Hymenoptera which is more vulnerable to insecticides than its host insects. In recent years, various countries have developed strategies to bring out the use of plant-protection inputs closer to a more sustainable agriculture in accordance with the basic requirements of the integrated pest management (IPM) programmes (European Commission, 2009) [9] aimed at integrating the chemical and biological control of target pests. However, the control of these notorious pests still largely depends on the pesticides that are not universally compatible with beneficial arthropods (Campiche *et al*. 2006; Dawar *et al*. 2016) [2, 6]. Therefore, more selective insecticides are needed to improve the ecological system by conserving the beneficial fauna. Moreover, *Trichogramma* spp. could be the best indicator species to know the potential negative impact of different insecticides (Consoli *et al*. 2001) [5]. Application of selective insecticides to control crop pests could be useful in conservation of natural enemy fauna and some novel insecticides are potentially more harmful to the target pest but not to bio-control agent, thus playing significant role in conservation of biological control agents in agricultural ecosystem. The integration of chemical and biological methods requires a thorough knowledge of both factors and its compatible effects on bio-control agents when employed at farmers field. The study was carried out as a relative toxicity of various insecticides against *T. japonicum* with an objective to search out the comparatively least toxic insecticide against *T. japonicum*, as a one of the best components of IPM programme.

Material and Methods

The present study was undertaken at Biocontrol Laboratory, Department of Entomology, N.M. College of Agriculture, Navsari Agricultural Universtiy, Navsari (Gujarat) India. Insecticidal solutions were prepared in glass jar (1 litre capacity) (Table-1) and tested for relative toxicity on egg parasitization and adult emergence of *T. japonicum*.

Laboratory culture of *C. cephalonica* and egg parasitoid, *T. japonicum*

The rice moth, *Corcyra cephalonica* (Stainton) and egg parasitoid, *T. japonicum* were mass reared at Biocontrol Laboratory, Department of Entomology, NMCA, Nau, Navsari. The mass production of *C. cephalonica* was made as per the methodology given by Naganna and Shinde (2017) [19]. Moreover, the preparation of trichocard and mass culture of *T. japonicum* was made as per the techniques given by Mohapatra and Shinde (2021) [18].

Table 1: Insecticidal treatment used against *T. japonicum*

Treat No.	Insecticides/ treatments	Formulations (As per CIB&RC)	Conc. (%)	Tested dose	Spray fluid
T_1	Flubendiamide	39.35 SC	0.005	0.12 ml	1000 ml
T_2	Chlorantraniliprole	18.5 SC	0.003	0.16 ml	1000 ml
T_3	Fipronil	5 SC	0.007	1.4 ml	1000 ml
T_4	Thiacloprid	21.7 SC	0.01	0.46 ml	1000 ml
T_5	Quinalphos	25 EC	0.025	1.0 ml	1000 ml
T_6	Lambda- cyhalothrin	5 EC	0.0005	0.1 ml	1000 ml
T_7	Thiamethoxam	25 WG	0.003	0.12 g	1000 ml
T_8	Control (Treated with water)	--	--	--	1000 ml

Impact on egg parasitization of *T. japonicum*

Freshly laid U.V sterilized 100 *Corcyra* eggs were pasted on white card broad paper strip (5 × 3 cm) with the help of gum acacia (50% diluted). It was then air-dried thoroughly for 15 minutes and the prepared strips were dipped in targeted insecticide solution for the period five seconds. For the control treatment, card paper stripes were dipped in water. Strips were further air dried for 15 minutes and thereafter it was exposed to five freshly emerged as well as pre-mated females for 24 hours. After 24 hours, strips were taken out and kept in other same sized vials (6.5 × 5 cm) individually for further development. Number of eggs parasitized was worked out as per following formula;

$$\text{Parasitization (\%)} = \frac{\text{Total number of blackened eggs}}{\text{Total number of eggs taken for investigation}} \times 100$$

The obtained data on parasitization (%) were converted into arcsine transformed values and then subjected to CRD analysis to interpret the results. All the tested insecticides were categorized into four different safety categories and scored as per as reduction in parasitization compared to control.

Sr. No.	Safety category	Reduction in parasitization over control (%)	Toxicity score
1	Harmless	<30	1
2	Slightly harmful	30 to 79	2
3	Moderately harmful	80 to 99	3
4	Harmful	>99	4
(Hassan *et al.*, 2000) [11]			

Impact on adult emergence of *T. japonicum*

A total of thirty-two white card broad paper strips (5 cm H × 3 cm W) glued with U.V sterilized *Corcyra* eggs (100 numbers) were prepared and mass exposed to females of *Trichogramma* females for the period of 24 hours. After four days when the eggs colour changed to black, the strips having parasitized eggs were dipped into insecticidal solution for five seconds. Control treatment was considered only with dipping the strips in water. Further, all such strips were then air dried for 15 minutes and kept individually in same sized plastic vials (6.5 cm H × 5 cm D). The emerged adults of *Trichogramma* were counted (irrespective of sex) and reported. Thus, adult emergence (%) was worked as per following formula;

$$\text{Adult emergence (\%)} = \frac{\text{Total number of adults emerged}}{\text{Total number of blackened eggs taken for investigation}} \times 100$$

The data on adult emergence (%) was also converted into arcsine transformed values by employing CRD analysis. The insecticides were categorized into four categories as per methodology given by Hassan *et al.* (2000) [11] and as per reduction in adult emergence compared to control treatment.

Results

Impact on egg parasitization of *T. japonicum*

The egg parasitoid, *T. japonicum* parasitize egg mass of rice yellow stem borer as well as eggs of various lepidopteran pests in various cropping ecosystems and there are chances of contact with the sprayed insecticides and may kill the parasitoids thereby reduction in egg parasitization under the field condition. To ascertain the effect of target insecticides on parasitization, an experiment was laid out with pre-treated stripes of *Corcyra* egg wherein such cards were offered to females of *T. japonicum* for parasitization. The data on egg parasitization (%) are presented in Table 2 and graphically depicted in Fig 01 and given below.

The result clearly indicated that all the tested insecticides registered significantly lower egg parasitization as compared to control treatment (83.50%). The next best treatment with maximum egg parasitization after control treatment was registered with chlorantraniliprole 18.5 SC (79.50%) and it was further at par with thiacloprid 21.7 SC (78.25%) and thiamethoxam 25 WG (77.75%). In addition, treatment of flubendiamide 39.35 SC and fipronil 5 SC showed 74.25 and 32.75 per cent egg parasitization, respectively. However, lambda-cyhalothrin 5 EC showed significant reduction in parasitic performance of the female of *T. japonicum* with 2.25 per cent egg parasitization. Moreover, cent per cent reduction in egg parasitization was seen with quinalphos 25 EC.

Looking to the above results, the ascending order of different insecticides in terms of their toxic effects on egg parasitization of the female of *T. japonicum* were control treatment < chlorantraniliprole 18.5 SC < thiacloprid 21.7 SC < thiamethoxam 25 WG < flubendiamide 39.35 SC < fipronil 5 SC < lambda-cyhalothrin 5 EC < quinalphos 25 EC. Based on reduction in egg parasitization due to various insecticides over control treatment, the ranking of the insecticides were made and are presented as hereunder.

Toxicity score	Category	Perceived insecticides
1	Harmless (<30% reduction in parasitization over control)	Flubendiamide 39.35 SC Chlorantraniliprole 18.5 SC Thiacloprid 21.7 SC Thiamethoxam 25 WG
2	Slightly harmful (30 to 79% reduction in parasitization over control)	Fipronil 5 SC
3	Moderately harmful (80 to 99% reduction in parasitization over control)	Lambda-cyhalothrin 5 EC
4	Harmful (>99% reduction in parasitization over control)	Quinalphos 25 EC

From the above data, it can be seen that flubendiamide 39.35 SC, chlorantraniliprole 18.5 SC, thiacloprid 21.7 SC and thiamethoxam 25 WG along with the control treatment were found safer and categorized as harmless insecticide to the egg parasitization of female wasp (*T. japonicum*). However, fipronil 5 SC was slightly harmful insecticide and lambda-cyhalothrin 5 EC was moderately harmful insecticide to the egg parasitization of female *T. japonicum*. Furthermore, quinalphos 25 EC was grouped as harmful insecticide to the egg parasitization of *T. japonicum*

Table 2: Relative toxicity of different insecticides on egg parasitization of *T. japonicum*

Treat. No.	Treatments	Conc. (%)	Egg parasitization* (%)	Reduction in egg parasitization over control (%)	Toxicity score
T_1	Flubendiamide 39.35 SC	0.005	59.53 (74.25)	11.08	1
T_2	Chlorantraniliprole 18.5 SC	0.003	63.09 (79.50)	4.79	1
T_3	Fipronil 5 SC	0.007	34.89 (32.75)	60.78	2
T_4	Thiacloprid 21.7 SC	0.01	62.25 (78.25)	6.29	1
T_5	Quinalphos 25 EC	0.025	0.25 (0.00)	100.00	4
T_6	Lambda- cyhalothrin 5 EC	0.0005	8.38 (2.25)	97.31	3
T_7	Thiamethoxam 25 WG	0.003	61.89 (77.75)	6.89	1
T_8	Control (Water)	--	66.06 (83.50)	--	--
S. Em ±			0.90	--	--
C.D. at 5%			2.61	--	--
C.V. (%)			4.00	--	--

* Figures outside the parentheses are arc sine transformed values while those insides are original values

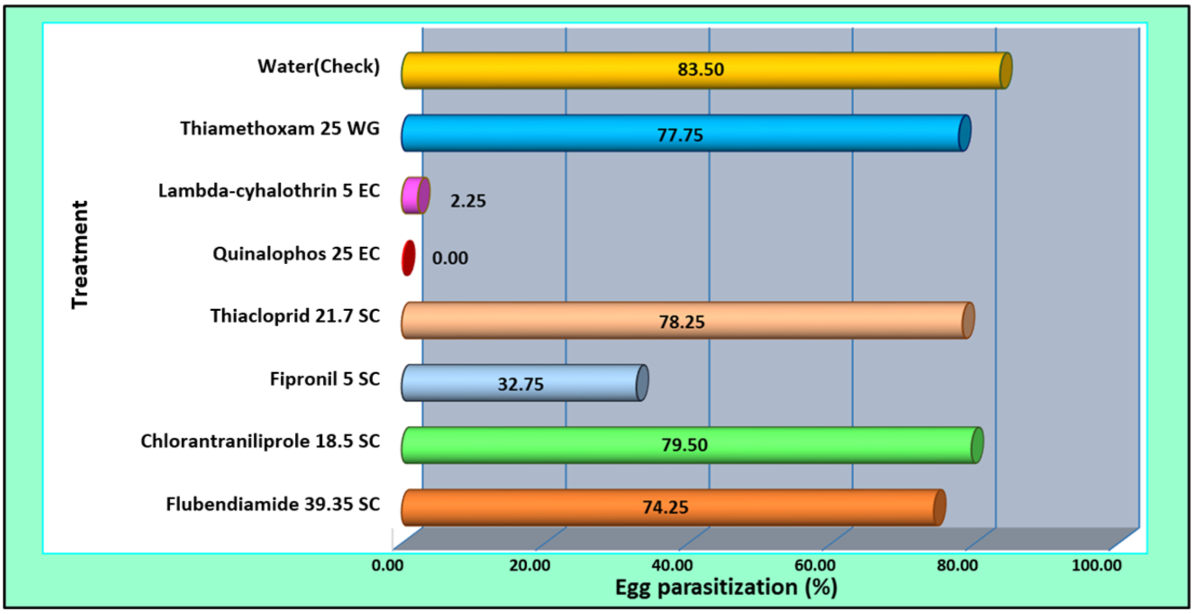

Fig 1: Relative toxicity of different insecticides on egg parasitization of *T. japonicum* under laboratory condition

Impact on adult emergence of *T. japonicum*

The sprayed insecticide may come in contact with *T. japonicum* and thereby reduces the emergence rate. Thus, to find out the adverse effect of the target insecticides on the adult emergence, a study was carried out pre-parasitized *Corcyra* eggs. The data adult emergence (%) are presented in Table 3 and depicted in Fig 02.

The results obtained during the present investigation revealed that among the various insecticides tested on *T. japonicum* parasitized eggs of *Corcyra*, control treatment had maximum adult emergence with 95.58 per cent followed by chlorantraniliprole 18.5 SC (91.79%). This was followed by thiacloprid 21.7 SC (88.97%), flubendiamide 39.35 SC (88.90%) and thiamethoxam 25 WG (88.12%) and all were at par. In addition, the treatment of lambda-cyhalothrin 5 EC and fipronil 5 SC registered 82.71 and 80.00 per cent adult emergence, respectively and both were found at par with each other. However, the strong negative impact on adult emergence of *T. japonicum* was recorded with quinalphos 25 EC (2.59% adult emergence).

It can be seen from the above findings, the ascending order of different insecticides in terms of their toxic effects on adult emergence of the *T. japonicum* were control treatment < chlorantraniliprole 18.5 SC < thiacloprid 21.7 SC < flubendiamide 39.35 SC < thiamethoxam 25 WG < lambda-cyhalothrin 5 EC < fipronil 5 SC < quinalphos 25 EC. Based on reduction in adult emergence over control, the various insecticides were grouped and are presented as hereunder.

Toxicity score	Category	Perceived insecticides
1	Harmless (<30% reduction in adult emergence over control)	Flubendiamide 39.35 SC Chlorantraniliprole 18.5 SC Fipronil 5 SC Thiacloprid 21.7 SC Lambda-cyhalothrin 5 EC Thiamethoxam 25 WG
2	Slightly harmful (30 to 79% reduction in adult emergence over control)	Nil
3	Moderately harmful (80 to 99% reduction in adult emergence over control)	Quinalphos 25 EC
4	Harmful (>99% reduction in adult emergence over control)	Nil

From the above data, it can be seen that quinalphos 25 EC caused severe reduction in the adult emergence of *T. japonicum* and was categorized as moderately harmful for adult emergence of parasitic wasp. However, all the remaining insecticides *viz.,* flubendiamide 39.35 SC, chlorantraniliprole 18.5 SC, fipronil 5 SC, thiacloprid 21.7 SC, lambda-cyhalothrin 5 EC and thiamethoxam 25 WG along with the control treatment were found safer and considered as harmless based on the adult emergence of *T. japonicum*.

Table 3: Relative toxicity of different insecticides on adult emergence of *T. japonicum* under laboratory condition

Treat. No.	Treatments	Conc. (%)	Adult emergence* (%)	Reduction in adult emergence over control (%)	Toxicity score
T_1	Flubendiamide 39.35 SC	0.005	70.63 (88.90)	6.99	1
T_2	Chlorantraniliprole 18.5 SC	0.003	73.49 (91.79)	3.97	1
T_3	Fipronil 5 SC	0.007	63.44 (80.00)	16.30	1
T_4	Thiacloprid 21.7 SC	0.01	70.77 (88.97)	6.92	1
T_5	Quinalphos 25 EC	0.025	8.93 (2.59)	97.29	3
T_6	Lambda- cyhalothrin 5 EC	0.0005	65.48 (82.71)	13.46	1
T_7	Thiamethoxam 25 WG	0.003	69.87 (88.12)	7.81	1
T_8	Control (Water)	--	77.88 (95.58)	--	--
S. Em ±			1.10	--	--
C.D. at 5%			3.21	--	--
C.V. (%)			3.52	--	--

* Figures outside the parentheses are arc sine transformed values while those insides are original values

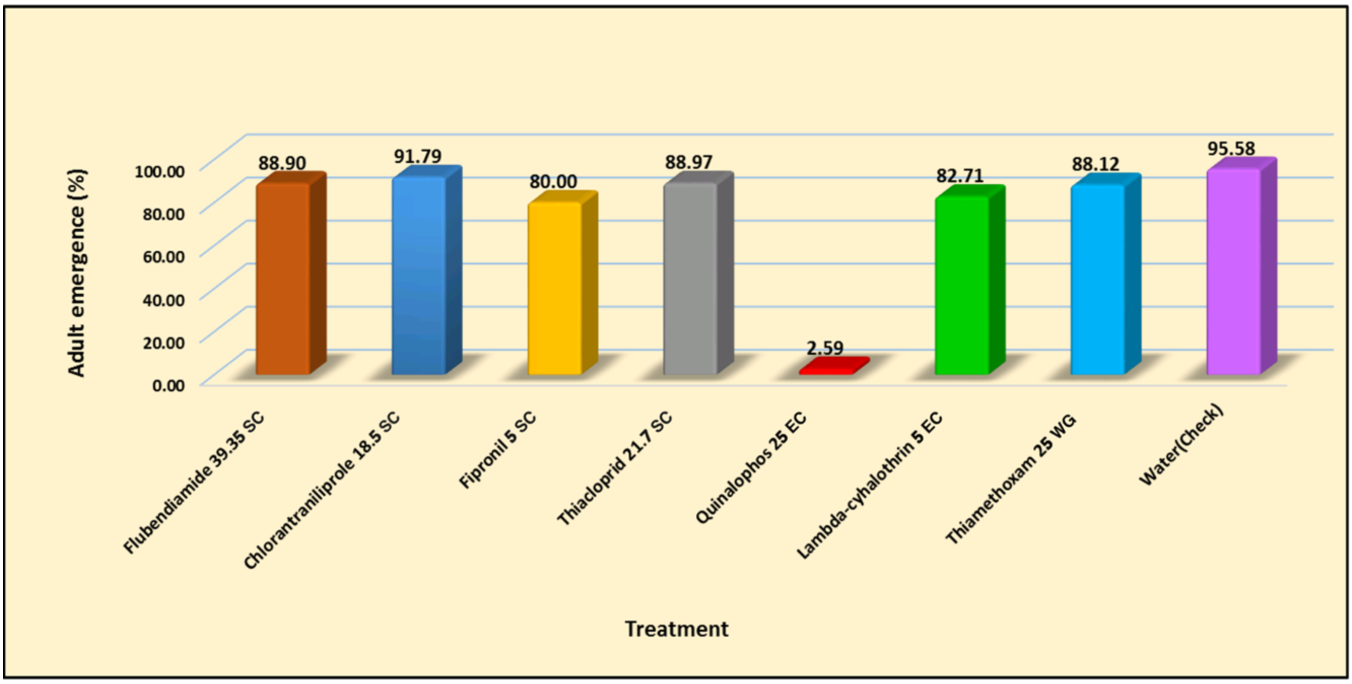

Fig 2: Relative toxicity of different insecticides on adult emergence of *T. japonicum* under laboratory condition

Discussion

Impact on egg parasitization of *T. japonicum*

The results of present investigation revealed that the impact of relative toxicity of various insecticides on the egg parasitization of *T. japonicum* varied to a great extent. The present investigation is akin with the findings of Deshmukh *et al.* (2018) [8] and Pawar *et al.* (2020) [21] who listed chlorantraniliprole and flubendiamide as harmless to the egg parasitization of *T. japonicum*. Further, Sattar *et al*. (2011) [23] reported that flubendiamide was found harmless to egg parasitization of *T. chilonis* showing highest rate of parasitization by *T. chilonis* female (7.48 eggs/female) among the tested insecticides. Fontes *et al.* (2018) [10] categorized chlorantraniliprole as harmless to egg parasitization of *T. achaeae*. The findings of De Paiva *et al.* (2018) [7] supports the present study who reported that chlorantraniliprole and flubendiamide were categorized as harmless to the egg parasitization of *T. pretiosum*. According to Sudhanan *et al.* (2014) [26], among all the tested insecticides, flubendiamide (84.16, 80.20 and 80.04% parasitization at 50, 60 and 70 g a.i/ ha, respectively) recorded the maximum egg parasitism by *T. chilonis* which was followed by chlorantraniliprole (75.86%). Thus, these reports are in line with the present findings.

The present findings are in tally with the Khan and Khan (2015) [15] who revealed that fipronil was categorized as slightly harmful to the egg parasitization of *T. chilonis* and Singh and Varma (1986) [24] who noticed that complete inhibition of egg parasitization by female *T. brasiliensis* was seen when host eggs were treated with quinalphos. In contrast to above workers, Sudhanan *et al.* (2014) [26] recorded that fipronil had 73.23 per cent egg parasitization and found comparatively safer to the egg parasitization of *T. chilonis* which disagrees with the present study. Further, Fontes *et al.* (2018) [10] showed that thiamethoxam and lambda-cyhalothrin were considered as slightly harmful and moderately harmful to the egg parasitization of *T. achaeae*, respectively which is more or less similar to the present investigation. Furthermore, Pawar *et al.* (2020) [21] showed that based upon the reduction in egg parasitization over control, lambda-cyhalothrin was categorized as moderately harmful while thiamethoxam and thiacloprid were considered as slightly harmful to the egg parasitization of *T. japonicum* which partly follows the present findings. De Paiva *et al.* (2018) [7] noted that lambda-cyhalothrin + thiamethoxam was found moderately harmful to the egg parasitization of *T. pretiosum* which is in accordance with the present findings. Moreover, Jiang *et al.* (2019) [14] reported that thiacloprid was found toxic to the host egg parasitism of *T. dendrolimi*, *T. ostriniae* and slightly toxic to egg parasitism of *T. confusum* whereas thiamethoxam showed significant reduction in parasitism of all the three Trichogrammatid species. Abdulhay and Rathi (2014) [1] noted that thiacloprid categorized as slightly harmful to the egg parasitization of *T. evanescens* which deviates from the present findings as thiacloprid and thiamethoxam were existed as harmless to the egg parasitization of *T. japonicum* during the present investigation. The findings of above scientists support the present investigation while difference in the relative toxicity of various insecticides on egg parasitization of *Trichogramma* wasp might be due to change in test insect, mode of action of tested insecticides, host eggs used for investigation.

Impact on adult emergence of *T. japonicum*

The close perusal of the data showed that all the tested insecticides had little or no selectivity towards adult emergence of *T. japonicum*. The findings are in accordance with Sattar *et al.* (2011) [23] who showed that flubendiamide was harmless for adult emergence of *T. chilonis* when treated at egg, larva and pupal stages of parasitoid inside *H. armigera* eggs whereas Hussain *et al.* (2012) [12] observed that flubendiamide and chlorantraniliprole were found to be safer to the adult emergence of *T. chilonis* when treated with the respective insecticide at 1st, 2nd, 3rd, 4th, 5th and 7th days after parasitization which is also in support of the present investigation. Sudhanan *et al.* (2014) [26] who noticed the highest adult emergence of *T. chionis* with flubendiamide (89.70, 87.30 and 82.91% adult emergence at 50, 60 and 70 g a.i/ ha, respectively) among all the tested insecticides followed by chlorantraniliprole (81.78%) and fipronil (78.24%) which are comparable to the present work. The present investigations are akin with Wahengbam *et al.* (2018) [28] who reported that chlorantraniliprole and thiacloprid were regarded as harmless to adult emergence of *T. chilonis* and *T. pretiosum.* Furthermore, De Paiva *et al.* (2018) [7] reported that chlorantraniliprole and flubendiamide were considered as harmless to the adult emergence of *T. pretiosum*. Hence, the present findings are in support with previous workers. Abdulhay and Rathi (2014) [1] disagrees with the above findings who registered that thiacloprid noted 60.75 per cent the reduction in adult emergence over control and categorized as slightly harmful to the adult emergence of *T. evanescens.* Another deviation from the present findings is Khan and Khan (2015) [15] who revealed that fipronil was proved to be harmful to the adult emergence of *T. chilonis* while fipronil was registered as harmless during the course of present investigation. Varma and Singh (1987) [27] reported that quinalphos caused complete inhibition of adult emergence of *T. brasiliensis* from the parasitized eggs which agrees with the present investigation.

Moreover, the present findings are more or less similar with Ko *et al.* (2015) [16] who reported that thiamethoxam was considered as harmless to adult emergence of *T. chilonis* when treated at egg and prepupal stages of parasitoid while it was found slightly harmful when treated at larval stage of the parasitoid. Another support to present study is provided by Carvalho *et al.* (2006) [3] who found that lambda-cyhalothrin, thiamethoxam and thiacloprid were categorized as harmless to the adult emergence of *T. pretiosum*. In contrast to this, Charles *et al.* (2000) [4] showed that lamda-cyhalothrin adversely affected the adult emergence of *T. exiguum* (1.4%) from the parasitized eggs of *H. zea*. Moreover, a great discrepancy is seen between the present findings and De Paiva *et al.* (2018) [7] who reported that lambda-cyhalothrin + thiamethoxam was found slightly harmful to the adult emergence of *T. pretiosum* when eggs were treated with the respective insecticide at 8th day after parasitization. The findings of above workers provide support to the present work. The disparity among the relative toxicity of different insecticides on adult emergence of Trichogrammatid parasitoid might be due to different species of *Trichogramma*, doses and mode of action of insecticides under study, methodology employed and prevailing weather condition during the investigation.

Conclusion
The present investigation indicated that there were some insecticides under testing found harmful where as some were considered comparatively safer to the *T. japonicum*. Therefore, application of moderately harmful to harmful insecticides should be avoided at least up to 72 hrs after release of parasitoid as well as during the peak activity period of parasitoid under field condition. Moreover, keeping the view of possible effect of the tested insecticides on parasitoid, the integration of these insecticides should be considered with utmost care during the implementation of integrated pest management programme.

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