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Comparative toxicity of certain insecticides against *Chrysoperla* Spp. (Neuroptera: chrysopidae) under laboratory conditions

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Abstract
Toxicity of newer insecticides *viz*., chlorantraniliprole 18.5 SC, flubendiamide 20 WG, buprofezin 25 SC, lambda cyhalothrin 9.5 + thiamethoxam 12.6 ZC, lambda cyhalothrin 5 EC, azadirachtin 5 w/w, thiamethoxam 25 WG, thiacloprid 21.7 SC were tested against different larval stages of *Chrysoperla spp.* under laboratory conditions. The observations on percent mortality of 1st, 2nd and 3rd instar of *C. spp*. larvae were recorded after 24, 48 and 72 hrs of application. The results showed that lambda cyhalothrin + thiamethoxam was found toxic to all instars of *Chrysopa* larvae whereas azadirachtin was found safest insecticide against all instar of *Chrysopa* larvae at all treatment intervals. Considering ecofreindliness, it can be concluded that insecticides azadirachtin, buprofezin and flubendiamide were found most safer to all instars of *C. spp.* which can be taken successfully in the IPM programmes whereas, lambda cyhalothrin + thiamethoxam was found most harmful to all instars of larvae.

Keywords: *Chrysoperla spp.*, mortality, larvae, toxicity, insecticides

Introduction

Biological control is involvement of parasitoids, predators and pathogens in maintaining other organisms density at a lower average level than would occur in their absence (DeBach, 1965) [4]. Bioagent can cause substantial decrease in pest population numbers (Hassell, 1978) [12]. In different agro-ecosystem the Green lacewing is an important natural enemy, belonging to family Chrysopidae, order Neuroptera. Green lacewing is a native North American species (Garland, 1985) [8]. *Chrysoperla carnea* is predominant species of green lacewing, having green cylindrical body, transparent wing with light green veins, long filiform antennae, golden eyes and stalked eggs that offer protection from predation (Pedigo, 1989) [24]. Larval stage is active predatory stage (Michaud, 2001) [19]. Larvae of *Chrysoperla carnea* is polyphagous, voracious feeder of Cotton aphids, *Aphis gossypii* Glover; Corn earworm, *Helicoverpa zea* Boddie (Lingren *et al*., 1968) [16]; Colarado potato beetle, *Leptinotarsa decemlineata* (Say) (Nordlund *et al.*, 1991) [22].

In agroecosystems, various biocontrol agents are one of the important components of the pest management, which have proved as best alternative over several insecticides (Singh, 2015) [26]. The insecticides commonly used to control the pest population have indirect effect on these natural enemies. Constant disturbances to agricultural ecosystem using chemical pesticides upset the natural balance causing pest up surgence (Prema *et al.*, 2016) [25]. The impact of synthetic pesticides on the environment, the beneficial arthropods and the human health by exposure to these chemicals are issues of growing concern (Garzon *et al*., 2015) [9]. The need for more selective insecticides was one of the key themes during the evolution of poison free management of insect-pests (Sparks, 2013) [29]. The purpose of work reported here was to evaluate the effects of insectcides on larvae of *C. carnea* under laboratory conditions.

Materials and Methods

Laboratory studies were conducted to find out the toxicity of newer group of insecticides on *Chrysoperla sp.* (carnea- group) during Kharif 2017-2018. The rearing of the host insect and predator was done under controlled room temperature and relative humidity conditions ranging

Between 26 ± 2^0c and 60 ± 5%, respectively and commercial formulations of insecticides used for the study were procured from the market. Mass multiplication of *Chrysopa* was done in the laboratory to obtain healthy culture of the test predator. The initial culture was obtained from the already established culture of *Chrysopa* in Biocontrol laboratory, Entomology Section College of Agriculture, Nagpur and was further multiplied on the factitious laboratory host, eggs of rice moth. To obtain the eggs of *Corcyra cephalonica* throughout the experimental period, rearing of rice moth was done in the laboratory and the culture was maintained on sorghum based artificial diet.

Table 1: Treatment details:

Tr. No.	Treatment name	Conc. (%)
T1	Chlorantraniliprole 18.5 SC	0.005
T2	Flubendiamide 20 WG	0.01
T3	Buprofezin 25 SC	0.05
T4	Lambda cyhalothrin 9.5 + thiamethoxam 12.6 ZC	0.008
T5	Lambda cyhalothrin 5 EC	0.003
T6	Azadirachtin 5 w/w	0.002
T7	Thiamethoxam 25 WG	0.005
T8	Thiacloprid 21.7 SC	0.004
T9	Control (water spray)	-

The insecticides were classified in different categories on the bases of per cent mortality of larvae, as suggested by IOBC/WPRS (Sterk *et al*. 1999) [30] as under:
Harmless (toxicity class 1) = less than 30 % mortality,
Slightly harmful (toxicity class 2) = 30-79 % mortality,
Moderately harmful (toxicity class 3) = 80-89 % mortality,
Harmful (toxicity class 4) = more than 90 % mortality.

Treatment of larvae
Leaf dip assay was used to treat the larvae, as it more closely to the field exposure. Fresh, medium size and unsprayed leaves of cotton (*Gossypium hirsutum*) variety Bt- cotton were collected from 60 days old plants. Leaves were cut into same diameter of petri plates. Leaf discs were dipped for 5 seconds into insecticide solutions and in water for control. All treated and control leaf discs were allowed to air dry and placed in Petri plates (1.25 cm dia. and 0.75 cm deep). Water moistened filter paper placed underneath of each leaf disc. The larvae of 1^{st}, 2^{nd} and 3^{rd} instars were placed in Petri plates separately. Irradiated eggs of *Corcyra cephalonica* was sprinkled in each Petri plate as a food for larvae. The treated larvae were considered dead when they no longer moved, twitched or when being touched 2-3 times with a brush. Mortality data was recorded at end of each exposed and subsequent instars after 24, 48 and 72 hrs up to pupation.
The mean per cent mortality of larvae in different treatments were calculated by the formula given here under:

$$\text{Mean per cent mortality of larvae} = \frac{\text{Number of larvae dead}}{\text{Number of larvae released}} \times 100$$

Statistical Analysis
The data on all relevant observation thus obtained were subjected for appropriate statistical analysis, Gomez and Gomez (1984) [10]. The corrected per cent mortalities were transformed to arcsine percentage and subjected to statistical analysis adopting Completely Randomized Design (CRD).

Results and Discussion

The results obtained from the present investigation as well as relevant discussion have been summarized under following heads:

Effect of various insecticides on per cent mortality of first instar *Chrysoperla spp.* larvae
The figures presented in table 2 exhibited that the toxicity of different insecticides against first instar *Chrysopa* larvae varied significantly after different time intervals and increased significantly with the increase in exposure period from 24 hrs to 72 hrs.
The mortality data showed that the safest insecticide was azadirachtin with 26.66 per cent mortality of *Chrysopa* larvae and differed significantly with control, where as maximum mortality of first instar larvae of *Chrysopa* was obtained with lambda cyhalothrin + thiamethoxam (90.00 %) followed by thiamethoxam, lambda cyhalothrin, chlorantraniliprole, thiachloprid, flubendiamide and buprofezin with 86.66, 83.33, 80.00, 53.33, 36.66 and 33.33 per cent, respectively up to 72 hrs exposure period. Similar results were also reported by, Aggarwal and Brar (2005) [1] reported that neemazal T/S 1.0% at 200, 400, 800 mg/l caused mortality with 12.33, 33.36 and 45.33 per cent first instar larvae of *C. carnea.* However, Erkilic and Ygnn (1997) [7] and Liu *et al.* (2000) [17] stated that buprofezin was harmless with very limited detrimental effects on natural enemies and did not affect development of instars and pupae of *Chrysoperla rufilabris*, respectively. Ullah *et al.* (2017) [31] observed that flubendiamide caused 43.33 mortality of the 1^{st} instar larvae. Ahmad *et al*. (2014) [2] and Moens *et al*. (2011) [20] reported that thiacloprid was safer to natural enemies and larvae of diptran hoverfly, *Episyrphus balteaus* and predatory mite, *N. fallacis* in the laboratory conditions, respectively. Hussain *et al.* (2012) [14] and Huggi and Mallapur (2016) [13] found that chlorantraniliprole as intermediately toxic which caused 72 and 85.00 per cent mortality of larvae of *C. carnea* respectively. Govindaraj *et al.* (2015) [11] found that lambda cyhalothrin @ 25 g a.i. ha[1] causing 83.33 per cent grub mortality. Khuram and Ashfaq (2011) [15] assessed that thiamethoxam (0.12 g/l) caused toxicity to 1^{st} instar larvae of *C. carnea* of about 62.5 per cent mortality under laboratory conditions.

Table 2: Effect of various insecticides on per cent mortality of first instar of *Chrysoperla spp.* Larvae

Tr. No	Treatments	Dose/l.	1^{st} instar larval mortality (%)		
			24 hrs	48 hrs	72 hrs
T_1	Chlorantraniliprole 18.5 SC	0.27ml	23.33 (28.86)	50.00 (45.00)	80.00 (63.43)
T_2	Flubendiamide 20 WG	0.5gm	16.66 (24.04)	23.33 (28.86)	36.66 (37.23)
T_3	Buprofezin 25 SC	2ml	13.33 (21.39)	20.00 (26.57)	33.33(35.24)
T_4	Lambda cyhalothrin 9.5 + thiamethoxam 12.6 ZC	0.36ml	33.33 (35.24)	60.00 (50.77)	90.00 (71.57)
T_5	Lambda cyhalothrin 5 EC	0.6ml	26.66 (31.05)	53.33 (46.89)	83.33 (65.88)
T_6	Azadirachtin 5 w/w	0.4ml	10.00 (18.43)	16.66 (24.04)	26.66 (31.05)
T_7	Thiamethoxam 25 WG	0.2gm	30.00 (33.21)	56.66 (48.79)	86.66 (68.28)
T_8	Thiachloprid 21.7 SC	0.18ml	20.00 (26.57)	40.00 (39.23)	53.33 (46.89)

T9	Control	Water	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	'F' test		Sig.	Sig.	Sig.
	SE (m) ±		1.78	1.62	1.91
	CD at 5%		5.17	4.71	5.55

(Values in the parenthesis are arc sine transformed)

Effect of various insecticides on per cent mortality of second instar *Chrysoperla spp.* larvae.

The data contained in Table 3 revealed that the mortality of third instar *Chrysopa* larvae increased significantly with the increase in exposure period from 24 hrs to 72 hrs.

The mortality data showed that the safest insecticide was azadirachtin with 23.33 per cent mortality of *Chrysopa* larvae and differed significantly with control, where as maximum mortality of first instar larvae of *Chrysopa* was obtained with lambda cyhalothrin + thiamethoxam (73.33 %) followed by, lambda cyhalothrin, thiamethoxam, chlorantraniliprole, thiachloprid, flubendiamide and buprofezin with 73.33, 70.00, 56.66, 53.33, 46.66, 33.33 and 26.66 per cent larval mortality, respectively, up to 72 hrs exposure period.

Table 3: Effect of various insecticides on per cent mortality of second instar *Chrysoperla spp.* larvae.

Tr. No	Treatments	Dose/l	2nd instar larval mortality (%)		
			24 hrs	48 hrs	72 hrs
T1	Chlorantraniliprole 18.5 SC	0.27ml	23.33 (28.86)	36.66 (37.23)	53.33(46.89)
T2	Flubendiamide 20 WG	0.5gm	16.66 (24.04)	26.66 (31.05)	33.33 (35.24)
T3	Buprofezin 25 SC	2ml	13.33 (21.39)	23.33 (28.86)	26.66 (31.05)
T4	Lambda cyhalothrin 9.5 + thiamethoxam 12.6 ZC	0.36ml	33.33 (35.24)	50.00 (45.00)	73.33 (58.89)
T5	Lambda cyhalothrin 5 EC	0.6ml	30.00 (33.21)	46.66 (43.05)	70.00 (56.79)
T6	Azadirachtin 5 w/w	0.4ml	10.00 (18.43)	16.66 (24.04)	23.33 (28.86)
T7	Thiamethoxam 25 WG	0.2gm	26.66 (31.05)	43.33 (41.15)	56.66 (48.79)
T8	Thiachloprid 21.7 SC	0.18ml	20.00 (26.57)	33.33 (35.24)	46.66 (43.05)
T9	Control	Water	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	'F' test		Sig.	Sig.	Sig.
	SE (m) ±		1.78	1.90	1.82
	CD at 5%		5.17	5.53	5.31

(Values in the parenthesis are arc sine transformed)

Similar results were also reported by, Shah *et al*. (2012) [27] proved neembaan as the safest, showing less than 50% mortality even at higher doses. Meghannavar and Kambrekar (2016) [18] in their studies observed the toxicity of insecticides to second instar grub of *Chrysoperla zastrowi sillemi* and classified buprofezin 50 SC @ 0.25 ml/l as harmless. Hussain *et al.* (2012) [14] found that flubendiamide and chlorantraniliprole caused 56.00 and 64 per cent mortality of 2nd instar larvae respectively where as, Amarsekare and Shearer (2013) [3] observed that lambda cyhalothrin caused significant mortality to both species of second instar *Chrysoperla carnea* and *Chrysoperla johnsoni* with 73.3 and 66.7 per cent mortality at 10 DAT (Days After Treatment). El-Zahi and El-Zahi (2012) [6] found that thiamethoxam caused 42.18% mortality against second instar larvae of *C. carnea* and Nasreen *et al.* (2005) [21] also found that thiamethoxam as moderately harmful to *C. carnea* larvae at lower concentration (C1) and was toxic at recommended and higher concentrations which is in line with the present studies.

Effect of various insecticides on per cent mortality of third instar *Chrysoperla spp.* larvae.

The data contained in Table 4 revealed that the mortality of third instar *Chrysopa* larvae increased significantly with the increase in exposure period from 24 hrs to 72 hrs.

After 72 hrs exposure period, the order of toxicity of tested insecticides *viz*., lambda cyhalothrin + thiamethoxam, lambda cyhalothrin, chlorantraniliprole, thiamethoxam, thiachloprid, flubendiamide and buprofezin was recorded with 63.33, 46.66, 43.33, 36.66, 33.33, 26.66, 20.00 per cent larval mortality, respectively. However, application of azadirachtin was significantly less toxic to the third instar larvae of *Chrysopa* with 16.66 per cent larval mortality at 72 hrs, respectively. Thus, data presented in table 4 showed that as the larvae became larger in size from first to third instar, their tolerance to insecticides also increased.

Table 4: Effect of various insecticides on per cent mortality of third instar *Chrysoperla spp.* larvae.

Tr. No.	Treatments	Dose/lit.	3rd instar larval mortality (%)		
			24 hrs	48 hrs	72 hrs
T1	Chlorantraniliprole 18.5 SC	0.27ml	23.33 (28.86)	36.66 (37.23)	43.33(41.15)
T2	Flubendiamide 20 WG	0.5gm	16.66 (24.04)	23.33(28.86)	26.66(31.05)
T3	Buprofezin 25 SC	2ml	13.33(21.39)	16.66 (24.04)	20.00 (26.57)
T4	Lambda cyhalothrin 9.5 + thiamethoxam 12.6 ZC	0.36ml	33.33 (35.24)	50.00 (45.00)	63.33 (52.71)
T5	Lambda cyhalothrin 5 EC	0.6ml	30.00 (33.21)	40.00 (39.23)	46.66 (43.05)
T6	Azadirachtin 5 w/w	0.4ml	10.00 (18.43)	13.33 (21.39)	16.66 (24.04)
T7	Thiamethoxam 25 WG	0.2gm	26.66 (31.05)	33.33 (35.24)	36.66 (37.23)
T8	Thiachloprid 21.7 SC	0.18ml	20.00 (26.57)	30.00 (33.21)	33.33 (35.24)
T9	Control	Water	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	'F' test		Sig.	Sig.	Sig.
	SE (m) ±		1.78	1.75	1.87
	CD at 5%		5.17	5.09	5.45

(Values in the parenthesis are arc sine transformed)

Similar results were also reported by, Aggarwal and Brar (2005) [1] reported that neemazal T/S 1.0% at 200, 400 800 mg/l caused 8.33, 10.33 and 16.86 per cent mortality of third instar larvae of *C. carnea*. Erkilic and Ygnn (1997) [7] who showed no or very limited detrimental effects on natural enemies, whereas Hussain *et al.* (2012); Garzon *et al.* (2015) and Ullah *et al.* (2017) [14, 9, 31] reported that flubendiamide and chlorantraniliprole which caused 52.00 and 68 per cent post treatment mortality of 3rd instar larvae and flubendiamide was found safer to 3rd instar larvae causing 6-36 per cent mortality, respectively. Omar *et al.* (2002) and Devi *et al.* (2014) [23, 5] reported that undesirable effect of thiamethoxam against *C. carnea* which decreased 31.00 and 26.67 per cent 3rd instar larval mortality. Silva *et al.* (2017) [28] classified lambda cyhalothrin as harmful (Class 4) in toxicity classes which is line with the present findings.

Conclusion
Considering ecofriendliness, it can be concluded from the present investigation that insecticides azadirachtin, buprofezin and flubendiamide were found most safer to all instars of *Chrysoperla spp.* which can be taken successfully in the IPM programmes. Where as lambda cyhalothrin + thiamethoxam was found most harmful to all instars of larvae

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