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Compatibility potential of *Steinernema bicornatum* with insecticides in laboratory condition

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Abstract
The present investigation was studied with the title "Compatibility potential of *Steinernema bicornatum* with insecticides in laboratory condition" and the work was carried out in the Post Graduate laboratory of Entomology Section, College of Agriculture Nagpur during 2020-21 with aim to test the compatibility of CIB registered insecticides with entomopathogenic nematode *Steinernema bicornatum*. The compatibility of *S. bicornatum* with combination of thiamethoxam 25% WG, diafenthiuron 50% WP, imidacloprid 17.8% SL, chlorantraniliprole 18.5% SC, indoxacarb 14.5% SC, cypermethrin 10% EC, chlorpyriphos 20% EC with double the recommended dose was found compatible with all treated insecticides except cypermethrin 10% EC and chlorpyriphos 20% EC which were found incompatible with *S. bicornatum.*

Keywords: compatibility, *Steinernema bicornatum*, entomopathogenic nematodes

Introduction
The widespread use of broad-spectrum pesticides against potential pests has resulted in the appearance of pesticide resistance in many pests, phytotoxicity, pesticide residue problems on plant products, growing cost of the crop production and overall negative environmental impacts on human and animal health (Dalvi *et al*., 2011) [4]. Entomopathogenic nematodes of the families Steinernematidae and Heterorhabditidae are soil inhabiting insect pathogens that possess potential as biological control agents (Gaugler and Kaya, 1990; Gaugler, 2002) [8, 7]. Nematodes are exposed to a variety of agrochemicals and formulations of biological control agents, when applied as part of an IPM programme that could be toxic and impair the nematode performance. EPNs could be effective in integrated pest management (IPM) programs as long-term suppressive agents used in combination with quick knockdown products such as commercially available insecticides. Nematode tolerance to agrochemicals allows the possibility of tank mixing nematodes with other agrochemical and biopesticide formulations that will save time and labour costs and achieve better control of a target pest species (Koppenhofer and Grewal 2005) [13, 14]. The broad host range of EPNs is advantageous, as the possibility exists of controlling more than one pest species in a single IPM programme. Overuse of chemical insecticides is common when insect's pest incidence is severe. Hence, with increased awareness among farmers to adopt safer control measures there is increasing emphasis on integrated pest management (IPM) where biological control agents can be deployed for management of insect pest. This has been well established with this pest in laboratory and field conditions. However, before an IPM technique for the control of this pest can be brought out, it is important that the compatibility of the nematodes with insecticides registered against this pest needs to be established. Moreover, chemical insecticide developing industries often do not test product toxicity to entomopathogens, only safety for predators and parasitoids is established (Alves *et al*., 1998) [1]. Hence, research is needed to know whether the insecticides are having any deleterious effect on EPNs before combining them with insecticides. The present investigation was carried out to evaluate compatibility of *S. bicornatum* with chemical insecticides registered against *Spodoptera litura* under laboratory conditions, so as to enable the integration of these control methods into effective management strategies. This work reports the effect of direct exposure to insecticide solutions on the survivability, infectivity and reproduction of *S. bicornatum.*

Material and Methods

Methodology

1. Stock solution at double the recommended concentration of the pesticide was prepared in distilled water. The suspension of infective juveniles was prepared in distilled water with a concentration of 2000 IJ/ml and one ml of nematode suspension was transferred to each container.
2. One ml solution of pesticide was added to the nematode suspension in each container so that final pesticide concentration was equal to the recommended concentration.
3. The recommended doses of pesticides are as per the Central Insecticide Board and Registration Committee, Faridabad, Haryana, India. Distilled water without chemical was used as control. The treatments were replicated four times.
4. The plates were kept at 25 ± 1°C. The mortality of IJs was recorded after 12 hrs, 24 hrs, 48 hrs and 72 hrs.
5. The observation was taken by taking 50 µl aliquots from each container and observed under the stereo zoom microscope.

Method of recording observations

The observations were recorded for nematode mortality, Straight IJs having no motion and not responding to prodding were counted as dead. The interpretations of observations on the compatibility of nematodes with chemicals were made based on the record of proportion of nematodes dead stage.

Highly compatible: If more than 86% per cent of the nematode's survival recorded as highly compatible with insecticide.

Compatible: If nematode survival per cent was found between 70-85% then recorded as compatible with insecticide.

Least compatible: If nematode survival per cent was found 50-70% then recorded as least compatible with insecticide.

Incompatible: If nematode survival per cent was found < 50% then recorded as incompatible with insecticide.

Treatment Details

Sr. No.	Trade name	Technical name	Group	% a.i.	Recommended dose (per liter)	Source
1.	Actara®	Thiamethoxam	NN	25 WG	0.2 gm	Syngenta India Ltd.
2.	Pegasus®	Diafenthiuron	Thiourea	50 WP	2.5 gm	Syngenta India Ltd.
3.	Confidor®	Imidacloprid	NN	17.8 SL	0.1 ml	Bayer Crop Science Ltd.
4.	Coragen®	Chlorantraniliprole	D	18.5 SC	0.3 ml	FMC India Pvt. Ltd.
5.	king Doxa®	Indoxacarb	O	14.5 SC	0.65 ml	Gharda Chemicals Ltd.
6.	Cymbush®	Cypermethrin	SP	25 EC	0.6 ml	Syngenta India Ltd.
7.	Excel ®	Chlorpyriphos	OP	20 EC	5 ml	Moti Insecticides Pvt Ltd.
8.	Control	Distilled water	-	-	-	-

(*CIB&RC list of label claim insecticides updated on 30.11.2020 (http://ppqs.gov.in/divisions/cib-rc/major-uses-of-pesticides) # Per ha dose is for dilution in 500 litres of water.)

Statistical Analysis

Survivors' nematode data were expressed in percentage. All data were arcsine transformed prior to statistical analysis performed with OPSTAT statistical software available on Hissar Agricultural University, Hissar.

$$\text{Per cent Survival (\%)} = \frac{\text{Total no.of nematode IJs} - \text{No.of Died IJs of nematodes}}{\text{Total No.of nematode IJs}} \times 100$$

Results and Discussion

The results of the studies made on the "Compatibility potential of *Steinernema bicornatum* with insecticides in laboratory condition" at COA Nagpur during 2020-21 have been presented and discussed aspect wise in accordance with the available literature.

To know the compatibility of CIB registered insecticides with entomopathogenic nematodes, *Steinernema bicornatum*

An experiment was conducted to study the effect of different insecticides on entomopathogenic nematodes *S. bicornatum* in laboratory condition. For these Seven insecticides viz., Thiamethoxam 25% WG (T_1), Diafenthiuron 50% WP (T_2), Imidacloprid 17.8% SL (T_3), Chlorantraniliprole 18.5% SC (T_4), Indoxacarb 14.5% SC (T_5), Cypermethrin 25% EC (T_6), Chlorpyriphos 20% EC (T_7) and water as control (T_8) were used at their doubled of the recommended dose as mentioned in the material and methods. Observations on per cent survival of IJs of *S. bicornatum* after 12 hrs, 24 hrs, 48 hrs and 72 hrs were recorded and presented in Table-1 (fig.1).

Per cent survival of *S. bicornatum*

Results presented in Table-1 (fig.-1) revealed that the per cent survival of *S. bicornatum* with different pesticides after 12 hrs, 24 hrs, 48 hrs and 72 hrs was observed and the results obtained were presented under following subheads:

Survivals after 12 hrs of exposure

In this observation the per cent Survival of *S. bicornatum* was observed in the treatments. In control, there was no mortality of nematodes. Among all insecticides tested, maximum per cent survival was recorded in Imidacloprid 17.8% SL (90.93%) i.e., highly compatible with *S. bicornatum* followed by Chlorantraniliprole 18.5% SC (85.70%), Diafenthiuron 50%WP (84.70%), Cypermethrin 25% EC (83.70%), Thiamethoxam25% WG (83.60) which at par with each other. After 12 hrs, all most all Insecticides were compatible with *S. bicornatum.* The minimum per cent survival observed in Chlorpyriphos 20% EC (82.20) and Indoxacarb 14.5% SC (81.05) and they were at par with each other.

Survivals after 24 hrs of exposure

In this observation the survival percentage observed in all treatments and per cent survival observed in untreated control. Highly compatible with *S. bicornatum* and found significantly differed followed by Chlorantraniliprole 18.5% SC (83.45%), Thiamethoxam 25% WG (82.50) Indoxacarb 14.5% SC (81.05%) and found at par with each other. Among all treatments minimum per cent survival was observed in Chlorpyriphos 20% EC (26.95%) which was least compatible with *S. bicornatum* followed by Cypermethrin 25% EC (49.05) and Diafenthiuron 50% WP (74.60%) respectively, which differed significantly with each other.

Survivals after 48 hrs of exposure

In this observation survival percentage (99.10%) of *S. bicornatum* was significantly higher in untreated control as compared to all treatments. Among insecticides tested maximum per cent survivals were observed in Imidacloprid 17.8% SL (86.15%) i.e., highly compatible with *S. bicornatum.* Chlorantraniliprole 18.5% SC (78.95%) followed by Thiamethoxam 25% WG (76.15%), Diafenthiuron 50% WP (73.70%), was at par with each other. Among all treatments minimum per cent survival was observed in Chlorpyriphos 20% EC (17.56%) i.e., least compatible with *S. bicornatum* followed by Cypermethrin 25% EC (40.80%) and Indoxacarb 14.5% SC (63.90%) respectively, which differed significantly to each other.

Survivals after 72 hrs of exposure

The survival per cent (98.30%) of *S. bicornatum* was significantly higher in untreated control as compared to all treatments. Among insecticides Imidacloprid17.8% SL (83.65%) showed maximum survival per cent i.e., highly compatible with *S. bicornatum* and found significantly superior than other insecticides. Thiamethoxam 25% WG (76.60%) followed by Diafenthiuron 50% WP (72.55%), Chlorantra- niliprole 18.5% SC (65.05%) and in that order found at par with each other. Among all treatments minimum survival per cent recorded in Chlorpyriphos 20% EC (13.75%) i.e., least compatible with *S. bicornatum* followed by Cypermethrin 25% EC (39.15%) and Indoxacarb 14.5% SC (60.75%) respectively, which differed significantly to each other.

Table 1: Survival per cent of *S. bicornatum* at 12 hrs, 24 hrs, 48 hrs and 72 hrs of exposure to the treatments

Sr. No	Treatments	Survival per cent of *S. bicornatum*			
		12 Hrs	24 Hrs	48Hrs	72Hrs
1	IJs+Thiamethoxam 25% WG	83.60 (66.11)	82.50 (65.31)	76.15 (60.92)	72.60 (58.65)
2	IJs+ Diafenthiuron 50% WP	84.70 (66.95)	74.60 (59.77)	73.70 (59.19)	72.55 (58.48)
3	IJs+Imidacloprid 17.8% SL	90.93 (72.44)	88.88 (70.49)	86.15 (68.23)	83.65 (66.27)
4	IJs+Chlorantraniliprole 18.5% SC	85.70 (67.76)	83.45 (66.01)	78.95 (62.70)	65.05 (53.75)
5	IJs+ Indoxacarb 14.5% SC	81.10 (64.42)	81.05 (64.27)	63.90 (53.06)	60.75 (51.20)
6	IJs+ Cypermethrin 25% EC	83.70 (66.24)	49.05 (44.45)	40.80 (39.56)	39.15 (38.64)
7	IJs+ Chlorpyriphos 20% EC	82.20 (65.02)	26.95 (31.19)	17.55 (24.68)	13.75 (21.63)
8	Control (IJs+ Distilled water)	100.00 (90.00)	99.95 (89.35)	99.10 (84.67)	98.30 (82.47)
9	F Test	sig**	sig**	sig**	sig**
10	SE (m) ±	1.01	1.45	1.80	1.78
11	SE(d) ±	1.43	2.05	2.55	2.52
12	CD@ 5%	2.97	4.27	5.29	5.24
13	CV	2.90	4.74	6.37	6.63

(Figures in parentheses are arcsine transformed values; **F test highly significant at 1% level of significance)

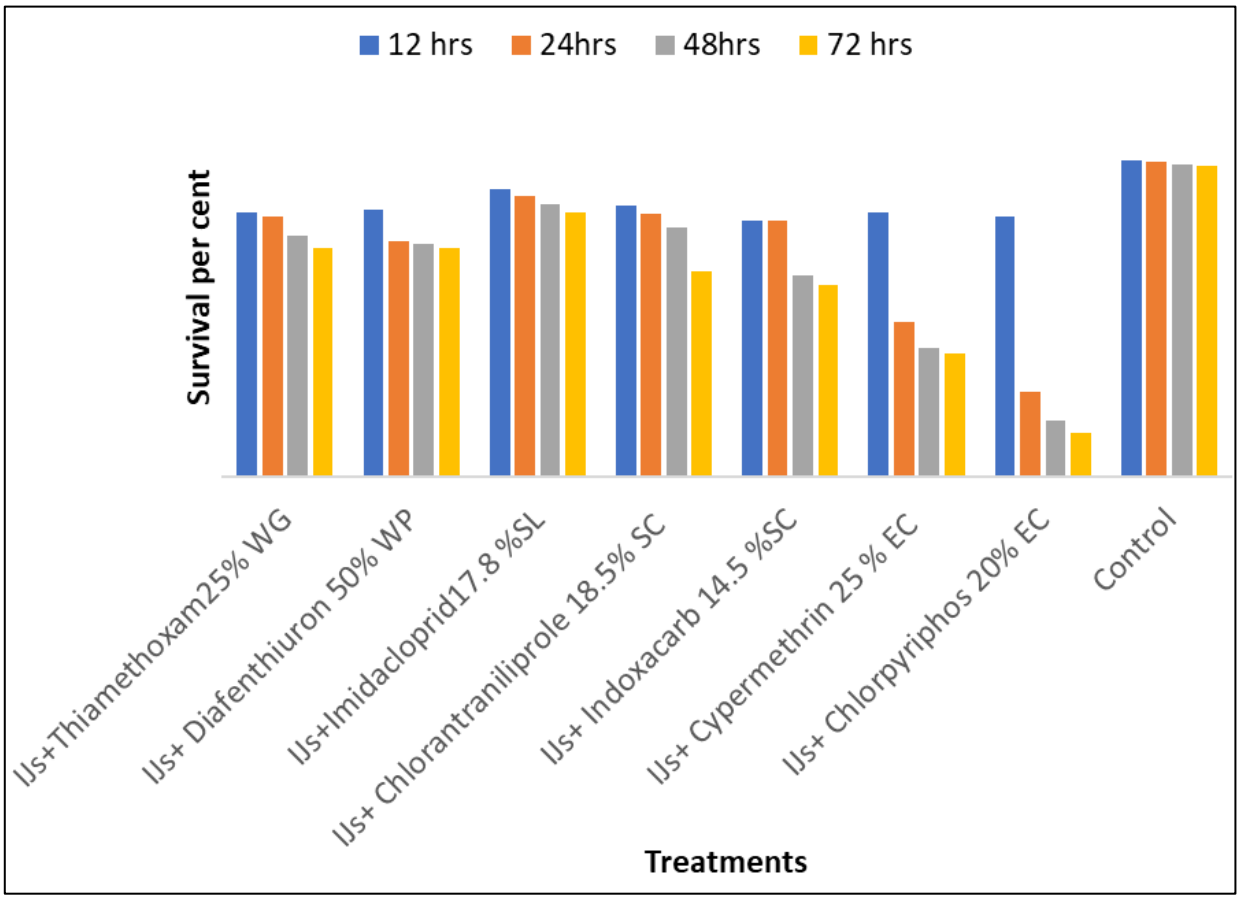

Fig 1: Survival percentage of *S. bicornatum* at 12 hrs, 24 hrs, 48 hrs and 72 hrs exposure to insecticides

Discussion

Similar studies in these regards were made by Anes and Sudershan Ganguly (2011) [2] they studied on the compatibility of entomopathogenic nematode, *Steinernema thermophilum* with four different doses of the twenty-six pesticidal formulations and reported that more than 98% survival of the infective juveniles of *S. thermophilum* was recorded even after 7 days of treatment in all the tested doses of formulations except Hostathion, Dursban, Thiodan which were non compatible. Gitanjali Devi (2019) [9] also reported that entomopathogenic nematodes were relatively resistant to many pesticides in recommended dosage, besides showing synergy between EPNs and chemicals insecticides. Laznik and Trdan (2014) [16] also reported that *S. carpocapsae* and *S. kraussei* are sensitive to all tested insecticides and *Steinernema feltiae* was compatible with imidacloprid. Head Langton (2000) [11] and Cuthbertson *et al.* (2003) [3] have suggested the combined use of pesticide with entomopathogenic nematodes for insect control in integrated pest management.

Conclusion

From the studies conducted it can be concluded that the compatibility of EPN isolate *S. bicornatum* with insecticidal combination namely, Thiamethoxam 25% WG (T_1), Diafenthiuron 50%WP (T_2), Imidacloprid 17.8% SL (T_3), Chlorantraniliprole 18.5% SC (T_4), Indoxacarb 14.5% SC (T_5) were compatible. However, Cypermethrin 25% EC (T_6) and Chlorpyriphos 20% EC (T7) were found to be incompatible.

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