Chapter

Management of *Spodoptera litura* (Fab.) in Green Gram (*Vigna radiata* L.) through Entomo-Pathogenic Nematode

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Abstract

Green gram is most important legume crop and richest source of 24% easily digestible protein. The green gram is attacked by number of insect pests but Spodoptera litura is more serious pest. The uses of entomopathogenic nematodes (EPN) as a biological control agent of insect pests are more effective. EPNs have been found effective for the management of tobacco caterpillar and are used as bio insecticides against a number of lepidopteran pests. The mass multiplication of *Steinernema carpocapsae* can be done on rice moth (*Corcyra cephalonica*), greater wax moth (Galleria mellonella), gram pod borer (Helicoverpa armigera) and tobacco caterpillar (Spodoptera litura). Infectivity of entomopathogenic nematode, S. carpocapsae against tobacco caterpillar was studied and observation was recorded after every day up to 10 days with different inoculum levels *viz.*, 10,000, 15,000 and 20,000 IJs/plant of S. carpocapsae. The experimental results revealed that maximum 82.50% mortality of S. litura was observed at inoculum level 20,000 IJs/ plant of S. carpocapsae after 9th day of inoculation followed by 75.00% mortality at inoculum level 15,000 IJs/plant. While, minimum 67.50% mortality was recorded at inoculum level 10,000 IJs/plant. Therefore, it was concluded that the mortality of insect larvae increased with an increase in the inoculum levels and period of exposure.

Keywords: green gram, infectivity, mass multiplication, *Spodoptera litura*, *Steinernema carpocapsae*

1. Introduction

Green gram (*Vigna radiata*) also known as mung bean, is native to India and Central Asia. The food legumes were grown by farmers since millennia providing nutritionally balanced food to the people of India [1] and many other countries in the world. Pulses occupy a unique position in economy of our nation being the major source of proteins. The major pulse crops that have been domesticated and are under cultivation include, green gram, black gram, chickpea, cowpea, pigeon pea, horse gram, lentil, moth bean, and pea.

Green gram is an important source of easily digestible high quality protein for vegetarians. It contains 24% protein, 0.326% phosphorus, 0.0073% iron, 0.00039%

carotene, 0.0021% of niacin [2]. Researchers has pointed out that plant protection remains a most neglected aspect in pulse cultivation; further stating that only 5–6% of the growers adopt plant protection measures in only 1.5% of the total area under this crop. The green gram is attacked by number of insect pests *viz*. *Helicoverpa armigera*, *Spodoptera litura*, *Maruca vitrata*, *Etiella zinckenella*, *Mylabris phalerata*. They cause significant damage to green gram including foliage and pods. The losses caused to green gram come to about 20%.

Spodoptera litura (Lepidoptera: Noctuidae) is a serious polyphagous pest of several cultivated crops and has attained global importance. The losses caused by *S. litura* on mung bean is much more severe as this pest has been reported to cause skelatalization of leaves in early stage and severe defoliation in later stage thus reducing the photosynthetic capacity of plants. Tobacco caterpillar (*Spodoptera litura*) has a wide host range of more than 120 host plants including crops (green gram, tobacco, soybean, castor, maize, sorghum, groundnut, linseed and mustard), vegetables (tomato, okra, brinjal and cucurbits) weeds and ornamental plants and the losses caused to these crops may range from 20 to 30% [3]. The caterpillars may eat entire leaves, and even flowers and fruits. The caterpillar burrows into the soil several centimeters and pupates without a cocoon. The pupal stage lasts either a few weeks or several months, depending upon time of year. The average life cycle is completed in about 25 days.

Realizing the role of these pests as limiting factor in agricultural productivity, several methods were developed and incorporated in to management program of the economically important pest. Out of these, use of insecticides could initially catch up to the growers because of their ready availability, ability to suppress pest's populations quickly and increasing productivity. Widespread development of resistance to chemical insecticides including the widely used pyrethroids has been reported in *S. litura* [4]. In addition to the development of resistance in pests, indiscriminate use of pesticides has grossly poisoned almost each and every component of the biosphere, including resurgence of pests and reduction of natural enemies in agro ecosystems, allowing rapid rebound of target and minor pests.

Use of insecticides although found effective however, looking into the adverse effect of chemical insecticides, several bioagents have been tried time to time to manage this pest but none of them could give desirable results.

Biological control of pests using entomopathogenic nematodes (EPNs) may prove to be an ideal alternative to other bioagent earlier used they have long term effect, without any harmful effect on non-target organisms. EPNs are potential agents as they serve as vectors of bacteria, achieve a quick kill of target insect pests, have broader host range, highly virulent, possess chemoreceptor's and can be cultured easily *in vitro* and *vivo*. EPNs can be easily applied using standard application equipment and are compatible with many chemical pesticides. The EPNs of the families *Steinernematidae* and *Heterorhabditidae* are potentially useful for biological control in agriculture systems [5]. The infective juveniles (IJs) of these families are free living, non-feeding and have the ability to search out their hosts. They have the potential for long-term establishment in soil through recycling of infected insects larvae. The importance of entomopathogenic nematode as a key component for the management of pests.

2. Mass multiplication of Steinernema carpocapsae on different hosts

Mass multiplication of *Steinernema carpocapsae* was done on rice moth (*Corcyra cephalonica*), greater wax moth (*Galleria mellonella*), gram pod borer (*Helicoverpa*

armigera) and tobacco caterpillar (*Spodoptera litura*). The infective juveniles of *S. carpocapsae* were released @ 100 IJs/larvae into the petri plate having 4th instar larvae of different insect hosts allowing them to enter into the insect body. Harvesting of EPN's population was done after 10 days of inoculation using white trap method up to 5 days.

Results have indicated that, on the basis of per mg. body weight of cadaver maximum 572.00 IJs of *S. carpocapsae* were produced on *G. mellonella*, followed by 568.00 IJs and 554.00 IJs on *S. litura* and *H. armigera* respectively. Whereas, minimum 542.00 IJs on *C. cephalonica*. Therefore, *G. mellonella* was the most suitable host for mass production of *S. carpocapsae* (**Table 1**).

2.1 Rice moth (Corcyra cephalonica)

The data on yield of IJs presented in **Table 2** showed that maximum 60212.0 IJs of *S. carpocapsae* were produced on large sized larvae (14–16 mm) with mean body weight of 134 mg/larvae followed by 48320.0 IJs from medium sized larvae (10–12 mm) and 39635.0 IJs from small sized larvae (6–8 mm).

2.2 Greater wax moth (Galleria mellonella)

The data on yield of IJs presented in **Table 1** showed that maximum 100240.0 IJs of *S. carpocapsae* were produced on large sized larvae (18–20 mm) with mean body weight of 202 mg/larvae followed by 66036.0 IJs from medium sized larvae (13–15 mm) and 49252.0 IJs from small sized larvae (10–12 mm).

S. no.	Size of larvae (mm)	Mean weight (mg/larvae)	IJs harvested/larvae	IJs/mg body weight of cadaver
1.	Small (10–12)	86	49252.00	572.75
2.	Medium (13–15)	131	66036.00	504.25
3.	Large (18–20)	202	100240.00	496.25
	SEm±	3.543	662.457	8.504
	CD (5%)	11.334	2119.316	27.205
	CV (%)	5.07	1.84	3.24
noculum level	= 100 IJs/larvae, replication	= 4 times.		

Table 1.

Yield of Steinernema carpocapsae from the larvae of greater wax moth (Galleria mellonella).

S. no.	Size of larvae (mm)	Mean weight (mg/larvae)	IJs harvested/ larvae	IJs/mg body weight of cadaver
1.	Small (6–8)	73	39635.00	542.94
2.	Medium (10–12)	90	48320.00	538.50
3.	Large (14–16)	134	60212.00	449.50
	SEm±	2.465	1163.214	4.976
	CD (5%)	7.886	3721.324	15.919
·	CV (%)	4.98	4.72	1.95

Inoculum level = 100 IJs/larvae, replication = 4 times.

 Table 2.

 Yield of Steinernema carpocapsae from the larvae of rice moth (Corcyra cephalonica).

2.3 Gram pod borer (Helicoverpa armigera)

The data on yield of IJs presented in **Table 3** showed that maximum 115362.0 IJs of *S. carpocapsae* were produced on large sized larvae (30–32 mm) with mean body weight of 274 mg/larvae followed by 106070.0 IJs from medium sized larvae (25–27 mm) and 113590.0 IJs from small sized larvae (20–22 mm) (**Figures 1** and **2**).

S. no.	Size of larvae (mm)	Mean weight (mg/larvae)	IJs harvested/ larvae	IJs/mg body weight of cadaver
1.	Small (20–22)	205	113590.00	554.25
2.	Medium (25–27)	236	106070.00	449.25
3.	Large (30–32)	274	115362.00	421.00
	SEm±	3.976	1795.069	7.535
	CD (5%)	12.719	5742.737	24.105
	CV (%)	3.34	3.21	3.17

Inoculum level = 100 IJs/larvae, replication = 4 times.

Table 3.

Yield of Steinernema carpocapsae from the larvae of gram pod borer (Helicoverpa armigera).

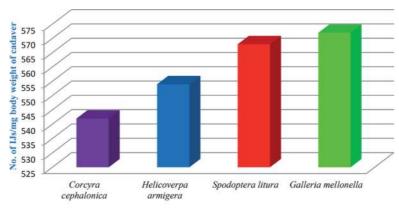




Figure 1. Mass multiplication of Steinernema carpocapsae on different hosts.

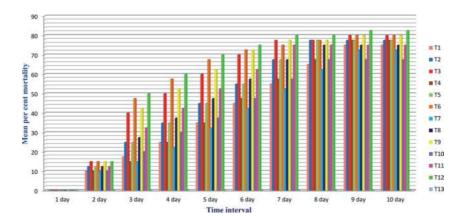


Figure 2.

Infectivity of Steinernema carpocapsae recovered from different hosts (a) Corcyra cephalonica, (b) Galleria mellonella, (c) Helicoverpa armigera and (d) Spodoptera litura against Spodoptera litura infecting green gram.

2.4 Tobacco caterpillar (Spodoptera litura)

The data on yield of IJs presented in **Table 4** showed that maximum 201280.0 IJs of *S. carpocapsae* were produced on large sized larvae (26–28 mm) with mean body weight of 430 mg/larvae followed by 200900.0 IJs from medium sized larvae (22–24 mm) and 193140.0 IJs from small sized larvae (18–20 mm) (**Figures 3** and **4**).

S. no.	Size of larvae (mm)	Mean weight (mg/larvae)	IJs harvested/ larvae	IJs/mg body weight of cadaver
1.	Small (18–20)	340	193140.00	568.00
2.	Medium (22–24)	400	200900.00	502.00
3.	Large (26–28)	430	201280.00	468.25
	SEm±	8.808	4406.542	11.159
	CD (5%)	28.179	14097.29	35.698
	CV (%)	4.52	4.44	4.35

Table 4.

Yield of Steinernema carpocapsae from the larvae of tobacco caterpillar (Spodoptera litura).

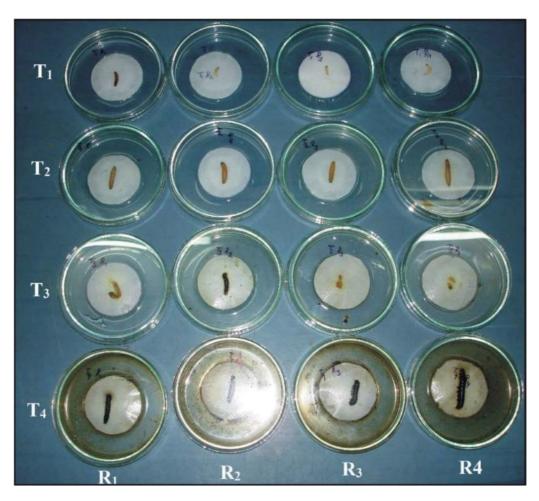


Figure 3. Mass multiplication of Steinernema carpocapsae on different hosts.

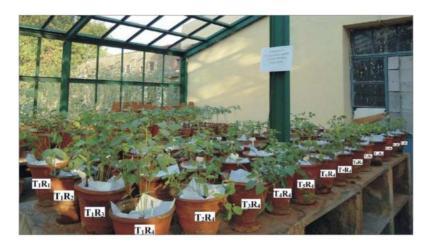




Figure 4.

Infectivity of Steinernema carpocapsae recovered from different hosts against Spodoptera litura infecting green gram.

3. Infectivity of *Steinernema carpocapsae* recovered from different hosts against *Spodoptera litura* infecting green gram

Experiment was conducted to find out the infectivity of *S. carpocapsae* recovered from different natural hosts *viz*. *Corcyra cephalonica*, *Galleria mellonella*, *Helicoverpa armigera* and *Spodoptera litura* at different inoculum levels 10,000, 15,000 and 20,000. The mean percent mortality was recorded after every day up to 10 days.

3.1 After 1st day

The experimental results presented in **Table 5** revealed that there was no mortality of insect larvae, by inoculating IJs recovered from natural hosts *viz*. *C. cephalonica*, *G. mellonella*, *H. armigera* and *S. litura*.

3.2 After 2nd day

Results showed that 15.00, 12.50 and 10.00% mortality of *S. litura* was achieved at inoculum levels 20,000, 15,000 and 10,000 IJs/plant respectively, with populations recovered from *C. cephalonica*, *G. mellonella*, *H. armigera* and *S. litura*.

5	I reauments				Me	Mean percent mortality at different intervals	ty at different	intervals			
ю.		1 day	2 day	3 day	4 day	5 day	6 day	7 day	8 day	9 day	10 day
1.	$S_1 D_1$	0.00	10.00 (18.43)	17.50 (24.16)	25.00 (29.89)	35.00 (36.22)	45.00 (42.12)	55.00 (47.88)	65.00 (53.78)	75.00 (60.11)	75.00 (60.11)
5	$S_1 D_2$	0.00	12.50 (20.47)	25.00 (29.89)	35.00 (36.22)	45.00 (42.12)	55.00 (47.88)	67.50 (55.28)	77.50 (61.77)	77.50 (61.77)	77.50 (61.77)
	$S_1 D_3$	0.00	15.00 (22.50)	40.00 (39.17)	50.00 (45.00)	60.00 (50.83)	70.00 (56.95)	77.50 (61.77)	77.50 (61.77)	80.00 (63.43)	80.00 (63.43)
4.	$S_2 D_1$	0.00	10.00 (18.43)	15.00 (22.50)	25.00 (29.89)	35.00 (36.12)	47.50 (43.56)	57.50 (49.33)	67.50 (55.28)	77.50 (61.77)	77.50 (61.77)
Ŀ.	$S_2 D_2$	0.00	12.50 (20.47)	25.00 (29.89)	35.00 (36.22)	45.00 (42.12)	55.00 (47.88)	67.50 (55.28)	77.50 (61.77)	77.50 (61.77)	77.50 (61.77)
6.	$S_2 D_3$	0.00	15.00 (22.50)	47.50 (43.56)	57.50 (49.33)	67.50 (55.28)	72.50 (58.45)	75.00 (60.11)	77.50 (61.77)	80.00 (63.43)	80.00 (63.43)
7.	$S_3 D_1$	0.00	10.00 (18.43)	15.00 (22.50)	22.50 (28.23)	32.50 (34.72)	42.50 (40.67)	52.50 (46.44)	62.50 (52.27)	72.50 (58.45)	72.50 (58.45)
×.	$S_3 D_2$	0.00	12.50 (20.47)	27.50 (31.39)	37.50 (37.66)	47.50 (43.56)	57.50 (49.39)	67.50 (55.28)	75.00 (60.11)	75.00 (60.11)	75.00 (60.11)
6	$S_3 D_3$	0.00	15.00 (22.50)	42.50 (40.61)	52.50 (46.44)	62.50 (52.34)	72.50 (58.61)	77.50 (61.77)	77.50 (61.77)	80.00 (63.43)	80.00 (63.43)
10.	$S_4 D_1$	0.00	10.00 (18.43)	20.00 (26.19)	30.00 (33.05)	37.50 (37.73)	47.50 (43.56)	57.50 (49.33)	67.50 (55.28)	67.50 (55.28)	67.50 (55.28)
11.	$S_4 D_2$	0.00	12.50 (20.47)	32.50 (37.42)	42.50 (40.67)	52.50 (46.44)	62.50 (52.27)	75.00 (60.11)	75.00 (60.11)	75.00 (60.11)	75.00 (60.11)
12.	S4 D3	0.00	15.00 (22.50)	50.00 (45.00)	60.00 (50.77)	70.00 (56.79)	75.00 (60.11)	80.00 (63.43)	80.00 (63.43)	82.50 (65.47)	82.50 (65.47)
13.	Control	0.00	0.00	0.00	0.00	0.00	00.00	0.00	0.00	0.00	0.00

s.	Treatments				Me	an percent morta	Mean percent mortality at different intervals	tervals			
no.		1 day 2 day	2 day	3 day	4 day	5 day	6 day	7 day	8 day	9 day	10 day
	SEm ±	0.00	1.703	2.217	1.817	1.707	1.860	1.443	1.486	1.445	1.445
	CD (5%)	0.00	4.872	6.342	5.197	4.882	5.322	4.128	4.251	4.134	4.134
	CV (%)	0.00	18.03	14.80	10.19	8.30	8.04	5.63	5.45	5.11	5.11
Replication = 4 times, EPN population prod	Replication = 4 times, (10 larvae/pot). EPN population produced on different host: S_1 = Corcyra cephalonica; S_2 = Galleria mellonella; S_3 = Helicoverpa armigera; S_4 = Spodoptera litura. Doses: D_1 = 10,000 Js/plant; D_2 = 15,000 Js/plant;	ae/pot). different host:	S ₁ = Covcyra cep	$halonica; S_2 = Gal$	leria mellonella; S	S ₃ = Helicoverpa a	<i>"migera;</i> S ₄ = Spoc	loptera litura. Do	ses: D ₁ = 10,000) IJs/plant; $D_2 =$	15,000 IJs/pl

 $D_3 = 20,000$ IJs/plant. Ē

 Table 5.

 Infectivity of Steinernema carpocapsae recovered from different hosts against Spodoptera litura infecting green gram.

3.3 After 3rd day

Data pertaining to mean percent mortality of *S. litura* presented in **Table 5** revealed that maximum 50.00% mortality of *S. litura* was observed at an inoculums level of 20,000 IJs/plant of *S. carpocapsae* obtained from *S. litura*, followed by 47.50% mortality at 20,000 IJs/plant produced on *G. mellonella*. Whereas, mini-mum 15.00% mortality at 10,000 IJs/plant recovered from both *G. mellonella*, and *H. armigera*.

3.4 After 4th day

Results showed in **Table 5** revealed that maximum 60.00% mortality of *S. litura* was recorded at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* recovered from *S. litura*, followed by 57.50% mortality at 20,000 IJs/plant produced on *G. mellonella*. Whereas, minimum 22.50% mortality at 10,000 IJs/plant obtained from *H. armigera*.

3.5 After 5th day

Maximum 70.00% mortality of *S. litura* was recorded at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* obtained from *S. litura*, followed by 67.50% mortality at 20,000 IJs/plant produced on *G. mellonella*, whereas, minimum 32.50% mortality at 10,000 IJs/plant recovered from *H. armigera*.

3.6 After 6th day

Data pertaining to mean percent mortality of *S. litura* presented in **Table 5** revealed that maximum 75.00% mortality of *S. litura* was recorded at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* recovered from *S. litura*, followed by 72.50% mortality at 20,000 IJs/plant produced on *G. mellonella* as well as *H. armigera*. While, minimum 42.50% mortality at 10,000 IJs/plant obtained from *H. armigera*.

3.7 After 7th day

Maximum 80.00% mortality of *S. litura* was observed at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* obtained from *S. litura*, followed by 77.50% mortality recorded at 20,000 IJs/plant produced on *C. cephalonica* and *H. armigera*, while minimum 52.50% mortality at 10,000 IJs/plant recovered from *H. armigera*.

3.8 After 8th day

Results showed in **Table 5** revealed that maximum 80.00% mortality of *S. litura* recorded at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* obtained from *S. litura*, followed by 77.50% mortality at 20,000 IJs/plant produced on *H. armigera*, *C. cephalonica* and *G. mellonella* and at 15,000 IJs/plant recovered from *C. cephalonica* and *G. mellonella*. Whereas, minimum 62.50% mortality at 10,000 IJs/plant recovered from *H. armigera*.

3.9 After 9th day

Data pertaining to mean percent mortality of *S. litura* presented in **Table 5** revealed that maximum 82.50% mortality of *S. litura* was recorded at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* obtained from *S. litura*, followed by

80.00% mortality at 20,000 IJs/plant recovered from *C. cephalonica*, *G. mellonella* and *H. armigera*. Whereas, minimum 72.50% mortality recorded at 10,000 IJs/plant recovered from *H. armigera*.

3.10 After 10th day

Maximum 82.50% mortality of *S. litura* was recorded at an inoculum level of 20,000 IJs/plant of *S. carpocapsae* recovered from *S. litura*, followed by 80.00% mortality was recorded at 20,000 IJs/plant produced on *C. cephalonica*, *G. mellonella* and *H. armigera*. Whereas, minimum 72.50% mortality at 10,000 IJs/plant obtained from *H. armigera*.

4. Conclusion

EPNs are excellent biocontrol agents for insect pests. When an EPN is used against a pest insect, it is critical to match the right nematode species against the target pest. Biotic agents including nematode pathogens, predators and other soil organisms, as well as abiotic factors such as ultraviolet radiation, soil moisture/relative humidity, temperature, etc., can affect EPN application efficacy. Recently, improvement of nematode formulation, application equipment or approaches, and strain improvement have been made to enhance EPN application efficacy. Additional research toward lowering product costs, increasing product availability, enhancing ease-ofuse, and improving efficacy and carryover effect will stimulate the extensive use of EPNs in biocontrol. With these advances EPNs will serve to reduce chemical insecticide inputs and contribute to the stabilization of crop yields and the environment.

In this chapter, we studied about the effect of host on multiplication and temperature on infectivity of *S. carpocapsae* against *S. litura* on green gram. Studies on mass multiplication of *Steinernema carpocapsae* was done on rice moth (*Corcyra cephalonica*), greater wax moth (*Galleria mellonella*), gram pod borer (*Helicoverpa armigera*) and tobacco caterpillar (*Spodoptera litura*). Results have indicated that on the basis of per mg body weight of cadaver maximum, *S. carpocapsae* was obtained from *G. mellonella*, followed *S. litura* and *H. armigera* respectively, whereas minimum IJs recovered from *C. cephalonica*. Therefore, it was concluded that on the basis of per mg body weight of cadaver *G. mellonella* was the most suitable host for mass production of *S. carpocapsae*.

When we studied about infectivity of *S. carpocapsae* against tobacco caterpillar (*S. litura*) under pot condition on green gram with different inoculum levels with different population of *S. carpocapsae* produce on natural hosts, the experimental results revealed that maximum percent mortality of *S. litura* was observed at 20000 IJs of *S. carpocapsae* recovered from *S. litura* after 9th days followed by 20,000 IJs recovered from *C. cephalonica*, *G. mellonella* and *H. armigera*. While, minimum percent mortality was recorded at 10,000 IJs recovered from *H. armigera*.

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