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Pathogenicity of entomopathogenic nematode isolate Steinernema *bicornatum* against red cotton bug *Dysdercus cingulatus*

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Abstract

Entomopathogenic nematodes (EPNs) of the genera Steinernema *bicornatum* are obligate and lethal insect parasites. In last decade they were widely used as biological control agents for pest insects of commercial crops, therefore research in this area is directly linked to agriculture. In this study, the pathogenicity of nematode species (*Steinernema bicornatum*) was tested against red cotton bug. The pathogenicity of these isolates was evaluated on the red cotton bug (Hemiptera: Pyrrhocoridae) at concentrations (50 IJs/100 μ l per nymph) in the laboratory at room temperature. Maximum mortality rate (95%) was achieved from the *Steinernema bicornatum* isolate at the lowest concentration after 48 h of post inoculation. While these isolates showed high pathogenicity on *G. mellonella* last instar ranging from 63 to 100% at the concentration of 50 IJs/100 μ l, 48 h post-inoculation. After 3 days of nymphal mortality, population recovery from treated red cotton bug nymph was upto 22 IJs/100 μ l *Steinernema bicornatum* suspension, whereas population count was obtained around 150-200 IJs/100 μ l from laboratory host *Galleria mellonella*. The results showed that recovery of nematode population very low in red cotton bug as compare to Greater wax moth.

Keywords: Pathogenicity, entomopathogenic nematodes, Steinernema bicornatum, Red cotton bug

Introduction

The red cotton bug, *Dysdercus cingulatus* belonging to the family Pyrrhocoridae of the order Hemiptera is an important pest of cotton. Red cotton bug is one of the important insects found damaging cotton crop in various states *viz.*, Gujarat, Maharashtra, Uttar Pradesh, Bihar, Madhya Pradesh and Tamil Nadu. The red cotton bug is called by various vernacular names in different parts of India such as Chainpa in Punjab, Behna in Kanpur, Kappa poka in Orissa, Lal chingum in Uttar Pradesh and Lal chusiya in Gujarat. The nymphs and adults suck up the juice from the seed of either green or ripe cotton bolls. Both are voracious feeder and cause extensive damage to the crop. The damaged bolls remain shrivelled and vitality of plant and the quality of the lint is also severally affected in case of heavy infestation. The insect remained very active during October to March. Entomopathogenic nematodes in the genera *Steinernema* and *Heterorhabditis* show strong pathogenicity to a wide range of insects. Infective juveniles (IJs) seek insect hosts in soil, and invade them through their natural openings or through the skin. After invasion, IJs release their symbiotic bacteria, which produce insect toxins, into the hemocoel of the host there by rapidly killing the host (Dowds and Peters, 2002)^[1].

Methods and Materials

Nematodes

Steinernema bicornatum were maintained on post-graduation laboratory host Greater wax moth and used for the experiments.

Test Insect

Collection of red cotton bug from cotton field. Laboratory Assessments The following experiments were designed and conducted with the objective of assessing the pathogenicity of entomopathogenic nematode isolates *Steinernema bicornatum* against red cotton bug in the laboratory.

Dose Response Experiments

Dose response assessment is very efficient in the determination of the pathogenic ability of nematode strains against specific hosts. The purpose of this experiment was to obtain the mean number of IJs penetrated and established inside the infected insect hosts (Fan and Hominick, 1991; Westerman and Stapel, 1992) ^[2, 3]. The experiment was performed on small plastic plates of 4cm diameter. The entomopathogenic nematodes Steinernema bicornatum isolate, one treatment dose of infective juveniles (IJs) (50) was used. The 20 number Red cotton bug were introduced each plastic plate after addition of the nematode suspensions containing specific doses. Treatment was replicated five times. All plates were kept in the dark inside at room temperature. Assessment of the results was performed by counting the number of infective juveniles which penetrated the red cotton bug at dose.

Reproduction Capability inside the Red cotton bug

The reproductive capability of an entomopathogenic nematode isolate *Steinernema bicornatum* refers to its ability to reproduce inside insect hosts. This assay was found to be efficient in assessing the infectivity of nematode strains against insect pests (Elawad *et al.*, 2001). Treatment was replicated five times. Three days after infection with nematode strains, dead red cotton bug were transferred individually to White Traps (White, 1927) ^[4]. Emerging

juveniles were collected and counted over a 20 days period.

Results and Discussion

Dose Response Experiments

Nematode isolates assessed by dose response have shown mortality rates to the red cotton bug. The nematode isolate *Steinernema bicornatum* showed high mortality against the red cotton bug. Entomopathogenic nematodes (EPNs) have been described from 23 nematode families. Out of these Steinernematidae and Heterorhabditidae have received the most attention because they possess many of the attributes of effective biological control agents (Kaya and Gaugler, 1993; Grewal *et al.*, 2005) ^[5, 6] and have been utilized as classical, conservational and augmentative biological control agents

Reproduction Capability inside the red cotton bug

A significant effect in the number of IJs produced from nematode strain. The number of nematode infective juveniles was recorded which indicates a powerful infectivity against the adults of the red cotton bug while the least number of infective juveniles was produced. Emerging IJs were counted every day (Fig. 2) but Table. 2 contains combined data up to 20 days. An infected nymph produced a total of about 11,000,12,500,10,000,9,000,11,000 IJs per nymph of red cotton bug, respectively. Overall average of IJs per replication was about (10700).

Table 1: Percent mortality of red cotton bug (nymph) after 72Hrs

Sr. No.	Total No. of treated insects	No. of IJs/nymph	No of Dead Insects	Percent mortality
1	20	50IJs/nymph	20	100%
2	20	50IJs/nymph	20	100%
3	20	50IJs/nymph	18	90%
4	20	50IJs/nymph	19	95%
5	20	50IJs/nymph	18	90%
6	20	Control (Water)	4	20%

Table 2: Population count Entomopathogenic nematode from insects

Sr. no.	Replication	Population count/100 μl	Total count in one week
1	\mathbf{R}_1	22	11,000
2	R ₂	25	12,500
3	R ₃	20	10,000
4	R 4	18	9,000
5	R 5	22	11,000
6	Control	00	00

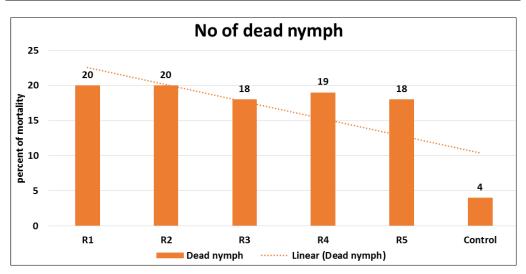


Fig 1: Percent mortality of red cotton bug (nymph)

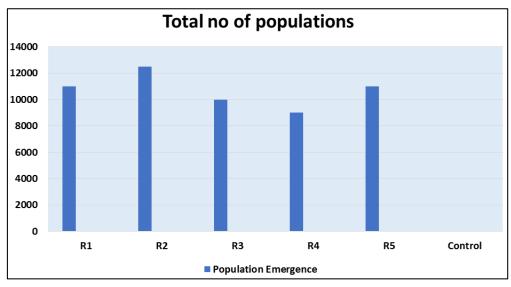


Fig 2: Reproduction potential in red cotton bug

Conclusion

All results of this research have clearly indicated that the entomopathogenic nematode *Steinernema bicornatum* possess great pathogenic capability to infect and kill red cotton Bug. The recovery of entomopathogenic nematodes very low in red cotton bug as compare to Greater wax moth. We strongly recommend the adoption of applications of targeted sprays of nematode suspensions during the peak period of infestation of RCBs as an effective strategy in combating the red cotton bug in Cotton fields.

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