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EFFECT OF INSECT PATHOGENIC FUNGI *ZOOPHTHORA RADICANS* ON THE GROWTH OF LARVAE AND PUPA OF *CNAPHALOCROCIS MEDINALIS*

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Abstract

The pupation percentage, larval length, larval duration, larval weight, pupal weight of *Cnaphalocrocis medinalis* was significantly reduced with different fungal concentrations of *Zoophthora radicans.* Least pupation percentage was noticed in positive control treatment of cypermethri n followed by neem plus when compared to fungal treatments. Malformed pupal stages were observed in the fungal treatments. All the cypermethrin treated pupae were malformed and healthy adult emergence percentage from pupae was very high in untreated and no adults emergence was observed in cypermethrin treatment.

***Key words*:** *Zoophthora radicans,* fungus*, Cnaphalocrocis medinalis*, growth

Introduction

Rice leaf folder, *Cnaphalocrocis medinalis* (Guenee) is one of the most important insect pests in Indian subcontinent (Gunathilagaraj and Gopalan, 1986). This pest has been reported to attain the major pest status in important paddy growing areas (Murugesan and Chelliah, 1987). Bhanu and Reddy (2008) reported that the leaf folder affected the crop adversely under favourable conditions causing 60% loss in India. The synthetic chemical pesticides have been the widely used approach to reduce the estimated 45% gross crop loss due to pests and diseases that amounted to 290 billion rupees per annum. More and more quantities of synthetic pesticides are being used for agricultural intensification to feed on ever growing population. Due to ill effects of insecticides, it becomes inevitable to find alternate, viable and effective biopesticides. Among the other bio control agents, fungal pathogens are increasingly found potential against many crop pests (Dhaliwal, 2008). An entomophthoralean fungus, *Z. radicans* has been reported from India infecting naturally leaf folder larvae (Parthasarathy, 1997). However, the research on *Z. radicans* against rice leaf folder was scanty in India and present studies were conducted to know the effect of this fungal pathogen against larvae and pupa of rice leaf folder, *Cnaphalocrocis medinalis*

Materials and Methods

Collection and Rearing of Pest

The larvae of *C. medinalis* were collected from infested rice plants from Annamalainagar in Cuddalore district. The larvae collected were maintained in the laboratory at 22 ± 2°C and 70 –75 % relative humidity. The larvae were reared on potted rice plants.

Bioassay

Spore suspension was prepared from 15 days old culture of *Z. radicans* on SMA medium. The fungal surface was scraped using a sterile loop with 10 ml of sterile distilled water having 0.02% Tween 80 as a wetting agent (Rombach *et al.,* 1986). The suspension was then filtered through sterile muslin cloth to eliminate the medium (Sasidharan and Varma, 2005). Spore concentration of the filtrate was determined using a Neubauer's haemocytometer. This served as the stock suspension. Different spore concentration was prepared by adding sterile 0.02% Tween 80 in distilled water. Spore suspension of *Z. radicans* at four different concentrations, 2.4×10^7, 2.4×10^6, 2.4×10^5 and 2.4×10^4 spores/ml was prepared and tested for its efficacy on third instar larvae, pupae and adults of *Z. radicans.*

Growth Inhibition of Larvae

For bioassay, spraying method was adopted. Nine ml of different spore concentrations of *Z. radicans* was sprayed against rice leaf folder larvae. Ten larvae were used per replication. The larvae were treated with sterile distilled water and 0.006 % (v/v) of neem product and cypermethrin. These three served as positive control. After treatment, the larvae were allowed to feed on rice leaves. Each treatment was replicated thrice. Growth parameters namely larval duration (days), larval length, larval weight and pupation (%) were recorded (Hafez *et al.,* 1994).

Growth inhibition of pupae

Four different spore concentrations of *Z. radicans* with three replications each were used for infecting the pupa of *C. medinalis.* The pupae were sprayed with 10 ml of respective fungal spore suspensions using hand atomizer. The pupae were treated with sterile distilled water and 0.006 % (v/v) neem product and

cypermethrin. These three served as positive control. The growth of surviving pupa was recorded up to adult emergence for the parameters, such as pupal duration (days), pupal weight (mg), pupal length (cm) and adult emergence (%) (Hafez *et al.,*1994). All the treatments were replicated four times and the experiment was conducted in CRBD.

Results and Discussion

Larval Growth

Among the different fungal concentrations, the least pupation was noticed in 2.4 × 10^7 and 2.4 ×10^4 (43.33 %) as against 100 % in untreated. The variation between different treatments was significant. The larval mortality was observed after 3 -7 days of fungal treatment. The *C. medinalis* length ranged from 2.70–3.90 cm in the different treatments. The least larval length (2.70 cm) was recorded in 2.4 × 10^5 spore concentration as against the highest in the untreated (3.90 cm). Similarly, among the fungal treatments, the larval weight was least (357.00 mg) in 2.4 × 10^5 treatment compared to untreated 396.20 mg. The larval duration was 7.30 days in 2.4 × 10^6 as against the untreated (7.90 days)

Pupal Growth

The pupal weight ranged from 163.50 to 205.10 mg. Among the cypermethrin and neem plus tested, Neemplus recorded least pupal weight (163.50 mg) as against the untreated recording 205.10 mg. Among the different fungal treatments least pupal weight (184.10 mg) was recorded in 2.4 x 10^5 spore concentration/ml as against the untreated (205.10 mg). In general there was not much variation in the pupal length. It was least (1.30 cm) in 2.4 x 10^5, 2.4 x 10^6 and 2.4 x 10^7 spore concentrations which was on par with cypermethrin (1.30 cm). Malformed pupal stages were observed in the fungal treatments. All the cypermethrin treated pupae are malformed when compared to the untreated (20.00%). Among the fungal concentrations, 2.4 ×10^4 spores/ml caused more malformed pupae (86.60%). Healthy adult emergence percentage from pupae was very high (80.00%) in untreated and no adults emergence was observed in cypermethrin treatment. Among the fungal concentrations, more adult's emergence was observed in 2.4 x 10^5 spores / ml. In the bioassay studies, the larval mortality was observed 3-7 days after the fungal treatment. With entomophthoralean fungi, unicellular yeast-like cells with chitinous walls (hyphal bodies) spread throughout the insect obtaining nutrients, leading to the death of the host by physiological starvation about 3-7 days after infection (Pell, 1993).

In general there was not much variation in the pupal length of *C. medinalis.* It was least (1.3 cm) in 2.4 x 10^5, 2.4 x 10^6 and 2.4 x 10^7 spore concentrations which was on par with cypermethrin (1.3 cm). This suppressive effect may be due to the inhibitory action on mitochondrial respiration by affecting the NADH-Cytochrome C-reductase and complex-I of insect mitochondria (Londershausen *et al.,* 1991).

The pupal duration was 8.0 days in 2.4 × 10^5 as against the untreated (8.5 days). In contrast, the pupal duration was prolonged in *B. bassiana* treated pupae of *Phthorimaea operculella* compared with the control (Hafez *et al.,* 1994).

Table1 : Effect of *Zoophthora radicans* on the growth of larvae of *Cnaphalocrocis medinalis*

Treatments		*C.medinalis* Larval growth parameters (Third instar)			
		Larval length (cm)	**Larval weight (mg)**	**Larval duration (days)**	**Pupation percentage**
Control	Untreated	3.90	396.00	7.90	100.00 (90.00)^a
	Cypermethrin (0.006 v/v)	3.00	340.50	7.30	36.70 (37.22)^g
	Neemplus (0.006v/v)	3.10	346.20	7.80	40.00 (39.19)^f
Z.radicans (Spores/ml)	2.4×10^4	2.80	361.20	7.60	43.30 (41.14)^{cd}
	2.4×10^5	2.70	357.00	7.70	46.70 (42.84)^c
	2.4×10^6	2.90	366.90	7.30	50.00 (44.99)^b
	2.4×10^7	3.00	367.50	7.50	43.30 (41.14)^{ce}
CD($p_{=0.05}$)		0.29	3.16	0.17	1.74
SE		0.10	1.11	0.05	0.64

Each value is a mean of four replications

Figures in parenthesis are arcsin transformed

In a column means followed by a common letter are not significantly different (P=0.05) by DMRT

Table 2: Effect of *Zoophthora radicans* on the growth of pupae of *Cnaphalocrocis medinalis*

Treatments		Pupal Growth Parameters			Adults Emergence Percentage	
		Pupal Weight (mg)	Pupal Length (cm)	Pupal Duration (Days)	Healthy	Malformed or dead pupa
Control	Untreated	205.1	1.5	8.5	80.0 (62.89)[a]	20.0 (26.55)[g]
	Cypermethrin	167.6	1.3	0.0	0.0 (0.0)[g]	100.0 (90.00)[a]
	Neemplus	163.5	1.4	9.0	11.1 (19.26)[f]	88.89 (70.05)[b]
Z.radicans (Spores/ml)	2.4×10^4	186.3	1.4	8.7	13.1 (21.10)[e]	86.60 (68.43)[bc]
	2.4×10^5	184.1	1.3	8.0	36.1 (36.86)[b]	63.9 (53.03)[f]
	2.4×10^6	204.5	1.3	8.5	27.8 (31.80)[c]	72.20 (58.10)[de]
	2.4×10^7	189.3	1.3	8.5	25.0 (30.00)[d]	75.0 (59.31)[d]
$CD(p_{=0.05})$		0.58	0.19	0.80	0.65	3.04
SE		0.19	0.03	0.55	0.22	1.06

Each value is a mean of four replications
Figures in parenthesis are arcsin transformed
In a column means followed by a common letter are not significantly different (P=0.05) by DMRT

Comparison between the different fungal treatments revealed that least pupal weight (184.1 mg) was recorded. The decrease in the juvenile hormone titre and its associated disturbances in oogenesis, larval-pupal and pupal-adult moults were interpreted as an interference with moulting hormone pools (Rembold *et al.,* 1982). Decrease in juvenile hormone influences the storage proteins and fat which were highly essential for metamorphosis, moulting and reproduction (Palli and Locke, 1987; Koul and Isman, 1991).

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