



Original Research Article**DOI: 10.26479/2018.0404.18****EFFECT OF NEEM AND GARLIC BASED BIOPESTICIDES ON THE REPRODUCTIVE PARAMETERS OF *EARIAS VITTELLA* (FABR.) (LEPIDOPTERA: NOCTUIDAE)****Pratibha, Rajendra Singh***Department of Zoology, Deendayal Upadhyay Gorakhpur University Gorakhpur, U.P. India

ABSTRACT: The effect of sublethal doses (20% and 60% of LC₅₀) of neem derivatives such as NeemGold (NG), aqueous extract of neem leaves (AEN) and aqueous extract of garlic bulb (AEG) at different concentrations, were observed on fecundity, incubation periods, hatchability and larval pupal as well as total mortality of *Earias vittella* (Fabr.) (Lepidoptera: Noctuidae). Results demonstrated that all the biopesticides significantly reduced the fecundity and hatchability of the *Earias vittella*. larval, pupal mortality of *Earias vittella* significantly increased with increase in concentration of the biopesticides tested. After 24 h exposure, at 20% sublethal dose of LC₅₀ of AEG, AEN and NG, the fecundity reduction were 177.8, 172.67 and 169.50 eggs/female respectively. At 60% sublethal dose the fecundity reduction was much more, i.e. 165.50 for AEG, 143.83 for AEN and 139.67 eggs/female for NeemGold. Incubation periods insignificantly affected by exposure periods as well as by biopesticides tested. Similarly, hatchability reduced upto 41.61 per cent on 60% sublethal dose of NeemGold after 24 h. Larval mortality increased after application of AEG 38.46 per cent, AEN 51.53 per cent and NG 59.46 per cent. Pupal mortality were increased after 24 h by the application of AEG upto 51.63 per cent AEN 59.35 per cent and NG 70.65 per cent. Alongwith this total mortality increased after 96 h of exposure period after applying AEG upto 69.56 per cent AEN 80.07 per cent and NG caused highest 88.23 per cent of total mortality.

KEYWORDS: *Earias vittella*, Biopesticides, Fecundity, Eggs, Hatchability, Total mortality.

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1. INTRODUCTION

The shoot and fruit borer, *Earias vittella* (Fab.) (Lepidoptera : Noctuidae) is one of the major insect pest of okra (*Abelmoschus esculentus*) and is cultivated mostly in tropical and subtropical area of the world [1,2]. Upto 90% infestation on okra depending on season was reported by Misra, *et al.* (2002) [3]. Biopesticides are plant derived pesticides and have been reported to have pronounced effect on the development, growth, adult emergence, fecundity, fertility, and embryogenesis resulting in effective control of pest insects [4]. The essential oils of tulsi (*Ocimum basilicum* L.), neem (*Azadirachta indica* A. Juss.) and eucalyptus (*Eucalyptus rostrata* Schlecht) have been demonstrated to inhibit substantially the egg hatchability of *E. vittella* [5, 6]. *A. indica* works as larvicidal, growth regulator, antipupational, repellent and oviposition inhibitor against *E. vitella* [5,6,7,8,9,10]. These studies demonstrated that the biopesticides diminish egg laying rate, egg hatchability, pupation, percentages of adult emergence, ovipositional period and fecundity of okra moth. Previous studies have shown that garlic also possess some insecticidal properties [11]. The advantage of using biopesticides is that they are easily available and have low or no toxicity to humans [12]. The deleterious effects of plant products on insects can be manifested in several manners including toxicity, mortality, antifeedant growth inhibitor, suppression of reproductive behavior and reduction of fecundity and fertility, growth inhibition, perturbation of reproductive behaviour [13]. The present work is an attempt to explore the efficacy of locally available NeemGold (NG), aqueous extract of neem leaves (AEN) and aqueous extract of garlic (AEG) to observe the fecundity, incubation periods, hatchability and mortality of *E. vittella* in the climatic condition of northeastern Uttar Pradesh.

2. MATERIALS AND METHODS

The *E. vitella* were reared following the standard method [14] in the laboratory. The adult moths were procured from the insectaries. The adults were genderwise identified and paired to mate in glass beakers (250 ml). After mating, they were placed in oviposition cages (30x30x30 cm) for egg laying. After hatching, the young larvae were reared on fresh fruits of okra at room temperature in the month of June-July 2016.

2.1 Preparation Of Biopesticides

2.1.1. Aqueous Extract Of Garlic Bulb

AEG was prepared by grinding 1 kg of AEGs with 1 l of distilled water. The extract was squeezed through fine meshed rayon cloth and finally filtered through Whatman filter paper. The filtrate (w/v) was used as test biopesticides.

2.1.2. Aqueous Extract Of Neem Leaves

AEN was prepared by grinding 1 kg of fresh leaves of neem with 1 l of distilled water. The extract was squeezed through fine meshed rayon cloth and finally filtered through Whatman filter paper. The filtrate (w/v) was used as test biopesticides.

2.1.3. Neem Gold Preparation

NG was purchased from local market. NeemGold contains Azadiractin A 0.03%, NG 90.57%, Hydroxy EL 5.00%, Epichorohydrin 0.50% and Aromex 3.90%, and is manufactured by Foliage Crop Solution Private Limited, Chennai, 600006, India. For preparation of test solution, 1 ml of NeemGold was dissolved in 100 ml of distilled water.

2.2. Experimental Sets Up

Six replicates each of eight treatments were setup and fecundity, incubation periods, hatchability and larval-pupal mortality was observed. For this, mated females were allowed to oviposit to observe the number of eggs laid in oviposition cages. LC_{50} for each treatment of biopesticides was calculated [15] and 20% and 60% of 24h, 48h, 72h and 96 h of LC_{50} of given biopesticides were applied on fresh leaves of okra plant and dried under the fan and in case of control, leaves were dipped in distilled water and dried accordingly. The females were allowed to deposit their eggs on these leaves. Treated leaves kept in the beakers having moist filter paper at their bottoms at alternate day. The numbers of eggs laid were counted within 3-4 days of egg laying to observe total fecundity. These eggs were kept under observation until hatching. After hatching, twenty neonate larvae were transferred gently on bisected fresh pods with the help of camel brush treated and dried under fan with biopesticides as mentioned earlier until pupation and pods were changed an alternate days. The data regarding the hatchability, incubation period, larval and pupal mortality were recorded.

3. RESULTS AND DISCUSSION

The present study revealed that the spraying of biopesticides on the eggs and feed of insect with the selected biopesticides at different concentration and doses reduces the female fertility, viability of eggs and that affect the egg hatchability and total survivability. The advantages of botanical pesticides mainly depend upon their quick degradation and lack of persistence and bioaccumulation in the ecosystem, which have been key problems in chemical pesticide use. The deleterious effects of neem products on lepidopteran pests have been well documented by many researchers in past [16,17,18,19].

3.1. Effect On Fecundity

Data relating to the effect of three biopesticides on the fecundity of *E. vittella* at different sublethal doses, i.e., 20% and 60% of LC_{50} calculated at different exposure periods viz. 24, 48, 72, 96 h, are shown in Table 1. The data show that AEG significantly affect the fecundity of *E. vittella* at its different doses. Female laid 177.8 ± 12.18 eggs/female and 165.50 ± 8.90 eggs/female at 20% and 60% of LC_{50} of 24 h while in control setup it laid 269.50 ± 9.24 eggs/female, which were significantly more than that of such doses of 96 h exposure periods. Similar results were observed for other biopesticides tested i.e., AEN and NG (Table 1). Comparison of toxicity of these three biopesticides demonstrated that NG is more detrimental in reducing the fecundity of *E. vittella* at different sublethal doses followed by AEN and AEG. At very low dose, i.e., 20% of LC_{50} all the

biopesticides behave alike while at 60% of LC₅₀ NG seems to be more toxic than that of AEN and AEG. The vapour of the leaves of neem (*A. indica*) have significantly inhibited egg hatchability of *E. vittella* [20].

Table 1. Comparison of different biopesticidal sublethal dose (20% and 60%) on the fecundity of *Earias vittella* at varying exposure periods. Values are expressed as mean \pm SE.

Different hours of exposure of LC _{50s}	Percentage of sublethal doses	AEG	AEN	NG	F-values
24	20	177.8 \pm 12.78	172.67 \pm 6.33	169.5 \pm 9.24	For 20% F _{conc} =24.73*
	60	165.5 \pm 8.90	143.83 \pm 8.06	139.67 \pm 11.17	
48	20	180.67 \pm 12.78	191.17 \pm 6.91	183.17 \pm 8.36	F _{bp} =0.58*
	60	168.6 \pm 10.54	169 \pm 9.33	159.17 \pm 10.21	
72	20	197.67 \pm 12.67	201.33 \pm 6.34	200.17 \pm 11.45	For 60% F _{conc} =2.10*
	60	180.67 \pm 14.72	189.67 \pm 7.46	171.33 \pm 11.17	
96	20	230.8 \pm 12.48	210.33 \pm 10.14	205.17 \pm 8.43	F _{bp} =32.04* *P < 0.05
	60	194.33 \pm 11.80	197.5 \pm 10.63	190.83 \pm 10.25	

3.2. Effect On Incubation Periods

Data regarding the effect of three biopesticides on the incubation periods of *E. vittella* at different sublethal doses 20% and 60% of LC₅₀ of different exposure periods, viz. 24 h, 48 h, 72 h, 96 h, are given in Table 2. The results demonstrated that there was no effect of sublethal doses of all biopesticides tested on the incubation periods of *E. vittella*.

3.3. Effect On Eggs Hatchability

Data analysis concerning the effect of three biopesticides on the per cent hatchability of eggs of *E. vittella* at different sublethal doses 20% and 60% of LC₅₀ of different exposure periods (viz. 24 h, 48 h, 72 h, 96 h), are shown in Table 3, which shows that AEG at different sublethal doses significantly influence the hatchability percentage of eggs of *E. vittella*. 71.20 \pm 1.49 per cent and 62.60 \pm 1.48 per cent eggs hatched at sublethal doses 20% and 60% of LC₅₀ of 24 h exposure period, respectively. Comparable results were observed for other biopesticides tested i.e. AEN and NG (Table 3). Comparison of these three biopesticides demonstrated that NG is more effective in decreasing percentage of hatchability of eggs of *E. vittella* followed by AEN and AEG. In control setup 90.75 \pm 1.98 per cent eggs were hatched. It has been concluded that two commercial neem based formulations (Nimbecidine and Neemazal) have the potential for use in the management of *E. vittella* [21]. The hatchability of eggs laid was significantly reduced (25-50%) and showed concentration dependent effect.

Table 2: Comparison of different biopesticidal sublethal dose (20% and 60%) on the incubation period of *Earias vitella* at varying exposure periods. Values are expressed as mean \pm SE.

Different hours of exposure of LC _{50s}	Percentage of sublethal doses	AEG	AEN	NG	F-values
24	20	3.50 \pm 0.44	3.67 \pm 0.21	4.17 \pm 0.17	For 20% F _{conc} =0.72*
	60	4.17 \pm 0.40	3.50 \pm 0.43	4.33 \pm 0.33	
48	20	4.17 \pm 0.52	3.83 \pm 0.31	4.33 \pm 0.33	F _{bp} =3.14*
	60	4.00 \pm 0.47	3.33 \pm 0.33	3.83 \pm 0.17	
72	20	3.33 \pm 0.43	3.67 \pm 0.21	3.50 \pm 0.22	For 60% F _{conc} =6.93*
	60	3.67 \pm 0.43	3.00 \pm 0.26	3.50 \pm 0.34	
96	20	3.8 \pm 0.40	3.50 \pm 0.20	3.50 \pm 0.34	F _{bp} =5.78* *P > 0.05
	60	4.17 \pm 0.22	3.33 \pm 0.21	3.33 \pm 0.19	

Table 3. Comparison of different biopesticidal sublethal dose (20% and 60%) on the per cent egg hatchability of *Earias vitella* at varying exposure periods. Values are expressed as mean \pm SE.

Different hours of exposure of LC _{50s}	Percentage of sublethal doses	AEG	AEN	NG	F-values
24	20	71.20 \pm 1.49	52.97 \pm 1.07	46.57 \pm 1.05	For 20% F _{conc} =21.43*
	60	62.60 \pm 1.48	46.73 \pm 1.02	41.61 \pm 1.18	
48	20	73.51 \pm 1.49	56.02 \pm 1.01	60.53 \pm 1.14	F _{bp} =22.68*
	60	66.54 \pm 1.90	54.57 \pm 1.20	56.05 \pm 1.11	
72	20	79.31 \pm 1.38	67.90 \pm 0.83	61.35 \pm 1.22	For 60% F _{conc} =24.54*
	60	74.91 \pm 1.63	63.70 \pm 2.58	59.79 \pm 1.21	
96	20	85.49 \pm 1.60	74.25 \pm 1.18	65.53 \pm 0.92	F _{bp} =50.77* *P < 0.05
	60	78.00 \pm 1.30	70.08 \pm 1.22	63.60 \pm 1.14	

3.4. Effect On Larval Mortality

Results of the effect of three biopesticides on per cent larval mortality of *Earias vittella* at different sublethal doses (viz. 20% and 60% of LC₅₀ of different exposure periods i.e. 24 h, 48 h, 72 h, 96 h), are displayed in Table 4. It shows that AEG at different sublethal doses significantly influence the percentage of larval mortality of *E. vittella* at 20% of LC₅₀, 31.23 \pm 1.90 per cent and at 60% of LC₅₀

of 24 h exposure period 38.46 ± 7.49 per cent larval mortality were observed. With increase of concentration of the biopesticides, percentage of larval mortality increased. Similar trend were also observed for the other biopesticides tested i.e. AEN and NG. Comparison of these three biopesticides demonstrated that NG is more lethal to the larvae followed by AEN and AEG. Per cent larval mortality of *E. vittella* was 26.53 ± 6.04 after the application of NG by applying 20% of LC_{50} of 96 h exposure period, while larval mortality increased up to 51.66 ± 6.23 per cent after applying 20% of LC_{50} of 24 h exposure period. Similar trend was observed for 60% of LC_{50} dose. AEG showed least toxicity on the per cent larval mortality of *E. vittella*. In control, 18.64 ± 2.65 per cent larval mortality was observed.

Table 4. Comparison of different biopesticidal sublethal dose (20% and 60%) on the per cent larval mortality of *Earias vittella* at varying exposure periods. Values are expressed as mean \pm SE.

Different hours of exposure of LC_{50s}	Percentage of sublethal doses	AEG	AEN	NG	F-values
24	20	28.62 ± 6.17	35.11 ± 5.54	51.66 ± 6.23	For 20% $F_{conc} = 30.04^*$ $F_{bp} = 88.65^*$ For 60% $F_{conc} = 34.03^*$ $F_{bp} = 109.97^*$ $*P < 0.05$
	60	38.46 ± 7.49	51.53 ± 4.49	59.46 ± 6.02	
48	20	31.23 ± 1.90	47.06 ± 5.59	41.76 ± 5.16	
	60	33.33 ± 2.50	40.99 ± 5.97	46.67 ± 5.41	
72	20	20.50 ± 3.60	26.54 ± 3.94	34.82 ± 3.29	
	60	28.95 ± 3.54	35.74 ± 3.82	41.35 ± 4.34	
96	20	10.88 ± 2.15	18.80 ± 2.32	26.53 ± 6.04	
	60	18.14 ± 3.49	25.48 ± 2.39	31.10 ± 3.46	

3.5. Effect on Pupal Mortality

The effect of three biopesticides on per cent pupal mortality of *Earias vittella* at different sublethal doses (i.e. 20% and 60% of LC_{50} of different exposure periods viz. 24 h, 48 h, 72 h, 96 h), are shown in Table 5. AEG at different sublethal doses significantly influence the pupal mortality of *E. vittella*. At 20% 46.81 ± 7.43 per cent and at 60% 51.63 ± 6.18 per cent pupal mortality of LC_{50} of 24 h showed in Table 5. Similar results were also observed for the other biopesticides tested i.e. AEN and NG. Comparison of these three biopesticides demonstrated that NG is more toxic to the pupal mortality of *E. vittella* followed by AEN and AEG. Pupal mortality of *Earias vittella* was 37.89 ± 3.12 per cent after the application of NG by applying 20% of LC_{50} of 96 h exposure, increased upto 68.40 ± 7.18 per cent after applying 20% of LC_{50} of 24 h exposure period. Similar tendency was observed for 60% of LC_{50} concentration. AEG showed least toxicity on the pupal mortality of *E. vittella*.

Table 5. Comparison of different biopesticidal sublethal dose (20% and 60%) on the per cent pupal mortality of *Earias vitella* at varying exposure periods. Values are expressed as mean \pm SE.

Different hours of exposure of LC _{50s}	Percentage of sublethal doses	AEG	AEN	NG	F-values
24	20	46.81 \pm 7.43	53.24 \pm 7.43	68.40 \pm 7.18	For 20% F _{conc} = 43.11*
	60	51.63 \pm 6.18	59.35 \pm 2.76	70.65 \pm 7.16	
48	20	38.06 \pm 4.22	45.17 \pm 1.69	53.08 \pm 2.94	F _{bp} = 125.71*
	60	45.05 \pm 7.47	50.17 \pm 5.22	58.79 \pm 7.16	
72	20	30.05 \pm 3.60	36.61 \pm 4.98	45.94 \pm 4.04	For 60% F _{conc} = 27.68*
	60	39.00 \pm 6.62	43.40 \pm 4.32	51.11 \pm 6.80	
96	20	24.66 \pm 5.50	29.91 \pm 5.22	37.89 \pm 3.12	F _{bp} = 105.86*
	60	30.83 \pm 4.36	37.01 \pm 1.60	40.69 \pm 3.63	

*P < 0.05

3.6. Total Percent Mortality

Effect of three biopesticides on total mortality of *E. vittella* at different sublethal doses of 20% and 60% of LC₅₀ of different exposure periods (viz. 24 h, 48 h, 72 h, 96 h), are given in Table 6. Table shows that AEG at different doses, significantly affect the per cent total mortality of *E. vittella*. At 20%, 32.97 \pm 4.64 and at 60% of LC₅₀ of 24 h it increased upto 69.56 \pm 7.05 per cent. Similar trend were also observed for the other biopesticides tested i.e. AEN and NG. Comparison of these three biopesticides demonstrated that NG is more affective followed by AEN and AEG. Total mortality of *E. vittella* was 85.52 \pm 3.74 per cent after application of 20% of LC₅₀ of 24 h exposure period, increased upto 88.23 \pm 3.98 per cent at 60% of LC₅₀ of 24 h exposure period after the application of NG. Similar trend were observed for AEN and AEG (Table 6). In Control setup, 22.63 \pm 5.92 per cent total mortality was observed. Whereas AEG showed less toxicity on the total mortality than that of other two biopesticides against *E. vittella*. The sub lethal doses of essential oil showed that it reduced adult emergence of meal moth *Plodia interpunctella* (Lepidoptera: Pyralidae) (Hübner) in laboratory condition, longevity of male and female insects, fecundity and fertility of females. Evaluation of toxic vapors on adult insect was also considered and LC₂₅, LC₅₀ and LC₇₅ were estimated to be 6.35%, 8.13% and 10.45% [22]. When larvae or adults were exposed with neem and eucalyptus or garlic and mint oil a significant reduction were observed in reproductive potential in breeding pairs of *Corecya cephalonica* (Stainton) (Lepidoptera: Pyralidae [23,24]. The result is general demonstrated statistically significant (p<0.005) effect of the biopesticides tested i.e. AEG, AEN and NG, on fecundity, hatchability and per cent larval and pupal mortality of *E. vittella*. Both effective doses as well as type of biopesticides significantly affected these parameters.

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Table 6. Comparison of different biopesticidal sublethal dose (20% and 60%) on the per cent total mortality of *Earias vitella* at varying exposure periods. Values are expressed as mean \pm SE.

Different hours of exposure of LC _{50s}	Percentage of sublethal doses	AEG	AEN	NG	F-values
24	20	63.21 \pm 5.80	73.92 \pm 5.86	85.52 \pm 3.74	For 20% F _{conc} =207.05*
	60	69.56 \pm 7.05	80.07 \pm 3.96	88.23 \pm 3.98	
48	20	55.54 \pm 5.66	64.37 \pm 3.44	72.88 \pm 2.58	F _{bp} =577.28*
	60	63.69 \pm 4.41	71.05 \pm 3.18	77.02 \pm 5.86	
72	20	44.47 \pm 3.32	53.48 \pm 4.59	64.80 \pm 3.25	For 60% F _{conc} =113.49*
	60	56.53 \pm 5.82	64.17 \pm 2.06	72.15 \pm 2.80	
96	20	32.97 \pm 4.64	43.13 \pm 4.54	54.76 \pm 3.35	F _{bp} =412.58*
	60	43.33 \pm 4.56	53.01 \pm 2.17	58.80 \pm 4.23	

4. CONCLUSION

The present findings suggest that freshly prepared AEG which has allicin as its main biologically active compound possesses a potentially vital insecticidal effect on *E.vittella* compared with the control. Thus, neem leaves extract, garlic bulb extract and NeemGold offers significant promise for combating the threat of bollworms to farmers in developing countries. The major thrust of this work is its adaptability for use by small scale farmers plagued by the challenge of not being able to afford conventional costly pesticides in the market. There is need for further investigations to identify the other neem and garlic juice constituents (apart from azadiractin and allicin respectively) with toxic effects on *E. vittella*, and to elucidate the precise mechanisms by which they exert their insecticidal effects.

6. CONFLICT OF INTEREST

I declare that no financial interest or any conflict of interest exists.

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