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Development of the predator *Eocanthecona furcellata* on different proportions of nucleopolyhedrovirus infected *Spodoptera litura* larvae and potential for predator dissemination of virus in the field

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Abstract The present work aimed to determine the impact of nucleopolyhedrovirus (NPV) on the development of *Eocanthecona furcellata* Wolff and to measure the infectivity of NPV discharged through its faeces. Developmental time, survival rate, sex ratio and incubation period of the predator reared on different proportions of healthy and NPV infected *S. litura* larvae as lifetime meal did not vary significantly. However, when the proportion of infected prey in the predator's lifetime meal exceeded 50 % significant reductions in body weight, fecundity, longevity and percent egg hatchability were noticed. The virus did not show detrimental effect on the developmental biology of the predator in the subsequent generations when offspring emerging from nymphs fed only on virus infected prey throughout their life were reared on 50 % of infected prey. Therefore, adverse effects on *E. furcellata* are unlikely in the field as the predator is not expected to consume more than 50 % of infected prey even if the virus application is done at recommended dose. Under field conditions, it was found that increased proportions of infected prey enhanced the dissemination of viral inoculum through the faeces of the predators and the subsequent infection in *S. litura* larvae ranged from 22 to 52 %.

Keywords Biological control · Bioinsecticides · Entomopathogens · Pentatomidae

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Introduction

Baculoviruses have been recognized to have potential as biological control agents against *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae) (Moscardi 1999; Moscardi et al. 2011; Zhong et al. 2011). However, the ability of baculoviruses to persist and spread in agro-ecosystems is a key factor in successful suppression of pest populations. The environmentally stable occlusion body (OB) stage of baculoviruses persists in the soil and on phylloplanes and is amplified in arthropod hosts that consume and are infected by them. The subsequent release of infectious virus OBs from dead larvae is the primary method by which virus is

dispersed and transmitted to healthy hosts. Additionally, predator mediated baculovirus dispersal has been demonstrated to be an important mechanism for virus dissemination in a diversity of crop and forest habitats (Smirnoff 1959; Biever et al. 1982; Young and Yearian 1987, 1992; Entwistle et al. 1993; Fuxa and Richter 1994). The mechanism for dispersal may be twofold. First, predators that consume baculovirus infected larvae may excrete viable viral OBs in their faeces for periods of several days following the meal (Beekman 1980; Young and Yearian 1987; Vasconcelos et al. 1996). Second, natural enemies may become superficially contaminated during the consumption of baculovirus infected larvae and may physically disperse the virus over plant surfaces (Sait et al. 1996).

Insect viruses play a major role in pest management but their effective integration into such systems depends on their compatibility with other components of the systems. If an entomopathogen such as nucleopolyhedrovirus (NPV) is to be used for the control of an insect pest, one must first assess the impact of the pathogen on non target beneficial insects. One advantage of the virus over conventional pesticides is that it is non-toxic to the natural enemies of the target pest (Szewczyk et al. 2006). However, this does not mean that applications of virus will not have adverse effects on predator populations. An understanding of such interactions is therefore necessary to predict the environmental effects of NPV applications on predator populations. Among the promising natural enemies of *S. litura*, the predatory stink bug, *Eocanthecona furcellata* Wolff (Hemiptera: Pentatomidae) is regarded as an important predator in cotton, chickpea and vegetable fields in Southeast Asia, Japan, India, and Taiwan (Yi and Kyi 2000; Nyunt 2001). Since *E. furcellata* can easily be reared on larvae of *Pieris rapae* Linnaeus (Lepidoptera: Pieridae) (Chu 1975) and frozen preserved larvae of *S. litura* (Yasuda and Wakamura 1992), its augmentative releases are recommended in Southeast Asia (Suasa-ard 1988; Aganon and Romero 2008). Baculoviruses and *E. furcellata* are known to coexist in crop ecosystems and sometimes their application and release (Siri 2007; Nyunt 2008) are made simultaneously or sequentially, but the ecological implications of their co-existence have not been studied in the field. Previous studies have shown that some parasitoids and predators could be detrimentally affected by the pathogen infecting their prey (Cossentine and Lewis 1988; Sajap and Lewis 1988; Ruberson et al. 1991). There are reports, however, showing that predators were unaffected by the pathogens (Kaya 1982; Young and Hamm 1985; Sajap 1989). Occasionally, the predators disseminated the pathogen through their faeces into the ecosystem (Capinera and Barbosa 1975; Young and Hamm 1985; Sajap and Lewis 1988). However, in most of these studies the predators were provided either healthy or virus infected prey only as a lifetime meal (Ruberson et al. 1991) or healthy prey during nymphal period and infected prey during adult stage and vice versa (Sajap et al. 1999). Although the consumption of infected larvae and subsequent release of viable virus by pentatomids are likely to occur in nature, the ecological implications of these phenomena have not been studied under field conditions. However, in-depth understanding of impact of baculoviruses on non target organisms, as well as their potential dispersal by such organisms is important for successful biological control in fields where the predator is not expected to consume infected or healthy prey only during its life cycle. Here we studied the impact of NPV on the development and some biological attributes (survival rate, body weight, sex ratio, pre-oviposition period, oviposition period, fecundity, incubation period, percent egg hatchability and longevity) of *E. furcellata* and measured the infectivity of NPV discharged in the faeces of *E. furcellata* in the field when different proportions of healthy and virus infected *S. litura* larvae were offered as prey to *E. furcellata* during its life-cycle. This information will be indicative of the compatibility of NPV and *E. furcellata* in an integrated pest management system and likelihood of virus disseminated by the predator to be acquired by host larvae in field situations.

Materials and methods

Host insect *Spodoptera litura*

The pest used in this study was obtained from the offspring of 34th generation female moths obtained from the Indian Agriculture Research Institute, New Delhi. The culture was maintained at 28 ± 2 °C, 60 % RH, and 16L:8D photoperiod on the artificial diet as suggested by Shorey and Hale (1965) with certain modifications (Gupta et al. 2010). Egg masses were collected and surface-sterilized with 0.01 %

sodium hypochlorite. The larvae were reared in a plastic container (30 × 20 × 10 cm^3) lined with a paper towel at the bottom. The diet and the paper were changed every 24 h.

Predator *Eocanthecona furcellata*

The predator used in this study was obtained from the offspring of 20th generation female bugs. The insect culture was started with eggs collected from *Parthenium hysterophorus* leaves (Gupta et al. 2004). The predators were maintained in the laboratory at 28 ± 2 °C, 60 % RH, and 16L:8D photoperiod, and fed every 24 h with healthy *S. litura* larvae.

Virus preparation

The multiple NPV used in this study was originally isolated from naturally field-infected *S. litura* larvae collected from tomato fields in Jammu, India (Monobrullah et al. 2007). The virus was propagated in *S. litura* larvae maintained on a semi synthetic diet based on chick pea flour. Viral occlusion bodies (OBs) were extracted by homogenizing virus-killed larvae in 0.1 % sodium dodecyl sulphate (SDS), followed by filtration through muslin cloth and subsequent pelleting through continuous sucrose gradient centrifugation for 1 h at 50,000×*g*. After several washes in TE (10 mm Tris–HCl, pH 8, 115 1 mm EDTA) the OBs were resuspended in 0.75 ml of distilled water and stored in aliquots at −20 °C. The virus was quantified using a haemocytometer and phase contrast microscope at ×800 magnification.

Development and reproduction of *E. furcellata* on different proportions of healthy and NPV infected *S. litura* larvae

The developmental biology of *E. furcellata* was compared by feeding predators different proportions of healthy and virus infected (48 h post-infection) *S. litura* larvae. For this, five treatment groups of predators were maintained on different proportions of healthy and virus-infected larvae of *S. litura.* The predators (both nymphs and adults) fed on healthy larvae of *S. litura* throughout their lives constituted the control group. The rest of four treatment groups were fed on different proportions of healthy and infected *S. litura* larvae i.e., 75 % healthy and 25 % infected, 50 % healthy and 50 % infected, 25 % healthy and 75 % infected and infected prey only. For each treatment group, 20 newly hatched nymphs were placed in each of three replicate arenas (Petri dishes 80 × 15 mm^2) supplemented with a tender shoot of *P. hysterophorus* wrapped in wet cotton, giving a total of 60 individual predatory nymphs for each treatment. Prey densities were maintained uniformly by providing the nymphs with fresh larvae and tender shoots every day. After second moult, the nymphs were reared individually in plastic containers (65 × 35 mm^2) and fed on host larvae only. To obtain infected larval prey 10 µl suspension of NPV, containing 1 × 10^7 polyhedra ml^{-1} was placed on each diet disc (1 mm thick × 4 mm diameter). The virus contaminated diet discs were offered to 3rd instar larvae of *S. litura* individually in plastic containers (65 × 35 mm^2) for 24 h to acquire the virus. More larvae were inoculated than required for experiments and only those larvae that had eaten the entire diet disc within 24 h were selected and transferred to fresh uncontaminated diet. Larvae fed on diet discs only constituted control prey. Developmental time, survival rate and body weight for each developmental stage of *E. furcellata* were recorded. Sex ratio was recorded at adult emergence. In order to study pre oviposition and oviposition period, total fecundity, and adult longevity of *E. furcellata,* ten pairs of newly emerged adult bugs were selected from each group on the basis of their weight (males and females were restricted to 65–68 and 70–73 mg, respectively). Each adult pair was kept individually for mating in a glass jar (30 × 20 × 10 cm^3). These experimental adults were assigned to five groups and were offered the same type of prey as fed to nymphs. Observations on incubation period and hatchability of eggs (proportion of hatched eggs of those surviving the incubation) were recorded for each group.

To evaluate the effect of feeding on virus infected prey on subsequent predator generations, a laboratory cohort of 200 newly emerged nymphs were randomly selected and reared on virus infected prey only as lifetime meal. This was termed parental group (control). From this group, ten randomly selected females were mated to an equal number of males separately in glass jars (30 × 20 × 10 cm^3) and their resulting progeny were reared for four generations. In each generation, a group of two hundred newly emerged nymphs were reared on equal proportions of healthy

and virus infected larvae as lifetime meal. Under field conditions predators have equal probability of feeding on healthy and virus infected prey. In each generation, body weight at emergence, longevity, fecundity and percent egg hatchability were recorded.

Virus transmission by *E. furcullata* under field conditions

To determine the ability of *E. furcellata* as an agent of virus dissemination in the field, an experiment was performed on six-weeks-old cabbage (*Brassica oleracea* L. var. *Capitata* cv. golden acre) plants that were grown in a single plot (20 × 20 m^2). Of plants with no signs of *S. litura* infestations, 150 were randomly selected and allocated to one of five treatments. Treatments involved placing one *E. furcellata* adult on the top third portion of the cabbage plant which was then enclosed by a nylon mesh bag 35 cm tall and 30 cm wide, gently tied at the base to minimize insect movement. One day prior to the trial *E. furcellata* adult were fed on a single meal of different proportions of healthy and virus-infected *S. litura* larvae (48 h post-infection) i.e. 75 % healthy and 25 % infected, 50 % healthy and 50 % infected, 25 % healthy and 75 % infected and infected prey only respectively. Predators provided with healthy *S. litura* larvae only constituted the control. Each treatment was replicated 25 times. After 48 h, the bags were opened and predators were removed. The plants were carefully inspected for presence of predator excretion spots which almost exclusively (99 % of observations) were found on the upper side of leaves and never on stems. Each experimental plant was then infested with five healthy second instar *S. litura* larvae taken from the laboratory culture as natural infestation levels rarely exceeded three larvae per plant. The bags were replaced and tied as mentioned above. After six days, the bags were removed and *S. litura* larvae were recovered. The larvae were then reared individually on semi synthetic diet and checked daily for virus-induced mortality for up to seven days. All virus deaths were confirmed by microscopic examination through Giemsa staining (Richards et al. 1999).

A set of five additional plants per treatment were maintained for quantification of OBs and their bioassay. The excretion spots from the leaves of additional plants were counted and collected using a sterile toothpick. All spots from each individual plant were suspended together in 100 µl of sterile distilled water using a vortex mixer. This suspension was examined microscopically to determine quantitatively the presence of viral OBs. Each suspension was then bioassayed for infectivity by feeding 1 µl to each of 20 second-instar *S. litura* using the diet plug method in compartmentalized acrylic made insect rearing system designed by National Bureau of Agriculturally Important Insects (NBAII) Bangalore, India. The controls were consisting of excretion spots voided by predators treated similarly after eating healthy larvae. Larvae were kept individually on diet at 28 ± 2 °C, and checked daily for up to ten days and data were expressed as percent infectivity.

Data analysis

All analyses were performed utilizing SPSS version 16. Data on developmental period, weight, survival rate, pre-oviposition period, oviposition period, total fecundity, incubation period of eggs, percent egg hatchability and adult longevity of *E. furcellata* fed on different proportions of healthy and NPV infected prey were subjected to ANOVA followed by Tukey's post-hoc test for comparison of means. The data on sex ratio was subjected to χ^2 test. The data on total excretion spots, recovery of *S. litura* larvae from cabbage plants, mean number of OBs in excreta per plant, and percent infectivity of NPV in excreta of *E. furcellata* was analysed using one-way ANOVA.

Results

Impact of NPV infected prey on *E. furcullata* developmental stages and reproduction

The virus did not show detrimental effect on the developmental time of the surviving nymphs as no significant difference ($F = 0.526$; $df = 4, 295$; $P = 0.717$) was found between the developmental period of nymphs reared on different proportions of healthy and NPV infected *S. litura* larvae. Regardless of the proportion of prey consumed, the male and female nymphs required a mean ± SE of 19.8 ± 0.24 and 19.4 ± 0.20 days, respectively, to reach the adult stage. The mean developmental times for the first, second, third, fourth and fifth stadia were 2.4 ± 0.06, 3.4 ± 0.06, 4.0 ± 0.07, 4.0 ± 0.10 and 5.4 ± 0.12 days, respectively.

It was found that the proportion of healthy and infected prey that was offered as lifetime meal to nymphs and adults of *E. furcellata* significantly affected their body weight in all the nymphal stages except 1st instar (Table 1). The weights of *E. furcullata* were reduced when lifetime meal proportions exceeded 50 %, the differences being significant for 2nd ($F = 5.568$; $df = 4, 295$; $P < 0.001$), 3rd ($F = 5.012$; $df = 4, 295$; $P < 0.001$), 4th ($F = 7.652$; $df = 4, 295$; $P < 0.001$), 5th ($F = 6.864$; $df = 4, 295$; $P < 0.001$) instars, adult male ($F = 4.360$; $df = 4, 295$; $P = 0.002$) and female ($F = 4.450$; $df = 4, 295$; $P = 0.002$) of the predator.

No significant difference ($F = 1.390$; $df = 4, 295$; $P = 0.237$) was observed in the survival rate of nymphs and adults that were fed on different proportions of healthy and virus-infected *S. litura* larvae. Regardless of the type of prey offered the mean survival rate for the first and second instar were 83.6 and 88.8 % respectively while no mortality was observed in 3rd to 5th instar and adults of the predator.

The virus infected meal did not show detrimental effect on the sex ratio ($\chi^2 = 1.280$; $df = 1$; $P = 0.258$). However, when the infected prey proportions in lifetime meals exceeded 50 %, the longevity of male ($F = 5.115$; $df = 4, 45$; $P = 0.002$) and female ($F = 4.397$; $df = 4, 45$; $P = 0.004$) was significantly shortened by more than three and four days, respectively, pre-oviposition period was significantly prolonged ($F = 12.874$; $df = 4, 45$; $P < 0.001$) by more than 2–4 days while a significant reduction in oviposition period ($F = 10.834$; $df = 4, 45$; $P < 0.001$) was observed (Table 2).

Proportions of healthy and infected prey fed during nymphal and adult development had an influence on the production of egg masses of females ($F = 7.820$; $df = 4, 45$; $P < 0.001$). However, this reduction was significant only when the infected prey proportion comprised of more than 50 %. A similar trend was observed for total numbers of eggs with a percent reduction compared to control ranging from 35 to 37 %. However, the incubation period of *E. furcellata* eggs did not vary significantly among treatments ($F = 1.764$; $df = 4, 45$; $P = 0.152$) with a mean incubation period ranging from 5.2 ± 0.4 to 6.5 ± 0.4 days. The percent egg hatchability of females that emerged from nymphs fed on healthy, 25 % infected and 50 % infected of *S. litura* larvae did not differ significantly, but hatchability was significantly reduced ($F = 5.244$; $df = 4, 45$; $P = 0.001$) when proportions of infected larvae in lifetime meals exceeded 50 %. The percent reduction over healthy larvae ranged from 21 to 23 % (Table 3).

The virus infected prey did not show detrimental effect on the developmental biology of the predators in subsequent generations. Although significant reductions were observed in the body weight ($F = 21.223$; $df = 4, 995$; $P < 0.001$), longevity ($F = 9.944$; $df = 4, 995$; $P < 0.001$), fecundity ($F = 19.285$; $df = 4, 995$; $P < 0.001$) and percent egg hatchability ($F = 40.137$; $df = 4, 995$; $P < 0.001$) of adults emerging from nymphs fed on virus infected prey only throughout their life, these parameters were not significantly affected ($F = 1.321$; $df = 4, 995$; $P = 0.260$) in the subsequent four generations that were fed on equal proportions of healthy and infected prey as lifetime meal.

Table 1 Body weight (mean ± SE) of nymphs and adults and sex ratio at emergence of *E. furcellata* adults fed on different proportions of healthy and virus-infected *S. litura* larvae ($n = 60$ per treatment)

Proportion of infected prey in lifetime meal	Weight (mg)							Sex ratio (proportion of male)
	1st instar	2nd instar	3rd instar	4th instar	5th instar	Male	Female	
0	0.7 ± 0.03	2.3 ± 0.11b	9.2 ± 0.31b	21.4 ± 0.79b	55.1 ± 2.86b	73.6 ± 3.35b	81.1 ± 2.17b	0.44 ± 0.01
0.25	0.6 ± 0.03	2.2 ± 0.12b	9.0 ± 0.37b	20.9 ± 0.81b	53.5 ± 2.81b	72.6 ± 2.95b	80.1 ± 2.41b	0.47 ± 0.02
0.50	0.5 ± 0.05	2.1 ± 0.13b	8.8 ± 0.44b	20.1 ± 0.83b	53.0 ± 2.98b	72.0 ± 2.36b	79.0 ± 3.62b	0.42 ± 0.01
0.75	0.6 ± 0.04	1.7 ± 0.11a	7.5 ± 0.44a	17.4 ± 0.92a	44.0 ± 1.84a	62.1 ± 1.73a	69.7 ± 3.77a	0.46 ± 0.02
1.00	0.6 ± 0.03	1.7 ± 0.09a	7.2 ± 0.41a	16.0 ± 0.94a	40.7 ± 1.31a	59.6 ± 2.27a	66.4 ± 3.87a	0.46 ± 0.02

Means in the same column followed by different letters are significantly different (ANOVA; $P < 0.05$)

Table 2 Longevity and reproductive attributes (mean ± SE) of *E. furcellata* adults fed on different proportions of healthy and virus-infected *S. litura* larvae as lifetime meals ($n = 10$ per treatment)

Proportion of infected prey in lifetime meal	Longevity male (days)	Longevity female (days)	Pre-oviposition period (days)	Oviposition period (days)
0	18.7 ± 0.54b	22.1 ± 1.18b	7.6 ± 0.33a	11.9 ± 0.42b
0.25	18.3 ± 0.73b	21.6 ± 1.12b	7.9 ± 0.37a	11.5 ± 0.43b
0.50	17.7 ± 0.74b	21.5 ± 1.15b	8.7 ± 0.44a	11.1 ± 0.47b
0.75	15.2 ± 0.84a	17.9 ± 0.83a	10.2 ± 0.53b	6.0 ± 0.52a
1.00	15.1 ± 0.89a	17.2 ± 0.84a	11.4 ± 0.61c	5.2 ± 0.42a

Means in the same column followed by different letters are significantly different (ANOVA; $P < 0.05$)

Table 3 Mean fecundity, incubation period and egg hatchability of *E. furcellata* fed on different proportions of healthy and virus-infected *S. litura* larvae as lifetime meals ($n = 10$ per treatment)

Proportion of infected prey in lifetime meal	No. of egg masses laid	Total no. of eggs laid	Incubation period (days)	Egg hatchability (%)
0	5.8 ± 0.35b	173.5 ± 10.59b	6.2 ± 0.41	83.4 ± 3.12b
0.25	5.2 ± 0.40b	164.4 ± 9.31b	6.4 ± 0.43	77.8 ± 3.00b
0.50	5.3 ± 0.34b	147.2 ± 10.77b	7.3 ± 0.49	78.5 ± 3.04b
0.75	3.8 ± 0.41a	111.1 ± 11.32a	6.5 ± 0.52	62.5 ± 2.54a
1.00	3.6 ± 0.16a	109.5 ± 9.77a	7.5 ± 0.40	60.2 ± 2.12a

Means in the same column followed by different letters are significantly different (ANOVA; $P < 0.05$)

Virus transmission by *E. furcellata* in the field

The proportion of infected prey as single meal offered to predators prior to release on cabbage plants had significant effect on the total number of excretion spots ($F = 20.742$; $df = 4, 120$; $P < 0.001$). Six days post-release, recovery of *S. litura* larvae from cabbage plants did not differ significantly between treatments ($F = 1.245$; $df = 4, 120$; $P = 0.295$). However, prevalence of virus infection in larvae that were recovered from the field differed significantly between treatments ($F = 3.514$; $df = 4, 120$; $P = 0.009$) (Table 4). The percent infection due to virus in recovered *S. litura* larvae ranged from 22 to 52 % when the proportion of infected prey in the predators meal increased from 25 to 100 %. No virus infection was observed in larvae collected from control plants.

Collection of excretion spots from the additional plants in the field revealed that *E. furcellata* voided substantial amount of OBs in excreta and within 48 h of virus fed meal, all the excretion spots voided by virus fed predators were positive for virus. The total number of excretion spots varied significantly ($F = 18.332$; $df = 4, 20$; $P < 0.001$) between treatments with their numbers increasing with increase in the proportion of infected prey offered to the predator. The mean number of OBs in excreta per plant increased significantly ($F = 5.381$; $df = 4,20$; $P = 0.004$) from 1.3×10^2 to 1.2×10^6 with their mean infectivity against *S. litura* observed through bioassay ranging from 28 to 61 % as the proportion of infected prey offered to the predators increased from 25 to 100 %. No OBs were detected in the faeces of predators fed on healthy larvae (Fig. 1).

Discussion

The present study revealed that ingestion of ≤50 % of NPV-infected prey during the lifespan of *E. furcellata* had no detrimental impact on the development of the predator. However, significant reductions in the nymphal (except first instar) and adult weight were observed as the infected prey proportion in total meal consumed during the lifespan exceeded 50 %. The abundance of OBs in the body of prey

Table 4 Total number of excretion spots per plant voided by *E. furcellata* adults within 48 h of single meal on different proportions of healthy and virus infected prey and their infectivity after the predators were removed from the plants and subsequently infested with five healthy *S. litura* second instar larvae per plant ($n = 25$ per treatment)

Proportion of infected prey offered	Total no. of excretion spots (mean ± SE)	No. of larvae recovered after six days (mean ± SE)	Per cent infection in *S. litura* larvae (mean ± SE)
0	2.7 ± 0.18a	4.3 ± 0.14	0.00a
0.25	4.2 ± 0.25b	4.4 ± 0.13	22.0 ± 1.81b
0.50	5.1 ± 0.12b	4.3 ± 0.14	37.0 ± 2.56c
0.75	7.2 ± 0.31c	4.5 ± 0.14	41.0 ± 3.14c
1.00	8.2 ± 0.42d	4.4 ± 0.14	52.0 ± 2.81d

Means in the same column followed by different letters are significantly different (ANOVA; $P < 0.05$)

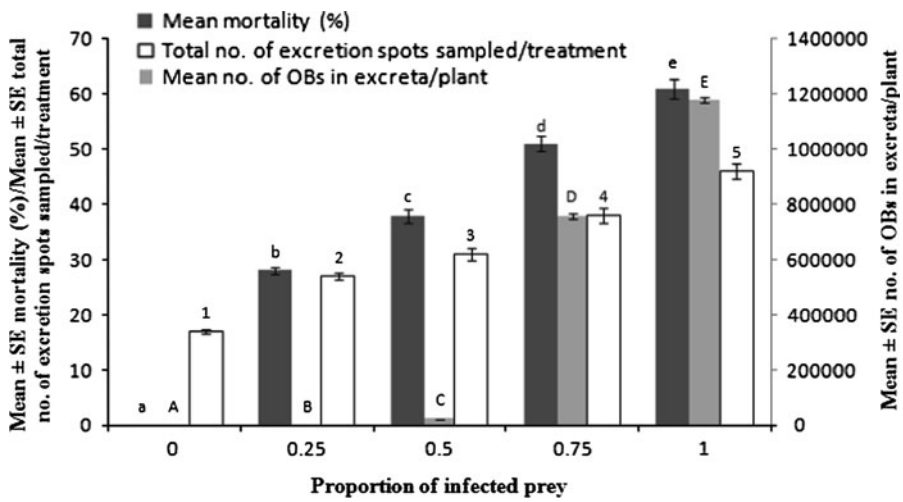

Fig. 1 Mortality (mean ± SE) of *S. litura* by OBs (mean ± SE) voided in *E. furcellata* excreta collected from additional cabbage plants ($n = 5$ per treatment) within 48 h of meal, offered as different proportions of healthy and virus infected prey and total no. of excretion spots (mean ± SE) sampled per treatment. Different letters and digits (lower case letters for mortality, capital letters for number of OBs and digits for total number of excretion spots sampled per treatment) above the bars indicate significant differences ($P < 0.05$; ANOVA)

probably reduced the assimilable proportion of proteins in haemolymph and fat body as suggested by Young and Yearian (1987), Ruberson et al. (1991) and therefore caused a nutritional imbalance to the predator. The pre-oviposition period was significantly delayed in females that emerged from nymphs fed on more than 50 % of infected prey in lifetime meals. This extended pre-oviposition period of virus fed bugs could have biological significance for mobile predators as they may increase the distance that the females are able to cover before they begin to lay eggs. The observed reduction of fecundity in females fed on infected larvae can be attributed to insufficient amount of protein needed for oogenesis (Mcfarlane 1985) or underdeveloped spermatocytes and oocytes in the developing ovaries possibly due to improper oocyte vitellogenesis (Rothman and Myers 1994). The current findings could indicate that an optimum level of protein reserves is essential in the post-nymphal development. While this level could be accumulated by the bugs which received about half of their lifetime meal as infected prey, beyond this limit the predatory bugs were not able to store optimum level of protein reserves. A significant reduction in the percent egg hatchability of *E. furcellata* that were fed on virus-infected *S. litura*

larvae (lifetime meal comprising >50 % infected prey) was observed in the present study.

The reduced adult weight, longevity and fecundity of insects are of greater concern as they are associated with reduced fitness which would have long-term negative impacts on population dynamics (Vickerman and Trumble 2003). However, as the predator did not show significant loss in weight, longevity and fecundity when provided ≤50 % proportion of infected prey in lifetime meal, it is possible that the predator was able to compensate nutritional balance with the remaining amount of healthy food. It can be implied that being a generalist predator, *E. furcellata* is not expected to consume larger proportions of infected prey than 50 % even if the virus application is done at the recommended dose. Also, a considerable proportion of OBs are inactivated in the field by ultraviolet radiation (Szewczyk et al. 2009), thereby reducing larval infection and therefore the amount of infected prey available to the predator. Thus, virus is not likely to have a significant effect on growth and development of *E. furcellata* in the field.

An indication of whether hemipteran predators play a significant role as passive disease vectors is demonstrated by the results from the field experiment. Previous experiments involving release of virus-fed predators into the field that were subsequently infested with host larvae resulted in low levels of infection (<5 %) of larvae (Vasconcelos et al. 1996; Castillejos et al. 2001). However, these experiments do not represent the natural field conditions as it is unlikely that predators will feed on infected prey only. In the present study it was found that increased proportions of infected prey enhanced the dissemination of viral inoculum through the faeces of the predators and the subsequent infection in *S. litura* larvae ranged from 22 to 52 %. This is quite contrary to earlier studies about negligible to very low levels of mortality as reported by Vasconcelos et al. (1996) using three carabid species, *Harpalus rufipes* De Geer, *Pterostichus melanarius* Illiger and *Agonum dorsale* Pont. The high mortality reported here was presumably due to higher mobility of *E. furcellata* and resultingly more contact with prey compared to carabids in the field (Vasconcelos et al. 1996). The contribution of the predators in baculovirus dissemination can only be ascertained by more extensive laboratory and field studies.

Although ingestion of >50 % of NPV infected prey in lifetime meals by *E. furcellata* could influence the body weight, fecundity, longevity and percent egg hatchability, such effects appear to be of short term and do not sustain in the subsequent generations. Hence the adverse effects on the developmental biology of *E. furcellata* might not occur in the field. From the present study, it can be concluded that field applications of NPV at recommended dose will not lead to deleterious effects on *E. furcellata*. Therefore this predator has the potential to be used in integrated pest management by predation as well as by dispersing the pathogens.

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