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Comparative Effect of Natural and Artificial Larval Diets on Biology of *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae)

Muzammil Sattar and Ghulam Hussain Abro
Nuclear Institute of Agriculture, Tando Jam (MS), and Department of Entomology, Sindh Agriculture University, Tando Jam, Pakistan (GHA)

Abstract.- A semi-solid artificial diet was compared with a factitious host *Sitotroga cerealella* (Olivier) eggs for economic mass rearing and commercial use of green lacewings *Chrysoperla carnea* (Stephens) in release programs against a variety of pests. The results revealed that highest percent larval survival and adult emergence were 93.25±0.53 and 91.50±0.63 on artificial diet compared with 85.25±1.30 and 81.75±1.60 on *S. cerealella* eggs, respectively. The larval period was much longer (10.00±0.20 days) on artificial diet than on natural diet of frozen eggs of *S. cerealella* (8.50±0.20 days) during sixth generation. The mean pupal weight of *C. carnea* fed artificial diet was 8.3±0.0 mg and for those fed on *S. cerealella* eggs, it was 8.1±0.0 mg in sixth generation. Similarly, fecundity per female was higher (397.10±3.60) for adults fed on artificial diet than it was for the larvae fed frozen eggs. The fertility of eggs of *C. carnea* diet-reared individuals was significantly higher ($P<0.05$) than *S. cerealella*-reared individuals. Almost all biological parameters of *C. carnea* studied, were higher in generation six, compared with generation one in both natural as well as artificial diets.

Key words: Green lacewings, mass production, natural and artificial larval diet.

INTRODUCTION

The introduction of exotic natural enemies for purposes of suppressing populations of an invasive pest species has been described (Bellows, 2001). Such studies would provide insight to further improve the effectiveness of indigenous natural enemies and might also improve our ability to predict predators whose introduction might be required for supplementary control of pests (Ridgway *et al*., 1970; Nordlund and Correa, 1995). Some invasive pests may be effectively controlled by generalist predators within a time frame, thus 'rear and release' strategy for pest management, such as conservation biological control is more appropriate (Michaud, 2002). Use of green lacewing, *Chrysoperla carnea* (Stephen) proved effective for management of many pests such as aphids, coccids, mites, mealy bugs, lepidopteran eggs and small larvae and a variety of other slow or non-moving soft-bodied arthropods (Hydron and Whitcomb, 1979; Zaki *et al*., 1999; Singh *et al*., 2003). Protocols for culturing of chrysopids using natural and artificial diets are available (Finney, 1948, 1950; Ridgway *et al*., 1970; Morrison *et al*., 1975; Morrison, 1977a; Cohen, 1992). Natural larval diets include eggs of the lepidopterans, *Phthorimaea operculella* (Zeller) (Finney 1948, 1950) and *Sitotroga cerealella* (Olivier) (Gelechiidae) (Ridgway *et al*., 1970; Morrison *et al*., 1975; Morrison, 1977a), *Anagasta kuehniella* (Zeller) (Pyralidae) (Zaki and Gesraha, 2001), *P. operculella* (Finney, 1948, 1950), honey bees, aphids (Tauber *et al*., 2000) and artificial diets (Zhang *et al*., 2004; Vanderzant, 1969; Cohen and Smith, 1998; Yazlovetsky, 1992).

To enhance egg laying efficiency for mass production of *C. carnea*, several larval diets have been developed. Zhang *et al*. (2004) and Vanderzant (1969) developed liquid and semi-liquid diets made primarily from enzymatic hydrolysate, casein, sugar, vitamins and water, but results were not satisfactory. Another artificial diet which contained wax-coated yeast hydrolysate droplets resulted in high mortality and was costly (Ridgway *et al*., 1970; Morrison *et al*., 1975; Morrison, 1977b). Vanderzant's diet was improved by adding vitamins and minerals, but all the larvae reared on this diet did not reach adulthood (Young *et al*., 1999, 2000). A semi-solid diet developed by Cohen and Smith (1998) containing protein, lipid, carbohydrate, cholesterol and water, proved better than *Sitotroga cerealella* eggs, in

* Corresponding Author: msattar73@yahoo.com
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increasing the colony size, and enhancing the predating ability of *C. carnea* larvae.

From the series of previous experiments (Sattar *et al*., 2007), a highly successful semi-solid artificial larval diet based on diet of Cohen and Smith (1998) and Cohen (1999) was developed for the predaceous green lacewing. This diet is full of nutrition and easy in presenting, was compared with frozen eggs of *S. cerealella* up to 18 generations. The impact of artificial diet was studied on all biological parameters uptil adult stage, and the results of generation one have been compared with those of generation six in the present paper.

MATERIALS AND METHODS

Procedure of diet preparation

The artificial diet based on Cohen and Smith (1998) and patented by Cohen (1999) was used in the present experiments.

Feeding artificial and natural diet to Chrysoperla carnea *larvae*

Culture of *C. carnea* larvae was obtained from the Bio-control laboratory, Nuclear Institute of Agriculture, Tando Jam, reared on factitious host *Sitotroga cerealella* (Olivier) frozen eggs. Culture of *S. cerealella* was mass reared at 27±2ºC and 65±5% RH with a photoperiod of L:D 16:8. Fresh eggs were daily collected from rearing glass jars, sieved, weighed and frozen at 4ºC for at least 24 hours to kill the embryo and then thawed before use as reported by Nordlund and Morrison (1992). Fifty newly hatched (less than 3h old) first instar unfed larvae for each treatment were randomly selected, placed individually in transparent plastic tubes (3mm dia and 7mm length) and provided with weighed diet (0.02 g) with the help of sterilized fine camel hairbrush. The tubes were sealed at both ends with impulse sealer.

The control was likewise run in parallel for which 0.1mg *S. cerealella* eggs per larva were placed in plastic tubes and sealed at both ends. Fresh diet/fresh eggs were provided in both the treatments after every four days till pupation. Larvae were observed daily for recording their larval period. After completion of larval period, plastic tube was cut from both ends and pupae were collected and kept in glass Petri dishes (1.5 cm height with 9.00 dia) with fresh green leaves of *Medicago sativa* to provide moisture inside Petri dishes. Pupae were observed daily for recording adult emergence. All emerged adults were counted and identified after 3rd day of emergence.

Many generations were produced with the artificial diet. In this paper the biological parameters of first generation has been compared with those of sixth generation for their competitiveness as a robust and vigorous predators and compared with the natural factitious host diet.

Adult feeding and oviposition

Adults were paired and placed in medium sized glass chimneys (10.2 cm height having 7.9 cm lower end and 6.6 cm upper end diameter) used as oviposition cage for single pair of adults. Eight replications were made to record the fecundity of both treatments.

A standardised diet containing protein hydrolysate, honey, yeast, sugar and water was provided on paper cards (4 mm in width and 6 mm in length) in droplets and wet cotton was provided as water in glass vials. Adults laid eggs on black muslin cloth, which was used as lid of the chimney. Eggs were harvested/ shaved with the help of razor from black cover as well as from walls of rearing chimneys and kept in folded black muslin cloth along with *S. cerealella* eggs as a diet to be used by freshly hatched larvae.

Statistical analysis

The whole data were statistically analysed and means were compared by t-test (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

The results indicate that artificial diet is a suitable alternative for *C. carnea* larvae. The protein, lipid, carbohydrate and water content of diet were similar to the nutrient profile of lepidopteran eggs (Cohen, 1992) and gross composition of lepidopteran larvae (Cohen and Smith, 1998).

Table I shows comparison of various biological parameters of *C. carnea* reared on artificial and natural diet. The percent fertility of

Table II.- **Biological parameters of *C. carnea* on natural and artificial diets.**

Parameters	Natural diet		Artificial diet	
	1st Generation	6th Generation	1st Generation	6th Generation
Incubation period (days)	3.75±0.25	3.75±0.16	3.62±0.18	3.50±0.19
Fertility (%)	83.50±1.30 b	87.25±0.65 b	94.00±0.53 a	94.75±0.53 a
Larval period (days)	10.10±0.20 b	10.06±0.12 b	12.27±0.23 a	12.41±0.17 a
Larval survival (%)	80.25±1.40 b	85.25±1.30 b	91.50±0.63 a	93.25±0.53 a
Pupal period (days)	9.65±0.13 b	9.62±0.15 b	10.30±0.13 a	10.20±0.11 a
Emergence (%)	77.25±1.60 b	81.75±1.60 b	89.50±0.73 a	91.50±0.63 a
Sex-ratio	1:1.38±0.05 b	1:1.40±0.04 b	1:1.55±0.04 a	1:1.68±0.08 a
Longevity				
Male	21.25±0.59	21.63±0.73 b	21.88±0.52	24.88±0.55 a
Female	32.88±0.93	33.13±0.52 b	34.50±0.57	36.88±0.44 a

Figures followed by same letter in a column are not significantly different at 5% DMRT.

eggs was significantly higher (t=7.23, P=0.001, df=14) for artificial diet fed insects compared with *C. carnea* reared on *S. cerealella* eggs. The larval development period of first generation was almost similar to that of 6th generation and it was significantly longer (t=7.16, P=0.001, df=83) on artificial diet compared with those fed on *S. cerealella* eggs. The larval survival was significantly high in 6th generation, compared to that of 1st generation (t=5.68, P=0.001, df=86). Pupal period of *C. carnea* from larvae reared on artificial diet in 1st and 6th generations was significantly longer than those fed on *S. cerealella* eggs (t=3.59, P=0.001, df=81 1st generation and t=3.09, P=0.001, df=84, 6th generation). Significantly more adults emerged from artificial diet in both generations, compared with natural diet (t=5.60, P=0.001, df=84, 6th generation). Female longevity of insects from natural diet of *S. cerealella* eggs was not significantly different (t=1.49, P=0.16, df=64) from insects of artificial diet in generation one but the same was significant in generation 6 (t=5.53, P=0.001, df=67).

Table II shows pupal weight and the fecundity of *C. carnea* reared on artificial and natural diets. There was no significant difference in pupal weight of *C. carnea* developed from larvae feeding on natural and artificial diets in both generations (t=0.61, P=0.55, df=14; t=1.13, P=0.28, df=14, 1st and 6th generations, respectively), but the pupae which developed on artificial diet were comparatively heavier in weight than the pupae of natural diet. Significantly more eggs were laid by females of artificial diet treatment than natural diet (t=4.16, P=0.001, df=14; t=7.17, P=0.001, df=14 first and sixth generation, respectively) in both generations.

Table II.- **Average pupal weight and fecundity of *Chrysoperla carnea* on natural and artificial diets.**

Diets	Av. pupal wt. (mg) Generation		Fecundity Generation	
	1st	6th	1st	6th
Natural	8.10±0.00	8.10±0.00	363.50±4.60b	365.75±2.50b
Artificial	8.20±0.00	8.30±0.00	385.38±2.60a	397.10±3.60a

Figures followed by same letter in a column are not significantly different at 5% DMRT

While preparing artificial diet for entomorphages, use of inert non-digestive fillers such as agar, cellulose etc should be avoided (Singh, 1977). This semi-solid diet supported continuous production of *C. carnea* that were aggressive predators. The viability of predator was normal up to 18 generations (data not shown). It has been reported that quantitative food inadequacy, inadequate nutrition usually results in great changes in the metabolism, behaviour and other characteristics of insect's vital activity (Yazlovetsky, 1992) and changes in the quality also affect fecundity, fertility and sex ratio in progeny (Hagley and Barber, 1992; Uckan and Gulel, 2000). Performance on artificial diet was generally superior or equal to that on the factitious host in present study. No reduction in population numbers in early generations was observed when insects were subjected to feeding on artificial diet (Bartlett, 1984; Cohen, 1992). This may be due to fact that the diet was a very nutrient-rich mixture of dietry components, especially lipids and was comparable to prey eggs and larvae in provision of nutrition (Cohen, 1992; Cohen and Smith, 1998; Florkin and Jeuniaux, 1974). Hen egg in the diet provided lipoproteins and phospholipids, which are an excellent replacement for insect components (Adams, 1975; Cohen and Smith, 1998).

Present artificial diet was protein-rich diet and *C. carnea* thrived better than its factitious host *S. cerealella* eggs diet. *C. carnea* fed on artificial diet had higher egg fertility, larval survival and adult emergence than natural diet of *S. cerealella* eggs. Pupae and adults of *C. carnea* developed from artificial diet weighed more and laid significantly more eggs than those developed on natural diet of *S. cerealella* eggs. Similar results were obtained by Cohen and Smith (1998) for another species *Chrysoperla rufilabris*.

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