

Effect of Prey Density on Biology and Functional Response of *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae)

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Abstract.- Effect of prey density on biology and functional response of green lacewing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) was studied in the laboratory of Entomology Section of Agricultural Research Institute, Dera Ismail Khan at 25±1°C, 65±5% RH and 10:14 light : dark regime. Newly emerged larvae of *C. carnea* were fed 20, 30, 40, 50, 60, 70, 80, 90 and 100 fresh eggs of *Sitotroga cerealella* (Lepidoptera: Gelechiidae) in 9 cm petri dishes. It was observed that the prey density had a significant effect on positive consumption rate, development and fecundity of *C. carnea*. In general maximum consumption with shortest developmental time, maximum fecundity and longest adult longevity were observed as prey density increased. In all treatments, predatory potential was high when the prey density was raised. Daily predation rate of *C. carnea* increased slowly during the first two instars and reached to its peak in the third larval instar. Although, *C. carnea* completed its development at all prey densities, the increase in prey densities reduced developmental time and mortality. Lacewing larvae provided with an overabundance of *S. cerealella* eggs developed faster than the larvae provided with fewer eggs. Lacewing fed during larval stage with 20 eggs/day showed lowest fecundity with the increase in prey density. A smaller intrinsic rate of increase was due to the fact that the population fed at a low prey density had prolonged developmental time, higher mortality rate in immature stages as well as a low daily rate of progeny.

Key words: *Chrysoperla carnea*, *Sitotroga cerealella*, prey density, functional response

INTRODUCTION

Green lacewing, *Chrysoperla carnea* (Stephens) commonly known as “aphid lion” is predominately important and widely distributed in Pakistan (Afzal and Khan, 1978) and other parts of the world (Geetha and Swamiappan, 1998). It is considered a prominent general predator that feeds on a variety of insect pests of field crops, vegetables and fruit orchards. Because of its voracious feeding on soft bodied insects e.g., aphids, caterpillars, leafhoppers, psyllids, mealybugs, white flies, thrips, insect eggs, spiders and mites, it is considered as an important component of IPM program (Rashid *et al.*, 2012).

The adult is non-predacious and only feeds on honeydew and pollen. It has high reproductive potential and long oviposition period (Dean and Satasook, 1983). Typical adults are 2 cm long with long net like wings, slender pale green bodies and

golden eyes. Female lays oval green eggs and are placed singly at the top of a thin “hair-like” support or filament.

Larvae are “alligator” shaped with long forceps-like curved tubular mandibles and have colorations ranging from grey to brown. The tubular mandibles inserted into insect body and suck the insect contents (blood) and probably for that nature it is known as “aphid lion”. Atlihan *et al.* (2004) noticed that as prey densities increased, *C. carnea* larvae increased its food consumption. It shows higher predation on older larval stages than younger ones. During development, each larva of *C. carnea* consumed an average of 732.35 eggs of *Corcyra cephalonica*, 662.53 eggs of *Heliothis armigera*, 419.18 *Aphis gossypii*, 409.55 neonates of *H. armigera*, 329.70 pupae of *Bemisia tabaci* and 288.45 nymphs of *Amrasca biguttula* (Balasubramani and Swamiappan, 1994). Pupa is formed inside a spherical silken cocoon that is attached to vegetation.

C. carnea can successfully be reared on eggs of different hosts (Morrison *et al.*, 1975; Gautam, 1994). However, eggs of *S. cerealella* proved to be the best larval diet. *C. carnea* larvae completed their

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life span in 13.9 days with just 15% mortality, when fed on eggs of *S. cerealella* eggs. The pre-oviposition period was 3.4 days and total eggs laid were as high as 713 (Syed *et al.*, 2008).

Depending upon the environmental conditions and food of the female, the eggs hatching period can vary from 2-3 days, larval period last for 2-3 weeks and passes through three instars. Patel and Vyas (1985) studied the biology of predator *Chrysopa scelestes* in the laboratory using eggs of *Corcyra cephalonica*. The egg period averaged 2.96 days. The three larval instars averaged 2.26, 2.14 and 2.30 days, respectively and complete larval stage averaged 6.77 days. The full fed larvae pupate in a spherical silken cocoon usually attached to vegetation. The pre-pupal and pupal stages last about 5-8 days. Rasheed (1991) observed the average period of egg, larval and cocoon stages of green lacewing feeding on *Amrasca devastans* were 3.18 ± 0.13 , 9.5 ± 0.22 , 13.8 ± 0.13 days, respectively.

The efficiency of lacewings for management of pests can be affected by many factors like pest type, distribution of the pest (*e.g.*, within and among plants), weather, crop, number of predators released, stage of predator released and the predator/prey ratio. Effectiveness of *C. carnea* as biological control agent has been demonstrated in field crops, orchards and in green houses (Hagley and Miles, 1987).

The predation phenomena of the *C. carnea* are sometime not as simple as mentioned above but change with varying prey densities. It has been observed with many insect and small animal predators that when prey population increased, prey consumption also increased and consumption rate is the function of food density. Such a changing behaviour ultimately affect the predator's release pattern in a bio- control program and needs to be studied for better understanding under different ecosystems. Therefore, a study was designed to evaluate the predation rate of laboratory reared *C. carnea* on *Sitotroga* eggs with the following objectives: (i) to determine predation rate of all larval instars under nine prey density levels; (ii) to study effect of prey density on the biology of *C. carnea* and (iii) to validate predation rate using type II functional response model.

MATERIALS AND METHODS

Studies were carried out at Entomological Research Laboratories, Agricultural Research Institute, Dera Ismail Khan during the year 2009. The experiment was designed to test the effect of prey densities on prey consumption and biology of *C. carnea* under controlled environmental conditions. *S. cerealella* eggs were used as prey for *C. carnea*. *S. cerealella* and *C. carnea* cultures were maintained in the laboratory under controlled environmental conditions.

Sitotroga cerealella culture

S. cerealella culture was maintained on whole wheat grain. The wheat grains were sterilized by dipping in boiling water for 5 min and then air drying. The grains were kept in freezer (0°C) for 2 hours and stored for culture in refrigerator. The baker's yeast (5g) was added to 700g of sterilized wheat grains and kept in plastic jars with lid lined with muslin cloth for ventilation. *S. cerealella* freshly laid eggs (3g) were added to the culture jar. One jar was prepared every day to collect the eggs. The emerging young larvae were fed on grains and adults were collected after 30 days with the help of power operated aspirator in a collecting glass jar and transferred to an adult jar with a fine mesh (size 40) on one side to facilitate the egg laying. The jars containing adults were placed on its side with mesh down on trays containing laundry starch to receive eggs, which were collected by sieving the starch in 100 mesh sieve. The freshly collected eggs were used for the experiment and recycle of the culture.

Chrysoperla carnea culture

C. carnea culture was maintained in the laboratory by placing eggs of *C. carnea* and 1000-1500 *Sitotroga* eggs in crystal clear gelatin capsule of 500 mg size. The eggs of *S. cerealella* were sufficient for lacewing larvae to survive till pupation. The capsules were de-caped as larvae turned into pupae and observed daily for adult emergence. Adults emerged from the pupae in the de-caped capsules were shifted daily to 30 × 30 × 30 cm plastic cage with mesh on one side. The adult diet was prepared by mixing 70 ml of water, 25 ml of honey and 5g of yeast. The adults of *C. carnea*

were fed on diet every morning by spotting the viscous diet solution on a plastic sheet measuring 5×5 cm. The inside top of the cage was lined with black card sheet to facilitate the *C.* for egg laying. The sheet was removed every day and eggs on sheet were taken by cutting the egg stalk with the help of sharp razor blade. The environmental conditions of insect rearing rooms were maintained at 25±3°C and 65±5% RH and 10:14 light: dark regime.

Experimental procedure

The experiment was designed in a completely randomized (CRD) design. There were nine (food densities) treatments, viz., 20, 30, 40, 50, 60, 70, 80, 90, 100 *Sitotroga* eggs/insect/day. Each treatment was replicated three times. Each replication has three samples (insects). Freshly harvested *C. carnea* eggs were placed in 9cm petri dish sealed with parafilm to avoid desiccation and were observed daily.

Newly emerged larvae of *C. carnea* were transferred into 500mg capacity crystal clear gelatin capsules with counted number of fresh *S. cerealella* eggs for each food density/ replication. The larvae of *C. carnea* were transferred very carefully to the capsules with the help of fine camel hair brush. Capsule contents were observed under stereo microscope every day to find out the number of unconsumed eggs and any change in larval biology i.e., molting or pupation etc. The numbers of unconsumed eggs were subtracted from the total number of offered eggs (prey density) and data were recorded on daily basis. Biological parameters like, the duration of development of each larval instar, pupation, adult emergence and mortality occurring in each treatment was recorded daily in all prey densities. After pupation, each pupa was observed for adult emergence and recorded. The emerging adults were transferred to a 10 l plastic jar. The jars were placed vertically and upper half was lined with black card sheet to collect eggs and the lid was replaced with muslin cloth covering for ventilation etc. The adults were fed daily on thick viscous solution of water + honey + yeast. The adult's jars were observed every 24 h for egg laying. The black card sheets with eggs on were removed carefully and eggs were removed with sharp razor blade, counted and recorded daily. This practice was

continued till the death of last adult *C. carnea*. The mortality was recorded and each dead insect was observed under stereo microscope for the sex orientation. After the death of all insects in each cage average lifespan of all insects was calculated (from the daily observation data) and recorded.

Analysis of the data was done with T-Test, ANOVA, F-test by employing software MSTATC and means were separated with least significant difference test (LSD) for the daily consumption data, whereas following functional response model (Holling, 1959) was fitted and observed and projected values were plotted against prey density to validate hypothesis.

$$f(R) = \frac{aR}{1 + ahR}$$

Where *f*, intake rate; *R*, food or rate at which the consumer encounters food items; *a*, per unit of food density is called the attack rate; *h*, the average time spent on processing of food item is called the handling time.

Handling time

Handling time is defined as the time spent searching, catching, and consuming each prey item plus the time spent preparing to search for the next prey item, and was calculated by starving the *C. carnea* larvae for 15 min and allowed in a petridish with *S. cerealella* eggs to consume. The moment the larvae caught the eggs stop watch turned on and after consumption till its reach to next egg, the time was measured and recorded. Six larvae observed three times every day till pupation.

RESULTS

Effect of food density on

C. carnea consumption

The larvae of *C. carnea* responded to increasing prey densities with increasing food consumption and older larval stages displayed a higher rate of predation than younger ones (Table I). The consumption rate increased progressively during each day. During 8th day in 80 eggs/day treatment consumption rate decreased from 100.00 to 77.78 because in this treatment most of the larvae

Table I.- Effect of various food densities on the food consumption of *C. carnea* at various intervals (days).

Treatments	Number of eggs consumed						
	2 nd day	3 rd day	4 th day	5 th day	6 th day	7 th day	8 th day
20 eggs	16.42 f	19.42 g	18.33 g	18.53 h	18.33 i	18.92 i	18.78 i
30 eggs	27.11 e	29.08 f	27.69 f	28.44 g	29.89 h	29.80 h	30.00 h
40 eggs	37.00 cd	34.88 ef	36.00 e	38.66 f	40.00 g	39.77 g	40.00 g
50 eggs	33.39 d	41.77 e	45.78 d	46.64 e	46.11 f	45.44 f	49.67 f
60 eggs	31.78 de	53.17 e	50.89 d	53.78 d	60.00 e	60.00 e	59.89 e
70 eggs	43.00 c	59.33 d	61.77 c	62.66 c	70.00 d	69.67 d	70.00 d
80 eggs	49.22 b	63.75 cd	65.11 c	71.44 b	45.67 c	79.77 c	79.55 c
90 eggs	58.78 a	75.00 b	77.11 b	76.77 b	90.00 b	89.55 b	90.00 b
100 eggs	63.66 a	84.11 a	92.66 a	83.67 a	99.33 a	100.0 a	77.78 a
LSD	6.107	7.091	5.625	6.137	4.804	4.742	1.406
CV	8.81	8.01	6.15	6.64	4.72	4.63	1.36

Each value is a mean of three replicates.

Means followed by the same letters are not significantly different from each other at α (0.05)

Table II.- Effect of food density on pre-oviposition period, total reproductive days, total number of eggs and male female ratio of *C. carnea*.

Treatments	Pre-oviposition period (days)	Reproductive days	Total eggs laid	Eggs/Female	Male:Female
20 eggs	9	2	14	7.0	8:2
30 eggs	10	2	12	6.0	8:2
40 eggs	9	2	24	8.0	12:3
50 eggs	8	3	60	15.0	12:4
60 eggs	6	4	92	15.3	10:6
70 eggs	5	6	408	25.5	7:16
80 eggs	4	9	672	42.0	8:16
90 eggs	5	6	721	45.0	10:16
100 eggs	7	5	297	29.7	12:10

Each value is mean of three replicates.

had gone into pupation while in other treatments maximum food consumption was recorded during 8th day.

Biological parameters

Food density had pronounced effects on the biological parameters of *C. carnea*. Shortest pre-oviposition period was recorded in 80 eggs/day treatment, whereas; longest pre-oviposition was observed in case of 50 eggs/day treatment. Similar trend was also observed in case of reproductive days. On an average, a single female laid maximum number of eggs when fed on 80 eggs/day and minimum number of eggs were recorded when it was fed on 50 eggs/day. While converting into the total number of eggs per female it was noticed that

maximum number of eggs were laid by female *C. carnea* when offered 90 eggs per day, whereas; minimum number of total eggs were recorded when it was reared on 40 and 60 eggs/day.

The maximum number of male was noticed in 40 eggs and 50 eggs/day treatments whereas; minimum number of male was recorded in 70 and 80 eggs/day treatments (Table II).

Duration of developmental stages

Food density directly affected the 3rd instar larval duration, pupal period and adult longevity of *C. carnea*. Increased prey densities reduced developmental time and mortality rate of *C. carnea* (Table III). Maximum duration of larval period was recorded in 20 eggs/day treatment whereas;

Table III.- Effect of food density on duration (in days) of different larval instars, pupal period, adult and total longevity (days) of *C. carnea*.

Treatments	1 st instar	2 nd instar	3 rd instar	Pupal period	Adult longevity	Total longevity
20 eggs	5.78 ^{NS}	2.056 cde	9.00 a	10.22 a	0.72 e	27.78 a
30 eggs	5.67	1.556 de	5.56 b	9.89 a	5.00 d	27.67 a
40 eggs	5.56	0.889 e	5.67 b	6.89 b	6.56 cd	25.56 bc
50 eggs	5.56	2.111 bcde	3.50 c	7.39 b	9.67 bc	28.22 a
60 eggs	5.56	2.889 abcd	1.78 d	6.67 b	9.78 bc	24.67 c
70 eggs	5.67	3.667 ab	1.00 d	6.22 b	10.56 a	27.11 ab
80 eggs	5.56	3.889 a	0.89 d	6.89 b	10.67 a	27.89 a
90 eggs	5.78	3.444 abc	1.11 d	6.78 b	9.33 ab	26.44 abc
100 eggs	5.78	3.33 abc	0.56 d	7.22 b	8.55 abc	25.44 bc
LSD		1.576	1.683	1.488	2.473	1.850
CV	6.48	34.39	30.11	11.35	18.69	

Each value is a mean of three replications.

NS, Non significant.

Means followed by the same letters with in a column are not significantly different from each other ($\alpha < 0.05$, LSD test).

minimum duration was recorded in 100 eggs/day treatment. In case of pupal period, it was shortest in 70 eggs/day treatment whereas; longest pupation period was recorded in 20 eggs/day treatment. Longest adult longevity was observed in 80 eggs/day treatments while shortest adult longevity was noticed in 20 eggs/day treatments. Maximum total longevity was recorded in 50 eggs/day treatment whereas; minimum total longevity was observed in 60 eggs/day treatment.

Cumulative adult mortality

Figure 1 showed that the increased prey density declined mortality rate and increased the longevity of *C. carnea*. The mortality of the predator started from day 18 and ended on day 33. During day 18, mortality had started in 20 eggs/day treatment, whereas; in case of 50, 80 and 100 eggs/day treatment mortality of all insects occurred on day 33.

Functional response

The statistical analysis (Table I) showed that prey consumption by larvae of all age group is a function of prey densities and were significantly positively correlated. The density responsiveness exhibited by all age group larvae and two larval instars of *C. carnea* to varying prey (eggs) densities depicts the type II functional response described by Holling (1959). The prey consumed daily by per *C.*

carnea larvae increased rapidly with initial increase in prey density, and thereafter increased at a decreasing rate (Fig. 2A-G). The increase in the number of eggs eaten by *C. carnea* at high prey density may be the result of several factors acting simultaneously.

It was noticed from the data that younger *C. carnea* larvae consumed less food in all prey densities than older larvae, probably due to smaller in size larvae has less mobility and prey handling efficiency than a larger sized larvae or older larvae.

DISCUSSION

Results of the present studies indicated that prey density had remarkable effect on consumption rate, development, mortality and fecundity of *C. carnea*. Larvae of *C. carnea* responded to increasing prey densities with increasing food consumption rate. Older larval stages displayed a higher rate of predation than younger ones. The above findings were in conformity with Zheng *et al.* (1993) who reported that individual lacewing larvae provided with higher number of Mediterranean flour moth (*Anagasta kuehniella*) eggs had a significantly higher feeding potential.

The total number of *S. cerealella* eggs consumed by *C. carnea* from 1st instar larvae up to pupation increased with increasing prey density up to 100 eggs at which highest consumption was

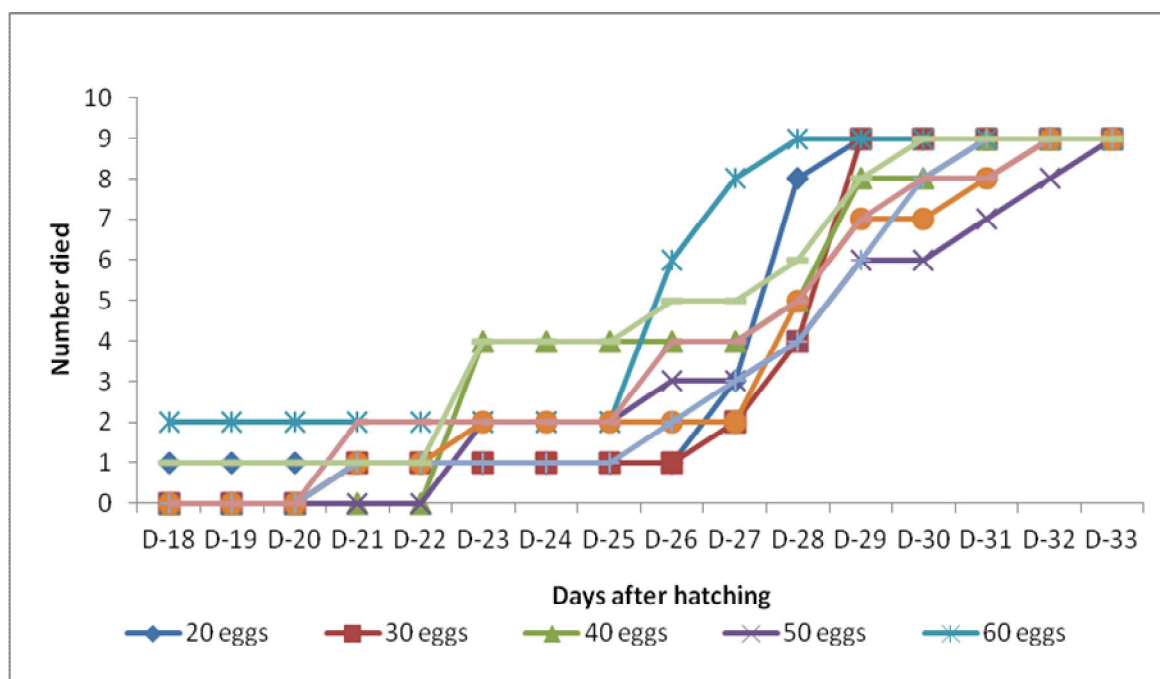


Fig. 1. Cumulative mortality of *C. carnea* as affected by different food densities

recorded. Similar results were shown by Hassanpour *et al.* (2009) who studied functional response of three larval instars of *C. carnea* on adult females of *T. urticae*. Results of the present studies revealed that the larvae of *C. carnea*, especially the last instar, have a good predation potential and probably its larger size facilitated its increased dietary requirement resulted in more prey consumption than 2nd and 1 instars. These results are in accordance with Klingen *et al.* (2009) who studied the predation rate of *C. carnea* on eggs and first instar larvae of the lepidopterous species *Mamestra brassicae* (L.) including the prey's influence on survival and development. In both cases the daily predation rate of *C. carnea* increased slowly during the first two instars and reached a peak in the third larval instar. During the third instar 87% and 85% of the total numbers of *M. brassicae* eggs and larvae were consumed.

In the present trials, it was noticed that although, *C. carnea* larvae completed development in each of the seven prey densities, increase in prey density reduced development time and mortality

rate. Similar results were shown by Pooja *et al.* (2005) who recorded the biology of *C. carnea* (Stephens) on different host insects. Shortest adult period (36.67 days) of *C. carnea* was recorded when fed on unsterilized eggs of *Corcyra cephalonica* while longest adult period of (49.67 days) was recorded on neonates of *Helicoverpa armigera*. The maximum larval period (15.33 days) were recorded on neonates of *Clostera fulgurita* and shortest larval period (6.17 days) recorded on sterilized eggs of *C. cephalonica*. It was also observed that development of *C. carnea* larvae on eggs of different hosts required less duration than the neonates of different lepidopterans.

Prey density had a considerable effect on fecundity of female *C. carnea*. This was in conformity with Atlihan *et al.* (2004) who reported that effect of different prey densities on fecundity was lowest for females (*C. carnea*) fed as larvae with 5 prey per day and increased with increasing prey density. A smaller intrinsic rate of increase was due to the fact that the population fed at a low prey density had a prolonged developmental time, higher

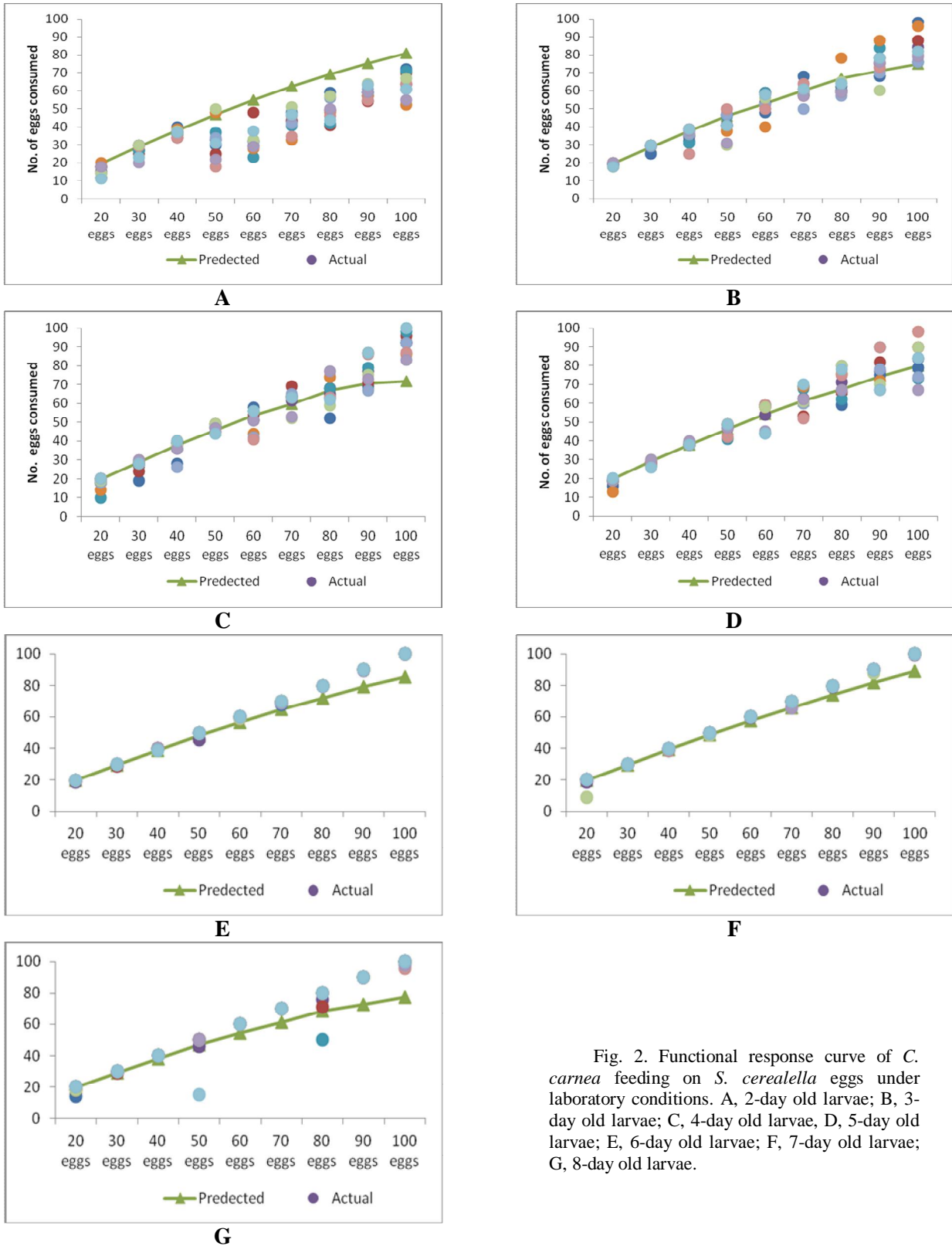


Fig. 2. Functional response curve of *C. carnea* feeding on *S. cerealella* eggs under laboratory conditions. A, 2-day old larvae; B, 3-day old larvae; C, 4-day old larvae; D, 5-day old larvae; E, 6-day old larvae; F, 7-day old larvae; G, 8-day old larvae.

mortality rate in immature stages as well as a low daily rate of progeny.

Results of the present study revealed that *C. carnea* has considerable potential for biological control program, the present studies give a rough estimate of release rate on the bases of prey density in the field or the infestation level. The prey density not only affects the prey consumption but also alter the male female ratio, which ultimately most likely to affect the future rate of increase of predator and subsequent predation as well. The findings still need a further confirmation and in detail studies at field level considering larger varying ecological factors in views.

Type II functional response model best fitted to the data obtained during the present study i.e. predation of *C. carnea* on *S. cerealella* eggs, which is the most commonly observed model in predators including insects (Biao *et al.*, 2008; Iason *et al.*, 2002). The predator larvae showed an increasing trend with increase in prey density, however, the instantly increase decreased at higher prey density which coincide with the findings of (Dhuyo and Soomro, 2008; Saleh *et al.*, 2010). Present study gives some idea that how *C. carnea* larvae at its various developmental stages responds to changing prey density under laboratory conditions. Although, the area was small and limiting the predator's movement and the prey egg was in immobile stage. Since field conditions may affect predation potential, behaviour, searching rate and handling time etc. of the predator, it is utmost important to couple the lab studies with field data by some future researcher for more realistic recommendations on field release rate, and forecasting the pest status using the predator-prey density.

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