

**Biological parameters and feeding efficiency of *Chrysoperla Carneastephens* (Neuroptera: Chrysopidae) feed on Citrus mealy bug *Planococcus Citri* (Risso) (Hemiptera: Pseudococcidae) under controlled conditions**

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**Abstract**

Experiment was conducted to check the biological parameters and feeding efficiency of *Chrysoperla carnea* fed on *Planococcus citri* mealy bug under controlled environment (24±1 °C with 60 ± 5 % RH), during 2018. Results regarding developmental time indicated that egg incubation, larva and pupal periods were 3.60±0.1, 9.65±0.19 and 6.01±0.17 days respectively. Apparent mortality (100qx) was maximum (11.11) for pupa and minimum (5.00) was during 3<sup>rd</sup> instar. Life expectancy (ex) for egg phase was highest (5.69) and lowest for pupal phase (1.88). Killing power (K-value) was found minimum (0.02) for 1<sup>st</sup> and 3<sup>rd</sup> instar while maximum (0.2) was recorded for adult stage. Feeding on artificial diet consisted of honey + sugar + Yeast + honey, the pre-oviposition, oviposition and post oviposition periods recorded for *C. carnea* were 8.2±0.41, 31.4±0.50 and 5±0.30 days, respectively. First and last mortality were observed at the age of 13 and 45 days while fast step down decrease was observed after 27 days when fed on the same food. Total number of eggs laid per female were 341.2±6.90 eggs. Main feeding efficiency and efficiency per day feeding on two different host stages were also found for *C. carnea*. The maximum (233.85±4.43 and 75.25±3.13) crawlers and adult were consumed by larva stage while minimum (27.10±1.35 and 8.70±0.48) crawlers and adult were consumed by 1<sup>st</sup> instar. The consumption day<sup>-1</sup> were highest (37.06± 0.28 and 9.67± 0.68) for 3<sup>rd</sup> instar while the 1<sup>st</sup> instar consumed lowest 9.67±0.56 and 2.89±0.21 crawlers and adult respectively.

**Keywords:** Green lace wing, *Chrysoperla carnea*, Citrus mealybug, *Planococcus citri*, biological attributes, feeding efficiency

**Introduction**

The green lacewing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) is a cosmopolitan predator found in a spacious range of agricultural habitats. Previously, many researchers have deliberated the potential of predators in managing the insect population in the fields. The introduction of *C. carnea* in pest management program can increase the success against pest up to one third [25]. The larval stage of green lacewing is voracious and effective bio-control agent for various agricultural insect pests [11]. Single larva consumes up to 500 aphids in its entire life and the green lacewings have proved their importance in the natural control of many soft body insect pests [12].

Being *C. carnea* a generalist predator of various insect pests, it also feed on different species of Mealy bugs. A lot of work has been carried out on *C. carnea* biology, feeding potential and effect of artificial diets. The same parameters further needed a consolidated study on various natural foods including mealy bugs to elaborate most effective rearing tools of *C. carnea*. The same was considered by previous

researchers as [22] reported feeding potential of *C. carnea* against different species of Mealy bug. Larvae of *C. carnea* are highly voracious predator of mealy bug's crawlers particularly first instars nymphs [13, 18]. *C. carnea* reported from Cotton crop feeding on different insect pests such larvae feed on eggs, small larvae of lepidopteron and premature stage of sucking insect [19].

Citrus mealybug, *Planococcus citri* (Risso) (Hemiptera: Pseudococcidae) is an important polyphagous pest attacking several horticultural and agricultural crops [6] including citrus and mango [1], Coffee [20] and Grape vine [5]. In Pakistan citrus mealybug is a major pest of citrus plants and highly sporadic pest attacks a wide variety of citrus cultivars [9]. It reduced the quantity and as well as fruit quality by sucking at base of fruit stalks, calyx and fruits, which results in dropping of fruits. In addition, it causes the accumulation of honeydew and sooty mould (a fungus) growth on different parts of plants such as on fruits and leaves [24].

Chemical control of mealy bug is more in practice worldwide. The frequent and wide use of insecticides is not

only incessantly harm the environment but also develop the resistance in many insects against pesticides [19]. To overcome these problems, it is highly necessary to search an alternative method for the control of insect pests.

Keeping in view the importance of green lacewing as bio-control agent the present study was intended to check the biological attributes and feeding efficiency of *Chrysoperla carnea* against Citrus mealy bug *Planococcus citri*.

**Materials and Methods**

The present study was conducted under laboratory condition of 24±1°C with 60 ± 5% relative humidity at Insectary, Insect Pest Management Program, Department of Plant and Environmental Protection, National Agricultural Research Centre, Islamabad. Before conducting experiments stock culture of predator was maintained on artificial diet containing yeast + sugar + honey + water (2:1:1:6) while culture of its host insect, Citrus mealy bug was maintained on brinjal plant.

**Biological parameters, age and stage specific life table parameters of immature stages of *C. carnea* feed on *P. citri* mealy bug**

100 eggs of the same age were collected from stock culture of *C. carnea* maintained on *P. citri*. 10 eggs of the same age were kept in petri dishes and the experiments were replicated 10 times. Percent egg hatching was calculated in each replicate. After hatching, first instar larvae of *C. carnea* were separately transferred in transparent larval rearing vials. The vials were covered with muslin cloth tightly. Sufficient amount of mix nymph instars of citrus mealy bug crawlers were provided on infected leaves of brinjal inside the vials on daily basis. In each vial old diet was replaced with new diet after 24hr. This process was continued unless all larvae reached pupal stages.

The following method was used for construction of life table parameters

- Developmental duration and molting of each larvae instar
- Pre-Pupal and pupae duration(days)
- Duration from egg to adult emergence(days)

**Data analysis**

The data on the developmental duration of different stages were subjected to analysis of variance with two factor factorial design using Statistix 8.1 package. Means were compared using LSD) test at 5% level of significance using two way ANOVA.

**Age Specific Life-Table parameters calculation**

Alive and dead insects emerged from 100 eggs were documented day by day. The following methods were used for construction of life table parameters of *C. carnea*.

Mortality during age (x) is formulated in equation  $100 q_x = (d_x/l_x) \times 100$

Whereas

100 q<sub>x</sub> represents Mortality during age

x represents Age of insect (days)

l<sub>x</sub> represents insects lived at start of interval

d<sub>x</sub> represents insects died during interval

e<sub>x</sub> represents average life left over for individual of age x, and was calculated by  $e_x = T_x / l_x$

Where, L<sub>x</sub> and T<sub>x</sub> were calculated as;

L<sub>x</sub> represents Live insects among age x and x +1, and will be determined by  $T_x = l_x + (l_{x+1}) + (l_{x+2}) + \dots + l_w$ , while, l<sub>w</sub> is the last age interval.

**Stage Specific Life-Table**

The following standard heads were used to make stage specific life table.

l<sub>x</sub> shows number of survived insects at initial of age interval (x)

x represents insect age in days

While, d<sub>x</sub> represent mortality in particular stage

The following life table parameters was computed from the above table's data.

**(i) Apparent Mortality (100 q<sub>x</sub>)**

It shows the dead insects percentage were determined by the formula

$$\text{Apparent Mortality} = \frac{d_x}{l_x} \times 100$$

**(ii) Survival Fraction (S<sub>x</sub>)**

The apparent mortality data was applied for the approximation of the stage specific survival fraction (S<sub>x</sub>) of each stage with the equation:

$$S_x \text{ of particular stage} = \frac{\text{lx of succeeding stage}}{\text{lx of that particular stage}}$$

**(iii) Mortality Survivor Ratio (MSR)**

This is the increase in population of insect and was determined;

$$\text{MSR} = \frac{\text{Mortality in particular stage}}{\text{lx of subsequent stage}}$$

**(iv) Indispensable Mortality (IM)**

IM = total adults emerged x (MSR of specific stage of insect)

**(v) K-values**

K values were computed by the difference in consecutive values for (log l<sub>x</sub>). The mortality of total generation was determined by addition up the k values of different growth stages of insect, which is selected as "K".

$$K = k_E + k_{L1} + k_{L2} + k_{L3} + k_{L4} + k_{PP} + k_P$$

Where k<sub>E</sub>, k<sub>L1</sub>, k<sub>L2</sub>, k<sub>L3</sub>, k<sub>L4</sub>, k<sub>PP</sub>,

And k<sub>P</sub> are the k-values at egg, 1<sup>st</sup> to 4<sup>th</sup> instars, larvae and pupal stages.

**Feeding efficiency of larval instars of *C. carnea* feed on different stages of *P. citri* Mealy bug**

To find out the feeding potential and developmental time of larval instars of *C. carnea*, a total of 40 newly emerged larvae were collected from stock culture, shifted into transparent vials separately and covered with fine muslin cloth. 20 newly emerged larvae were provided mix (1<sup>st</sup> to 3<sup>rd</sup>) instar's crawler mealy bug on daily basis. Initially the first instar larvae were provided 20 mix crawlers and 5 adult mealy bugs in each vial. Number of consumed, dead and unconsumed mealy bugs were counted with binocular microscope after each 24 hours and replaced with fresh diet at time of observations in each vial. The number of crawlers was increased as the age of the larvae proceeds. The

experiment was continued till all larvae entered in to pupal stage. If at any stage the larvae were found dead they were replaced with new larvae from stock culture.

Data was recorded on the following parameters

- Number of consumed crawlers per each larval instar and total larvae
- Number of consumed adult mealy bug per each larval instars/ larvae
- Number of consumed crawlers and adult mealy bug per each larval instars and larvae/ day

**Data analysis**

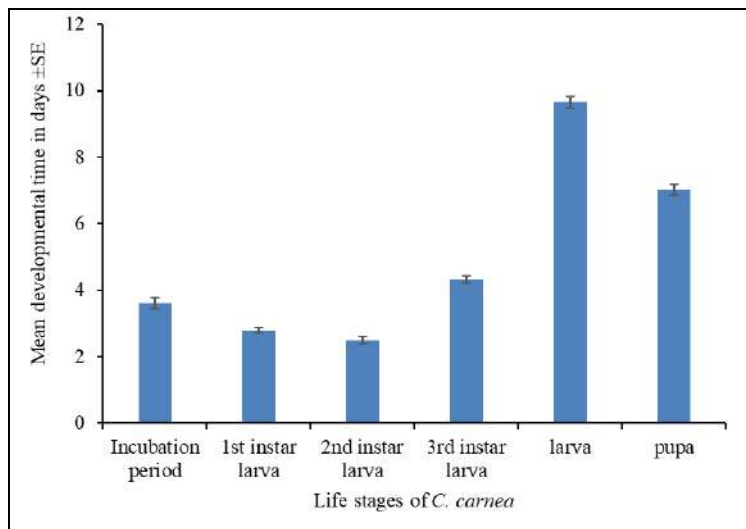
The data on the developmental duration of different stages were subjected to analysis of variance with two factor

factorial design using Statistix 8.1 package. Means were compared using LSD) test at 5% level of significance using two way ANOVA.

**Results and Discussion**

**Developmental duration in days of *C. carnea* larval instars feed on Crawlers, Citrus Mealy bug, *P. citri***

The results showed (Fig-1) the mean developmental time of life stages of *C. carnea*. The egg incubation period was 3.60±0.1 days, 1<sup>st</sup> instar larval was 2.80±0.08 days, 2<sup>nd</sup> instar larval was 2.50±0.11 days, 3<sup>rd</sup> instar larval 4.31±0.11 days, larval was 9.65±0.19 days and pupal duration was 6.01±0.17 days.



**Fig 1:** Developmental duration in days of *C. carnea* larval instars feed on Crawlers, Citrus Mealy bug, *P. citri*

**Age and stage specific life table parameters of immature stages of *C. carnea* feed on *P. citri* mealy bug**

Results pertaining in Table 3 revealed that the apparent (noticeable) mortality during egg, first, second, third larval instars, pre-pupal and pupal phases were 9, 5.49, 6.97, 5, 5.26 and 11.11, respectively. The survival fraction (Sx) recorded during these were recorded 0.91, 0.94, 0.93, 0.95, 0.94 and 0.88, respectively. Similarly mortality survival ratio (MSR) noted during the same phases were 0.098, 0.058, 0.075, 0.052, 0.055 and 0.125, respectively. The

recorded Indispensable mortality (IM) during egg phase, first, second, third larval instars were 6.272, 3.712, 4.8, 3.328, 3.52 and 8, respectively. It is obvious from the results that life expectancy(ex) for egg phase was highest (5.69) and lowest for pupal phase (1.88). The same was noticed 5.15, 4.39 and 3.65, respectively, during first second, third larval instars. The k-value at egg phase and pupal phase was highest (0.05) and lowest (0.02) at first and third larval instar, respectively. The total K value for the whole generation was 0.2.

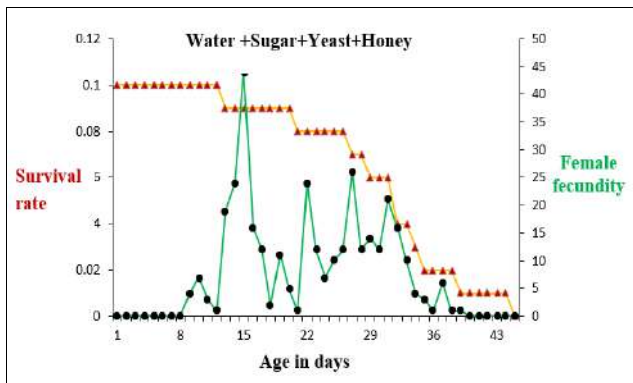
**Table 3:** Age and Stage specific life table parameters of immature stages of *C. carnea* feed on *P. citri* mealy bug

Stage X	Lx	Dx	Lx	100qx	Sx	Tx	MSR	IM	Log lx	Ex	k-values
Egg	100	9	95.5	9	0.91	569	0.098	6.272	2	5.69	0.05
1 <sup>st</sup> instar	91	5	88.5	5.49	0.94	469	0.058	3.712	1.95	5.15	0.02
2 <sup>nd</sup> instar	86	6	83	6.97	0.93	378	0.075	4.8	1.93	4.39	0.03
3 <sup>rd</sup> instar	80	4	78	5	0.95	292	0.052	3.328	1.90	3.65	0.02
Pre-pupa	76	4	74	5.26	0.94	212	0.055	3.52	1.88	2.78	0.03
Pupa	72	8	68	11.11	0.88	136	0.125	8	1.85	1.88	0.05
Adult	64	64							1.80		0.2

- Where lx = No. surviving at the beginning of the stage
- Dx = No. dying In each stage
- Lx = No. alive b/w age x and x+1
- 100qx = Apparent mortality
- Sx = Survival fraction
- MSR = Mortality/ survivor ratio
- IM = Indispensable mortality
- Ex = Life expectancy
- K = killing power

**Effect of artificial diet (honey + sugar + Yeast + honey) on developmental durations and reproductive potential of adult female *C. carnea***

The results revealed that pre-oviposition period was 8.2±0.41 days on artificial diet consisted of honey+sugar+Yeast+honey (Fig-2). Similarly oviposition and post oviposition periods recorded for *C. carnea* were 31.4±0.50 and 5 ± 0.30 days. It provided a male longevity of 27.4± 0.49 days and female longevity of 44.6±0.74 days. The results indicate that total number of eggs laid per female were 341.2±6.90 on this diet while the no of eggs laid per female per day was 10.87± 0.53.



**Fig 2:** Age and stage specific mortality of female *C. carnea* on artificial diet (days)

**Age and stage specific mortality and fecundity of *C. carnea* female feed on artificial diet**

The first mortality (dx) was observed at the age of 13 when fed on artificial diet containing Water + Sugar + Yeast + Honey (Fig. 2). The second and 3<sup>rd</sup> mortality were observed at the age of 21 and 27 days. After attaining the age of 27 days and further step down decreased were observed in sample population. The last mortality was observed at the age of 45 days in sample population. Similarly the first egg was laid by female at the age of day 9 and the maximum (44) eggs were laid at the age of 15 day of female and then step up and down increase and decrease were observed.

**Feeding efficiency of different larval instars of *C. carnea*, feed on *P. citri* mealy bug crawlers and adult stages**

Results pertaining in table-4 indicate that feeding efficiency of first larval instar of *C. carnea* were significantly different (27.10±1.35) and (8.70±0.48) when feed on crawlers and adults mealy bug, respectively. It further explains that the feeding efficiency of first larval instar was significantly different from all other instars of *C. carnea* when feed on two different stages of host insects. Feeding efficiency of second larval instar of *C. carnea* were statistically significant (46.9±1.82) and (20.10±1.30) from each other feeding on two different stages of the host. It was also significant from all other instars when this vary stage fed on adult mealy bugs. Significant difference was observed between and among the feeding efficiency of 3<sup>rd</sup> larval instar (159.75±4.3) and (46.45±2.8) and all the rest of larval stages except 2<sup>nd</sup> larval instar when it fed on crawlers and adults of mealy bugs, respectively. Feeding efficiency of larvae was also significantly different (233.85±4.43 and 75.25±3.13) from each other when it were fed on two

different host's stages.

**Table 4:** Mean feeding Efficiency±SE of *C. carnea* larval instars feed on two different stages (Crawlers and adult) of Citrus Mealy bug, *P. citri*

Developmental stage	Host stage (Mealy bug)	
	Crawler	Adult
	Mean feeding efficiency± SE	Mean feeding efficiency± SE
1 <sup>st</sup> instar	27.10±1.35e	8.70±0.48g
2 <sup>nd</sup> instar	46.9±1.82d	20.10±1.30f
3 <sup>rd</sup> instar	159.75±4.3b	46.45±2.8d
Larvae	233.85±4.43a	75.25±3.13c
LSD values ≤0.05	6.6914	

Mean with different lower case letters row and column wise are significantly different from each other (Two way ANOVA) using LSD test at p value less than ≤ 0.05

**Feeding efficiency per day of different larval instars of *C. carnea*, feed on *P. citri* mealy bug crawlers and adult stages**

The results indicate that feeding efficiency per day of first larval instar of *C. carnea* were (9.67±0.56 and 2.89±0.21) crawlers and adult mealy bug respectively (Table-5). It was significantly different from each other when feed on two different stages of host insect. It further indicates that that feeding efficiency per day of first larval instar was significantly different from all other instars of *C. carnea* except third larval instar when feed on adult mealy bug. Significantly different feeding efficiency was observed for 2<sup>nd</sup> larval instar (18.76±0.62 and 7.58±0.18) when fed on crawlers and adults of mealy bugs, respectively. It was also statistically significant from the rest of instars except larval stage when fed on adult mealy bug. Significant difference was observed between 3<sup>rd</sup> larval instars (37.06± 0.28 and 9.67± 0.68) feeding on tow host's stages and also among all other instars except 1<sup>st</sup> instar when fed on mix crawlers of mealy bug. The feeding efficiency per day of larval stage per day of *C. carnea* were (24.23± 0.35 and 7.22±0.29) crawlers and adult mealy bug respectively. The results further elaborate that feeding efficiency per day of larval stage was significantly different from all other instars of *C. carnea* except second larval instar when feed on adult mealy bug.

**Table 5:** Mean feeding efficiency per day ±SE of *C. carnea* larval instars feed on two different stages (Crawlers and adult) of Citrus Mealy bug, *P. citri*

Developmental stage	Host stage (Mealy bug)	
	Crawler	Adult
	Mean feeding efficiency/day ± SE	Mean feeding efficiency/day ± SE
1 <sup>st</sup> instar	9.67±0.56d	2.89±0.21f
2 <sup>nd</sup> instar	18.76±0.62c	7.58±0.18e
3 <sup>rd</sup> instar	37.06± 0.28a	9.67± 0.68d
Larvae	24.23± 0.35b	7.22±0.29e
LSD values ≤0.05	1.23	

Mean with different lower case letters row and column wise are significantly different from each other (Two way ANOVA) using LSD test at p value less than ≤ 0.05

## Discussion

The current sections have described the results and discussion regarding developmental time and feeding efficiency of predator *C. carnea* against Citrus Mealy bug *P. citri*. In addition studies were also conducted on the effect of artificial diets on adult female biology and reproductive potential for economically mass rearing of the predator under controlled conditions.

The results of the present investigation indicate that *C. carnea* successfully completed their life stages on *P. citri*. The larval duration was  $9.65 \pm 0.19$  days when feed on crawlers and adult mealy bug respectively. Previously, researchers have elaborated different developmental time for various stages of the predator, *C. carnea*.<sup>[21]</sup> reported completion of *C. carnea*' larval stage in 8.2, 10.0 and 12.0 days when fed on *S. cerealella* eggs, *Chiloinfuscatellus* and *Aleurolobus Barodensis*, respectively. Concisely, <sup>[4]</sup> reported that total larval duration of *C. carnea* was  $6.92 \pm 0.13$  on *C. cephalonica* <sup>[2]</sup> found that total larval developmental time was ( $15.13 \pm 0.35$ ) and ( $13.60 \pm 0.31$ ) on *A. fabae* and *E. kuehniella* eggs <sup>[15]</sup>. Reported that total developmental duration from egg to adult emergence was 23.1 days when feed on *B. brassicae* aphids. Similarly, <sup>[16, 26]</sup> reported that *C. carnea* completed their duration from egg to adult emergence was 18.6 and 22.7 days. Differences in the developmental durations may be due to different host insects which were provided to the predator in experiments. Our results of life table coincided with that of <sup>[10]</sup> who reported that life expectancy (ex) were maximum (5.24) and minimum (1.24) days during egg phase and pupal phase when feed on *C. cephalonica* eggs. It were also in close conformation for maximum life expectancy during egg phase and minimum during pupal phase which indicated that the life expectancy was maximum at initial stage and with the passage of time the life expectancy stepped down till the final stages. During the course of the present study different artificial diet was tested consisted of water + sugar + Yeast + Honey, for its effect on developmental time and reproductive potential of adult female *C. carnea*. The present results revealed that pre-oviposition period was  $8.2 \pm 0.41$  days on artificial diet consisted of honey+sugar+Yeast+honey. Similarly oviposition and post oviposition periods recorded for *C. carnea* were  $31.4 \pm 0.50$  and  $5 \pm 0.30$  days which provided a male longevity of  $27.4 \pm 0.49$  days and female longevity of  $44.6 \pm 0.74$  days. The results further indicate that total number of eggs laid per female were  $341.2 \pm 6.90$  on this diet while the no of eggs laid per female per day was  $10.87 \pm 0.53$ . Previously researchers including <sup>[23]</sup> found comparatively close relation regarding pre-oviposition, oviposition post-oviposition, fecundity and longevity periods when the insects were provided various artificial diets <sup>[17]</sup>. Determined the impact of six artificial diets and showed that the mixture of honey, yeast and extract of *A. kuehniella* eggs, provided promising results on biological aspects of the predator as compare to other tested diets <sup>[3]</sup>. Reported that for increasing the population of *C. carnea* in the agricultural ecosystems, artificial diets can be used in their habitats. They showed that by spraying artificial diets in agricultural ecosystems, adults of *C. carnea* can mate and lay eggs and control the pests better. The present result regarding feeding efficiency of larval stage of *C. carnea* were  $233.85 \pm 4.43$  and

$75.25 \pm 3.13$ . The results of the past workers <sup>[14]</sup> who results indicated that feeding potential was  $219.02 \pm 2.18$  on *P. solenopsis*. Feeding efficiency per day was also determined to be  $43.17 \pm 0.92$  when provided the same host insect. Similarly <sup>[8]</sup> reported that feeding potential was  $192.1 \pm 2.57$  and  $479.7 \pm 4.08$  on 3<sup>rd</sup> nymph stages of *S. graminum* and *B. tabaci* <sup>[7]</sup>. found that the predatory potential of the predator was higher in the older instars than the younger ones. The estimated handling time was somewhat lower for the third instar because of the higher prey consumption.

## Conclusion

It is concluded from results of the present study that *C. carnea* successfully completed their life cycle on citrus mealy bug fecundity rate per female was maximum ( $341.2 \pm 6.90$ ) and the larva was a voracious predator of mealy bug's crawlers and predator can be utilised for the management of citrus mealy bug as a part of successful IPM programme.

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