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Invitro assessment of green lacewing *Chrysoperla carnea* feeding performance on cotton aphid species

[a] Khalil Ahmed Memon, [a] Aslam Bukero, [a] Shafique Ahmed Memon, [b] Arif Ali, [a] Abdul Aziz Bukero, [c] Naimatullah Koondhar, [a] Fatima Imtiaz, [b] Gulkhanda Parwaiz

[a] Department of Entomology, Faculty of Crop Protection, Sindh Agriculture University Tandojam, Sindh, Pakistan,
[b] Department of Entomology, Faculty of Agriculture, Lasbela University of Agriculture, Water and Marine Science, Lasbela, Balochistan, Pakistan,
[c] Department of Plant Pathology, Faculty of Agriculture, Lasbela University of Agriculture, Water and Marine Science, Lasbela, Balochistan, Pakistan.

*Corresponding Author's Email Address: arifalirao@gmail.com

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ABSTRACT

Cotton is the most widespread profitable non-food crop in the world; in Pakistan it contributes more than 65% to the total foreign exchange earnings of the country. Cotton aphids reduced leaf area by 58% and shoot biomass by 45% so due to cotton aphid cotton reduce their yield and reduction of fiber quality. Cotton aphis has been resistance against number of synthetic pesticides because of their continuously uses. Natural enemies such as green lace wing is the most suitable alternate method to control aphid species, it's safe and clean methods for human being and environmental. To the management of aphid's species green lacewing (*Chrysoperla carnea*) has been used as biological control against, *Aphis gossypii, A. nerii, Urolecen carthemi*, and *Brevicoryne brassicae* at 26 ± 2°C with 65±5% humidity. The result indicated that maximum feeding efficiency was observed in 3rd instar 23.66 ± 0.6/day on *A. gossypii* followed by 2nd and 1st instars of *C. carnea*, respectively. Whereas, the highest per stage consumption rate was recorded in 3rd instar 91± 1.79/stage when reared on *A. gossypii* followed by 2nd and 1st instars of *C. carnea*, respectively. In the 1st, 2nd and 3rd larval instars of *C. carnea* preferred *A. gosypii* followed by *B. brassicae*, *U. carthemi* and *A. nerii*, respectively. The analysis of variance revealed that there were highly significant ($P<0.05$) difference in feeding efficiency in the larval instar of *C. carnea* on aphid species. The result further revealed that the development period (DP-day) and rate development (RD-day) significantly ($P<0.05$) different in larval instars and pupa of *C. carnea* on different aphid species. Similarly, maximum adult emergence % was observed on *A. gossypii* (92%), following by *B. brassicae* (87%), *U. carthemi* (84%) and *A. nerii* (80%). A statistically Significant ($P<0.05$), difference was observed among all of given treatment. This predator can be used to control aphis species in organic cotton.

Keywords: Green lacewing, natural enemies, *Aphis nerii*, *Urolecen carthemi*, *Aphis gossypii*, *Brevicoryne brassicae*.

INTRODUCTION: Cotton belongs to the genus Gossypium, referred as the prime fiber crop, has been grown in tropical, subtropical, and semiarid regions of the world (Tarazi *et al.*, 2019). Fiber as the major product of cotton has been extensively used in textile industry, and required millions of cotton bales yearly (Shahzad *et al.*, 2019). Utilization of cotton vegetable oil has been increased along with the demand of cotton seed meal for livestock feed. Pakistan economy is largely depends on agriculture sector which has contributed about 19.2% of the country's Gross Domestic Product (GDP), provided 38.5% employment, and the livelihood of 70% population depends on agriculture (Shahzad *et al.*, 2022). Cotton has an imperative role to boost the agriculture based economic growth of the country. Remarkably, 50% of the total industrial labor along with more than 60% of the total exports comes from cotton crop product chain (Abbas and Waheed, 2017). Pakistan is one of the leading countries of the production of cotton, exportation, and consumption in the world. Cotton is widely cultivated in several regions of Pakistan, in which Punjab and Sindh provinces are the main growing belts, and Punjab is the leading province in terms of the total cultivated area and the total production of cotton bales (Shuli et al. 2018). Numbers of insect pest has been attacked on cotton crops but cotton aphid (*Aphis gossypii*) is considered as a major pest of cotton worldwide. Nymphs and adults use their piercing-sucking mouthparts to feed on cotton plants which are causing substantial damages to stems and leaves. This feeding behavior results in significant nutrient loss, impacting overall plant health and productivity. Additionally, cotton aphids excrete honeydew and transmit viral diseases. Their short life cycle and rapid reproduction rate lead to multiple generations per year. Cotton producers must use efficient management methods since the combined impacts of these variables seriously impair cotton productivity and quality (Zhang et al., 2020). Pests pose a constant threat to human interests, and pesticides are the most widely used method for pest control. In 2022, total pesticides use in agriculture was 3.70 million tonnes (Mt) of active ingredients, marking a 4 percent increase with respect to 2021, a 13% increase in a decade, and a doubling since 1990, to manage harmful pests (FAO, 2021). However, pesticide use has numerous adverse effects on human health and the environment, often leading to pest resurgence and the unintended killing of beneficial organisms (Weathersbee and Mckenzie, 2005).

Biological control is an environmentally sound and economically efficient approach to pest management that utilizes natural enemies. These natural enemies are employed in various biological control strategies, including classical, augmentative, and inundative releases (Tauber et al., 2000). In recent years, green lacewings have shown compatibility as biological control agents for suppressing pest populations below economic thresholds. As a result, integrated pest management programs that incorporate biological control have been widely adopted to manage a variety of damaging pests in diverse crops worldwide (Canard et al., 1984). Lacewings are generalist predators and with a voracious feeding habit against eggs as well as mature and immature stages of soft bodied insect pests such as white flies, mealybugs, spider mite, thrips, leaf hopper, aphids, and caterpillars, that are released as an efficient bio-logical agent against phytophagous arthropods (McEwen, 2001). The predatory potential of C. carnea proceeding lettuce aphids has been examined (Shrestha, 2011), but as such there is no available information regarding prey preference of predator to F. occidentalis and *N. ribisnigri* which appear in lettuce field. The preference of predator to prey directly affects their controlling efficiency towards different prey species (Xu and Enkegard 2009), thus information about preference is very essential to evaluate the potential of predator in situation where different insect species are available in the crop (Enkegard et al. 2001).

OBJECTIVE: The present study is planned to determine the feeding efficiency of C. carnea against cotton aphid and it results was compared with, Akk aphid, safflower aphid, and cabbage aphid.

MATERIAL AND METHODS: The present experiment was carried out in the Bio-control Research Laboratory of Entomology Department, Sindh Agriculture University (SAU), and Tando jam. The newly emerged (n=500) larvae of *C. carnea* were collected from stock culture of laboratory. Natural diets was comprised of cotton aphid, akk aphid, safflower aphid, and cabbage aphid were collected from respective host plants. The natural diet (aphids) was provided to the larvae. There were four treatments including i.e. *A. nerii*, (T_1) *U. carthemitheo*,(T_2), *A. gossypii* (T_3), *B. brassicae* (T_4) each treatment replicated five times.

Experimental design: The experimental design was Complete Randomized Design (CRD). The temperature (26±2^{0}C) and relative humidity (65±5%) were maintained. After hatching green lacewing from eggs (n=100) larvae were shifted in new petri dishes. Each grub was feed by aphid species and data was collected after 24 h. The feeding efficiency, development period 1st, 2nd and 3rd instars (DP^{-days}) and all 3 instars rate development (RD^{-days}), was noted on daily basis. After completing larval stage pupal (n=50) were collected from treated larvae and data were subjected for statistical analysis by using one way annova with the help of (Statistix. 8.1) software. Means was compared with Tukey test (*P<0.05*).

RESULTS: The maximum feeding rate was recorded 9.46 ± 0.13/day on *A. gosypii* followed by 7.6 ± 0.41/day, 3.66 ± 0.18/day and 2.3 ± 0.13/day observed on *A. nerii*, *B. brassicae* and *U. carthemi*, respectively (table 1). The highest per stage consumption rate was recorded as 28.4 ± 0.4/stage when reared on *A. gossypii* followed by 22.8 ± 0.41/stage, 11 ± 0.55/stage and 4.6 ± 0.51/stage observed on *A. nerii*, *B. brassicae* and *U. carthemi*, respectively (table 2). It was observed that first instar larvae voraciously feed on *A. gossypii*, because it is small in size as compare to rest aphid species, easy in grasp, however, minimum feeding choice was recorded on *U. carthemi*, this species bigger in size as compare to rest of aphid species. The analysis of variance showed that there were highly significant (P<0.05) difference in feeding performance in the described instars on aphid species. The maximum devouring rate of second instar was seen 17.13 ± 0.73/day on *A. gossypii* followed by 10.1± 0.6/day, 8.33 ± 0.72/day and 7.9 ± 1.13/day were recorded on *A. nerii*, *B. brassicae* and *U. carthemi*, respectively (table 1). In the same way per stage feeding rate was observed in (table-2) when fed with *A. gossypii* 51.4 ± 2.2/stage on followed by 25 ± 2.17/stage, 20.2 ± 1.2/stage and 15.8 ± 2.27/stage observed on *B. brassicae*, *A. nerii* and *U. carthemi*, respectively. The same pattern of feeding efficiency of second instar was found as in the 1st instar on varoius aphid species.

Life Stages	*Aphis nerii* (T_1)	*Urolecen carthemi* (T_2)	*Aphis gossypii* (T_3)	*Brevicoryne brassica* (T_4)
First instar	7.6 ± 0.41b	2.3 ± 0.25cd	9.46 ± 0.13a	3.66 ± 0.18c
Second instar	10.1 ± 0.60b	7.9 ± 1.13cd	17.13 ± 0.73a	8.33 ± 0.72c
Third instar	12 ± 0.56cd	15.3 ± 1.39b	23.66 ± 0.6a	13.9 ± 0.93c

Table 1: Feeding efficiency per day of immature stages of *C. carnea* on selected aphid species under control conditions (temperature 26±2, relative humidity 65±5%). Values (mean ± SE) in given column letters are significantly different by Tukey test (p<0.05).

Life Stages	*Aphis nerii* (T_1)	*Urolecen carthemi* (T_2)	*Aphis gossypii* (T_3)	*Brevicoryne brassica* (T_4)
First instar	22.8 ± 1.24b	4.6 ± 0.51d	28.4 ± 0.40a	11 ± 0.55c
Second instar	20.2 ± 1.20c	15.8 ± 2.27d	51.4 ± 2.20a	25 ± 2.17b
Third instar	60 ± 2.79d	71 ± 1.79c	91.8 ± 8.36a	83.4 ± 5.64b

Table. 2. Feeding efficiency per stage of immature stages of *C. carnea* on selected aphid species under control conditions (temperature 26±2, relative humidity 65±5%). Values (mean ± SE) in given column letters are significantly different by Tukey test (p<0.05).

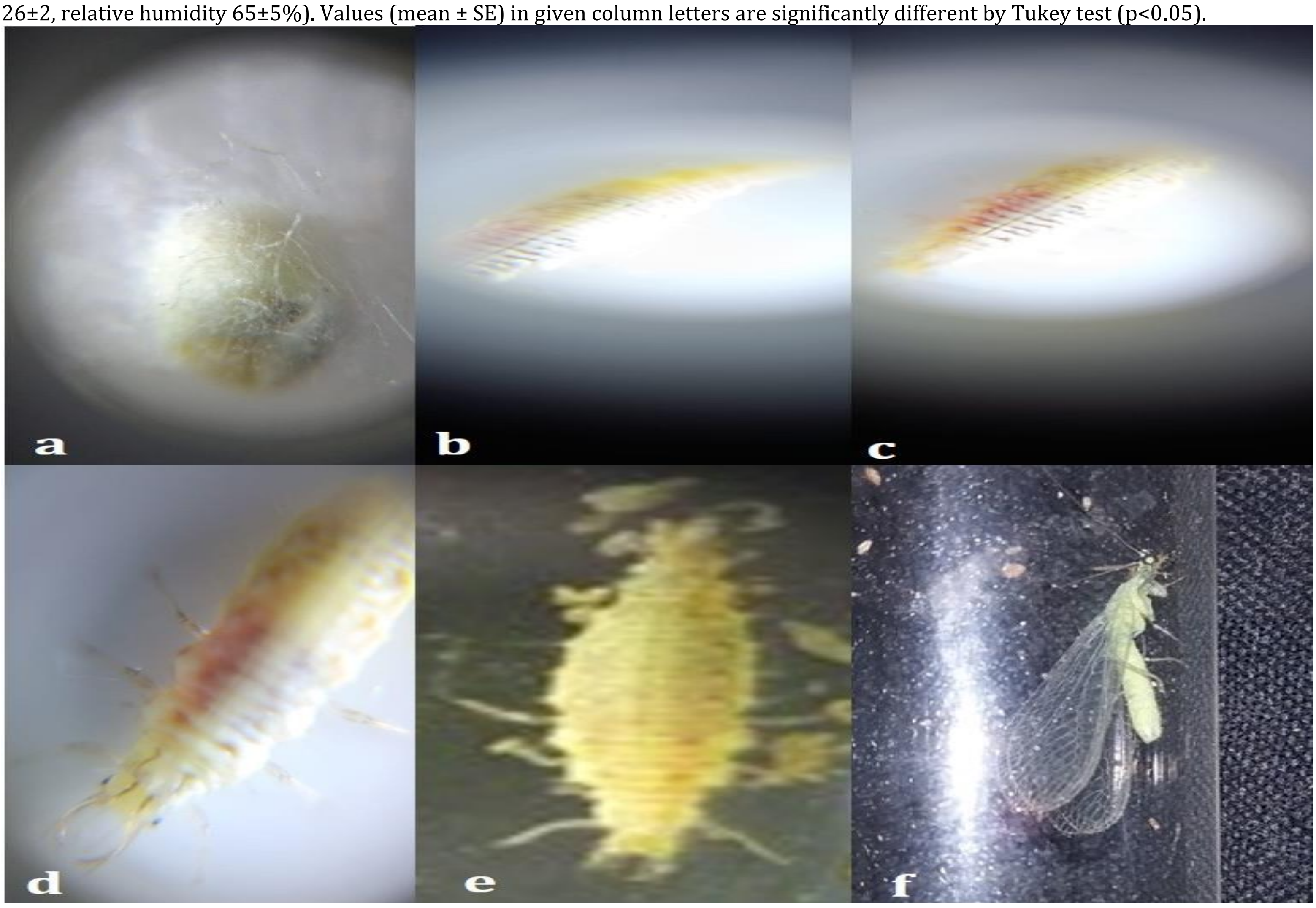

Figure 1: Egg(a), 1st instar (b), 2nd instar (c), 3rd instar (d), feeding on aphid (e) and adult of *Chrysoperla carnea*

The feeding performance were significantly (P<0.05) different among all aphid species, but in non-significant difference was pointed among *B. brassicae* and *U. carthemi* aphid species in per day consumption. The third instar larva feeding voraciously on aphid species. The maximum feeding rate was recorded 23.66 ± 0.6/day on *A. gossypii* followed by 15.3 ± 1.39/day, 13.9 ± 0.72/day and 12 ± 0.56/day were recorded on *U. carthemi*, *B. brassicae* and *A. nerii*, respectively (table 1). Per stage, the highest consumption rate was recorded on *A. gossypii* 91.8 ± 8.36/stage on followed by 83.4 ± 5.64/stage, 71 ± 1.79/stage and 60 ± 2.79/stage observed on *B. brassicae*, *U. carthemi*, and *A. nerii*, respectively (table 2). In the light of above result the feeding efficiency of 3rd instar were significantly (P<0.05) different on aphid species.

Development of period of 1st instar was recorded 3.2 ± 0.26 days on *A. nerii* followed by 2.8 ± 0.26 days, 2.4 ± 0.03 days and 2.2 ± 0.26 days were recorded on *B. brassicae*, *A. gossypii* and *U. carthemi*, respectively. There were no significant (P<0.05) different between *B. brassicae*, *A. gossypii* and *U. carthemi*, whereas, one species of aphid, *A. nerii* significantly (P<0.05) different from rest aphid species. The second instar lasted 2.8 ± 0.26 days on *B. brassicae* followed by 2.6 ± 0.32 days, 2.2 ± 0.26 days and 2.2 ± 0.26 days were recorded on *A. gossypii*, *A. nerii* and *U. carthemi*, respectively. There were no significant (P<0.05) different among aphid species. The highest development in 3rd instar was observed 5.6 ± 0.32 and 5.6 ± 0.32 days on *U. carthemi* and *B. brassicae* followed by 4.6 ± 0.32 days, 2.8 ± 0.26 days on *A. nerii* and *A. gossypii*, respectively. The analysis of variance showed that there were no significant (P<0.05) different between aphid species, *U. carthemi* and *B. brassicae*. Whereas, significant (P<0.05) difference was found among *A. nerii* and *A. gossypii*.

The maximum development in pupa was observed 5.4 ± 0.32 days when reared on *U. carthemi* followed by 4.4 ± 0.32 days, 3.4 ± 0.52, 3.2 ± 0.26 days on *A. nerii*, *B. brassicae*, *A. gossypii*, respectively (table 3). The analysis of variance showed that there no significant (P<0.05) different between aphid *B. brassicae* and *A. gossypii*. While, *U. carthemi* was significantly (P<0.05) difference among aphid species. A maximum adult emergence % was observed on *A. gossypii* (92%), following by *B. brassicae* (87%), *U. carthemi* (84%) and *A. nerii* (80%). A statistically Significant (P<0.05), difference was observed among all of given treatment.

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Life stages	*Aphis nerii*		*Urolecen carthemi*		*Aphis gossypii*		*Brevicoryne brassicae*	
	DP-day*	RD-day*	DP-day*	RD-day*	DP-day*	RD-day*	DP-day*	RD-day*
First instar	3.20 cd	0.31	2.20 e	0.45	2.40 e	0.42	2.80 cde	0.36
Second instar	2.20 e	0.45	2.20 e	0.45	2.60 de	0.38	2.80 cde	0.36
Third instar	4.60 b	0.22	5.60 a	0.18	2.80 cde	0.36	5.60 a	0.18
Pupa	4.40 b	0.23	5.40 a	0.19	3.20 cd	0.31	3.40 c	0.29
Adults Emergence	80d		84c		92a		87b	

Table 3. Development period (days) and adults emergence % of *C. carnea* on selected aphid species under control conditions (temeprature 26±2, humidity 65±5%). Values (mean ± SE) in given column letters are significantly different by Tukey test ($p<0.05$).

DISCUSSIONS: The findings of our present results indicated the feeding efficiency and development rate of immature stages of *C.. carnea* on selected aphid species. It was observed that highest per day and per stage consumption rate of 1st, 2nd and 3rd larval instars were recorded on *A. gossypii* followed by *A. nerii, B. brassicae* and *U. carthemi*, respectively. It was observed that 3rd instar larvae voraciously feed on aphid species followed by 2nd and 1st instars. The 1st instar larvae preferred aphid species because it is small in size as compare to rest aphid species, easy in grasp. The result of present study indicated that highest development of period (days) was recorded in 1st instar on *A. nerii* followed by on *B. brassicae*, *A. gossypii* and *U. carthemi*, respectively. There were no significant (P<0.05) different between *B. brassicae*, *A. gossypii* and *U. carthemi*, whereas, one species of aphid, *A. nerii* significantly (P<0.05) different from rest aphid species. The result further revealed that the maximum development period (days) in pupa was seen on *U. carthemi* followed by *A. nerii*, *B. brassicae*, *A. gossypii*, respectively. The analysis of variance showed that there no significant (P<0.05) different between aphid species, *B. brassicae* and *A. gossypii*. While, *U. carthemi* was significantly (P<0.05) difference among aphid species.

The findings of present study have the conformity with Saminathan et al. (2003), Jagadish and Jayaramaiah (2004) and Udin et al., (2019) they reported that when the development of larval instars of C. carnea increased the predatory efficiency also increased with its growth rate from 1st to 3rd instar the feeding rate was found better in all the prey species (aphids). This is also having strong agreements with those of Singh and Manojkumar (2000) they described that the 1st, 2nd and 3rd instars larvae of *C. carnea* devoured 11.48, 79.52 and 83 *L. erysimi*, respectively. Similarly, Singh and Hamid (1998), reported that the 1st, 2nd and 3rd instars larvae of *C. carnea* devoured an average of 21.68, 76.92 and 160.92 cabbage aphids, respectively. During total larval period 15.57 days only a single larva devoured about 259.22 aphids. Rana and Srivastava (1998), sattar and Zahra (2022), depicted in their findings that the larvae of *C. carnea* consumed highest number of aphids. The discrepancy in feeding potential noticed by earlier workers might be attributed due to the variation in stage (instar) of the prey (aphid) used, prey density and environmental conditions prevailed during the study period. According to Satpathy et al. (2001) whose findings showed that the at highest prey density food consumption rate was also more. The results of present study have the more or less agreements with the report of Rana and Srivastava (1998) who revealed that aphid consumption positively correlated with food preferences in order to *L. erysimi* followed by *A.craccivora* and *B. brassicae*. Our findins strongly supported by Bansod et al. (2001) and Liu and Chen (2001) they reported that the predatory larvae of *C. carnea* preferred *A. gossypii* as compare to *L. erysimi* and *U. compositae* as food (prey). Therefore, *C. carnea* efficiently utilized in cotton ecosystem than others as it also feeds on bollworm eggs and neonate larvae except on aphids and other sucking pests. Our findings have the partial agreements with those of Bukero, et al. (2014) who reported that *C. carnea* larvae devoured highest nymphs of banana aphids during life span followed by safflower Aphid and jassids. The 1st instar larvae lasted for 3 days and then transformed in to subsequent stage. However, 2nd and 3rd instars non- significantly rest 2 days to transform into consequent instars, respectively. There was no significant (P<0.05) difference was recorded in rate of development (0.33-day) of 1st instar lavae followed by 2nd (0.22- day) and 3rd instar larvae (0.22- day) reared on *P. nigronervosa*, *U. carthami* and *E. devastans*. However, LSD further revealed that *C. carnea* larvae developed non-significantly on safflower aphid and jassid prey species.

CONCLUSIONS: Feeding efficiency of larval instars of *C. carnea* reared on *A. gossypii* was relatively higher as compared to those on *B. brassicae*, *U. carthemi* and *A. nerii*, respectively. The highest consumption rate was found in 3rd instar larva of *C. carnea* followed by 1st and 2nd instar, respectively. With the development of age, the *C. carnea* larvae food consumption increased simultaneously. The DP-day and RD-day were found different in larval instars and pupa of *C. carnea* reared on aphid species. The shortest development was recorded on cotton *A. gossypii* as compare to other aphid species. It is suggested that the *C. carnea* larvae should be reared on natural food (aphids) in bio-control laboratories for their mass production. This is the best strategy of IPM to release this bio-control agent in field of organic cotton and other crops for the management of devastating arthropod pests.

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