



## BIOLOGY OF *CHEILOMENES SEXMACULATA* (F.) (COLEOPTERA: COCCINELLIDAE) ON COTTON APHID *APHIS GOSSYPYII* GLOVER

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### ABSTRACT

The six spotted ladybeetle, *Cheilomenes sexmaculata* (Fabricius) is one of the important native aphidophagous species widely distributed all over the country. It is known to prey on different soft bodied insects preferably aphids. Hence it attracted attention for its predatory potential on many important aphid pests, cotton aphid *Aphis gossypii* Glover as being one of them. Keeping in view of this tri-trophic combination, investigations have been carried out to know the prey-host specific biology of *C. sexmaculata* on *A. gossypii* hosted by cotton. Cotton aphid *A. gossypii* was found to be the most suitable host for *C. sexmaculata* as evidenced by faster development ( $6.21 \pm 0.03$  days), more longevity of female ( $67 \pm 4.7$  days) and high fecundity (756.5 eggs/ female). These positive biological attributes of *C. sexmaculata* on *A. gossypii* could contribute to its mass production for augmentative biological control.

**Key words:** Biological control, ladybird beetle, morphometry, predator, prey

Cotton aphid, *Aphis gossypii* Glover is an important pest of field and horticultural crops with host range of about 320 plant species belonging to 46 families (Blackman and Eastop, 2000). However, cotton, *Gossypium hirsutum* L., remains highly susceptible to cotton aphid (Ebert and Cartwright, 1997). After introduction of Bt cotton major share of insecticide was directed towards the management of aphids and other sucking pests. Frequent application of chemical insecticides has led to the development of insecticide resistance. This called for more emphasis on biological pest suppression of aphids using predators (Sarmiento et al., 2007; Van Emden and Harrington, 2007).

The six spotted ladybeetle, *Cheilomenes sexmaculata* (Fabricius) is one of the most important native species of coccinellid beetles with high biocontrol potential against aphids (Srikanth and Lakkundi, 1990; Omkar and Pervez, 2016). With respect to its feasibility in biological pest control, it has shown intrinsic advantage over two coexisting lady beetles *Coccinella septempunctata* L. and *C. transversalis* Fab. (Omkar et al., 2005). Biology of coccinellid varies with aphid prey species (Omkar et al., 2009) and may also vary with host plant on which prey was reared (Rakshan and Ahmed, 2013). Even though some studies were done on biology of *C. sexmaculata* (Ng, 1991; Rai et al., 2003; Omkar et al., 2005; Tank and Korat, 2007; Ali et al., 2012; Priyadarshini et al., 2016), studies using *A. gossypii* as prey are limited or inadequate. Hence, the

present investigations have been carried out to know the biology and morphometry of *C. sexmaculata* on *A. gossypii*.

### MATERIALS AND METHODS

Cotton plants (*G. hirsutum*) of variety RCH-650 were grown in the plastic pots ( $\varnothing$  20 cm) filled with fine loamy soil and coco peat in equal quantity by sowing in July, 2018. These plants were kept in screen cages to avoid cross infestation and maintained at controlled conditions of  $25 \pm 1$  °C,  $65 \pm 5\%$  RH and 16 hr of photo period in environmental chambers (Rem®). Cotton aphid, *A. gossypii* was collected from the cotton fields of ICAR-Indian Agricultural Research Institute (IARI), New Delhi. Parthenogenetic adult females were transferred on to the potted cotton plants to establish aphid population and maintained as stock culture throughout the study period (Oliveira et al., 2010). Some of the potted cotton plants having enough aphid population were used for rearing of *C. sexmaculata*. Field collected adult beetles of *C. sexmaculata* were released on to the cotton plants in the screen cages having *A. gossypii* colonies. Eggs laid by second generation beetles were used for the study of biology and morphometry.

The study on biology of *C. sexmaculata* was conducted in the Biological Control Laboratory, Division of Entomology, ICAR-IARI at controlled conditions of  $25 \pm 1$  °C,  $65 \pm 5\%$  RH and photo period

of 14 hr. Eggs laid by adult beetles were kept in the petri plate to observe incubation period and hatching percent. Growth and development were observed by providing aphid on cotton leaf substrate kept in a petri plate (100 mm Ø). Freshly collected cotton leaf was spread upside down in the petri plate, mixed population of second and third instar nymphs of aphid were gently transferred on to the leaf and allowed to settle. Newly hatched grub of *C. sexmaculata* was transferred on to the leaf (Dixon and Agarwala, 1999). Agarose based Hoagland's media containing essential plant nutrients was used to keep the prolonged freshness of leaf. In each petri plate 5 ml of Hoagland's media (1.5 %) was poured at one side by keeping in slant position and kept for solidifying. Cut end of petiole of cotton leaf was inserted in to Hoagland's media before transferring the aphids. Total 60 plates were maintained as replicates. Aphids were replaced every day with fresh ones where as cotton leaf for every three days. The grubs were observed for every 3 hr for moulting, pre-pupation, pupation, adult emergence and mortality. For I<sup>st</sup> instar grub 50, II<sup>nd</sup> instar 100, III<sup>rd</sup> and IV<sup>th</sup> instar 150 aphids were provided as prey/ day.

The same procedure was followed for adult beetles. One day old adult beetles were transferred individually in to a petri plate containing 150 aphids. After pre-mating period, pairing was done and allowed for 24 hr, and then the pair was separated and maintained individually till their mortality. Different biological parameters viz., fecundity, incubation period, hatching per cent, duration of each stage, sex ratio, oviposition period, longevity and mortality were recorded. During each stage, observed under stereo-binocular microscope for colour pattern. To ascertain proper growth, morphometrical measurements were recorded for each stage using Nikon microscope (SMZ 800N) having camera (USB 500) with software, "VIMAGE 2016".

## RESULTS AND DISCUSSION

The details of biology and morphometrics are given in Table 1. Eggs were laid mostly on cotton leaf sometimes on the surface of petri plate in clusters of 6 to 24 eggs. Freshly laid eggs were cigar shaped, yellow in colour with smooth chorion and without any reticulations. With advancement of age, eggs changed to pale yellow then turned greyish and became completely black just before hatching. Length of eggs varied from 0.88 to 1.18 mm with a mean of  $1.06 \pm 0.06$  mm and the breadth ranged from 0.34 to 0.45 mm with a mean of  $0.38 \pm 0.02$  mm. Incubation period

showed consistency and ranged from 3 to 3.6 days with a mean of  $3.2 \pm 0.1$  days. In a cluster, almost all eggs hatched simultaneously without any lagging period, particularly in small clusters of 6 to 12 eggs. Hatching percentage varied from 69.2 to 100 percent with a mean of  $89.9 \pm 4.7$ .

The grubs of *C. sexmaculata* completed development through four instars with three moults then went under pupation with considerable pre-pupation period. Freshly hatched grubs were dark grey in colour with shining dark head capsule and legs. Length and breadth of grub ranged from 1.33 to 1.64 mm and 0.38 to 0.47 mm, respectively. Mean length and breadth was  $1.49 \pm 0.05$  and  $0.42 \pm 0.01$  mm, respectively. Mean width of head capsule of I<sup>st</sup> instar grub was  $0.23 \pm 0.01$  mm. Second instar grubs were greyish black in colour with black head capsule and legs, and white coloured patches on first and fourth abdominal segments. Length and breadth of II<sup>nd</sup> instar grub ranged from 3.94 to 4.42 mm and 0.58 to 0.75 mm, respectively. Mean length and breadth was  $4.13 \pm 0.09$  and  $0.68 \pm 0.03$  mm, respectively. Mean width of head capsule was  $0.45 \pm 0.03$  mm.

Third instar grub was glistening black in colour with development of white spots on mid-dorsal line of meso thorax, meta thorax and first abdominal segments. Length and breadth of III<sup>rd</sup> instar grub ranged from 5.26 to 6.13 mm and 0.76 to 0.88 mm, respectively. Mean length and breadth was  $5.74 \pm 0.15$  and  $0.83 \pm 0.02$  mm, respectively. Mean width of head capsule was  $0.55 \pm 0.02$  mm. Fourth instar grubs were yellowish black in colour with black colour legs. Length and breadth of IV<sup>th</sup> instar grub was ranged from 6.94 to 7.63 mm and 1.28 to 1.43 mm, respectively. Mean length and breadth was  $7.37 \pm 0.12$  and  $1.36 \pm 0.02$  mm, respectively. Mean width of head capsule was  $0.68 \pm 0.02$  mm. The duration of I<sup>st</sup>, II<sup>nd</sup>, III<sup>rd</sup> and IV<sup>th</sup> instar grub ranged from 1.625 to 1.875, 1 to 1.375, 1.125 to 1.625 and 1.75 to 2.25 days, respectively with a mean duration of  $1.729 \pm 0.018$ ,  $1.161 \pm 0.022$ ,  $1.323 \pm 0.072$  and  $2 \pm 0.027$  days, respectively. The total grub period ranged from 6 to 6.375 days with mean developmental period of  $6.214 \pm 0.03$  days. Pre-pupation period was  $1.318 \pm 0.05$  days.

The results of present study signify the suitability of *A. gossypii* for *C. sexmaculata* as shown faster development of grub in just 6.21 days. The faster development may be attributed to the quality of prey possessing essential nutrients required for growth and development of predator. Even though many studies

Table 1. Biology and morphometrics of *C. sexmaculata* on *A. gossypii*

S. No.	Particulars	Period (Days)			Measurements			Number observed
		Minimum	Maximum	Mean±SE	Length (mm)±SE	Breadth (mm)±SE	Head width (mm)±SE	
1	Egg	3.0	3.6	3.2±0.1	1.06±0.06	0.38±0.02	-	60
2	First instar	1.625	1.875	1.729±0.018	1.49±0.05	0.42±0.01	0.23±0.01	60
3	Second instar	1.000	1.375	1.161±0.022	4.13±0.09	0.68±0.03	0.45±0.03	56
4	Third instar	1.125	1.625	1.323±0.022	5.74±0.15	0.83±0.02	0.55±0.02	55
5	Fourth instar	1.750	2.250	2.000±0.027	7.37±0.12	1.36±0.02	0.65±0.02	54
6	Total grub period	6.000	6.375	6.214±0.030	-	-	-	54
7	Pre-pupa	1.000	1.875	1.318±0.050	-	-	-	54
8	Pupa	3.250	3.875	3.585±0.043	3.83±0.13	2.32±0.1	-	54
9	Adult-Male	19.0	86.0	65.1±7.3	4.23±0.09	3.75±0.09	-	24
10	Adult-Female	34.0	87.0	67.0±4.7	5.35±0.25	4.06±0.15	-	26
11	Pre-oviposition	5.0	8.0	6.0±0.4	-	-	-	26
12	Oviposition	20.0	49.0	42.0±3.3	-	-	-	26
13	Post-oviposition	9.0	36.0	19.0±3.1	-	-	-	26
14	Total life cycle of male	97.25	30.12	75.56±8.76	-	-	-	24
15	Total life cycle of female	98.0	45.0	78.06±5.96	-	-	-	26
16	Survival of grub (%)	-	-	90.0	-	-	-	60
17	Survival of pupa (%)	-	-	92.6	-	-	-	54
18	Adult emergence (%)	-	-	83.33	-	-	-	60
19	Sex ratio (Male:Female)	-	-	0.92:1	-	-	-	54
20	Fecundity (No. of eggs/female)	241	1377	756.5±128.9	-	-	-	26
21	Hatching %	69.2	100.0	94.9±4.7	-	-	-	100 clusters

have reported *A. craccivora* as the most suitable prey for ladybird beetles, viz. *C. septempunctata* (Hodek, 1960), *Propylea dissecta* Mulsant (Omkar and Mishra, 2005) and *C. sexmaculata* (Omkar and Bind, 2004), the present combination of predator-prey was missed. Suitability of prey depends on lady bird species and also varies with predator-prey combination (Omkar et al., 2009). Interestingly, Sugiura and Takada (1998) reported *A. craccivora* as the most suitable prey for *C. sexmaculata* compared to *A. gossypii* but the host species used was a pumpkin *Cucurbita moschata* Duchesne.

It was evidenced that the prey suitability and developmental period also influenced by host species on which the prey was reared as earlier studied by Rattanapun (2012) who reported significant variation in developmental period of *C. sexmaculata* on *A. gossypii* when reared on two different host species. Similarly Rakshan and Ahmed (2013) found morphological and developmental variations in *C. sexmaculata* on *A. craccivora* reared on different hosts. Faster development of *C. sexmaculata* on *A. gossypii* found

in present study was strongly supported by Omkar et al. (2009) who found fastest development of lady beetle *Anegleis cardoni* (Weise) on *A. gossypii* followed by *A. craccivora* and *Lipaphis erysimi* Kalt and reported *A. gossypii* as most suitable prey.

Freshly formed pupae were whitish yellow then turned to pale orange yellow. Pupae had symmetrically arranged black spots on each segment. Mean length and breadth of pupae was  $3.83 \pm 0.13$  mm and  $2.32 \pm 0.1$  mm., respectively. Pupation period ranged from 3.25 to 3.875 days with mean of  $3.585 \pm 0.043$  days. Newly emerged adults were yellowish, which later turned to shining yellow with black spots. Later adult spotted zigzag markings on elytra. Length and breadth of male ranged from 3.98 to 4.47 and 3.56 to 4.02 mm with a mean of  $4.23 \pm 0.09$  and  $3.75 \pm 0.09$  mm, respectively. Whereas, in case of female, it ranged from 4.46 to 5.84 and 3.57 to 4.46 mm, with a mean of  $5.35 \pm 0.25$  and  $4.06 \pm 0.15$  mm, respectively. After adult emergence, three days of pre-mating period was observed whereas, pairing was noticed from fourth day onwards. Total longevity of male and female ranged from 19 to 86 and

34 to 87 days, respectively. However, mean duration of longevity was noticed as  $65.1 \pm 7.3$  and  $67 \pm 4.7$  days. Despite of shorter duration of grub stage, longevity of adults was found on higher side. Adult longevity was higher when compared with earlier studies as reported from 30 to 40 days (Tank and Korat, 2007; Priyadarshini et al., 2016; Sanghani et al., 2017). However, Singh et al. (2008) reported higher longevity (92.25 days) which is in accordance with present study. Male has shown longer duration compared to female as reported earlier (Tank and Korat, 2007; Priyadarshini et al., 2016). In a total life span of 67 days of female,  $6 \pm 0.4$  days were devoted for pre-oviposition,  $42 \pm 3.3$  days for oviposition and  $19 \pm 3.1$  days for post oviposition.

During the mean oviposition period of 42 days, fecundity has been found to increase for first 30 days and there after decreased gradually by exhibiting a triangular fecundity function (Dixon and Agarwala, 2002). The highest per day fecundity for individual was 72 whereas, the mean fecundity was found to be  $18.01 \pm 0.88$  eggs/day/female. The total fecundity varied between 241 and 1377 with pooled mean of  $756.5 \pm 128.96$  eggs/female. Higher longevity and oviposition period were contributed to yield higher fecundity (756.5). High fecundity and oviposition period of *C. sexmaculata* on *A. craccivora* was also reported by Omkar et al. (2005). Present results are in closer agreement with Rajamohan and Jayaraj (1973) who reported fecundity of 717.7 for *C. sexmaculata* when reared on *A. craccivora*. Total life cycle was completed with a mean duration of  $78.06 \pm 5.96$  days in female and was found higher than male ( $75.56 \pm 8.76$ ). Survival percentage of egg, larva and pupa were 94.9, 90.0 and 92.59, respectively. Morphometrical observations recorded in the present study were in line with the earlier reports (Rai et al., 2003; Tank and Korat, 2007; Rakshan and Ahmed, 2013) without much deviation indicates proper growth and development of *C. sexmaculata* on *A. gossypii*.

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