BIOLOGY, BEHAVIOUR AND FUNCTIONAL RESPONSE OF *CYDNOCORIS GILVUS* BRUM. (HETEROPTERA: REDUVIIDAE: HARPACTORINAE) A PREDATOR OF TEA MOSQUITO BUG (*HELOPELTIS ANTONII* SIGN.) ON CASHEW IN INDIA

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Abstract: Helopeltis spp. (Hemiptera: Miridae) are major sucking pests of cashew (Anacardium occidentale L.) in India. Cydnocoris gilvus Brum. (Heteroptera: Reduviidae: Harpactorinae) is recorded as a potential predator of Helopeltis spp. Biology, mating behaviour and functional response of C. gilvus were studied by rearing in the laboratory (temperature 26–28 $^{\circ}\text{C};$ relative humidity 89–94 %) with wax moth, Galleria mellonella, larvae. Based on laboratory rearing, the fecundity was 56.33 eggs in 8.67 batches per female. The average stadial period was 37.3 days, with a maximum of 11 days for V instar and a minimum of 4.5 days for III instars. C. gilvus took 45.5 days to complete a generation. The innate capacity of natural increase was 0.07 with a gross reproduction of 67.8 females per female. The adult exhibited a pin and jab mode of predation in a sequence of actions. The sequential action of mating comprised arousal (1.32 min), approach (12.30 min), riding over (140.48 min) and copulation (85.40 min). The predator responded to increasing prey density by killing more prev than at lower prev densities

Keywords: Anacardium occidentale, developmental stages, Galleria mellonella, Helopeltis antonii, predator.

Cashew (Anacardium occidentale L.) is a native of Brazil, and it was introduced into the western coast of India by Portuguese travelers during the 16th century. India's share in world cashew area is 22.50% and the share in world cashew production is 20.74% (DCR 2011). Cashew is presently grown in an area of 0.982 million hectares with an annual production of about 0.728 million tonnes of raw cashewnuts in the country (DCCD 2013). Infestation by insects has been identified as a major factor responsible for low productivity in cashew (Sundararaju 1993). The main insect affecting cashew is the Tea Mosquito Bug (TMB), Helopeltis antonii Signoret (Hemiptera: Miridae). Both nymphs and adults damage tender shoots, inflorescence, immature nuts and apples at various stages of development, resulting in a yield loss of 30-50% (Devasahayam & Nair 1986).

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Reduviids (Hemiptera: Reduviidae) are recorded as potential natural predators of *Helopeltis* spp. on cashew (Devasahayam & Nair 1986; Stonedahl 1991; Sundararaju 1996). Five species of reduviids, viz., *Sycanus collaris* Fab., *Sphedanolestes signatus* Dist., *Endochus inornatus* Stal, *Irantha armipes* Stal and *Occamus typicus* Dist., were reported as predators of *Helopeltis antonii* on cashew in India (Sundararaju 1984). *E. inornatus* was reported to feed on 20 individuals of *H. antonii* per day in cashew plantations (Sundararaju 1984; Devasahayam & Nair 1986). Vennison & Ambrose (1990) accounted for the significance of *S. signatus* in controlling *Helopeltis* spp. The biological control programme against *Helopeltis* spp. using reduviids has been reported from Southeast Asia and Pacific region (Rao et al. 1971).

Reduviids can be successfully used as effective biological control agents of important agricultural pests (Ambrose 2003; Sahayaraj et al. 2006). Biological parameters were reported for *Rhynocoris kumarii* Ambrose and Livingstone (Ambrose 2000), *Sphedanolestes minusculus* Berg. (Ambrose et al. 2006), *Endochus migratorius* Dist. (Ambrose et al. 2007), *S. himalayensis* Dist., *S. signatus* Dist. (Vennison & Ambrose 1990) and *S. variabilis* Dist. (Ambrose et al. 2009). Although the biologies of a few species of Oriental reduviids are known, there are still plenty of other species to be explored (Sahayaraj 2012).

During the survey for natural predators of *Helopeltis* spp. in cashew plantations we recorded *Cydnocoris gilvus* Burmeister (Heteroptera: Reduviidae: Harpactorinae). Earlier, the prey mediated feeding response of *C. gilvus* was reported and it is described in the checklist of Indian assassin bugs (Venkatesan et al. 1997). The knowledge on bioecology, behaviour, and pest suppression efficacy of any organism is a prerequisite for its utilization as a biological control agent. Hence, we reared *C. gilvus* in the laboratory using wax moth, *Galleria mellonella* L. (Lepidoptera: Pyralidae), larvae as prey, and examined its biology, behaviour and the functional response towards its natural prey, *Helopeltis antonii*.

MATERIAL AND METHODS

Laboratory culture of *C. gilvus*: The nymphs of *C. gilvus* were collected from cashew plantations of Directorate of Cashew Research (DCR), Puttur (12.45°N & 75.4°E; elevation 90m) in Karnataka State, southern India. They were reared in glass rearing bottles (500ml capacity) using larvae of wax moth, under laboratory conditions (temperature 26–28 °C; relative humidity 89–94 %). The virgin males and females that emerged in the laboratory were allowed to mate in glass rearing

bottles. Only adults reared in the laboratory were used in the experimental studies.

The containers were carefully examined at regular intervals to record the number of eggs laid. Ejection of spermatophore capsules by mated females confirmed successful copulation. The eggs laid were allowed as such to hatch in the same bottles, which were kept over wet cotton swabs for maintaining optimum humidity (85%). The cotton swabs were changed periodically in order to prevent fungal attack. Mated females were maintained individually in order to record the number of batches of eggs and the number of eggs in each batch. Twentyfive newly hatched nymphs were separated soon after eclosion and reared individually with first and second instar (5–6 mm) larvae of wax moth. As the nymphal development advanced the fourth and fifth instar larvae (10–15 mm) of the wax moth were supplied.

We observed eclosion, fecundity, hatchability, ecdysis, nymphal mortality, adult emergence, sex ratio and adult longevity from the adults that emerged in the laboratory for two generations. The life table parameters were obtained according to the methods of Atwal & Bains (1974). Observations from hatching of eggs till the emergence and death of adults were made daily, which provided the values for a life table (lx).

Predatory behaviour of *C. gilvus*: Predatory behaviour of *C. gilvus* towards wax moth larvae and *H. antonii* adults was observed in a span of 24 hours. The extent of juvenile cannibalism in *C. gilvus* was also recorded.

Mating behaviour of *C. gilvus*: The mating behaviour of sex starved *C. gilvus* was studied in laboratory conditions. The time taken for the sequential acts was observed.

Laboratory mass culture of H. antonii: The H. antonii gravid females were collected from the cashew plantations of DCR. They were allowed to lay eggs in the laboratory on potted cashew seedlings, which were confined in perforated tubular cages (30×7.5 cm) made from transparent polyester film (thickness 175 micron). The respiratory processes of eggs projecting from the surface of the bark were indicative of the presence of eggs. Immediately after hatching, the nymphs were transferred into nymphal rearing cages (size: 15×15×20 cm and thickness: 18 gauge) developed by Sundararaju & John (1992). Nymphal rearing cages consisted of four glass vials of 5ml capacity fixed on a small aluminum stand with a handle of 15cm height fixed at the centre. A tender cashew shoot was kept erect inside each vial filled with water and the opening of the vial was closed with wet absorbent cotton. Every third day another

aluminum stand along with four fresh tender shoots was placed adjacent to the already existing aluminum stand without transferring/disturbing the nymphs feeding on the shoots kept on the previous day. In the rearing cage, two side provisions were fixed with cloth sleeve in order to facilitate the removal of adults after final moulting

Functional response: Laboratory raised and starved for 24 h adults of *C. gilvus* were used in this experiment. The functional response was assessed separately at four different prey densities viz., 1,2,4 and 8 prey/predator of its natural prey, H. antonii for five days in rearing glass bottles (500ml). Six replicates were made for each category. At 24 hr interval the number of prey killed was recorded and the prey number was maintained constant by the introduction of fresh prey throughout the experiment. "Disc" equation of Holling (1959) $Y' = \alpha$ Tt-by X was used to describe the functional response of C. gilvus to H. antonii.

Where

X = Prey density

Y = Total number of prey killed in given period of time (Tt)

Tt = Total time in days for which prey was exposed to the predator

b = Handing time in days

 α = Rate of discovery per unit of searching time (y/x / Ts)

x/y = Attack ratio

Ts = Searching time in days

Linear regression analysis was used to explore the relationship between the prey density and number of prey attacked, searching time and attack ratio.

RESULTS

Biology: Cydnocoris gilvus laid broadly oval, orange yellow coloured eggs in batches on the bottom of the rearing bottles, each egg vertically glued to the substratum (Image 1). Females laid 8.67±0.67 batches of eggs with a total number of 56.33±7.88 eggs. The batch size varied from the mean minimum of 3.33±0.33 per batch to the mean maximum of 11.33±1.45 per batch. The fertilized eggs turn into dark reddish chorion with eyespots prior to hatching whereas unfertilized eggs become shrunken after a few days. The eggs hatched after 8.17±0.31 d. The pre-oviposition period of C. gilvus was 11.00±0.37 d and females lived only 3.83±0.17 d (post-oviposition period) after laying its last batch of eggs (Table 1). The oviposition index value observed was 67.07%.

The newly hatched nymphs were fragile and they became tanned in 3-4 hr after emergence and thereafter start feeding, showing preference to small and sluggish larvae. The developmental duration of I, II, III, IV and V instars were 9.50±1.45, 6.33±0.33, 4.50±0.62, 6.00±0.68 and 11.00±1.88 d, respectively (Table 2). Survival percentage of first instar was comparatively lower than

Ш ш IV (11.0mm) (3.1mm) (5.1mm) (7.2mm) Image 1. Cydnocoris gilvus: (a) fresh eggs, (b) eggs hatching, (c) hatched eggs and (d) I-V nymphal instars



Table 1. Longevity and oviposition pattern of C. gilvus (n = 45)

Parameters	C. gilvus (mean±SE)			
Adult male longevity (d)	41.67±4.69			
Adult female longevity (d)	42.00±4.98			
Pre-oviposition (d)	11.00±0.37			
Post-oviposition (d)	3.83±0.17			
Oviposition (d)	28.17±4.47			
Oviposition index (%) *	67.07			
Total number of batches of eggs	8.67±0.67			
Total number of eggs laid (Fecundity)	56.33±7.88			
Average number of eggs /batch	6.44±0.99			
Minimum number of eggs / batch	3.33±0.33			
Maximum number of eggs/ batch	11.33±1.45			
Hatching percentage (%)	93.57±4.84			

* Percentage of egg-laying days in total lifespan of female

Table 2. Incubation and developmental period of C. gilvus

Parameters	Mean±SE (n=45)	Range (d)			
Incubation period (d)	8.17±0.31	7–9			
Stadial period (d)/ Per cent survival					
I	6–14				
Ш	6.33±0.33 (100.00)	5–7			
Ш	4.50±0.62 (77.78)	3–7			
IV	6.00±0.68 (100.00)	3–8			
V	11.00±1.88 (85.00)	7–20			
Male (d)	38.00±3.00	32–41			
Female (d)	36.67±2.92	31–41			
Total (d)	37.33±0.67 (82.17)	31–41			
Sex ratio (M:F)	1:0.8				

Values in parentheses indicate the survival percentage.

other instars.

The developmental period of females was faster (36.67 \pm 2.96 d) than males (38.00 \pm 3.00 d) (Fig. 1). *C. gilvus* took 45.5 \pm 0.92 days to complete a generation.

The innate capacity of natural increase (rc) was 0.07 with a gross reproduction (mx) of 67.80 females per female. Mean length of generation (Tc) was 52.26 days (Table 3).

Predatory behaviour: *C. gilvus* exhibited a pin and jab mode of predation in a sequence of acts. The sequential action of predatory behaviour was observed in a span of 24 hr as follows: arousal - approach - capturing - rostral probing - paralysing - sucking - postpredatory behaviour. The nymphs showed no cannibalistic behaviour when they were mass reared (Image 2).

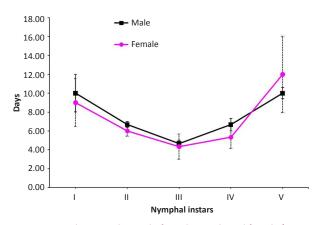


Figure 1. Developmental period of *C. gilvus* male and female (mean \pm SE)



Image 2. C. gilvus nymphs predating on wax moth larva

Table 3. Life table parameters of *Cydnocoris gilvus* on *G. mellonella* under laboratory conditions

Parameters	Formula	
Gross reproduction (GR) (females/female/day)	∑mx	67.80
Net reproduction (R_{o})	∑lxmx	48.80
Mean length of generation (T_c)	$\sum x x x x x x /_{Ro}$	52.26
Innate capacity of increase in numbers (rc) (females/female/day)	$\log e^{Ro}/T_{Tc}$	0.07
Hypothetical F ₂ females	(Ro) ²	2381.44

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Prey density (x)	Prey attacked (y)	Max 'y' (k)	Days /y b = Tt/k	All y's days (by)	Searching days Ts = Tt–by	Attack ratio y/x	Rate of discovery γ/x/Ts = α	Disc equation y'= α (Tt– by) x	y'
1	0.48			0.92	4.08	0.48	0.12	y' = 0.12 (4.08) x 1	0.49
2	1.89	2.60	1.92	3.63	1.37	0.95	0.69	y' = 0.69 (4.08) x 2	5.63
4	2.40			4.62	0.38	0.60	1.56	y' = 1.56 (4.08) x 4	25.46
8	2.60			5.00	0.00	0.33	-	-	

Table 4. Functional response of C. gilvus towards H. antonii



Image 3. *C. gilvus*: (a) male, (b) female, (c) riding over and (d) copulation

Mating behaviour: The sequential act of mating behaviour observed in *C. gilvus* as follows:

Arousal - approach - riding over - copulation - post copulatory acts: The matured C. gilvus adults (4-5 days old) were aroused immediately by the sight of the opposite sex in 1.32±0.29 min. The aroused male chased the females within a span of 12.3±1.34 min with extended antenna movement. Sometimes the females escaped from the approaching male. The male placed its legs on the female after approaching. Males rode over the females with extended rostrum for 140.48±5.48 min. Riding over was seen longer than all other mating behaviors (Image 3c). Copulation was 85.40±2.99 min in duration. It remained motionless during copulation and exhibited pterothorax rostral pinning. Drooping down of antennae by both the sexes was observed at the termination of copulation and thereafter separation of mating partners. After separation both male and female moved away from the place of copulation (Image 3d). The post copulatory acts such as genitalia

brushing, antennal grooming, cleaning the legs, and wing beating were observed in both sex partners. Post copulatory acts lasted for 5.70±0.37 min. Successful completion of copulation was evidenced by the ejection of spermatophore capsule by female after termination of copulation.

Functional response: *C. gilvus* responded to increasing prey density by killing more prey than at lower prey densities (Table 4). The number of prey killed (y) by the individual predator increased as the prey density (x) increased from one prey/predator to eight prey /predator. This was further confirmed by the positive correlation (r=0.63) obtained between prey density and prey killed. The maximum predation was represented by 'k' value (2.60). The highest attack ratio was observed at the density of two prey/predator and the lowest attack ratio was found at the density of eight prey /predator. Hence, the attack ratio decreased as the prey density increased (r=-0.63). A negative correlation was obtained between prey density and searching time (r=- 0.35) of the predator at all prey densities.

DISCUSSION

Biological studies on reduviids and their utilization in biological control of insect pests have been gaining momentum in India and other countries in recent years (Sahayaraj 2007; Schaefer 2010).

The laboratory culture technique standardized using wax moth larva as prey was found to be similar to the other reduviid rearing techniques (Vennison & Ambrose 1990; Ambrose 1999; Ambrose 2000; Sahayaraj 2012).

C. gilvus laid eggs on the bottom and sides of the rearing bottles and muslin cloth as reported in *Edocla slateri* Distant (Vennison & Ambrose 1986), *Coranus soosaii* Ambrose & Vennison (Vennison 1989) and *C. spiniscutis* Reuter (Claver & Reegan 2010). The fecundity of *C. gilvus* was higher than *Sphedanolestes* spp. (15.33±6.41 eggs) (Vennison & Ambrose 1990) while lower than that of *Rhynocoris marginatus* Fab.

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Image 4. C. gilvus: (a) male and (b) female predating on H. antonii

(208.3±3.9 eggs) (Sahayaraj & Sathiamoorthi 2002) and *C. spiniscutis* (173.72±11.67 eggs) (Claver & Reegan 2010).

The egg incubation period was shorter than other harpactorines like *R. kumarii* (10 d), *Sycanus collaris* Fab. (15 d) and *Panthous bimaculatus* Dist. (21 d) (Sahayaraj 2012) and higher than *R. marginatus* (6-7 d) (Sahayaraj & Sathiamoorthy 2002) and *S. variabilis* (6.92±0.29 d) (Ambrose et al. 2009). The longest stadium was the fifth instar and the shortest was the third instar. Harpactorines generally have the shortest stadial period for II and III instars (Das 1996).

The total nymphal developmental period of *C. gilvus* was closer to *S. variabilis* (37.33±4.40 d) (Ambrose et al. 2009) and shorter than that of *S. collaris* (75.67±9.06), *R. kumarii* (88.30±3.60) and *P. bimaculatus* (101.12±2.30) (Sahayaraj 2012). The higher mortality of first instar nymphs might be owing to a higher susceptibility to mechanical injuries and temperature variations.

The male biased sex ratio was observed in harpactorines as well as nonharpactorine reduviids (Ambrose 1999; Ambrose et al. 2007).

The sequential pattern of pin and jab mode of predation observed in *C. gilvus* was similar to that of several other harpactorine reduviids (Ambrose 1999). Congregational feeding and cannibalism observed in many reduviids (Das & Ambrose 2008) were not observed in *C. gilvus* when they were mass reared. Though cannibalism is common among adults and nymphs, the degree of intensity of such behaviour vary considerably among different subfamilies and species

(Ambrose 1999).

Mating behaviour was reported in *Ectomocoris tibialis* Dist. and *Acanthespis pedestris* Stal (Ambrose & Livingstone 1978), *Rhynocoris kumarii* (Ambrose & Livingstone 1987a) and *Coranus vitellinus* Dist. (Ambrose & Livingstone 1987b). The duration of riding over was significantly longer than arousal, approach and copulation. *C. gilvus* mated in dorsoventral position as reported in *Sycanus reclinatus* Doh. and *Coranus soosaii* Ambrose & Vennison (Vennison 1989) and in several other harpactorine reduviids (Ambrose 1999).

The number of prey killed (y) by the individual predator increased as the prey density (x) increased, thus exhibiting the typical functional response of the second model of Holling's 'disc' equation (1959). The type II functional response is typical for most heteropteran predators (Cohen & Byrne 1992). Awadallah et al. (1984) working on Allaeocranum biannulipes Montr. of a stored product pest, Ambrose et al. (2000) on R. marginatus of a pest of pigeon peas and Claver et al. (2004) on C. spiniscutis Reut. Spodoptera litura Fab. and Helicoverpa armigera Hub. (Noctuidae: Lepidoptera) observed a similar response. An indirectly proportional relationship was found between the attack ratio and the prey density which is similar to the results obtained by Ambrose et al. (2009) in S. variabilis. It is assumed that when the prey density increases, predator took less time on nonsearching activities, resulting in discerning decline in attack rate until the hunger is established (Claver et al. 2004). A negative correlation was also obtained between prey density and searching time as

the results of Ambrose et al. (2009).

In conclusion the study suggested that *C. gilvus* can be multiplied within 50 days for a generation in the laboratory. Cannibalistic behaviour was not observed, which is ideal for the mass culture of this predator. The present study on the functional response suggested that *C. gilvus* is capable of reducing pest numbers (Image 4). However, the efficacy of *C. gilvus* in biocontrol of *Helopeltis* spp. needs further investigation.

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