

**Biology, Behavior, Functional Response and
Molecular Characterization of Rihirbus
Trochantericus Stal Var. Luteous (Hemiptera:
Reduviidae: Harpactorinae) a Potential Predator of
Helopeltis Spp. (Hemiptera: Miridae)**

Author(s): P. S. Bhat , K. K. Srikumar , T. N. Raviprasad , K.
Vanitha , K. B. Rebijith , and R. Asokan

Source: Entomological News, 123(4):264-277. 2013.

Published By: The American Entomological Society

DOI: <http://dx.doi.org/10.3157/021.123.0409>

URL: <http://www.bioone.org/doi/full/10.3157/021.123.0409>

BioOne (www.bioone.org) is a nonprofit, online aggregation of core research in the biological, ecological, and environmental sciences. BioOne provides a sustainable online platform for over 170 journals and books published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Web site, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/page/terms_of_use.

Usage of BioOne content is strictly limited to personal, educational, and non-commercial use. Commercial inquiries or rights and permissions

requests should be directed to the individual publisher as copyright holder. BioOne does not make any warranties about the suitability, accuracy, completeness, or appropriateness of any content appearing on the site. All content appearing on the site is the property of the individual publisher and is subject to the publisher's terms and conditions. BioOne is a nonprofit organization that operates on a non-profit basis to maximize access to critical research.

BIOLOGY, BEHAVIOR, FUNCTIONAL RESPONSE AND MOLECULAR CHARACTERIZATION OF *RIHIRBUS TROCHANTERICUS* STAL VAR. *LUTEOUS* (HEMIPTERA: REDUVIIDAE: HARPACTORINAE) A POTENTIAL PREDATOR OF *HELOPELTIS* SPP. (HEMIPTERA: MIRIDAE)¹

P. S. Bhat,² K. K. Srikumar,^{2,3} T. N. Raviprasad,² K. Vanitha,² K. B. Rebijith,⁴ and R. Asokan⁴

ABSTRACT: Reduviid species are recorded as indigenous natural enemies of tea mosquito bug (*Helopeltis* spp.), which is one of the major economically important pests of cashew. *Rihirbus trochantericus* laid eggs singly as well as in groups of up to 26 eggs in 3 to 7 clusters per female. The incubation period was 13.00 ± 0.69 days. The stadal durations of I, II, III, IV and V nymphs were 12.39 ± 1.13 , 7.00 ± 0.39 , 7.56 ± 0.35 , 9.28 ± 0.64 and 12.78 ± 1.27 days, respectively. Adult males and females survived for 107.13 ± 2.70 and 117.9 ± 3.83 days, respectively and their sex ratio was 1: 0.7. The sequential acts of predation as well as mating conform to those of Harpactorine reduviids. *R. trochantericus* exhibited Holling's type II functional response. The molecular characterization of *R. trochantericus* will be highly useful in confirming the identity of the species in any of its life stages.

KEYWORDS: *R. trochantericus*, biological parameters, functional response, barcode, cashew

INTRODUCTION

Several insect pests have been recorded on cashew (*Anacardium occidentale* L.) in India (Sundararaju, 1993), prominent among which is the tea mosquito bug (TMB), *Helopeltis* spp. (Hemiptera: Miridae). Reduviidae constitute an important group of predatory insects that could be most successfully harnessed as effective biological control agents.

Naik and Sundararaju (1982) recorded *Endochus inornatus* Stal as a predator of *H. antonii* Sign. Sundararaju (1984) also reported five species of Reduviidae: *Sycanus collaris* F., *Sphedanolestes signatus* Dist., *Endochus inornatus* Stal, *Irantha armipes* Stal and *Occamus typicus* Dist., as predators of *H. antonii* on cashew in India. All these predators were capable of devouring 1 to 5 TMB nymphs or adults in a day. Attempts to mass-rear these reduviids under laboratory conditions revealed that their mass culture is amenable if nymphal mortality could be reduced to a minimum. *S. signatus* is an alate, entomophagous reduviid found in the scrub jungles and agroecosystems of southern India (Distant, 1904). It is also found as a potential biocontrol agent on the cashew pest, *H. antonii* (Sundararaju, 1984; Vennison and Ambrose, 1990). Reduviids should be conserved and augmented to be effectively utilized in Integrated Pest Manage-

¹ Received on July 16, 2013. Accepted on October 26, 2013.

² Division of Entomology, Directorate of Cashew Research (DCR), Puttur, Karnataka, India.

³ Corresponding author: sreeku08@gmail.com

⁴ Division of Biotechnology, Indian Institute of Horticultural Research, (IIHR), Bangalore, India.

ment (IPM) programs (Ambrose, 1999; Ambrose, 2000; Ambrose, 2003; Ambrose et al., 2000; Ambrose et al., 2006).

R. trochantericus Stal var. *luteous* (Hemiptera: Reduviidae: Harpactorinae) is one of the common predators recorded in the cashew ecosystem. The subfamily Harpactorinae is the largest but most poorly studied subfamily of the Reduviidae (Cai and Tomokuni, 2003). More than 300 genera and 2000 species are known (Putshkov and Putshkov, 1985; Maldonado, 1990). Most of the Oriental species of this subfamily are listed in the work of Stal (1874), Distant (1904) and Miller (1940). Though the biology of a few species of Oriental reduviids is known (Ambrose and Livingstone, 1979; Vennison and Ambrose, 1990), our knowledge of the natural history of Oriental reduviids is scanty. Biological parameters are reported for *Rhynocoris kumarii* Ambrose and Livingstone (Ambrose, 2000), *S. minusculus* Berg. (Ambrose et al., 2006), *E. migratorius* Dist. (Ambrose et al., 2007), *S. himalayensis* Dist., *S. signatus* Dist. (Vennison and Ambrose, 1990), and *S. variabilis* Dist. (Ambrose et al., 2009), but still there is no such documentation for *R. trochantericus* even though it is reported in the checklist of Indian assassin bugs (Ambrose, 2006; Biswas et al., 1994; Biswas et al., 2010).

In this paper, we document for the first time the biology and behavior of *R. trochantericus*. Functional response was examined to determine the intake rate of prey. The mitochondrial cytochrome c oxidase (mtCOI) gene of all developmental stages was also characterized. The current study is a prerequisite for its utilization as a biological control agent.

MATERIALS AND METHODS

BIOLOGY

The nymphs of *R. trochantericus* were collected from cashew plantations of the Directorate of Cashew Research, Puttur (12.45° N latitude, 75.4° E longitude and 90 m above MSL) in the Karnataka State of Southern India. They were brought to the laboratory and reared during September to December, 2012 (temperature 26-28°C; relative humidity 89-94%) in separate glass bottles (500 ml capacity) using larvae of the wax moth, *Galleria mellonella* L. (Lepidoptera: Pyralidae).

The males and females that emerged were allowed to mate in the glass rearing bottles. 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 were allowed to hatch in the same bottles, kept over wet cotton swabs for maintaining optimum humidity (85%). The cotton swabs were changed periodically in order to prevent fungal infestation. Mated females were maintained individually in order to record the number of batches of eggs and the number of eggs in each batch. Subsequently, incubation period, stadia period, nymphal mortality, fecundity, longevity and sex ratio for two generations were recorded.

BEHAVIOR

The mating and predatory behaviors of *R. trochantericus* were studied under laboratory conditions by direct observation. Predatory behavior consists of stimulus-response mediated sequences of events, initiated by moving prey.

FUNCTIONAL RESPONSE

The functional response of *R. trochantericus* was assessed separately at six different prey densities: 1, 2, 4, 6, 8 and 10 prey/predator of both wax moth larvae and its natural prey (TMB) for 5 days in glass rearing bottles. Five replicates were maintained for each category. At 24 h intervals the number of prey killed was recorded and the prey number was maintained at a constant level by the introduction of fresh prey throughout the experiment.

In the present study the 'disc' equation of Holling (1959) was used to describe the functional response of *R. trochantericus*.

The following parameters were used for obtaining the 'disc' equation:

x = prey density

y = total number of prey killed in given period of time (T_t)

y/x = attack ratio

T_t = total time in days when prey was exposed to the predator

b = time spent handling each prey by the predator (T_t / k)

a = rate of discovery per unit of searching time $[(y/x)/T_s]$

The handling time 'b' was estimated as the time spent for pursuing, subduing and feeding on each prey. The maximum predation was represented by the 'k' value and it was restricted to the higher prey density.

Another parameter 'a', the rate of discovery, was defined as the proportion of the prey attacked successfully by the predator per unit of searching time. Assuming that the predatory efficiency is proportional to the prey density and to the time spent by the predator in searching the prey (T_s), the expression of relationship is:

$$y = a T_s x \quad (1)$$

Since time available for searching is not a constant, it is deducted from the total time (T_t) by the time spent for handling the prey. If one presumes that each prey item requires a constant amount of time 'b' for consumption, then

$$T_s = T_t - by \quad (2)$$

Substituting (2) in (1), Holling's 'disc' equation is

$$y = a (T_t - by) x \quad (3)$$

The data were subjected to linear regression analysis (Daniel, 1987).

MOLECULAR CHARACTERIZATION

The molecular identification of *R. trochantericus* was based on the core principle of generating DNA barcode using mitochondrial cytochrome c oxidase I (mtCOI) gene. Total DNA was isolated from individual *R. trochantericus* in various life stages (egg, nymphal instars and adult) using a part by modified CTAB method (Saghai Maroof et al., 1984). The rest of the specimen was used as the specimen voucher and deposited in National Pusa Collection (NPC), New Delhi. A part of the specimen was ground with 1ml of 2% cetyl trimethyl ammonium bromide (CTAB), 100mM Tris-HCl (pH-8.0), 1.4 M sodium chloride, 20mM EDTA and 2% of 2-mercaptoethanol. The suspension was incubated at 65°C for 1-2 hours and then an equal volume of chloroform: isoamyl alcohol (24:1) solution was added. The suspension was centrifuged at 6000 rpm for 15 minutes. The aqueous layer was transferred to a fresh 2ml micro centrifuge tube taking care not to disturb the middle protein interface. DNA was precipitated by the addition of 20µl of 0.3 M sodium acetate and equal volume of ice-cold 95% ethyl alcohol. The precipitated DNA was spun at 8000 rpm for 10 minutes and the resultant DNA pellet was washed with 70% ethyl alcohol. This was centrifuged at 8000 rpm for 10 minutes and finally the pellet was dissolved in 50µl DNase, RNase free molecular biology water. The genomic DNA was visualized using 1% agarose gel and diluted with sterile water to get a working solution of 20-25ng/µl.

Polymerase chain reaction (PCR) was carried out in a thermal cycler (AB- Applied Biosystems) with the following cycling parameters: 94°C for 4 minutes as initial denaturation followed by 35 cycles of 94°C for 30 seconds, 47°C for 45 seconds, 72°C for 45 seconds and 72°C for 20 minutes as a final extension. The universal primer specific to mitochondrial cytochrome oxidase I (mtCOI) used for the amplification resulted in an approximately 700 bp fragment. PCR was performed in 25µl reaction volume containing 20 picomoles of each primer, 10mM Tris-HCl (pH-8.3), 50mM KCl, 2.5mM MgCl₂, 0.25 mM of each dNTP and 0.5 U of Taq DNA polymerase (Fermentas Life Sciences). The amplified product was resolved in 1% agarose gel and the remaining PCR product was eluted using Nucleospin Extract II according to the manufacturers protocol (MN, Germany) which is sequenced in an automated sequencer (ABI Prism 310; Applied Biosystems, USA) using M₁₃ universal primer both in forward and reverse direction.

RESULTS AND DISCUSSION

R. trochantericus laid elongately oval dark brown eggs (67.50 ± 15.01) with flower-like opercular architecture and glued basally to the substratum. Females laid eggs singly as well as in groups of up to 26 eggs in 3 to 7 clusters per female as observed in Harpactorines (Ambrose, 1999; Ambrose et al., 2006) (Fig. 1). The fecundity was higher than in other Harpactorines like *Sphedanolestes* spp. (15.33 ± 6.41 eggs) (Vennison and Ambrose, 1990) but lower than that of *R. mar-*

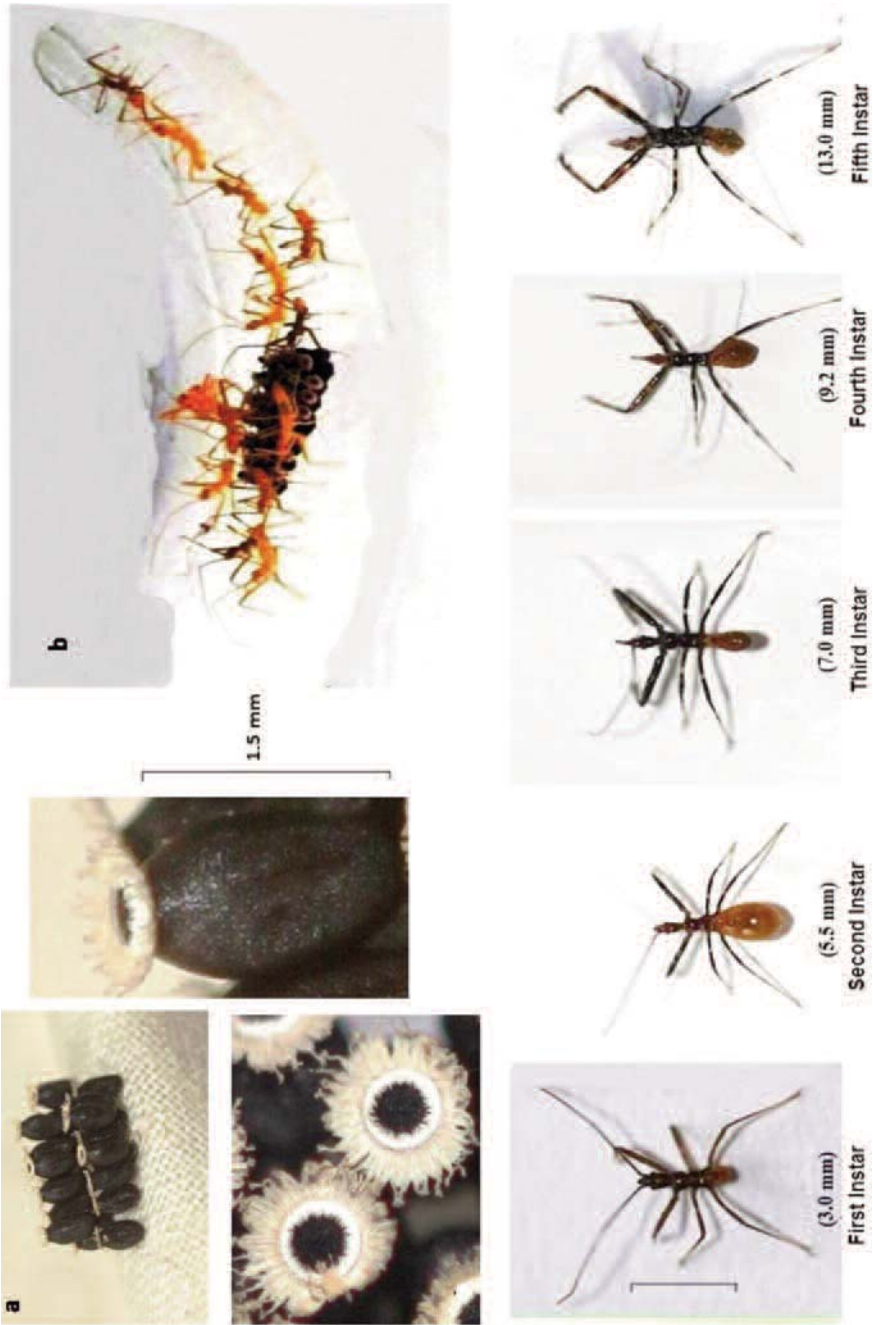


Fig. 1. a. Eggs of *Rihirbus trochantericus*, b. newly hatched nymphs, c. I-V nymphal instars.

Table 1. Biological parameters of *R. trochantericus* fed on wax moth larvae under laboratory condition (n = 47; $\bar{x} \pm SE$)

Parameters	(in days)
Incubation period (days)	13.00 \pm 0.69
Stadial period (days)	
I instar	12.39 \pm 1.13
II instar	7.00 \pm 0.39
III instar	7.56 \pm 0.35
IV instar	9.28 \pm 0.64
V instar	12.78 \pm 1.27
I -V instars	49.00 \pm 2.48
Total stadial period (days)	
Male	46.13 \pm 2.35
Female	51.30 \pm 4.02
Fecundity/female (no.)	67.50 \pm 15.01
Hatchability (%)	78.89
Survival rate (I-V) (%)	40.00
Sex ratio (female: male)	1:0.7
Preoviposition period (days)	17.88 \pm 0.72
Oviposition period (days)	34.62 \pm 3.49
Postoviposition period (days)	9.75 \pm 0.70
Adult longevity (days)	
Male	61.00 \pm 3.12
Female	66.60 \pm 5.73
Total longevity (days)	
Male	107.13 \pm 2.70
Female	117.90 \pm 3.83

ginatus Fab. (208.3 \pm 3.9 eggs) (Sahayaraj and Sathiamoorthi, 2002). The fertilized egg did not show any color change prior to hatching whereas the unfertilized eggs became shrunken.

The preoviposition period of *R. trochantericus* was shorter (17.88 \pm 0.72 days) than that of *R. marginatus* (33.3 days) and *R. kumarii* (26.0 days), and closer to *R. fuscipes* Fab. (19.0 days). The eggs hatched in 13.00 \pm 0.69 days and mean per cent egg hatchability was 78.89. A higher hatching percentage is a diagnostic feature of Harpactorines (Ambrose, 1999; Ambrose et al., 2006).

The incubation period of *R. trochantericus* was longer than *C. spiniscutis* Bergröth (4.66 \pm 0.77 days) (Claver and Reegan, 2010), *S. signatus* (9.6 \pm 0.86 days) (Vennison and Ambrose, 1990) and *Sycanus collaris* Fab. (15.0 days) but shorter than *Panthous bimaculatus* Dist. (21.0 days) (Sahayaraj, 2012). The eclosion last-

ed for about 3 to 4 minutes. The newly hatched nymphal instars started feeding 6 to 7 hrs after eclosion, showing a preference for small and sluggish prey.

The stadial durations of I, II, III, IV and V nymphal instars were 12.39 ± 1.13 , 7.00 ± 0.39 , 7.56 ± 0.35 , 9.28 ± 0.64 and 12.78 ± 1.27 days, respectively (Table 1).

The total developmental period of *R. trochantericus* from egg to adult was 49.00 ± 2.48 days. It was shorter than that of *S. collaris* (75.67 ± 9.06), *R. kumarii* (88.30 ± 3.60) and *Panthous bimaculatus* Distant (101.12 ± 2.30) (Sahayaraj, 2012). Abnormal hatching and molting caused 60% nymphal mortality from I to V instars and thus the nymphal instars had a survival rate of 40%. The nymphal mortality of *R. trochantericus* was lower when compared to *Sphecanolestes pubinotum* Reuter (89.30%) and greater than that of *S. minusculus* Bergroth (21.06%) and *S. himalayensis* (13.0%) (Ambrose, 1999). The adult male longevity and total male longevity were shorter (61.00 ± 3.12 and 107.13 ± 2.70 days) than that of the female (66.60 ± 5.73 and 117.9 ± 3.83 days). The pre-oviposition and postoviposition periods were 17.88 ± 0.72 and 9.75 ± 0.70 days, respectively. The oviposition period of *R. trochantericus* lasted for 34.62 ± 3.49 days. The laboratory-emerged adults exhibited female biased sex ratio (1:0.67). Among harpactorines, the female biased sex ratio was also observed in *S. collaris* (1:0.67), *R. kumarii* (1: 0.50) and *P. bimaculatus* (1:0.60) (Sahayaraj, 2012). Our laboratory breeding experiments indicated that *R. trochantericus* is a multi-voltine species.

There exists distinct sexual dimorphism in *R. trochantericus* in size, shape and color. Females are larger (19.2-20.1 mm length) and stout with a conical abdominal base, whereas males are relatively small (13.2-15.1 mm), and lean with a round abdominal base. In addition, there is a distinct coloration difference; females are uniformly black while males have dull brownish yellow in the rostrum, thorax, pronotum, scutellum, anterior wings and legs up to the femur (Fig. 2).

R. trochantericus attacked prey in a sequential pattern: arousal -approach -capturing -probing -piercing and sucking (Fig. 3). The post-predatory cleaning was also observed as in other nontibial pad harpactorine reduviids (Ambrose, 1999).

R. trochantericus mated in the laboratory in the following sequence: arousal-approach-riding over-extension of genitalia-copulation, -ejection of spermatophore capsule by the female, and post mating cleaning. The mating behavior of *R. trochantericus* with the characteristic riding over phenomenon represented its harpactorine character (Ambrose, 1999).

The reduviid responded to increasing prey density of wax moth larvae and TMB by killing more prey than at lower prey densities (Table 2 and Table 3). It exhibited a typical functional response and thus established the applicability of the second model of Holling's 'disc' equation. The type II functional response is typical of most heteropteran predators (Cohen and Byrne, 1992). The number of prey killed (y) by the individual predator increased as the prey density (x) increased from one prey/predator (Fig. 4). This was further confirmed by the pos-

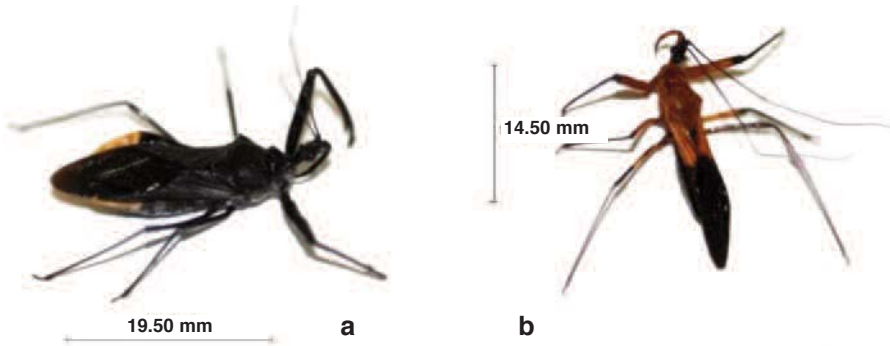


Fig. 2. a. ♀ *Rihirbus trochantericus*, b. ♂ *R. trochantericus*



Fig. 3. *R. trochantericus* feeding on; a. on wax moth larva, b. on TMB.

itive correlation ($r = 0.9867$ for wax moth larvae and $r = 0.9738$ for TMB) obtained between the prey density and prey killed. A similar result (positive correlation between prey density and prey killed) was obtained by Claver et al. (2004) and Ravichandran and Ambrose (2006).

Increase in the number of prey killed by an individual predator as a function of increasing prey density confirmed earlier reports of Ambrose and Claver (1997) and Ravichandran and Ambrose (2006). The maximum predation was represented by 'k' value ($r = 1.92$ for wax moth larvae and $r = 3.88$ for TMB)

Table 2. Functional response of *R. trochantericus* to wax moth larva

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 ($\frac{y}{x} = a$)	Disc equation (y')
1	0.32			0.83	4.17	0.32	0.08	
2	0.60			1.56	3.44	0.30	0.09	
4	1.08	1.92	2.60	2.81	2.19	0.27	0.12	$y' = 1.16(5 - 2.60y)x$
6	1.36			3.54	1.46	0.23	0.16	
8	1.80			4.69	0.31	0.23	0.72	
10	1.92			5.00	0.00	0.19		

Table 3. Functional response of *R. trochantericus* to TMB

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 ($\frac{y}{x} = a$)	Disc equation (y')
1	0.60			0.77	4.23	0.60	0.14	
2	1.44			1.86	3.14	0.72	0.23	
4	2.16	3.88	1.29	2.78	2.22	0.54	0.24	$y' = 2.03(5 - 1.29y)x$
6	3.16			4.07	0.93	0.53	0.57	
8	3.48			4.48	0.52	0.44	0.84	
10	3.88			5.00	0.00	0.39	0.00	

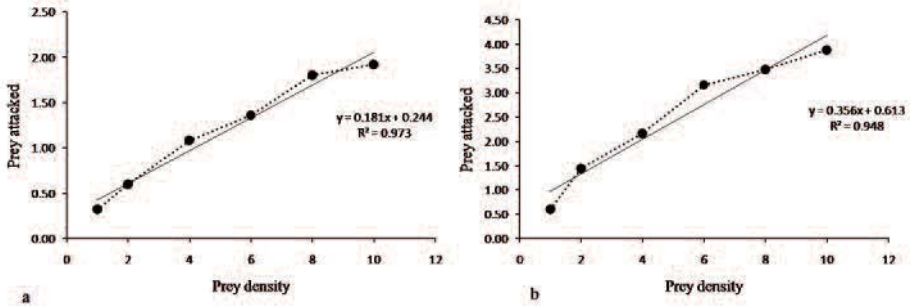


Fig. 4. Linear regression between Prey density and Prey attacked (a) wax moth larvae (b) TMB.

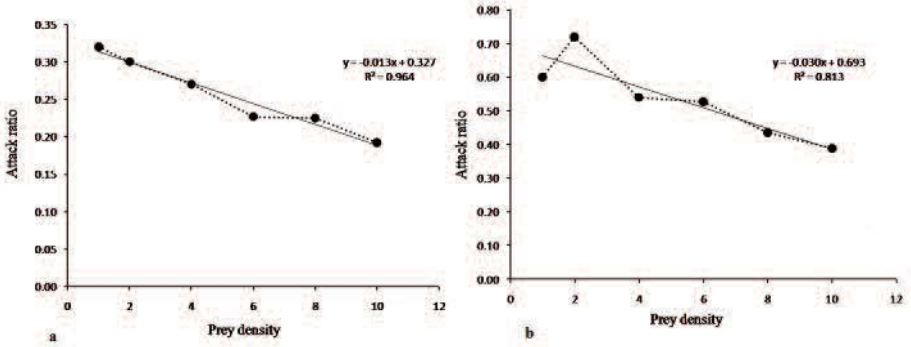


Fig. 5. Linear regression between Prey density and Attack ratio (a) wax moth larvae (b) TMB.

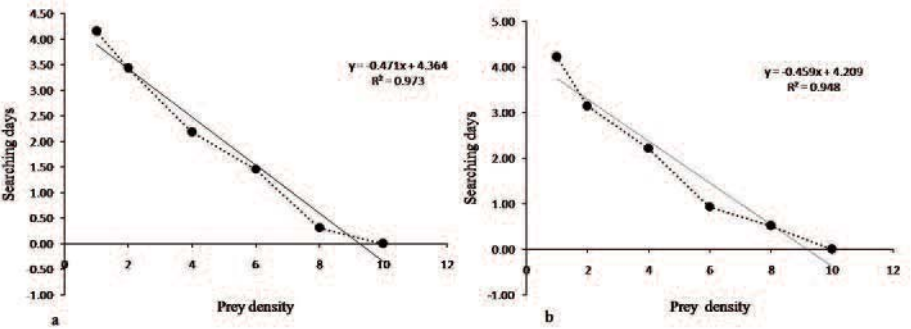


Fig. 6. Linear regression between Prey density and searching days (a) wax moth larvae (b) TMB.

which was always found restricted to the higher prey densities. The highest attack ratio was observed at the density of 1 and 2 prey/predator and the lowest attack ratio was found at the density of 10 prey/predator. Hence, the attack ratio decreased as the prey density increased ($r = -0.9818$ for wax moth larvae and $r = -0.9019$ for TMB) (Fig. 5). An indirectly proportional relationship was found between the attack ratio and the prey density which is similar to the observations of Ambrose et al. (2009) in *S. variabilis*. It is presumed that the predator took less time on nonsearching activities, which in return might have caused a perceptible decline in the attack rate until hunger was established (Claver et al., 2004). This type of searching time was also observed in *R. marginatus* to *Clavigralla gibbosa* Spinola and *Hieroglyphus banian* (F.) (Ambrose et al., 2000). Hassell et al. (1976) stated that the attack rate decreased with increasing prey density with predators having a type II functional response. A negative correlation was obtained between prey density and searching time ($r = -0.9867$ for wax moth larvae and $r = -0.9738$ for TMB) of the predator at all prey densities (Fig. 6). At a density of six TMB, the searching efficacy and rate of consumption were at their maximum. Thus it can be estimated that *R. trochantericus* released at a ratio of 1:6 (predator-prey) may be optimal to realize the biological control potential of this predator in the cashew ecosystem.

Mitochondrial COI gene was sequenced successfully from all the different life stages (Fig. 7). A comparison of the sequences for all the life stages of *R. trochantericus* showed no differences or mismatches, indicating that the barcode developed was unique without sequencing errors. A total fragment size of 658 bp DNA sequence was deposited in NCBI-GenBank database with accession number KC 834736. Sequences were confirmed by similarity search in GenBank Blastn. Since the Folmer region primers (LCO-1490 and HCO-2198) (Hebert et al., 2003a, b) are highly conserved, the same applied to *R. trochantericus*. Evidence of nuclear copies was not observed which was supported by the absence of stop codons within the sequences and the base composition was similar with no indels across various life stages. Thus the present studies indicate that the barcode developed will be useful in confirming the identity of the species in any of its life stages.

This is the first description of the biology and behavior of *R. trochantericus*. The molecular characterization made of *R. trochantericus* will be highly useful in confirming the identity of the species in any of its life stages. The findings may help to improve the future IPM strategies against *Helopeltis* in cashew.

ACKNOWLEDGEMENTS

The senior author is grateful to the Indian Council of Agricultural Research, Government of India, for financial assistance (OPR on management of sucking pests of horticultural crops). The authors are grateful to the Director, Directorate of Cashew Research, for institutional facilities. Our thanks are due to Dr. Dunston P. Ambrose, Entomology Research Unit, St. Xavier's College, Palayamkottai, for morphologically identifying *R. trochantericus*.

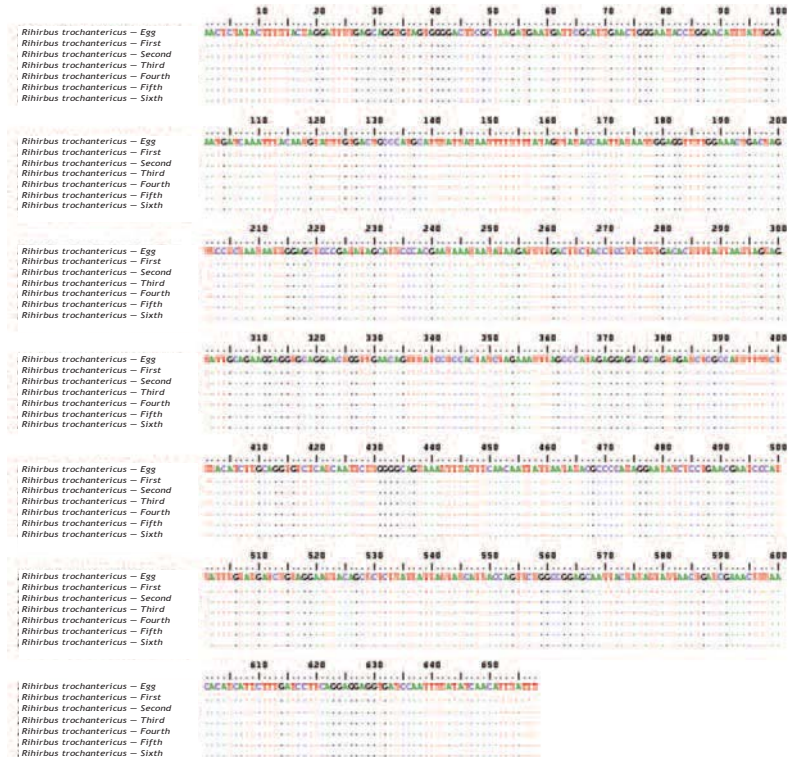


Fig. 7. Consensus sequence of 658 bp from the mitochondrial cytochrome oxidase I (mtCOI) gene for the *R. trochantericus* in their various life stages viz. egg, different instars and adult.

LITERATURE CITED

- Ambrose, D. P.** 1999. Assassin bugs. New Hampshire, USA: Science publishers and New Delhi, India: Oxford and IBH Publishing Co. Pvt. Ltd. pp. 337.
- Ambrose, D. P.** 2000. Substrata impact on mass rearing of the reduviid predator *Rhynocoris kumarii* Ambrose and Livingstone (Heteroptera: Reduviidae). *Journal of Entomological Research* 24: 337-342.
- Ambrose, D. P.** 2003. Biocontrol potential of assassin bugs (Hemiptera: Reduviidae). *Journal of Experimental Zoology* 6: 1-44.
- Ambrose D. P.** 2006. A checklist of Indian assassin bugs (Insecta: Hemiptera: Reduviidae) with taxonomic status, distribution and diagnostic morphological characteristics. *Zoos Print Journal* 21: 2388-2406.
- Ambrose, D. P. and D. Livingstone.** 1979. On the bioecology of *Lophocephala guerini* Laporte. (Reduviidae: Harpactorinae) a coprophagous reduviid from Palghat gap. *Journal of Natural History* 13: 581-588.

- Ambrose, D. P. and M. A. Claver.** 1997. Functional and numerical response of the reduviid predator, *Rhynocoris fuscipes* F. (Het., Reduviidae) to cotton leafworm *Spodoptera litura* F. (Lep., Noctuidae). *Journal of Applied Entomology* 121: 331-336.
- Ambrose, D. P., M. A. Claver, and P. Mariappan.** 2000. Functional response of *Rhynocoris marginatus* (Heteroptera: Reduviidae) to two pests of pigeon pea (*Cajanus cajan*). *Indian Journal of Agricultural Sciences* 70: 630-632.
- Ambrose, D. P., S. P. Kumar, K. Nagarajan, S. M. Das, and B. Ravichandran.** 2006. Redescription, biology, life table, behaviour and ecotypic diversity of *Sphedanolestes minusculus* Bergroth (Hemiptera: Reduviidae). *Entomologica Croatia* 10: 47-66.
- Ambrose, D. P., S. K. Gunaseelan, J. V. Singh, B. Ravichandran, and K. Nagarajan.** 2007. Redescription, biology and behaviour of a harpactorine assassin bug *Endochus migratorius* Distant. *Hexapoda* 14: 12-21.
- Ambrose, D. P., S. J. Rajan, K. Nagarajan, V. J. Singh, and S. S. Krishnan.** 2009. Biology, behaviour and functional response of *Sphedanolestes variabilis* Distant (Insecta: Hemiptera: Reduviidae: Harpactorinae), a potential predator of lepidopteran pests. *Entomologica Croatia* 13: 33-44.
- Biswas, B., G. C. Sen, and L. K. Ghosh.** 1994. (Hemiptera: Heteroptera: Reduviidae) State Fauna Series 3: Fauna of West Bengal 5: 369-411.
- Biswas, B. and B. Animesh.** 2010. (Hemiptera: Heteroptera: Reduviidae) State Fauna Series, Fauna of Uttarakhand 18: 255-268.
- Cai, W. and M. Tomokuni.** 2003. *Camptibia obscura*, gen. and sp. n. (Heteroptera: Reduviidae: Harpactorinae) from China. *European Journal of Entomology* 100: 181-185.
- Claver, M. A., M. S. A. Muthu, B. Ravichandran, and D. P. Ambrose.** 2004. Behaviour, prey preference and functional response of *Coranus spiniscutis* (Reuter), a potential predator of tomato insect pests. *Pest Management in Horticultural Ecosystem* 10: 19-27.
- Claver, M. A. and Reegan A. D.** 2010. Biology and mating behaviour of *Coranus spiniscutis* Reuter (Hemiptera: Reduviidae), a key predator of rice gandhi bug *Leptocorisa varicornis* Fabricius. *Journal of Biopesticides* 3: 437-440.
- Cohen, A. C. and D. W. Byrne.** 1992. *Geocoris punctipes* and predator of *Bemisia tabacci*, a laboratory evaluation. *Entomologica Experimentalis et Applicata* 64: 195-202.
- Daniel, W.** 1987. *Biostatistics: A foundation analysis in the health sciences*. John Wiley & Sons, New York. 734 pp.
- Distant, W. L.** 1904. *The Fauna of British India, including Ceylon and Burma*. Ryn. 2. Taylor & Francis, London. pp. 196-403.
- Hassell, M. P., J. H. Lawton, and J. R. Beddington.** 1976. The components of arthropod predation 1. The prey death rate. *Journal of Animal Ecology* 45: 135-164.
- Hebert, P. D. N., A. Cywinska, S. L. Ball, and J. R. deWaard.** 2003a. Biological identifications through DNA barcodes. *Proceedings of the Royal Society of London, Series B* 270: 313-321.
- Hebert, P. D. N., S. Ratnasingham, and J. R. deWaard.** 2003b. Barcoding animal life: cytochrome c oxidase subunit 1 divergences among closely related species. *Proceedings of the Royal Society of London Series B* 270: S96-S99.
- Holling, C. S.** 1959. Some characteristics of simple types of predation and parasitism. *Canadian Entomologist* 91: 385-398.

- Maldonado, C. J.** 1990. Systematic catalogue of the Reduviidae of the world (Insecta: Heteroptera). A special edition of Caribbean Journal of Science. 694 pp.
- Miller, N. C. E.** 1940. New genera and species of Malaysian Reduviidae. Part I. Journal of the Federated Malay States Museums 18: 419-599.
- Naik, B. G. and D. Sundararaju.** 1982. In CPCRI Annual Report. Central Plantation Crops Research Institute, Kasaragod, India. 134 pp.
- Putshkov V. G. and P. V. Putshkov.** 1985. A catalogue of assassin bugs genera of the world (Heteroptera: Reduviidae). VINITI, Moskva. 138 pp.
- Ravichandran, B. and D. P. Ambrose.** 2006. Functional response of a reduviid predator *Acanthaspis pedestris* Stal (Hemiptera: Reduviidae) on three lepidopteran insect pests. Entomon 31: 1-9.
- Sahayaraj, K.** 2012. Artificial rearing on the nymphal developmental time and survival of three reduviid predators of Western Ghats, Tamil Nadu. Journal of Biopesticides 5: 218-221.
- Sahayaraj, K. and P. Sathiamoorthi.** 2002. Influence of different diets of *Corcyra cephalonica* on life history of a reduviid predator *Rhynocoris marginatus* (Fab.) Journal of Central European Agriculture 3: 54-61.
- Saghai Maroof, M. A., K. M. Soliman, R. A. Jorgensen, and R. W. Allard.** 1984. Ribosomal DNA spacer-length polymorphisms in barley: Mendelian inheritance, chromosomal location and population dynamics. Proceedings of the National Academy of Science, USA 81: 8014-8018.
- Stal, C.** 1874. *Enumeratio Hemipterorum* 4. K. Svenska Vetensk Acad. Handl. (N.F) 11: 1-182.
- Sundararaju, D.** 1984. Cashew pests and their natural enemies in Goa. Journal of Plantation Crops 12: 38-46.
- Vennison S. J. and D. P. Ambrose.** 1990. Biology and behaviour of *Sphedanolestes signatus* Distant (Insecta: Heteroptera: Reduviidae) a potential predator of *Helopeltis antonii* Signoret. Uttar Pradesh Journal of Zoology 10: 30-43.