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First record of a rare masked bee *Hylaeus* (*Indialaeus*) *strenuus* (Cameron, 1897) from south India with an updated checklist of *Hylaeus* species (Hymenoptera, Colletidae)

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ABSTRACT: A rare species of masked bee *Hylaeus strenuus* (Cameron, 1897) is reported for the first time from south India. Diagnosis of the species together with the illustrations of morphological characters is presented. Images of the trap nest and nest parameters are provided. A revised checklist of all the known species of the genus *Hylaeus* from south India along with species distribution map is also provided.

KEYWORDS: Apoidea, new report, diagnosis, checklist, distribution

INTRODUCTION

Bees in the genus *Hylaeus* Fabricius, 1793 (Family Colletidae) are one of the small sized bees (forewing length: 5–8mm) which are relatively hairless and wasp-like, with a prominent yellow/white paraocular markings on the face (yellow faced bees or masked bees). These are also called plasterer bees because of their unique nature of lining their nest cells with a self-secreted cellophane-like material. Cell lining is made of lipids and proteins which is a transparent water proof membrane, insoluble in different solvents and resistant to fungal attack (Almeida, 2008). Unlike most bees, *Hylaeus* carry pollen internally in the crop instead of on body hairs (Michener, 2007). Many species nest in preexisting cavities in plant stems and twigs, plant galls, beetle borings, old cells of bees and wasps and some nest in the ground (Michener, 2007; Scheuchl and Willner, 2016). *Hylaeus* bees are poorly known from the Oriental region in general (Snelling, 1980; Dathe, 2011; Magnacca *et al*., 2011). According to Ascher and Pickering (2023), though there are about 768 species of the genus *Hylaeus* worldwide only 18 species are recorded from India, six of which occur in south India (Saini *et al*., 2021).

In this paper, *H.* (*Indialaeus*) *strenuus* (Cameron, 1897) is reported, not previously documented to occur in south India. This species shows rare distribution across its range, with earlier records from Gujarat, Sikkim and West Bengal (Saini *et al*., 2021). In 2007, this species was reported from O'ahu, Hawaii (United States), as a recent introduction from India (Dathe, 2011; Magnacca *et al*., 2011, 2013). Nothing is known about its biology in its native range. However, like *Ceratina*

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smaragdula (Fabricius, 1787) (Apidae), this species is reported to nest in twigs and collect pollen from *Scaevola sericea* Vahl and *Scaevola taccada* (Gaertn.) Roxb. (Goodeniaceae), *Heliotropium foertherianum* (Blanco) Mabb. (Boraginaceae), *Erythrina sandwicensis* O. Deg. (Fabaceae) and *Metrosideros polymorpha* Gaudich. (Myrtaceae) in Hawaii (Magnacca *et al*., 2011; Magnacca and King, 2013). During a survey in south India, for the first time this species was collected from artificial trap nests installed in Chintamani, Karnataka. Brief diagnosis of the species together with the illustrations of morphological characters is given. Nest parameters are discussed. An updated checklist of species of the genus *Hylaeus* from south India is presented, with a distribution map.

MATERIALS AND METHODS

Specimens were obtained from artificial trap-nest (Bamboo stem pieces 34.00cm long and 2.00cm outer diameter) that were installed in the bee park (13°20’08"N; 78°04’55"E, 858m) at College of Sericulture, Chintamani, Karnataka, India (Fig. 3D). The study site recorded a mean maximum temperature of 30.10°C, mean minimum temperature of 18.58°C, mean precipitation of 153.69 mm, mean relative humidity of 71.81per cent, and wind velocity of 7.90 km h^{-1}.

Identification of bees followed original descriptions and keys (Cameron, 1897; Snelling, 1980; Dathe, 2011; Saini *et al*., 2021). For external morphological studies, a Nikon SMZ 800N microscope was used. Digital colour images of important diagnostic characters and habitus of species were taken using Leica SAPO stereomicroscope with Flexacam C3 camera attachment. Images were edited with Adobe Photoshop CS (Version 9.0). Morphological characters used mainly follow the terminology used by Michener (2007), Snelling (1980), Dathe (2011) and Magnacca *et al*. (2011). All measurements were taken as the maximal length of body parts measured using a Leica SAPO stereomicroscope. Body length was measured from the anterior margin of head to the posterior margin of metasomal tergum 2 (T2). Head: length- mid-ocellus to apical margin of clypeus and maximum width - between outer margins of eyes as in front view. Nest parameters like number of cells built, cell length, the diameter of the entrance, type of the material used for remodeling nests and cell partitions and other parameters were recorded.

Voucher specimens from this study are deposited in the Insect Museum, Department of Entomology, University of Agricultural Sciences, Gandhi Krishi Vignana Kendra, Bangaluru Karnataka, India. The checklist along with geographical distribution data of species was prepared by extracting published faunal records (Cameron, 1897; Snelling, 1980; Dathe, 2010; Dathe, 2011; Magnacca *et al*., 2011; Saini *et al*., 2021) and world checklist (Ascher and Pickering, 2023). Mapinfo Professional 7.5SCP was used for generating the distribution map of species.

RESULTS AND DISCUSSION

Systematic accounts

Family: Colletidae

Sub family: Hylaeinae

Genus: *Hylaeus* Fabricius, 1793

Subgenus: *Indialaeus*

Prosopis strenua Cameron, 1897. Loc. typ.: Barrackpore (Barakpur), West Bengal, India.

= *Hylaeus (Indialaeus) strenuus* (Cameron, 1897) sensu Dathe (2010)

***Hylaeus* (*Indialaeus*) *strenuus* (Cameron, 1897) Figs. 1, 2**

Hylaeus strenuus (Cameron, 1897)

Prosopis striatifrons Cameron, 1897: 89♀ (syn.) according to Dathe (2010: 66- 67).

Prosopis strenua Cameron, 1897: 91♂(syn.) according to Dathe (2010: 66- 67).

Braunsapis chandrai Gupta & Sharma in Gupta *et al*., 2015: 373 –♂(syn.) according to Saini *et al*., 2021

Material examined: INDIA: Karnataka: Chintamani, 13°20’08"N; 78°04’55"E, 858 m, 4♀ and 1♂, 25.iii.2022, Trap nest, Coll. Arati Pannure; 1♀, 14. iv.2023, Coll. Manjunath, K.L.; 1♀, 1.vi.2023, Coll. Arati Pannure; 865m, 13°16' N; 78°

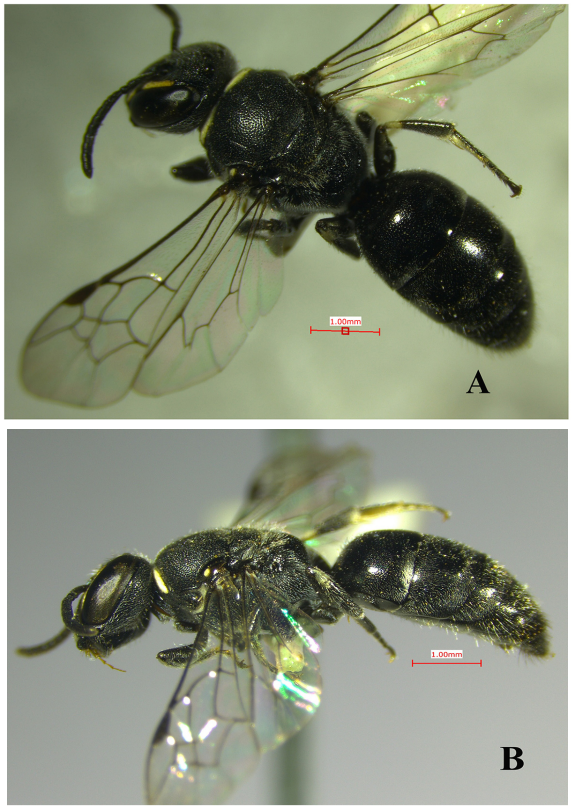

Fig. 1 Habitus of *Hylaeus (Indialaeus) strenuus* (Cameron, 1897), -Female
A - dorsal view; B - lateral view. Scale bar=1mm

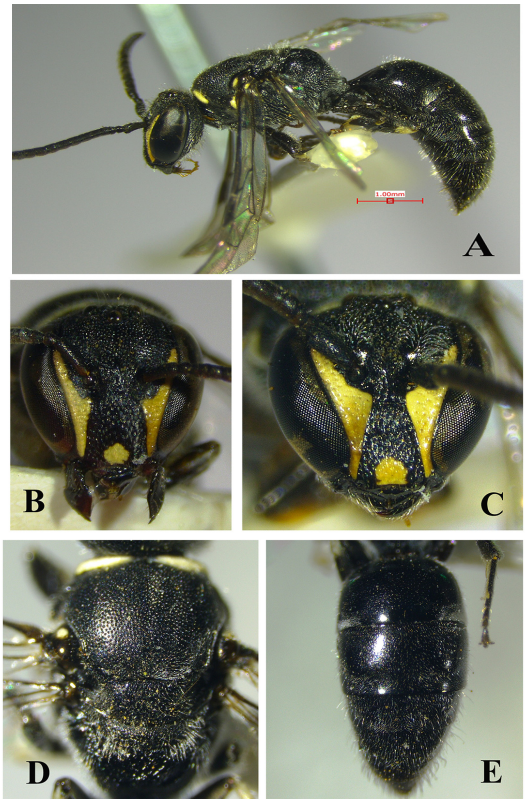

Fig. 2 A, Habitus of *Hylaeus* (*Indialaeus*) *strenuus* (Cameron, 1897), Male, lateral view; B, C, head, frontal view (Facial maculation patterns of female (B) and male (C)); D, Mesosoma (Female); E, Abdomen (Female)

12' E, 1♀, 22.iii.2023, Coll. Pampareddy.

Measurements: Female. Body length: 4.40-4.80 mm (Head + mesosoma + T1+T2). Head: median length- 1.45mm (n=4) & maximum width - 1.83 (n=4). Scutum median length – 1.19 and maximum width 1.57. Total length of fore wing 5.04mm.

Male. Body length: 4.27 mm (n=1). Head: median length- 1.40mm and maximum width - 1.72 (n=4). Scutum median length – 0.97mm and maximum width 1.50. Total length of fore wing 4.60mm.

Diagnosis: The species differs from the other known species of the *Hylaeus* in India by the following characters: This species possesses strong and coarse punctures on head and mesosoma. Mesopleura with smooth ground sculpture, highly shiny especially in males. Clypeus striate-punctate; supraclypeal area and area above it on frons finely strigate. Orbits strongly convergent below. Lateral margin of clypeus distinctly separated from inner eye margin (Figs. 2B, C). Mandibles clearly bidentate; preapical notch of mandible strong, a distinct preapical tooth present. **Female** mandibles are unusually broad (Fig. 2B). Median area of the propodeum is rugose only medially, with distinct smooth impunctate area laterad; bordered by a stout transverse semicircular keel/carina which is incomplete medially. **Males:** Copulatory apparatus with long setae; apical lobe of sternum 8 with setae. Males have long, attenuate gonoforceps which protrude slightly beyond the apex of the abdomen even when retracted. Both sexes have first tergum with apicolateral pubescent patch of appressed, plumose pubescence; the apical area of second metasomal tergite distinctly depressed, with the posterior rim shining, impunctate, and reflexed upward (Fig. 2E).

Fig. 3 A, Linear array of nest cells of *H.* (*Indialaeus*) *strenuus* with cellophane-like cell lining in bamboo trap nest. B , C, individual cells showing length of the cell and empty space between cells. D. Artificial trap-nests (Bee hotel) at bee park at College of Sericulture, Chintamani, Karnataka, India from where nests of *H.* (*Indialaeus*) *strenuous* collected

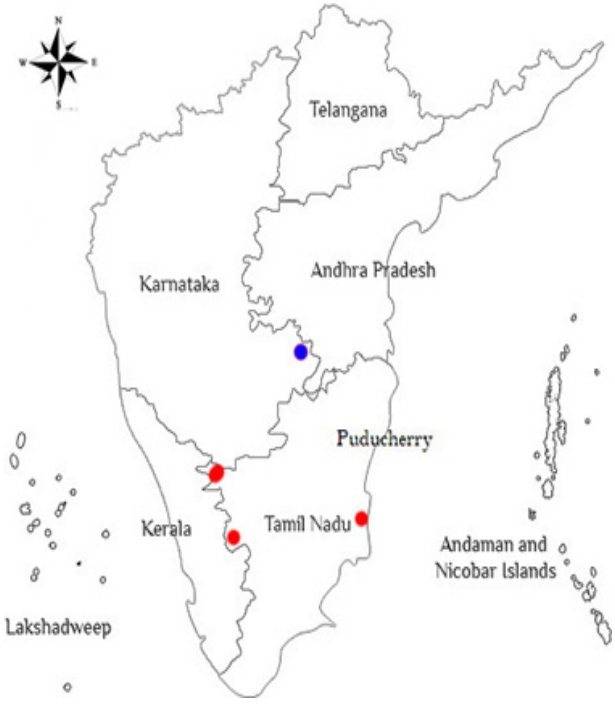

Fig. 4 Distribution map of *Hylaeus* species in south India. Blue point in the map (●) indicates location of Chintamani, Karnataka from where *Hylaeus* (*Indialaeus*) *strenuus* (Cameron, 1897) was collected. Red points (●) indicate the places where the other species of *Hyalaeus* have been recorded

Table 1. Nest parameters of *H. strenuus*

Cell No.	Length of the cell (mm)	Empty space between cells (mm)
1	5.69	1.20
2	5.75	1.24
3	5.75	1.24
4	5.74	1.24
5	5.74	2 cells: 3.09; Left cell: 1.84 Right cell: 1.24
6	5.73	0.94
7	5.73	0.67
8	5.82	1.24
9	5.62	1.08
10	5.62	1.08
11	5.62	1.08
12	6.24	1.02
13	6.24	-
Mean ± STD	5.79 mm ± 0.21 (n=13)	1.11 ± 0.17 (n=12)

Coloration: Blackish. The following ivory: Both males and females have elongate lateral face marks and a small anteromedial clypeal mark (Fig. 2B–C); pronotal collar interrupted in middle (Fig. 2D); posterior part of pronotal lobe in male (Fig. 2A); an anterior spot on tegulae (Fig. 2D). Female: Only hind tibiae with ivory spot at base. Mandibles black, ferruginous at tip. Flagellum dark brown, slightly paler beneath. Wings clear, veins and stigma brown; apex of fore wings little smoky.

Nest parameters: Trap nest made of bamboo occupied by *H. strenuus* in March, 2022 was collected and stored in an emergence cage. From the nest, 4 females and one male emerged during last week of March & first week of April, 2022. In 2023, 3 females were collected from artificial trap nest (at bee hotel, Fig. 3D) during March to June. The traps had an entrance diameter of 3.8 mm and nest was 95-98 mm long. Trap nest consisted of linear series of 13 cylindrical sack like cells (Fig. 3A-C). This species constructed cells out of a clear single-layered cellophane-like transparent lining which was lightly adhered to the trap nest walls. Brood cell length varied between 5.62 – 6.24 mm with the average brood cell length of 5.79 mm ± 0.21 (n=13) (Table 1). The length of the empty space between cells varied from 0.67 mm to 1.24 mm with the average length of 1.11 ± 0.17 (n=12). Nest was observed only after the emergence of the adults, no vestibular and intercalary cells were differentiated. However, after the empty space of fifth cell, a short cell of 1.84 mm length was observed.

Peak activity Period: March to June

Distribution: India: Gujarat, Jharkhand, Karnataka (new record), Sikkim, and West Bengal. Elsewhere: United States (Hawaii), Singapore?

Checklist of known species of *Hyalaeus* from south India:

1. *H.* (*Indialaeus*) *parmatus* Snelling, 1980

H. parmatus Snelling, 1980: 10♂.

Distribution: India: Tamil Nadu (Ascher and Pickering, 2023).

2. *H.* (*Indialaeus*) *peltates* Snelling, 1980

H.peltates Snelling, 1980: 12♂.

H. eurygnathus Snelling, 1980: 14♀ (syn.) according to Dathe, (2011: 257)

Distribution: India: Karnataka, Tamil Nadu (Saini *et al*., 2021; Ascher and Pickering, 2023).

3. *H.* (*Indialaeus*) *sedens* Snelling, 1980

H. sedens Snelling, 1980: 13♀.

Distribution: India: Puducherry, Tamil Nadu (Saini *et al*., 2021). Elsewhere: Sri Lanka (Karunaratne *et al*., 2005; Ascher and Pickering, 2023).

4. *H.* (*Indialaeus*) *strenuus* (Cameron, 1897)

H. strenuus (Cameron, 1897)

Prosopis striatifrons Cameron, 1897: 89♀ (syn.). according to Dathe (2010: 66- 67).

P. strenua Cameron, 1897: 91♂ (syn.). according to Dathe (2010: 66- 67).

Braunsapis chandrai Gupta & Sharma in Gupta *et al*., 2015: 373 –♂(syn.) according to Saini *et al.,* 2021

Distribution: India: Gujarat, Jharkhand, **Karnataka** (new record), Sikkim, West Bengal (Ascher and Pickering, 2023). Elsewhere: United States (Hawaii) (Magnacca *et al*., 2011; 2013; Ascher and Pickering, 2023), Singapore? (Ascher *et al*., 2022).

5. *H.* (*Indialaeus*) *thyreus* Snelling, 1980

H. thyreus Snelling, 1980: 7♂.

Distribution: India: Tamil Nadu (Ascher and Pickering, 2023).

6. *H. oresbius* Snelling, 1980

H. oresbius Snelling, 1980: 5♂.

Distribution: India: Tamil Nadu (Ascher and Pickering, 2023).

7. *H. porcatus* Snelling, 1980

H. porcatus Snelling, 1980: 7♂.

Distribution: India: Tamil Nadu (Ascher and Pickering, 2023).

Of the 18 species that are known from India, seven

occur in south India including a newly recorded *H. strenuus*. Most of the Indian species of *Hylaeus* are known only from the type specimens (majority from males only) or at the most, from two or three localities. Based on earlier records from India it can be assumed that *H.* (*Indialaeus*) *strenuus* is clearly a rare species and is known by only a few recent records and small extent of occurrence from most parts of its range despite many years of intensive field surveys. Little is known about the biology of this species. Therefore, the documentation of this species from south India assumes significance for taking up conservation strategies and management.

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Indian record of the old world psyllid *Heterotrioza chenopodii* (Reuter) (Hemiptera, Psylloidea, Triozidae) on quinoa *Chenopodium quinoa* (Amaranthaceae)

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ABSTRACT: The Old-World Psyllid, *Heterotrioza chenopodii* (Hemiptera, Psylloidea, Triozidae) was found infesting quinoa, *Chenopodium quinoa* (Amaranthaceae) crop. The psyllids were identified using taxonomic traits and further confirmation with mitochondrial marker based molecular approach. The information on damage and its life stages is reported along with the phylogenetic information of the species. The identity analysis in NCBI indicated that the MT-COI sequences of *H. chenopodii* were 96 per cent identical to the previously deposited sequences with NCBI and five submissions with accession numbers were made *viz*., OP735496, OP740826, OP740828, OP740829 and OP740830. The phylogenetic tree represented the similarities in the analyzed sequences Indian populations are found to be merged in between the other similar global populations.

KEYWORDS: Triozid , taxonomic traits, MT-COI analysis, phylogenetic information

INTRODUCTION

Jumping plant-lice (psyllids), belong to Hemiptera; Psylloidea, are phloem sap-sucking insects that severely damage their host plants (Burckhardt *et al.,* 2006 a,b; Dzokou *et al.,* 2009). Approximately 4000 species in eight families so far around the world are described in Psylloidea and primarily habituated in the tropics and southern temperate zones (Burckhardt and Ouvrard 2012; Spodek *et al.,* 2017; Burckhardt *et al.,* 2022).The nymphs and adults inject their saliva into plant tissues, causing the entire plant to degenerate, leaves to warp, and leaves and stems to necroses (Dzokou *et al.,*2009). They make the plants sick and excretes honeydew which invites sooty mould growth, impairing plant growth (Burckhardt *et al.,* 2004). Numerous species of psyllids have the ability to cause galls in their host plants. Other psyllids are also known to be the carriers of bacterial and phytoplasma plant diseases (Mathur, 1975; Burckhardt and Lauterer, 1997).

The species from Triozidae family, *Heterotrioza chenopodii* (Reuter, 1876) reported in this study, is associated with Amaranthaceae (Halperin *et al.,*

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1982; El Nasr and Abd-Rabou, 2012). It feeds on a variety of Amaranthaceae plants including *Amaranthus* sp., *Atriplex halimus*, *A. tatarica*, *Chenopodium album*, *C. glaucum*, *C. quinoa*, *Halimione portulacoides*, *Beta vulgaris* and *Spinacia oleracea* (Mathur, 1975; Aguiar and Martin, 1999; Spodek *et al.,* 2017; Ouvrard, 2020). Initially recorded from Finland (Reuter, 1876), *H. chenopodii* is now globally reported in new geographical locations (Mathur, 1975; Horton *et al.,* 2018; Mifsud, 2020; Ouvrard, 2020; Percy *et al.,* 2020; Haouas *et al.,* 2021; Soliman *et al.,* 2021). *H. chenopodii* induces leaf deformations and yellowing on the host plants. Early-instar nymphs induce the plant tissue to produce galls on their hosts while feeding from within the leaf folds, whereas fourth and fifth instars feed freely on leaves, stems, petioles and inflorescences (Lauterer, 1982). The nymphal stage rarely moves and prefers feeding on occluded surfaces. At severe stages of infestation crop death was also observed (Soliman *et al.,* 2021). In this paper, the occurrence of *H. chenopodii* into southern India is reported. The species identity was confirmed by examination of morphological traits and molecular markers, mitochondrial cytochrome oxidase I gene (MT-COI). The sequences were deposited to National Centre for Biotechnology Information (NCBI) to obtain accession numbers.

MATERIALS AND METHODS

Specimen collection: Adult and immature stages of *H. chenopodii* were collected from *Chenopodium quinoa* plants grown in TNAU, Coimbatore (Latitude: 11.008114N, Longitude: 76.932394E) during August 2022. Psyllids were collected by tapping infected plants over a white beating sheet to dislodge different lifestages and aspirating dislodged insects into vials (Fig. 1). The collected psyllids were transferred using a fine brush to a labelled vial containing absolute ethanol. The collected specimens were stored under -20^{o}C until further processes. The collected live specimens were immobilized at 4 ^{o}C for 5 minutes and then, the photographs of live adult and immature insects were taken using Leica M-205A Encoded Stereo Microscope (Leica Microsystems, Wetzlar, Germany).

Morphological trait examination: The specimens were cleared for mounting onto microscope slides by immersion in KOH (10%) at room temperature and then washing with distilled H_2O. The terminal segments of the abdomen were removed from a specimen using insect pins and placed in lactic acid on a microscope slide. The structure was photographed at 4, 10, 40 and 100X using a phase contrast microscope (Euromex iScope, The Netherlands). Measured 10–15 specimens of both sexes. The mounted specimens were identified using descriptions as found at https://bugguide.net/node/ and the standard morphological keys down to species level given by Wheeler and Hoebeke (1997). Confirmed the identification of *H. chenopodii* from the descriptions and illustrations provided for Old-World populations of psyllid (Lauterer, 1982).

Diagnostic traits included wing venation; shape and size of the egg; adult body coloration, leg and antennal colour and morphology of the fifth instar nymph, including features of the marginal sectasetae. Measurements of the adult, egg and fifth-instar nymphs were made using the Leica M-205A Encoded Stereo Microscope equipped with an ocular micrometer. The measurements were taken largely to supplement descriptions of

H. chenopodii available in the literature. Total body length was measured for the fifth-instar nymph by placing specimens in a drop of alcohol on a microscope slide beneath a cover slip. Marginal sectasetae were examined in fifth-instar nymphs by placing a specimen dorsal side up on a microscope slide in KOH (10%). The slide was gently heated for 1 h to dissolve wax filaments. Number of marginal sectasetae on the head, forewing pad, hindwingpad, and abdomen were determined by examination of the digital images.

DNA isolation: Total genomic DNA was extracted from egg, nymph and adult psyllids separately by a rapid standard method (Montero Pau *et al.,* 2008). Briefly, a single specimen in a micro centrifuge tube was ground using 30 ìl of tissue lysis buffer (10N NaOH and 0.5M Na EDTA at pH 8.0) by crushing the sample and homogenized. The lysate was mixed by vortex and the homogenate was incubated at

65°C for 30 min. To inactivate the lysis buffer, an equal volume of neutralizing buffer (10 mM Tris-HCL, pH 5.0) was added and incubated at 95°C for 15 min followed by centrifugation for 5 min at 12000 rpm. Finally, the DNA aliquot was stored until further processes. Quantification of DNA was done in NanodropOne™ (Thermoscientific, Massachusetts, United States) at A260 nm before polymerizing reactions. The MT-COI was amplified using the primers LCO (5' GGTCAACAAATCATAAAGATATTGG 3') and HCO (5' TAAACTTCAGGG TGACCAA AAAATCA 3') in polymerase chain reaction (PCR). PCR conditions of initial denaturation at 94°C for 5 min, followed by 30 cycles of (i) denaturation at 94°C for 1min, (ii) annealing at 50°C for 1min, (iii) elongation at 72°C for 1min and a final extension step at 72°C for 10 min in Eppendorf Mastercycler™ thermocycler (Eppendorf, Hamburg, Germany) (Hoy and Jeyaprakash, 2005). The amplified DNA products were resolved on 1 per cent agarose gel stained with ethidium bromide (10 mg/ml) and visualized in a gel documentation system (Bio-Rad®, Hercules, USA) for quality and then, the obtained PCR product after purificationwas checked for quantity in NanodropOne™ at A260 nm. Then, it was sent to the sequencing facility at Eurofins India Pvt. Ltd., Bangalore, India, and was sequenced in both directions utilizing double pass sanger dideoxy DNA sequencing method with MT-COI forward and reverse primers provided.

Sequence and phylogeny analysis: The raw sequence chromatograms were manually checked then assembled and edited using Chromas Version 2.6.5. (Technelysium Pty Ltd, Brisbane, Australia). The identity of the species was checked by homology search in BLASTn search tool (https://blast.ncbi.nlm.nih.gov/Blast.cgi) for the MT-COI gene. The processed sequences were deposited to NCBI website to obtain accession numbers. The partial sequences showing 99-100 per cent similarity to *H. chenopodii* MT-COI were retrieved from NCBI (MG988841, MW630128, MT021799, MT162454, MT162455 and MG401358) for further analysis. In order to investigate the genetic link, a total of twelve (retrieved 7 no. and generated 5 no.) 435bp MT-COI sequences were aligned using ClustalW after being modified with Bio edit. MEGA X Software (Molecular Evolutionary Genetics Analysis, Version X) was used to carry out the phylogenetic analysis based on the ML statistical approach (Tamura *et al.,* 2013; Kumar *et al.,* 2018). The T92 distance model (Tamura 3-parameter) was used to select the nucleotide mismatch for each region. The Neighbour-Joining (NJ) phylogenetic tree approach was used to create the phylogenetic tree using 1000 bootstrap replications to evaluate the branches' dependability (Saitou and Nei, 1987). Because they are closely related species, brown plant hopper *Nilaparvata lugens* (Accession number: AF222883) sequence from NCBI GenBank was used as an outgroup to illustrate the difference in lineage diversion.

RESULTS AND DISCUSSION

Taxonomic characterization of *H. chenopodii* different life stages:

The eggs are swollen without micropyle, about 0.30 - 0.32 mm long and 0.14 - 0.15 mm broad. The front end of the egg is slightly elevated from the leaf surface and taper to a visible point, with the base of the egg extensively in touch with the leaf surface. Eggs are attached to leaves by a short pedicel (Figs. 2, 7).

Nymph body length varies from 0.7-1.0mm and width varies from 0.4-0.6mm (Fig. 9). When nymphs are in their first instar, they are bright yellow with red ocelli. As they grow, the colour changes to a pale yellowish-green with powdery coating (Figs. 3, 7). Nymphs rarely move and prefer the abaxial leaf surface. The forewing pads were extended anteriorly into humeral lobes. Body covered with setae all around (Fig. 9).

As for adult, both sexes have complete wings. Head and thorax are dark brown to black in mature specimens and yellowish brown in younger specimens (Fig. 4); abdomen yellowish green to darker green (Figs. 5, 6). The length of the adult body can vary from 1.3-1.9mm. Their legs are mostly yellow. Antenna varies in pigmentation, but generally segments II-V pale and the remaining

Fig. 1 Field symptoms of *H. chenopodii* on *C. quinoa* plants, marginal rolls on leaves

Fig. 2 Eggs of *H. chenopodii*

Fig. 3 Nymphal stage of *H. chenopodii*

Fig. 4 Newly emerged adult of *H. chenopodii*

Fig. 5 Matured adult of *H. chenopodii*

Fig. 6 Older adult of *H. chenopodii* with abdominal coloration

Fig. 7 Nymphs emerging from eggs of *H. chenopodii*

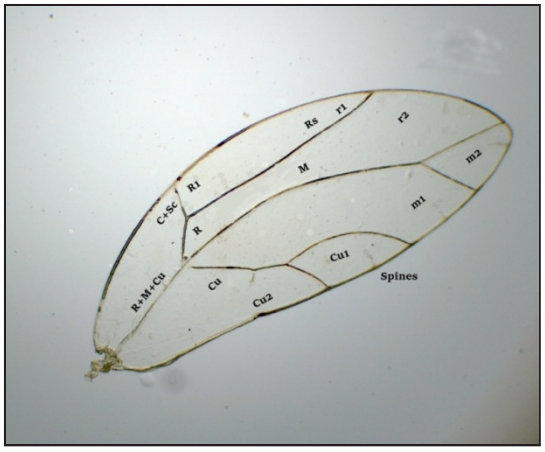

Fig. 8 Wing venation of adult *H. chenopodii*

Fig. 9 Nymphal *H. chenopodii* with sectasetae

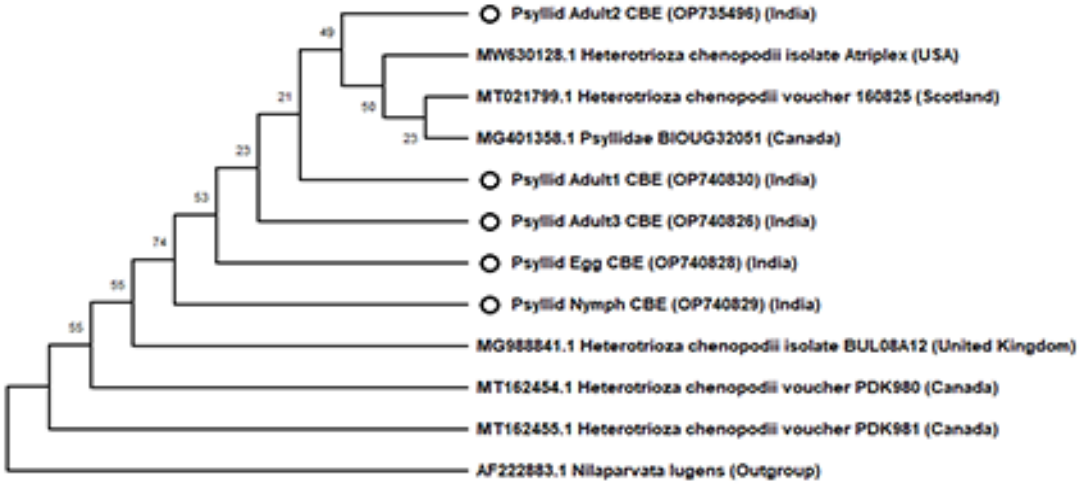

Fig. 10. Dendrogram representation of Neighbour-Joining tree based on 435 bp alignment of 11sequences of the MT-COI gene of *H.chenopodii*. The sequences obtained in this study are indicated in circles

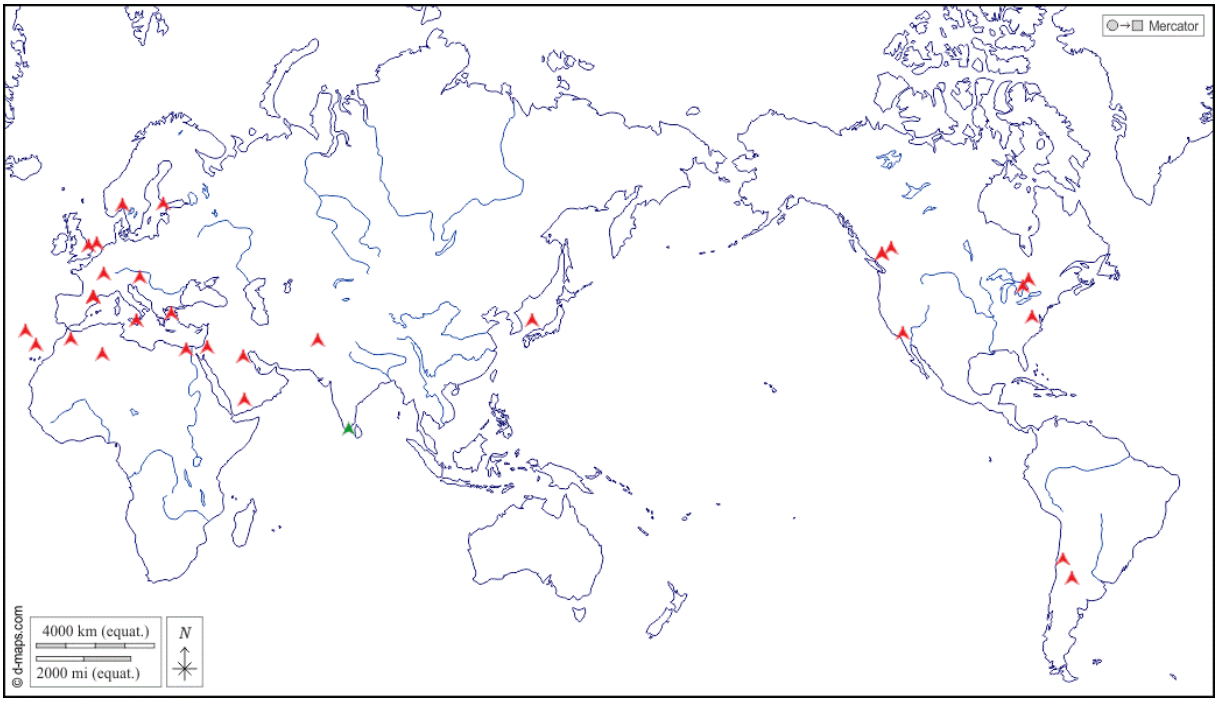

Fig. 11 Graphical display depicting global expansion range of *H. chenopodii*. The red triangles denote the record of *H. chenopodii* from that geographical range. Graphic updated using the reference of Global Biodiversity Information Facility (GBIF)

segments are brown or black.Wings are hyaline with trifurcation of veins (R+M+Cu1) which is typical to Triozidae. The forewing is distinctly broader in the middle, narrowing to an acute apex. Forewing is having a reduced venation and hind wings are filled with minute spinules and with slight brown colour pattern. The vein arising from humeral area (R+M+Cu1), connects the coastal and subcoastal veins with radius. In the forewing, veins Cul and M have a common stem, each arising separately from common origin at vein R (Fig. 8). The apical area of wing contains three apicular spines at median and cubital areas. Forewing with surface spinules largely confined to basal half of wing. Generally abdomen is green in colour for adults. After the last nymphal molt, emerging young adults are initially pale green, but soon the dorsal colorationby maturation become darker with considerable variation in the degree and intensity of colours. In older specimens, dorsum was often found to be uniformly dark brown coloured on the head and thorax, and also on the terminal part of the antennae such colour variations can be seen.

Molecular characterization and Phylogeny:

The identity analysis in NCBI indicated that the MT-COI sequences of *H. chenopodii* were 96 per cent identical to the previously deposited sequences of *H. chenopodii* with NCBI. The generated sequences were further processed and deposited to the NCBI website andaccession numbers assigned were OP735496, OP740826, OP740828, OP740829 and OP740830. The phylogenetic tree shows a single clade of sequences which represented the similarities in the analyzed sequences (Fig. 10). Every sequence had converted into a single clade with 9 subclades excluding the out group. Out of these subclades, eight were with a single sequence and only one subclade was further divided into three. The Indian populations are found to be merged in between other similar populations. *H. chenopodii* is having historical evidences of distribution into sub- continental or continental scales out of their native range (Fig. 11). It was recorded in several countries present in temperate Asia, North Africa, Europe, and the Middle East

(Mifsud, 2020; Percy *et al.,* 2020) and reached the terrain areas of Pakistan too (Wheeler and Hoebeke, 1997, 2013). Now, the insect's geographical range isextended to tropical southern India, on *C. quinoa,* a pseudo-cereal that is highly nutritive and has been cultivated to utilize its foliage. Currently in India, the crop has been gaining attention for human consumption because of its medicinal value as an analgesic, anti-inflammatory and protein supplement (Mujica *et al.,* 2003; Bhargava *et al.,* 2006). Infestation of *H. chenopodii* may affect the productivity ofquinoa and may also other members of the Amaranthaceae, which are seriously infested by this pest in other parts of the globe.

*H. chenopodii*is found to be an effective biological control agent against weeds (Haouas *et al.,* 2021). Hence, a careful assessment of the host plant preference among Amaranthaceae members may be required for further pest risk analysis.The presently reported psyllid has no specific history in this geographical region and sequences of *H. chenopodii* generated are highly similar to the global population. The recent phylogenomic analysis by Percy *et al.* (2018) has grouped the *Heterotrioza* with numerous triozids that are exclusively found in the Austro-Pacific region denoting the eastern range origin of the genus *Heterotrioza*. Hence, it is almost certain that their presence in peninsular India is as the result of recent immigration (Percy *et al.,* 2012; Percy *et al.,* 2020; Burckhardt *et al.,* 2021).

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Grasshoppers, crickets and katydids of Kerala, an updated checklist for the order Orthoptera

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ABSTRACT: An updated checklist of the order Orthoptera of Kerala is provided. Eighty-five species have been added to the existing checklist. A total of 215 species and 21 subspecies belonging to 154 genera under 18 families of two suborders are enumerated along with their distributional data across the state. Suborders Caelifera and Ensifera are represented by 127 and 88 species, respectively.

KEYWORDS: Species, Caelifera, Ensifera, distributional data

INTRODUCTION

Orthoptera is a familiar and significant insect group found in diverse ecosystems (Bhowmik and Rui, 1982), comprising grasshoppers, locusts, crickets, mole crickets and katydids. Besides being conspicuous pests, they are also significant as primary consumers, prey to predators and good indicators of changing environmental conditions (Bazelet and Samways, 2011; Gangwere and Muralirangan, 1997; Belovsky and Slade, 2017). The order consists of more than 29,410 valid species belonging to 43 families under two suborders, Caelifera and Ensifera (Cigliano *et al.* 2023). In India, no major taxonomic compilation is available on the Orthoptera fauna except for a review by Chandra *et al.* (2010) and an annotated checklist of Orthoptera from India by Shishodia *et al.* (2010) in which 1033 species were reported. Recently Gupta and Chandra (2020) compiled a checklist of Orthoptera of Western Ghats. The grasshoppers of India mostly recognised as agricultural pests, are least explored after the colonial researchers (Bhaskar *et al.*, 2019). Notable taxonomic works on Indian Orthoptera were done by Kirby (1914), Hancock (1915), Uvarov (1921, 1929), Hebard (1929), Henry (1940), Chopard (1969), Tandon (1976) and Bhowmik (1977, 1985). Many other significant works on Orthoptera were conducted in various states of India (Tandon and Shishodia, 1977; Vasanth, 1993; Tandon and Hazra, 1998; Shishodia, 1999, 2000; Chitra *et al*., 2000; Shishodia and Kulkarni, 2002; Dey and Hazra, 2003; Thakur *et al*., 2004; Kulkarni and Shishodia, 2005; Senthilkumar *et al*., 2006; Senthilkumar, 2010; Usmani *et al*., 2010; Nayeem and Usmani, 2012; Akhtar and Usmani, 2014; Kumar and Usmani, 2015; Bhaskar *et al.,* 2022). Significant studies on

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Orthopterans fauna of Kerala were done by Shishodia and Hazra (1986), Vasanth (1991), Shishodia and Kulakrni (2002), Priya and Naredran (2003), Prabakar and Radhakrishnan (2005), Bhaskar *et al*. (2018, 2019, 2020 a,b,c), Meena *et al.* (2021), Hiremath and Prathapan (2021) and Jaiswara *et al.* (2021, 2022). Bhaskar *et al*. (2018) compiled the first-ever checklist of Orthoptera of Kerala, in which 130 species were recorded. Later, six new Orthopteran species were recorded from Kerala (Bhaskar *et al*., 2020c; Meena *et al*., 2021; Hiremath and Prathapan, 2021 and Jaiswara *et al.*, 2021, 2022). In addition, some studies on orthopteran diversity were also conducted (Kumar *et al*., 2018; Bhaskar *et al*., 2019; Thasnim and Bijoy, 2021). Gupta and Chandra (2020) compiled a checklist of Orthoptera of Western Ghats in which 147 species were enlisted from Kerala. Based on the literature review and taking into account of Indian Orthoptera-type specimens deposited in the European Natural History Museums an updated checklist of Orthoptera of Kerala is presented.

MATERIALS AND METHODS

This checklist was prepared by reviewing available taxonomic literature on Indian Orthoptera from 1914 to 2023 including Kirby (1914), Hancock (1915), Bhowmik (1977), Shishodia and Hazra (1986), Tandon (1988), Vasanth (1991), Wagan and Kevan (1992), Shishodia and Kulakarni (2002), Priya and Naredran (2003), Mathew (2004), Mathew *et al*. (2004), Prabakar and Radhakrishnan (2004, 2005), Mandal *et al*. (2007), Shishodia *et al.* (2010), Chandra and Gupta (2013), Srinivasan and Prabakar (2013), Ravi *et al*. (2014), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019, 2020 a, b, c), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Jaiswara *et al.* (2021, 2022), Hiremath and Prathapan (2021), Meena *et al.* (2021), and Sreeja *et al*. (2023). To confirm the species identity, the Indian Orthoptera type specimens deposited in Natural History Museum, London, U.K. (BMNH); the Muséum National d'Histoire Naturelle, Paris, France (MNHN); Museo Nacional de Ciencias Naturales, Madrid, Spain (MNCN); Muséum d'histoire naturelle, Geneva, Switzerland (MHNG) were considered. The type specimens were studied from 2016 to 2019 by Dr Dhaneesh Bhaskar with the support of the Orthoptera Species File. Species taxonomic hierarchy was followed by the latest Orthoptera Species File Online (OSF) version (Cigliano *et al.,* 2023).

Abbreviations used: NP - National Park, WS - Wildlife Sanctuary, TR - Tiger Reserve, BR - Biosphere Reserve

RESULTS AND DISCUSSION

A modified Checklist of Orthoptera of Kerala is prepared in which 215 species and 21 subspecies belonging to 154 genera under 18 families of two suborders are enlisted. Suborder Caelifera is represented by 127 species under 88 genera and 6 families. Suborder Ensifera is represented by 88 species under 66 genera and 12 families. In comparison to the existing checklist of Orthoptera of Kerala (Bhaskar *et al.,* 2018), the number of species has risen by 85. The reported species from Kerala are mentioned along with where they are found with source (Table 1).

An annotated checklist for the Orthoptera fauna of Kerala, India is provided covering historical documents and the Orthoptera species list in all valid scientific publications from the colonial period to date. Altogether 215 species belonging to 154 genera under 18 families of two suborders are enlisted as the Orthoptera diversity in Kerala, India. Suborder Caelifera is represented by 127 species and Ensifera by 88 species. Compared to the existing checklist (Bhaskar *et al.*, 2018), the species number rose from 130 to 215, an increase of 85 species. Protected areas in Kerala were least explored for its orthopteran fauna. Only few works; Shishodia and Hazra (1986), Shishodia and Kulakarni (2002), Mathew (2004) and Bhaskar *et al.* (2019) are available. By reviewing these literatures 30 species from Silent Valley NP, 54 species from Parambikulam TR and 22 species from Eravikulam NP are enlisted in this checklist. The number of species in such a small geographic area indicates the richness of Orthoptera fauna in the Indian subcontinent, especially in the Western Ghats region

Table 1. Updated Checklist of Orthopteran in Kerala

No.	Species	Distribution	Source
Suborder: CAELIFERA; Infraorder: ACRIDIDEA; Superfamily: ACRIDOIDEA Family: ACRIDIDAE MacLeay, 1821: Subfamily ACRIDINAE MacLeay, 1821			
Genus *Bababuddinia* Bolívar, 1917			
1.	*Bababuddinia bizonata* Bolívar, 1917	Palakkad (Silent Valley NP)	Shishodia and Hazra (1986), Mathew (2004), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Carliola* Uvarov, 1939			
2.	*Carliola carinata* (Uvarov, 1929)	Palakkad (Parambikulam TR) Idukki (Eravikulam NP), Ernakulam, Alappuzha	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
Genus *Paraduronia* Bolívar, 1909			
3.	*Paraduronia carinata* (Bolívar, 1902)	Nilgiri BR	Mathew (2004), Cigliano *et al.* (2023)
Tribe Acridini MacLeay, 1821			
Genus *Acrida* Linnaeus, 1758			
4.	*Acrida exaltata* (Walker, 1859)	Palakkad (Parambikulam TR), Thiruvananthapuram, Kollam, Alappuzha, Malappuram, Wayanad, Kozhikode, Kannur, Kasaragod, Ernakulam, Kottayam, Pathanamthitta, Idukki, Thrissur	Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al*. (2023)
5.	*A. gigantea* (Herbst, 1786)	Palakkad (Parambikulam TR), Thrissur, Thiruvananthapuram, Kollam, Malappuram, Kozhikode, Kasaragod, Ernakulam, Kottayam, Wayanad	Mathew (2004), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Phlaeobini Brunner von Wattenwyl, 1893			
Genus *Phlaeoba* Stål, 1861			
6. a	*Phlaeoba angustidorsis angustidorsis* Bolívar, 1902	Kerala.	Prabakar and Radhakrishnan (2005), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023),
6. b	*P. antennata antennata* Brunner von Wattenwyl, 1893	Palakkad (Parambikulam TR, Silent Valley NP), Wayanad, Ernakulam, Alappuzha	Shishodia and Hazra (1986), Shishodia *et al.* (2010), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
7.	*P. infumata* Brunner von Wattenwyl, 1893	Palakkad (Parambikulam TR), Malappuram, Ernakulam, Idukki, Nilgiri BR, Alappuzha	Mathew (2004), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al*. (2023)
8.	*P. panteli* Bolívar, 1902	Palakkad (Parambikulam TR), Ernakulam, Alappuzha	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)

Genus *Zygophlaeoba* Bolívar, 1902			
9.	*Zygophlaeoba sinuatocollis* Bolívar, 1902	Kerala	Priya and Narendran (2003), Bhaskar *et al.* (2018), Cigliano *et al*. (2023)
10.	*Z. collina* Uvarov, 1929	Nilgiri BR	Mathew (2004), Cigliano *et al.* (2023)
Subfamily CALLIPTAMINAE Jacobson, 1905			
Genus *Acorypha* Krauss, 1877			
11.	*Acorypha glaucopsis* (Walker, 1870)	Ernakulam, Thrissur	Shishodia *et al.* (2010), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily CATANTOPINAE Brunner von Wattenwyl, 1893			
Genus *Bambusacris* Henry, 1933			
12.	*Bambusacris travancora* Henry, 1940	Palakkad (Parambikulam TR), Idukki (Eravikulam NP)	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
Genus *Mopla* Henry, 1940			
13.	*Mopla guttata* Henry, 1940	Palakkad (Parambikulam TR)	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019, 2020a), Gupta and Chandra (2020), Cigliano *et al*. (2023)
14.	*M. rubra* Henry, 1940	Kerala	Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Naraikadua* Henry, 1940			
15.	*Naraikadua charmichaelae* Henry, 1940	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Palniacris* Henry, 1940			
16.	*Palniacris maculatus* Henry, 1940	Palakkad (Parambikulam TR, Silent Valley NP), Idukki (Eravikulam NP)	Shishodia and Hazra (1986), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Catantopini Brunner von Wattenwyl, 1893			
Genus *Diabolocatantops* Jago, 1984			
17.	*Diabolocatantops innotabilis* (Walker, 1870)	Palakkad (Parambikulam TR), Thrissur, Malappuram, Ernakulam, Alappuzha	Priya and Narendran (2003), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
18.	*D. pinguis* (Stål, 1861)	Alappuzha, Malappuram	Shishodia *et al.* (2010), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Stenocatantops* Dirsh, 1953			
19.	*Stenocatantops splendens* (Thunberg, 1815)	Thrissur, Wayanad, Kannur, Kottayam, Palakkad	Priya and Narendran (2003), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)

Genus *Xenocatantops* Dirsh, 1953			
20.	*Xenocatantops henryi* (Bolívar, 1917)	Kollam (Shendurney WS), Palakkad (Silent Valley NP), Thrissur	Shishodia and Hazra (1986), Mathew (2004), Mathew *et al*. (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
21.	*X. humilis* (Serville, 1838)	Palakkad (Parambikulam TR, Silent Valley NP), Idukki (Eravikulam NP), Wayanad, Kannur, Alappuzha, Ernakulam, Nilgiri BR	Shishodia and Hazra (1986), Mathew (2004), Mathew *et al*. (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
22.	*X. karnyi* (Kirby, 1910)	Ernakulam, Kottayam, Thrissur	Kumar *et al*. (2018), Cigliano *et al.* (2023)
Tribe Oxyrrhepini Tinkham, 1940			
Genus *Oxyrrhepes* Stål, 1873			
23.	*Oxyrrhepes meyeri* Willemse, 1936	Palakkad (Silent Valley NP)	Priya and Narendran (2003), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
24.	*O. obtusa* (Haan, 1842)	Palakkad (Silent Valley NP), Idukki	Shishodia and Hazra (1986), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Paraconophymatini Otte, 1995			
Genus *Paraconophyma* Uvarov, 1921			
25.	*Paraconophyma scabra* (Walker, 1870)	Kerala	Priya and Narendran (2003), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Subfamily COPTACRINAE Brunner von Wattenwyl, 1893			
Genus *Coptacra* Stål, 1873			
26.	*Coptacra ensifera* Bolívar, 1902	Palakkad (Silent Valley NP), Kozhikode, Malappuram	Shishodia and Hazra (1986), Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
27.	*C. punctaria* (Walker, 1870)	Palakkad (Silent Valley NP), Kozhikode	Shishodia and Hazra (1986), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Epistaurus* Bolívar, 1889			
28.	*Epistaurus sinetyi* Bolívar, 1902	Malappuram, Ernakulam, Thrissur	Priya and Narendran (2003), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Cigliano *et al.* (2023)
Genus *Eucoptacra* Bolívar, 1902			
29.	*Eucoptacra binghami* Uvarov, 1921	Palakkad (Silent Valley NP)	Shishodia and Hazra (1986), Mathew (2004), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
30.	*E. praemorsa* (Stål, 1861)	Malappuram, Kozhikode, Wayanad	Shishodia *et al.* (2010), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Subfamily CYRTACANTHACRIDINAE Kirby, 1910			
Genus *Pachyacris* Uvarov, 1923			
31.	*Pachyacris vinosa* (Walker, 1870)	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
32.	*P. violascens* (Walker, 1870)	Malappuram, Kannur	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
Tribe Cyrtacanthacridini Kirby, 1910			
Genus *Anacridium* Uvarov, 1923			
33.	*Anacridium flavescens* (Fabricius, 1793)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Chondracris* Uvarov, 1923			
34.	*Chondracris rosea* (De Geer, 1773)	Palakkad (Parambikulam TR, Silent Valley NP), Thrissur	Shishodia *et al.* (2010), Bhaskar *et al.* (2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Cyrtacanthacris* Walker, 1870			
35.	*Cyrtacanthacris tatarica tatarica* (Linnaeus, 1758)	Palakkad (Parambikulam TR, Silent Valley NP), Eravikulam NP (Idukki), Malappuram, Ernakulam, Thrissur	Shishodia and Hazra (1986), Shishodia and Kulkarni (2002), Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Patanga* Uvarov, 1923			
36.	*Patanga succincta* (Johannson, 1763)	Palakkad (Parambikulam TR), Nilgiri BR	Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Schistocerca* Stål, 1873			
37.	*Schistocerca gregaria* (Forskål, 1775)	Malappuram	Bhaskar *et al.* (2020b)
Subfamily Eyprepocnemidinae Brunner von Wattenwyl, 1893			
Genus *Cataloipus* Bolívar, 1890			
38.	*Choroedocus illustris* (Walker, 1870)	Palakkad (Parambikulam TR), Kannur, Thrissur, Wayanad	Prabakar and Radhakrishnan (2005), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
39.	*C. robustus* (Serville, 1838)	Thrissur	Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
40.	*Cataloipus indicus* Uvarov, 1942	Kerala	Nayeem and Usmani (2012), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Genus *Tylotropidius* Stål, 1873			
41.	*Tylotropidius varicornis* (Walker, 1870)	Palakkad (Parambikulam TR, Silent Valley NP), Idukki (Eravikulam NP), Ernakulam, Wayanad, Nilgiri BR	Shishodia and Hazra (1986), Shishodia and Kulkarni (2002), Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Nayeem and Usmani (2012), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Eyprepocnemidini Brunner von Wattenwyl, 1893			
Genus *Eyprepocnemis* Fieber, 1853			
42.	*Eyprepocnemis alacris alacris* (Serville, 1838)	Palakkad (Silent Valley NP), Malappuram, Ernakulam, Thrissur, Alappuzha	Shishodia and Hazra (1986), Mathew (2004), Shishodia *et al.* (2010), Nayeem and Usmani (2012), Srinivasan and Prabakar (2013), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
43.	*E. rosea* Uvarov, 1942	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Subfamily GOMPHOCERINAE Fieber, 1853			
Genus *Madurea* Bolívar, 1902			
44.	*Madurea cephalotes* Bolívar, 1902	Nilgiri BR	Mathew (2004), Cigliano *et al.* (2023)
Tribe Arcypterini Bolívar, 1914			
Genus *Aulacobothrus* Bolívar, 1902			
45. a	*Aulacobothrus luteipes luteipes*(Walker, 1871)	Malappuram	Priya and Narendran (2003), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
45. b	*A. luteipes infernus* Bolívar, 1902	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
46.	*A. taeniatus* Bolívar, 1902	Palakkad (Parambikulam TR), Malappuram	Priya and Narendran (2003), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
47.	*A. socius* Bolívar, 1902	Palakkad (Parambikulam TR), Wayanad	Priya and Narendran (2003), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
48.	*A. svenhedini* Sjöstedt, 1933	Wayanad	Priya and Narendran (2003), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Leionotacris* Jago, 1996			
49.	*Leionotacris bolivari* (Uvarov, 1921)	Kerala	Priya and Narendran (2003), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Dociostaurini Mistshenko, 1974			
Genus *Leva* Bolívar, 1909			
50.	*Leva indica* (Bolívar, 1902)	Nilgiri BR	Mathew (2004), Cigliano *et al.* (2023)
Subfamily HEMIACRIDINAE Dirsh, 1956			
Genus *Calamippa* Henry, 1940			
51.	*Calamippa prasina* (Bolívar, 1902)	Palakkad (Silent Valley NP)	Shishodia and Hazra (1986), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Siruvania* Henry, 1940			
52.	*Siruvania dimorpha* Henry, 1940	Palakkad (ParambikulamTR), dukki (Eravikulam NP), Malappuram, Kozhikode, Kannur	Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)

Tribe Hieroglyphini Bolívar, 1912			
Genus *Hieroglyphus* Krauss, 1877			
53.	*Hieroglyphus banian* (Fabricius, 1798)	Palakkad (Parambikulam TR), Kollam, Alappuzha, Wayanad, Kozhikode, Kannur, Kasaragod, Ernakulam, Thrissur, Malappuram	Priya and Narendran (2003), Mathew (2004), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
54.	*H. nigrorepletus* Bolívar, 1912	Thiruvananthapuram, Kollam, Malappuram, Kozhikode, Kasaragod, Ernakulam, Kottayam, Thrissur, Idukki	Priya and Narendran (2003), Mathew (2004), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Parahieroglyphus* Carl, 1916			
55.	*Parahieroglyphus colemani* (Bolívar, 1912)	Kollam, Kozhikode	Kumar *et al*. (2018).
Tribe Leptacrini Johnston, 1956			
Genus *Leptacris* Walker, 1870			
56.	*Leptacris filiformis* Walker, 1870	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
Subfamily OEDIPODINAE Walker, 1871			
Genus *Chloebora* Saussure, 1884			
57.	*Chloebora grossa* Saussure, 1884	Malappuram	Kumar *et al*. (2018), Cigliano *et al.* (2023)
Genus *Dittopternis* Saussure, 1884			
58.	*Dittopternis venusta* (Walker, 1870	Palakkad (ParambikulamTR), Idukki (Eravikulam NP), Malappuram, Thrissur, Alappuzha, Ernakulam	Kumar *et al*. (2018), Bhaskar *et al.* (2019), Cigliano *et al.* (2023)
Genus *Morphacris* Walker, 1870			
59.	*Morphacris fasciata* (Thunberg, 1815)	Palakkad	Kumar *et al*. (2018), Cigliano *et al.* (2023)
Tribe Acrotylini Johnston, 1956			
Genus *Acrotylus* Fieber, 1853			
60.	*Acrotylus insubricus* (Scopoli, 1786)	Thiruvananthapuram,Kollam, Malappuram, Kozhikode, Kasaragod, Ernakulam, Pathanamthitta, Idukki	Kumar *et al*. (2018), Cigliano *et al.* (2023)
Tribe Epacromiini Brunner von Wattenwyl, 1893			
Genus *Aiolopus* Fieber, 1853			
61.	*Aiolopus simulatrix* (Walker, 1870)	Ernakulam, Pathanamthitta, Thrissur, Palakkad	Kumar *et al*. (2018), Cigliano *et al.* (2023)
62.	*A. thalassinus tamulus* (Fabricius, 1798)	Palakkad, Thiruvananthapuram, Kollam, Wayanad, Kannur, Kasaragod, Ernakulam, Thrissur, Nilgiri BR	Mathew (2004), Shishodia *et al.* (2010), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Locustini Kirby, 1825			
Genus *Gastrimargus* Saussure, 1884			
63.	*Gastrimargus africanus africanus* (Saussure, 1888)	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
64.	*G. marmoratus* (Thunberg, 1815)	Thrissur	Priya and Narendran (2003), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Genus *Locusta* Linnaeus, 1758			
65.	*Locusta migratoria migratoria* (Linnaeus, 1758)	Kerala	Priya and Narendran (2003), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Genus *Oedaleus* Fieber, 1853			
66.	*Oedaleus abruptus* (Thunberg, 1815)	Palakkad, Thrissur, Pathanamthitta, Idukki, Malappuram, Nilgiri BR	Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
Genus *Pternoscirta* Saussure, 1884			
67.	*Pternoscirta cinctifemur* (Walker, 1859)	Palakkad (Silent Valley NP), Malappuram, Kozhikode, Ernakulam, Kottayam, Pathanamthitta, Idukki, Thrissur, Nilgiri BR	Shishodia and Hazra (1986), Mathew (2004), Shishodia *et al.* (2010), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Parapleurini Brunner von Wattenwyl, 1893			
Genus *Ceracris* Walker, 1870			
68.	*Ceracris nigricornis nigricornis* Walker, 1870	Thiruvananthapuram, Kollam, Kannur, Kasaragod, Ernakulam, Kottayam, Idukki, Thrissur	Kumar *et al*. (2018), Cigliano *et al.* (2023)
69.	*C. striata* Uvarov, 1925	Ernakulam, Alappuzha	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Tribe Sphingonotini Johnston, 1956			
Genus *Sphingonotus* Fieber, 1852			
70.	*Sphingonotus* (*Sphingonotus*) *longipennis* Saussure, 1884	Kerala	Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Trilophidiini Shumakov, 1963			
Genus *Trilophidia* Stål, 1873			
71.	*Trilophidia annulata* (Thunberg, 1815)	Palakkad (Silent Valley NP, Parambikulam TR), Thrissur, Thiruvananthapuram, Kollam, Alappuzha, Wayanad, Kannur, Kasaragod, Ernakulam, Kottayam, Pathanamthitta, Idukki, Malappuram, Nilgiri BR	Shishodia and Hazra (1986), Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)

Subfamily OXYINAE Brunner von Wattenwyl, 1893			
Genus *Chitaura* Bolívar, 1918			
72.	*Chitaura indica* Uvarov, 1929	Idukki (Eravikulam NP), Palakkad (Silent Valley NP), Kozhikode, Wayanad, Kannur, Kottayam, Ernakulam, Alappuzha, Nilgiri BR	Tandon (1988), Shishodia and Hazra (1986), Shishodia and Kulkarni (2002), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Kumar *et al*. (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Hygracris* Uvarov, 1921			
73.	*Hygracris malabaricus* Willemse, 1962	Palakkad (Parambikulam TR)	Tandon (1988), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Oxyini Brunner von Wattenwyl, 1893			
Genus *Gesonula* Uvarov, 1940			
74.	*Gesonula punctifrons* (Stål, 1861)	Palakkad (Parambikulam TR), Ernakulam, Kottayam, Thrissur	Tandon (1988), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Oxya* Serville, 1831			
75.	*Oxya chinensis* (Thunberg, 1815)	Alappuzha, Ernakulam	Shishodia *et al.* (2010), Kumar *et al*. (2018), Cigliano *et al.* (2023)
76.	*O. fuscovittata* (Marschall, 1836)	Palakkad (Parambikulam TR), Thrissur, Thiruvananthapuram, Kollam, Alappuzha, Wayanad, Kannur, Kasaragod	Tandon (1988), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
77.	*O. grandis* Willemse, 1925	Alappuzha, Wayanad, Kannur	Tandon (1988), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
78.	*O. hyla* Serville, 1831	Palakkad (Silent Valley NP, Parambikulam TR), Idukki (Eravikulam NP), Nilgiri BR, Thrissur, Ernakulam, Kottayam, Pathanamthitta, Alappuzha	Tandon (1988), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
79.	*O. japonica japonica* (Thunberg, 1824)	Palakkad (Parambikulam TR), Idukki, Kottayam, Thiruvananthapuram, Kollam, Alappuzha, Wayanad, Kannur, Kasaragod, Thrissur	Tandon (1988), Priya and Narendran (2003), Shishodia *et al.* (2010), Nayeem and Usmani (2012), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
80.	*O. nitidula* (Walker, 1870)	Palakkad (Silent Valley NP), Thrissur	Shishodia and Hazra (1986), Priya and Narendran (2003), Mandal *et al*. (2007), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
81.	*O. velox* (Fabricius, 1787)	Alappuzha, Wayanad, Kannur, Ernakulam, Thrissur	Mathew (2004), Kumar *et al*. (2018), Cigliano *et al.* (2023)

Subfamily SPATHOSTERNINAE Rehn, 1957;Tribe Spathosternini Rehn, 1957			
Genus *Spathosternum* Krauss, 1877			
82.	*Spathosternum prasiniferum prasiniferum* (Walker, 1871)	Palakkad (Parambikulam TR), Thrissur, Thiruvananthapuram, Kollam, Alappuzha, Malappuram, Wayanad, Kozhikode, Kannur, Kasaragod, Ernakulam, Kottayam, Pathanamthitta, Nilgiri BR	Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Nayeem and Usmani (2012), Srinivasan and Prabakar (2013), Kumar *et al*. (2018), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
Subfamily TERATODINAE Brunner von Wattenwyl, 1893			
Genus *Teratodes* Brullé, 1835			
83.	*Teratodes monticollis* (Gray, 1832)	Palakkad (Parambikulam TR), Idukki (Eravikulam NP), Thrissur	Mathew (2004), Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
Subfamily TROPIDOPOLINAE Jacobson, 1905;Tribe Tristriini Mistshenko, 1945			
Genus *Tristria* Stål, 1873			
84.	*Tristria pulvinata* (Uvarov, 1921)	Palakkad (Silent Valley NP)	Shishodia and Hazra (1986), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Superfamily EUMASTACOIDEA Burr, 1899; Family CHOROTYPIDAE Stål, 1873 Subfamily CHOROTYPINAE Stål, 1873; Tribe Chorotypini Stål, 1873			
Genus *Burrinia* Bolívar, 1930			
85.	*Burrinia burri* (Bolívar, 1914)	Palakkad (Parambikulam TR), Ernakulam, Alappuzha	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Phyllochoreia* Westwood, 1839			
86.	*Phyllochoreia ramakrishnai* Bolívar, 1914	Palakkad (Parambikulam TR)	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
87.	*P. unicolor* Westwood, 1839	Palakkad (Parambikulam TR), Malabar	Kirby (1914), Mathew (2004), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
88.	*P. westwoodi* Bolívar, 1930	Kerala	Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily PRIONACANTHINAE Descamps, 1973; Tribe Prionacanthini Descamps, 1973			
Genus *Prionacantha* Henry, 1940			
89.	*Prionacantha picta* Henry, 1940	Idukki (Eravikulam NP)	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Family MASTACIDEIDAE Rehn, 1948; Subfamily MASTACIDEINAE Rehn, 1948			
Genus *Mastacides* Bolívar, 1899			
90.	*Mastacides nilgirisicus* Bolívar, 1914	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
Genus *Paramastacides* Descamps, 1974			
91.	*Paramastacides ramachendrai* (Bolívar, 1930)	Palakkad (Parambikulam TR), Idukki (Eravikulam NP)	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)

	Superfamily PYRGOMORPHOIDEA Brunner von Wattenwyl, 1874; Family PYRGOMORPHIDAE Brunner von Wattenwyl, 1874; Subfamily ORTHACRIDINAE Bolívar, 1905; Tribe Orthacridini Bolívar, 1905		
Genus *Neorthacris* Kevan & Singh, 1964			
92.	*Neorthacris acuticeps acuticeps* (Bolívar, 1902)	Palakkad (Parambikulam TR), Thrissur, Malappuram, Ernakulam, Alappuzha	Priya and Narendran (2003), Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
93.	*N. acuticeps nilgirensis* (Uvarov, 1929)	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
94.	*N. malabarensis* Singh & Kevan, 1965	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
	Subfamily PYRGOMORPHINAE Brunner von Wattenwyl, 1874 Tribe Atractomorphini Bolívar, 1905		
Genus *Atractomorpha* Saussure, 1862			
95.	*Atractomorpha crenulata* (Fabricius, 1793)	Palakkad (Silent Valley NP, Parambikulam TR), Idukki (Eravikulam NP), Kottayam, Thrissur, Malappuram, Ernakulam, Alappuzha	Shishodia and Hazra (1986), Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Srinivasan and Prabakar (2013), Ravi *et al*. (2014), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Thasnim and Bijoy (2021), Cigliano *et al.* (2023)
	Tribe Chrotogonini Bolívar, 1884		
Genus *Chrotogonu*s Serville, 1838			
96.	*Chrotogonus (Chrotogonus) oxypterus* (Blanchard, 1836)	Palakkad (Parambikulam TR), Malabar, Malappuram	Kirby (1914), Shishodia and Hazra (1986), Priya and Narendran (2003), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
97.	*C. (C.) trachypterus trachypterus* (Blanchard, 1836)	Kozhikode	Priya and Narendran (2003), Bhaskar *et al.* (2018, 2019), Cigliano *et al.* (2023)
	Tribe Poekilocerini Burmeister, 1840		
Genus *Poekilocerus* Serville, 1831			
98.	*Poekilocerus pictus* (Fabricius, 1775)	Palakkad (Parambikulam TR), Idukki (Eravikulam NP)	Priya and Narendran (2003), Mathew (2004), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
	Tribe Pseudomorphacridini Kevan & Akbar, 1964		
Genus *Pseudomorphacris* Carl, 1916			
99.	*Pseudomorphacris notata* (Brunner von Wattenwyl, 1893)	Idukki (Eravikulam NP), Kozhikode	Prabakar and Radhakrisnan (2005), Bhaskar *et al.* (2018), Gupta and Chandra (2020)
	Tribe Pyrgomorphini Brunner von Wattenwyl, 1874		
Genus *Zarytes* Bolívar, 1904			
100.	*Zarytes squalinus squalinus* (Saussure, 1884)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Taphronotini Bolívar, 1904			
Genus *Aularches* Stål, 1873			
101.	*Aularches miliaris miliaris* (Linnaeus, 1758)	Palakkad (Silent Valley NP, Parambikulam TR), Wayanad, Thrissur	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2019), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Superfamily TETRIGOIDEA Rambur, 1838; Family TETRIGIDAE Rambur, 1838			
Genus *Bolotettix* Hancock, 1907 [The genus formerly placed under subfamily SCELIMENINAE, but now remains separately under the family Tetrigidae, without subfamily placement (Cigliano *et al.,* 2023)]			
102.	*Bolotettix* anomalus Hancock, 1907	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Tribe Criotettigini Kevan, 1966 [Tribe Criotettigini was formerly placed under the subfamily SCELIMENINAE and now remains separately under the family TETRIGIDAE without subfamily placement (Cigliano *et al.,* 2023)]			
Genus *Criotettix* Bolívar, 1887			
103.	*Criotettix fastiditus* Bolívar, 1917	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Tribe Thoradontini Kevan, 1966 [Tribe Thoradontini was formerly placed under the subfamily SCELIMENINAE and now remains separately under the family Tetrigidae without subfamily placement (Cigliano *et al.*, 2023)]			
Genus *Eucriotettix* Hebard, 1930			
104.	*Eucriotettix flavopictus* (Bolívar, 1902)	Palakkad (Silent Valley NP, Parambikulam TR), Idukki (Eravikulam NP)	Shishodia and Hazra (1986), Shishodia *et al.* (2010), Bhaskar *et al.* (2018, 2019), Gupta and Chandra (2020), Cigliano *et al.* (2023).
105.	*E spinilobus* (Hancock, 1904)	Thiruvananthapuram	Kirby (1914), Hancock (1915), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
106.	*E. tricarinatus* (Bolívar, 1887)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily CLADONOTINAE Bolívar, 1887; Tribe Cladonotini Bolívar, 1887			
Genus *Deltonotus* Hancock, 1904			
107.	*Deltonotus gibbiceps* (Bolívar, 1902)	Palakkad (Silent Valley NP, Parambikulam TR), Idukki (Eravikulam NP)	Shishodia and Hazra (1986), Shishodia and Kulkarni (2002), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019, 2020c, 2022), Gupta and Chandra (2020), Cigliano *et al.* (2023)
108	*D. subcucullatus* (Walker, 1871)	Parambikulam TR	Bhaskar *et al.* (2020c; Bhaskar and Skeja, 2022)
Genus *Hancockella* Uvarov, 1940			
109.	*Hancockella portentosa* (Kirby, 1914)	Kollam (Thenmala), Thiruvananthapuram	Shishodia and Hazra (1986), Shishodia and Kulkarni (2002), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018, 2019, 2020c), Bhaskar and Skeja, 2022), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Epitettigini Storozhenko, 2023			
Genus *Epitettix* Hancock, 1907			
110.	*Epitettix tumilus* Günther, 1939	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020)
Tribe Xerophyllini Günther, 1979			
Genus *Tettilobus* Hancock, 1909			
111.	*Tettilobus prashadi* Günther, 1938	Kerala	Bhaskar *et al.* (2018, 2020c), Bhaskar and Skeja (2022), Cigliano *et al.* (2023)
112.	*Tettilobus trishula* Skejo, Bhaskar & Stermšek, 2020	Idukki (Eravikulam NP)	Bhaskar *et al.* (2020c), Bhaskar and Skeja (2022), Cigliano *et al.* (2023)
Subfamily METRODORINAE Bolívar, 1887; Tribe Cleostratini Bolívar, 1887			
Genus *Indomiriatra* Tinkham, 1939			
113.	*Indomiriatra provertex* (Hancock, 1912)	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2019), Cigliano *et al.* (2023)
Genus *Miriatroides* Zheng & Jiang, 2002			
114.	*Miriatroides gravelyi* (Günther, 1939)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Subfamily SCELIMENINAE Bolívar, 1887; Tribe Scelimenini Bolívar, 1887			
Genus *Euscelimena* Günther, 1938			
115.	*Euscelimena gavialis* (Saussure, 1862)	Palakkad (Parambikulam TR), Thiruvananthapuram, Kollam (Thenmala)	Kirby (1914), Hancock (1915), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018, 2019), Bhakar and Skeja (2022), Cigliano *et al.* (2023)
116.	*E. harpago* (Serville, 1838)	Palakkad (Parambikulam TR), Idukki (Eravikulam NP)	Bhaskar *et al.* (2018, 2019), Bhaskar and Skeja (2022), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Scelimena* Serville, 1838			
117.	*Scelimena producta* (Serville, 1838)	Thiruvananthapuram	Kirby (1914), Mathew (2004), Gupta and Chandra (2020)
Subfamily TETRIGINAE Rambur, 1838			
Genus *Ergatettix* Kirby, 1914			
118.	*Ergatettix dorsiferus* (Walker, 1871)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Coptotettix* Bolívar, 1887			
119.	*Coptotettix hancocki* (Kirby, 1910)	Kerala	Bhaskar *et al.* (2018)
Genus *Hedotettix* Bolívar, 1887			
120.	*Hedotettix gracilis* (Haan, 1843)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Tetrigini Rambur, 1838			
Genus *Euparatettix* Hancock, 1904			
121.	*Euparatettix personatus* (Bolívar, 1887)	Palakkad (Parambikulam TR)	Wagan and Kevan (1992), Bhaskar *et al.* (2019), Cigliano *et al.* (2023)

Genus *Paratettix* Bolívar, 1887			
122.	*Paratettix cingalensis* (Walker, 1871)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily TRIPETALOCERINAE Bolívar, 1887; Tribe Tripetalocerini Bolívar, 1887			
Genus *Tripetalocera* Westwood, 1834			
123.	*Tripetalocera ferruginea* Westwood, 1834	Thiruvananthapuram	Kirby (1914), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Infraorder TRIDACTYLIDEA; Superfamily TRIDACTYLOIDEA Brullé, 1835 Family TRIDACTYLIDAE Brullé, 1835; Subfamily DENTRIDACTYLINAE Günther, 1979			
Genus *Bruntridactylus* Günther, 1979			
124.	*Bruntridactylus saussurei* (Chopard, 1933)	Palakkad (Parambikulam TR)	Bhaskar *et al.* (2019), Cigliano *et al.* (2023)
Subfamily Tridactylinae Brullé, 1835			
Genus *Xya* Latreille, 1809			
125.	*Xya castetsi* (Bolívar, 1900)	Palakkad (ParambikulamTR)	Bhaskar *et al.* (2019), Cigliano *et al.* (2023)
126.	*X. riparia* (Saussure, 1877)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020)
127.	*X. variegata* (Latreille, 1809)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Suborder ENSIFERA; Infraorder GRYLLIDEA Superfamily GRYLLOIDEA Laicharting, 1781; Subfamily PTEROPLISTINAE Chopard, 1936 [Subfamily PTEROPLISTINAE was formerly placed under the family GRYLLIDAE but now remains separately under superfamily GRYLLOIDEA, without family placement (Cigliano *et al.,* 2023)]			
Genus *Pteroplistes* Brunner von Wattenwyl, 1873			
128.	*Pteroplistes* (*Pteroplistes*) *platycleis* Bolívar, 1900	Thiruvananthapuram, Ernakulam	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Family GRYLLIDAE Laicharting, 1781 Subfamily GRYLLINAE Laicharting, 1781			
Genus *Callogryllus* Sjöstedt, 1910			
129.	*Callogryllus bilineatus* (Bolívar, 1900)	Thiruvananthapuram	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
130.	*C. orientalis* (Bolívar, 1900)	Thiruvananthapuram	Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Itaropsis* Chopard, 1925			
131.	*Itaropsis tenella* (Walker, 1869)	Kerala	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Meristoblemmus* Jones & Chopard, 1936			
132.	*Meristoblemmus lobifrons* Jones & Chopard, 1936	Thiruvananthapuram	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Gryllini Laicharting, 1781			
Genus *Acheta* Fabricius, 1775			
133.	*Acheta domesticus* (Linnaeus, 1758)	Ernakulam, Alappuzha	Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Cophogryllus* Saussure, 1877			
134.	*Cophogryllus maindroni* Chopard, 1928	Thiruvananthapuram	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Genus *Gryllodes* Saussure, 1874			
135.	*Gryllodes sigillatus* (Walker, 1869)	Ernakulam, Alappuzha	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Gryllus* Linnaeus, 1758			
136.	*Gryllus* (*Gryllus*) *bimaculatus* De Geer, 1773	Ernakulam	Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Gymnogryllus* Saussure, 1877			
137.	*Gymnogryllus kashmirensis* Bhowmik, 1967	Kerala	Shishodia *et al.* (2010), Cigliano *et al.* (2023)
Genus *Phonarellus* Gorochov, 1983			
138.	*Phonarellus* (*Phonarellus*) *humeralis* (Walker, 1871)	Kozhikode	Chopard (1969), Bhowmik (1977), Vasanth (1991), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
139.	*P.* (*P.*) *minor* (Chopard, 1959)	Kerala	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Plebeiogryllus* Randell, 1964			
140.	*Plebeiogryllus guttiventris guttiventris* (Walker, 1871)	Palakkad (Silent Valley NP), Malappuram, Thiruvananthapuram, Ernakulam (Kochi)	Chopard (1969), Shishodia and Hazra (1986), Vasanth (1991), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Scapsipedoides* Chopard, 1936			
141.	*Scapsipedoides macrocephalus* Chopard, 1936	Kerala	Chopard (1969), Mathew (2004)
Genus *Sphecogryllus* Chopard, 1933			
142.	*Sphecogryllus armatus* Chopard, 1933	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Genus *Tarbinskiellus* Gorochov, 1983			
143.	*Tarbinskiellus portentosus* (Lichtenstein, 1796)	Thrissur	Mathew (2004), Cigliano *et al.* (2023)

Genus *Teleogryllus* Chopard, 1961			
144.	*Teleogryllus* (*Teleogryllus*) *gravelyi* (Chopard, 1928)	Palakkad (Parambikulam TR), Thiruvananthapuram, Ernakulam	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et a*l. (2023)
145.	*T.* (*Brachyteleogryllus*) *occipitalis occipitalis* (Serville, 1838)	Kozhikode	Vasanth (1991), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
146.	*T.* (*B.*) *rohinae* Jaiswara & Jain, 2021	Kasaragod	Jaiswara *et al*. (2021), Cigliano *et al.* (2023)
147.	*T.* (*Macroteleogryllus*) *mitratus* (Burmeister, 1838)	Kozhikode, Wayanad, Thiruvananthapuram	Chopard (1969), Vasanth (1991), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al*. (2023)
Genus *Velarifictorus* Randell, 1964			
148.	*Velarifictorus* (*Velarifictorus*) *aspersus* (Walker, 1869)	Palakkad (Silent Valley NP), Kozhikode	Shishodia and Hazra (1986), Vasanth (1991), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
149.	*V.* (*V.*) *fallax* (Chopard, 1969)	Malappuram	Vasanth (1991), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al*. (2023)
150.	*V.* (*V.*) *sahyadrensis* Vasanth, 1991	Wayanad	Vasanth (1991), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
151.	*V.* (*V.*) *saussurei* (Chopard, 1969)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020)
Tribe Modicogryllini Otte & Alexander, 1983			
Genus *Gryllopsis* Chopard, 1928			
152.	*Gryllopsis femorata* Chopard, 1935	Palakkad (Silent Valley NP)	Shishodia and Hazra (1986), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Modicogryllus* Chopard, 1961			
153.	*Modicogryllus* (*Modicogryllus*) *pallipes* (Chopard, 1925)	Kozhikode	Bhowmik (1977), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
154.	*M.* (*M.*) *signifrons* (Walker, 1869)	Kozhikode	Vasanth (1991), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
155.	*M.* (*Promodicogryllus*) *confirmatus* (Walker, 1859)	Palakkad (Silent Valley NP)	Shishodia and Hazra (1986), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Subfamily ENEOPTERINAE Saussure, 1874; Tribe Xenogryllini Robillard, 2004			
Genus *Indigryllus* Robillard & Jaiswara, 2019			
156.	*Indigryllus sagani* Jaiswara & Robillard, 2022	Kasaragod	Jaiswara *et al.* (2022), Cigliano *et al.* (2023)
Subfamily ITARINAE Shiraki, 1930			
Genus *Itara* Walker, 1869			
157.	*Itara* (*Itara*) *minor* Chopard, 1925	Kozhikode	Bhowmik (1977), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
158.	*I.* (*Gryllitara*) *pilosa* Meena & Swaminathan, 2021	Idukki (Vagamon)	Meena *et al.* (2021), Cigliano *et al.* (2023)
Subfamily LANDREVINAE Saussure, 1878; Tribe Landrevini Saussure, 1878			
Genus *Duolandrevus* Kirby, 1906			
159.	*Duolandrevus nairi* Vasanth, 1991	Kozhikode	Vasanth (1991), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily SCLEROGRYLLINAE Gorochov, 1985; Tribe Sclerogryllini Gorochov, 1985			
Genus *Sclerogryllus* Gorochov, 1985			
160.	*Sclerogryllus coriaceus* (Haan, 1844)	Kozhikode, Thiruvananthapuram, Ernakulam	Chopard (1969), Bhowmik (1977), Vasanth (1991), Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
161.	*S. punctatus* (Brunner von Wattenwyl, 1893)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Family MOGOPLISTIDAE Costa, 1855; Subfamily MOGOPLISTINAE Costa, 1855 Tribe MOGOPLISTINI Costa, 1855			
Genus *Derectaotus* Chopard, 1936			
162.	*Derectaotus indicus* (Chopard, 1928)	Kerala	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Family OECANTHIDAE Blanchard, 1845; Subfamily EUSCYRTINAE Gorochov, 1985			
Genus *Euscyrtus* Guérin-Méneville, 1844			
163.	*Euscyrtus* (*Osus*) *hemelytrus* (Haan, 1844)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020)
Genus *Patiscus* Stål, 1877			
164.	*Patiscus quadripunctatus* Bolívar, 1900	Thiruvananthapuram	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily PODOSCIRTINAE Saussure, 1878; Tribe Podoscirtini Saussure, 1878			
Genus *Indotrella* Gorochov, 2003			
165.	*Indotrella maindroni* (Chopard, 1928)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Genus *Madasumma* Walker, 1869			
166.	*Madasumma keralensis* Vasanth, 1991	Kannur	Vasanth (1991), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Mnesibulus* Stål, 1877			
167.	*Mnesibulus* (*Mnesibulus*) *fuscipennis* Chopard, 1928	Kerala	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Prozvenella* Gorochov, 2002			
168.	*Prozvenella saussureana* (Chopard, 1969)	Kozhikode	Vasanth (1991), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020)
169.	*P. soror* (Chopard, 1969)	Kozhikode (Kakkayam Reserve Forest)	Prabakar and Radhakrishnan (2004), Cigliano *et al.* (2023)
Family PHALANGOPSIDAE Blanchard, 1845; Subfamily PHALANGOPSINAE Blanchard, 1845 Tribe Phalangopsini Blanchard, 1845			
Genus *Arachnomimus* Saussure, 1897			
170.	*Arachnomimus* (*Arachnomimus*) *maindroni* (Chopard, 1969)	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
171.	*A.* (*A.*) *nietneri* (Saussure, 1878)	Ernakulam, Thiruvananthapuram	Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020)
Genus *Kempiola* Uvarov, 1940			
172.	*Kempiola flavipunctata* Desutter-Grandcolas, 2012	Kerala	Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily PARAGRYLLINAE Desutter-Grandcolas, 1987; Tribe Paragryllini Desutter-Grandcolas, 1987			
Genus *Pseudendacustes* Chopard, 1928			
173.	*Pseudendacustes gravelyi* Chopard, 1928	Thiruvananthapuram	Chopard (1969), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Family TRIGONIDIIDAE Saussure, 1874; Subfamily NEMOBIINAE Saussure, 1877			
Genus *Homonemobius* Chopard, 1935			
174.	*Homonemobius monomorphus* (Bolívar, 1900)	Thiruvananthapuram	Chopard (1969), Mathew (2004), Cigliano *et al.* (2023)
Tribe Burcini Gorochov, 1986			
Genus *Paranemobius* Saussure, 1877			
175.	*Paranemobius pictus* (Saussure, 1877)	Kerala	Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020)
176.	*P. vicinus* Chopard, 1928	Palakkad (Parambikulam TR), Ernakulam (Kochi), Kozhikode, Thiruvananthapuram	Chopard (1969), Bhowmik (1977), Vasanth (1991), Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Pteronemobiini Vickery, 1973			
Genus *Dianemobius* Vickery, 1973			
177.	*Dianemobius fascipes* (Walker, 1869)	Palakkad (Silent Valley NP), Kozhikode	Chopard (1969), Bhowmik (1977), Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Pteronemobius* Jacobson, 1904			
178.	*Pteronemobius* (*Pteronemobius*) *heydenii concolor* (Walker, 1871)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
179.	*Pteronemobius* (*Pteronemobius*) *pantelchopardorum* Shishodia & Varshney, 1987	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily TRIGONIDIINAE Saussure, 1874; Tribe Trigonidiini Saussure, 1874			
Genus *Metioche* Stål, 1877			
180.	*Metioche* (*Metioche*) *gigas* (Bolívar, 1900)	Thiruvananthapuram	Mathew (2004), Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Trigonidium* Rambur, 1838			
181.	*Trigonidium* (*Trigonidium*) *humbertianum* (Saussure, 1878)	Thiruvananthapuram	Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Superfamily GRYLLOTALPOIDEa Leach, 1815; Family GRYLLOTALPIDAE Leach, 1815 Subfamily GRYLLOTALPINAE Leach, 1815; Tribe Gryllotalpini Leach, 1815			
Genus *Gryllotalpa* Latreille, 1802			
182.	*Gryllotalpa africana* Palisot de Beauvois, 1820	Ernakulam, Thrissur, Alappuzha	Mathew (2004), Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et a*l. (2023)
Family MYRMECOPHILIDAE Saussure, 1874; Subfamily MYRMECOPHILINAE Saussure, 1874 Tribe Myrmecophilini Saussure, 1874			
Genus *Myrmecophilus* Berthold, 1827			
183.	*Myrmecophilus* (*Myrmophilina*) *albicinctus* Chopard, 1924	Ernakulam, Alappuzha	Chopard (1969), Mathew (2004), Bhaskar *et al.* (2018), Cigliano *et al*. (2023)
184.	*M.* (*Myrmophilina*) *americanus* Saussure, 1877	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020)
Infraorder TETTIGONIIDEA; Superfamily Stenopelmatoidea Burmeister, 1838 Family Gryllacrididae Blanchard, 1845; Subfamily Gryllacridinae Blanchard, 1845 Tribe Capnogryllacridini Cadena-Castañeda, 2019			
Genus *Capnogryllacris* Karny, 1937			
185.	*Capnogryllacris nigripennis* (Gerstaecker, 1860)	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)

Genus *Diaphanogryllacris* Karny, 1937			
186.	*Diaphanogryllacris dravida* (Karny, 1929)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Eremini Cadena-Castañeda, 2019			
Genus *Eremus* Brunner von Wattenwyl, 1888			
187.	*Eremus basalis gravelyanus* (Griffini, 1915)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
188.	*E. longicauda* Pictet & Saussure, 1893	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Gryllacridini Blanchard, 1845			
Genus *Eugryllacris* Karny, 1937			
189.	*Eugryllacris panteli* (Bolívar, 1900)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Gryllacris* Serville, 1831			
190.	*Gryllacris* (*Pardogryllacris*) *spuria* Brunner von Wattenwyl, 1888	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020)
Genus *Melaneremus* Karny, 1937			
191.	*Melaneremus canillii* (Griffini, 1915)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al*. (2023)
Family ANOSTOSTOMATIDAE Saussure, 1859			
Genus *Hypocophoides* Karny, 1930			
192.	*Hypocophoides biforaminatus* (Griffini, 1914)	Ernakulam	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al*. (2023)
193.	*H. indicus* (Bolívar, 1900)	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al*. (2023)
Family STENOPELMATIDAE Burmeister, 1838; Subfamily STENOPELMATINAE Burmeister, 1838 Tribe Oryctopterini Gorochov, 1988			
Genus *Oryctopterus* Karny, 1937			
194.	*Oryctopterus varuna* Hiremath & Prathapan, 2021	Thiruvananthapuram (Vellayani)	Hiremath and Prathapan, 2021, Cigliano *et al*. (2023)
195.	*O. yeshwanthi* Hiremath & Prathapan, 2021	Thiruvananthapuram (Kallar)	Hiremath and Prathapan, 2021, Cigliano *et al*. (2023)
Superfamily TETTIGONIOIDEA Krauss, 1902; Family TETTIGONIIDAE Krauss, 1902 Subfamily CONOCEPHALINAE Kirby & Spence, 1826; Tribe Agraeciini Redtenbacher, 1891			
Genus *Gonatacanthus* Karny, 1907			
196.	*Gonatacanthus pulcher* (Bolívar, 1900)	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)

Tribe Conocephalini Kirby & Spence, 1826			
Genus *Conocephalus* Thunberg, 1815			
197.	*Conocephalus* (*Anisoptera*) *maculatus* (Le Guillou, 1841)	Kerala	Shishodia *et al.* (2010), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Copiphorini Karny, 1912			
Genus *Euconocephalus* Karny, 1907			
198.	*Euconocephalus pallidus* (Redtenbacher, 1891)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily MECOPODINAE Walker, 1871			
Genus *Strongyloderus* Westwood, 1834			
199.	*Strongyloderus serraticollis* Westwood, 1834	Kerala	Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Mecopodini Walker, 1871			
Genus *Mecopoda* Serville, 1831			
200.	*Mecopoda elongata* (Linnaeus, 1758)	Idukki (Eravikulam NP)	Shishodia and Kulkarni (2002), Mathew (2004), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily MECONEMATINAE Burmeister, 1838; Tribe Meconematini Burmeister, 1838			
Genus *Xiphidiopsis* Redtenbacher, 1891			
201.	*Xiphidiopsis* (*Xiphidiopsis*) *malabarica* Kevan & Jin, 1993	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily PHANEROPTERINAE Burmeister, 1838			
Genus *Execholyrus* Henry, 1940			
202.	*Execholyrus allector* Henry, 1940	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Ducetiini Brunner von Wattenwyl, 1878			
Genus *Ducetia* Stål, 1874			
203.	*Ducetia japonica* (Thunberg, 1815)	Idukki (Eravikulam NP), Kottayam	Shishodia and Kulkarni (2002), Mathew (2004), Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Elimaeini Brunner von Wattenwyl, 1891			
Genus *Elimaea* Stål, 1874			
204.	*Elimaea bidentata* Brunner von Wattenwyl, 1878	Kerala	Bhaskar *et al.* (2018), Gupta and Chandra (2020), Cigliano *et al.* (2023)
205.	*E.* (*Neoelimaea*) *melanocantha* (Walker, 1869)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Orthelimaea* Karny, 1926			
206.	*Orthelimaea securigera* (Brunner von Wattenwyl, 1878)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)

Tribe Letanini Hebard, 1922			
Genus *Letana* Walker, 1869			
207.	*Letana bulbosa* Ingrisch, 1990	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Phaneropterini Burmeister, 1838			
Genus *Phaneroptera* Serville, 1831			
208.	*Phaneroptera* (*Phaneroptera*) *gracilis* Burmeister, 1838	Kerala	Gupta and Chandra (2020), Cigliano *et al.* (2023)
Subfamily PSEUDOPHYLLINAE Burmeister, 1838; Tribe Cymatomerini Brunner von Wattenwyl, 1895			
Genus *Tegra* Walker, 1870			
209.	*Tegra viridivitta* (Walker, 1870)	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)
Tribe Phyllomimini Brunner von Wattenwyl, 1895			
Genus *Acanthoprion* Pictet & Saussure, 1892			
210.	*Acanthoprion suspectum* (Brunner von Wattenwyl, 1895)	Kerala	Shishodia *et al.* (2010), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Phyllomimus* Stål, 1873			
211.	*Phyllomimus* (*Phyllomimus*) *nodulosus* Bolívar, 1900	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Genus *Pirmeda* Henry, 1940			
212.	*Pirmeda tenmalaia* Henry, 1940	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
213.	*P. travancora* Henry, 1940	Kerala	Shishodia *et al.* (2010), Chandra and Gupta (2013), Gupta and Chandra (2020), Cigliano *et al.* (2023)
Tribe Pseudophyllini Burmeister, 1838			
Genus *Onomarchus* Stål, 1874			
214.	*Onomarchus uninotatus* (Serville, 1838)	Thrissur	Sreeja *et a*l. (2023)
Family RHAPHIDOPHORIDAE Walker, 1869;Subfamily RHAPHIDOPHORINAE Walker, 1869 Tribe Rhaphidophorini Walker, 1869			
Genus *Rhaphidophora* Serville, 1838			
215.	*Rhaphidophora indica* Gorochov, 2013	Kerala	Bhaskar *et al.* (2018), Cigliano *et al.* (2023)

and questions the underestimated number of total Orthoptera species in the entire country given the uniqueness of the biodiversity in the Indian subcontinent and the relative lack of taxonomic expertise.

Solitarious individuals of *Schistocerca gregaria* were seen in some parts of southern states of India (Tamil Nadu, Karnataka and Kerala) during 2020. In Kerala *S. gregaria* solitary individual was recorded from Malappuram district, Kerala (Bhaskar *et al*., 2020b).

Some non-peer-reviewed journal articles with questionable distribution of taxa (as per Cigliano *et al.,* 2023) were not considered for this work. Even though "Malabar" is an officially obsolete name, it is used in this checklist because distribution status of some species recorded as Malabar and no other details are available (Kirby, 1914).

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Growth dilution and its effect on pesticide dynamics in okra

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ABSTRACT: Dissipation studies at single and double doses of chlorantraniliprole, thiamethoxam (25 and 50 g ai ha^{-1}), and imidacloprid (20 and 40 g ai ha^{-1}) were conducted on okra fruits following field application, and the residues were estimated using LC-MS/MS. The initial deposit of 0.42 and 0.80 mg kg^{-1} of chlorantraniliprole dissipated below quantitation level on the tenth day at single and double dosages. For thiamethoxam, the initial deposits of 0.42 and 0.71 mg kg^{-1} reached below quantitation level on tenth day at single dosage and on fifteenth day at double dosage; and for imidacloprid, the initial deposits are 0.10 and 0.16 mg kg^{-1} which dissipated below quantification level on fifth day. Growth dilution plays a significant role in the rate of dissipation of thiamethoxam when compared with imidacloprid and chlorantraniliprole when simulated dissipation rate due to growth dilution was calculated. The half-life of chlorantraniliprole, thiamethoxam and imidacloprid were 1.94 and 1.72 days, 1.88 and 1.99 days, 1.13 and 1.05 days, respectively for single and double dosages. Chlorantraniliprole, thiamethoxam and imidacloprid were found to be the safer insecticides with calculated waiting period of less than one day for single dosage.

KEYWORDS: Chlorantraniliprole, thiamethoxam, imidacloprid, dissipation, half-life, waiting period

INTRODUCTION

Okra (*Abelmoschus esculentus* L.) is an important vegetable crop of the tropical and subtropical regions of the world. However, the crop growth and yield is reduced by the attack of a number of pests like *Earias vittella* (Fab.), *Helicoverpa armigera* (Hubner) *Bemisia tabaci* Gennadius, *Phenacoccus solenopsis* Tinsley and so on. Hence application of pesticide is needed for the control of pests on this crop. The indiscriminate use of these pesticides will result in serious implications to man and his environment. Awareness of the dissipation dynamics of any insecticide is essential for determining their fate in the environment. Chlorantraniliprole, an anthranilic diamide class of insecticides, activates ryanodine receptors and stimulates calcium ion release from muscle cells causing paralysis and death in chewing insect pests. Neonicotinoids viz., thiamethoxam and imidacloprid with neurotoxic mode of action exhibit a variety of lethal and sublethal effects on insect feeding, oviposition and fecundity in Lepidoptera, Coleoptera and Hemiptera. Chlorantraniliprole is a reduced-risk pesticide which can be used to control lepidopteran pests while the neonicotinoids can be used for controlling beetles and sucking pests in

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okra. The combined application of pesticides with different action mechanisms can prolong the service life of pesticides and can reduce the generation of resistance (Wei *et al.,* 2020).

The rate of dissipation of insecticides is dependent on several factors including degradation, plant absorption, environmental factors, mode of application, plant species, growth dilution among which growth dilution plays a significant role. The effect of growth dilution in total dissipation is least studied and hence the present study was undertaken to gather quantitative information on the effect of growth dilution on dissipation behaviour of the commonly used insecticides chlorantraniliprole, thiamethoxam and imidacloprid on okra fruits.

MATERIALS AND METHODS

Chemicals and reagents: Analytical standards of chlorantraniliprole (purity 97.84 %), procured from Dr.Ehrenstorfer, Germany, thiamethoxam (purity 99.3%) and imidacloprid (purity, 99.9 %) were purchased from Sigma Aldrich. The formulations were obtained from E.I. DuPont India Pvt Ltd, Bayer Crop Science India Ltd and Insecticides (India) Ltd. Solvents like acetonitrile, anhydrous sodium sulfate, and anhydrous magnesium sulfate (ACS reagent grade) were obtained from Merck Germany and primary secondary amine (PSA) was obtained from Agilent Technologies, USA.

Preparation of standard solution: Method validation was carried out at 0.01, 0.1 and 0.5 mg kg^{-1} levels initially to test the efficiency of extraction and clean up procedures and to standardize the procedure for residue estimation. Standard stock solutions of chlorantraniliprole, thiamethoxam and imidacloprid (1000 ìg ml^{-1}) were prepared in LC-MS grade methanol and were serially diluted to obtain the solutions required for preparing a calibration curve (0.50, 0.25, 0.10, 0.075, 0.05, 0.025, and 0.0l ìg ml^{-1}).

Instrument Parameters: The chromatographic separation was achieved using Waters Acquity UPLC system equipped with a reverse phase Atlantis C-18 (2.1 mm × 100 mm, 5-micron particle size) column. The operation of the LC gradient involved the following two eluent components: A) 10 per cent methanol in water/ 0.1 per cent formic acid/5 m M ammonium acetate; B) 10 per cent water in methanol + 0.1 per cent formic acid + 5 m M ammonium acetate. The gradient elution was: 0 min. isocratic 20 per cent B, 0.0–4.0 min. linear from 20 to 90 per cent B, 4.0–5 min. linear from 90 to 95 per cent B, and 5–6 min. linear from 95 to 100 per cent B, with 6–8 min. for initial conditions of 20 per cent B. The flow rate was kept constant at 0.8 mL min^{-1}. and injection volume was 10 µL. The column temperature was kept at 40°C. The effluent from the LC system was introduced into AB Sciex API 3200 MS/MS system equipped with an electronspray ionization interface (ESI), operating in the positive ion mode. The source parameters were temperature 600°C; ion source gas (GS1) 50 psi, ion source gas (GS2) 60 psi, ion spray voltage 5,500 V, curtain gas 13 psi. The retention time for chlorantraniliprole, thiamethoxam and imidacloprid were 2.98, 0.91 and 1.1 minute, respectively.

Field experiment: Okra was raised in a farmer's field at Maranalloor, Thiruvananthapuram district and the trial was laid out in randomized block design plots of size 25 m^2 with three replicates. The crop was sprayed with chlorantranilirole (Coragen 18.5 SC) at 25 and 50 g ai ha^{-1}, thiamethoxam (Arrow 25 WG) at 25 and 50 g ai ha^{-1} and Imidacloprid (Confidor 17.8 SL) at 20 and 40 g ai ha^{-1} during the fruiting stage.

Extraction and clean up (QuEChERS method): The insecticide residues were estimated after collecting 500 g of tender fruits randomly on 0, 1, 3, 5, 7, 10, and 15 days after insecticide spray. The extraction and clean up was done as per the modified QuEChERS method (Anastassiades *et al.* (2003)). 250 g of the chopped samples per replicate were macerated in a blender. Ten grams of the ground samples were then taken from each replicate in a 50 ml centrifuge tube, 20 ml of HPLC grade acetonitrile was added and the sample was homogenised in a high speed homogenizer (Heidolph Silent Crusher-M) at 14000 rpm for 3-4 min. The sample was then mixed for 2 min on a rotospin after adding 4.5 g sodium chloride

(activated) and was then centrifuged for 5 min at 2,500 rpm. 12 ml clear upper layer of the sample was transferred into a 50- ml centrifuge tube prefilled with 5 g pre-activated sodium sulphate, and vortexed for 2 min. The extract was cleaned up by dispersive solid phase extraction (DSPE). Upper layer (8 ml) was transferred into a test tube containing 0.125 g primary secondary amine (PSA) and 0.8 g anhydrous magnesium sulfate and was again vortexed for 2 min, and centrifuged for 5 min at 2,500 rpm. Five mL of the supernatant liquid was evaporated to dryness under a gentle stream of nitrogen at 40°C and 7.5 psi flow rate. The residue was then reconstituted in 2 mL of methanol and filtered through a 0.2-micron filter for UPLC-MS/MS analysis.

Simulated dissipation rate due to growth dilution for each day is calculated by multiplying initial residue of pesticide with percentage weight gain of okra fruits from fruit setting stage to maturing (Miles et al. (1964)). This simulated data was compared with field data using excel spreadsheet. Half-life of the insecticides was calculated as per the procedure outlined by Hoskins (1961) and was done by Microsoft Excel 2007 spreadsheet.

RESULTS AND DISCUSSION

Efficiency of the method

The fortification at levels of 0.01, 0.05 and 0.1 mg kg^{-1} gave a good recovery percentage of 88 to 115, 92 to 118, 89 to 120 of chlorantraniliprole, imidacloprid and thiamethoxam, respectively (Table1). Hence the recovery and precision confirmed to the acceptable limits (recovery percentage: 70-120 and relative standard deviation values: below 20), establishing the suitability of the method. Matrix matched calibration was done and was found that and compared to the standard in pure solvent, in the calibration range of 0.01 to 0.50 ìg ml^{-1} (Figs.4, 5). The limit of quantitation (LOQ) for all these three insecticides were found to be 0.01 mg kg^{-1} and the limit of detection (LOD) being 0.005 mg kg^{-1}.

Persistence of chlorantraniliprole, imidacloprid and thiamethoxam

The data on the persistence of chlorantraniliprole (Fig.1), imidacloprid (Fig.3) and thiamethoxam (Fig.2) when applied at standard and double dosage on okra fruits revealed that the chlorantraniliprole and imidacloprid residues persisted for 10 days and 3 days at both dosages respectively, thiamethoxam residues persisted for 7 days and 10 days, at standard and double dosage, respectively. The initial deposit of chlorantraniliprole for single and double dose was 0.42 and 0.80 mg kg^{-1} which dissipated to 0.20 and 0.59 mg kg^{-1} respectively on the first day. However, the simulated values were 0.23 and 0.45 on first day which shows that there are environmental factors also along with growth dilution that will be affecting the dissipation of chlorantraniliprole. From the date of flowering, requires about five to seven days for the okra fruits to mature. Within this short period okra variety Anakomban grows at a faster rate ie., from 1 to 45

Table 1. Recovery of insecticides from okra

Dose (mgkg^{-1})	Recovery (%)	Precision (RSD)
Chlorantraniliprole		
0.01	88.4	15.6
0.05	106.2	16.2
0.1	115.3	14.8
Thiamethoxam		
0.01	89.7	20.6
0.05	120.0	11.3
0.1	117.6	16.4
Imidacloprid		
0.01	92.7	18.7
0.05	119	15.2
0.1	118.6	16.0

Table 2. Dissipation equations, half-life and waiting period of insecticides at two dosages

Dosage (g a.i.ha^{-1})	Regression equation	Half-life ($t_{1/2}$)	Waiting period (days)
Chlorantraniliprole			
25	y = -0.16X+1.59	1.94	0.74
50	y = -0.18X+1.89	1.72	2.36
Thiamethoxam			
25	y = -0.16X+1.58	1.88	-0.7
50	y = -0.15X+1.88	1.99	1.2
Imidacloprid			
20	y = -0.27X+1.00	1.13	-4.8
40	y = -0.29X+1.27	1.05	-3.5

g and showing growth dilution up to 97 per cent that may result in the rapid decline of the insecticide. Hopkins *et al.* (1952) also examined the importance of growth as a factor in the reduction of residues on alfalfa in New York state. The degradation half-life of chlorantraniliprole was less than one day and the waiting period calculated from the residue dissipation data and compared with FSSAI MRL was found to be less than one day for standard dose and 2.4 day for double dose. Similar results were obtained in another dissipation study of chlorantraniliprole (Coragen 18.5 SC) in okra fruits at single and double doses of 30 and 60 g ai ha $^{-1}$, and the initial residues were 0.48 and 0.91 mg kg^{-1}, respectively which reached below detectable level of 0.01 mg kg $^{-1}$ on the 10th day. Half-life of chlorantraniliprole were 0 and 1.20 days, respectively (Vijayasree *et al*., 2015). In another study by Singla *et al*., 2020 it was shown that the residues dissipated to below the limit of quantification (LOQ) of 0.03 mg kg^{-1} after 7 and 10 d of the application of insecticide at the two doses respectively. The half-lives (t1/2) and waiting periods of chlorantraniliprole in okra were calculated to be 2.27 and 2.45 d and 0 and 1d, at the recommended and double the recommended dosages respectively.

The initial deposit of imidacloprid was 0.10 and 0.16 mg kg^{-1}, dissipated to the residue concentration of 0.04 and 0.12 mg kg^{-1} respectively whereas the initial concentration of thiamethoxam was 0.42 and 0.71 mgkg^{-1} and 0.24 and 0.66 mg kg^{-1} on first day for single and double dose, respectively. The simulated rate of dissipation due to growth dilution at single and double dosages were 0.23 and 0.40 and 0.06 and 0.09 on the first day for thiamethoxam and imidacloprid, respectively. Hence the growth factor would have significantly contributed for the dissipation of thiamethoxam unlike imidacloprid. The degradation half-life of imidacloprid and thiamethoxam was 1.1 and 1.05 days and 1.88 and 1.99 days, respectively. The waiting period calculated for thiamethoxam was found to be less than one day for single dosage and 1.2 days for

Table 3. Simulated rate of dissipation of insecticide in okra due to growth dilution at two dosages

DAS	EGd (%)	Mean residue (Field) mgkg^{-1}		Residue (Simulated) mgkg^{-1}	
		x	2x	x	2x
chlorantranilprole					
0		0.42	0.80		
1	44	0.20	0.59	0.23	0.45
3	87	0.14	0.15	0.05	0.10
5	97	0.10	0.16	0.01	0.02
thiamethoxam					
0		0.42	0.71		
1	44	0.24	0.66	0.23	0.40
3	87	0.12	0.22	0.05	0.09
5	97	0.05	0.15	0.01	0.02
imidacloprid					
0		0.10	0.16		
1	44	0.05	0.12	0.06	0.09
3	87	0.02	0.03	0.01	0.02
5	97	BDL	0.01	0.00	0.00

Fig.1 Dissipation of chlorantraniliprole in okra

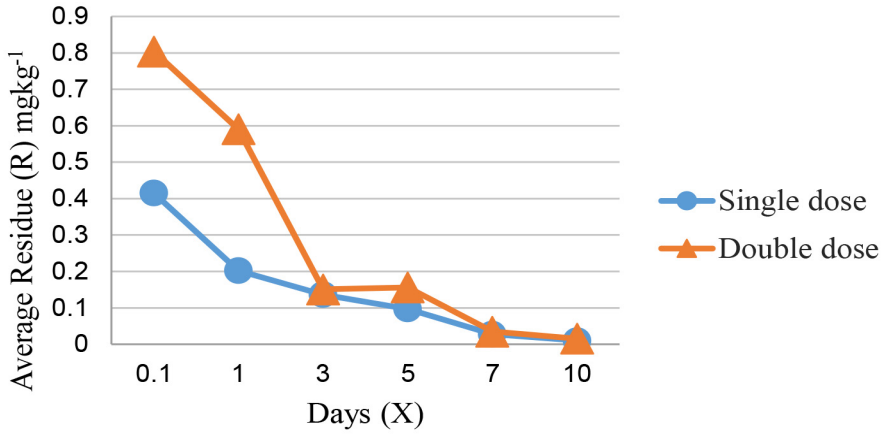


Fig. 2 Dissipation of thiamethoxam in okra

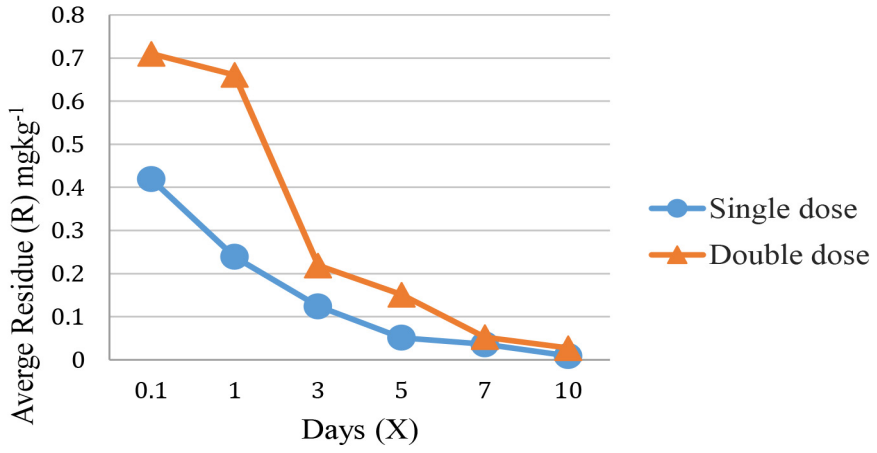
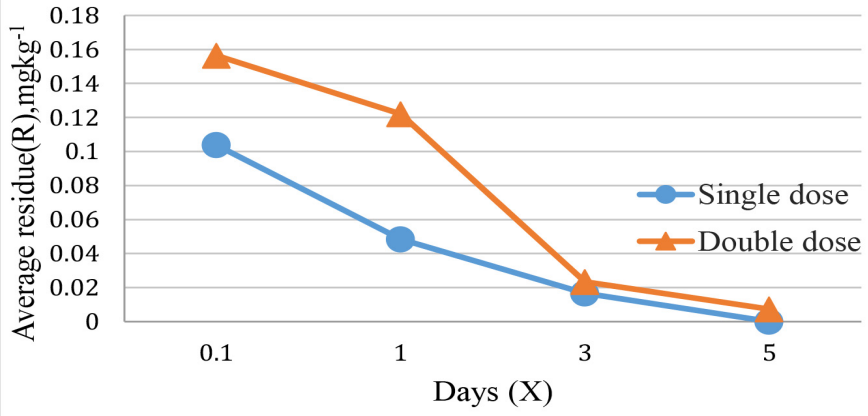


Fig.3 Dissipation of imidacloprid in okra



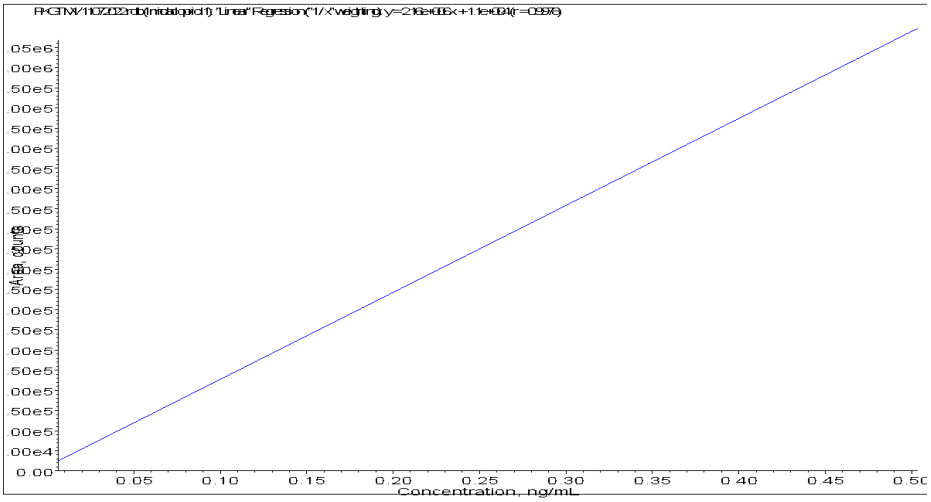


Fig. 4 linearity curve of Imidacloprid in LC-MS/MS

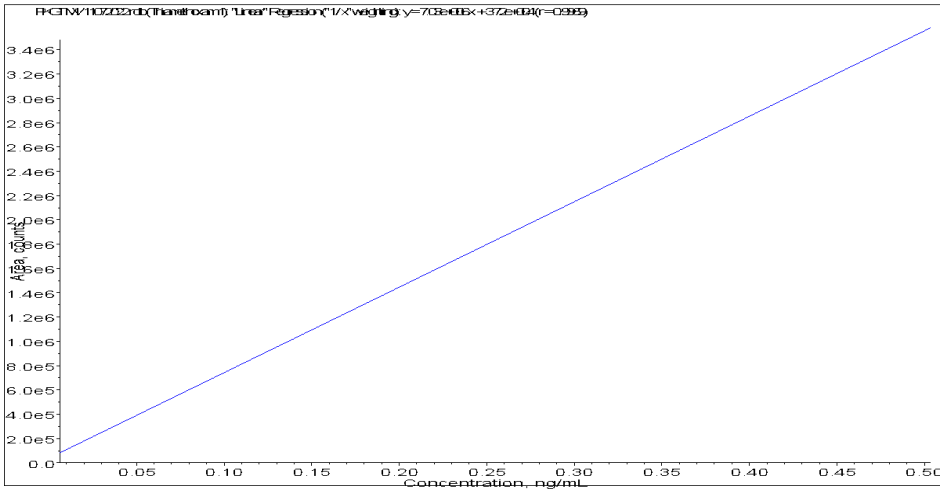


Fig. 5 linearity curve of thiamethoxam in LC-MS/MS

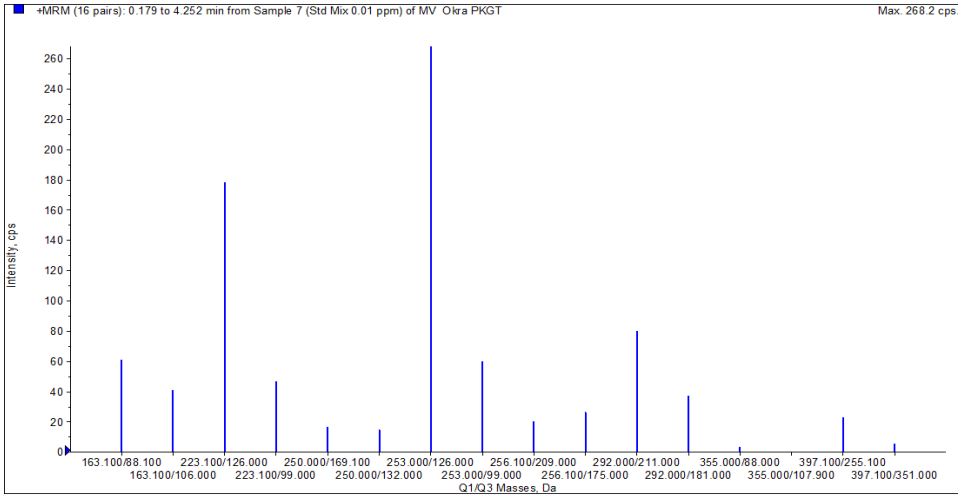


Fig. 6 MRM spectrum of pesticides: Imidacloprid, Thiamethoxam and Chlorantraniliprole

double dosage when compared with FSSAI MRL value (0.5 mgKg^{-1}). However, it was less than one day for both the dosages of imidacloprid as the FSSAI MRL value is 2 mgkg^{-1}. Dissipation studies by Pandit, 2016 found that the residues of imidacloprid in okra declined to below detection level (BDL) within 7 days in fruits when sprayed at the rate of 24.5 g a.i. ha^{-1} and the half-life (t1/2) ranged between 1.76 - 2.07 days in fruit. Joshi, 2019 reported that the initial deposit of 1.02 mg kg^{-1} residues of imidacloprid applied at dose of 20g a.i. ha^{-1} in okra dissipated in 7 days. According to Sharma, 2016 imidacloprid residues were the highest at the first picking after spray and lowest at the third picking and were in the range of 0.10 to 0.16 ug g^{-1} at the recommended dose of 0.3 mL L^{-1}. According to Chauhan, 2013 dissipation behaviour of thiamethoxam applied at recommended dose of 25 g ai ha^{-1} showed that the initial deposits of 0.245 mg kg^{-1} reached below detectable level of 0.005 mg kg^{-1} at 15 days after application with a half-life period of 1.47 day while Shalaby,2016 reported that okra fruits could be consumed safely after 15 days of treatment with thiamethoxam.

Among the different insecticides studied, it was found that growth dilution plays a significant role in the rate of dissipation of thiamethoxam. Even though, the presence of residues of chlorantraniliprole, imidacloprid and thiamethoxam were detected even after three days in okra, the observance of prescribed waiting period is a way of reducing the risk of the residue problems. And it was revealed that chlorantraniliprole thiamethoxam and imidacloprid are safer insecticides that can be used in okra when the waiting period was worked out.

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Studies on biology and preference of *Helicoverpa armigera* (Hubner) on different hosts and evaluation of botanicals for its management

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ABSTRACT: The biology and fitness of *Helicoverpa armigera* (Hubner) was studied on bottle gourd, lady's finger, chilli and tomato. The total larval duration was found to be 20, 22.5, 20.3 and 18.3 days on tomato, chilli, lady's finger and bottle gourd respectively. The total life cycle was of 35.25, 40.7, 34.2 and 31.3 days on tomato, chilli, lady finger and bottle gourd respectively. Shortest life cycle was observed on bottle gourd and longest on chilli. Feeding preference and fitness of *H. armigera* revealed that it preferred bottle gourd over other host plants. On bottle gourd, the pest recorded highest mean larval weight gain (0.112g/ day), while the lowest was on chilli (0.090g/ day). The feeding period was 8.073 days on bottle gourd and 8.266 days on chilli. Average food ingested on bottle gourd, lady finger, chilli and tomato were 3.083, 2.347, 2.076 and 1.988g respectively. Bioefficacy of botanicals (5 % aqueous extracts of periwinkle, giloy, tulsi and lantana) against *H. armigera* by leaf dip bioassay using leaves of tomato showed that the average food ingested in periwinkle, giloy, tulsi, lantana and control was 0.644, 0.944, 1.038, 0.985 and 2.297g respectively. This is the first report evidencing the insecticidal properties of aqueous extracts of giloy against *H. armigera.* © 2023 Association for Advancement of Entomology

KEY WORDS: Host preference, life cycle, insecticidal activity, periwinkle, giloy, tulsi, lantana

INTRODUCTION

Helicoverpa armigera (Hubner) is a polyphagous and cosmopolitan pest that affects important crops including cotton, pigeon pea, chickpea, corn, tomato, sorghum, millet, okra and sunflower (Manjunath *et al.*, 1989; Sharma, 2001). *H. armigera* is widely distributed all over Asia, central and southern Europe, America, Africa, and Australia (Tay *et al*., 2013). In India, this pest has been reported to cause 14-50 per cent damage on cotton (Kaushik *et al.,* 1969) and around 90 per cent damage in pulses (Patil *et al*., 2018). Even though there are numerous methods for reducing pests, each has its own set of advantages and disadvantages. Commercially available synthetic pesticides are reported to inflict severe environmental repercussions. Phytochemicals, mainly botanicals are currently a part of interest because of their successful application in plant protection as potential biocontrol

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agents. Hence the current study focuses on studies on biology and behavior of *H. armigera* on different host plants and to screen the bioefficacy of botanicals from commonly available plant species in managing *H. armigera* in laboratory bioassays.

MATERIALS AND METHODS

Mass culturing and biology of *Helicoverpa armigera* on different host plants

H. armigera culture was established with eggs purchased from NBAII, Bangaluru. Eggs were maintained at 25±2°C and 65±5 per cent relative humidity (RH). On hatching the neonates were placed in separate cups (7cm diameter x 15cm height) containing feed material (leaves of host plants under study [tomato (*Solanum lycopersicum* Linn.), bottle gourd (*Lagenaria siceraria* (Molina) Standl.), chilli (*Capsicum annuum* Linn.*)* and ladys finger (*Abelmoschus esculentus* (Linn.) Moench)] to avoid cannibalism and the cups were covered with muslin cloth. The insect was mass cultured using standard procedure with few modifications (Boopal *et al*., 2014). When larvae entered pre pupal stage, one-third of plastic cups were filled with moist sand, which provides optimal condition for pupation. When adults emerged, they were allowed into oviposition cage (20 x 20 x 20cm) for mating and egg laying. Honey solution (10 %) with few drops of vitamin E was provided as adult feed.

Evaluation of the biology of *H. armigera* on tomato, bottle gourd, chilli and ladys finger was carried out. Host plants were raised in plastic pots (60 x 30 x 30cm) without application of any agrochemicals for crop protection. Healthy plants of uniform growth and age 30 days after sowing (DAS) was provided as feed for first instar larvae in the cups. Total of three replications for each treatment and 10 larvae per replication were maintained. The insect was observed for their larval duration, pupal period, percent adult emergence, sex ratio and adult longevity.

Fitness analysis of *H. armigera* on different hosts

To study the host fitness, fully opened leaves from four hosts (tomato, bottle gourd, chilli and ladys finger) of uniform age 30 days after sowing (DAS) were taken weighed and placed in plastic cups. Three replications were maintained for each treatment with 10 larvae per replication. First instar larvae were weighed and released into individual cups and covered with muslin cloth. Twelve hours once fresh feed was provided and observations were made on larval weight gain, amount of feed consumed, weight of feces excreted etc. Also relative growth rate, approximate digestibility and consumption index was calculated as per the standard protocol (Waldbauer, 1968).

1. Relative Growth Rate (RGR) =

$$\frac{\text{Weight gained by the larva}}{\text{Mean larval weight} \times \text{feeding period (days)}}$$

2. Approximate Digestibility (AD) =

$$\frac{\text{Weight of food ingested - Weight of faeces}}{\text{Weight of food ingested}} \times 100$$

3. Consumption index (CI) =

$$\frac{\text{Weight of food ingested by the larva}}{\text{Mean larval weight} \times \text{Feeding period (days)}}$$

4. Efficiency of conversion of ingested food to body substance (ECI) =

$$\frac{\text{Weight gained by larva}}{\text{Weight of food ingested}} \times 100$$

5. Efficiency of conversion of digested food to body substance (ECD) =

$$\frac{\text{Weight gained by larva}}{\text{Weight of food ingested} - \text{Weight of feces}} \times 100$$

Bioefficacy of botanicals against *H. armigera* in laboratory bioassay

Fourth instar larvae of *H. armigera* were used for the bioassays. Leaves of giloy (*Tinospora cordifolia* (Willd.) Miers), periwinkle (*Catharanthus roseus* Linn.), tulsi (*Ocimum tenuiflorum* Linn.), and lantana (*Lantana camara* Linn.) were shade dried and grinded into powder. 2.5g powder of each plant was soaked in 50ml of water overnight to prepare 5 per cent of solution and then it was filtered with muslin cloth to get

aqueous extract of the botanicals used for study. Host plant leaves were weighed and dipped in each extract of botanicals by leaf dip method for bioassays and provided as feed to the larvae. In each treatment, four replications with one larva per replication were maintained. After every 24h remaining feed was weighed and replaced with new feed and the process was continued and observations were made until larval mortality to evaluate bioefficacy of the aqueous extract (5%) of each botanical tested.

Statistical analysis: Completely randomized design (CRD) was adapted for all the laboratory experiments. All the data obtained was subjected to an analysis of variance (ANOVA). The means were separated by Duncan's New Multiple Range Test (DMRT) and statistical analysis was carried out using SPSS v.26.

RESULTS AND DISCUSSION

Biology of *H. armigera* on different host plants

Observations for larval development of each instar, total larval period, per cent pupation, pupal period, per cent adult emergence, adult longevity and sex ratio were recorded (Table 1). Growth index of *H. armigera* was 2.66, 1.48, 2.955 and 3.311 on tomato, chilli, ladys finger and bottle gourd respectively

Fitness of *H. armigera* on different host plants

Studies showed that weight gained by larvae was maximum on bottle gourd and ladys finger, followed by tomato and chilli. However, the mean larval weight on bottle gourd, ladys finger, chilli and tomato were on par with each other. Food ingested was found to be highest on bottle gourd and weight of

Table 1. Biology of *H. armigera* on different hosts

Life stages*	Tomato	Chilli	Ladys finger	Bottle gourd
Larval period				
1st Instar	2.07±0.061	2.2 ± 0.040	1.9 ± 0.081	1.76 ± 0.028
2nd Instar	2.8 ± 0.040	3.02 ± 0.20	2.62 ± 0.102	2.65 ±0.122
3rd Instar	3.65± 0.04	3.95 ± 0.085	3.9 ± 0.081	3.35 ±0.122
4th Instar	3.3 ± 0.081	3.82 ± 0.102	3.75 ± 0.040	3.17 ± 0.020
5th Instar	3.87 ± 0.20	4.87 ± 0.061	4.13 ± 0.012	3.85 ± 0.040
6th Instar	4.15 ± 0.04	4.7 ± 0.081	4.06 ± 0.053	3.55 ± 0.122
Total period	20 ± 0.60	22.5 ± 0.721	20.3 ± 0.623	18.3 ± 0.516
Pupation(%)	53.3	33.3	60	60.6
Pupal period	12.6 ± 0.081	13.6 ± 0.204	11.6 ± 0.040	10.3 ± 0.081
Adult emergence%	25	20	27.7	30
Male longevity	9 ± 4.5	8 ± 4	10 ± 5	11 ± 5.5
Female longevity	10.3±0.408	10 ± 5	10.5 ± 0.353	11.4 ± 0
Sex ratio	3:1	1:1	4:1	5:1
Life cycle	35.25±0.93	40.7 ± 0.081	34.2 ± 1.306	31.3 ± 0.326
Growth index	2.66±0.269	1.48 ± 0.028	2.955 ± 0.018	3.311 ± 0.086

Mean ± SE/ period (days); each treatment was replicated three times with 10 larvae per replication

faeces excreted was found to be lowest on chilli. The consumption index (CI), relative growth rate (RGR) and efficiency of conversion of ingested food to body (ECI) were statistically similar on all the hosts. Efficiency of conversion of digested food to body (ECD) was lowest in chilli and highest in bottle gourd and approximate digestibility (AD) was highest in chilli (Table 2).

Bioefficacy of botanicals against *H. armigera* in laboratory bioassays

Studies on bioefficacy of aqueous extracts of periwinkle, giloy, lantana and tulsi by leaf dip bioassay using tomato leaves showed that the larvae consumed less on plants treated with botanical extracts. Mean larval feeding per day was 0.140, 0.144, 0.165, 0.179 and 0.366g on periwinkle, giloy, tulsi, lantana and control respectively. Larval mortality in days recorded in each treatment showed mortality occurred earlier in larvae forced to feed on diet containing periwinkle extract (5.2 days), followed by those on lantana, giloy, tulsi which were 6.0, 6.25, 6.7 days respectively while the larvae feeding on untreated leaves survived and also pupated (Table 3). In addition to larval mortality, larvae fed on tomato leaves treated with botanicals

Table 2. Effect of different host plants on the growth parameters (mean ± SE) of *Helicoverpa armigera*

Host*	Wt. gained (g)	Larval wt.(g)	Food ingested (g)	Wt. of faeces(g)	Feeding days	CI	RGR	ECI	ECD	AD
Bottle gourd	1.081± 0.070 (1.257)^{b}	0.112± 0.004 (0.782)^{a}	3.083± 0.169 (1.892)^{b}	0.816± 0.982 (1.024)^{b}	8.073± 0.322 (2.927)^{a}	9.577± 2.935 (3.174)^{a}	0.977± 0.029 (1.215)^{a}	28.518± 1.225 (5.386)^{a}	40.970± 3.124 (6.439)^{b}	78.168± 1.386 (8.869)^{a}
Lady finger	0.885± 0.059 (1.170)^{ab}	0.08± 0.003 (0.766)^{a}	2.347± 0.095 (1.687)^{a}	0.468± 0.032 (0.983)^{ab}	7.5± 0.173 (2.828)^{a}	12.105± 0.670 (3.550)^{a}	1.080± 0.118 (1.256)^{a}	27.880± 1.147 (5.327)^{a}	35.316± 1.702 (5.984)^{ab}	80.778± 3.093 (9.015)^{ab}
Chilli	0.703 ± 0.096 (1.096)^{a}	0.090± 0.024 (0.768)^{a}	2.076± 0.159 (1.604)^{a}	0.127± 0.018 (0.791)^{a}	8.266± 0.612 (2.960)^{a}	8.647± 0.844 (3.024)^{a}	1.014± 0.005 (1.230)^{a}	27.251± 2.048 (5.267)^{a}	29.439± 2.635 (5.471)^{a}	94.899± 0.051 (9.767)^{c}
Tomato	0.796± 0.148 (1.138)^{a}	0.076±0 (0.75)a	1.988± 0.0527 (1.577)^{a}	0.317± 0.174 (0.903)^{ab}	7.133± 0.163 (2.762)^{a}	11.337± 1.115 (3.440)^{a}	1.019± 0.006 (1.232)^{a}	26.905± 1.222 (5.234)^{a}	36.355± 1.637 (5.866)^{ab}	85.844± 0.530 (9.292)^{b}

In column, means followed by a common letter are not significantly different by DMRT (P=0.05); Figures in parentheses are square root transformed values; *Replicated three times with 10 larvae per replication

Table 3. Bioefficacy of botanicals (5% aqueous extracts) on *H. armigera* (Mean± S.E.)

Treatments	Total food ingested (g)*	Food ingested per day (g)*	Mortality (Days)*
T1 (Periwinkle)	0.664 ± 0.123(1.078)^{a}	0.140 ± 0.035(0.8)^{a}	5.2 ±1.767(2.397)^{b}
T2 (Giloy)	0.944 ± 0.009(1.201)^{a}	0.144 ± 0.009(0.802)^{a}	6.25 ±0.353(2.598)^{b}
T3 (Tulsi)	1.038 ± 0.353(1.240)^{a}	0.165 ± 0.014(0.815)^{a}	6.7 ± 2.121(2.692)^{b}
T4 (Lantana)	0.985 ± 0.072(1.218)^{a}	0.179 ± 0.035(0.824)^{a}	6 ± 1.414(2.549)^{b}
Control	2.297 ±0.211(1.672)^{b}	0.366 ± 0.067(0.930)^{b}	0 ± 0(0.707)^{a}

In column, means followed by a common letter are not significantly different by DMRT (P=0.05) ; Figures in parenthesis as square root transformed values; * Mean of four replications.

also showed abnormalities like swollen body, early pupation and malformation in pupal stage.

Studies on biology showed that the eggs of *Helicoverpa* was hemispherical in shape and yellowish white in color and later turns darker before hatching. Similar findings have been reported by Ali *et al*. (2009), Patel *et al*. (2011), Sharma *et al*. (2019). In the present investigation it was observed that the larvae underwent six larval instars which were similar to Gandhiya *et al*. (2014) and Sharma *et al*. (2019). The total larval period on chilli found as 22.5 days which was on par similar to findings of Patil *et al*. (2018) where they observed total larval period of 21-25 days on chilli. Total larval period of ladys finger, tomato, and bottle gourd was found to be 20.3, 20.0, and 18.3 days respectively. In the experiments the initial color of pupa was light green to yellow which later turned into dark brown which is similar to the report of Singh (2014). The pupal period was highest on chilli (13.6 days), followed by tomato (12.6 days), lady finger (11.6 days) and bottle gourd (10.3 days). Percent pupation and adult emergence were higher in bottle gourd and lady finger followed by tomato and chilli. Similar trend was observed with respect to adult longevity where that reared-on bottle gourd had highest adult longevity in case of both males and females and females lived longer than males. Previous studies also reported variations in total larval period, pupal period, percent pupation, adult emergence and adult longevity of *Helicoverpa* grown on different hosts (Ali *et al*., 2009; Patel *et al*., 2011). In the current study, *Helicoverpa* fed on bottle gourd had shortest life cycle of 31.3 days and those fed on chilli had the longest lifecycle with 40.7 days. Also the larval weight gain was greater on bottle gourd with 1.081 g and lowest on chilli with 0.703 g. The value of ECI was higher on bottle gourd with 28.518 per cent followed by lady finger, chilli and tomato with 27.880, 27.251 and 26.905 per cent respectively. ECD was found higher on bottle gourd (40.97%) and lower on chilli (29.439%). In this study, the RGR of the test insects was on par on all the host plants used for the study. However, AD was highest in chilli (94.899%) and lowest on bottle gourd (78.168%). Hemati *et al*. (2012) also reported AD of *Helicoverpa* varied with host plants. *Helicoverpa* preferred bottle gourd than other hosts. This might be because bottle gourd leaves are more succulent and remains moist than tomato, chilli and lady finger for long time and provides adequate amount of moisture and nutrition to the larvae and help them to complete their life cycle early. Chilli was the least preferred host as it showed high larval mortality and less larval weight gain with longer life cycle (egg to adult emergence). This might be because of xenobiotic resistance or plants metabolites present. Based on larval weight gained, feeding period with shorter life cycle of *Helicoverpa* reared on bottle gourd, it was observed to be the most preferred host among the plants tested. Thus, it is evident that host plant composition affects insect biology and duration of various biostages.

In the current study aqueous leaf extracts (5%) of all the four plants namely periwinkle, giloy, tulsi, and lantana tested gave better reduction in larval feeding over control. *Helicoverpa l*arvae feeding on treated leaves consumed two times less than those on control. Optimal feeding (2.297g) with no mortality in total experimental period was recorded on untreated leaves while leaves treated with periwinkle extract recorded lowest feeding (0.664g) and early mortality (5.2 days) followed by Giloy where larval feeding per day was 0.944g and mortality occurred in 6.25 days. This was followed by tulsi and lantana. This is the first report on insecticidal properties of aqueous extract of giloy against *H. armigera*. Simmonds *et al*. (2001) reported high antifeeding compounds which are extracted from different plants and used against *Helicoverpa* larvae. Ramya *et al*. (2008) reported 90 per cent feeding resistance on host plant treated with *Datura stromonium* and *Calotropis procera* aqueous extracts. Insecticidal properties of neem have been well established against many insects including *Helicoverpa*. In the current study there were reduced larval feeding, larval edema, larval mortality, pupal malformation and pupal mortality of insects fed with food poisoned with botanicals. This is due to the presence alkaloids, steroids, sesquiterpenes, saponins, tannins, flavonoids, aliphatics with vincristine and vinblastine like compounds present in these botanicals which has

multiple mode of action on insects such as feeding and ovipositional deterrents, acting as antimetabolies, molting inhibitors etc., meddling with insect, growth, metamorphosis and reproduction (Wahengbam *et al*., 2021). Current investigation provides with basic understanding of biology and host preference of *Helicoverpa* so as to use them appropriately in crop rotation and integrated pest management programs. The study shows that aqueous extracts of periwinkle, giloy, tulsi and lantana could effectively be used to manage *Helicoverpa* thereby providing farmers with a simple extraction technique which is a cheaper and ecofriendly option of pest management.

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Randomized detection of *kdr* allele frequencies in wild populations of *Aedes aegypti* (Diptera, Culicidae) in Colombo District, Sri Lanka

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ABSTRACT: Sri Lanka is one of the most affected countries in South Asia by dengue fever, with the number of dengue cases increasing over the last five years. The main strategy for managing disease outbreaks is to reduce infected vector populations with pyrethroid insecticides. However, extensive pyrethroid exposure has resulted in an increase in the selection of knockdown resistance mutations in *Aedes aegypti* (Linnaeus) (Diptera, Culicidae) voltage-gated sodium channel (*vgsc*) gene that confer pyrethroid resistance. Colombo district records the highest dengue incidence across the country each year, thus a failed vector control program will be a major threat to public health. Multiplexed Allele-specific PCR was used to genotype *kdr* alleles in wild *Ae. aegypti* mosquitoes obtained via random sampling from Wellawatte, Borella, and Battaramulla areas in the Colombo district. This study presents the co-occurrence of F1534C and V1016G *kdr* mutations from a randomized population in the Colombo district. 1534C mutant allele was predominant (with a 56.7% frequency) and 1016G was prevalent in 32.5 per cent of the population. The heterozygous mutant 1016VG genotype showed the highest distribution (with a 65% frequency) and the incidence of 1534FC was 56.7 per cent. Interestingly, 1016GG was completely absent and the FC/VG mutation combination had the highest incidence with 46.7 per cent. Furthermore, 82.36 per cent of individuals with the 1534FC genotype also had the 1016VG genotype, indicating a high prevalence of pyrethroid resistance in the studied population.

KEYWORDS: Pyrethroid, mutations, knockdown resistance, alleles, genotype

INTRODUCTION

Dengue, a prevalent arboviral disease in Sri Lanka, has reported approximately 65,000 cases annually over the past decade (Epidemiology Unit, 2022). Notably, the Colombo district, an urbanized area with unplanned human constructions, consistently reports the highest number of dengue cases in the island (Malavige *et al.,* 2021). The main vector responsible for transmitting the dengue virus

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(DENV) is *Aedes aegypti* (Linnaeus) (Diptera, Culicidae) (WHO, 2009). In the absence of an effective drug or vaccine for DENV infection, vector control is the main strategy in controlling the infections (WHO, 2009). Sri Lanka primarily employs space spraying using pyrethroids as the main vector control strategy (Karunaratne *et al*., 2013). Pyrethroids can bind to voltage-gated sodium channels (*vgsc)* in the nervous system of *Ae. aegypti*, thus hindering the maintenance of voltage difference across the membrane. Consequently, the mosquito will be imperiled to rapid paralysis and sudden death. This phenomenon is often referred to as "Knockdown" (Dong, 2014). However, the emergence of resistance to pyrethroids in *Aedes* species has been documented in various regions globally, including Asia, the Americas, the Middle East, and Africa (Moyes *et al*., 2017; Mashlawi *et al*., 2022). Thus, the efficacy of this approach has been compromised due to the widespread occurrence of knockdown resistance (*kdr*) mutations in *Ae. aegypti* populations (Fernando *et al*., 2018, 2020). Notably, a total of 15-point mutations occurring at the *vgsc* have been associated with *kdr* globally, with five confirmed to confer pyrethroid resistance, namely S989P, I1011M, V1016G/I, and F1534C, along with the recently discovered V410L (Du *et al*., 2016; Haddi *et al*., 2017).

Among these mutations, F1534C and V1016G have shown extensive distribution in various populations worldwide, with a notable occurrence in Asian *Ae. aegypti* populations (Linss *et al*., 2014; Vera-Maloof *et al*., 2015; Al Nazawi *et al*., 2017; Brito *et al*., 2018; Fernando *et al*., 2018; Ranathunge *et al*., 2021). Furthermore, evidence suggests that their co-occurrence confers a higher level of pyrethroid resistance than when they occur singularly (Du *et al*., 2013, 2016; Linss *et al*., 2014; Vera-Maloof *et al*., 2015; Saavedra-Rodriguez *et al*., 2018). To monitor the efficiency of current vector control strategies, regular assessments of pyrethroid resistance in *Ae. aegypti* populations are essential (Kushwah *et al*., 2020; Wuliandari *et al*., 2020). Genotyping *kdr* alleles, especially those confirmed to confer pyrethroid resistance, is a valuable tool in predicting the efficacy of pyrethroids in the field (Du *et al*., 2013, 2016; Linss *et al*., 2014; Vera-Maloof *et al*., 2015; Saavedra-Rodriguez *et al*., 2018). However, most studies have focused on genotyping pyrethroid-resistant individuals, emphasizing the need for randomized sampling from wild-caught adult populations to provide an accurate representation of *kdr* mutation distribution in a specific area (Du *et al*., 2016; Linss *et al*., 2014). The present study documents the co-occurrence of F1534C and V1016G *kdr* mutations, their distribution, and mutation associations in randomized populations from the Colombo district, Sri Lanka.

MATERIALS AND METHODS

Mosquito sampling and rearing: Wellawatte (6.8741°N; 79.8605°E), Borella (6.9122°N; 79.8829°E), and Battaramulla (6.897994°N; 79.922287°E), localities in the Colombo district were selected for sample collection as they were among the areas with the highest reported dengue cases on the island during 2021 (Epidemiology Unit, 2022). Also, these sites are routinely sprayed with permethrin and deltamethrin (National Dengue Control Unit Sri Lanka, 2016). Preimaginal stages (eggs, larvae, and pupae) (F_0) of *Ae. aegypti* mosquitoes were collected from January to April 2022, by placing around 20-30 ovitraps in 20-25 randomly selected neighboring houses from each locality for 5-7 days. Each house was accommodated with a maximum of two ovitraps at a distance between 5 to 10m apart from each other based on the structural design of the house. Ovitraps were set up considering favored breeding places of *Ae. aegypti*, such as dark, shady places with more human presence and less exposure to direct sunlight (Brown *et al.,* 2011; Rakotoarivony and Schaffner, 2012). The collected samples were transported to the insectary at the Centre for Biotechnology, University of Sri Jayewardenepura. The eggs collected from all the localities were hatched in separate containers to avoid any contamination of samples. Subsequently, the emerging larvae were fed with high-protein fish feed, and once emerged; the adults were supplied with a 10 per cent sucrose solution. All larvae and adults (F_{-0}) were maintained at 28 ± 2^0C with a relative humidity of 75 ± 10 per cent. Female *Ae. aegypti* were morphologically

identified to the species level based on the thorax patterns (WHO, 2020). Identified female *Ae. aegypti* adults were killed by deep freezing, and DNA was extracted from single mosquitoes by modified phenol-chloroform DNA extraction protocol (Ballinger-Crabtree *et al.,* 1992).

Genotyping of F1534C and V1016G *kdr* mutations using Multiplex Allele-specific PCR: A total of 60 samples from three collection sites were screened for *kdr* mutations at 1016 and 1534 mutation sites following a multiplex allele-specific (MAS) PCR protocol developed by Saingamsook *et al.* (2017). The PCR primer pair used, the region amplified in the *vgsc* gene, and the product sizes are shown in Table 1. Each PCR reaction was performed in a 25µl volume containing: 5ng of DNA sample, 2µM of $MgCl_2$ (Promega®),7.7µl of *Taq* Ready mix (2X) (FastGene®), and primers (Sigma-Aldrich Solutions®): Gly1016f (1.25µM), Val1016r (0.625µM), Gly1016r (1.25µM), c1534-f (0.625µM), c1534-r (0.625µM), Ae1534F-r (0.25µM) and Ae1534C-f (1.25µM). The amplification consisted of 92 °C for a 2 min heat activation step, followed by 35 cycles of 95 °C for 30 s, 55 °C for 30 s, and 72 °C for 30 s with a 2 min final extension step at 72 °C. PCR products were loaded onto 3 per cent (TBE) agarose gel and electrophoresis was conducted for 50min at 100V with a 50 bp DNA ladder (Promega USA). The scoring of *kdr* alleles was done according to Fig.1 and table 2.

Statistical Analysis: Allele frequencies, genotype frequencies, and Hardy-Weinberg equilibrium of F1534C and V1016G loci were calculated using the HW-test software (Santos *et al.,* 2020). Hardy–Weinberg equilibrium (HWE) of *kdr*-alleles in a population was tested using Fisher's exact test. Wright's inbreeding coefficient was calculated using the formula F = (He - Ho/He), where 'He' is expected heterozygosity and 'Ho' is observed heterozygosity. Pairwise linkage disequilibrium coefficients and associated chi-squared tests between F1534C and V1016G loci were calculated using LINKDOS (Garnier-Gere and Dillmann, 1992) following previously described guidelines (Saavedra-Rodriguez *et al.,* 2018; Vera-Maloof *et al.,* 2015). Genotype data from adjacent sites Wellawatte and Borella were pooled to increase the sample size for detecting linkage disequilibrium. The prevalence of all six genotype combinations was calculated and graphed by Microsoft Excel (2018). The proportion of 1016VG genotype distribution in individuals who are heterozygous mutants for F1534C mutation was calculated and visualized by R Studio Team (2022).

RESULTS

MAS PCR genotyping of *kdr* mutations of *Ae. aegypti* populations:

Genotyping results of *kdr* alleles at loci 1534 and 1016 carried out on 60 field-collected F_0 populations from all collection sites, indicated that F1534C was the most widespread *kdr* point mutation with 56.7 per cent of the individuals having the heterozygous mutant genotype 1534FC and 28.3 per cent of them being homozygous mutant 1534CC while only 15 per cent were homozygous wild type 1534FF. Overall, the V1016G mutation was less common compared to F1534C. Among the individuals 65 per cent had the heterozygous mutant genotype 1016VG, while only 35 per cent of them had homozygous wild type 1016VV and surprisingly, homozygous mutant genotype 1016GG was absent in all collection sites (Table 3).

Mutation combinations:

Six out of nine possible genotype combinations were observed in the 60 samples from all three locations. Frequency of each of the nine bi-locus combinations is depicted in Fig. 2. Only 10 per cent of the population had wild-type alleles at both 1534 and 1016 sites. Double mutant combinations were found only as FC/VG and CC/VG. The most common bi-locus genotype combination was heterozygous double mutant FC/VG with a percentage of 46.7 per cent (28 out of 60 individuals). Homozygous mutant 1534CC with homozygous wild type 1016VV (CC/VV) was the second most common combination (15%); and 13.3 per cent of the individuals had a double mutant CC/VG combination. Both FF/VV and FC/VV were present in 10 per cent while 5 per cent of the population had FF/VG combinations. No individual

Table 1. Sequences of Primers used in this study (Saingamsook *et al.,* 2017)

Primer	Primer sequence (5'-3')	Product size	[a]Exon
1016 genotyping			
Gly1016f	ACCGACAAATTGTTTCCC		15-16[b]
Val1016r	[short GC tail][c]AGCAAGGCTAAGAAAAGGTTAATTA	60	16
Gly1016r	[long GC tail][d]AGCAAGGCTAAGAAAAGGTTAACTC	80	16
1534 genotyping			
c1534-f	GCGTACCTGTGTCTGTTCCA	368	23
c1534-r	GGCTTCTTCGAGCCCATCTT		24
Ae1534F-r	GCGTGAAGAACGACCCGA	232	24
Ae1534C-f	CCTCTACTTTGTGTTCTTCATCATCTG	180	24

[a]Exon from the *Ae. aegypti* VGSC gene. This transcript corresponds to VectorBase Transcript ID AAEL006019
[b]Intron between exon 15 and 16
[C]Short GC tail sequence: 5^2 -GCG GGC-3'
[d]Long GC tail sequence: 5^2 -GCG GGC AGG GCG GCG GGG GCG GGG CC-3^2

was found to be a homozygous mutant for both F1534C and V1016G mutations. Homozygous 1534CC was only found in conjunction with homozygous wild type 1016VV and heterozygous mutant 1016VG at a combined frequency of 28.33 per cent (17 out of 60 individuals). No individual had a homozygous mutant 1016GG genotype from any collection site. Heterozygous mutant 1534FC was found only in combination with heterozygous mutant 1016VG and homozygous wild type 1016VV at a combined frequency of 56.7 per cent. In contrast, the heterozygous mutant 1016VG genotype was found in combination with all three genotypes of the 1534 mutation including 1534FF, 1534FC, and 1534CC at a combined frequency of 65 per cent from all collection sites. Also, VG was the most expressed genotype with a 65 per cent frequency.

Three patterns of mutational associations were identified: i). Nearly all (82.36%) of heterozygous mutant 1534FC individuals were heterozygous mutant for 1016VG loci, while the remaining (17.64%) were homozygous wild type for 1016VV; ii). Almost half of the individuals with (47.06%) homozygous mutant 1534CC had heterozygous mutant 1016VG genotype whereas the rest (52.94%) of the 1534CC population were having homozygous wild type 1016VV; and iii). None of the individuals had homozygous mutant 1016GG genotypes in all three collection sites.

Table 2. Criteria for the scoring of multiplex PCR amplified 1534 and 1016 alleles using agarose gel electrophoresis

Genotype	PCR bands present and their size (bp)
1534	
FF	Only one band at 232bp
FC	Two bands at 232 and 180bp
CC	only one band at 180bp
1016	
VV	Only one band at 60bp
VG	Two bands at 60 and 80 bp
GG	Only one band at 80bp

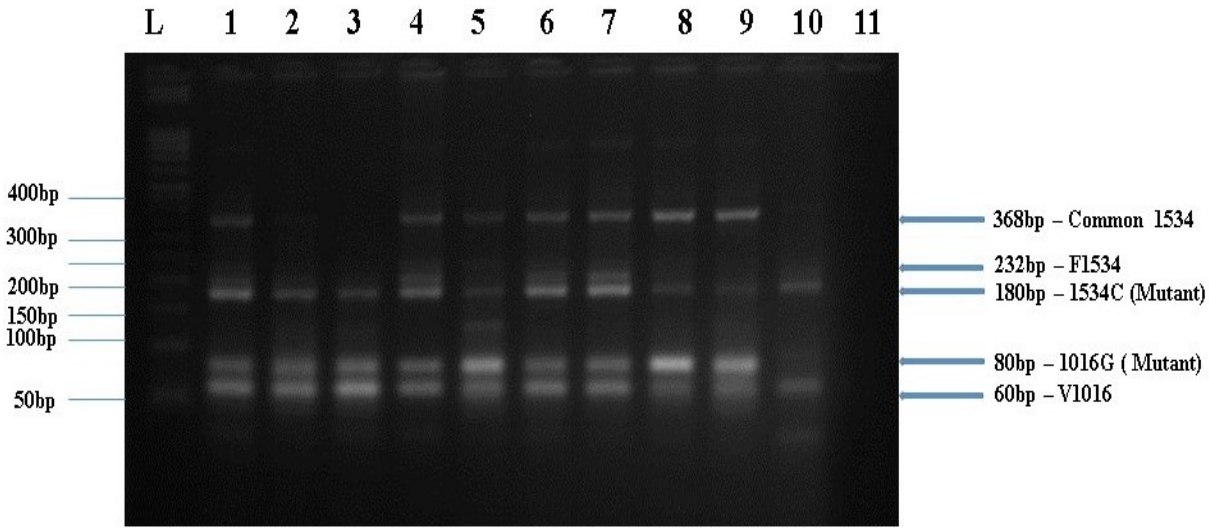

Fig. 1 Gel electrophoresis results. Heterozygous mutant genotypes for both F1534C and V1016G *kdr* mutations are shown in the 1st,4th,5th, 6th and 7th lanes. The 2nd,3rd,8th and 9th lanes show a genotype that is heterozygous mutant for the 1016 allele but homozygous mutant for the 1534 allele. The 10th lane contains homozygous mutant for 1534 allele and homozygous wildtype for 1016 allele. Lane L is the low molecular weight DNA ladder. The last lane (11) contains the negative control in which PCR water was used as the template in the multiplex PCR reaction

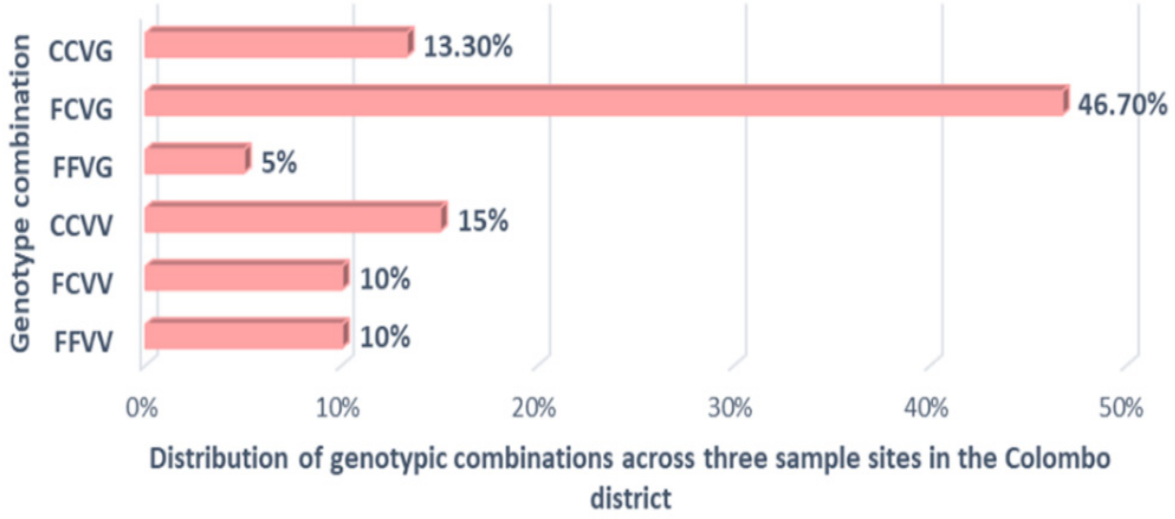

Fig. 2 Distribution of genotypic combinations across three sample sites in the Colombo district

Inbreeding coefficient and linkage disequilibrium:

Except for Battaramulla samples, the other two sites were in Hardy-Weinberg equilibrium for 1534 loci but only Wellawatte samples were in Hardy-Weinberg equilibrium for 1016 loci. When combined across sites, only the F1534C mutation was in Hardy-Weinberg equilibrium (P= 0.297) while the V1016G mutation showed a significant deviation from the equilibrium ($P< 10^{-4}$). Analysis of the Wright's inbreeding coefficient for F1534C and

V1016G *kdr* alleles revealed a negative value indicating the presence of higher heterozygotes than expected (*F*= -0.1547 and *F*= -0.4814 respectively). The results from Pairwise linkage disequilibrium analysis revealed that the linkage disequilibrium between F1534C and V1016G mutations is not consistently significant in the studied population (Table 3).

DISCUSSION

Identification of *kdr* mutations in natural populations of *Ae. aegypti* in the Colombo district can be useful in predicting pyrethroid resistance because it is routinely undergoing pyrethroid-adulticides spraying and also reporting the highest number of dengue cases every year. F1534C *kdr* mutation was first detected during 2016 (Fernando *et al.,* 2017) while V1016G was first reported in 2018 (Fernando *et al.,* 2018) in Sri Lankan *Ae. aegypti* populations. Following that F1534C and V1016G have been identified in pyrethroid-resistant *Ae. aegypti* as well as wild populations from numerous studies conducted in several areas across the island (Fernando *et al.,* 2017, 2018, 2020; Hegoda, 2017; Ranathunge *et al.,* 2019, 2021).

The present study documents the presence of two of the most prevalent *kdr* mutations F1534C and V1016G which have been established to confer pyrethroid resistance in the *vgsc* of *Ae. aegypti* (Du, 2013; Hirata *et al.,* 2014; Vera-Maloof *et al.,* 2015). In the current study, heterozygous mutant genotypes 1534FC and 1016VG predominated in the population with 56.6 and 65 per cent respectively, whereas only 1534CC was present as a homozygous mutant genotype with 28.33 per cent while 1016GG was absent in the population. These results are in line with a previous study finding where the 1534FC genotype was dominating the Colombo district samples with 43.7 and 56.8 per cent in 2018 and 2019 respectively (Ranathunge *et al.,* 2021). However, wild-type 1016VV was significantly higher in Colombo district populations with 79.1 and 74.5 per cent in 2018 and 2019 correspondingly (Ranathunge *et al.,* 2021).

Mutant allele 1534C frequency was significantly higher (56.7%), while in 1016G mutant allele it was lower (32.5%). Nonetheless, the VG genotype was present in 65 per cent of the population suggesting the presence of higher heterozygotes than expected with a negative value achieved for Wright's inbreeding coefficient (F= -0.4814). This is the highest reported VG percentage so far by random sampling from a wild population of *Ae. aegypti* in Sri Lanka. Double heterozygous genotype combination, FC/VG was found to have the highest prevalence across the sample sites (46.7%). Furthermore, the frequency of an individual with FC having VG genotype was always significantly higher for all the sites; while for Battaramulla sample, it was 100 per cent. Taken together, these results suggest strong evidence of an increased *kdr* incidence considering random sampling from a wild population and the relatively small sample sizes being examined.

Table 3. Result of *kdr* genotyping of *Ae. aegypti* in field population (F_0): distribution of allelic frequencies and their compliance to Hardy–Weinberg equilibrium and distribution of mutation combinations

Collection Site	F1534C allele frequencies				V1016G allele frequencies				PHWE test		WiC	
	FF (WT)	FC (MT)	CC (MT)	C allelle	VV (WT)	VG (MT)	GG (MT)	G allele	F1534 C	V1016 G	F1534 C	V1016 G
Wellawatte (n=20)	0.4	0.45	0.15	0.375	0.6	0.4	0.0	0.2	1.000	0.538		
Borella (n=20)	0.05	0.5	0.45	0.7	0.3	0.7	0.0	0.35	0.613	0.045		
Battaramulla (n=20)	0.0	0.75	0.25	0.625	0.15	0.85	0.0	0.425	0.015	0.001		
Total(60)	0.15	0.567	0.283	0.567	0.35	0.65	0.0	0.325	0.297	<10-4	-0.1547	-0.4814

WT = Wild type; MT = Mutant type; PHWE = P value of Fisher's exact test; WiC = Wright's in breeding coefficient

The high frequency of a mutant allele for F1534C mutation is similar to the findings from previous studies however with different sample sizes from the Colombo district (Fernando *et al.,* 2020; Ranathunge *et al.,* 2021) as well as from the Gampaha district (Fernando *et al.,* 2018) and Galle (Fernando *et al.,* 2020). Similarly, a significantly high frequency for 1534C was reported in a bioassay study with 63.9 per cent in permethrin and 57.5 per cent in deltamethrin-resistant samples from the Colombo district (Fernando *et al.,* 2018). Also, several studies established that F1534C can alone elicit resistance towards permethrin (Du *et al.,* 2013; Hirata *et al.,* 2014). Meanwhile, a high level of pyrethroid has been confirmed when V1016G is present with F1534C mutation in combination (Kushwah *et al.,* 2020; Plernsub *et al.,* 2016). This scenario has also been confirmed in Sri Lankan populations (Fernando *et al.,* 2020; Hegoda, 2017).

However, all the previous local studies had a lower 1016G allele frequency in any district than the results of this study (Fernando *et al.,* 2018, 2020; Ranathunge *et al.,* 2021). Similarly, the homozygous mutant 1016GG genotype was not detected from any site in this study. This lower frequency of the 1016G allele is likely due to more exposure to type I pyrethroids (permethrin) compared to type II pyrethroids (deltamethrin) in these collection sites, and/or the V1016G mutation has not yet been established in the population and is still evolving. However, this account must be approached with some caution due to the smaller sample sizes tested here and for the previous studies, not all the analyses were done in wild populations and possible inbreeding might have occurred.

When considering the global distribution of *kdr* mutations, the 1534C mutant allele frequency was particularly high in India (Kushwah *et al.,* 2020; Saha *et al.,* 2019), Taiwan (Biduda *et al.,* 2019), and Malaysian (Akhir *et al.,* 2022; Ishak *et al.,* 2015) *Ae. aegypti* populations. However, studies in Myanmar (Kawada *et al.,* 2014; Naw *et al.,* 2020), Africa (Ayres *et al.,* 2020), and Indonesia (Wuliandari *et al.,* 2020) demonstrated a high prevalence of V1016G mutation. Findings of a 12-year linkage disequilibrium analysis in the Mexican population suggested the 1534C mutation is the one that was selected first with low fitness cost thus facilitating a combination of *kdr* mutation to establish and confer a high level of pyrethroid resistance over time (Du *et al.,* 2016; Vera-Maloof *et al.,* 2015). Similarly, F1534C happened to be the first *kdr* mutation reported in the Sri Lankan population, and only after that, other *kdr* mutations including S989P and V1016G were reported. Interestingly 71.79% of VG individuals possessed FC genotype in this study and also several other studies demonstrated a high proportion of FC genotype in individuals who were heterozygous for V1016G mutation from Sri Lankan (Fernando *et al.,* 2020; Ranathunge *et al.,* 2021) as well as global populations (Kushwah *et al.,* 2020; Wuliandari *et al.,* 2020) thus supporting the hypothesis from Vera-Maloof *et al.* (2015). Also, it has been suggested that in Asian populations, 989P+1016G and 1016G haplotypes are more likely to emerge from the F1534C haplotype (Cosme *et al.,* 2020; Du *et al.,* 2016). Taken together, these findings suggest the constant high frequency of F1534C mutation in Sri Lankan populations may likely establish the origin and dispersion of S989P and V1016G mutations thus resulting in strong resistance to both types I and II pyrethroids.

Co-occurrence of S989P and V1016G in domain II of the *vgsc* gene was prevalent in Asian *Ae. aegypti* populations (Cosme *et al.,* 2020; Wuliandari *et al.,* 2020) while obtaining a perfect linkage disequilibrium between two mutations (Kushwah *et al.,* 2020; Wuliandari *et al.,* 2020). Also, this observation was later recorded in some Arabian populations as well (Al Nazawi *et al.,* 2017). Further, the combination of S989P and V1016G has exhibited a synergistic effect to confer resistance to deltamethrin (Hirata *et al.,* 2014; Srisawat *et al.,* 2010). However, most of the Asian populations have not shown a significant linkage disequilibrium between F1534C and V1016 (Cosme *et al.,* 2020; Du *et al.,* 2016; Kushwah *et al.,* 2020;). Likewise, this study reported, no significant linkage disequilibrium between F1534C and V1016G in both pooled samples. Therefore, it is likely that in Asian populations S989P+V1016G combination is more

prevalent than the F1534C+V1016G combination (Cosme *et al.,* 2020). More *kdr* genotyping analyses in the future will confirm the association between F1534C with V1016G and S989P with 1016G in Sri Lankan *Ae. aegypti* populations. Intriguingly, the F1534C mutation has always been in a high frequency while the 1016G mutant allele has been under 50% in all the local studies done up to date. However, in the Sri Lankan population, 1534C is more prevalent than 989P (Fernando *et al.,* 2018; Ranathunge *et al.,* 2021) and field identifications with larger sample sizes will explain this further. Even though previously published *kdr* occurrence in Sri Lankan *Ae. aegypti* populations are limited to a few studies, there has been a notable increase in *kdr* mutant allele frequencies through the years. Especially, a rapid increase was visible in 1534C allele frequency in 2017 compared to the 2015 collection from 17.5 (n=39) to 80.2 per cent (n=48) in the Colombo district (Fernando *et al.,* 2020). Another early study demonstrated a 71.5 per cent (n=156) frequency for the 1534C allele when combined across three sample sites from the Colombo district which had an extreme level of resistance to permethrin (Hegoda, 2017). In a recent study, F1534C and S989P *kdr* mutations were found from randomly selected *Ae. aegypti* samples from the Colombo district showed resistance towards Cyfluthrin, Permethrin, ë-Cyhalothrin, and DDT (Nugapola *et al.,* 2021).

The Colombo district has been reporting dramatic *kdr* incidence as opposed to other areas on the island through the years (Fernando *et al.,*2018,2020; Ranathunge *et al.,* 2019, 2021) and the highest recorded pyrethroid resistance in *Ae. aegypti* populations (Fernando *et al.,* 2018, 2020; Nugapola *et al.,* 2021) while being accounted for the area with the highest number of reported dengue cases every year (Epidemiology Unit, 2022; National Dengue Control Unit-Disease, 2022) Moreover, a lot of studies have explained that compared to *Ae. albopictus,* pyrethroid resistance in *Ae. aegypti* occur mainly due to *kdr* mutations rather than metabolic resistance (Leong *et al.,* 2019; Nugapola *et al.,* 2021). These collective pieces of evidence suggest the rapid increase in F1534C is not likely because of genetic drift or founder effect but caused by the convergent selection for resistance to DDT and/or pyrethroids. Therefore, on-field diagnosis of *kdr* alleles is vital in predicting pyrethroid resistance ideally before every spray season.

The findings suggest a possibility of high pyrethroid resistance. An excess of heterozygosity reported for both F1534C and V1016G mutations likely indicates an emerging selection of *kdr* alleles due to the constant exposure to the pyrethroids.

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Effect of heat shock on embryonic development and its impact on commercial traits of silkworm *Bombyx mori* L.

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ABSTRACT: The early stage of an organism – embryonic stage, architects all the post embryonic developments which are governed by genetic and environmental conditions, but the effect of hot events during that stage remain obscure in the silkworm *Bombyx mori* L. Thus, APM1; a multivoltine parental breed of a ruling CB and APHO1 silkworm breed developed through induction of thermotolerance, and APHO1 breed to examine the impact of heat shock (HS) on the embryo and resultant larvae. Different developmental stages of embryo were exposed to varied HS temperatures for 2 h followed by a 2 h recovery period. After HS the eggs and the resultant larvae were reared under normal environmental conditions. Interestingly, 45^{0}C although determined as lethal temperature yielded vibrant larvae. Whereas APM1 and APHO1 eggs heat shocked at 35^{0}C exhibited increased hatching (91.66 and 69.33%), larval weight (1.72 and 3.33 g), effective rate of rearing (72.39 and 81.93%), cocoon weight (1.01 and1.6 g), shell weight (0.12 and 0.29 g), shell ratio (13.11 and 20.52%) and pupal weights (0.87 and1.29g) when compared to control APM1 and APHO1. Besides increased total protein content, expression of 205 kDa, 90 kDa and 70 kDa heat shock proteins and the glycogen content was found more on day - 3 compared to day - 2 in the embryos of APM1 and APHO1 which eventually declined as the embryonic development proceeded to hatching. This work shows that APM1 and APHO1 eggs had shown profound response to HS temperatures exhibiting varied acquired thermotolerance to overcome fluctuating environmental condition.

KEY WORDS: Strains, APM1, APHO1, thermotolerance, protein, glycogen changes

INTRODUCTION

The sericulture industry has contributed significantly to the economic development of many countries due to the commercial importance of silk in the textile world as silkworms are easy to rear under domestication. Thus, the silkworm, *Bombyx mori* was not only exploited over a long period for cocoon

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production but also widely used in basic research, biotechnology and as a molecular model insect. In tropical countries like India, to achieve sustainable sericulture, silkworm strains that withstand high temperature and frequent fluctuations are in need. But, the domesticated silkworm, *B. mori,* is highly sensitive to the fluctuating environmental conditions during different stages of development due to poikilothermic nature. However, sensitivity varies among different silkworm races, strains, and breeds. Comparatively, bivoltine silkworm breeds have enough potential for the production of superior cocoons qualitatively and quantitatively over multivoltines but cannot thrive well in adverse or fluctuating environmental conditions. In India, the temperature and humidity vary from season to season, region to region, while fluctuation occur between dawn to dusk. Sericulturists experience a great risk in rearing silkworms. The fluctuated environment leads to improper growth of embryo, poor hatching, weak larvae, inferior cocoons and ultimately production of low-grade silk. Even, few hours of elevated temperature ~40° C and above in rearing house causes significant damage in commercial and biological traits of silkworm *B. mori* (Manjunatha *et al*., 2005 and Vasudha *et al*., 2006).

The effect of temperature on the development of *B. mori* has been studied extensively both in larval and embryonic stages but not much information is available on the impact of HS on hatching of the embryo which is an index of embryonic development (Manjunatha *et al*., 2005, 2008). In the field of sericulture, breeders agree that, the development of thermotolerant bivoltine breeds which are suitable for high temperature environment and yet productive by following conventional breeding strategy is a difficult task. Resistance to high temperature has been recognized as a heritable characteristic in silkworm and the possibility for the temperature tolerant silkworm races were suggested by "Kato" as early as 1989. Therefore, means other than the conventional breeding methods are to be adopted to attain the goal. With the aid of modern biotechnological tools, it may be possible to quantify the factors responsible for the expression of temperature tolerance. It was found recently that supplementation of spermidine in micromolar concentrations helps in thermotolerance (Anugata et al, 2022). The increased thermotolerance in spermidine supplemented silkworms was due to elevated levels of caldopentamine (Anugata et al, 2023). However, the mulberry silkworm is one of the most thermal-sensitive organisms. Intensive and careful domestication over centuries has apparently deprived this taxon of opportunities to acquire thermo-tolerance.

The terms heat shock and thermotolerance, acclimation and hardening are commonly used to describe the changes in an organism living state caused by external environments and treatments (Bowler, 2005; Loeschchke and Soresen, 2005; Langerspetz, 2006). Unfortunately, the usage of these terms in silkworm research has not been well defined and requires systematic study to draw a line between them. Comparatively, heat shock response among different silkworm races/strains of the polyvoltine, bivoltine and univoltine varies significantly and thermotolerance increases as the larval development proceeds (Vasudha *et al*., 2006; Manjunatha *et al*., 2010; Raju *et al.*, 2018; Shou-Min Fang *et al.*, 2021).

It is well known that improper management of light, temperature and humidity during embryonic developments affects the characters in successive generations. The effect of temperature on the development of silkworm larvae has been studied extensively, but there is a need to study the effect of temperature on the growth and development of silkworm embryos. Very few reports are available on the impact of heat shock on hatching, biochemical composition, biological and commercial traits that pre-determine the embryonic stage.

With this backdrop, the current investigation was undertaken using APM1 and APHO1 silkworm breeds, to evaluate the effect of HS on embryonic development of *B. mori* eggs which decides the post embryonic development, protein expression and commercial traits. Results showed that embryos exposed to HS during embryonic stages acquired varied thermos- tolerance.

MATERIALS AND METHODS

The newly emerged moths of silkworm, *B. mori* strains, APM1 and APHO1 (both for heat shock treatment and for control batch) were drawn from the germplasm of the APSSRDI for the preparation of loose eggs towards present investigation. Fertile and unfertilized eggs were separated by brine treatment. The fertile eggs thus collected were incubated in a natural room environment without using any instruments to maintain optimum temperature and relative humidity.

HS incubation was performed from day-2 after oviposition till blue egg stage at 24 hr intervals. About 40 eggs in each replication were exposed to HS temperatures of 35, 40 and 45°C in the water bath for 2 h followed by 2 h recovery period at room temperature. A control batch in three replications were maintained at room temperature that ranged from 28 to 31°C with relative humidity of 56 – 67 per cent. After heat shock all the eggs including control batch were preserved in the plastic tray that was covered with paraffin paper and wet foam pads until blue egg stage. They were transferred to the black box for 24 h, and the next day all the eggs were exposed to light for hatching.

The eggs of HS treatment along with control batches were brushed separately in triplicates and reared until spinning under natural environmental conditions with temperature fluctuations from 26 to 31°C and relative humidity of 56 to 84 per cent (Vasudha *et al.,* 2006).

Analysis of biochemical, biological, and commercial traits: Sensitivity to varied HS temperatures was measured based on per cent hatching as an index of embryonic development. Protein was extracted and quantified by using biophotometer (Lowry *et al*., 1951). The SDS-PAGE was performed as stated by Weber and Osborn (1969) with necessary changes as suggested by Vasudha *et al*. (2006). Carbohydrate (glycogen) was estimated by Anthrone method following the earlier protocol (Sadasivan *et al.,* 2009). Effective rate of rearing (ERR), larval weight, cocoon weight, pupal weight, shell weight and shell ratio were recorded for analyzing the commercial traits.

Data were statistically analyzed using statistical software package (IBM-SPSS) version 23. Kolmogorov – Smirnov test was applied to check the data distribution. Variables were normally distributed. Two-way ANOVA was utilized to study the effect of time, temperature, and their interaction on the studied variables. Least significant difference (LSD, at $p < 0.05$) was used to check for significant differences among the studied groups. Multiple linear regression analysis was utilized to illustrate the relationship between time, temperature, species, and the studied parameters.

RESULTS

Changes in the protein profile due to heat shock at the embryonic stage of *B. mori:*

Quantitative changes: The APM1 and APHO1 eggs exhibited significant variations in the protein content after HS treatment. In APM1, high protein content was seen in day - 6 eggs HS at 35 °C, while in control it was low. Comparatively, the protein content found increased in day - 2, -3, and -4 eggs HS at 35 °C; day - 2, -4, and -7 eggs HS at 40 °C; day - 2, -3, -4 and -6 eggs HS at 45 °C, whereas it was declined in other days of respective eggs HS in 35, 40 and 45 °C (Fig. 1).

The bivoltine silkworm strain APHO1 was also responding to HS at 35 °C with increased protein content at day - 4, -3 and -8 respectively compared to control. The protein content in the eggs exposed to HS at 40 and 45°C was found to increase on day -3, -4, -5, -7 and -8 and day -4, -6 and -8 respectively. Whereas the protein content decreased in other batches of eggs HS at 35, 40 and 45°C compared to control (Fig. 1).

Qualitative changes: Further, to check if there are qualitative changes, the protein isolated from the embryos of the breeds APM1 and APHO1 under control and high temperature conditions was separated on SDS-PAGE gel. Protein profile of day - 2 showed a total of 18-19 protein bands in APM1 and APHO1 whereas on day - 6, 16-17 bands are seen. The 4 significant protein bands referable to vitellin-L, egg specific protein, vitellin-H, and 30kDa protein are similar between the

strains but differ in intensity, higher intensity was seen in 45°C treated embryos. A couple of extra protein groups were seen on day - 6 which may be a mark of the improvement of new organs in the egg. A protein band of 205kDa was found overexpressed in both the strains on day 2, but 18 kDa and 19 kDa proteins were found degrading in both the strains on day - 6 treated embryos. In addition, 90kDa and 70 kDa heat shock proteins were also found more expressed in APM1 in comparison to APHO1 but the rate of expression intensity differed (Fig. 2). Results showed several qualitative changes in embryos after HS.

Changes in the glycogen content due to heat shock at embryonic stage of *B. mori.:*

Glycogen content in embryos of APM1 and APHO1 found declined from day - 3 till hatching both in control and HS batches. In APM1, the highest glycogen content was noticed in day - 3 old embryos which decreased until hatching in control batch of eggs. However, the content of glycogen was more on day - 3 eggs of all the HS induced eggs, but their quantity declined as the embryonic development proceeded. Interestingly, the quantum of glycogen was found variable either increased or decreased in the different developmental stages of embryos exposed to different HS temperatures. Accordingly, the glycogen content found increased in all the HS induced eggs at 35, 40 and 45°C in day 5 old embryos compared to control (Fig. 3).

The glycogen content in APHO1 was also found to be increased on day - 7 eggs subjected to HS at 35, 40 and 45°C compared to control. Whereas in HS induced eggs of different age group the glycogen content was less compared to control.

Determination of sensitivity to heat shock at the embryonic stage of *B. mori.:*

The hatching of eggs of polyvoltine-APM1 and bivoltine-APHO1 silkworm strains was determined as it is an important index to determine the sensitivity towards HS treatment. The results state that, the hatching of eggs of polyvoltine-APM1 and bivoltine-APHO1 silkworm strains increased on day - 5, -6 and -7 at 35°C HS but it declined at 40°C HS on all the days.

Concomitantly, the bivoltine silkworm strain APHO1 also showed positive response to HS at 35°C with increased hatching against control. In addition, the eggs exposed to HS at 40°C HS was also increased hatching on day - 3 and -6, respectively, while other days eggs were shown reduced hatching compared to control (Fig. 4), which is statistically significant at $P< 0.01$. More interestingly, delayed hatching was recorded from the eggs subjected for HS at 45°C and quite a few larvae hatched later recovered well.

Changes in the larval growth due to heat shock at embryonic stage:

The larval growth as influenced by HS at different embryonic stages was measured based on their weight from day - 2 till blue egg stage. Interestingly, increased weight was observed not only in the larvae derived from day - 6 embryo HS at 35°C but also at 40°C compared to control batch which is significant at $p < 0.01$. Correspondingly, larvae survived after HS of day - 2 till day - 6.

In case of APHO1, an average weight of the larvae obtained from the embryos HS at 35°C. Increase in weight was recorded in the larvae derived from day - 2 embryo HS at 35°C compared to control which is significant at $P < 0.01$. The larvae which survived after HS of embryos on day - 3 at 45°C exhibited better growth compared to control (Figs. 5, 6).

Changes in the Effective Rate of Rearing (ERR) due to Heat shock at embryonic stage:

The ERR denotes for the larvae succeeding to spin cocoons. Eventually, the silkworm larvae derived from HS induced eggs of APM1 and APHO1 were reared under natural environmental conditions prevailed in the rearing house. Interestingly, 2 to 34 per cent of improvement in ERR was recorded in the population derived from different age group of APM1 eggs HS at 35 40 and 45^0C with highest ERR 34.39 per cent in day - 5 eggs HS at 40^0C (Fig. 7).

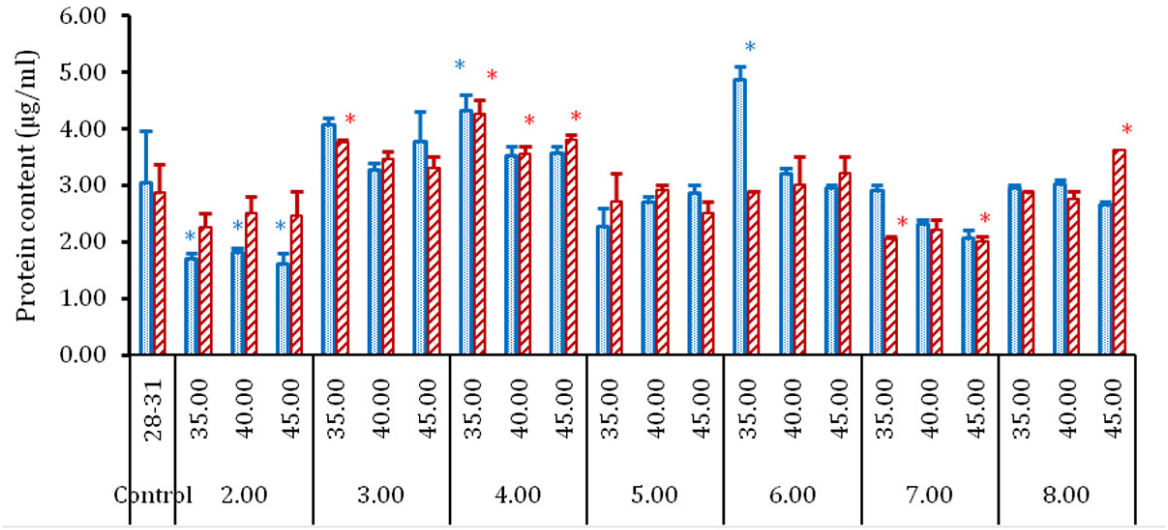

Fig. 1 The protein content (µg/ml) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (day - 1, 2, 3, 4,5, 6, 7 and 8) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

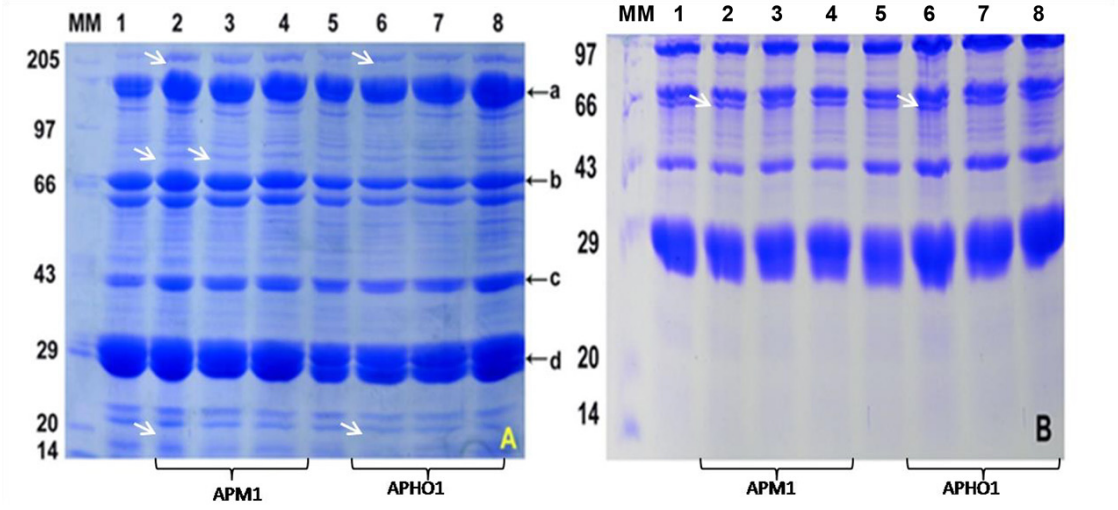

Fig. 2 Differential expression of protein profile of silkworm egg of APM1(Lanes 1 to 4: Lane 1 – Control, Lane 2 – 35°C, Lane 3 – 40°C , Lane 4 - 45°C) and APHO1(Lanes 5 to 8: Lane 5 – Control, Lane 6 – 35°C, Lane 7 – 40°C, Lane 8 - 45°C). Gel A-Day2 and B-Day6 -Arrows show the difference in expression of proteins compared to control. Numbers shown at the top of the lanes represent different days.
Band a – vitellin-H; Band b-Egg specific protein; Band c-vitellin-L; Band d-30kDa proteins

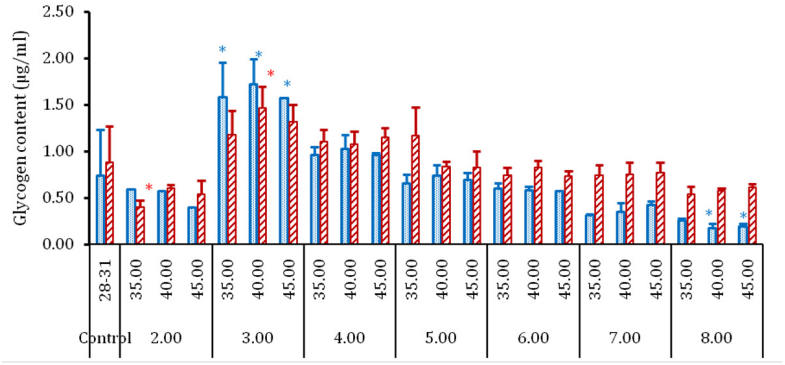

Fig 3. The glycogen content (µg/ml) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (day - 1, 2, 3, 4,5, 6, 7 and 8) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

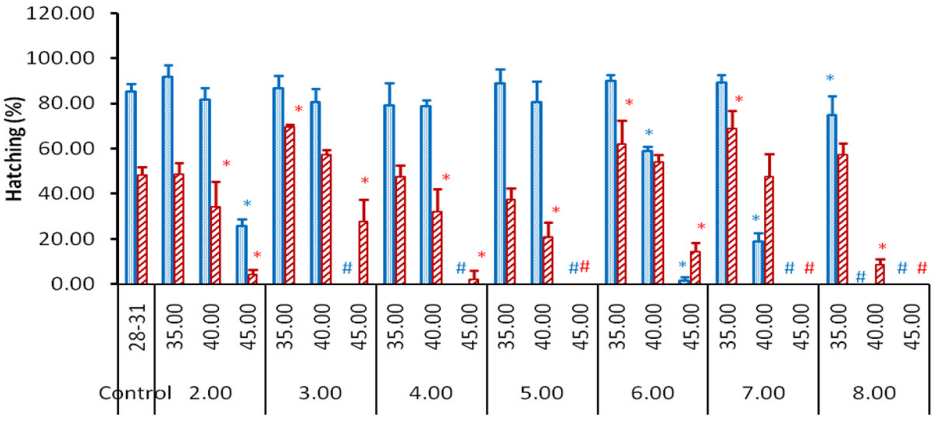

Fig 4. The hatching (%) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (day - 1, 2, 3, 4, 5, 6, 7 and 8). (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. #: represents mortality. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

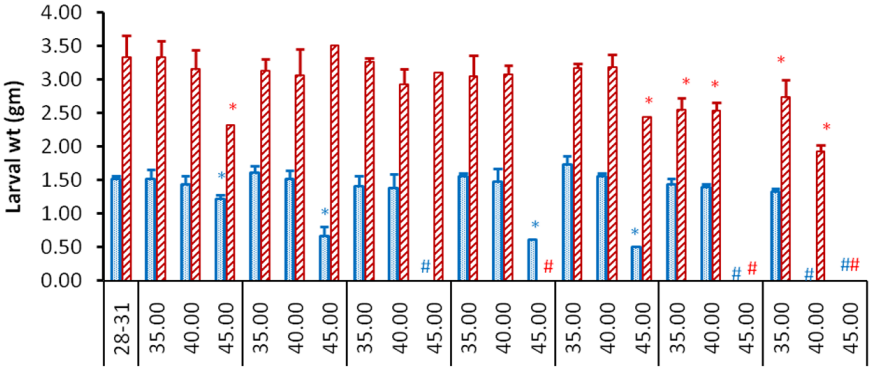

Fig 5. The larval weight (g) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (1, 2, 3, 4, 5, 6, 7 and 8 days). Blue bar represents APM1 and Red bar represents APHO1 breed. Each bar represents mean ± standard error of mean. #: represents mortality. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

ERR was severely affected due to HS at embryonic stage of APHO1, wherein good cocoons although produced but comparatively lower yield than control (81.63%). More interestingly, larvae derived from the eggs of APM1 and APHO1 HS at 45^0C recovered well and spun good cocoons and their ERR although appears to be low in comparison with control but found quite significant.

Cocoon weight: Weight of the cocoon spun by the APM1 silkworm larvae derived from HS induced embryos on day - 2 and - 4 at 35, 40 and 45°C was found significantly increased compared to control. The highest weight of the cocoon was observed in day - 4 embryo HS at 35°C. An average weight of cocoon was observed that corresponds to day - 2, 3, 4, 5, 6, 7 and 8 embryos HS at 40°C respectively that significant at P <0.01. Comparatively, the larvae survived upon HS of eggs at 45°C spun the cocoon which showed increased weight against control. (Fig. 8)

Concomitantly, the bivoltine silkworm strain APHO1 was also showed positive response to HS at 35°C with increased cocoon weight on day - 3 embryo subjected to HS against control More interestingly, larvae derived from the day - 3 eggs HS at 45^0C recovered well and spun good cocoon.

Shell Weight: The cocoon shell weight also unequivocally affected as that of cocoon weight due to fluctuated environmental conditions in the rearing house. As a result, shell weight in control was 0.11g and 0.31g in APM1and APHO1 respectively. However, larvae survived after HS at 45^0C during embryonic stage were spun the cocoons with an average shell weight in APM1and APHO1 by having marginal difference against their respective control batches (Fig. 9).

Cocoon shell ratio: The cocoon shell ratio was also correspondingly affected as that of cocoon and shell weight due to HS at embryonic stage both in APM1 and APHO1 silkworm strains. The cocoon

Table 1. Multiple linear regression (stepwise); explanatory variables entered were time (days), temperature (°C) and breed type to explain variations in the hatching percentage, larval weight, ERR, protein and glycogen levels

Dependent variable	Explanatory variables	â-coefficient			$t_{statistic}$	P-value
		Unstandardized	SE	Standardized		
Protein level	Temperature	-0.025	0.017	-0.133	-1.486	0.140
	Time	-0.015	0.034	-0.039	-0.438	0.662
	breed	-0.012	0.135	-0.008	-0.087	0.931
Glycogen content	Temperature	0.000	0.007	-0.005	-0.060	0.952
	Time	-0.092	0.015	**-0.470**	-6.039	0.000
	breed	0.145	0.061	0.185	2.382	0.019
Hatching (%)	**Temperature**	-6.53	0.384	**-0.796**	-16.99	0.000
	breed	-15.85	3.14	-0.236	-5.05	0.000
	Time	-3.51	0.784	-0.209	-4.47	0.000
Larval weight (gm)	breed	1.43	0.112	**0.629**	12.76	0.000
	Temperature	-0.124	0.014	-0.447	-9.06	0.000
	Time	-0.188	0.028	-0.330	-6.69	0.000
ERR	**Temperature**	-4.23	0.494	**-0.571**	-8.56	0.000
	Time	-5.41	1.00	-0.357	-5.36	0.000
	breed	7.023	4.00	0.116	1.754	0.082

â: represents regression coefficient with the negative signs indicating inverse association between the studied variables

shell ratio highest in the population derived from the eggs of APM1 HS at 35, 40 and 45°C respectively, which is significant at P <0.01. Concomitantly, no improvement was recorded in 35 and 40°C but at 45°C the increased shell ratio was recorded against control in the population derived from the day - 6 embryos of APHO1, which is significant at P <0.01. Obviously, induction of HS at 35, 40 and 45°C during different days of embryonic development affected the cocoon shell ratio both in APM1 and APHO1 of *B. mori* (Fig.10).

Pupal weight: Weight of the pupa, as an index of its growth, showed highest weight in the population derived the embryos of HS at 35 and 40°C on day - 7 and day - 2 respectively, which is significant at P <0.01. More importantly, the APM1 and APHO1 silkworm embryo HS at 45°C on day - 2 and - 3 were also exhibited marginal difference in weight by their respective controls (Fig. 11)

Tables 1 and 2 clarify the results of stepwise multiple linear regression analysis applied to identify the explanatory variables associated with the changes in the studied dependent variables. The strongest predictor of cocoon weight, shell ratio, and pupal weight was the temperature. There was a negative association between temperature and the change in these variables. However, each change in shell weight was highly associated with breed type. Thus, the shell weight in APHO1 was 0.132 higher than in APM1.There were strong negative associations of temperature with both the percentage of hatching and ERR. Moreover, the larval weight was higher in APHO1 than APM1 by 1.43. On the other hand, no association was observed between the protein levels and any of the explanatory variables. Time was the strongest predictor for change in the glycogen content. As each increase in time by one day, there was a decrease by 0.092 in the glycogen content.

DISCUSSION

The sericulture industry has contributed significantly to the economic development of many countries due to the commercial importance of silk in the Textile World and easy to rear silkworms under domestication. Due to continuous domestication, *B. mori* larvae lost their tolerance to high temperature and resistance to diseases. Thus, heavy loss of cocoon crop has been experienced by the farmers during critical climatic conditions. In view of this commercial importance, as well as to evaluate thermotolerance based on biochemical constituents that associated with it.

We have selected popular parental silkworm strains APM1 and APHO1 to examine the impact of HS during embryonic development, which determine the post embryonic development and cocoon characteristics for the first time as most of the studies were confined to either egg and/or larval stages (Manjunatha *et al.,* 2010). Since, it is well known that temperature plays a significant role on growth and productivity of silkworm (Sujatha *et al.,* 2001; Howrelia, 2011) a correlation study was carried out between APM1 and APHO1 in relation to heat shock response of embryos, growth and development of silkworm larvae and cocoon characteristics. It is evident from the earlier studies also that induction of HS for an hour at temperature ranging from 35 to 45°C and above has great impact on embryonic development (Manjunatha *et al*., 2005) in terms of hatching but its influence on post embryonic stages was not studied. Further, early embryonic stages until end of blastokinesis were found to be sensitive -to HS compared to late embryonic stages. In support of this, Coulon and Mathelin (1991) also opined that the initiation phase (from 72 to 120 h) is more sensitive to stress than deep phase.

Towards this, while eggs of APM1 and APHO1 subjected for HS at 35, 40 and 45°C for 2 h with 2 h recovery from day - 2 to blue egg stage (day - 8 or 9) at 24h intervals exhibited changes in the per cent of hatching revealing 45°C as lethal temperature for both the silkworm strains. The highest of 91.6 per cent hatching while noticed in day - 2 eggs of APM1 subjected for HS at 35°C, APHO1 was also showed positive response at same HS temperature with an improvement of 43.4 per cent against control (48.33%) denoting that exposure of silkworm eggs to mild HS temperature of 35°C induces highest hatching than control. Interestingly, weight of larvae derived from the eggs

Fig. 6 Effect of heat shock during embryonic development in relation to larval growth of *Bombyx mori* strain
Left side - (i) APM1 and right side - (ii) APHO1: A-Control, B-35, C-40 and D-45ºC

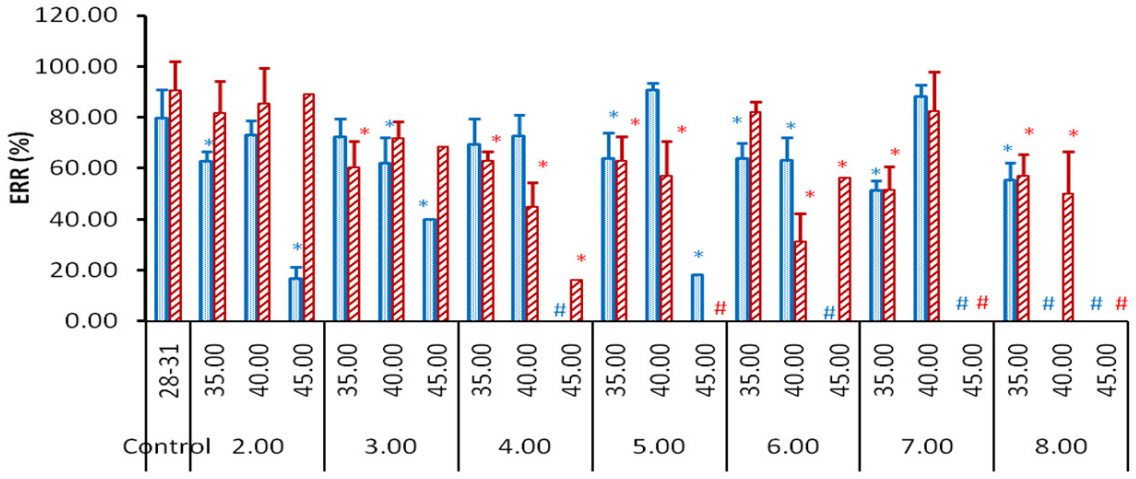

Fig. 7 ERR (%) of two species APM1 and APHO1, under different temperatures (35, 40 and 45ºC) and after different time intervals (day - 1, 2, 3, 4, 5, 6, 7 and 8) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. #: represents mortality. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

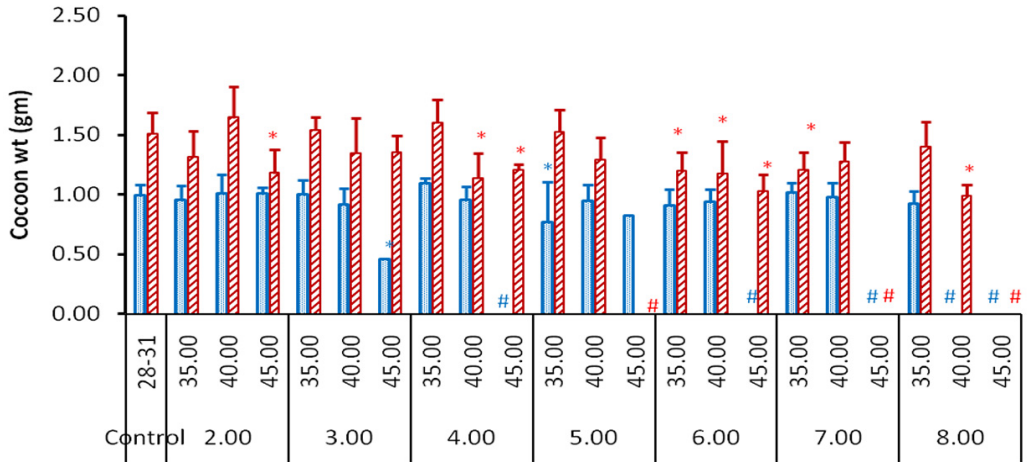

Fig. 8 Cocoon weight (g) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (1, 2, 3, 4, 5, 6, 7 and 8 days) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. #: represents mortality. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

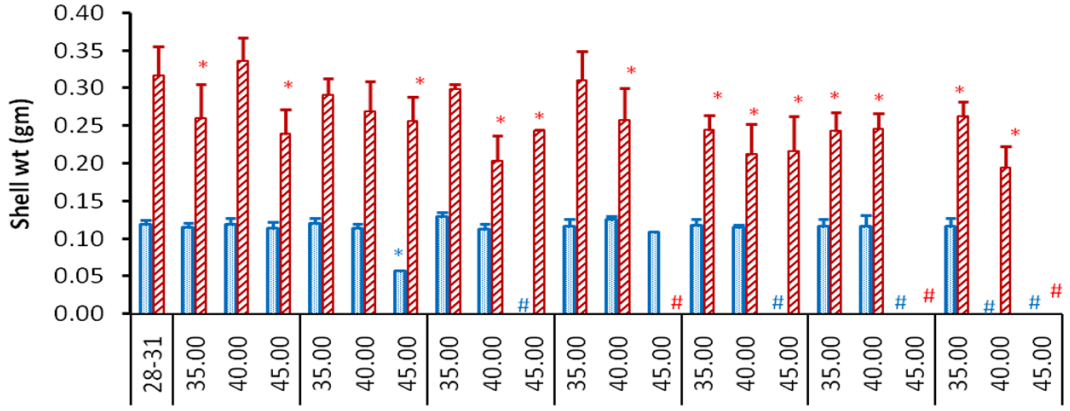

Fig. 9 The shell weight (g) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (day - 1, 2, 3, 4, 5, 6, 7 and 8) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. #: represents mortality. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

of APM1 HS at 35 and 40°C showed better growth with 22.63 and 12.31 than APHO1 against their respective controls. Whereas larval weight declined in HS induced embryos at 45°C compared to control indicating the lethal effect of high temperature on embryonic development that inturn produced either dead or weak larvae. Notably, some of the larvae hatched out after HS at 45°C although showed slow growth by taking longer duration but recovered well and grow either equal or much better than control exhibiting their acquired thermotolerance and emerged as healthy moth. Thus, the phenomenon of heat shock/thermotolerance that expressed in the survivals upon HS could be a potent breeding material for development of silkworm strains with acquired thermotolerance for tropics.

In APM1, 4.8 µg ml^{-1} (high) of protein content was seen in day - 6 eggs HS at 35°C, which is found to be decline in other days of eggs HS at 35, 40 and

Table 2. Multiple linear regression (stepwise); explanatory variables entered were time (days), temperature (°C) and breed type to explain variations in the pupal weight, shell ratio, shell weight, and cocoon weight

Dependent variable	Explanatory variables	ß-coefficient			$t_{statisti}$	P-value
		Unstandardized	SE	Standardized		
Cocoon weight (gm)	Temperature	-0.068	0.005	-0.538	-13.28	0.000
	breed	0.411	0.042	0.401	9.89	0.000
	Time	-0.097	0.010	-0.378	-9.32	0.000
Shell weight (gm)	breed	0.132	0.007	0.643	18.32	0.000
	Temperature	-0.011	0.001	-0.428	-12.19	0.000
	Time	-0.016	0.002	-0.311	-8.849	0.000
Shell ratio (%)	Temperature	-1.082	0.089	-0.554	-12.11	0.000
	breed	4.826	0.730	0.302	6.61	0.000
	time	-1.163	0.182	-0.291	-6.37	0.000
Pupal weight (gm)	Temperature	-0.056	0.009	-0.536	-12.83	0.000
	Time	-0.084	0.004	-0.393	-9.41	0.000
	breed	0.299	0.036	0.352	8.42	0.000

ß:represents regression coefficient with the negative signs indicating inverse association between the studied variables

45 °C than that of control (3.9 µg ml^{-1}). Similarly, the bivoltine silkworm strain APHO1 was also responding to HS at 35, 40 and 45°C with increased protein content ranging from 2.05 (day-7) to 4.25µg ml^{-1} (day-4), 2.25 (day-7) to 3.55µg/ml (day-4) and 2.05 (day-7) to 3.80 µg ml^{-1} (day-4) compared to control (2.7 µg ml^{-1}) indicating varied effect of HS on different embryo stages of *B. mori.* On the other hand, the protein content was also found to decline up to -29.31 (45°C) and -16.6 (35°C) in APM1 and APHO1 indicating the sensitivity to HS that facilitates unfolding of proteins in the embryos of APM1 and APHO1.

Furthermore, the increased quantity of protein observed in the present study is due to synthesis or over expression of 205 kDa, 90 kDa and 70 kDa heat shock proteins both in the APM1 and APHO1 HS at 35, 40 and 45°C compared to control. The expression of HSPs might involve in protecting the silkworm embryos from the fluctuated environmental condition prevailed during incubation period that resulted in increased percent of hatching compared to control as has been observed in the HS induced larvae (Manjunatha *et al*., 2010; Shabir and Manjunatha 2010; Vasuhdha *et al.*, 2006; Shou-Min Fang *et al.*, 2021).

Interestingly, the glycogen content was found more on day - 3 compared to day - 2 in the embryos of APM1 and APHO1 eventually declined as the embryonic development proceeds until hatching indicating their utilization as a source of energy. This was also observed in the embryos of NB4D2 and PM silkworm strains (Manjunatha *et al*., 2008), since carbohydrates and proteins play a vital role in the development, morphogenesis and intermediary metabolic pathway of insects (Wyatt *et al*., 1978). However, the glycogen content found variable in all the HS induced eggs while it increased in 35°C (0.65 mg ml^{-1}), 40°C (0.73 mg ml^{-1}) and 45°C (0.69 mg ml^{-1}) HS induced day-5 eggs of PM compared to control (0.63 mg ml^{-1}). The biochemical process in increased content of glycogen in the HS induced embryos is unclear and offers detailed investigation. Meanwhile, the glycogen content was found to decrease compared to control, which can be attributed that the HS induced embryos might have utilized more energy to overcome the thermal stress and might be insufficient to facilitate normal

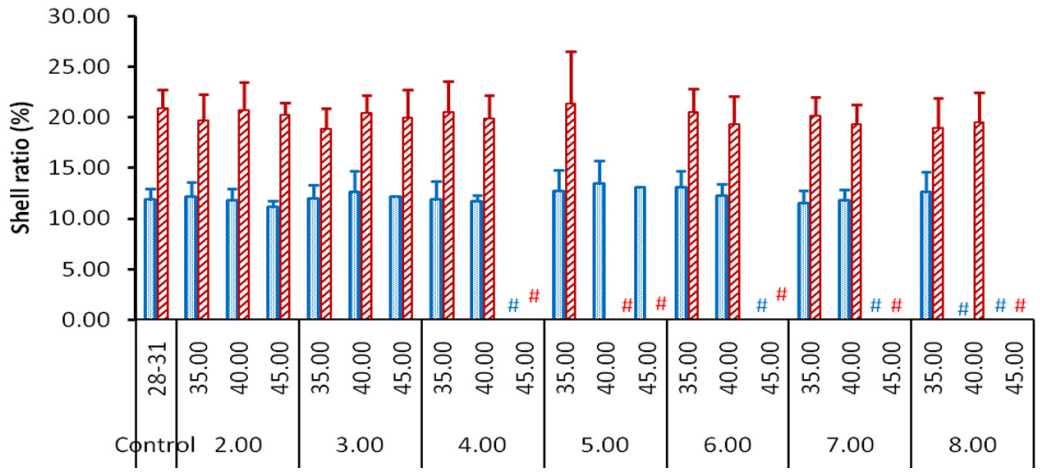

Fig. 10 The shell ratio (%) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (day - 1, 2, 3, 4, 5, 6, 7 and 8) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. #: represents mortality. *: represent significant ($p<0.05$) difference, as compared to the corresponding control

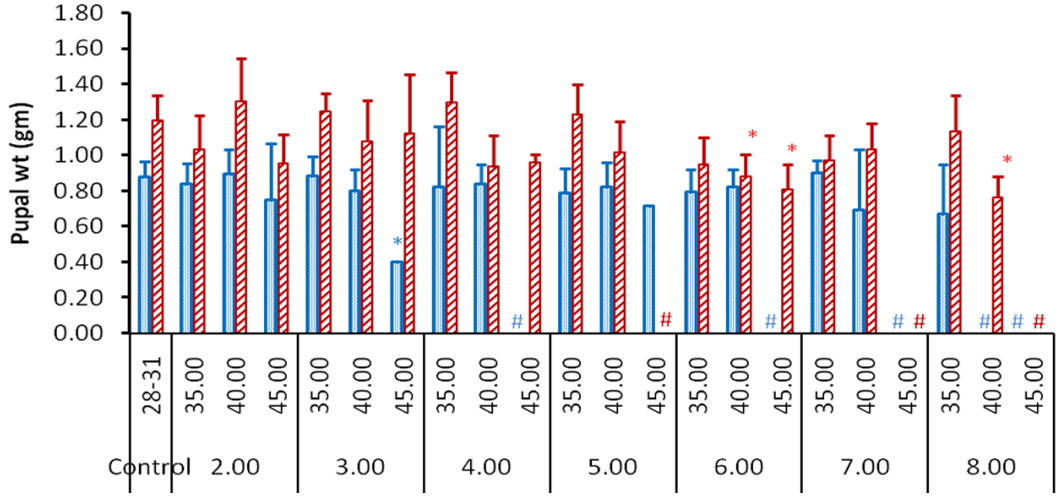

Fig. 11 The pupal weight (g) of two species APM1 and APHO1, under different temperatures (35, 40 and 45°C) and after different time intervals (day - 1, 2, 3, 4,5, 6, 7 and 8) (Blue bar represents APM1 and Red bar represents APHO1 breed). Each bar represents mean ± standard error of mean. #: represents mortality *: represent significant ($p<0.05$) difference, as compared to the corresponding control

hatching that resulted in embryonic death within the egg or at the time hatching or after hatching or embryos develop as weak larvae. Thus, it is suggested that since the silkworm embryos are highly sensitive to fluctuating environmental conditions they should be preserved under optimum conditions or even 2 h of thermal stress above threshold cause embryonic death or weak larvae which intern might affect other post embryonic developments and cocoon traits.

Based on these findings we conclude that eggs of the APM1 and APHO1 when exposed to critical temperature even for 2 h affect the hatching, altered

the protein and glycogen content, larval weight, ERR, cocoon weight, shell weight and pupal weight. However, mild HS either at 35 or 40°C at specific stage of the egg might facilitate the embryo to exhibit acquired tolerance to fluctuated environmental conditions and produce good quality cocoons. In addition, induction of HS at 45°C might produce thermotolerant silkworm strains or which can be used as potent parental breeding materials for development of thermotolerant silkworm strains for commercial exploitation in tropics.

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Diversity and distribution of true flies (Diptera) of Kuldiha Wildlife Sanctuary, Odisha, India: Functional roles based on ecological guilds

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ABSTRACT: Proper understanding of ecological dynamics of faunal components, whether it is a large mammal or a tiny insect of any ecosystem including forests, plays an important role in the eco-management of any eco-zone. Dipteran insects constituting a major faunal group among the entomo-diversity of any forest ecosystem portray significant functional roles in determining the stability in the ecosystem functioning of the respective ecosystem. The present paper has attempted to document the diversity of dipteran insects inhabiting a tropical deciduous forest of the extended part of Deccan Biogeographic Zone in the eastern part of India, the Kuldiha Wildlife Sanctuary alongside indicating its habitat preference and distribution patterns. A total of 34 species under 19 families of the order Diptera were recorded from different habitats of the studied forest areas, of which three species are considered new reports from the state of Odisha, India. Out of the three selected eco-zones, the deep forest area having a higher density of sal trees (*Shorea robusta*) revealed less species richness but high relative abundance, whereas the barren grazing land demonstrated higher species richness with low relative abundance. The eco-zone with wetlands and associated vegetation have shown moderate species richness and diversity of dipteran insects. Three contrasting seasons (pre-monsoon, monsoon, and post-monsoon) of this region have also demonstrated different patterns of diversity and density of this group of insects which have been segregated into several feeding guilds in tune with the seasonal availability of food resources.

KEYWORDS: Feeding guilds, habitat preference, seasonality, new records

INTRODUCTION

Kuldiha Wildlife Sanctuary (KWLS), covering an area of approximately 272.75 km^2, is one of the stable and well-protected eco-zones of India. This area was declared as a sanctuary in the year of 1984 under the territorial jurisdiction of Baripada division, comprising Tenda reserve forest, Kuldiha reserve forest, and Devgiri reserve forest intermingling with other adjoining forest land of Nilagiri sub-division extending up to Simlipal National Park, Odisha, India. Being a well-protected tropical deciduous forest, this KWLS provides shelter to a wide variety of fauna, of which insects represent the most diverse faunal group. True flies (Insecta: Diptera), being one of the major

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and the least explored faunal group of the insect community, plays a series of ecological functions within an ecosystem.

However, the faunal group of this well-protected and ecologically managed KWLS is explored by only a very few studies on mammals (Debata *et al.*, 2013; Debata and Swain, 2017, 2018a; Mohapatra *et al.*, 2013), herpetofauna (Rout *et al.*, 2016a), birds (Das and Debata, 2018; Ghosh *et al.*, 2018) and flying squirrel (Ghosh *et al.*, 2023). Overall diversity of mammals and avifauna in KWLS was earlier reported by Murmu *et al.* (2013). Some scanty studies also revealed the possibilities of use of medicinal plants among the local people (Saravanan *et al.*, 2017, 2018). Unfortunately, very few attempts have been made to explore the insects and their ecology in KWLS, except for some very recent ones by Parui *et al.* (2015) on ants, Paria *et al.* (2018) on butterflies, Debata and Swain (2018b) on Odonata, and Ganguly *et al.* (2022) on termites.

The functional diversity forming the necessary linkages among different individuals within a species population and different species within a biotic community can counterbalance the damages caused by the loss of another species in ecosystem processes and patterns and thereby ensure the ecological stability of the ecosystem. In a biotic community, different functionally similar (belonging to the same trophic level) and dissimilar species (belonging to different trophic levels) having access to the same pool of resources can compensate for the loss of one species and prohibit any reduction in the use of that resource pool because of the increasing populations of other species that are present in the same ecosystem, which will accordingly simply increase their use of that same resource. The term guild refers to the differential behavioural patterns in respect of their differential forms of feeding strategies and thereby encompasses groups of potentially competing species, not cooperating ones. The concepts of guilds rest on functional manifestations of a group of species that exploit the same class of environmental resources in a similar way depending on their phylogeny and resource requirements (Chakraborty, 2020).

Several studies on the dipteran community, including other insect groups, have been conducted in different parts of Odisha (Parui and Datta, 1987; Nandi, 1977; Joseph and Parui, 1987; Veer *et al.*, 2002; Srinivasan and Jambulingam, 2013; Shety *et al.*, 2018), but no such works have so far been reported of dipteran insects from KWLS. In such a context, the present paper has attempted to report the diversity and distributional patterns of dipteran insects from this KWLS, with ecological notes on their occurrence in different habitats, alongside highlighting the new records of three dipteran species from the state of Odisha, India.

MATERIALS AND METHODS

The KWLS (Fig. 1) is situated between $21^0 20'$ to $21^0 30'$ N; $86^0 30'$ to $86^0 45'$ E, merging with the Similipal Reserve Forest in Odisha. The vegetation of this forest fringe is characteristic of tropical deciduous forest and is dominated mostly by *Shorea robusta*, Sal trees (Champion and Seth, 1968). A recent study reveals KWLS as the home of 108 plant species, of which 38 species are trees, 38 species are shrubs, and 32 species are herbs (Rout et al. 2016b). Depending on several ecological characteristics like vegetation type, green coverage area, water bodies, and anthropogenic involvements, three categories of land cover types (LCT) were identified (Table 1, Fig. 2), which differentially support the dipteran faunal diversity of this geographical region. Two sample sites were selected in each LCT for observing and documenting the dipteran community. The climatic condition in this deciduous forest portrays three contrasting seasons, i.e., pre-monsoon (March–June), monsoon (July–October), and post-monsoon (November–February). The average temperature ranges from 8°C in post-monsoon to 42°C in pre-monsoon (Debata and Swain, 2018a).

The survey was conducted in this forest range once in each season (for three days) to explore the dipteran diversity of the area along with their ecological activities. The collection and observation of the dipteran species have been made under the canopies of forest vegetation, flowers, elephant dung, and also in the cowsheds of the local residents. Covering all kinds of habitat types during three

consecutive seasons (pre-monsoon, monsoon, and post-monsoon) between the years 2017 and 2018. The observations have been made by the simple transect walk method in each of the land cover types of the study area. The mosquito vectors were collected with the help of an aspirator. The collected insect materials were then identified up to species or genus level in the laboratory with the stereo-zoom microscope following the guidelines of standard literature (several volumes of Fauna of British India, Fauna of India, and other relevant literatures). The map of the study area has been made with the help of Google Earth.

The calculation of seasonal relative abundance of each representative dipteran families were done based on the observation of only adult dipteran insects by using the following formula:

$$Relative\ abundance = \frac{a_1}{\sum_{i-1}^{n} a_1} \times 100\ (\%)$$

Where, a_1 is the number of adults of species 1 on a particular site; $\sum_{i-1}^{n} a_1$ is the total numbers of adult observed of all species on a site.

To quantify the differences in diversity among different LCTs, some diversity indices have been deduced, like the Shannon-Weiner Index (*H'*), Simpson's Dominance Index (D), Margalef's Species Richness (R), and the Berger-Parker Index. All the statistical calculations were performed with the help of Microsoft Excel 2013 and PAST version 2.17. Cluster analysis was executed by using the paired group algorithm (UPGMA) and Jaccard Similarity measure, on the basis of presence and absence of insect species among the studied habitat types (LCTs).

RESULTS AND DISCUSSION

Diversity and ecological distribution of true flies according to habitat and resource utilization:

During the present study, a total of 328 individual dipteran insects representing 34 species under 19 families were recorded in and around Kuldiha Wildlife Sanctuary (KWLS) (Table 2). Among the different families under the order Diptera, Culicidae shares the most species (seven species, 20.59%), followed by Syrphidae (six species, 17.65%), Muscidae (three species, 8.82%), and both Stratiomyidae and Sarcophagidae (two species, 5.89%). The other insect orders possess only one representative (Table 2).

Depending on the different ecological characteristics of habitats, the study area was categorised into three LCTs. Among these, the barren grazing land mostly along the roadside (LCT-3) displayed maximum species richness with 28 species (82.35% of total dipteran species), followed by the wetland-associated vegetation (LCT-1) with 17 species (50% of total dipteran species) among 34 species. The deeper parts of the forest (LCT-2) revealed the least species richness with only 13 species, which shared about 38.24% of total dipteran species (Fig. 2). Depending on the present-absent matrix, the similarity and distance analysis (Jaccard cluster analysis) was computed according to the habitat preferences of dipteran species, which portrayed the occurrence of several groups of species (Fig. 3). It was observed that the deep forest area showed high dipteran abundance but less species diversity, mainly because of the ecological homogeneity of that studied eco-zone. At the same time, the wetland-associated vegetation shows the relatively lowest relative abundance (14.63%) but modest diversity of dipteran species because it offers higher habitat heterogeneity but less than roadside grasslands, which have maximum ecological heterogeneity coupled with higher diversity. In consideration of the relative abundance of dipteran insects in different habitats, LCT–3 (56.4%) is designated as the most abundant habitat, followed by LCT–2 (28.96%) and LCT–1 (14.63%) (Fig. 4)

The diversity indices were calculated for all three types of habitat observed in the study area (Table 3). The Shannon diversity index was found to be maximum in the LCT-3 (barren grazing land and roadside), followed by the LCT-1 (wetland-associated vegetation), and then the LCT-2 (deep forest). The species dominance and Berger-Parker index were observed to be highest in contrast to Margalef's species richness index, which was found lowest in the LCT-2 (deep forest). The species

Table 1. Different habitats or Land cover types (LCT) within Kuldiha Wild Life Sanctuary, Odisha, India, with ecological characteristics

No.	Category of LCT	Code	Characteristics
01	Wetland associated vegetation	LCT-1	There are several small waterbodies spread over the sanctuary including a large dam namely Rissia Dam. The vegetation near the waterbodies observed for the study.
02	Deep forest	LCT-2	Mostly dominated by Sal tree with other trees and shrubs.
03	Barren grazing land	LCT-3	The grazing lands of animals and the roadside area was categorized under same LCT for similar kind of anthropogenic and faunal interference. Some local people also resides within this area.

richness was calculated at its maximum in the LCT-3 (barren grazing land and roadside).

Moreover, based on the resource partitioning, all these flies tended to enjoy a particular ecological habitat with its own ecological distinctiveness, which was categorised as their feeding guild. Some of these flies are specialists, which depend on a single feeding guild, whereas several others are generalist species, which depend on more than one feeding guild to utilise resources. Twelve species were recorded from each of the two feeding guilds, i.e., flower visitors and saprophytes/decomposers. Eleven species were spotted as leaf or trunk inhabitants of different plant species across the KWLS, whereas six species were found as blood suckers or hematophagous (Table 2).

Seasonal distribution:

The seasonal distribution pattern of the dipteran insects in and around Kuldiha WLS clearly depicted the maximum abundance in the pre-monsoon, except for Culicidae and Sarcophagidae. The abundance of the culicids (mosquitoes) was very low in the pre-monsoon compared to the monsoon and post-monsoon. The sarcophagid flies were mainly abundant during the post-monsoon, while they were found in very minimal numbers during the monsoon. Another important observation was reported in the case of the dipteran species under the family Stratiomyidae (soldier flies), which were found only during the pre-monsoon in the study area (Fig. 5).

New records:

Following three dipteran species, among all the reported ones, were encountered for the first time in the state of Odisha, India: *Hermetia illucens* (Linnaeus, 1758), *Mimegralla albimana* (Doleschall, 1856), and *Dideopsis aegrota* (Fabricius, 1805), which belong to the families Stratiomyidae, Micropezidae, and Syrphidae, respectively.

Order Diptera; Sub order Brachycera

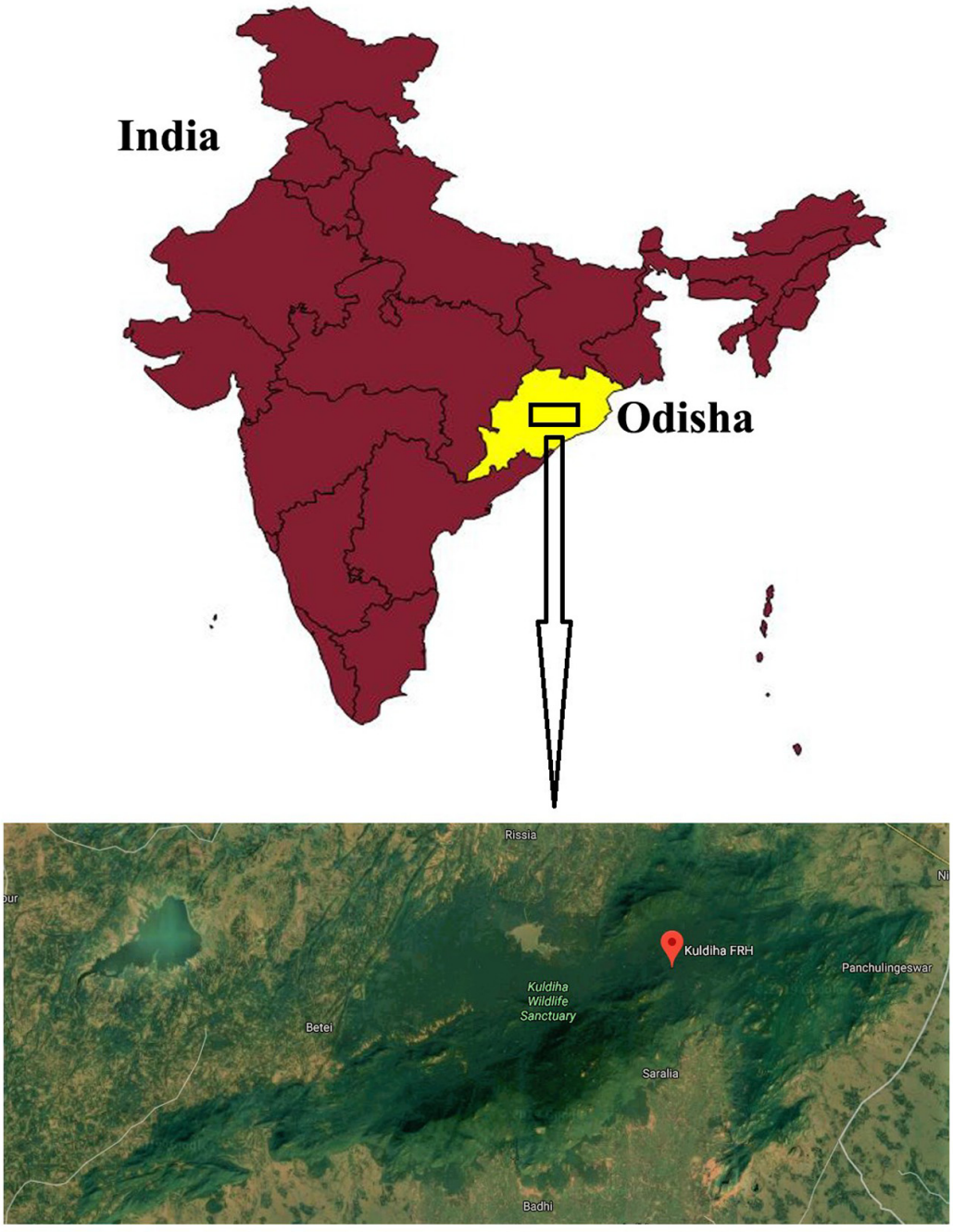
Family Stratiomyidae

1. *Hermetia illucens* (Linnaeus, 1758)*

1758. *Musca illucens* Linnaeus. *Systema naturae* Ed. 10, vol 1: 589

Type-locality: South America

Distribution: India: Assam, Odisha (Present record), Karnataka, Kerala, Maharashtra, Manipur, Punjab, Sikkim, Tamil Nadu, Uttar Pradesh, West Bengal. Elsewhere: Widespread in the World, nearly cosmopolitan.



Kuldiha Wildlife Sanctuary

Fig. 1: Map of the Study Area

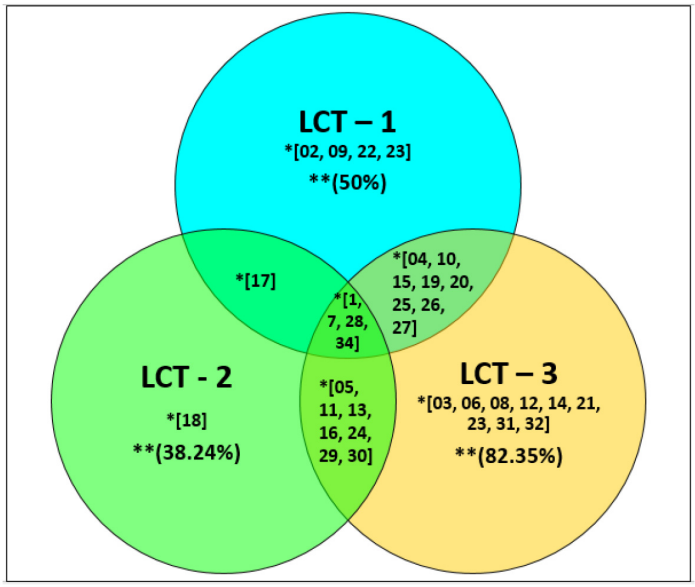


Fig. 2 Species distribution in different land cover types in and around KuldihaWLS [*numbers within third parentheses refer the species serial numbers mentioned in the Table no. 2 and
**percentages within the first parentheses refer the percentage of species composition in each LCTs among the total dipteran species]

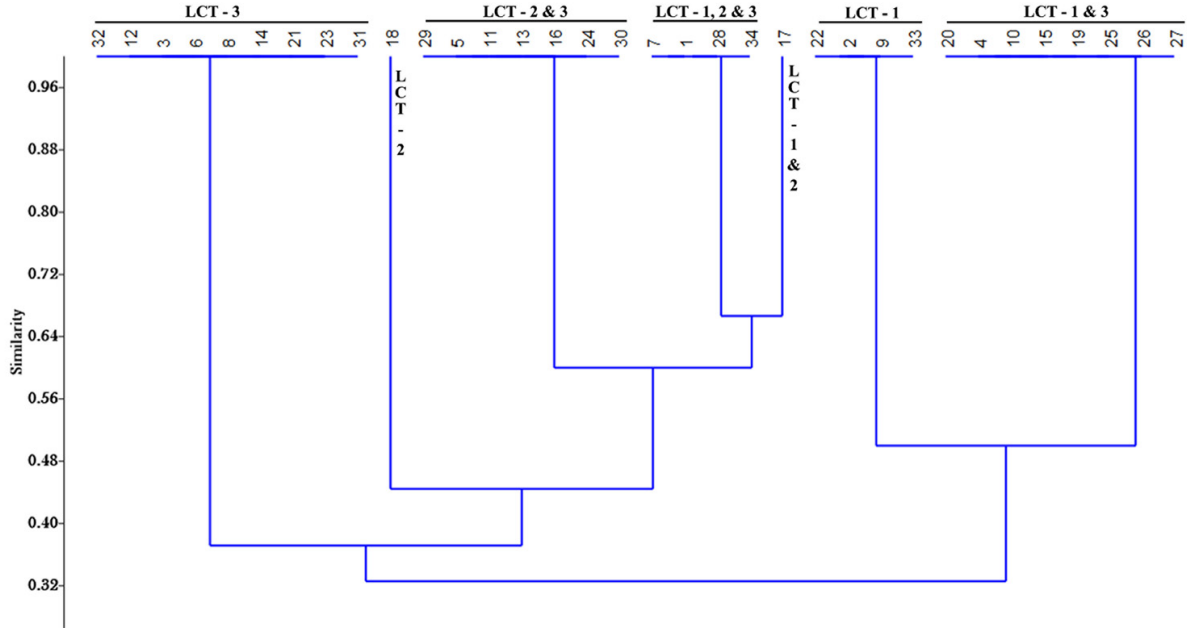


Fig. 3 Cluster analysis (Jaccardsimilarity measure with UPGMA method) of the dipteran species on the basis of their habitat preferences, where '1' indicates maximum similarity and '0' indicates no similarity [the numeric digits, i.e. 1-34 in the figure refers to the serial numbers of dipteran species in the Table no. 2]

Table 2. List of the dipteran species from different land cover types of Kuldiha Wild Life Santuary, Odisha, India ['+' denotes present and '-' denotes absent]

Sl. No.	Name of the species	LCT-1	LCT-2	LCT-3
	Family Tipulidae			
01	*Pselliophora* sp.	+	+	+
	Family Limoniidae			
02	*Limonia* sp.	+	-	-
	Family Culicidae			
03	*Anopheles (Cellia) culifacies* Giles, 1902	-	-	+
04	*Anopheles (Cellia) subpictus* Grassi, 1899	+	-	+
05	*Anopheles (Cellia) fluviatilis* James, 1902	-	+	+
06	*Aedes (Stegomyia) albopictus* (Skuse, 1895)	-	-	+
07	*Armigeres* sp.	+	+	+
08	*Culex* sp.	-	-	+
09	*Mansonia(Mansonoides) annulifera* Theobald, 1901	+	-	-
	Family Chironomidae			
10	*Chironomus* sp.	+	-	+
	Family Stratiomyidae			
11	*Sargus metallinus* Fabricius, 1805	-	+	+
12	*Hermetia illucens* (Linnaeus, 1758)*	-	-	+
	Family Tabanidae			
13	*Tabanus (Tabanus) rubidus* Wiedemann, 1821	-	+	+
	Family Muscidae			
14	*Musca (Musca) domestica* Linnaeus, 1758	-	-	+
15	*Neomyia* sp.	+		+
16	*Atherigona (Acritochaeta) orientalis* Schiner, 1868	-	+	+
	Family Micropezidae			
17	*Mimegralla albimana* (Doleschall, 1856)*	+	+	-

	Family Syrphidae			
18	*Dideopsis aegrota* (Fabricius, 1805)	-	+	-
19	*Eristalinus (Eristalinus) polychromata* (Brunetti, 1923)*	+	-	+
20	*Eristalinus (Eristalinus) arvorum* (Fabricius, 1787)	+	-	+
21	*Paragus (Paragus) serratus* (Fabricius, 1805)	-	-	+
22	*Ischiodon scutellaris* (Fabricius, 1805)	+	-	-
23	*Episyrphus (Episyrphus) balteatus* (De Geer, 1776)	-	-	+
	Family Calliphoridae			
24	*Chrysomya megacephala* (Fabricius, 1794)	-	+	+
	Family Rhiniidae			
25	*Idiella mandarina* (Wiedemann, 1830)	+	-	+
	Family Ulididae			
26	*Physiphora aenea* (Fabricius, 1794)	+	-	+
	Family Phoridae			
27	*Megaselia (Megaselia) scalaris* (Loew, 1866)	+	-	+
	Family Tephritidae			
28	*Bactrocera (Zeugodacus) cucurbitae* (Coquillett, 1899)	+	+	+
	Family Sarcophagidae			
29	*Sarcophaga (Liosarcophaga) dux* Thomson, 1869	-	+	+
30	*Sarcophaga (Liosarcophaga) brevicornis* Ho, 1934	-	+	+
	Family Drosophilidae			
31	*Drosophila* sp.	-	-	+
	Family Sepsidae			
32	*Sepsis* sp.	-	-	+
	Family Asilidae			
33	*Philodicus femoralis* Ricardo, 1921	+	-	-
	Family Dolichopodidae			
34	*Chrysosoma* sp.	+	+	+

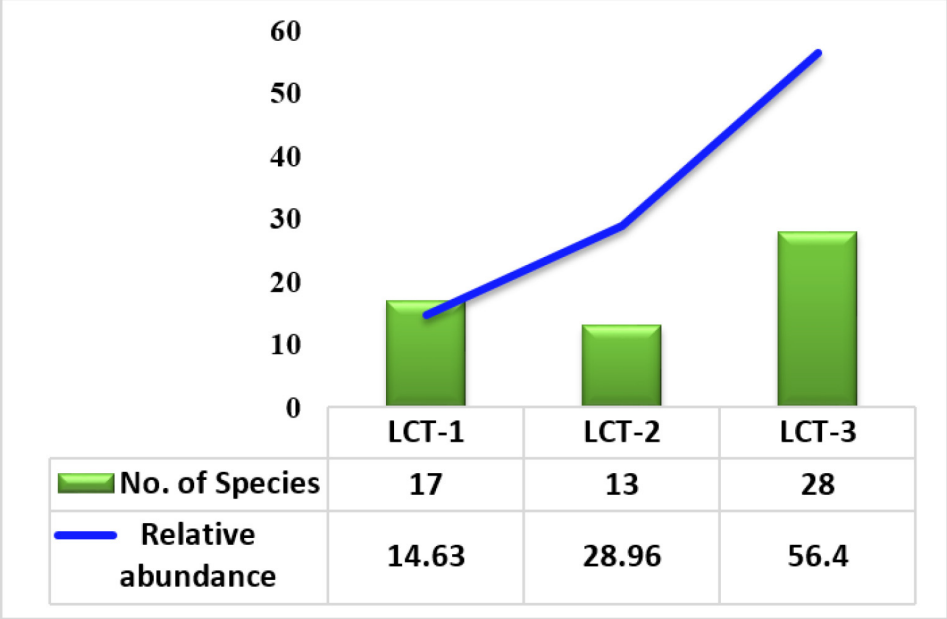


Fig. 4 Dipteran species diversity and relative abundance in different Land cover types (LCT-s) across the KuldihaWild Life sanctuary, Odisha, India

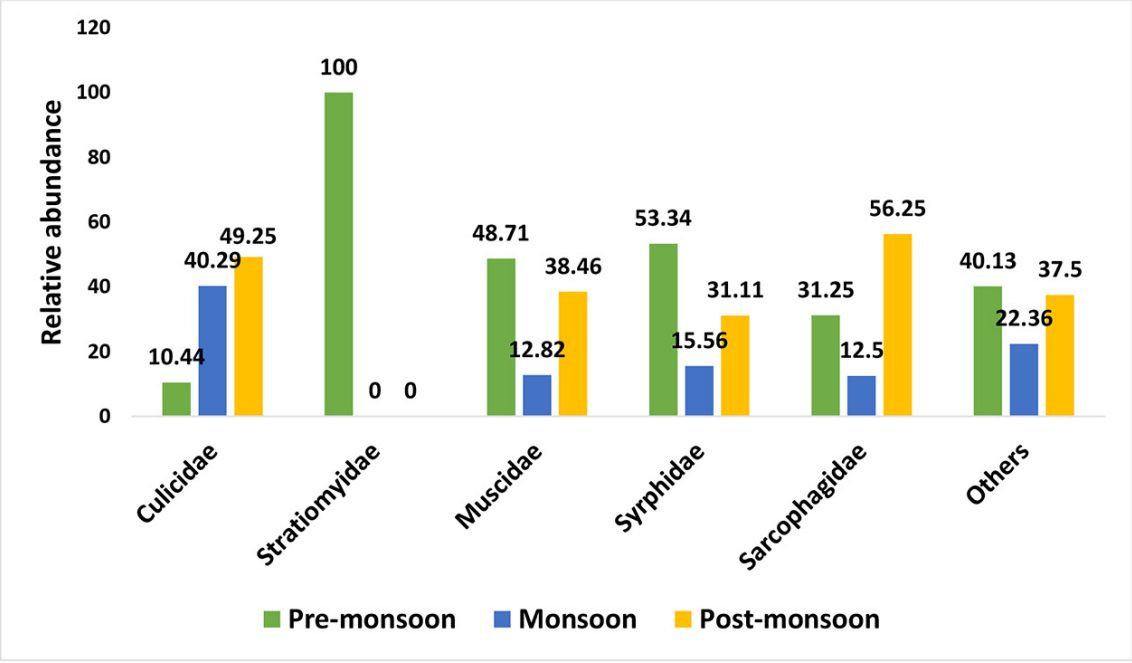


Fig. 5 Seasonal distribution of different dipteran species in and around KuldihaWild Life Sanctuary, Odisha, India

Remarks: Though it is native to the New World, but has been distributed throughout the World by anthropogenic commercial activities. The larvae of this species are utilized in processing of swine and hen manure, and food supplement of chicken which seems much cost effective (Diener *et al.,* 2011; Roy *et al.,* 2018). This species recorded here for the first time from the state of Odisha.

Family Micropezidae

2. *Mimegralla albimana* (Doleschall, 1856)*

1856. *Taenioptera albimana* Doleschall. *Eerste bijdrage tot dekennis der dipterologische fauna van Nederlandsch Indie. Natuurkd.Tijdschr. Ned.-Indie* 10: 413.

Type-locality: Indonesia: Java: Djokjakarta

Distribution: India: Assam, Mizoram, Odisha (Present record), Tripura, West Bengal. Elsewhere: Bangladesh, Java, Malaysia, Borneo, Japan, Belau, Guam, Micronesia, Myanmar, Northern Marianas, Papua New Guinea, Ryukus.

Remarks: Only eight species were reported of the family Micropezidae from India, of which 6 species were under the genus *Mimegralla* (Mitra et al. 2015c). This species also shows its distribution in other protected areas like, Sundarban Biosphere reserve (Mitra *et al.,* 2015a) and Bibhuti Bhusan Wildlife Sanctuary (Mitra *et al.,* 2015b). This is a new record for this state, Odisha.

Family Syrphidae

3. *Dideopsis aegrota* (Fabricius, 1805)

1805. *Eristalis aegrota* Fabricius, *Systema antliatorum secundum ordines, genera, species*. Xiv: 243.

Type-locality: India. Tamil Nadu: Tharangambadi

Distribution: India: Andaman and Nicobar islands, Arunachal Pradesh, Assam, Himachal Pradesh, Kerala, Karnataka, Meghalaya, Maharashtra, Madhya Pradesh, Odisha (Present record), Sikkim, Tamil Nadu, Uttarakhand and West Bengal. Elsewhere: Australia, Nepal, New Guinea.

Remarks: A widely distributed flower fly has been recorded for the first time from the state of Odisha.

The true flies are one of the most diverse insect orders, comprising about 1,59,294 described species (Pape *et al.,* 2011). However, the actual total number of extant fly species is many fold, most of which are still unexplored. The living dipteran species have been categorised into about 10,000 genera, 150 families, 22–32 super-families, 8–10 infra-orders, and 2 sub-orders (McAlpine and Wood 1989), and around 3100 fossil species have so far been described (Evenhuis 1994). Among the 150 families of the order Diptera, 85 families have been reported so far from India. These vast insect groups possess various types of ecological roles. Some of them are turning harmful to human and animal society, while some of them are beneficial to human society because of their functional contributions as pollinators, decomposers, bio-indicators, vectors, predators, and prey in the food chain and food web dynamics of the ecosystem. From the present study, 34 species under 19 families have been recorded from this KWLS, of which families Culicidae and Syrphidae have been reported as major groups in terms of species numbers as well as abundance.

Among the two major groups of dipteran insects from the study area, the family Culicidae includes some vector species, namely, *Anopheles (Cellia) culifacies* and *Anopheles (Cellia) fluviatilis* (Malaria vectors), *Aedes (Stegomyia) albopictus* (Dengue and Chikungunya vector), and *Mansonia (Mansonoides) annulifera* (Japanese Encephalitis vector) which are responsible for several deadly diseases in India (Tyagi *et al.,* 2015). Because of the favourable environmental conditions, members of the Culicidae family were most abundant during the post-monsoon season, followed by the monsoon season. Being ectotherms, the environmental factors like average temperature, relative humidity, and precipitation rates of any given area impart considerable impacts on eco-biology, especially on the development and life cycles of this insect fauna. Though only 1–10 per cent of the laid eggs emerge as adults (Aniedu *et al.,* 1993; Okogun, 2005), each life stage of these mosquitoes (egg, larva, pupa, and adult) is dependent on the temperature for its

developmental and mortality rates (Beck-Johnson *et al.,* 2013). As the temperature range during the post-monsoon and monsoon has appeared to be most favourable for the development of dipteran species, the relative abundance of this group was observed to be specifically higher compared to other dipteran groups from this area. One dipteran species, *Tabanus rubidus*, a tabanid fly in the family Tabanidae and notorious carrier of Surra disease in India, has also been reported from this KWLS, India (Basu *et al.,* 1952; Veer *et al.,* 2002).

Besides the deadly dipteran creatures that act as public health nuisances, some beneficial groups of dipteran insects were found in KWLS. The members of the family Syrphidae are one of those ecosystem service providers that, besides the honeybees, play a crucial role as pollinators (Mitra and Banerjee, 2007; Orford et al., 2015). Different species of flower flies (Syrphidae) were recorded in and around the KWLS from several plants as flower-visiting insects. They mainly visited flowers to feed themselves with nectar (myophily), which in turn enabled them to act as potential secondary or accidental pollinators. The flower flies or hover flies (Family Syrphidae) used to display their peak abundance during the pre-monsoon, while their abundance started declining drastically during the post-monsoon and monsoon. Hover flies are one of the major groups of flower-visiting insects and also act as pollinators for several wild plants, agricultural plants, ornamental plants, medicinal plants, etc. (Mitra and Banerjee, 2007; Klecka *et al.,* 2018). However, the fact remains that the roles in plant–pollinator interactions rendered by syrphid and some non-syrphid groups are often underappreciated (Inouye *et al.,* 2015). A detailed account of the roles of syrphid and non-syrphid dipteran groups as potential pollinators on a comparative basis is also available by studying 30 pollen-transport networks and 71 pollinator-visitation networks, which categorically indicate the importance of these forgotten flies in pollination (Orford *et al.,* 2015). The pre-monsoon season, with plenty of blooming flowers in the study area, including the flowers of the most dominant Sal trees, *Shorea robusta*, was seen to attract one syrphid fly, *Dideopsis aegrota*, in higher abundance, followed by other dipterans, hymenopteran insects, and lepidopteran insects in and around Kuldiha Wild Life Sanctuary (KWLS).

While observing the habitat-wise distribution of the dipteran species, it was noticed that the deep forest area (LCT-2) has fewer species diversity with a relatively high species abundance, which is supposed to be due to the homogeneity of the ecological conditions of the habitats, which are characterised by less penetration of sunlight, higher humidity, less availability of open space to fly, etc. Most of the dipteran insects were observed to flourish in either the blooming flowers or over the surface of the elephant dung. The elephant dungs were mostly visited by dipteran insects like *Chrysomya megacephala*, *Sarcophaga (Liosarcophaga) dux*, and *Neomyia* sp., along with several other insect groups like beetles (Coleoptera), ants (Hymenoptera), termites (Isoptera), etc. Certain species from families like Muscidae, Calliphoridae, Tabanidae, and Sarcophagidae could develop a close relation to human settlements (Chaiwong *et al.,* 2012; Valverde-Castro *et al.,* 2017), and thus their presence in high numbers and diversity were found at LCT–3. Aside from the presence of the greatest number of species and relatively higher abundance of mosquitoes in villages, it appears to be significant in terms of their availability of food as human blood, as most of them were found in LCT-1 as well.

Natural forests, specifically virgin evergreen forests, serve as the key reservoir of biodiversity, as they always signify their unique floral and faunal composition. Despite all the threats, any protected area holds rich biodiversity. Therefore, it is always essential to document the biodiversity of any ecosystem or protected area for the purposes of planning better eco-management and conservation. Increased species richness enhances the performance of entire communities but reduces the average contributions of individual species. This paper fulfils the preliminary knowledge of the dipteran diversity in and around the Kuldiha WLS along with the roles of habitat heterogeneity and favourable seasonal conditions to trigger both the diversity and abundance of dipteran flies. Moreover,

the present research study has also highlighted some functional roles of the major groups of dipterans, like the Culicidae and Syrphidae. Being an unexplored eco-zone, the biodiversity of this KWLS may attract the attention of several biodiversity experts who can explore different areas of biodiversity research in order to strengthen the biodiversity documentation process of the country, and the research outcomes from the present paper may certainly fill such a lacuna in the knowledge base by generating some baseline research information on the dipteran community of KWLS.

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A checklist of robber flies (Diptera, Asilidae) of Kerala, India

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ABSTRACT: A checklist of robber fly species reported in Kerala, India based on literature survey is provided. In this list, 87 species of robber flies representing 25 genera and eight subfamilies are enumerated. The diversity of robber flies in Kerala was highlighted. Most of the species were reported from the protected forest areas of Kerala such as Ponmudi, Anamalai hills, Idamalayar, Thekkadi, Valparai, Chembra peak, Nilambur, Peermade, Walayar, Tenmalai and Silent Valley of the Western Ghats.

KEYWORDS: Assassin flies, biodiversity, taxonomy, Western Ghats

INTRODUCTION

The Western Ghats is regarded as one of the eight biodiversity hotspots in the world and as a vulnerable ecological region. With its tall, dense tropical rain forests, Kerala, at the southernmost tip of India, boasts the most diverse vegetation of the region (Reddy *et al.,* 2016). Asilidae is the third most diverse family in the order Diptera and is commonly known as robber flies or assassin flies (Pape *et al*., 2011; Brown *et al*., 2018). They are a significant group of predators in all zoogeographical zones and contain 7531 species in 556 genera scattered throughout the world (Pape *et al*., 2011; Dikow, 2020). Currently a comprehensive information on the Asilidae of Kerala is not available. As a foundation on this fauna, a checklist of the asilid species previously recorded from Kerala was worked out.

MATERIALS AND METHODS

The checklist was prepared entirely based on a literature. Asilidae generic classification *sensu* Dikow (2009) is followed in this study. Reported details regarding the robber fly diversity of Kerala were collected from various sources in the literature. When the exact distribution of a taxon is unknown, it is recorded simply as Kerala.

RESULTS AND DISCUSSION

Subfamily Asilinae Latreille, 1802

Genus *Astochia* Becker, 1913

The genus *Astochia* Becker has a vast distribution in the Oriental region, which includes China, India, Indonesia (Java, Sumatra), Philippines, and Thailand (Scarbrough and Biglow, 2004).

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1. *Astochia pseudoguptai* Joseph and Parui, 1987

Distribution**:** Nilambur (Malappuram)

Source: Roy *et al*. (2020)

Genus *Clephydroneura* Becker, 1925

Robber flies of the genus *Clephydroneura* are primarily found in tropical Asia.

2. *Clephydroneura anambrevipennis* Oldroyd, 1938

Distribution: Thekkady (Idukki)

Source: Mathew (2004)

3. *Clephydroneura apicalis* Oldroyd, 1938

Distribution: Kerala

Source: Parui and Joseph (1994), Joseph and Parui (1995)

4. *Clephydroneura brevipennis* Oldroyd, 1938

Distribution: Nelliyampathy (Palakkad), Thekkady (Idukki)

Source: Joseph and Parui (1990b, 1997), KFRI (1999), Mathew (2004)

5. *Clephydroneura exilis* Oldroyd, 1938

Distribution: Thekkady (Idukki)

Source: Joseph and Parui (1984c)

6. *Clephydroneura oldroydi* Joseph and Parui, 1995

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1995)

Remarks: *Clephydroneura anamalaiensis* Joseph and Parui, 1979 has been reported from Cinchona hills according to Joseph and Parui (1990b) and Mathew (2004). But Cinchona hills is a part of the Anaimalai hills of Tamil Nadu side, and not in Kerala as wrongly stated by Joseph and Parui (1981: 216) in their original description, but correctly cited by Joseph and Parui for the holotype of *Laphria nathani* Joseph and Parui (1981: 217) in this same paper.

Genus *Heligmonevra* Bigot, 1858

The range of the genus is primarily limited to the Oriental and Afrotropical regions. The genus contains 34 species in the Oriental region, with the Philippines having the most with 26 species and India having 19 species placing it in second place (Joseph and Parui 1980; Scarbrough and Duncan 2004).

7. *Heligmoneura cheriyani* (Joseph and Parui, 1980)

Distribution**:** Anamalai hills

Source: Joseph and Parui (1990a, 1997), Mathew (2004)

8. *Heligmoneura indirae* (Joseph and Parui, 1997)

Distribution**:** Dhoni (Palakkad)

Source: Joseph and Parui (1997)

9. *Heligmoneura poonmudiensis* (Joseph and Parui, 1980)

Distribution: Ponmudi (Thiruvananthapuram), Idamalayar (Ernakulam)

Source: Joseph and Parui (1987b, 1990b), Mathew (2004)

Genus *Machimus* Loew, 1849

Most species are native to the Palearctic ecozone and Southern Asia. The genus is distributed in the Oriental, Palearctic, Nearctic and Afrotropical regions (Geller-Grimm, 2004).

10. *Machimus calicutensis* Joseph and Parui, 1986

Distribution: Chembra peak (Kozhikode)

Source**:** Joseph and Parui (1986a)

11. *Machimus hirtipes* Ricardo, 1919

Distribution: Chembra peak (Kozhikode)

Source**:** Joseph and Parui (1991, 1995)

12. *Machimus keralaensis* Joseph and Parui, 1986

Distribution: Anamalai hills

Source**:** Joseph and Parui (1990b), Mathew (2004)

13. *Machimus parvus* Ricardo, 1919

Distribution: Kerala

Source**:** Parui and Joseph (1994), Joseph and Parui (1997)

14. *Machimus smithi* Joseph and Parui, 1986

Distribution: Anamalai hills, Chembra peak (Kozhikode)

Source**:** Joseph and Parui (1990a)

Genus *Philodicus* Loew, 1848

The genus is well distributed in the Afrotropical region (Oldroyd, 1980) and in the Oriental region (Oldroyd, 1975).

15. *Philodicus ceylanicus* Schiner, 1868

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui (1990a, 1995, 1997)

16. *Philodicus chinensis* Schiner, 1868

Distribution: Kozhikode

Source**:** Joseph and Parui (1997)

17. *Philodicus femoralis* Ricardo, 1921

Distribution: Chalakudy (Thrissur), Nilgiri hills

Source**:** Joseph and Parui (1987b), Parui and Joseph (1994)

18. *Philodicus indicus* Joseph and Parui,1991

Distribution: Kozhikode

Source**:** Joseph and Parui (1997)

19. *Philodicus londti* Joseph and Parui,1991

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui (1991)

20. *Philodicus propinquus* Bromley, 1938

Distribution: Chavakkad (Thrissur)

Source: Bromley (1938)

21. *Philodicus pruthii* Bromley,1935

Distribution: Kerala

Source**:** Parui and Joseph (1994), Joseph and Parui (1997)

Genus *Promachus* Loew, 1848

The genus is distributed in all zoogeographical regions (Geller-Grimm, 2004).

22. *Promachus heteropterus* (Macquart, 1838)

Distribution: Malappuram

Source**:** KFRI (1999), Mathew (2004)

23. *Promachus leucotrichodes* Bigot, 1892

Distribution: Kannur

Source: KFRI (1999), Mathew (2004)

24. *Promachus maculatus* (Fabricius, 1775)

Distribution: Kerala

Source: Naskar *et al*. (2019)

25. *Promachus nedungaduensis* Tomasovic, 2013

Distribution: Nedungadu (Ernakulam)

Source: Tomasovic (2013)

26. *Promachus ramakrishnai (*Bromley, 1939)

Distribution: Taliparamba (Kannur)

Source: Joseph and Parui (1997), KFRI (1999), Mathew (2004)

27. *Promachus tristis* Bigot, 1892

Distribution: Kannur

Source: KFRI (1999), Mathew (2004)

28. *Promachus yerburiensis* Ricardo, 1920

Distribution: Kerala

Source: Menon (1976), Joseph and Parui (1981a, 1984a, 1995), Mathew (2004)

Subfamily Dasypogoninae Macquart, 1838

Genus *Pegesimallus* Loew, 1858

The genus is well distributed in the Afrotropical region and also in the Oriental region (Londt, 1980).

29. Pegesimallus volcatus (Walker, 1849)

Distribution: Ponmudi (Thiruvananthapuram)

Source**:** Joseph and Parui (1991)

Genus *Saropogon* Loew, 1847

Members of the genus are found in regions with temperate and tropical climates in the Palearctic and Nearctic regions. In addition, some species are known to exist in the Oriental, Australian, Neotropical and Afrotropical regions (Hull, 1962; Lehr, 1988).

30. *Saropogon hulli* Joseph and Parui, 1981

Distribution: Ponmudi (Thiruvananthapuram), Idamalayar (Ernakulam)

Source: Joseph and Parui (1981a, 1987b, 1990a, 1990b, 1991), Parui and Joseph (1994), Debabrata *et al*. (2016)

31. *Saropogon londti* Parui, 1999

Distribution: Nilgiri hills

Source: Parui (1999)

Subfamily Laphriinae Loew, 1847

Genus *Hyperechia* Schiner, 1866

The members of this genus are predominately distributed in the Afrotropical region, with only two species reported from the Oriental region (Joseph and Parui, 1998).

32. *Hyperechia xylocopiformis* (Walker, 1849)

Distribution: Vellayani (Thiruvananthapuram)

Source: Prathapan and Sankararaman (2022)

Genus *Laphria* Meigen, 1803

With more than 150 species the genus is widely distributed in all zoogeographical regions (Geller-Grimm, 2010).

33. *Laphria alternans* Wiedemann, 1828

Distribution: Valparai:Chalakudy (Thrissur)

Source: Joseph and Parui (1997, 1998), Chandra *et al.* (2020)

34. *Laphria fuscata* (Joseph and Parui, 1997)

Distribution: Munnar (Idukki)

Source: Joseph and Parui (1997, 1998)

35. *Laphria indica* Joseph and Parui, 1981

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1981c, 1990b, 1995, 1998), Mathew (2004)

36. *Laphria keralaensis* Joseph and Parui, 1981

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1981c, 1990b, 1995, 1998), Mathew (2004)

37. *Laphria nathani* Joseph and Parui, 1981

Distribution: Chembra peak (Kozhikode), Peermade (Kottayam), Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1981c, 1986b, 1990b, 1991, 1995, 1997, 1998), Parui and Joseph (1994), Mathew (2004), Debabrata *et al*. (2016), Chandra *et al.* (2020)

38. *Laphria valparaiensis* Joseph and Parui, 1997

Distribution: Valparai-Chalakudy (Thrissur)

Source: Joseph and Parui(1997)

Genus *Maira* Schiner, 1866

The genus is predominantly distributed in the Oriental and the Australian region (Joseph and Parui, 1998).

39. *Maira pseudoindiana* Joseph and Parui, 1995

Distribution: Ponmudi (Thiruvananthapuram)

Source**:** Joseph and Parui (1995, 1998)

Genus *Nusa* Walker, 1851

The genus is widely distributed in the Afrotropical and Oriental regions (Joseph and Parui, 1998).

40. *Nusa pseudoalbibasis* Joseph and Parui, 1987

Distribution: Tenmalai (Kollam)

Source: Joseph and Parui (1987c, 1990b, 1998), Debabrata *et al*. (2016), Chandra *et al.* (2020)

41. *Nusa sahai* (Joseph and Parui, 1997)

Distribution: Valparai: Chalakudy (Thrissur), Nilambur (Malappuram)

Source**:** Joseph and Parui (1997), Chandra *et al.* (2020)

Subfamily Leptogastrinae Schiner, 1868

Genus *Leptogaster* Meigen, 1803

A common genus distributed in all the zoogeographical regions of the world (Joseph and Parui, 1998).

42. *Leptogaster albimana* Walker, 1859

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1991, 1998), Chandra *et al.* (2020)

Genus *Lobus* Martin, 1972

The genus is mainly distributed in Afrotropical region and Oriental region (Naskar *et al.* 2018).

43. *Lobus jairami* Joseph and Parui, 1984

Distribution: Edapaliyam (Kottayam)

Source: Joseph and Parui (1984b, 1990b, 1998), Mathew (2004), Debabrata *et al*. (2016), Naskar *et al.* (2018), Chandra *et al.* (2020)

44. *Lobus keralae* Martin, 1972

Distribution: Walayar (Palakkad)

Source: Joseph and Parui (1990b, 1998), Mathew (2004), Naskar *et al.* (2018), Chandra *et al.* (2020)

45. *Lobus martini* Joseph and Parui, 1983

Distribution: Konnakuzhi (Thrissur), Kannimangalam (Eranakulam), Malayattoor (Eranakulam), Idamalaya (Eranakulam)

Source: Joseph and Parui (1983, 1990b, 1998), Mathew (2004), Debabrata *et al*. (2016), Naskar *et al.* (2018), Chandra *et al.* (2020)

Subfamily Ommatiinae Hardy, 1927

Genus *Cophinopoda* Hull, 1958

The genus is distributed in the Oriental, Australian, Palaearctic and Afrotropical regions (Geller-Grimm, 2004).

46. *Cophinopoda chinensis* (Fabricius, 1794)

Distribution: Kerala

Source: Joseph and Parui (1987b, 1998), Chandra *et al.* (2020)

Genus *Emphysomera* Schiner, 1866

Dark, small to medium-sized robber flies distributed in Oriental, Australian and Afrotropical regions (Scarbrough and Marascia, 1999).

47. *Emphysomera tectura* Scarbrough and Marascia, 1999

Distribution: Ponmudi (Thiruvananthapuram), Chembra peak (Kozhikode), Walayar (Palakkad)

Source**:** Scarbrough and Marascia (1999)

Genus *Michotamia* Macquart, 1838

The genus is distributed in Oriental, Australian and Afrotropical regions (Geller-Grimm, 2004).

48. *Michotamia aurata* (Fabricius, 1794)

Distribution: Thekkady (Idukki), Chembra peak (Kozhikode)

Source: Joseph and Parui (1984a, 1984c, 1991, 1997, 1998), Parui and Joseph (1994), Mathew (2004), Chandra *et al.* (2020)

49. *Michotamia fuscifemorata* Joseph and Parui, 1984

Distribution: Walayar (Palakkad), Thekkady (Idukki), Tenmalai (Thiruvananthapuram), Kumili (Idukki), Chalakudy (Thrissur)

Source: Joseph and Parui (1984c, 1986b, 1987b, 1990b, 1995, 1997, 1998), Mathew (2004), Debabrata *et al*. (2016), Chandra *et al.* (2020)

50. *Michotamia indiana* Joseph and Parui, 1981

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui (1981b, 1990b, 1997, 1998), Mathew (2004), Debabrata *et al*. (2016), Chandra *et al.* (2020)

Genus *Ommatius* Wiedemann, 1821

A common genus distributed in all the zoogeographical regions of the world (Geller-Grimm, 2004).

51. *Ommatius disparis* Scarbrough, 2007

Distribution: Kerala

Source: Chandra *et al.* (2020)

52. *Ommatius dravidicus* Joseph and Parui, 1983

Distribution: Anamalai hills

Source: Parui and Joseph (1983), Joseph and Parui (1995, 1998), Debabrata *et al*. (2016), Chandra *et al.* (2020)

53. *Ommatius hulli* Joseph and Parui, 1983

Distribution: Anamalai hills, Ponmudi (Thiruvananthapuram), Silent Valley (Palakkad)

Source: Parui and Joseph (1983), Joseph and Parui (1984a, 1984c,1998), Mathew (2004), Debabrata *et al*. (2016), Chandra *et al.* (2020)

54. *Ommatius indicus* Joseph and Parui, 1983

Distribution: Ponmudi (Thiruvananthapuram), Peermade (Kottayam), Munnar, Periyar (Idukki), Malayatur, Pandupara, Idamalayar (Ernakulam), Konnakuzhi,Poringalkuth (Thrissur), Nilambur (Malappuram), Kozhikode, Dhoni (Palakkad), Ranni (Pathanamthitta)

Source: Parui and Joseph (1983), Joseph and Parui (1987b, 1989, 1995, 1997, 1998), Debabrata *et al*. (2016), Chandra *et al.* (2020)

55. *Ommatius kodaikanalensis* Joseph and Parui, 1994

Distribution: Munnar (Idukki)

Source: Joseph and Parui (1997)

56. *Ommatius malabaricus* Joseph and Parui, 1985

Distribution: Taliparamba (Kannur)

Source: Joseph and Parui (1984a, 1990b, 1998), KFRI (1999), Mathew (2004), Debabrata *et al*. (2016), Chandra *et al.* (2020)

57. *Ommatius minor* Doleschall, 1857

Distribution: Anamalai hills, Chembra Peak (Kozhikode)

Source: Joseph and Parui (1984a, 1990a, 1991, 1997, 1998), Mathew (2004), Chandra *et al.* (2020)

58. *Ommatius pillaii* Joseph and Parui, 1986

Distribution: Silent Valley (Palakkad)

Source: Joseph and Parui (1986c, 1990b, 1998), Debabrata *et al*. (2016), Chandra *et al.* (2020)

59. *Ommatius tuberculatus* Joseph and Parui, 1983

Distribution: Ponmudi (Thiruvananthapuram), Chembra Peak (Kozhikode), Anamalai hills, Peermade, Periyar (Idukki)

Source: Parui and Joseph (1983), Joseph and Parui (1987b, 1990a, 1990b, 1991, 1995,1997, 1998), Chandra *et al.* (2020)

Genus *Pseudomerodontina* Joseph and Parui, 1976

The genus is reported only from the Oriental region (Geller-Grimm, 2004).

60. *Pseudomerodontina indica* Joseph and Parui, 1979

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui (1997, 1998), Chandra *et al.* (2020)

61. *Pseudomerodontina jayaraji* Joseph, 1976

Distribution: Maddathoray (Thiruvananthapuram)

Source: Joseph and Parui (1997, 1998), Chandra *et al.* (2020)

Subfamily Stenopogoninae Hull, 1962

Genus *Microstylum* Macquart, 1838

The members of this genus are distributed in Palearctic, Nearctic, Afrotropical and Oriental regions (Geller-Grimm, 2004).

62. *Microstylum ananthakrishnani* Joseph and Parui, 1984

Distribution: Ponmudi (Thiruvananthapuram), Silent valley (Palakkad)

Source: Joseph and Parui (1986c, 1987d, 1991), Parui and Joseph (1994), Mathew (2004), Debabrata *et al*. (2016)

63. *Microstylum bhattacharyai* Joseph and Parui, 1984

Distribution: Mudutailam (Thrissur)

Source: Joseph and Parui (1984c, 1989, 1990a, 1995), Mathew (2004)

64. *Microstylum varshneyi* Joseph and Parui, 1984

Distribution: Walayar (Palakkad)

Source**:** Joseph and Parui (1987a, 1989)

Genus *Scylaticus* Loew, 1858

The members of this genus are distributed in Palaearctic, Neotropical, Afrotropical and Oriental regions (Geller-Grimm, 2004).

65. *Scylaticus indicus* Bromley, 1939

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui (1991)

Genus *Stenopogon* Loew, 1847

The genus is distributed in the Palearctic, Nearctic,

Afrotropical and Oriental regions (Geller-Grimm, 2004).

66. Stenopogon cinchonaensis Joseph and Parui, 1981

Distribution: Anamalai hills

Source: Joseph and Parui (1990b), Mathew (2004), Debabrata *et al*. (2016)

67. *Stenopogon hulli* Joseph and Parui, 1981

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1987d)

68. *Stenopogon kherai* Joseph and Parui, 1976

Distribution: Anamalai hills

Source: Joseph and Parui (1990a, 1991)

69. *Stenopogon manii* Joseph and Parui, 1981

Distribution: Anamalai hills, Thenmala (Kollam), Peermade (Kottayam), Walayar (Palakkad)

Source: Joseph and Parui (1981b, 1984a, 1990a, 1990b, 1995), Parui and Joseph (1994), Mathew (2004), Debabrata *et al*. (2016)

70. *Stenopogon raven* (Bromley, 1938)

Distribution: Walayar (Palakkad)

Source: Parui and Joseph (1994), Joseph and Parui (1995)

Subfamily Stichopogoninae Hardy, 1930

Genus *Stichopogon* Loew, 1847

The genus is distributed in almost all zoogeographical regions (Geller-Grimm, 2004).

71. *Stichopogon inequalis* (Loew, 1847)

Distribution: Kozhikode

Source: Joseph and Parui (1998), Chandra *et al.* (2020)

72. *Stichopogon meridionalis* Oldroyd, 1948

Distribution: Kozhikode

Source: Joseph and Parui (1998), Chandra *et al.* (2020)

Subfamily Trigonomiminae Enderlein, 1914

Genus *Damalis* Fabricius, 1805

The genus is distributed in the Palearctic, Australasian, Afrotropical and Oriental regions (Geller-Grimm, 2004).

73. *Damalis artigasi* Joseph and Parui, 1984

Distribution: Idamalayar (Ernakulam), Peermade (Kottayam)

Source: Joseph and Parui (1984d, 1987b, 1995), Mathew (2004)

74. *Damalis anamaliensis* Scarbrough, 2007

Distribution: Kerala

Source: Chandra *et al.* (2020)

75. *Damalis calicutensis* Joseph and Parui, 1990

Distribution: Kozhikode

Source: Mathew (2004),Debabrata *et al*. (2016)

76. *Damalis cederholmi* Joseph and Parui, 1984

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1984d, 1990a, 1991, 1995), Mathew (2004)

77. *Damalis dimidiata* Joseph and Parui, 1990

Distribution: Kovalam beach (Thiruvananthapuram)

Source: Joseph and Parui (1997)

78. *Damalis dravidica* Joseph and Parui, 1984

Distribution: Ponmudi (Thiruvananthapuram), Kottayam

Source: Joseph and Parui (1984a, 1984d, 1986b, 1987d, 1995), Mathew (2004), Debabrata *et al*. (2016)

79. *Damalis dubia* Joseph and Parui, 1995

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1995)

80. *Damalis fusca* Walker, 1849

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1991, 1995), Parui and Joseph (1994)

81. *Damalis indica* Joseph and Parui, 1984

Distribution: Ponmudi (Thiruvananthapuram), Anamalai hills

Source: Joseph and Parui (1984d, 1990a, 1990b, 1995), Debabrata *et al*. (2016)

82. *Damalis keralaensis* Joseph and Parui, 1984

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1984d), Mathew (2004), Debabrata *et al*. (2016)

83. *Damalis kottayamensis* Joseph and Parui, 1995

Distribution: Peermade (Kottayam)

Source: Joseph and Parui(1995)

84. *Damalis mercaraensis* (Joseph and Parui, 1984)

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui(1995)

85. *Damalis pseudoartigasi* Joseph and Parui, 1989

Distribution: Ponmudi (Thiruvananthapuram)

Source: Joseph and Parui (1995)

86. *Damalis rufoabdominalis* Joseph and Parui, 1984

Distribution: Chembra peak (Kozhikode)

Source: Joseph and Parui (1984d, 1990b, 1995), Debabrata *et al*. (2016)

Genus *Trigonomima* Enderlein, 1914

The genus is distributed only in the Oriental region (Geller-Grimm, 2004).

87. *Trigonomima anamaliensis* Joseph and Parui, 1980

Distribution: Anamalai hills

Source: Joseph and Parui (1990a)

The diversity of robber flies in Kerala was highlighted by this checklist, which was fully based on literature review and suggests the presence of at least 87 species of robber flies in 25 genera. Most of the species were reported from the protected forest areas of Kerala such as Ponmudi, Anamalai hills, Idamalayar, Thekkadi, Valparai, Chembra peak, Nilambur, Peermade, Walayar, Tenmalai and Silent Valley, which mainly consist of wet evergreen forests, semi-evergreen forests, moist deciduous forests, dry deciduous forests and shola grassland complexes (Champion and Seth, 1968). A more thorough understanding of the robber fly diversity of Kerala will require extensive field surveys, examination of museum collections, and revisionary taxonomy.

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Seasonal population dynamics of *Aceria erineus* (Nalepa) (Acari, Eriophyidae) on walnut trees in Kashmir, India

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ABSTRACT: Population fluctuation of *Aceria erineus* in relation to abiotic factors on different walnut orchards of Kashmir was roving surveyed in 27 agricultural sites in nine districts of Kashmir during May 2018 to April 2019. In each district, three sites were selected randomly and the incidence of the mite was recorded at fortnightly intervals throughout the crop growth period. Field surveys indicated the presence of the pest in the entire surveyed region in varying intensity with maximum incidence recorded in Kupwara (21.77±1.39 mites per leaf), Baramulla (19.27±1.09 per leaf), Shopian (18.4±2.47 per leaf), Budgam (18.33±1.24 per leaf) and Pulwama (18.24±1.75 per leaf) respectively. Significant positive correlations of mite populations with minimum temperature were found in Shopian and all the districts of north Kashmir. Significant negative correlation was also found between mite population and rainfall except Anantnag district where it was non-significant. Maximum and minimum temperatures as well as sunshine hours had non-significant positive correlation, whereas relative humidity had non-significant negative correlation with the mite population.

KEYWORDS: Blister mites, abiotic factors, seasonal variations

The Jammu and Kashmir is the major walnut producing region in India and almost entire quantity (98%) of walnut is exported from this region. Jammu and Kashmir is the major walnut producing region contributing more than 85 percent of total production of the country. Common walnut (*Juglans regia* L.) is cultivated in the districts of Poonch, Pulwama, Anantnag, Ganderbal, Kulgam, Budgam, Kupwara, Baramulla and Srinagar. Among these districts, Shopian was on the forefront in walnut production in the past. The trend has changed and nowadays district Kupwara is leading in walnut production (Anonymous, 2015). Pest and disease management is a challenge in walnut because of giant size trees. Among the non- insect pests of crops, mites are probably the most notorious ones and gaining tremendous importance in the recent years owing to their devastating nature (Tabasum and Buhroo, 2022). Several eriophyoid mites are known to occur on walnut and other species of *Juglans* (Amrine and Stasny, 1994). Eriophyoids are obligatory plant feeders with unusual morphological, biological and behavioural specialization compared to other Acari (Skoracka *et al.,* 2010). Many of them are major pests of agricultural and ornamental crops, wild plants, grasses, and plants of urban and community forestry but rarely cause their death (Lindquist *et al.,* 1996).

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A number of eriophyoid species are considered to be main pests on some crops, while others are known to be a quarantine threat for several countries. *Aceria erineus* (Nalepa) (Acari, Eriophyidae) is a small, yellowish-white, slender, wormlike mite, measuring about 0.25mm long. They have a waxy white colour and generally can only be seen with a stereomicroscope. The walnut blister mite attacks the lower surface of the leaves of Persian (English) walnut (Miller and Thamson, 1937). The infestation caused by this mite is noticeable as shiny, convex swellings on the upper surface of the leaf blade and on the underside as patches of shallow, large, solitary concavities lined with felty, yellowish hairs, among which the mites are found. Walnut blister mites overwinter beneath bud scales. When spring time temperatures rise, the mites feed among the leaf hairs on the undersides of the leaves. Several generations occur during the summer, which attack new foliage as soon as it unfurls. Since the degree of incidence of blister mite changes with season, it is desirable to have a thorough understanding of the seasonal incidence of the mite for the development of suitable management programmes. Hence, an attempt was made to determine the effect of weather factors on the incidence and dynamics of this blister mite on walnut trees of Kashmir.

Studies on the seasonal variation of walnut blister mite *A. erineus* were carried out from May 2018 to April 2019 on different walnut orchards of Kashmir. Roving surveys were conducted in 27 agricultural sites in nine districts of Kashmir *viz.,* Anantnag, Pulwama, Shopian, Srinagar, Ganderbal, Budgam, Baramulla, Bandipora and Kupwara. The experimental plots were kept completely free from insecticidal applications. In each district, three sites were selected randomly and the incidence of the mite was recorded at regular intervals throughout the crop growth period. Five plants were selected randomly at each site and tagged as a replication. Altogether, six leaves from different layers (top, middle and bottom) from each main side of the tree canopy (north, south, east and west) were collected from fifteen randomly selected plants per sampling date, making a total 90 leaves collected fortnightly. After conducting on-site visual sampling of pests, the samples were put in polythene bags and tied loosely with rubber bands for subsequent transportation to laboratory for further observation under stereo binocular microscope. Data on the mite population was recorded throughout the crop growth period and mean number of mites per leaf was calculated for each sampling period. Concerning the significant factors emerged, ANOVA was carried out and a post hoc test Fisher's Least Significant Difference (LSD) was applied for the comparison of means of different fortnights within the district. Simultaneously, the weather data on maximum and minimum temperature (°C), relative humidity (%), sunshine (h), and rainfall (mm) were also collected from the Indian Meteorological Department (IMD) Rambagh Srinagar. The data on the mite population used in the analysis and correlation coefficient was worked out with above meterological parameters. All the statistical procedures were carried out using MS Excel software and SPSS software. To arrive at meaningful results with respect to the impact of weather parameters on the population dynamics of blister mite, the data collected from three sites of each district was pooled and the relationship between the mite population per leaf and the average weather parameters was worked out using correlation analysis at 5 per cent significance level.

Mite population density: Incidence of walnut blister mite was noticed in all the nine districts of Kashmir. However, the maximum leaf infestation was found in Kupwara district recording 21.77±1.39 mites per leaf, followed by Baramullah district (19.27±1.09 per leaf), Shopian district (18.4±2.47 per leaf), Budgam district (18.33±1.24 per leaf), and Pulwama district (8.24±1.75 per leaf). The low mite density was recorded in Srinagar (16.08±1.60 per leaf), Anantnag (16.75±1.42 per leaf), Ganderbal (17.57±1.79 per leaf), Bandipora (17.64±1.53 per leaf). The blister mite population commenced from the 19th standard week of May on walnut trees in all the surveyed locations. Sampling started soon after the first appearance of blisters in spring and continued until autumn. Mite populations showed an exponential growth upto the beginning of July, then the mite population declined in an inconsistent manner at the start of rainy season

Table 1. Pearson's correlation (r) between blister mite population and weather parameters in nine districts of Kashmir

Districts	T max	T min	Rf (mm)	RH1	RH2	SSH
Anantnag	0.821	0.734	-0.076	-0.076	-0.077	0.360
Pulwama	0.908	0.758	-0.031*	-0.052	-0.200	0.659
Shopian	0.864	0.58*	-0.133*	0.010	-0.271	0.593
Srinagar	0.811	0.667	-0.157*	-0.084	-0.133	0.320
Ganderbal	0.853	0.760	-0.062*	-0.109	-0.077	0.398
Budgam	0.855	0.804	-0.003*	-0.085	-0.031	0.436
Baramulla	0.806	0.72*	-0.055*	-0.042	-0.075	0.363
Bandipora	0.856	0.80*	-0.002*	-0.117	-0.057	0.415
Kupwara	0.922	0.76*	-0.001*	-0.072	-0.219	0.666

T max = Maximum temperature; T min = Minimum temperature; Rf (mm) = Rainfall; RH1= Relative humidity; RH2 = Relative humidity; SSH = Sunshine hours; * Significant correlations at $p < 0.05$

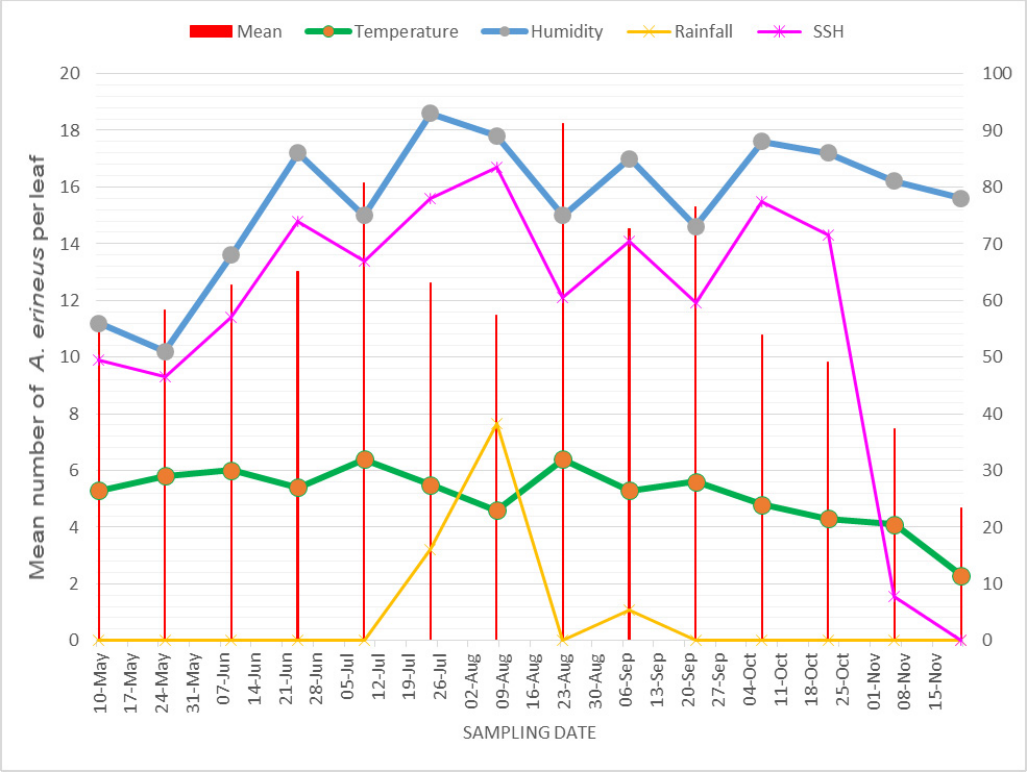

Fig. 1 Seasonal population dynamics of *Aceria erineus* on *Juglans regia* and fluctuations of temperature (°C), relative humidity (%), rainfall (mm) and sunshine (hours)

and as the at-mospheric relative humidity increased from mid july to the first week of August, again the population started to grow from mid August to September. Subsequently the population dropped rapidly in the month of october and remained at extreme low levels in November (Fig. 1). From December to March, the mite populations were not recorded because they undergo overwintering diapause. During the summer, the females migrate from the erineum to the terminal buds, where they overwinter (Keifer *et al.,* 1982).

Correlation between mite population and weather parameters: The relationship between *A. erineus* population and weather parameters was assessed through correlation analysis. Mite population showed non-significant positive correlation with maximum and minimum temperature in all the districts of valley with the exceptions of significant positive association with minimum temperature in Shopian district and all the districts of north Kashmir. With relative humidity, mite population showed non-significant negative correlation in all the districts of the valley. Significant negative correlation was also found between mite population and rainfall except Anantnag district where non-significant negative correlation was found. With Sunshine hours, mite population showed non-significant positive correlation in all the districts of Kashmir (Table 1).

The walnut growing regions of Kashmir showed slight to severe infestation with the maximum incidence in Kupwara district followed by Baramulla district and the least incidence was noticed in Srinagar and Ganderbal districts. The variations in mite incidence in the study locations may be due to the interplay of various biotic and abiotic factors that influence the pest population. The highest density and activity of *A. erineus* was observed during the beginning of July and late August. During these months, the average temperature and relative humidity were 32 °C, 75 percent respectively. The prevailing high temperature coupled with low humidity was observed to be very advantageous for the rapid multiplication of this species of mite.

The seasonal variation of this species is comparable to that of other eriophyids. The mite's incidence was recorded on walnut trees throughout the crop growth period, indicative of overlapping generations. *A. erineus* showed a bimodal dynamic, with two distinct population peaks during early July and late August, this behaviour has been reported for other eriophyids in temperate zones that produce malformations such as *Phytoptus phloeocoptes* (Nalepa) in peach and *A. cinerea* in walnut, where it has been observed that the greatest abundance of malformations and number of individuals occur in the months where summer begins and decreases as winter begins (Boczek 1974; Keifer *et al.,* 1982). The results are also in agreement with the findings of Canales *et al.* (2019) on soursop where two population peaks were described, but contrast the data coming from Italy where *Aculus schlechtendali* peak population was observed only in the month of July on apple orchard (Simoni *et al.* 2018); similar results have been reported for *Aculus schlechtendali* by Hoyt (1969), Herbert (1974), Easterbrook (1979) who found the populations peak in July or early August.

Populations of *A. erineus* were positively and non-significantly correlated with temperature in almost all the districts indicating that higher temperature would be ideal for the build-up of mite population. This is in conformity with the findings of Abou-Awad (1981) for *Eriophyes mangiferae*. Significant positive correlation of mite population with minimum temperature was found in Shopian and all the districts of North Kashmir, as shown by Ranjan and Ray (2015) for *A. litchi*, Abou-Awad *et al.* (2011) for *Calepitrimerus baileyi*, Abou-Awad *et al.* (2011) for *A. mangiferae* and *Metaculus mangiferae*. Not only temperature but also humidity affects the population growth of eriophyoid mites. In this study, relative humidity (morning and evening) had non-significant negative correlation with blister mite which is in accordance with the Ranjan and Ray (2015), Kamuran (2020) but in contrast to the findings of Abou-Awad *et al.* (2011) who found significant positive correlation between mite population and relative humidity. In addition, sunshine hours were found to be positively and non-significantly correlated with mite population

which agree with the findings of Thakur and Sharma (1990) for *A. litchi.* Significant negative correlation was also found between mite population and rainfall. This observation is in conformity with the findings of Nasareen and Ramani (2014) for *A. pongamiae* on *Pongamia pinnata*, Nasareen and Ramani (2015) for *A. doctersi* on *Cinnamomum verum* Ranjan and Ray (2015) for *A. litchi* on Litchi trees. Present investigations showed that in Kashmir valley, the incidence of *A. erineus* is high during the months of July and August, which indicates that appropriate plant protection measures should be applied during these months to prevent the crop loss.

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Effect of sowing dates and cultivars on the incidence of *Spodoptera frugiperda* (J.E. Smith) on maize (*Zea mays* L.) in Nagaland, India

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ABSTRACT: The Effect of sowing dates and cultivars on the incidence of fall army worm *Spodoptera frugiperda* (J.E. Smith) and yield attributes of maize (*Zea mays* L.) was carried out with five cultivars namely, Zarsi (local), Sipho (local), Ronimi (local), Khoi (local) and HQPM-1 (composite) and three different sowing dates (6th March, 21st March and 5th April). Among the different dates of sowing, 6th March recorded maximum pest incidence, while 5th April recorded the least on maize. The interaction between sowing dates and cultivars showed significant effect on the incidence of army worm at different days after sowing. The maize sown on 21st March, recorded the highest grain yield (4.12 t ha^{-1}). It can be suggested that manipulating the sowing date and growing of tolerant variety of maize such as HQPM1 can be an effective measure to manage exotic army worm infestation. Mid sowing of maize (21st March) and growing of local cultivars such as Sipho observed significantly better yield attributes which will ensure higher economic returns to the farmers.

KEY WORDS: Exotic army worm, months, pest incidence, grain yield

Maize (*Zea mays* L.) is an important cereal crop belonging to the family Gramineae. It is also called as the 'Queen of Cereals' which is the most widely cultivated cereal crop in India. It is a staple and an important source of carbohydrate in human diet and also serves as a source of animal feed (Onasanya *et al*., 2009), an important source of industrial and pharmaceutical production in the country (Olaniyan, 2015). The yield of maize is greatly affected by many insect pests. Out of 140 species of insect pests (army worm, stem borer, thrips, aphids, termites, white grub, seed corn maggots, root worms, Indian meal moth, grain borer and grain weevil during storage), only 12 species are the serious pests of maize causing damage from sowing to the harvesting and also in the storage conditions (Siddiqui and Marwaha, 1993). Exotic army worm *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera, Noctuidae), infests maize crop from emergence to tasseling, silking and cob formation stage. The caterpillars feed on leaves and stems of more than 80 plant species.

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Dates of planting significantly influence the growth, development and yield due to climate changes that occur during the cropping season (Dahmardeh, 2012). Manipulation of sowing dates of crops is an important cultural practice to avoid the peak infestation of insect pests on the crop. In this context, the information pertaining to dates of sowing (DOS) on the incidence of major pest infestation in maize and also in yield has been lacking in the regions of Nagaland.

A field experiment was carried out in the experimental farm of SASRD, Nagaland University, Medziphema campus, Nagaland during March to July, 2019 located at 25° 45′ 45′′N; 93° 53′ 04′′E, at an altitude of 304.8m above mean sea level, in the foot hills of Nagaland. The experimental site falls under sub-tropical with high humidity and moderate annual temperature range (21-32°C), having average annual rainfall (2000 - 3000mm) and RH (70-80%). The soil is sandy loam in texture, acidic in nature with pH ranging from 4.5-6.5. The treatments are three dates of sowing [6th March (D_1), 21st March (D_2) and 5th April (D_3)] and five cultivars [Zarsi (C_1), Sipho (C_2), Ronimi (C_3), Khoi (C_4) and HQPM-1(C_5)]. The experiment was carried out in Split Plot Design with three replications, keeping planting dates in the main plot and cultivar in the sub plots. The main plot was

Table 1. Effect of different sowing dates and cultivars on leaf infestation by *Spodoptera frugiperda* on maize

Treatments	Infestation (%)		
	30 DAS	45 DAS	60 DAS
Sowing dates			
D1: 6th March	16.33(23.65)	21.87(27.73)	26.00(30.55)
D_2: 21st March	13.80(21.43)	18.00(24.82)	22.67(28.21)
D_3: 5th April	10.53(18.58)	14.47(22.07)	18.67(25.37)
SEm±	0.27	0.32	0.36
CD (p=0.05)	1.04	1.26	1.43
Cultivars			
C_1: Zarsi	15.56(23.06)	20.78(27.00)	25.56(30.28)
C_2: Sipho	10.78(18.99)	14.89(22.49)	18.89(25.59)
C_3: Ronimi	20.56(26.93)	25.78(30.47)	30.00(33.18)
C_4: Khoi	13.33(21.34)	17.78(24.90)	22.22(28.09)
C_5: HQPM1	7.56(15.79)	11.33(19.51)	15.56(23.08)
SEm±	0.43	0.50	0.69
CD (p=0.05)	1.25	1.47	2.02

Note: Figures in the table are mean values and those in parentheses are angular transformed values

divided into 5 sub-plots to accommodate five cultivars. Recommended agronomic package of practices were followed for the crop cultivation. The seeds were sown with a spacing of 60 X 25cm in a 4.2 X 1m plot size maintaining a population of 24 plants per plot. At random, ten plants were tagged in each plot to observe incidence of *S. frugiperda* at 30, 45, and 60 days after sowing (DAS). The crop was subjected to natural infestation and infestation was calculated (No. of infested plant/ Total no. of plants × 100). The yield attributes were recorded and the cob yield and grain yield were converted in t ha^{-1}. The data obtained were then subjected to analysis of variance (ANOVA). F test was used to determine the significance of difference between the two means and in case F-test was significant, the critical difference (CD) was calculated for comparison.

Effect of sowing dates and cultivars on leaf infestation by exotic army worm on maize:

DOS showed significant effect on infestation by army worm. At 30 DAS, the highest infestation (16.33%) was observed in 1st DOS, while it was the lowest (10.53%) in 3rd DOS. At 45 DAS the infestation was maximum in1st DOS (21.87%), followed by 2nd DOS (18.00%) and 3rd DOS (14.47%). Similar trend was observed at 60 DAS, where the highest infestation (26.00%) was noted at 1st DOS followed by 2nd DOS (22.67%) and the 3rd DOS recorded the lowest infestation (18.67%) (Table 1).

The interaction between sowing dates and cultivars showed significant effect on the incidence of army worm. At 30 DAS, 'Ronimi' sown on the 1st DOS recorded highest infestation (23.00%) and the lowest (6.00%) on the variety HQPM-1 sown on the 2nd DOS. At 45 DAS also Ronimi recorded, the highest percentage (29.33) sown on the 1st DOS and the lowest (9.00) HQPM-1 which was sown on 3rd DOS (Table 1). At 60 DAS the interaction between 1st DOS interacting with Ronimi showed the highest infestation and the lowest infestation was observed on the interaction of D_2C_5 (13.33%) and D_3C_1 (13.33%) with the variety HQPM-1(Table 2).

Table 2. Interaction effect of different sowing dates and cultivars on infestation by *Spodoptera frugiperda* on maize

Dates x Cultivars	Leaf infestation (%)		
	30 DAS	45 DAS	60 DAS
D_1C_1	18.67 (25.60)	25.33 (30.18)	30.00 (33.21)
D_1C_2	14.33 (22.23)	19.67 (26.30)	23.33 (28.88)
D_1C_3	23.00 (28.65)	29.33 (32.79)	33.33 (35.25)
D_1C_4	15.33 (23.04)	19.33 (26.05)	23.33 (28.86)
D_1C_5	10.33 (18.75)	15.67 (23.30)	20.00 (26.57)
D_2C_1	17.67 (24.85)	21.33 (27.51)	26.67 (31.07)
D_2C_2	10.67 (19.06)	15.33 (23.05)	20.00 (26.57)
D_2C_3	20.33 (26.79)	25.67 (30.44)	30.00 (33.21)
D_2C_4	14.00 (21.94)	18.33 (25.34)	23.33 (28.86)
D_2C_5	6.33 (14.53)	9.33 (17.78)	13.33 (21.34)
D_3C_1	10.33 (18.73)	15.67 (23.30)	20.00 (26.57)
D_3C_2	7.33 (15.68)	9.67 (18.11)	13.33 (21.34)
D_3C_3	18.33 (25.34)	22.33 (28.19)	26.67 (31.07)
D_3C_4	10.67 (19.06)	15.67 (23.31)	20.00 (26.57)
D_3C_5	6.00 (14.09)	9.00 (17.44)	13.33 (21.34)
SEm±	0.74	0.87	1.20
CD (p=0.05)	2.17	2.55	3.50

Note: Figures in the table are mean values and those in parentheses are angular transformed values

Table 3. Effect of different sowing dates and cultivars on yield attributes of maize

Treatments	No. of cobs plant^{-1}	Cob length (cm)	Cob diameter (cm)	Fresh cob weight (g cob^{-1})	Cob yieldt ha^{-1}	Grain yieldt ha^{-1}
Sowing dates						
D_1: 6th March	1.48	12.00	4.41	176.49	4.17	3.13
D_2: 21st March	1.61	14.20	4.64	214.39	5.49	4.12
D_3: 5th April	1.55	13.53	4.49	194.84	4.79	3.59
Sem±	0.001	0.12	0.01	1.51	0.04	0.03
CD (p=0.05)	0.004	0.48	0.03	5.94	0.15	0.11
Cultivars						
C_1: Zarsi	1.45	13.44	4.52	194.45	4.49	3.37
C_2: Sipho	1.62	14.44	4.74	214.04	5.50	4.13
C_3: Ronimi	1.38	12.56	4.32	188.29	4.15	3.11
C_4: Khoi	1.55	12.00	4.26	180.00	4.45	3.34
C_5: HQPM1	1.73	13.78	4.72	199.42	5.49	4.12
SEm±	0.002	0.13	0.01	1.73	0.04	0.03
CD (p=0.05)	0.005	0.37	0.04	5.05	0.11	0.09

The present finding indicates that late sowing performed better in regards to infestation by exotic army worm than early sowing. HQPM-1 was the most tolerant among the cultivars which might be due to the difference in morphological character like compactness of leaf tissue, hard and tough stem and genetic variability which render the variety 'HQPM - 1' cultivar to escape the attack.

Effect of sowing dates and cultivars on yield attributes of maize:

The results obtained revealed that maize sown on D_2 (21st March) significantly recorded the highest number of cob per plant (1.61), cob length (14.20 cm), cob diameter (4.64 cm), fresh cob weight (214.39g cob^{-1}), cob yield (5.49 t ha^{-1}) and grain yield (4.12 t ha^{-1}). Early sowing D_1 (6th March) recorded lesser yield. Among the different cultivars, HQPM1 (C_5) recorded the highest number of cob per plant (1.73) while the least was observed in Ronimi (1.38). The cultivar Sipho (C_2) recorded significantly higher cob length (14.44cm), cob diameter (4.74cm), fresh cob weight (214.04g cob^{-1}), cob yield (5.50 t ha^{-1}) and grain yield (4.13 t ha^{-1}) as compared to other cultivars (Table 3). The results showed significant interaction effect between sowing dates and cultivars on yield attributes (Table 4). The interaction between D_2C_5 (DOS 21st March and cultivar HQPM-1) recorded the highest number of cobs per plant (1.80) and cob diameter (4.90cm). The highest cob length (15.33cm), fresh cob weight (234.40 g cob^{-1}), cob yield (6.23 t ha^{-1}) and grain yield (4.67 t ha^{-1}) was obtained from the interaction between DOS 21st March and cultivar Sipho. It was observed that the late sown 2nd DOS crop (21st march) proved the best one as it gave the highest grain yield of 4.12 t ha-1 than that sown early on 1st DOS (March). The findings of Chaudhary and Sharma (1992) are also in accordance with the present studies.

From the results obtained in the present study, it can be concluded that late sowing of maize (5th

Table 4. Interaction effect of different sowing dates and cultivars on yield attributes of maize

DOS x Culti vars	Cobs plant^{-1}	Cob length (cm)	Cob diam eter (cm)	Fresh cob weight (g cob^{-1})	Cob yield t ha^{-1}	Grain yield t ha^{-1}
D_1C_1	1.38	12.33	4.40	176.27	3.87	2.90
D_1C_2	1.56	13.67	4.63	200.38	4.95	3.71
D_1C_3	1.32	11.00	4.23	167.22	3.51	2.63
D_1C_4	1.48	10.33	4.20	158.47	3.75	2.81
D_1C_5	1.66	12.67	4.57	180.10	4.75	3.57
D_2C_1	1.52	14.33	4.67	212.88	5.12	3.84
D_2C_2	1.67	15.33	4.85	234.40	6.23	4.67
D_2C_3	1.45	13.67	4.43	204.72	4.71	3.53
D_2C_4	1.62	13.00	4.37	204.53	5.25	3.94
D_2C_5	1.80	14.67	4.90	215.40	6.15	4.62
D_3C_1	1.45	13.67	4.50	194.20	4.47	3.35
D_3C_2	1.62	14.33	4.75	207.35	5.33	4.00
D_3C_3	1.38	13.00	4.30	192.92	4.23	3.17
D_3C_4	1.55	12.67	4.20	177.00	4.35	3.27
D_3C_5	1.73	14.00	4.70	202.75	5.57	4.18
CD (p= 0.05)	0.009	0.63	0.07	8.75	0.20	0.15

April) and HQPM1 variety recorded significantly lower incidence of fall army worm at all stages of crop growth compared to early sowing. Therefore, it can be suggested that manipulating the sowing date and growing of tolerant variety of maize as HQPM1 can be an effective measure to manage exotic army worm infestation. Mid sowing of maize (21st March) and growing of local cultivars such as Sipho observed significantly better yield attributes which will ensure higher economic returns to the farmers.

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A new species of *Chalcis* Fabricius (Hymenoptera, Chalcididae) from south India

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ABSTRACT: The morphologically diverse and cosmopolitan chalcidid genus, *Chalcis* includes 59 species. Most of the species are distributed in the temperate regions of the Northern Hemisphere. Till date only two species were recorded from the Oriental region. A new species, *Chalcis biligiriensis* from scrub forests of Biligiri Rangaswamy Temple Tiger Reserve (BRT) of the Western Ghats (Karnataka, India) is described. In addition, detailed illustrations of *C. gibsoni* Narendran for the first time along with an illustrated key to the identification of the Oriental species are provided. © 2023 Association for Advancement of Entomology

KEYWORDS: Western Ghats, Chalcidoidea, taxonomic key, BR Hills, Oriental Region, malaise trap

The family Chalcididae (Hymenoptera, Chalcidoidea) includes more than 1500 species in over 90 genera globally (Noyes, 2023). This family contains eight subfamilies and nine tribes (Cruaud *et al*., 2020). The genus *Chalcis* belongs to the subfamily Chalcidinae (Bouček, 1988), contains 59 species (Saguiah *et al*., 2020; Noyes, 2023). Even though the natural history of *Chalcis* is not yet explored much, species were recorded as egg-larval or larval-pupal primary parasitoids of Stratiomyidae (Diptera) (Müller, 1908; Burks, 1940; Schremmer, 1960). Though species of *Chalcis* morphologically diverse, they can be recognized by the combination of following characters; mesotibial spur reduced or absent, tarsal claws only slightly curved in both male and female, pectinate basally in male, medial portion of female hypopygium sclerotized and most often extending posteriorly and male hypopygium enlarged and posteriorly emarginated (Delvare, 1992).

No taxonomic revision of *Chalcis* is carried out in a global scale. Literature about species revisions were available for the following regions; North America (Burks, 1940, 1977), New World (Delvare, 1992), Europe (Bouček 1951), Iran (Lotfalizadeh *et al*., 2012), Oriental region (Narendran, 1989), Japan (Habu, 1960), and Australasia (Bouček, 1988). The highest species diversity is recorded in the New World (Saguiah *et al*., 2020). Only two species were reported from the Oriental region; one from India and another from Taiwan (Narendran, 1989). In the present study, the third species of *Chalcis* is described with illustrations from the Oriental region. A key to the Oriental species is provided along with the illustration of *Chalcis gibsoni* Narendran.

Holotype was collected through the Malaise trap from Biligiri Rangaswamy Temple Tiger Reserve, Karnataka. After sorting the specimen was preserved

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in ethyl alcohol and later card mounted. Images of the holotype were taken with Keyence VHX-6000 and edited in Adobe Photoshop CS8. Holotype is deposited in the collection of ATREE Insect Museum, Bengaluru, India (AIMB). Morphological terminology followed Hymenoptera Anatomy Ontology (Yoder *et al*., 2010; http://glossary.hymao.org). The terminology for body sculpture follows Harris (1979).

Abbreviations used

AIMB: ATREE Insect Museum, Bengaluru, India

Fu1, Fu2: Funicular segments 1 and 2

MOD: Maximum diameter of median ocellus

OOL: Ocular-ocellar line or minimum distance between a lateral ocellus and eye margin

POL: Posterior ocellar line or minimal distance between lateral ocelli

SMV: Submarginal vein

MV: Marginal vein

PMV: Postmarginal vein

Chalcis Fabricius, 1787

Chalcis Fabricius, 1787: 272. Type species *Sphex sispes* Linnaeus, 1761, by subsequent designation of Westwood (1839: 65).

Smiera Spinola, 1811: 147. Type species *Sphex sispes* Linnaeus, 1761, by subsequent designation of Curtis (1833: 472). Synonymy by Gahan & Fagan (1923: 31).

Smicra Spinola, 1837: 1. Unjustified emendation of *Smiera*.

Diagnosis: Scape distinctly exceeding median ocellus (Figs. 1B, 4C); mandibular formula 2:3 or 3:3, the upper tooth larger and longer than the others; pre and post orbital carinae indistinct (Figs. 1B, 4C); frontogenal sulcus often present rarely absent (Figs. 1B, 4C); propodeum often with midlongitudinal carina (Figs. 1E, 5B); mesocoxa with short pubescence dorsolaterally (Fig. 1A); mesotibial spur at most as long as apical width of mesotibia, occasionally absent; metafemur with basal tooth long in most species or short in Oriental species (except in *C. gibsoni*) (Figs. 1A, 2C, 3A, 4A, 5C); metafemur mostly with (Figs. 3A, 5D) or without inner basal tooth (in *C. edentata*); tarsal claws usually slightly curved, sometimes falcate in female, apically bifid in males (Fig. 3B); petiole with dorsal tubercle like projection anteriorly and posteriorly (Figs. 1A, 2A, B, 4B, 5E); female hypopygium with median portion narrowly extended posteriorly with median portion distinct from the lateral areas, or thickened but only slightly extended posteriorly beyond the adjacent margins; apex of ovipositor with long setae; male hypopygium enlarged, flat or concave, with distal margin truncate to notched.

***Biology*:** Egg-larval or larval-pupal primary parasitoids of Stratiomyidae.

***Distribution*:** Cosmopolitan.

Key to Oriental species of *Chalcis* Fabricius

1. Hind femur fully black, without inner basal tooth; Gaster including petiole distinctly longer than thorax; head 1.4 × as wide as long; petiole with tubercle like projection anteriorly [Taiwan].....*Chalcis edentata* Narendran

- Hind femur with yellow spots basally and subapically (Figs. 1A, 2C, 3A, 4A, 5C), with distinct inner basal tooth (Figs. 3A, 5D); Gaster including petiole as long as thorax (Figs. 1A, 4A); head less than 1.4 × as wide as long (Figs. 1B, 4C); petiole with tubercle like projection anteriorly and posteriorly (Figs. 1A, 2A, B, 4B, 5E).....2

2. First tooth of outer ventral margin of hind femur long (Fig. 5C); propodeum without midlongitudinal carina basally (Fig. 5B); petiole long 4.2 × as long as maximum width (Fig. 5E); post marginal vein longer than marginal vein (Fig. 4B); second gastral tergite with three or more rows of setae (Figs. 4A, 5E) [India].....*Chalcis gibsoni* Narendran

- First tooth of outer ventral margin of hind femur short (Fig. 2C); propodeum with a short midlongitudinal carina basally (Fig. 1E); petiole short, 2.6 × as long as maximum width

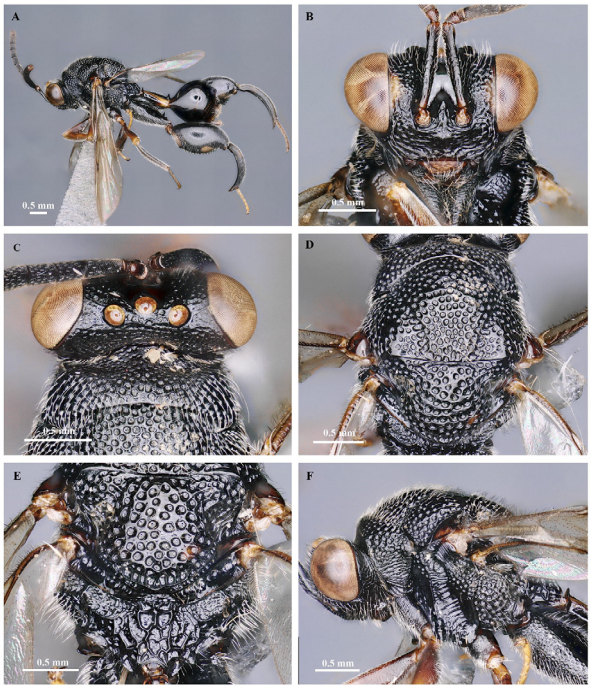

Fig. 1 *Chalcis biligiriensis* **sp. nov.**, holotype, female- A) Habitus, in lateral view; B) Head, in anterior view; C) Head, in dorsal view; D) Mesosoma, in dorsal view; E) Scutellum and propodeum, in dorsal view; F) Head and mesosoma, in lateral view

Fig. 2 *Chalcis biligiriensis* **sp. nov.**, holotype, female - A) Gaster, in lateral view; B) Gaster, in dorsal view; C) Hind femora, in lateral view; D) Fore wing

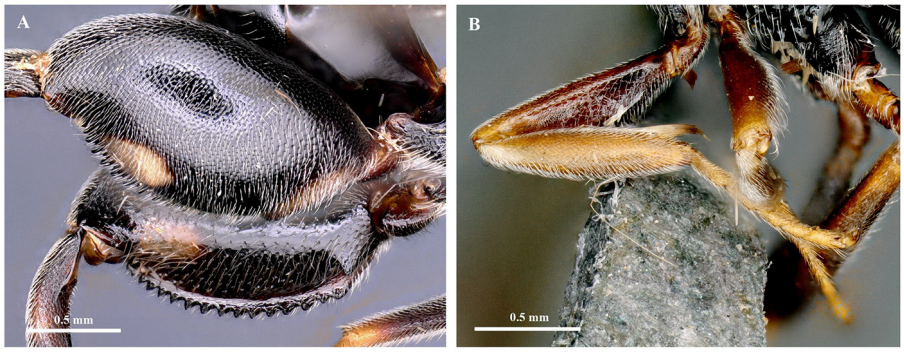

Fig. 3 *Chalcis biligiriensis* **sp. nov.**, holotype, female - A) Hind femora, in ventro-lateral view; B) Fore leg

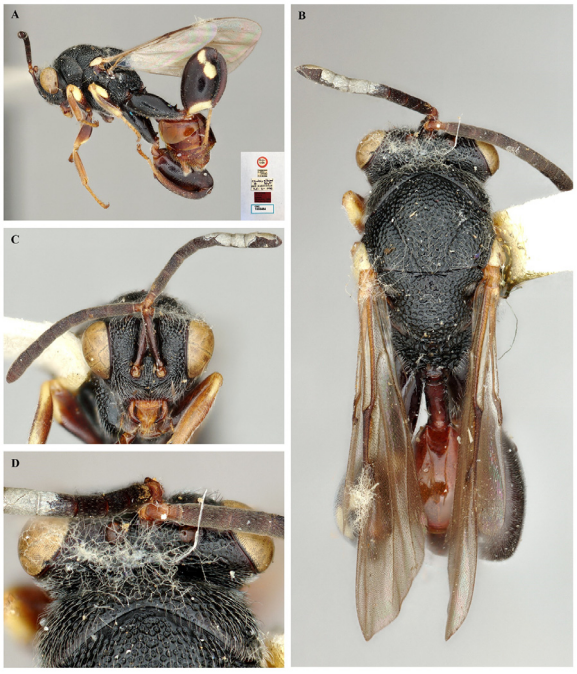

Fig. 4 *Chalcis gibsoni* Narendran, holotype, female - A) Habitus, in lateral view; B) Habitus, in dorsal view; C) Head, in anterior view; D) Head, in dorsal view

Fig. 5 *Chalcis gibsoni* Narendran, holotype, female - A) Head and mesosoma, in lateral view; B) Propodeum, in dorsal view; C) Hind femora, in lateral view; D) Hind femora, in ventro-lateral view; E) Gaster, in dorsal view

(Fig. 2B); post marginal vein shorter than marginal vein (Fig. 2D); second gastral tergite with single row of setae (Figs 1A, 2A) [India] .....*Chalcis biligiriensis* sp. nov.

***Chalcis biligiriensis* sp. nov. (Figs. 1–3)**

LSIDurn:lsid:zoobank.org:act:F49C0679-FBA4-4161-BFF6-CA06F043B972

Material examined.

Holotype. Female. INDIA: Karnataka, Chamarajanagar, Biligiri Rangaswamy Temple Tiger Reserve, scrub jungle, 77°06 55.1 E, 12°01 41.4 N; 31.v–15.vi.2005, Malaise trap, coll. D.R. Priyadarsanan.

Description. Female, holotype. Body length 5.4 mm.

Head. Lower face slightly bulging above clypeus, with shallow umbilicate fovea, interspaces broad and finely granulate; parascrobal area foveolate, interspaces moderately broad, smooth; median intumescence present; malar space 0.4 × eye height; malar sulcus distinct, straight, crenulated; gena smooth to rugulose, genal carina present; mandibular formula 2:3; antennal scrobe smooth, shiny and without transverse carina below median ocellus; interantennal projection without longitudinal carina anteriorly, with shallow narrow groove, posteriorly with more or less elongated triangular area; MOD:POL:OOL = 1: 2: 1. Scape 6.0 × as long as wide, inner face slightly sinuous; anellus about 0.5 × as long as wide; Fu1 longer than other funicular segments, about 2.5 × as long as wide and 1.4 × as long as Fu2 length.

Mesosoma. Mesoscutum sparsely setose, interspace mostly smooth and shiny, broad medially, 1.0 × diameter of umbilicate foveae; mesoscutellum convex, interspaces broad and smooth anteriorly, narrow and granulate posteriorly, frenal carina acutely convex medially, blade-like; mesopleuron with mesepisternum longitudinally striate dorsally, foveolate ventrally; propodeum rugose antero-

laterally with irregular median and submedian carinae, anterior and posterior costulae irregular, adpetiolar area with irregular fovea, anterosubmedial area irregularly areolate-rugose; tarsomeres 4 and 5 of all legs with pubescence similar to basal tarsomeres; protarsomeres 4 and 5 ventrally with pairs of distinct peg-like spines; protarsal claws falcate, ventrally with 5 basal spines followed by 1 wide and blunt tooth; mesocoxa sparsely setose posteriorly; mesotibial spur 0.7× as long as the width of the mesotibial apex; metacoxa smooth and shiny dorsally, puncticulate and interstices smooth and shiny ventrally, inner face glabrous; metafemur with outer face puncticulate and interstices smooth and shiny, ventrally without inner basal tooth but with 16 teeth along outer margin, outer basal tooth at most as large as and close to second tooth, the following teeth equal in size; metatibia with apical spine triangular and longer than the apical width of metatibia; metatarsomere 1. shorter than tarsomeres 2. Fore wing SMV:MV:PMV = 2.3: 1: 1.

Metasoma. Petiole about 2.6 × as long as wide, cylindrical, with dorsolateral carina at most along basal half, with a short pair of submedian carina basally, with dorsal tubercle like projection anteriorly and posteriorly, anterior projection pointed and posterior projection blunt in lateral view; gastral tergites smooth; second gastral tergite sparsely punctate submedially; apex ovipositor sheath rounded apically, with long setae.

Colour: Black, except eye, parascrobal area medially, ocelli, fore tibia basally, fore tarsus, metatibia apically, mid tarsus, posterior part of hind femur basally and subapically, outer face of hind tarsus, petiole dorsally, apico-laterally, ovipositor apically yellow.

Male. Unknown;

Biology. Unknown.

Distribution. India (Karnataka).

Comparative diagnosis: Apart from the differences mentioned in the key, the new species can be distinguished from *C. gibsoni* in having petiole yellow dorsally (reddish brown dorsally in *C. gibsoni*), apical half of fore and mid femora yellowish brown (yellow in *C. gibsoni*); gaster black (reddish brown in *C. gibsoni*), metafemur with 17 ventral teeth (with 19 ventral teeth in *C. gibsoni*), tegula reddish brown (yellow in *C. gibsoni* (Fig. 5A)), frons with a pair of yellow spots laterally (frons without yellow spots in *C. gibsoni*), POL 2.0 × as long as OOL (POL 1.6 × as long as OOL in *C. gibsoni* (Fig. 4D)), hind femora without yellow spot apico-dorsally (with yellow spot apico-dorsally in *C. gibsoni*).

Etymology: The species is named after the type locality, Biligiri Rangaswamy Temple Tiger Reserve, Karnataka, India.

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Invitro screening of leaf extracts of selected plants from Lamiaceae, Asteraceae and Fabaceae for mortality and repellency to *Odoiporus longicollis* (Olivier)

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ABSTRACT: A study was focused on biocidal and repellent efficiencies of leaf extracts of *Chromolaena odorata* (L) (Asteraceae), *Gliricidia sepium* (Jacq.) (Fabaceae), *Coleus aromaticus* Benth, *Hyptis suaveolens* (L.)Poit. (Lamiaceae) and *Artemisia vulgaris* L. (Asteraceae), against *Odoiporus longicollis*, banana pseudostem weevil. Ethanol leaf extracts of the above plants were primarily analysed for the mortality and repellency activity, the results showed that among the extracts *G. sepium* was most effective. Further *G. sepium* in various solvents like petroleum ether, chloroform, ethyl acetate, ethanol and water were analysed for the biocidal and repellency assays, and found that, the ethyl acetate solvent of *G. sepium* elicited maximum effectiveness. The ethyl acetate extract of *G. sepium* is found as a promising biopesticide.

KEY WORDS: *Gliricidia sepium,* ethyl acetate extract, biopesticide, banana stem weevil

Odoiporus longicollis (Olivier) (Coleoptera, Curculionidae), the banana pseudostem weevil (BPW) is a harmful pest which attacks banana plantations in South-East Asia and all the banana growing belts of India (Padmanaban and Sathiamoorthy, 2001). Due to the attack of this pest, the plant eventually becomes fragile resulting in premature falling due to tunnels made by the grubs in stems (Ravi and Palaniswami, 2002). Prasuna *et al.* (2008) reported that *O. longicollis* causes 10–90 per cent yield loss depending on the intensity of pest attack and management efficiency.

It is desirable to develop an appropriate biopesticide to control *O. longicollis*. The present investigation evaluated the impact of leaf extracts of five selected plants having aromatic properties such as *Chromolaena odorata* (L) (Asteraceae), *Gliricidia sepium* (Jacq.) (Fabaceae), *Coleus aromaticus* Benth, *Hyptis suaveolens* (L.)Poit. (Lamiaceae) and *Artemisia vulgaris* L. (Asteraceae), for their biocidal and repellent efficiencies against *O. longicollis*. Having higher efficacy, Ethyl acetate extract of *G.sepium* leaves was chosen for detailed studies as well as to isolate and characterize the active component. Adults as well as the fourth instar larvae were evaluated for their repellency activities. Biocidal efficiency

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studies of *C. odorata*, *G. sepium*, *C. aromaticus, H. suaveolens* and *A. vulgaris* on *O. longicollis,* had not been conducted earlier and hence exploration in this direction was found to be worthwhile.

O. longicollis adults were collected from the banana fields of Aruvippuram, Thiruvananthapuram district, Kerala, India. The weevils were reared in plastic containers (30cm × 20cm) along with pseudostem pieces of size 10cm × 10cm. Pseudostem accommodating eggs were reared in separate containers and the newly hatched first instar larvae were transferred onto different containers. Fresh pseudostem pieces weighing 1, 2, 3 and 4g were provided respectively for the first, second, third and fourth instars and these pseudostem pieces were regularly replaced on the third day. Mortality and repellency assays were performed using eight insects each in control and treated cultures. Each experiment was performed in eight replicates.

Leaves of *C. odorata, G. sepium, C. aromaticus, H. suaveolens and A. vulgaris* were collected from Aruvippuram, Thiruvananthapuram district, Kerala, India (8.4245° N; 77.0982° E). The plants were identified, leaves were washed thoroughly with fresh water and shade dried for 14 days. Soxhlet extraction of these leaves were done using ethanol as solvent. Serial extraction of the best effective plant leaves were done in various solvents like petroleum ether, chloroform, ethyl acetate, ethanol and water were analysed for the mortality and repellency assay to select the best solvent.

Table 1. LC 50 (mg ml^{-1}) values of selected plant extracts. (LCL: 95% lower confidence limit, UCL: 95% upper confidence limit)

No	Plant	LC_{50}	LCL	UCL
1	*Artemisia vulgaris*	519.874	438.164	726.493
2	*Coleus aromaticus*	407.249	348.283	532.345
3	*Gliricidia sepium*	149.705	110.403	178.565
4	*Chromolaena odorata*	382.238	346.464	438.172
5	*Hyptis suaveolens*	286.273	250.637	329.365

Fourth instar larvae of *O. longicollis* were selected for the experiment. Each larva was provided with pseudostem piece of weight 4g as food. The different plant extracts in ethanol solvent with varying doses of 100, 200, 300 and 400 mg ml^{-1} were separately spread on the pseudostem pieces. Insects devoid of any plant extract treatment were used as the control group. During the experiment, the pseudostem pieces were replaced every third day. Mortality of the larvae was noted after 12, 24, 48 and 72 h respectively. The *G.sepium* leaf extracts in various solvents with varying doses of 100, 200, 300 and 400 mg ml^{-1} were also analysed for the mortality assay.

Repellency assay was conducted using Whatman No1 filter paper with a diameter of 150mm. The filter papers were cut into two halves, followed by the uniform application of 1 mg ml^{-1} of the ethanol plant leaf extract of selected plants on one half. The other half was treated with ethanol alone and both halves were air-dried to evaporate the solvent. After complete evaporation, both halves were re-made into the original disc shape by using cello tape. The discs were then placed in petri dishes followed by the release of larvae at the centre.

Results obtained from mortality and repellency were corrected using Abott's correction formula (Abott, 1925). Statistical analysis of the results was performed by the statistical software SPSS V.23 for windows using Tukey's test and one way ANOVA with Duncan's post hoc pairwise comparisons.

Probit analysis of the leaves showed LC_{50} values of 519.874, 407.249, 149.705, 382.238 and 286.273 mg ml^{-1} respectively for the plants *A. vulgaris*, *C. aromaticus*, *G. sepium*, *C. odorata* and

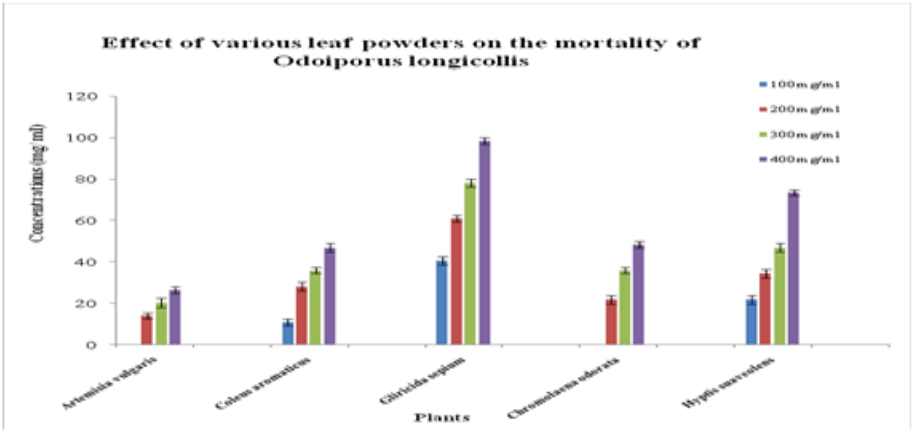

Fig. 1 Effect of various leaf powders on the mortality of 4th instar larvae of *O. longicollis*

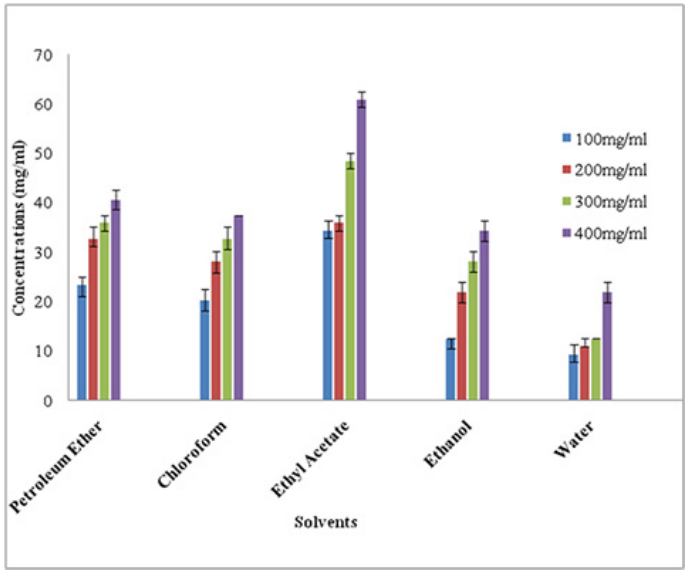

Fig. 2 Effect of various solvent leaf extracts of *G. sepium* on the mortality of fourth instar larvae of *O. longicollis*

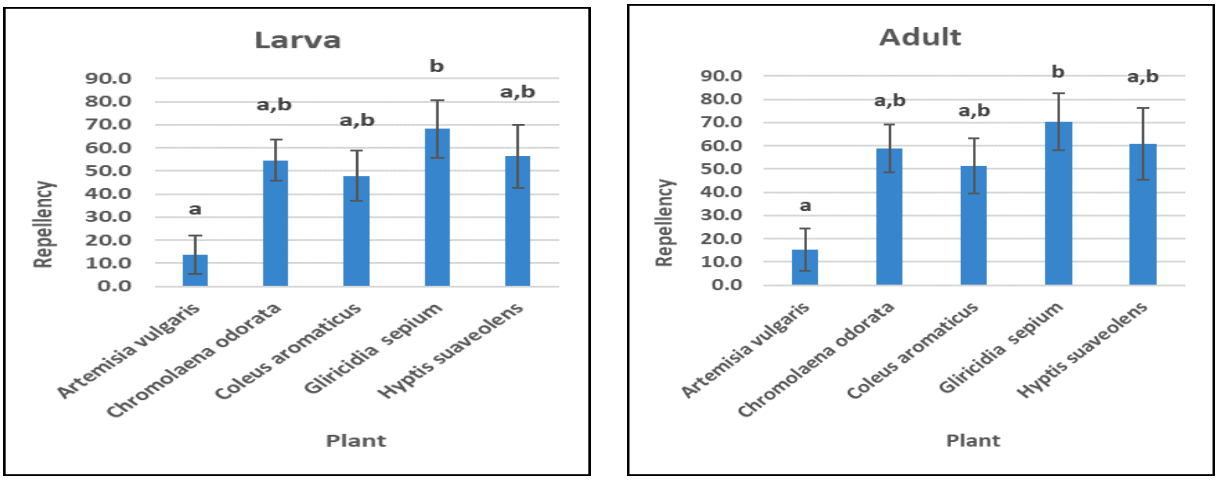

Fig. 3 Repellency activity of plant leaf extracts on the larvae and adults of *O. longicollis* (Different superscript letters in the graph indicate statistical significance at 5% level)

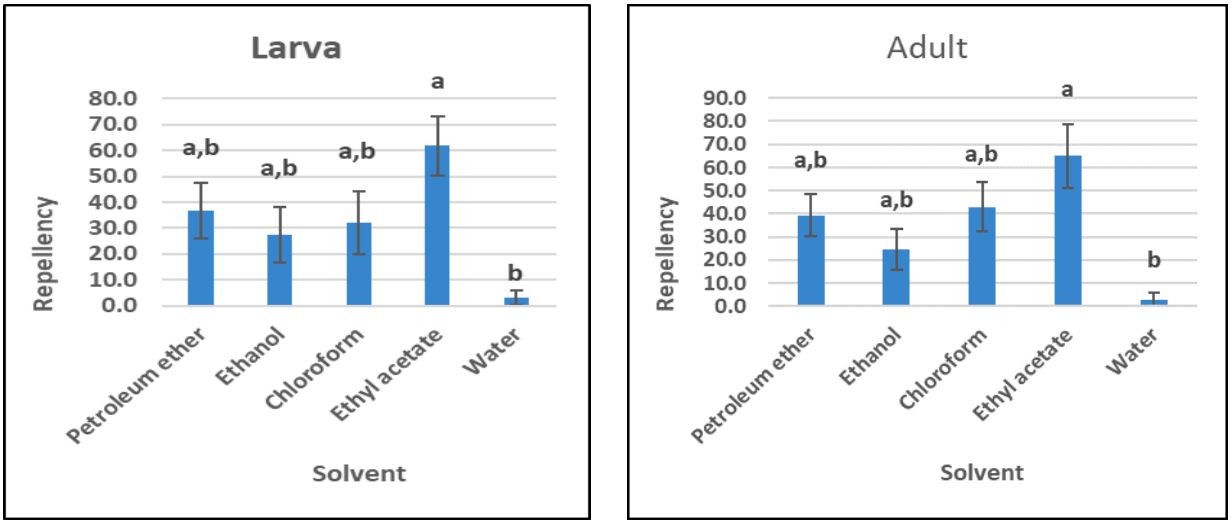

Fig. 4 Repellency activity of different solvent leaf extracts of *G. sepium* on *O. longicollis* (Different superscript letters in the graph indicate statistical significance at 5% level)

H. suaveolens (Table 1). Highest percentage of mortality with *G. sepium* leaf extract followed by *H. suaveolens, C. odorata, C. aromaticus and A. vulgaris*.

In the experiment on the effect of leaf powders on the mortality of 4th instar larvae of *O. longicollis*. *G. sepium* was found to elicit the maximum impact followed by *H. suaveolens*, *C. odorata*, *C. aromaticus* and *A. vulgaris* (Fig. 1).

In the leaf extract experiment maximum mortality was noted in the ethyl acetate solvent leaf extract of *G. sepium* followed by petroleum ether, chloroform, ethanol and water respectively (Fig. 2). Probit analysis of the different solvents revealed LC_{50} of solvent petroleum ether as 808.013 mg ml^{-1}, for chloroform 863.338, ethyl acetate 292.661, ethanol 982.994.

Among the different extracts, the ethyl acetate solvent extract exhibited maximum repellency followed by petroleum ether, chloroform and ethanol respectively. *G. sepium* exhibited maximum repellency, followed by *H. suaveolens*, *C. odorata*, *C. aromaticus* and *A. vulgaris*. Repellency to larva is statistically significant against the selected plants ($F_{4, 15}$ = 3.483, p value = 0.033) and in adult is also significant against the selected plants as determined by one-way ANOVA ($F_{4, 15}$ = 3.138, p value = 0.046). From Tukey's test, plant repellency showed maximum significance between *Artemisia vulgaris* and *G. sepium* (Fig. 3)

Solvent repellency in larva and adult was significant ($F_{4, 15}$ = 4.294, p value = 0.016 for larva, $F_{4, 15}$ = 5.586, p value = 0.006 for adult). The Tukey post hoc multiple comparison test shows that the solvent repellency has maximum significance between ethyl acetate and water (Fig. 4).

A. vulgaris, C. odorata, G. sepium, C. aromaticus and *H. suaveolens* elicited biocidal and repellency effects on *O. longicollis*. However, the maximum impact was displayed by the ethanol extract of *G. sepium* that the primary and secondary metabolites eluted out maximum in ethanol solvents.

The management of banana weevil is done primarily by the application of insecticides (Collins *et al*., 1991). Biological agents are an important alternative to minimize or replace the use of synthetic pesticides (David, 2008). *C. infortunatum, L. camara* and *C. alata* elicited biocidal activity in pseudostem weevil, of which *C. infortunatum* was found to be the most effective (Remya and Dayanandan, 2019).

Stem injection of monocrotophos, Azadirachtin and the application of *Beuveria bassiana* recorded the highest percent mortality (Irulandi *et al.,* 2012). Essential oils from stems of *T. purpurea* and *I. carnea* could be explored as natural repellents for the control of *O. longicollis* (Sahayaraj *et al.*, 2015).

The presence of phytochemicals in the ethanol extract of *G. sepium* elicited antibacterial activity (Ajaieoba, 2002). The potency of *G. sepium* was due to the presence of saponins, phenolic compounds, essential oils, and flavonoids (Akharay *et al.,* 2012).The phytochemical analysis *of C. odorata* showed the presence of tannins, saponins, flavanoids and alkaloids in the leaves of *C. odorata* which indicates larvicidal efficacy (Man, 2013). *G. sepium* leaf and flowers contains forty-two known compounds. Of these sixteen have been identified and quantified from the flower essential oil by GC-MS analysis (Kaniampady *et al.,* 2007). The essential oil from the leaves of *G. sepium* from Columbia was analysed by GC-FID and GC-MS (Celis *et al*., 2015). The major components are methyl-3(E)-pentenyl ether (11.55%), 3-methyl-2-butanol (10.65%), 3-methoxy hexane (10.14%), 1-(1-ethoxyethoxy)-2-hexene (9.72%), 2- decanol (8.97%), coumarin (8.07%) and hexadecanoic acid (5.16%) (Reddy *et al*., 2010).

Based on previous reports, natural agents which have the potential to elicit repellency and mortality in both the larvae and adult populations of weevils could be effectively developed as biopesticides. The present findings demonstrated significant repellency and mortality in both the larvae and adult populations of *O. longicollis*.

Ethyl acetate extract of *G. sepium* is identified as a potent pesticide against *O. longicollis* is a preliminary indication.

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Population dynamics of southern birdwing (*Troides minos* Cramer, 1779) in Sirumalai Reserve Forest, Eastern Ghats, India

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ABSTRACT: Southern birdwing *Troides minos* (Cramer, [1779]), a large swallow tail butterfly, state butterfly of Karnataka, is endemic to south India. *T. minos* was listed in CITES (Convention on International Trade in Endangered species of wild fauna and flora) and is of high conservation priority. IUCN also recommends close monitoring of *T. minos*. The population trend of *T. minos*, in Sirumalai Reserve Forest, Eastern Ghats was monitored for the period of twenty six months using the line transect method. Higher prevalence of *T. minos* butterflies were observed and recorded over the study. This study highlights the abundance of *T. minos* in certain months of the study period and peak abundance during the post monsoon period.

KEY WORDS: Papilionidae, swallow tail, abundance, IUCN, CITES, conservation

Papilionidae, the smallest butterfly family constitutes 550 species and is distributed worldwide (Kunte, 2020). India harbors 107 species of Papilionidae; amongst that, peninsular India has only 19 species. India occupies the 6th rank in a list of countries suitable for swallowtail conservation (Kehimkar, 2016). *Troides minos* (Cramer, [1779]), a striking swallowtail butterfly is the second largest butterfly in India with a wing span of 140-190mm. Members of Papilionidae are conspicuously large in size with similar body forms with long and slender legs, round and narrow head with black eyes and coiled proboscis. They also have a large thorax and even larger abdomen with contrasting longitudinal lines. Both wing surfaces have similar markings with subtle differences (Powell, 2009).

Most of the papilionidae species are forest dwelling species, except a few known to be associated with open habitats. *Troides* species are also one of the forest dependent species (Rajeswari and Jeyabalan, 2017). In previous studies, *T. minos* was declared as endemic to Western Ghats (Goankar, 1996). But in later studies it was confirmed that Southern

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birdwing is also present in few regions of Eastern Ghats (Sharmila and Thatheyus, 2014; Ponraman *et al*., 2015; Sundar rajan *et al*., 2016). IUCN (1990) recommends close monitoring of this species. In the present study, prevalence of *T.minos* was examined for twenty six months in Sirumalai Reserve Forest, Tamil Nadu, India.

Sirumalai is a dense forest region of about 60,000 acres, situated between 10°07' - 10°18' N latitude and 77°55' - 78°12' E longitude; Much of the Sirumalai is covered by deciduous forest and the lower slopes are covered by dry deciduous. In the present study Sirumalai Reserve Forest area was divided into eight transects, site 1 (Konganuthu), site 2 (Puli sathuodai), site 3 (Ulkombai saragam), site 4 (Kannimarkovil), site 5 (Vellode), site 6 (Kuranguthopu), site 7 (Ambathur beat), site 8 (Kadaman solai), based on different altitudes. Line transect sampling method (Pollard and Yates, 1993) was followed in the eight sites for monitoring the prevalence of *T.minos* within the forest reserve. Identification of *T. minos* was done by using standard keys from literature (Wynter-Blyth, 1957; Haribal, 1992). The influence of microclimatic factors like temperature, relative humidity, light intensity and wind speed were noted time to time during the observation period (Kunte, 1997).

Prevalence of *T. minos* was observed for a period of twenty six months from September 2020 to October 2022 and four hundred and fifty numbers of *T. minos* butterflies were observed. *T. minos* was more prevalent during the post monsoon months (October, November and December) of 2020, and *T. minos* population was absent during the months of summer (April, May and June) in 2021 (Fig.1). Relative abundance of *T.minos* was distributed irregularly throughout the year, but the greatest number of individuals were observed and recorded in post monsoon season. The uncertain species abundance may be due to the environmental stochasticity.

Climatic constancy is important for perseverance of butterfly population (Roy *et al.,* 2001). The underlying correlation between microclimatic factors and abundance of *T. minos* in Sirumalai Reserve Forest was analyzed by using Pearson correlation coefficient in PAST statistical software. The environmental factors humidity (r^2= 0.1598) and wind speed (r^2=0.25389) are positively correlated with *T. minos* abundance in Sirumalai Reserve Forest, whereas, temperature (r^2= -0.426) and light intensity (r^2= -0.3360) are negatively correlated (Table.1).

The effect of seasonal change is evident on the vegetation. The availability of host plants is the clear pre-requisite for the survival of the species. *Troides* species larvae are exclusive feeders of *Aristolochia* (Yao, 2015). *Aristolochia* species are one of the important plants, suitable for larval food for *Troides* sp and contain Aristolochic acids, causing both the adult and larva to be unpalatable to predators (Mebs and Scheinder, 2002). *Lantana camara*, an invasive species of Indian subcontinent, was the most prevalent nectar plant for *T.minos* in the study area. The greater availability of larval host plants and adult nectar sources at the season

Table 1. Pearson's Correlation coefficient between microclimatic factors and abundance of *Troides minos* in Sirumalai Reserve Forest

Microclimatic fators	Temp (^{0}C)	Humidity (%)	Wind speed(m/s)	Light intensity(lux)	Density of *T. minos*
Temperature (^{0}C)		-0.14741	-0.38006	0.46578	-0.42609
Humidity (%)	-0.14741		0.27154	-0.536	0.15989
Wind speed (m/s)	-0.38006	0.27154		-0.68414	0.25381
Light intensity (lux)	0.46578	-0.536	-0.68414		-0.33606
Density of *T. minos*	-0.42609	0.15989	0.25381	-0.33606	

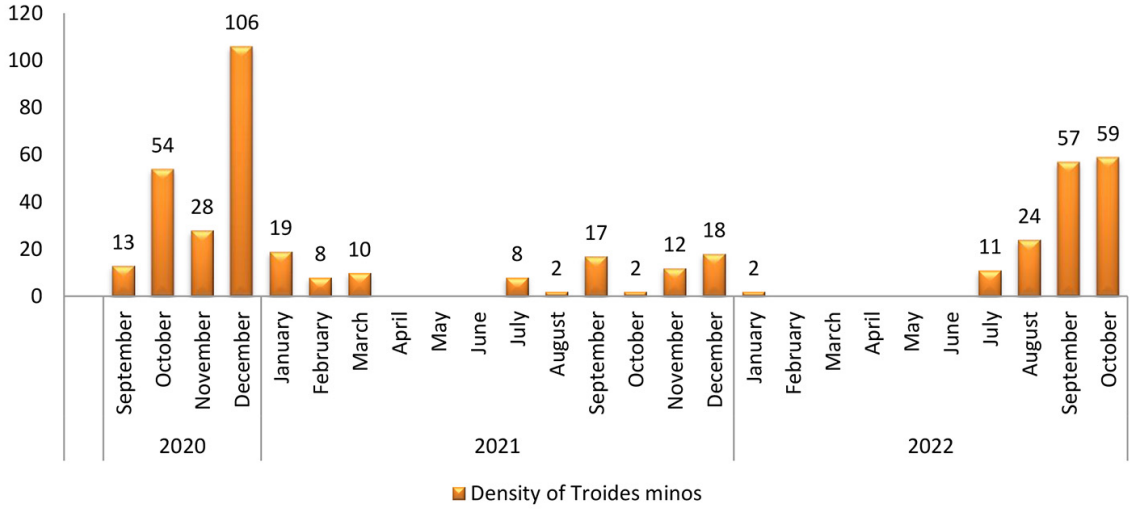

Fig.1 Prevalence of *Troides minos* during the study period in Sirumalai Reserve Forest

of wetter years may be the reason for migrants to replenish (Hu *et al*., 2021). Different butterfly species prefer different altitudes based on the availability of food source. Based on the study, *T.minos* prefers medium or higher altitude. Thick canopy in higher altitudes protects the butterflies from extreme conditions and increases its fitness (Pellet *et al*., 2012).

Population dynamics of Southern birdwing seems to be counter intuitive (Hu *et al.,* 2021). Dispersal of butterflies has an immense importance in setting up of population dynamics. Southern birdwing population might be prevalent in the study area due migration from some other areas. Studying other aspects of butterfly biology contributes to its conservation (Devi *et al*. 2021; Sharmila *et al*., 2022; Archana *et al*., 2022). Conservation measures are needed to save the species of *T. minos*. Encouraging plantations of suitable host and nectar plants within the forest reserve will help to augment the number of *T.minos* butterflies in Sirumalai Reserve Forest and in addition it will also help to improve ecotourism.

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A check list of blow fly fauna (Diptera, Calliphoridae) of Kerala including forensically significant species

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ABSTRACT: Seventeen species of blow fly belonging to four subfamilies and eight genera were recorded from Kerala based on field studies and literature. Out of the 17 species, seven are forensically significant, six are pollinators and two species each are carrion breeders and termite predators respectively. The distinguishing features and distribution of all the species are discussed.

KEYWORDS: Western Ghats, south India, Bengaliinae, Luciliinae, Chrysomyinae, Rhiniinae

Blow flies belonging to family Calliphoridae is encompassed of a group of flies having veterinary, ecological, medical and forensic significance with worldwide distribution. Currently, 1760 species of blow flies were reported (Bánki *et al.*, 2023) from all over the world. In India, the family is represented by nine subfamilies, 30 genera and 119 species (Nandi, 2004; Bharti, 2011). The subfamilies included are; Melanomyinae, Calliphorinae, Bengaliinae, Luciliinae, Rhiniinae, Helicoboscinae, Chrysomyinae, Ameniinae and Polleniinae.

Extensive work on the taxonomy of blow flies in India was done by Senior-White *et al.* (1940). One of the most significant contributions to the blow fly fauna of India was that of Nandi (2004). An updated checklist of blow flies in India was prepared by Bharti (2011) in which 119 blow fly species belonging to 30 genera were listed. Various aspects of biology (Subramanian and Mohan, 1980), ecology (Radhakrishnan *et al.,* 2012), new reports (Bharti *et al.,* 2014) and molecular identification (Bharti and Singh, 2017) of blow flies were studied. In Kerala, morphological and molecular identification of blow flies; *Chrysomya chani* (GenBank accession no: MW600494.1), *Chrysomya rufifacies* (GenBank accession No: OM019083.1), *Hemipyrellia ligurriens* (GenBank, accession no: MN831480.1) and *Chrysomya megacephala* (GenBank accession No: MW 522614) were done by Reject Paul and Binoy (2021, 2022).

The checklist was prepared based on field studies and literature survey (Senior-White *et al*., 1940; Nandi, 2004; Bharti, 2011; Bharti *et al.,* 2014; Bharti and Singh, 2017). A total of seventeen species belonging to four subfamilies and eight genera were recorded from Kerala. The description and distribution of these species with references (Senior-White *et al*., 1940, Bharti *et al.,* 2014) are discussed.

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Subfamily: Bengaliinae

Genus: *Bengalia* Robineau- Desvoidy, 1830

1. *Bengalia jejuna* (Fabricius, 1787)

Diagnosis: Absence of concavity on the posterior margin of eye, upper part of mesopimeron with 9-11 black setulae, broad cercus narrowing down to pointed tip, 3rd and 4th tergite with marginal bands, bacilliform sclerite with an oblique distal margin, distiphallus with a constriction at the middle of dorsal wall, broad distal lip process with broad wing like membranes.

Distribution: Kochi, Walayar, Thiruvananthapuram (Bharti, 2011)

2. *Bengalia surcoufi* (Senior-White, 1924)

Diagnosis: Brownish grey parafrontalia, first and second antennal segments reddish brown in colour, pale palpi with black bristles, all tergites black banded, pale yellowish squamae, tarsi tips are darkened.

Distribution: Kochi (Nandi, 2004)

Subfamily: Luciliinae

Genus: *Hemipyrellia,* Townsend, 1917

3. *Hemipyrellia ligurriens* (Wiedemann, 1830)

Diagnosis: Genae and parafrontalia silver white in colour, antennae tawny yellow to brownish in colour, orange palpi, short hairs on the edges of tergites and first visible sternite, bare stem vein, 1st longitudinal vein without any setulae, 3rd longitudinal vein with short setulae on dorsal and ventral aspects, upper squama with creamish white short cilia and lower squama with light brown cilia. Eyes were holoptic in males and dichoptic in females. The length of the third antennal segment is shorter than the distance between the eyes in males. In males, the parafrontalia was covered with silver white tomentum. In females, the frontal stripe is broader at the middle of frons than in male fly. In males, sparse short hairs are seen on the edges of tergites and first visible sternite.

Additional material examined: 5 males; 8 females; Collected and Identified by: Reject Paul M.P., Location: Thrissur (Kolangattukara; 10°34’29.4"N; 76°11’01.8"E), Palakkad (Vaniamkulam-II-10°45’32.1"N; 76°20’04.8"E), Ernakulam (Aimuri-10°08’52.8"N; 76°29’18.5"E).

Repository: Dept. of Zoology, St. Thomas’ College (Autonomous), Thrissur, Kerala.

Distribution: Foot of Nilgiri hills, Thrissur, Palakkad and Ernalulam, Kerala (Nandi, 2004; Reject Paul and Binoy, 2021).

Remarks: *Hemipyrellia ligurriens* is a forensically significant blow fly and in the current investigation, it was found to get attracted to decomposing pork meat and completing its life cycle.

Genus: *Lucilia* Robineau-Desvoidy, 1830

4. *Lucilia ampullacea* (Villeneuve, 1922)

Diagnosis: Third to fifth tergites without marginal band, basicostal scale brownish black, subcostal sclerite with upstanding hairs, and two post sutural acrosctichal, alar squama white with tuft of hair on the lower margin, and lower squama infuscated, tibiae black.

Distribution: Malabar Coast, Kerala (Nandi, 2004).

5. *L. papuensis* (Macquart, 1843)

Diagnosis: Frons broader than inter post ocelli distance, parafacialia broader than the third antennal segment, occiput with more than two irregular rows of black post ocular setae, posterior surface of post gena with black hairs, anterior pair of post sutural acrostichals present posterior to the second pair of post sutural dorsocentrals, alar and thoracic squama infuscated with a tuft of blackish brown hairs at the lower margin.

Distribution: Malabar Coast, Kerala (Nandi, 2004)

6. *Lucilia sericata* (Meigen, 1826)

Diagnosis: Parafrontalia with short decumbent bristles, cerebrale with 8-9 occipital bristles on either side, non arched tergites, tergites metallic golden green with sparse pruniosity, absence of tuft of long hairs on sternites, hypopygium inconspicuous.

Distribution: Calicut, Kerala (Nandi, 2004; Priya and Sebastian, 2015)

Subfamily: Chrysomyinae

Genus: *Chrysomya* Robineau-Desvoidy, 1830

7. *Chrysomya megacephala* (Fabricius, 1794)

Diagnosis: parafrontalia slightly narrower than the breadth of frons, and were covered with golden tomentum. Antennae, arista and palpi were orange, parafacialia and genae were completely orange in colour, anterior spiracles were dark brown in colour, sub costal sclerite covered with brown felted pubescence and also with small erect hairs, a row of setulae were seen on the upper posterior side on the stem vein. Upper calypter was with ventral hairs on the opaque white basal part. Eyes were holoptic in males and dichoptic in females. Facets of upper two-thirds in the male eyes were enlarged and was clearly demarcated from the smaller facets below. In females, the facets were uniformly small. In males, the parafrontalia was covered with golden tomentum. Outer vertical bristles were absent in males. In females, the frontal stripe is broader at the middle of frons than in male fly.

Additional material examined: 6 males; 9 females,Collected and Identified by: Reject Paul, M.P.

Location: Thrissur (Choolissery-10°35’45.0"N; 76°11’18.3"E), Palakkad (Palappuram-10°45’48.3 "N;76°24’54.1"E), Ernakulam (Kanjirakkad-10°07’37.8"N; 76°28’03"E).

Repository: Dept. of Zoology, St. Thomas’ College (Autonomous), Thrissur, Kerala.

Distribution: Calicut, Thrissur, Palakkad and Ernakulam in Kerala (Bharti and Singh, 2017; Reject Paul and Binoy, 2022).

Remarks: *Chrysomya megacephala* is a forensically significant blow fly and in the current investigation, it was found to get attracted to decomposing pork meat and completing its lifecycle.

8. *Chrysomya chani* (Kurahashi, 1979)

Diagnosis: Fuscous to black colour was present on the genae and parafacialia, setulae and hairs on parafacialia and parafrontalia were blackish in colour, brown to fuscous coloured 1st , 2nd and 3rd antennal segments were present, black hairs were seen on the venter of tergite V, small prothoracic spiracle was fuscous black in colour, black coloured epaulet and basicosta were present, dense basal tuft of black hairs were present on the subcostal sclerite, black setae were present on the upper margin of 3rd longitudinal vein, base of alar squamae was white in colour and ventrally it was bare except for fringe. Eyes were holoptic in males and dichoptic in females. Facets of upper two-thirds in the male eyes were enlarged and was clearly demarcated from the smaller facets below. In females, the facets were uniformly small. In males, the parafrontalia was covered with fuscous to black tomentum. In females, the frontal stripe is broader at the middle of frons than in male fly.

Additional material examined: 4 males; 7 females. Collected and Identified byReject Paul, M.P.

Location: Thrissur (Thangaloor; 10°37’35.5"N; 76°11’15.3"E), Palakkad (Varode-10°48’50.0"N; 76°22’47.2"E), Ernakulam (Kuruppampady-10°07’01.8"N; 76°30’12.6"E).

Repository: Dept. of Zoology, St. Thomas’ College (Autonomous), Thrissur, Kerala.

Distribution: Western Ghats, Thottilpalam, Calicut, Thrissur, Palakkad and Ernakulam, Kerala (Bharti, 2014; Bharti and Singh, 2017; Reject Paul and Binoy, 2022).

Remarks: *Chrysomya chani* is a forensically significant blow fly and in the current investigation, it was found to get attracted to decomposing pork meat and completing its lifecycle.

9. *Chrysomya nigripes* (Aubertin, 1932)

Diagnosis: Parafrontalia and parafacialia with grey tomentum, genae grey, antennae dark brown, anterior spiracle white, only one mesopisternal setae developed, all hairs on the tergite V black, prothoracic stigma white, hind margins of second and third tergites dark banded, basicostal scale dark brown, sub costal sclerite with pale hairs, squama white.

Distribution: Calicut, Kerala (Bharti and Singh, 2017).

10. *Chrysomya rufifacies* (Macquart, 1842)

Diagnosis: Third antennal segment is brownish red in colour on the inner surface. Parafrontalia was narrowed with a black colour in the upper half, lower half was covered with silver tomentum and was covered with upstanding white hairs, parafacialia and genae were light yellowish and covered with white hairs, anterior spiracle white, few white hairs were present on the tergite V, and upper squama was white in colour. The lower squama was slightly fuscous in colour with white hairs. Eyes were holoptic in males and dichoptic in females. In males the parafrontalia was reduced to a fine line covered with silver tomentum. The right and left paprafrontalia slightly narrower than the breadth of the frons. In females, the frontal stripe is broader at the middle of frons than in male fly. Median incision present on the posterior edge of tergite V of females.

Additional material examined: 6 males; 9 females - Collected and Identified by: Reject Paul, M.P.

Location: Thrissur (Thangaloor; 10°37’35.5"N; 76°11’15.3"E), Palakkad (Varode; 10°48’50.0"N; 76°22’47.2"E), Ernakulam (Kuruppampady; 10°07’01.8"N;76°30’12.6"E),

Repository: Dept. of Zoology, St. Thomas’ College (Autonomous), Thrissur, Kerala.

Distribution: Calicut, Thrissur, Palakkad and Ernakulam in Kerala (Nandi, 2004; Bharti and Singh, 2017; Reject Paul and Binoy, 2022)

Remarks: *Chrysomya rufifacies* is a forensically significant blow fly and in the current investigation, it was found to get attracted to decomposing pork meat and completing its lifecycle.

11. *Chrysomya albiceps* (Wiedemann, 1819)

Diagnosis: Third antennal segment blackish brown, prostigmal bristles absent, two mesoepisternal setae, dorsal part of thorax shine with a little dusting, anterior spiracle white, few white hairs on the posterior edge of tergite V with incision, black transverse narrow marginal bands on the 3rd and 4th tergites.

Distribution: Periyar Lake and Tiger Reserve, Thekkady, Kerala (Radhakrishnan *et al.*, 2012)

Subfamily: Rhiniinae

Genus: *Idiella* Braeuer and Berensteamn, 1889

12. *Idiella euidielloides* (Senior-White, 1922)

Diagnosis: Basicosta black, sternopleuron and mesopleuron with distinct piliferous spots, first and second tergite with few black lateral bristles, posteroventral surface of hind tibia with longer hairs, tibial hairs not exceeding the width of tibia.

Distribution: Cardamom Estate, Kerala (Arce et al., 2020)

13. *Idiella mandarina* (Wiedemann, 1830)

Diagnosis: Frontal stripe brownish black, white parafrontalia with black spots, genae shining black, antennae brown, black palpi, lower half of the occiput with dense hairs, pleurae with dense golden hairs, tibiae and first tarsal joint brown and rest of tarsi black.

Distribution: Thiruvananthapuram, Kerala (Nandi, 2004).

Genus: *Stomorhina* Rondani, 1861

14. *Stomorhina discolor* (Fabricius, 1794)

Diagnosis: Frontal stripe dark brown, parafacialia and parafrontalia white with shining black spots, epistome and genae shining black, antennae and palpi brown, green thorax densely grey dusted with small black spots, anterior lower mesopleuron and anterior sternopleuron glossy black, abdominal segments with black hind margins with a black median stripe, hind femur yellowish at base, tibiae and tarsi brownish yellow.

Distribution: Cardamom Estate, Kerala (Nandi, 2004).

Genus: *Cosmina* Robineau-Desvoidy, 1830

15. *Cosmina bicolor* (Walker, 1856)

Diagnosis: Parafrontalia greyish with black spots, parafacialia silvery white, antennae yellowish brown, palpi black, propleuron hairy, mesopleuron metallic green, sub median mesonotal stripes broad, abdominal segments with a median stripe, strong bristles close to the apex of fifth sternite, hypopygium without strong spines, epaulet reddish brown.

Distribution: Nilgiris, Kerala (Nandi, 2004).

16. *Cosmina simplex* (Walker, 1858)

Diagnosis: Parafacialia silvery white with black spots, parafrontalia greyish with shining black spots, genae shining black, antennae yellowish brown, thorax copper green with black spots, long bristles on the entire surface of fifth visible sternite, hypopygium with curved laterally directed spines.

Distribution: Kochi, Kerala (Nandi, 2004).

Genus: *Strongyloneura* Bigot, 1886

17. *Strongyloneura prolata* (Walker, 1860)

Diagnosis: Mesopleuron without bristle on its upper part, third sternite with- out tuft of hair, fourth sternite with tuft of hair, fifth sternite and hypopygium are well developed, last sternite projected posteriorly and widely uncovered by corresponding tergites, bend of vein M_{1+2} gently curved.

Distribution: Chalakudy, Kerala (Nandi, 2004).

Among the 17 species recorded, six species were pollinators (*Idiella euidielloides*, *I. mandarina*, *Stomorhina discolor*, *Cosmina bicolor*, *C. simplex* and *Strongyloneura prolata),* seven species forensically significant flies (*Chrysomya megacephala*, *C. chani*, *C. nigripes*, *C. rufifacies*, *C. albiceps*, *Hemipyrellia ligurriens* and *Lucilia sericata*), two species carrion breeders (*Lucilia ampullacea*, *L. papuensis*) and two species were termite predators (*Bengalia jejuna* and *B. surcoufi*) (Nandi, 2002).

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Acarine species associated with subterranean termites (Blattodea, Termitidae)

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ABSTRACT: Ten species of mites were found associated with three species of termites, *viz*, *Odontotermes obesus* Rambur, 1842, *O. feae* Wasmann, 1896 and *Nasutitermes gardneri* Snyder, 1933. The collected mite species conformed to nine genera, four families and two orders. Among the ten species collected, nine of them were from the order Sarcoptiformes and cohort Astigmatina, while only one belonged to the order Trombidiformes. Seven species collected conformed to the family Acaridae. All mite species exhibited a phoretic relationship with their respective host insects. One adult mite, *Premicrodispus paramaevi* Hosseininaveh and Hajiqanbar, 2015 and nine deutonymphs were collected and described.

KEY WORDS: Acaridae, Microdispidae, Suidasiidae, mite, deutonymphs, phoresy

Eusociality in termites converges along many lines with colony organization and highly social behaviour in the phylogenetically distinct insect order Hymenoptera. Termites have the potential to destroy the agriculture of tropical farmers and even make their way to our home to destroy the household materials (Korb, 2007). Termites are found associated with many organisms with which they share different types of associations like phoresy, parasitism, mutualism and commensalism (Wang *et al.*, 2002). Acarine associates of termites are a least explored area (Eickwort, 1990). Majority of the termite associated mites are from the cohort Astigmatina and the deutonymphal hypopi are the more frequently reported groups (Phillipsen and Coppel, 1977; Eraky 1998, 1999a, b, 2000, 2003; Fakeer *et al.,* 2014). O'Connor (2001) reviewed the various genera of the family Acaridae exhibiting termite association which are *Australhypopus, Machadoglyphus, Mahunkallinia, Mahunkaglyphus*, *Cosmoglyphus, Sancassania, Schweibea* and one unnamed genus. About 25 species of Mesostigmata has been reported in association with termites and termite nests (Hunter and Rosario, 1988). Records of heterostigmatic mites phoretic on termites are limited to some families like Scutacaridae, Pygmephoridae, Microdispidae, Dolichocybidae (Khaustov *et al.*, 2016, 2017, 2018a, b; Baumann and Ferragut, 2019).

Termites were collected from the termitarium from the agricultural fields of Thrissur, Palakkad and Malappuram districts of Kerala. The termites were collected in aerated plastic bottles either by using a moistened camel hairbrush or otherwise a part of termitarium containing the termites was carefully placed inside the bottle. Mites were carefully removed from the host insects and permanent slides

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Table 1. Mite species collected from the termites (Blattodea, Termitidae) with phoretic relationship

Termite species	Mite species	Family/Order of mite species	part preferred by the mite
Odontotermes feae Wasmann, 1896 Worker and soldier	1. *Premicrodispus paramaevi* (Hosseininaveh & Hajiqanbar, 2015)	Microdispidae: Trombidiformes	Sternal region
	2. *Rhizoglyphus vicantus* Manson, 1977	Acaridae: Sarcoptiformes	Dorsal head
	3. *Schweibea* sp.	Acaridae: Sarcoptiformes	Dorsal head
	4. *Caloglyphus subterraneousi* Fakeer *et al*., 2014	Acaridae: Sarcoptiformes	Dorsal head
O. obesus Rambur, 1842Worker and soldier	1. *Rhizoglyphus vicantus* Manson, 1977	Acaridae: Sarcoptiformes	Dorsal head
	2. *Sancassania boharti* (Cross, 1968)	Acaridae: Sarcoptiformes	Dorsal head
	3. *Histiostoma herbali* Eraky 2017	Histiostomatidae: Sarcoptiformes	Dorsal head
Nasutitermes gardneri Snyder, 1933 Worker and soldier	1. *Acarus solimani* Eraky 1999	Acaridae: Sarcoptiformes	Dorsal head
	2. *Acotyledon tariqi* Ashfaq *et al.,*1987	Acaridae: Sarcoptiformes	Dorsal head
	3. *Caloglyphus manuri* Negm, 2007	Acaridae: Sarcoptiformes	Dorsal head
	4. *Sapracarus* sp.	Suidasiidae: Sarcoptiformes	Dorsal head

were made by following standard procedure. (Walter and Krantz, 2009). The microscopic examination of termites revealed that mites are frequently associated with both the workers and soldiers of termites. A total of ten species belonging to nine genera, four families and two orders were collected from different body parts like dorsal head (Fig. 1) and sternal region of three termite species, *viz, Odontotermes feae* Wasmann, 1896, *O. obesus* Rambur, 1842 and *Nasutitermes gardneri* Snyder, 1933 (Table 1). Among the ten species collected, nine of them were from the order Sarcoptiformes and cohort Astigmatina while only one of them belonged to the order Trombidiformes. Seven among the ten species collected conformed to the family Acaridae. The acarine families like Microdispidae, Histiostomatidae and Suidasiidae were represented by single species. The mite

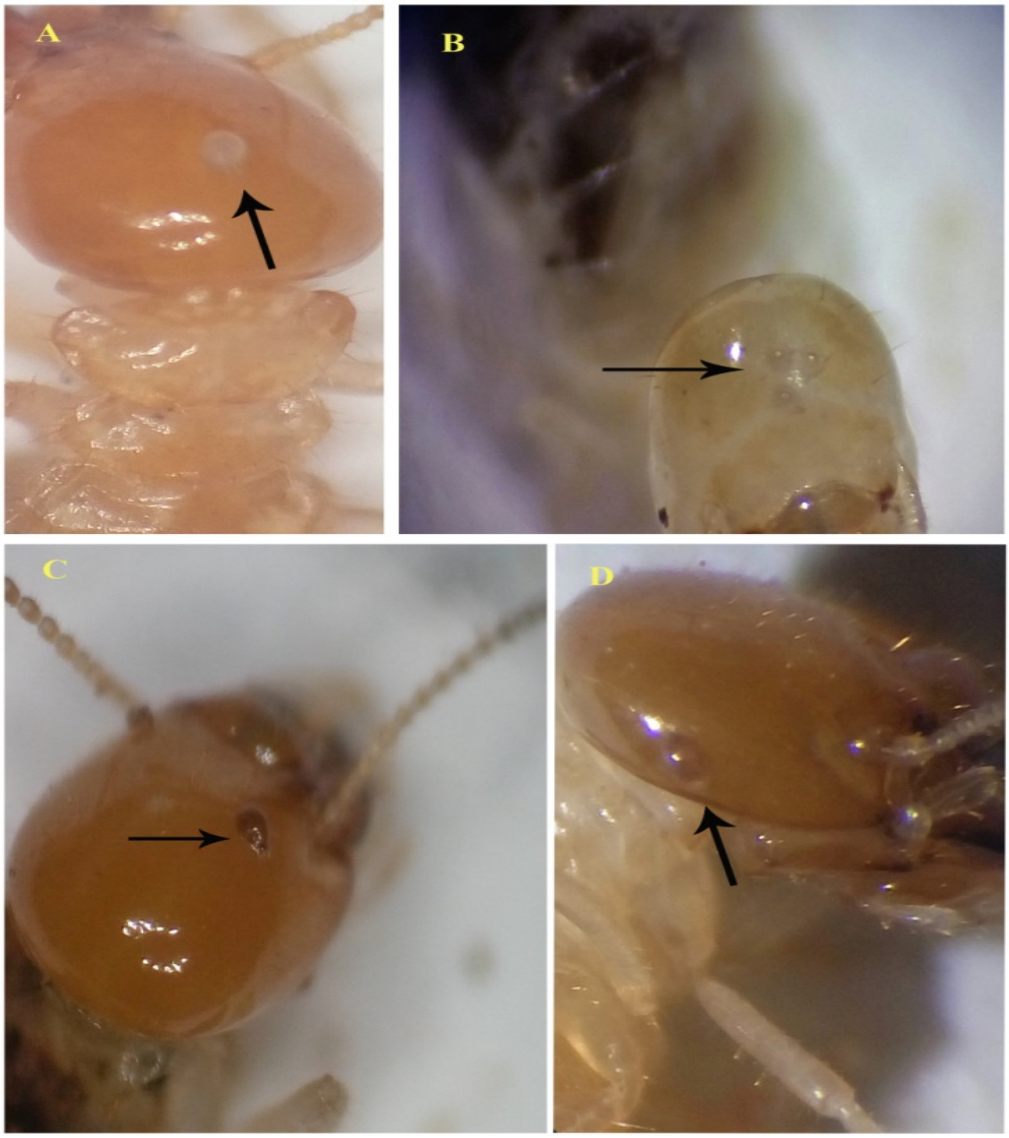

Fig. 1 A. *Histiostoma herbali* on *Odontotermes obesus*, *B. Acarus solimani* on *Nasutitermes gardneri*, *C. Caloglyphus subterraaneousi* on *Odontotermes feae*, *D. Sancassania boharti* on *Odontotermes obesus*

species were exclusively recovered in their deutonymphal stage except for one species, *Premicrodispus paramaevi* whose adult female was collected. All of the collected mite species except one species, *Rhizoglyphus vicantus* exhibited preference to a single host termite. Majority of the species preferred to attach on the dorsal head capsule of the host insect. The average number of mites collected from a single species was very low ranging from 1-2.

The present study reports 10 species of mites associated with three species of termites, all of which forms new host records. *Premicrodispus paramaevi* Hosseininaveh & Hajiqanbar, 2015 was recorded from *O. feae* is a new record and was earlier reported from the beetle *Lucanus ibericus* Motschulsky, 1845 (Hosseininaveh *et al*., 2015). The above finding concludes the wide host range of *P. paramaevi* ranging from Coleoptera to Blattodea. The mite, *Rhizoglyphus vicantus*

Manson, 1977 reported from *O. feae* and *O. obesus* in the present study has previous records from soil litter and plant materials (Barbosa and De Moraes, 2020). Likewise, *Histiostoma herbali* Eraky *et al.*, 2017, found phoretic on *O. obesus* in the present study has been previously reported from soil (Eraky *et al.*, 2017). The above two records help to draw the conclusion that mites present in the soil litter might have loaded on the termites for transport and hence phoretic in association. The report of *Acarus solimani* Eraky 1999 from *N. gardneri* in the present study is in agreement with the finding of Eraky (1999), where the author discovered *A. solimani* associated with an undetermined species of termite. The genus *Schweibea*, *Acotyledon, Caloglyphus, Sancassania* were found in association with other species of termites (O'Connor, 2001; Eraky *et al.*, 2015), which suggests that the above genera are constantly associated with termites.

Among the 10 species of mites collected, nine were found attached to the head capsule of the termite which is similar to the observation made by Wang *et al.* (2002), Myles (2002) and Silva *et al.* (2016) but the authors does not provide any particular reason for the high preference of mites towards head capsule. A similar observation was made by Behura (1956) where the hypopi of *Histiostoma polypori* (Oudemans 1914) exhibited high preference towards smooth cuticle of its host earwig and the author justified this finding that the hypopi clinging to the smooth surface of the host body is difficult to be detached. The above justification can be applied to the present observation since all the mites obtained from the head capsule was in the hypopus stage. The head capsule in the termite body offers a very smooth substratum for the hypopi to attach and can ensure a safe transport. A high incidence of astigmatid deutonymphal stage on termites implies that the mite embark on the termite body merely for transport, ie, phoresy. Members of Cohort Astigmatina exhibit heteromorphic stage called hypopus to aid in their phoretic dispersal (O'Connor, 2009), phoresy being three types; accidental, facultative and obligate as observed by Camerik (2009). The type of phoresy seen here is possibly facultative phoresy since facultative phoresy occur in relatively transient environments as observed by Camerik (2009) and termitarium offers more or less a transient habitat. The hypopus stage thus offers a free ride for the mite deutonymphs enabling them to explore different habitats where the soldiers and workers let them. Majority of mites were recovered from the family Acaridae and this observation further supports the observation that the phoresy here is not accidental since the acarid mites can be a constant associate of the fungal garden found in the termitarium and can utilise the ride offered by workers and soldiers in the termitarium as workers and soldiers are members which move out the colony for foraging and defence respectively. The constant association of acarid mites with fungus and decaying materials was observed by Zhang (2003) which again support the present observation of high incidence of acarid mites on the termite body.

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New distributional records of six Aphodiinae species (Coleoptera, Scarabaeidae) from south India

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ABSTRACT : Six species (*Platytomus indicus* (Balthasar, 1941), *P. nathani* Pittino & Mariani, 1986, *Rhyssemus karnatakaensis* Pittino, 1984, *R. procerus* Petrovitz, 1973, *R. loebli* Petrovitz, 1975 and *Neocalaphodius moestus* (Fabricius,1801)) of the subfamily Aphodiinae (Coleoptera, Scarabaeidae) are first report from Kerala and two first report from south India. Descriptions of species with images are provided. © 2023 Association for Advancement of Entomology

KEY WORDS: Dung beetles, *Neocalaphodius, Platytomus, Rhyssemus,* Kerala, first report

Subfamily Aphodiinae (Coleoptera, Scarabaeidae) is a predominant group of the dung beetle communities in the Palearctic and Nearctic region (Hanski, 1991; Lobo, 2000; Cabrero-Sañudo and Lobo, 2009) consisting of nearly 285 genera and 3,200 described species. 133 species of Aphodiinae are reported from India (Chandra, 1999) with 10 genera from south Indian region. In the present study, first record of one species of *Neocalaphodius,* two species of *Platytomus*, and three species of *Rhyssemus* from Kerala state and first record of one species of *Rhyssemus* from Karnataka state are reported.

Specimens were collected from agricultural fields at Pattambi (10°48'36''N; 76°11'21''E, 53m) of Palakkad district, Karuvambram west (11°06'56''N; 76°07'21''E, 66m) of Manjeri of Malappuram district and forests at Nelliyampathy (10°29'45''N; 76°42'21''E, 1020m) of Palakkad district, Parambikulam Tiger Reserve (10°26'31''N - 76°49'07''E, 569 m) of Palakkad district and Peechi (10°29'05''N; 76°26'01''E, 705m) of Thrissur district of Kerala State and from the Mookambika Wildlife Sanctuary (13°49'40''N; 74°44'46''E, 277m) of Udupi district of Karnataka State using the portable UV LED light traps and light sheet.

Tribe, genus and species level identification were done by using the keys provided in Schmidt (1908), Balthasar (1963), Pittino and Mariani (1986), specimens were studied using Labomed CZM6 microscope. Photographs were taken with Leica DFC 450 Camera and images were stalked using Leica V3.80. The images were post-processed using Adobe@ Photoshop@ CC. Identified specimens were deposited in 'National Zoological Collections' at Western Ghat Regional Centre, Zoological Survey of India, Kozhikode (ZSIK). New records are asterisked '*'.

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***1. Platytomus indicus* (Balthasar, 1941) (Fig. 1)**

Diastictus indicus Balthasar, 1941, 133: 170; 1964, 3: 543.

Description: Lateral clypeal margin widely rounded. Pronotum as wide as long, widest distinctly behind middle, obviously narrowed anteriorly, anterior angle narrow, lacking marginal furrow. Elytra widest behind the middle, finely and slightly striate; metasternum smooth, shiny and punctate, longitudinal furrow complete from base to apex. Fore tibia with a small basal tooth behind the third one.

Body length: 3 to 3.4mm.

Material examined: 13 ex., 23.iii.2022, light trap, Pattambi, Palakkad, Kerala, India, coll. K. A. Sobhana, ZSI/WGRC/I.R-INV.22729; 7 ex., 12.viii.2022, light attracted, Karuvambram west, Manjeri, Malappuram, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22888; 10 ex., 20.iii.2022, light trap, Peechi, Thrissur, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22488; 5 ex., 25.iii.2022, light trap, Nelliyampathy, Palakkad, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22257.

Distribution: India: Tamil Nadu: Coimbatore; Kerala*: Palakkad, Malappuram, Thrissur (Fig. 8).

Elsewhere: Not recorded.

Remarks: This species was earlier reported from Tamil Nadu (Balthasar, 1941) and it is the first record from Kerala state during the present study.

***2. Platytomus nathani* Pittino & Mariani, 1986 (Fig. 2)**

Platytomus nathani Pittino & Mariani, 1986, 3: 49.

Description: Dark reddish brown colour. Pronotum slightly narrower than elytral base. Metasternum unpunctate, lacking any trace of a longitudinal furrow. Hind femur about as wide as anterior one, slightly wider than middle femur. Pronotal and metasternal sculptures are extremely reduced.

Body length: 3.1 to 3.5mm.

Material examined: 10 ex., 23.iii.2022, light trap, Pattambi, Palakkad, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22728; 7 ex., 12.viii.2022, light attracted, Karuvambram west, Manjeri, Malappuram, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22888; 10 ex., 20.iii.2022, light trap, Peechi, Thrissur, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22488; 35 ex., 25.iii.2022, light trap, Nelliyampathy, Palakkad, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV. 22257.

Distribution: India: Tamil Nadu: Coimbatore; Kerala*: Palakkad, Malappuram, Thrissur.

Elsewhere: Not recorded.

Remarks: This species was earlier reported from the state of Tamil Nadu (Pittino & Mariani, 1986) and is the first record from the state of Kerala during the present study.

***3. Rhyssemus karnatakaensis* Pittino, 1984 (Fig. 3)**

Rhyssemus karnatakaensis Pittino, 1984, 2(6): 34.

Description: Uniformly black, except clypeal and pronotal margins in reddish brown. Anterior clypeal border roundly emarginated. Head with rounded and oblong-oval tubercles. Pronotum widest in the middle. Elytra oval, strongly convex, widest at the middle. Metasternum smooth, shiny, and punctate at middle with wide, deep, oval median depression and with distinct complete midline furrow.

Body length: 2.9 to 3.1mm.

Material examined: 1 ex., 23.iii.2022, light trap, Pattambi, Palakkad, Kerala, India, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22728; 1 ex., 20.iii.2022, light attracted, Mookambika WLS, Karnataka, coll. V.D. Hegde & party, ZSI/WGRC/I.R-INV.21894.

Distribution: India: Karnataka: Chikmangalore; Kerala*: Palakkad.

Elsewhere: Not recorded.

Remarks: This species was earlier reported from the state of Karnataka (Pittino, 1984) and it is the

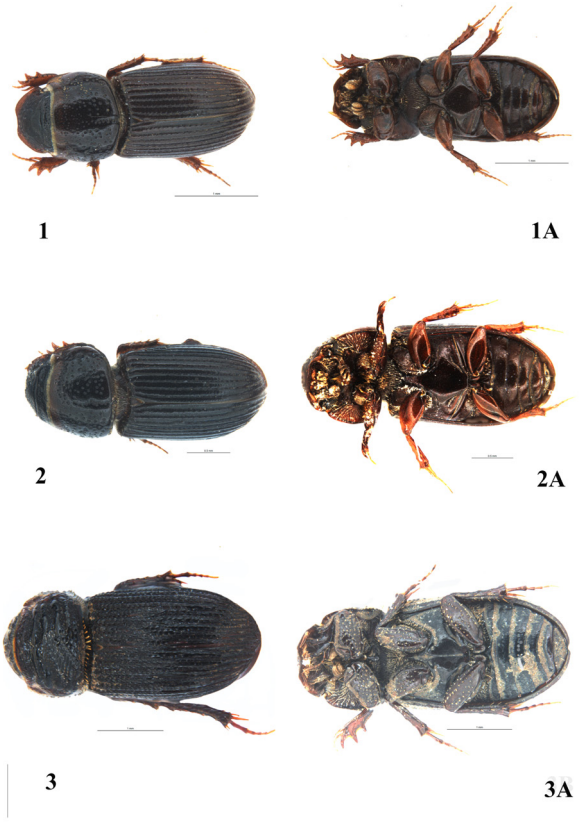

Fig. 1 Habitus of *Platytomus indicus* dorsal view, 1A- ventral view
Fig. 2 Habitus of *Platytomus nathani* dorsal view, 2A- ventral view
Fig. 3 Habitus of *Rhyssemus karnatakaensis* dorsal view, 3A- ventral view

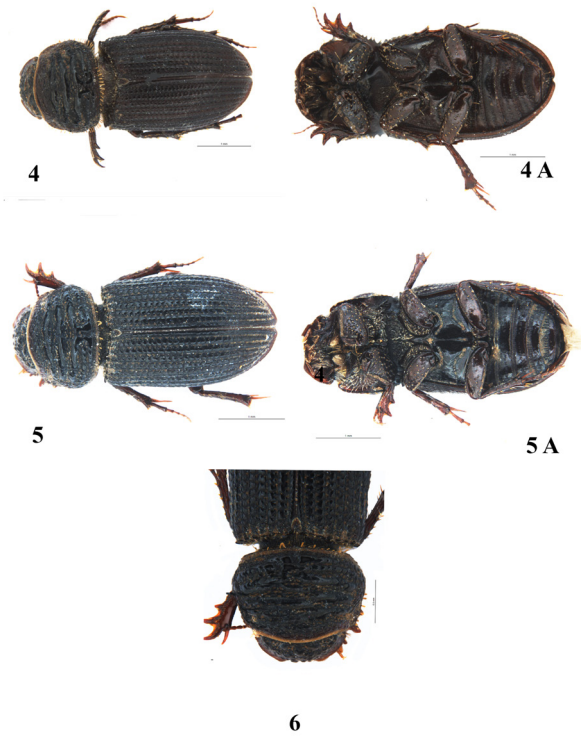

Fig. 4 Habitus of *Rhyssemus procerus* dorsal view, 4A - ventral view
Fig. 5 Habitus of *Rhyssemus loebli* dorsal view, 5A - ventral view
Fig. 6 Frontal view of *Rhyssemus loebli*

first record from the state of Kerala during the present study.

4. Rhyssemus procerus Petrovitz, 1973 (Fig. 4)

Rhyssemus procerus Petrovitz, 1973, 24: 306.
Rhyssemus tschadensis Dellacasa, 1988, 1: 423.

Description: Body slender, nearly parallel. Glossy, deep black in the front edge of clypeus. Pronotum widest in middle. Sides and bases of pronotum notched and ciliated with bristles that are thickened towards the tip. Pronotal structure consists of five transversal ridges and five transversal furrows. Elytra sub parallel with oval granules. Elytral striae with distinct humeral denticles. Ventral surface mostly glabrous. Meta- ventral plate glabrous and with complete midline furrow, narrow anteriorly and slightly reduce posteriorly, area surrounding midline furrow moderately concave. Pygidium with 2 pygidial macro setae.

Material examined: 3 ex., 23.iii.2022, light trap, Pattambi, Palakkad, Kerala, India, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22214; 2 ex., 30.x.2022, light trap, Parambikulam TR, Palakkad, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22256.

Body length: 2.9 to 3.1mm.

Distribution: India: New Delhi; Kerala*: Palakkad.

Elsewhere: Not recorded.

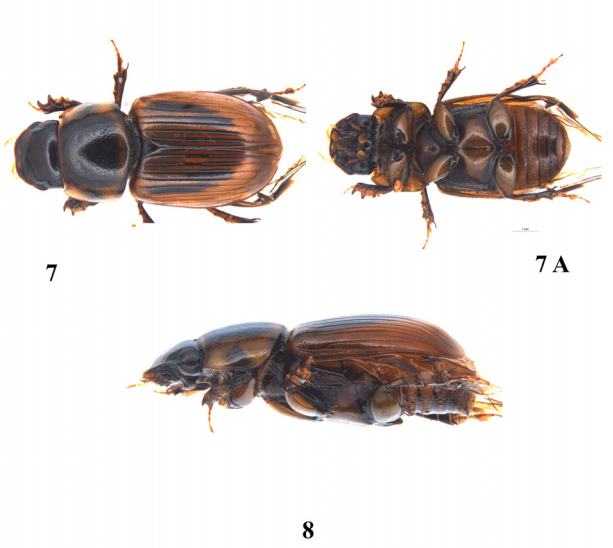

Fig. 7 Habitus of *Neocalaphodius moestus* dorsal view, 7A - ventral view
Fig. 8 Dorso-lateral view of *Neocalaphodius moestus*

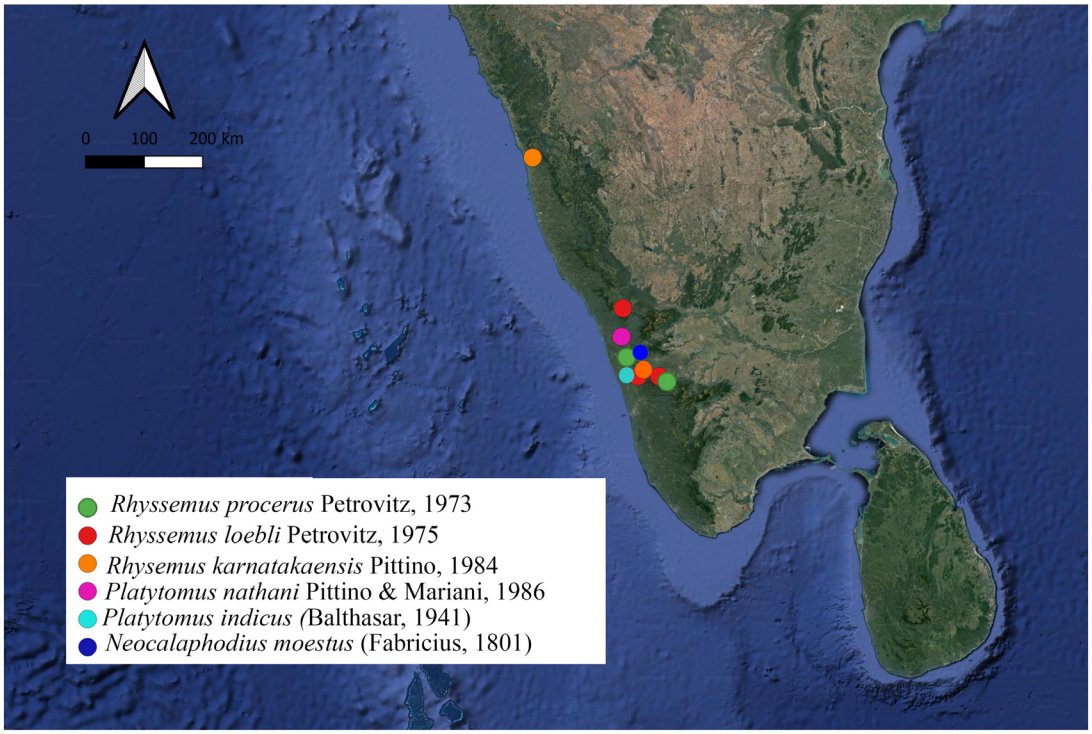

Fig. 9 Map showing the distribution pattern of subfamily Aphodiinae in the south Western Ghat Regions of Kerala

Remarks: This species was earlier reported from New Delhi (Petrovitz, 1973). It is the first report of the species from state of Kerala and also from south India during the present study.

***5. Rhyssemus loebli* Petrovitz, 1975 (Figs. 5, 6)**

Rhyssemus loebli, Petrovitz, 1975, 82: 617.
Rhyssemus loebli, Dellacasa, 1988, 1: 455.

Description: Dark brown, shining, and oblong oval. Clypeus is not dentate, but angulated each side. Clypeal surface with distinct transversal granules. Pronotum widest behind the middle. Elytra not sub parallel. Individual granules in discal elytral intervals arranged in 2 rows. Ventral surface is mostly glabrous and smooth.

Body length: 3.3 to 3.5mm.

Material examined: 8 ex., 23.iii.2022, light trap, Pattambi, Palakkad, Kerala, India, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22485; 3 ex., 12.viii.2022, light trap, Peechi, Thrissur, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.21894; 9 ex., 25.iii.2022, light trap, Nelliyampathy, Palakkad, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22256; 2 ex., light trap, Wayanad, Kerala, coll. K.A. Sobhana, ZSI/WGRC/I.R-INV.22213.

Distribution: India: Tamil Nadu: Udumalpet; Kerala*: Palakkad, Thrissur, Wayanad.

Elsewhere: Sri Lanka.

Remarks: This species was earlier reported from the state of Tamil Nadu and Sri Lanka (Petrovitz, 1975) and it is the first record from the state of Kerala during the present study.

6. *Neocalaphodius moestus* (Fabricius, 1801) (Figs. 7, 8)

Aphodius moestus Fabricius, 1801.

Aphodius mutans Walker, 1858.

Aphodius madagascariensis Harold, 1859.

Aphodius subvittatus Fairmaire, 1896.

Aphodius (Calaphodius) moestus infrasp. innotatus Endrodi, 1960.

Aphodius (Calaphodius) moestus infrasp. connectens Endrodi, 1960.

Description: Moderately convex, shiny. Head black-brown. Clypeus almost truncate anteriorly. Pronotum yellow-brown with large dark disc spot and small dark spots in the light side edge. Elytra yellow-brown, fairly deeply striped, stripes on the disc blackened, ventral parts and legs mostly tawny.

Body length: 5.3 to 5.5mm.

Material examined: 23 ex., 30.x.2022, light trap, Parambikulam TR, Palakkad, Kerala, coll. L. Bindu and party, ZSI/WGRC/I.R-INV.23608.

Distribution: **Afrotropical:** Republic of South Africa, Ghana, Sudan, Rwanda, Namibia, Ethiopia, Ivory Coast, Kenya, Chad, Niger, Angola, Botswana, Cameroon, Djibouti, Malawi, Senegal, Somalia, Tanzania, Zambia (Harold, 1859).

Oriental: India: Andaman and Nicobar Islands, Assam, Chhattisgarh, Haryana, Madhya Pradesh, Punjab, Rajasthan, Kerala*; Sri Lanka, Thailand, Myanmar, Laos, Cambodia, Malaysia), Madagascan (Madagascar, Comoros).

Palaearctic: Afghanistan, Pakistan, Turkey, Taiwan, Nepal, Tajikistan, Myanmar, Yemen: Socotra, India: Himachal Pradesh, Uttarakhand.

Remarks: Widespread speceis in Afrotropical, Oriental, and Palaearctic regions. It is the first report of the species from Kerala and south India during the present study.

The distribution pattern of subfamily Aphodiinae in the south Western Ghat Regions of Kerala is depicted in the map (Fig. 9).

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Distributional records of *Onthophagus germanus* Gillet, 1927 and *O. orissanus* Arrow, 1931 (Coleoptera, Scarabaeidae, Scarabaeinae) from south India

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ABSTRACT: Distribution records of two dung beetle species, *Onthophagus orissanus* Arrow, 1931 and *O. germanus* Gillet, 1927, from south India is provided. *O. orissanus* is reported for first time from south India and *O. germanus* is reported for the first time outside the moist south Western Ghats from the Malabar Coast region in Kerala. © 2023 Association for Advancement of Entomology

KEY WORDS: First report, South Western Ghats, Kerala

The dung beetles *Onthophagus orissanus* Arrow, 1931 and *Onthophagus germanus* Gillet, 1927comes belong to tribe Onthophagini, subfamily Scarabaeinae and family Scarabaeidae. 5700 species under 234 genera and 12 tribes are known within the subfamily Scarabaeinae (Krajcik, 2012). *Onthophagus* Latreille, 1802 is a speciose genus within the tribe Onthophagini, (Kharel *et al.,* 2020). In India, genus *Onthophagus* represented by 190 species and is the mega-diverse genus (Kharel *et al*., 2020). Known distribution of *Onthophagus orissanus* Arrow, 1931 includes Bihar, Madhya Pradesh, Maharashtra and Odisha and that of *Onthophagus germanus* Gillet, 1927 include Sikkim, Uttarakhand and West Bengal (Kharel *et al.*, 2020). Present study reveals the range extension of these species to south India and is reported for the first time from the state of Kerala.

Specimens were collected using pitfall traps during 2018-2019, from the sacred grove located in Koyilandy of Kozhikode district, Kerala, India (11° 29' 49" N; 75° 39' 51" E, 9m). The specimens were pinned, dried, labeled and then identified employing the keys of Arrow (1931) and Balthasar (1963). Specimens were examined under LEICA M205A stereo zoom microscope and imaged using LEICA DFC 500 digital camera attached to the microscope. The studied specimens are deposited at Zoological Survey of India, Western Ghat Regional Centre, Kozhikode, Kerala, India (ZSIK).

1. *Onthophagus orissanus* Arrow, 1931 (Figs.1, 2).

Onthophagus orissanus - Arrow, 1931: 257.

Onthophagus (Onthophagus) orissanus - Balthasar, 1963: 464

Material examined: 1♂, 1♀, Kerala, Kozhikode, Koyilandy, Muchukunnu, Kottayilkavu, (11° 29' 49" N; 75° 39' 51" E, 9m), 20.VII.2018, Coll. T.K. Viswanath.

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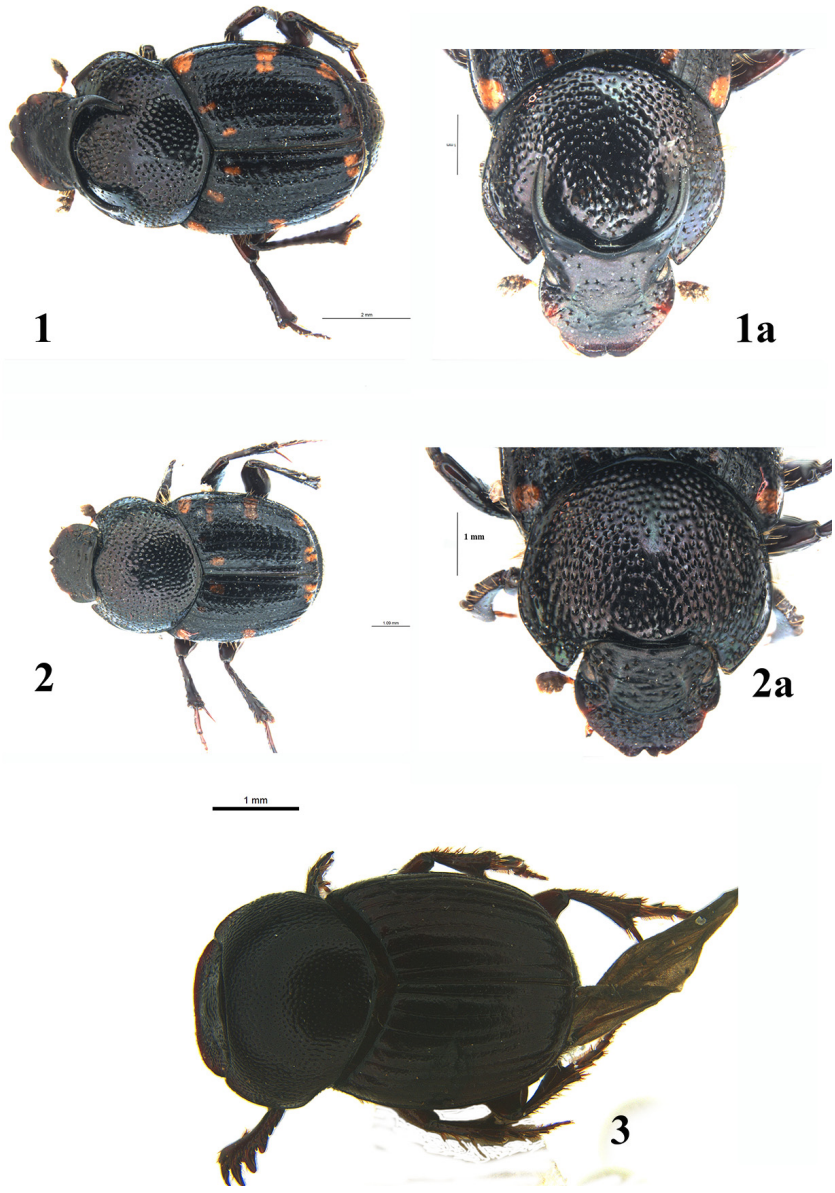

Fig. 1 *Onthophagus orissanus* Arrow, 1931 ♂, Fig. 1a *Onthophagus orissanus* Arrow, 1931 ♂ (Frontal view), Fig. 2 *Onthophagus orissanus* Arrow, 1931 ♀, Fig. 2a *Onthophagus orissanus* Arrow, 1931 ♀(Frontal view), Fig. 3 *Onthophagus germanus* Gillet, 1927 ♀

Diagnosis: Length: 6.5mm, width 4.0mm. Oval and convex. Head, pronotum, pygidium and lower surface dark coppery or metallic green in colour. Clypeus deeply notched and sharply bilobed in front. Head bears a pair of backwardly directed horns (Fig.1a) which are moderately broad at base slender at the end and united by a curved carina enclosing a half-circle at its posterior margin. In female, clypeus and forehead divided by a curved carina and the vertex of head bears another straight carina (Fig. 2a).

Distribution: India: Bihar, Kerala (New Record), Madhya Pradesh, Maharashtra and Odisha.

Remarks: *Onthophagus orissanus is* reported first time from south India. It is also the first record from the south Western Ghat region of Kerala State.

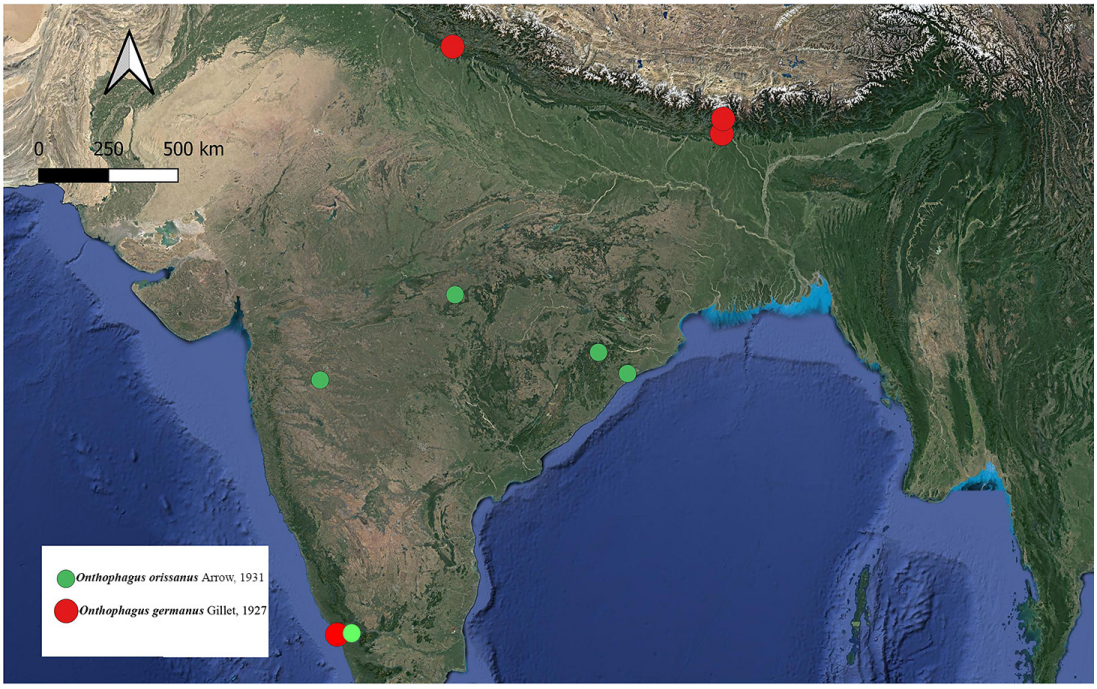

Fig. 4 Map showing the new distribution of *Onthophagus orissanus*

2. ***Onthophagus germanus*** **Gillet, 1927 (Fig.3).**

Onthophagus germanus Gillet. *Bull. Soc. Ent. Belg*. 1927, p. 254.

Material examined: 1♀, Kerala, Kozhikode, Koyilandy, Muchukunnu, Kottayilkavu, (11° 29' 49" N 75° 39' 51" E, 9m), 20.VII. 2018, Coll. T. K. Viswanath.

Diagnosis: Length 6-7mm, width 4mm. Black, Oval and highly convex, lower surface clothed with reddish hair. Head flat, broad, and densely rugose. Clypeal margin rounded with scarcely perceptible angulations in the middle. The front angles of the pronotum broadly rounded; lateral margins feebly rounded in front and sinuate behind. Elytra finely striate intervals flat and minutely punctate. Metasternum with a vertical, sharply compressed, and apically pointed process in front.

Distribution: India: Kerala (Malabar Coast; Shendurney and Wayanad in south Western Ghats montane rain forests ecoregion), Sikkim, Uttarakhand, West Bengal (Fig. 4).

Remarks: *Onthophagus germanus* was reported from south Western Ghats montane rain forests eco region (Sabu *et al.* 2011) in Kerala state with exact localities not provided by the earlier authors. It is the first record of the species outside the south Western Ghats and from the Malabar Coast in Kerala state referred as Malabar Coast moist deciduous forest ecoregion.

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Diversity of edible insects in Tuensang District, Nagaland, India

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ABSTRACT: This study provides important information on the population diversity of edible insects in Tuensang district, Nagaland, India. The region is known for its diverse group of entomofauna, which are used as a healthy food source by the local people. In the study to evaluate the edible insects of Tuensang district, Nagaland, 23 different varieties of insects were identified. The study highlights the importance of these insects as a food source for the local people, which could have implications for food security and sustainability in the region. © 2023 Association for Advancement of Entomology

KEYWORDS: Entomofauna, diversity, entomophagy, food source

Eating insects is a need in many underdeveloped countries. The edible insects have a healthy quantity of fats, proteins, carbohydrates, lipids, minerals and vitamins (Costa-Neto and Dunkel, 2016; Bernard and Womeni, 2017; Mozhui *et al*., 2017). In addition to serving as a supplemental meal or food additive in industrialised nations, edible insects may be utilised as a food source to enhance the nutritional condition of individuals living in poor countries (Van Huis *et al,.* 2013, 2015). In Nagaland, insects are an important part of the human diet historically and still insects are consumed. They include termites, cicadas, ant, wasp nests, grasshoppers, locusts, caterpillars, beetle larvae, and many aquatic insect species. Nagaland has a variety of animals, including a large diversity of insects connected to its native plants, and an abundance of natural resources. The rural populations are not always access to affordable, lengthy-to-prepare traditional sources of animal based protein. Insects are frequently eaten as a meat alternative. The ingestion of a number of edible insect species was passed down through the generations in Nagaland (Shantibal *et al.,* 2012; Pongener *et al.,* 2019).

The Tuensang district's edible insect population was not previously assessed. The current study allows us to recognize the diversity of edible entomofauna present in this area. The residents of Tuensang usually consume large amounts of edible insects that are gathered seasonally from a variety of habitats and used as a delicious meal. Despite the tribe's ignorance about the nutritional benefits of insects, the majority of people who live in villages consumed them as a nutritious seasonal meal.

The international boundary runs along Tuensang's eastern border, one of Nagaland's districts that is situated on the country's eastern side. Its neighbours

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Table 1. Edible insects of Tuensang district, Nagaland, India

No.	Common/ localname	ScientificName/ Order	Availability	Edible stage	*IUCN3.1
1	Honey bee/ Nau	*Apis cerana* (F)/ Hymenoptera	Through out the year	Eggs, Larva, pupa	LC
2	Honey bee/ Nau	*Apis florea* (F)/ Hymenoptera	Sept -Feb	Eggs, larva, pupa	LC
3.	Asian giant hornet/ Nau	*Vespa mandarinia* (Smith)/ Hymenoptera	Oct –feb	Eggs, Larva, pupa	LC
4.	Honey bee/ Nau	*Apis dorsata* (F)/ Hymenoptera	Sept –may	Eggs, larva, pupa	LC
5	Termites/ Lango	*Odontotermes obesus* (Rambur)/ Isoptera	Oct -Nov	Adult	LC
6	Cicada/ Onyung	*Cryptotympana facialis* (Walker)/ Hemiptera	June –August	Adult	LC
7	Giant cicada/ Onyung	*Quesada gigas* (Olivier)/ Hemiptera	June -August	Adult	LC
8	Dinorid bug/ Aubi labie	*Coridius singhalanus* (Distant)/ Hemiptera	Jan -march	Adult	LC
9	Katydids/ koksung	*Mecopoda nipponensis* (Walker)/ Orthoptera	Aug -Nov	Adult	LC
10	Gray bird grasshopper/ koksung	*Schistocerca nitens* (Thunberg, 1815)/ Orthoptera	June –oct	Adult	LC
11	Two –striped grasshopper/ koksung	*Melanoplus bivittatus* (Say)/ Orthoptera	June –oct	Adult	LC
12	Rice -field grasshopper/ koksung	*Oxya yezoensis* (Shiraki)/ Orthoptera	June –oct	Adult	LC
13	Red –legged grasshopper/ koksung	*Melanoplus femurrubrum* (De Geer)/ Orthoptera	June –oct	Adult	LC
14	Sulphur-winged grasshopper/ koksung	*Arphia sulphurea* (F))/ Orthoptera	June -oct	Adult	LC
15	Grasshopper/ koksung	*Oxya fuscovittata* (Marschall)/ Orthoptera	Aug- NoV	Adult	LC
16	Field cricket/ Kotshou moun shou	*Gryllus* sp./ Orthoptera	Aug –Nov	Adult	LC
17	Bush cricket/ koksung	*Mecopoda elongata* (L))/ Orthoptera	Aug – nov	Adult	LC
18	Grasshopper/ koksung	*Oxya hyla* (Servile)/ Orthoptera	June –oct	Adult	LC
19	Dragonfly/ Deipin	*Pantala flavescens* (F)/ Odonata	July –Nov	Nymph	LC
20	Dragonfly/ Deipin	*Orthetrum Sabina* (Drury)/ Odonata	July –Nov	Nymph	LC
21	Eri silkworm/ Eri yang	*Samia ricini* (Drury, 1773)/ Lepidoptera	Through out the year	Larva, pupa	LC
22	Carpenter worm/ Akyang	*Cossus* sp / Lepidoptera	July -Feb	larva	LC
23	Preying mantids/ Keipong	*Hierodula coarctata/* Mantodae	July -sept	Adult	LC

*Conserrvation status

to the north and east are Mon and Longleng District, respectively, as well as Mokokchung in the northwest, Zunheboto in the southwest, Kiphire in the south, and Myanmar in the east. This district's geography is made up of the Helipong Range, Yakur Range, Longtokur Range, Mangko Range, and Takhaya Range, which is distinguished by its high hills, deep gorges, and small valleys. The district Tuensang covers 4,228 square kilometres and can be found at latitudes of 26° 14¹ 8.67ºN and longitude of 94° 48¹ 47.47ºE, with elevations ranging from 800 to 3500m above mean sea level. The native inhabitants of this region are comparable to other Nagas in terms of their mongoloid characteristics. Changs, Khiamniungans, Sangtams, and Yimkhiung are the main tribes to call Tuensang district home. Although each tribe has its own native dialect, Nagamese serves as the "language" of communication in this territory. The district has three types of soil: alluvial soil, non-laterite red soil, and forest soil. Tuensang has an evergreen subtropical and temperate coniferous forest, which supports a diverse flora and fauna.

The largest and eastern most district of Nagaland is Tuensang, which is also blessed with a diverse entomofauna. Assessment of the edible insects in Tuensang District has not previously been conducted or examined, however the current research contributes to learning about the variety of edible entomofauna. By direct field investigation, interviews with locals, including employees, neighbours, villages, and hunters, as well as references from books, a total of 23 edible insect species from 7 Orders and 17 Genera were evaluated and identified from this study region (Table 1). The edible insects were gathered from different locations in this region based on their physical and systematic differences when they were in season and samples were kept in alcohol (70%) for subsequent characteristics reference. The images were taken using cameras. The mode of consumption is very well practised by them such as cooking or frying with local ingredients. The ingredients used were fermented bamboo shoot, dried bamboo shoot, local garlic and ginger.

The Order Hymenoptera members *Apis cerana*, *A. florea*, *A. dorsata*, and *Vespa mandariania* are available periodically in the Tuensang region. *A. cerana* species are abundantly available throughout the year and easily domesticated. The largest hornet, *V. mandariania*, is a wholesome diet by the locals of this area. *A. florae* is very tiny when compared to other honeybee varieties, it is known as the dwarf honey bee. Immature stages of *A. dorsata*, such as the eggs and larvae, are typically prepared with fermented bamboo shoot and local spices. They are the good source of honey and bee wax. Under order Othoptera, *O. yezoensis*, *S. nitens*, *M. bivittatus*, *M. femurrubrum*, *A.* sulphurea, *O. hyla*, *M. nipponensis* and *Mecopoda elongata* are found seasonally available. They are harvested in large quantities and commonly consumed in cooked or fried form with using local ingredients. Species of dragonfly *P. flavescens* and *Orthetrum sabina* Drury, 1770 are seasonally available and people prefer to collect only their nymph stage for consumption (Table 1).

Eri silkworm (*Samia cynthia ricini*) available throughout the year, as they are reared commonly in every tribal houses and is considered as their most delicious and nutritious food. It is an edible worm that is most commonly preferred by many people as they are considered as an environmentally friendly diet as they do not require a large amount of resources when they are raised. The carpenter worm (*Cossus sp*) is an edible insect; most commonly prefer their larva stage for consumption.

The termite species known as *O. obesus* (Isoptera) is most frequently consumed in fried form and makes for a nutritious meal. *C. singhalanus* (Hemiptera) known as stinkbugs is edible insect which is best consumed in chutney or cooked form and they are harvested during the month of January - March. *C. facialis* and *Q. gigas* (Hemiptera), commonly known as Cicada insect, harvested are in large quantities when available periodically by the farmers for consumption. The wings, digestive tract, and heads of the Mantodae species, *H. coarctata* are removed before the insect is ready for food. They are consumed in cooked or fried form with local ingredients.

The current study adds to the awareness of edible insect variety of Tuensang District, Nagaland, and aids in the nutritional worth. The Tuensang tribes collected the edible insects in larger quantities seasonally when they are available and consumed it as their primary source of sustenance..

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Impact of seed dressing insecticides on natural enemies of *Bt* cotton ecosystem

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ABSTRACT: Investigations were carried out on the effect of seed dressing chemicals on the beneficial predators in the *Bt* cotton ecosystem during 2021-22. All seed dressing insecticides were found safer to the natural enemies as the population of *Chrysoperla* and ladybird beetle were found comparable to population of untreated control treatment up to 37 days of seed treatment. Similarly, the spider population was also not affected up to 17 days of seed treatment. Thereafter, the population of natural enemies was found higher with the higher prey (sucking pests) populations in the untreated check compared to treatments of seed dressing chemicals and in later treatments, there was no significant difference. The maximum population of spiders (0.52/plant), *Chrysoperla* (0.42/plant) and ladybird beetle (0.42/plant) was observed in the untreated control. Yield data indicated that the treatment with imidacloprid 70 WG @ 3 g kg^{-1} obtained highest seed cotton yield (21.69 q ha^{-1}) and it was found superior over the other seed treatments.

KEYWORDS: *Chrysoperla*, ladybird beetle, spider, insecticides

Cotton (*Gossypium hirsutum* L.) is an important commercial fibre crop grown under diverse agro-climatic conditions and is called as 'White Gold' and also as King of Fibre. Owing to the introduction of *Bt* cotton having gene from *Bacillus thuringiensis* (Berliner) expressing delta endotoxin, the pest status of bollworm complex has declined (Peshin *et al*., 2007). Though genetically engineered *Bt* cotton provide effective management of bollworm complex but nowadays sucking pests *viz*., thrips, *Thrips tabaci* (Lindeman), leafhopper; *Amrasca biguttula biguttula* (Ishida), aphid, *Aphis gossypii* (Glover) and whitefly; *Bemisia tabaci* (Gennadius) attained the status of key pests in Gujarat and cause considerable damage (> 10%) to the cotton crop during its early stages of development resulting in pre-mature shedding of leaves and fruiting parts. In the early stage of growing the crop, farmers use foliar insecticides to avoid damage from these pests. These early foliar applications of insecticide often kill the natural enemies which then results in a resurgence of the pests. With the introduction of the systemic insecticides for seed treatment, farmers have been able to use them to protect their crop from the early season, sap- sucking insect pests. The effects of imidacloprid and thiamethoxam on sucking pests (Kagabu, 1999; Yamada *et al.,* 1999; Maienfisch

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Table 1. Effect of seed treatment of various chemicals on population of spider in *Bt* cotton

Treatments	Dose (kg^{-1} seed)	Average spider per plant- DAS										
		10	17	23	30	37	44	51	58	65	72	Pooled
Carbosulfan 25 DS	60 g	0.71^{a} (0.00)	0.73^{a} (0.03)	0.72^{a} (0.02)	0.74^{b} (0.04)	0.74^{b} (0.05)	0.74^{b} (0.04)	0.74^{b} (0.05)	0.74^{b} (0.05)	0.75^{b} (0.06)	0.77^{b} (0.09)	0.74^{b} (0.05)
Imidacloprid 70 WG	3 g	0.71^{a} (0.00)	0.74^{a} (0.04)	0.74^{a} (0.04)	0.75^{b} (0.06)	0.76^{b} (0.08)	0.77^{b} (0.09)	0.78^{b} (0.10)	0.76^{b} (0.08)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.75^{b} (0.06)
Imidacloprid 48 FS	8 ml	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{b} (0.00)	0.77^{b} (0.09)	0.79^{b} (0.12)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.74^{b} (0.04)
Imidacloprid + Hexaconazole 20 FS	2 ml	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.73^{b} (0.03)	0.73^{b} (0.04)	0.74^{b} (0.04)	0.74^{b} (0.05)	0.74^{b} (0.05)	0.73^{b} (0.03)	0.71^{b} (0.01)	0.72^{b} (0.02)
Thiamethoxam 30 FS	10 ml	0.71^{a} (0.00)	0.73^{a} (0.03)	0.74^{a} (0.04)	0.74^{b} (0.04)	0.75^{b} (0.06)	0.76^{b} (0.08)	0.77^{b} (0.09)	0.75^{b} (0.06)	0.75^{b} (0.06)	0.77^{b} (0.09)	0.74^{b} (0.05)
Thiamethoxam 70 WS	4 g	0.71^{a} (0.00)	0.73^{a} (0.03)	0.74^{a} (0.04)	0.74^{b} (0.04)	0.74^{b} (0.05)	0.74^{b} (0.04)	0.74^{b} (0.05)	0.74^{b} (0.05)	0.75^{b} (0.06)	0.77^{b} (0.09)	0.74^{b} (0.05)
Chlorantraniliprole 9.3 SC + lamda cyhalothrin 4.6 CS (13.9 ZC)	2.5 ml	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.74^{b} (0.04)	0.73^{b} (0.04)	0.74^{b} (0.05)	0.75^{b} (0.06)	0.74^{b} (0.05)	0.73^{b} (0.03)	0.72^{b} (0.01)	0.73^{b} (0.03)
Control	-	0.82^{a} (0.17)	0.86^{a} (0.24)	0.90^{a} (0.31)	0.95^{a} (0.41)	0.97^{a} (0.44)	1.02^{a} (0.55)	1.07^{a} (0.65)	1.12^{a} (0.75)	1.19^{a} (0.91)	1.19^{a} (0.91)	1.01^{a} (0.52)
CD at 5% (T)		NS	NS	0.11	0.12	0.12	0.11	0.11	0.11	0.12	0.11	0.04
CD at 5% (TxP)	-	-	-	-	-	-	-	-	-	-	-	0.10

Figures in parentheses are retransferred values, those outside are square root transformed values. In each column means followed by a same alphabet are not significantly different from each other

et al., 2001) and their effects on predators are well documented (Woolweber and Tietjen, 1999). However, very little information is available on the effect of these seed treatment insecticides on the *Chrysoperla,* ladybird beetle and spider under field conditions. Seed treatment insecticides are used commercially to protect against injury by early season sucking pests (Wilde *et al.,* 1999; Mckirdy and Jones, 1996). It is also effective at controlling many sucking insects, including aphids, thrips, jassid, whitefly, and mealybugs when used as a seed treatment (Harvey *et al.,* 1996) and is commonly used on several crops, including cotton (Hernandez *et al*., 1999). This study was initiated to gain confidence on the safety of these compounds against the predators in order to include these compounds for the management of early season sucking pests.

The present investigation was conducted during *Kharif* 2021 at Main Cotton Research Station, Navsari Agricultural University, Surat. Systemic insecticides were used as seed treatment under field condition on *Bt* cotton variety, Ajeet 155 BG II used. The untreated seed was used as control treatments. The experiment was laid out in a randomized block design with three replications. Treated and untreated seeds were sown by hand using a dibbing method on bed and furrow. The plots consisted of 18518 plants per hectare spaced

0.45cm within row and 1.20m between rows. No foliar spray application was given during the study period.

The required quantity of *Bt* cotton seeds and insecticides (Table 1) were put in polythene bag and mixed thoroughly. Few drops of water *i.e.* @ 2 ml 100 g^{-1} seed were sprinkled on the mixture of seeds and insecticide. The mixture was stirred frequently till uniform coating of insecticides occurred. The treated seeds were spread on a paper in a room and kept overnight for drying.

Five plants were selected randomly from each plots tagged. While plants located at border were avoided for recording observation. The total number of grubs and adult of lady bird beetle, *Chrysoperla* and spider was count. Observations were recorded on all 5 randomly selected plants up to 72 days at weekly intervals and data were subjected to statistical analysis. The cotton from each net plot (3.60 x 4.50cm) was picked at each picking and weighed separately. The picking was carried out till the end of season. Total yield from each plot was calculated and computed on hectare basis. The data collected during the course of experimentation were subjected to statistical analysis with appropriate transformation for interpretation of results in Randomized Block Design (RBD) in order

Table 2. Effect of seed treatment of various chemicals on population of *Chrysoperla* in *Bt* cotton

Treatments	Dose (kg^{-1} seed)	Average spider per plant- DAS										
		10	17	23	30	37	44	51	58	65	72	Pooled
Carbosulfan 25 DS	60 g	0.71^{b} (0.00)	0.73^{b} (0.03)	0.74^{b} (0.05)	0.75^{b} (0.06)	0.72^{b} (0.02)	0.78^{ab} (0.11)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.71^{b} (0.00)
Imidacloprid 70 WG	3 g	0.71^{b} (0.00)	0.74^{b} (0.04)	0.75^{b} (0.06)	0.76^{b} (0.07)	0.77^{ab} (0.09)	0.79^{ab} (0.12)	0.80^{b} (0.14)	0.78^{b} (0.11)	0.77^{b} (0.10)	0.79^{b} (0.13)	0.71^{b} (0.00)
Imidacloprid 48 FS	8 ml	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.77^{ab} (0.09)	0.78^{b} (0.10)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.71^{b} (0.00)
Imidacloprid1+ Hexaconazole 20 FS	2 ml	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.74^{b} (0.04)	0.76^{ab} (0.08)	0.75^{b} (0.07)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.01)	0.71^{b} (0.00)
Thiamethoxam 30 FS	10 ml	0.71^{b} (0.00)	0.74^{b} (0.04)	0.73^{b} (0.03)	0.76^{b} (0.07)	0.75^{ab} (0.06)	0.77^{b} (0.09)	0.78^{b} (0.10)	0.75^{b} (0.06)	0.75^{b} (0.06)	0.77^{b} (0.09)	0.71^{b} (0.00)
Thiamethoxam 70 WS	4 g	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.73^{ab} (0.03)	0.79^{ab} (0.12)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.77^{b} (0.09)	0.77^{b} (0.09)	0.71^{b} (0.00)
Chlorantraniliprole 9.3 SC + lamda cyhalothrin 4.6 CS (13.9 ZC)	2.5 ml	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.75^{b} (0.06)	0.76^{ab} (0.08)	0.75^{b} (0.07)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.72^{b} (0.01)	0.71^{b} (0.00)
Control	-	0.71^{a} (0.00)	0.81^{a} (0.16)	0.81^{a} (0.16)	0.86^{a} (0.24)	0.92^{a} (0.35)	0.97^{a} (0.44)	1.04^{a} (0.58)	1.13^{a} (0.78)	1.16^{a} (0.84)	1.19^{a} (0.91)	0.71^{a} (0.00)
CD at 5% (T)		NS	NS	NS	NS	NS	0.11	0.11	0.12	0.12	0.12	NS

Figures in parentheses are retransferred values, those outside are square root transformed values. In each column means followed by a same alphabet are not significantly different from each other

Table 3. Effect of seed treatment of various chemicals on population of ladybird beetle in *Bt* cotton

Treatments	Dose (kg^{-1} seed)	Average spider per plant- DAS										
		10	17	23	30	37	44	51	58	65	72	Pooled
Carbosulfan 25 DS	60 g	0.71^{a} (0.00)	0.73^{a} (0.03)	0.74^{a} (0.04)	0.74^{a} (0.04)	0.74^{ab} (0.05)	0.74^{b} (0.04)	0.74^{b} (0.05)	0.74^{b} (0.05)	0.75^{b} (0.06)	0.77^{b} (0.09)	0.74^{b} (0.05)
Imidacloprid 70 WG	3 g	0.71^{a} (0.00)	0.74^{a} (0.04)	0.75^{a} (0.06)	0.76^{a} (0.07)	0.77^{ab} (0.09)	0.79^{ab} (0.12)	0.75^{ab} (0.06)	0.78^{b} (0.11)	0.77^{b} (0.09)	0.77^{b} (0.09)	0.75^{b} (0.07)
Imidacloprid 48 FS	8 ml	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.75^{ab} (0.06)	0.79^{ab} (0.12)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.74^{b} (0.04)
Imidacloprid1 + Hexaconazole 20 FS	2 ml	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.75^{a} (0.06)	0.76^{ab} (0.08)	0.75^{b} (0.07)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.01)	0.72^{b} (0.02)
Thiamethoxam 30 FS	10 ml	0.71^{a} (0.00)	0.74^{a} (0.04)	0.75^{a} (0.06)	0.76^{a} (0.07)	0.72^{b} (0.02)	0.79^{ab} (0.12)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.74^{b} (0.05)
Thiamethoxam 70 WS	4 g	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.77^{ab} (0.09)	0.79^{ab} (0.12)	0.72^{b} (0.02)	0.72^{b} (0.02)	0.76^{b} (0.07)	0.77^{b} (0.09)	0.74^{b} (0.04)
Chlorantraniliprole 9.3 SC+lamda cyhalothrin 4.6 CS (13.9 ZC)	2.5 ml	0.71^{a} (0.00)	0.71^{a} (0.00)	0.71^{a} (0.00)	0.75^{a} (0.06)	0.76^{ab} (0.08)	0.75^{b} (0.07)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.71^{b} (0.00)	0.72^{b} (0.01)	0.72^{b} (0.02)
Control	-	0.82^{a} (0.17)	0.81^{a} (0.16)	0.81^{a} (0.16)	0.86^{a} (0.24)	0.92^{a} (0.35)	0.97^{a} (0.44)	1.04^{a} (0.58)	1.13^{a} (0.78)	1.16^{a} (0.84)	1.19^{a} (0.91)	0.96^{a} (0.42)
CD at 5% (T)		NS	NS	NS	NS	NS	0.11	0.11	0.11	0.12	0.11	0.05
CD at 5% (TxP)	-	-	-	-	-	-	-		-	-	-	0.10

Figures in parentheses are retransferred values, those outside are square root transformed values. In each column means followed by a same alphabet are not significantly different from each other

to test the level of significance among the various treatments.

Spiders: Pooled analysis of spider population showed that population ranging from 0.02 to 0.06 per plant in seed dressing chemical treatments while in control plots spider population was found 0.52 per plant. Significantly highest population of spider recorded in plots which were treated with imidacloprid 70 WG at 3 g kg^{-1} seed (0.06) except control. Significantly equal population (0.05) recorded in thiamethoxam 30 FS at 10 ml kg^{-1} seed, thiamethoxam 70 WS at 4 g kg^{-1} seed (3.26) and carbosulfan 25 DS at 60 g kg^{-1} seed (3.06). Among all chemical treatments, lowest spider population (0.02) recorded in plots which were treated with imidacloprid +hexaconazole 20 FS at 2 ml kg^{-1} (Table 1). Seed treatment of transgenic cotton with imidacloprid at 5 g kg^{-1} seed was not only safe but also attracted predators, *viz.,* Lynx spider, orb spider wolf and long-jawed spider in transgenic cotton (Kannan *et al*., 2004). Thakre *et al*. (2009) reported by the seed treatments of thiamethoxam at 4 g kg^{-1} seed and imidacloprid at 10 g kg^{-1} seed were proved safer to spider . Seed treatment with imidacloprid at 7.5 g kg^{-1} seed and thiamethoxam at 7.5 g kg^{-1} seed among the natural enemy complex spider was the dominant predators which were observed in good numbers in the cotton ecosystem (Sayala *et al*., 2009).

Chrysoperla: Pooled analysis of *Chrysoperla* population in various chemical treatments ranged from 0.02 to 0.07 per plant (Table 2). Significantly highest population of *Chrysoperla* was 0.52 per plant in control plots. Among seed dressing chemical treatments, significantly maximum population (0.06) of *Chrysoperla* recorded in plots which were treated with imidacloprid 70 WG at 3 g kg^{-1} seed and which was followed by thiamethoxam 30 FS at 10 ml kg^{-1} seed (0.06) and carbosulfan 25 DS at 60 g kg^{-1} seed (0.05). Seed treatment with thiamethoxam 70 WS at 4 g kg^{-1} seed (0.04) and imidacloprid 48 FS at 8 ml kg^{-1} seed (0.04) recorded equal population. Lowest population (0.02) of *Chrysoperla* recorded in plots which were treated chlorantraniliprole 9.3 SC+lamda cyhalothrin 4.6 CS (13.9 ZC) at 2.5 ml kg^{-1} seed and imidacloprid + hexaconazole 20 FS at 2 ml kg^{-1} seed. The seed treatments of thiamethoxam at 4 g kg^{-1} and imidacloprid at10 g kg^{-1} seed were proved safer to *Chrysoperla* (Thakre *et al.,* 2009). Seed treatment with imidacloprid at 7.5 g kg^{-1} seed and thiamethoxam at 7.5 g kg^{-1} seed, among the natural enemy complex *Chrysoperla* was the dominant predators which were observed in good numbers in the cotton ecosystem (Sayala *et al*., 2009). Seed treatment with imidacloprid 70 WS at 7 g kg^{-1} seed was conserved more number of *Chrysoperla* (Jayaprakash *et al*., 2015).

Ladybird beetle: In pooled analysis over period (Table 3), the population of ladybird beetle in various seed dressing chemical plots was ranging from 0.02 to 0.07. Population of ladybird beetle was recorded significantly maximum (0.42) per plant in control plots. Among all chemical treatments, significantly maximum population (0.07) of ladybird beetle recorded in plots which were treated with imidacloprid 70 WG at 3 g kg^{-1} seed it was followed by carbosulfan 25 DS at 60 g kg^{-1} seed (0.05). Significantly equal population was recorded in seed treatment with thiamethoxam 70 WS at 4 g kg^{-1} seed (0.04), and imidacloprid 48 FS at 8 ml kg^{-1} seed (0.04). Lowest population (0.02) of ladybird beetle recorded in plots which were treated with imidacloprid+hexaconazole 20 FS at 2 ml kg^{-1} seed and chlorantraniliprole 9.3 SC+lamda cyhalothrin 4.6 CS (13.9 ZC) at 2.5 ml kg^{-1} seed. The seed treatments of thiamethoxam 70 WS at 4 g kg^{-1} seed and imidacloprid 70 WS at10 g kg^{-1} seed were proved safer to ladybird beetle (Thakre *et al.,* 2009). Seed treatment with imidacloprid 70 WS at 7 g kg^{-1} seed was conserved more number of ladybird beetle (Jayaprakash *et al*., 2015).

Yield: Seed cotton yield was ranging from 17.90 to 21.69 q ha^{-1}. Imidacloprid 70 WG at 3 g kg^{-1} seed, recorded significantly higher yield (21.69 q ha^{-1}) and it was at par with carbosulfan 25 DS at 60 g kg^{-1} seed (19.99 q ha^{-1}), thiamethoxam 70 WS 4 g kg^{-1} seed (19.96 q ha^{-1}), imidacloprid 48 FS 8 ml kg^{-1} seed (19.66 q ha^{-1}), chlorantraniliprole (9.3%) + lamda cyhalothrin (46% ZC) at 2.5 ml kg^{-1} (18.95 q ha^{-1}), and imidacloprid (18.5%) + hexaconazole (1.5% FS) at 2 ml kg^{-1} seed (17.90 q ha^{-1}). Imidacloprid (18.5%) + hexaconazole (1.5% FS) at 2 ml kg^{-1}recorded lowest yield (15.42 q ha^{-1}), even lower than the control plots (16.95 q ha^{-1}). Amin *et al*. (2008) reported that seed treatment with gaucho at all threshold levels gave significantly higher yield and profitable benefit cost ratio. Seed cotton yield was maximum in imidacloprid 70 WS seed treatment with at 5.5 and 4.5 g kg^{-1}. CB9 cotton cultivar gave a higher benefit cost ratio, when seed were treated with imidacloprid 70 WS at 5.5 g kg^{-1} seed fuzzy seed (Hossain *et al*., 2012). Rao *et al*. (2014) reported maximum seed cotton yield in seed treatment with imidacloprid 70 WS followed by thiamethoxam 70 WS. Sanganna (2018) also recorded maximum seed cotton yield in seed treatment with imidacloprid 75 WS at 3.5 g kg^{-1} of seed followed by seed treated with carbosulfan 25 DS at 30 g. kg^{-1}.

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OBITUARY

Professor Rajan Asari
(1941-2023)

Professor Rajan Asari former Professor of Entomology, Kerala Agricultural University, born on May 28th, 1941 in the Pathanamthitta district, Kerala, passed away on 08-05-2023. He was the eldest son of Sri Ayyappan Asari P.A. and Smt. Bhargavi Ammal. He initially took up an administrative job at the Agriculture College, Vellanikkara, Trissur through the Public Service Commission. However, due to his deep-rooted passion for science and education, he took long leave to pursue higher studies in Zoology in the University College, Trivandrum and further in the Agricultural College Vellayani, Trivandrum for an MSc in Entomology.

Professor Rajan Asari was selected as a lecturer at the same college, soon after completing post-graduation. Throughout his career, he remained dedicated to the field of Zoology, particularly focusing on entomology and taxonomy. He often assisted students, faculty and researchers from various academic institutions in identifying insect species. Additionally, he had expertise in rodent biology.

During his service in Kerala Agricultural University, he worked at several important centres across Kerala, including the Integrated Farming Systems Research Station in Thiruvanandapuram, the Regional Agricultural Research Station in Kasargod, Kerala Agricultural University in Thrissur, and the Regional Agricultural Research Station in Kottayam. He constantly updated himself with the latest scientific inventions and discoveries, extending his passion beyond biological sciences to various other disciplines. He retired as a Professor of Entomology from the College of Agriculture, Vellayani, Thiruvananthapuram.

Professor Rajan Asari even pursued a course in the German language and obtained a diploma in homeopathic medicine. With a clear understanding of both scientific and spiritual aspects, he also earned a doctoral degree in Vasthu Sasthra after retirement.

He was one of the active members of the Executive Committee of the Association for Advancement of Entomology. The sudden demise of Professor Rajan Asari is a great loss to the AAE.

Executive Committee
Association for Advancement of Entomology

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