# A new species of pterodectine feather mites (Acarina, Analgoidea, Proctophyllodidae) from the Little Spiderhunter Arachnothera Iongirostra (Passeriformes, Nectariniidae) in Meghalaya, India 

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#### Abstract

The article describes a new species of the feather mite subfamily Pterodectinae from the Little Spiderhunter Arachnothera longirostra Temminck, 1826 (Passeriformes, Nectariniidae) in India. Pedanodectes angustilobus sp. n. differs from all known Pedanodectes species by having opisthosomal lobes short, at base wider than long, roughly rounded apically in males, and strongly elongated and narrowed lobar region with wide terminal appendages in females. A key to species of the genus Pedanodectes is presented.


## Keywords

Pterodectinae, Pedanodectes angustilobus, new species, systematics

## Introduction

Feather mites are commensals or ectoparasites permanently living on birds. In India, the diversity of feather mites is poorly investigated, and only 26 species have been mentioned so far in various taxonomic papers (Oudemans 1904, Bonnet 1924, Gaud and Mouchet 1963, Atyeo et al. 1972, Gaud 1972, McClure and Ratanaworabhan 1973, Gaud and Atyeo 1976, 1987, Santana 1976, Peterson et al. 1980, D'Souza and

Jagannath 1982, Atyeo 1984, Gaud et al. 1985, 1988, Dabert and Ehrnsberger 1998, Dabert 2003, Mironov et al. 2002, Putatunda et al. 2004, Constantinescu et al. 2014). Atyeo (in McClure and Ratanaworabhan 1973) reported five new species of feather mites collected in Asia from the host Arachnothera longirostra (including Pedanodectes species), but unfortunately this material has never been described.

In this paper, we describe a new species of Pedanodectes found on $A$. longirostra in India and we present a key for all species of this genus.

## Materials and methods

The material used in the present paper was collected in Meghalaya (India), in January 2014. The birds were trapped by means of ornithological mist nets, identified and visually checked for the presence of mites and, after mites were collected, released back to the wild. Mite specimens were placed in tubes with $95 \%$ ethanol. Later, in the laboratory, the mite specimens were cleared in lactic acid and mounted on microscope slides in Hoyer's medium. Drawings were made using an Olympus CX21 microscope, using a camera lucida drawing device.

The bird specimens were identified according to Rasmussen and Anderton (2012) and Grimmett et al. (2011), and the taxonomy of the birds used in the present paper follows Clements et al. (2013). The description of new species is given according to the current format used for species of pterodectine mites (Mironov and Fain 2003, Valim and Hernandes 2006, Mironov 2006, Mironov et al. 2008). The body chaetotaxy of mites follows that of Griffiths et al. (1990) with the modifications by Norton (1998) concerning coxal setae, and the chaetotaxy of legs follows Gaud and Atyeo (1996). The measuring techniques of particular structures used in the present paper were recently described by Mironov and Proctor (2009). We give the full set of measurements for the holotype (male) and a range of measurements for all corresponding paratypes. All measurements are in micrometers $(\mu \mathrm{m})$. The holotype and all paratypes of the new species are deposited in the Acarological Collection of the "Grigore Antipa" National Museum of Natural History, Bucharest, Romania.

## Results

## Family Proctophyllodidae Trouessart \& Mégnin, 1884 Subfamily Pterodectinae Park \& Atyeo, 1971

Genus Pedanodectes Park \& Atyeo, 1971

The genus currently includes six species, associated with birds of the order Passeriformes (families Nectariniidae, Malaconotidae, Cisticolidae and Platysteiridae) in Africa (see Table 1).

Table I. Pedanodectes species and their host associations.

| Mite species | Host species | Host family | Location | References |
| :---: | :---: | :---: | :---: | :---: |
| P. hologaster (Gaud, 1953) | Chalcomitra senegalensis (Linnaeus)*, Chalcomitra fuliginosas (Bechstein) | Nectariniidae | Central African Republic | Gaud 1953; Park and Atyeo 1971 |
| P. andrei (Till, 1954) | Tchagra senegalus (Linnaeus) | Malaconotidae | Mozambique | Till 1954 |
|  <br> Mouchet, 1957) | Deleornis fraseri cameroonensis (Bannerman) | Nectariniidae | Cameroon | Gaud and Mouchet 1957 |
| P. marginatus Mironov \& Kopij, 1997 | Camaroptera brachyura <br> (Vieillot) | Cisticolidae | South Africa | Mironov and Kopij 1997 |
| P. latior Mironov \& Kopij, 1997 | Platysteira peltata Sundevall | Platysteiridae | South Africa | Mironov and Kopij 1997 |
| P. blaszaki Mironov, 2008 | Cinnyris cupreus (Shaw) | Nectariniidae | South Africa | Mironov 2008 |
| P. angustilobus sp. n. | Arachnothera longirostra (Latham) | Nectariniidae | India | Present paper |

## Pedanodectes angustilobus Constantinescu, sp. n.

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Figs 1-4

Type material. Male holotype (ANA256), 3 male (ANA257, ANA258, ANA259) and 4 female (ANA260, ANA261, ANA262, ANA263) paratypes from the Little Spiderhunter Arachnothera longirostra Temminck, 1826 (Passeriformes, Nectariniidae); INDIA: Meghalaya, Jaintia Hills, Shnongrim village, ( $25^{\circ} 21^{\prime} 12.36^{\prime \prime N}, 92^{\circ} 31^{\prime} 3.06^{\prime \prime} \mathrm{E}$ ); 1151 m alt; 24.01.2014, collector Costică Adam.

Description. MALE (Figs 1A, B; 3A-D; holotype, range for 3 paratypes in parentheses): Length of idiosoma 316 (308-317), width 100 (100-104), length of hysterosoma 212 (207-212). Prodorsal shield divided into two parts by transversal band of soft tegument bearing setae se and si, antero-lateral extensions short and rounded, posterior margin slightly convex in median part, total length of shield 102 (102106), greatest width 90 (84-90), surface without ornamentation (Fig. 1A). Scapular setae se separated by 38 (36-40). Humeral shields absent, setae $c p$ situated ventrally, setae $c 2$ situated dorsally, in anterior angles of hysteronotal shield. Subhumeral setae c3 lanceolate, $20(19-20) \times 6(5-6)$. Length of hysteronotal shield from anterior margin to bases of setae h2 195 (199-203), greatest width in anterior part 88 (80-86), anterior margin concave, anterior angles acute, surface without ornamentation with strongly sclerotized transverse fold between hl setae. Opisthosomal lobes short, at base wider than long, roughly rounded apically, lobar apices bearing setae h2. Terminal cleft almost semicircular, with narrow membranous margin in anterior part, length of terminal cleft $10(12-13)$. Supranal concavity absent. Hysteronotal setae c1, d1, e1 absent; setae h3 narrowly lanceolate, length 24 (24-25), greatest width 6 (4-6); setae $h 2$ represented by macrosetae, length 130 (132-140), greatest width 5 (4-5); setae ps2 slightly thickened, 18 (12-16) long; setae ps1 filiform, minute, length 6 (5-6), situated slightly anterior to bases of setae $h 3$ and $h 2$, approximately equidistant


Figure I. Pedanodectes angustilobus sp. n., male holotype: A dorsal view of idiosoma B ventral view of idiosoma.
from inner and outer margins of opisthosomal lobe. Dorsal measurements: se-c2 72 (66-70), c2-d2 92 (90-104), d2-e2 68 (60-66), e2-h3 32 (32-34), h1-h3 10 (10-12), h2-h2 38 (38-42), h3-h3 26 (26-28), ps2-ps2 50 (46-49). Epimerites I fused into a Y, sternum about $1 / 2$ of the total length of epimerites, posterior end of sternum with pair


Figure 2. Pedanodectes angustilobus sp. n., female paratype: $\mathbf{A}$ dorsal view of idiosoma $\mathbf{B}$ ventral view of idiosoma.
of postero-lateral extensions not connected to epimerites II. Epimerites II short, not extending to level of sejugal furrow. Coxal fields I-III open, without wide sclerotized areas. Epimerites IVa long, their anterior ends extending to midlevel of epimerites IV, their basal parts connected by semicircular transverse sclerite (supposedly genital shield) and almost completely encircling genital apparatus (Fig. 1B). Genital arch 11 (11-12) long, $14(13-14)$ wide, basal sclerite of genital apparatus large, poorly sclerotized; aedeagus long, extending almost to anterior margin of terminal cleft, length of aedeagus from its anterior bend to tip $74(72-78)$. Genital papillae indistinct, adanal shields absent. Anal suckers $11(11-12)$ in diameter, corolla without indentations. Opisthoventral shields occupying lateral margin of opisthosoma, with narrow inner projection bearing seta $p s 3$. Ventral measurements: $3 a-4 b 22$ (25-30), 4b-4a 50 (44-50), $4 a-g 24(24-28), g-p s 332$ (28-31), ps3-ps3 40 (32-38), ps3-h3 24 (23-24).

Legs I slightly longer and thicker than legs II, femora I and II with ventro-basal crests (Fig. 3A, B). Seta $m G I I$ strongly thickened in basal half. Tarsus IV 18 (19-22) long, with apical claw and with small apico-ventral extensions bearing seta $r$ and $w$; setae $d$, $e$ button-like, seta $d$ bigger than $e$, situated in basal and apical parts of segment, respectively (Fig. 3D). Length of solenidia: $\omega 1$ II 15 (14-15), $\omega 1$ II 12 (13-16), $\varphi \mathrm{I} 42$ (34-46), $\varphi$ II 29 (32-34), ¢III 22 (22-24), $\varphi$ IV 32 (28-32).

FEMALE (Figs 2A, B; 4A-E; range for 4 paratypes): Length of idiosoma excluding terminal appendages 436-456, width 118-124, and length of hysterosoma 312340. Prodorsal shield divided into two parts by transversal band of soft tegument, bearing setae se and si, antero-lateral extensions short and angular, posterior margin concave, total length of shield 118-122, greatest width 104-112, surface without ornamentation (Fig. 2A). Scapular setae se separated by 48-50. Humeral shields absent, setae $c p$ situated ventrally, setae $c 2$ situated dorsally, in anterior angles of hysteronotal shield. Subhumeral setae $c 3$ lanceolate, $17-20 \times 5-6$. Hysteronotal shield divided into anterior hysteronotal shield and lobar shield. Anterior hysteronotal shield roughly rectangular, with anterior margin concave, greatest length 204-208, greatest width in anterior part 100-106, surface without ornamentation. Lobar shield elongated and narrowed, with well-developed lateral extensions bearing setae $h 2$, length of lobar shield 86-92, width at level of setae $h 258-62$. Terminal cleft narrow, almost parallelsided, with narrow elliptical part in anterior third, length 72-76, greatest width 3-4. Supranal concavity absent. Terminal appendages wide, their width in basal half similar to that of lobes. Hysteronotal setae $c 1, d 1, e 1$ absent; setae $h 1$ situated on lobar shield at level of anterior end of terminal cleft, setae $h 2$ spindle-shaped, without terminal filaments, $38-40 \times 6-9$. Setae ps1 situated approximately equidistant from outer and inner margins of opisthosomal lobes, setae h3 6-8 long. Dorsal measurements: se-c2 88-94, c2-d2 100-112, d2-e2 86-94, e2-h2 36-44, h2-h3 48-52, h1-h2 16-22, h1h1 18-22, h2-h2 36-40, h3-h3 20-24. Epimerites I fused as a Y, posterior end of sternum with small rounded lateral extensions not reaching epimerites II. Coxal fields I, II open, without heavily sclerotized areas, outer margins of epimerites I and II with narrow sclerotized areas (Fig. 2B). Epimerites IVa rudimentary. Translobar apodemes of opisthosomal lobes present, fused to each other anterior to terminal cleft. Epigynum

## A



Figure 3. Pedanodectes angustilobus sp. n., details: A-D legs I-IV of male, respectively, dorsal view.


Figure 4. Pedanodectes angustilobus sp. n., details: A-D legs I-IV of female, respectively $\mathbf{E}$ spermatheca and spermaducts, dorsal view. Abbreviations: pd - primary spermaduct; s - spermatheca; sd - secondary spermaduct.
horseshoe-shaped, outer margin with lateral extensions, greatest width 52-60. Head of spermatheca as in Fig. 4E, primary spermaduct with three different enlargements: balllike enlargement near very head of spermatheca; moderate enlargement with verrucous external surface in proximal $1 / 5$ of this duct; conical enlargement at copulatory opening. Secondary spermaducts short, not longer that ball-like enlargement of primary spermaduct. Copulatory opening situated ventral, posterior to anal opening. Distance between pseudanal setae: ps2-ps2 24-286, ps3-ps3 24-29, ps2-ps3 10-14.

Legs I longer and thicker than legs II, femora II with ventro-basal crests, genua III with dorso-basal crest (Fig. 4A-C). Genual setae $m G I$ and $m G I I$ noticeably thickened in basal half. Length of solenidia: $\omega 1$ II 17-18, $\omega 1$ II 13-16, $\varphi$ I 45-56, $\varphi$ II 32-38, $\varphi$ III 22-27, $\varphi$ IV 7-8.

Etymology. The specific epithet refers to the narrowed lobar region of the female, and it is an adjective in the nominative singular.

Remarks. Pedanodectes angustilobus sp. n. clearly differs from all previously described Pedanodectes species because its males have well expressed opisthosomal lobes, and females have elongated and narrowed lobar region, with wide terminal appendages. Among previously known species of the genus, males of $P$. angustilobus sp. n. appear to be closest to that of $P$. mesocaulus (Gaud \& Mouchet, 1957) from Deleornis fraseri cameroonensis (Bannerman, 1921) (Passeriformes: Nectariniidae). Males of both species have setae ps1 situated antero-lateral to the adanal suckers, epimerites I fused into Y, similar shape of opisthoventral shields and epimerites IVa with narrow anterior projections. Males of $P$. angustilobus sp. n. are easily to distinguish from those of $P$. mesocaulus by the following features: epimerites IVa have free the anterior projections and are connected posterior to the genital arch by a transversal sclerite, the tip of aedeagus does not extend beyond the posterior margin of idiosoma. In males of $P$. mesocaulus, the anterior projections of epimerites IVa are fused forming a pregenital sclerite while the transverse sclerite connecting the bases of these epimerites is absent, the tip of aedeagus extends beyond the posterior margin of idiosoma. Females of the new species are clearly different from those of the other species of the genus by the following unique combination of characters: the lobar region has the same width in anterior and posterior part; the terminal cleft is parallel-sided, with the margins almost touching, except for the anterior one third; and the terminal appendages are thick, their basal half is approximately as wide as opisthosomal lobes. In females of the other Pedanodectes species, the lobar region in the anterior part is wider than in the posterior part and the terminal appendages are narrower than lobes. The terminal cleft in females of the other species has the following shape: with the lateral margins parallel and almost touching in $P$. andrei and P. mesocaulus, with lateral margins sinuous and almost touching in certain parts in P. marginatus and $P$. latior; as a narrow inverted V in P. blaszaki, and as a narrow inverted U in $P$. hologaster.

## Discussion

According to the diagnosis of the genus Pedanodectes, some authors considered that males of this genus practically have no opisthosomal lobes and setae ps3 are usually situated lateral or postero-lateral to the adanal suckers (Gaud and Atyeo 1996, Mironov 2008, Hernandes and Valim 2014). However in the original definition of this genus, Park and Atyeo (1971), apparently based on a some undescribed material they had on the hand, mentioned that the presence of weakly developed opisthosomal lobes and the position of setae ps3 antero-lateral to the adanal suckers can be found in some species. The new species found on $A$. longirostra in India and described in this paper demonstrates the example of a Pedanodectes species having distinct opisthosomal lobes and setae ps3 situated antero-lateral to the anal suckers in males.

## Key to males of Pedanodectes

1 Epimerites I fused V-likely.......................................................................... 2

- Epimerites I fused Y-likely .......................................................................... 4

2 Postero-lateral extensions of epimerites I connected to epimerites II ...P. latior

- Postero-lateral extensions of epimerites I not connected to epimerites II..... 3

3 Tip of aedeagus extending beyond posterior margin of idiosoma ...P. andrei

- Tip of aedeagus not extending beyond posterior margin of idiosoma.
P. bologaster

4 Postero-lateral extensions of epimerites I present ........................................ 5

- Postero-lateral extensions of epimerites I absent.......................... P. blaszaki

5 Postero-lateral extensions of epimerites I connected to epimerites II .P. marginatus

- Postero-lateral extensions of epimerites I not connected to epimerites II..... 6

6 Opisthosomal lobes present; tip of aedeagus not extending beyond posterior margin of idiosoma
P. angustilobus sp. n.

- Opisthosomal lobes absent; tip of aedeagus extending beyond posterior margin of idiosoma.
P. mesocaulus


## Key to females of Pedanodectes

1 Width of terminal appendages in anterior half similar to that of opisthosomal lobes ......................................................................... P. angustilobus sp. n.

- Terminal appendages narrower than opisthosomal lobes............................. 2

2 Lateral margins of terminal cleft spaced in some parts or on their entire length

- Lateral margins of terminal cleft almost touching on their entire length ..... 6

3 Prodorsal shield split into anterior and posterior pieces at level of scapular setae
P. blaszaki

- Prodorsal shield entire ................................................................................. 4

4 Lateral margins of prodorsal shield entire.................................................... 5

- Lateral margins of prodorsal shield with deep incisions around setae se......... P. latior

5 Epimerites I fused V-likely ..................................................... P. bologaster

- Epimerites I fused Y-likely .....................................................P. marginatus

6 Posterior margin of anterior hysteronotal shield straight ................P. andrei

- Posterior margin of anterior hysteronotal shield concave........ P. mesocaulus


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# A new species of Paramunida Baba, 1988 from the Central Pacific Ocean and a new genus to accommodate P. granulata (Henderson, I885) 

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#### Abstract

The genus Paramunida belongs to the most diverse family of galatheoids and it is commonly reported from the continental slope across the Indian and Pacific Oceans. Examination of material collected by the NOAA RV Townsend Cromwell Cruise near Christmas (Kiritimati) Island, Kiribati, revealed the existence of a new species of Paramunida (P. haigae), which represents the fourth record of the genus for the Central Pacific. Furthermore, recent efforts to unravel phylogenetic relationships and diversification patterns in Paramunida revealed P? granulata (Henderson, 1885) to be the most basally diverging taxon within the genus. This species is clearly distinguished from other species of Paramunida by the spinulation of the carapace and the length of the distomesial spine of the second antennal peduncle article, which in combination with a high level of genetic divergence suggest that this species represents a separate monotypic lineage. A new genus, Hendersonida gen. n., is proposed to accommodate this species based on morphological and molecular evidence. An updated dichotomous identification key for all species of Paramunida is presented.


## Keywords

New species, Paramunida, new genus, Hendersonida, Munididae, squat lobster, morphology, phylogeny

## Introduction

Squat lobsters are abundant and highly visible crustaceans in the deep sea (Baba et al. 2008). Our understanding of the taxonomy and phylogeny of this speciose group has been revolutionized in the last three decades, mainly thanks to the numerous MU-SORSTOM-TDSB expeditions (Richer de Forges et al. 2013). Major rearrangements at higher classifications (Ahyong et al. 2010; Schnabel and Ahyong 2010), the description of 14 new genera (Macpherson and Baba 2011; Macpherson and Robainas-Barcia 2013) and many new species (Baba 2005) demonstrate the outstanding efforts of taxonomists to accurately describe and interpret squat lobster diversity.

The genus Paramunida Baba, 1988, recently transferred to the family Munididae (Ahyong et al. 2010), was established by K. Baba to accommodate seven species morphologically close to Munida Leach, 1820, but characterized by having a shortrostrum, carapace covered with spinules or granules, indistinct transverse striae, a well-developed distomesial spine on the first article of the antennal peduncle and the male gonopods present only on the second abdominal somite. In following years, several species were described from New Caledonia and adjacent waters (e.g., Cabezas et al. 2009; Macpherson 1993, 1996) and the most recent taxonomic revisions revealed the surprising existence of 14 new species (Cabezas and Chan 2014; Cabezas et al. 2010).

The genus includes 40 genetically distinct yet morphologically very similar species (Cabezas et al. 2010). Interestingly, the species Paramunida granulata (Henderson, 1885) is unique in having a granulated carapace and the distomesial spine of antennal article 2 very long, almost reaching the end of the anterior prolongation of article 1 (Baba 1988; Cabezas et al. 2010). A previous phylogenetic study reported this species as the earliest offshoot within the genus in the early Oligocene (Cabezas et al. 2012), which along with the above-mentioned marked morphological differences and a high genetic divergence indicate that this species followed an independent evolutionary trajectory (Cabezas et al. 2010; Cabezas et al. 2012; Machordom and Macpherson 2004). To reflect these findings, we herein propose a new genus, Hendersonida gen. n.

Furthermore, during a recent visit to Los Angeles County Museum of Natural History, some Paramunida specimens previously identified as Munida hawaiiensis (Baba, 1981) were discovered to be an undescribed species. The material examined was collected by the NOAA ship RV Townsend Cromwell in Christmas (Kiritimati) Island, Kiribati, in the Central Pacific Ocean. To date, only the endemic species $P$. hawaiiensis (Baba, 1981) from Hawaii, P. spatula Macpherson, 2006 from the Austral Archipelago and P. echinata Macpherson, 1999 from the Marquesas Islands are known from Central Pacific waters. Therefore, the new species described here is the fourth record of the genus for the region. Finally, we present an updated dichotomous key to species of Paramunida.

## Material examined

We studied material collected by the NOAA RV Townsend Cromwell Cruise during Febru-ary-March 1973 in the Central Pacific Ocean. The new described species in this study is deposited in Los Angeles County Museum of Natural History, Los Angeles (LACM). The terminology used mainly follows Baba et al. (2011). The size of the carapace is indicated as the postorbital carapace length measured along the dorsal midline from the posterior margin of the orbit to the posterior margin of the carapace. The length of the antennular and antennal articles is measured excluding distal spines along their lateral margins; the width is measured at midlength of each article. The abbreviations used are: P1 = first pereopod (chelipeds), $\mathrm{P} 2-\mathrm{P} 4=$ second to fourth pereopods (first to third walking legs).

## Molecular data

The phylogenetic tree presented in this study was obtained from Cabezas and Chan (2014). The new species described here failed amplification because material was preserved in formalin, so no molecular comparison is provided.

## Family Munididae Ahyong, Baba, Macpherson \& Poore, 2010

## Genus Paramunida Baba, 1988

Paramunida Baba, 1988: 175 (gender: feminine). - Poore 2004: 239. - Baba 2005: 197. - Baba et al. 2008: 171 (compilation of species). - Baba et al. 2009: 277. - Cabezas et al. 2010: 5. - Macpherson and Baba 2011: 60.

Diagnosis. (modified from Baba et al. 2009) Carapace as long as wide; dorsal surface covered with spinules, indistinct transverse striae; posterior margin with some spines; rostrum short, basally subtriangular, distally ending in spine; supraocular spines small, clearly not reaching midlength of rostrum and falling short the end of the corneae; margin between rostral and supraocular spines straight or slightly concave; anterolateral spines well developed at front near anterolateral angles, reaching the level between rostrum and supraocular spines; lateral margins with some spines. Eyes large, maximum corneal diameter about one-third distance between anterolateral spines. Lateral margin of antennular article 1 with distal slender portion about half as long as proximal inflated portion, with 2 distal small spines. Antennal peduncle with anterior prolongation of article 1 spiniform; article 2 with distomesial spine never reaching end of anterior prolongation of article 1. P1-P4 long and slender, squamate; P2-P4 dactyli slender, curved and unarmed on flexor margin. Male gonopods only present on the second abdominal somite.

Type species. Paramunida setigera Baba, 1988; by original designation.
Remarks. The Munida scabra group was recognized by K. Baba in 1981. It included five species - M. scabra (Henderson, 1885), M. granulata (Henderson, 1885), M. proxima (Henderson, 1885), M. tricarinata (Alcock, 1894) and M. hawaiiensis (Baba, 1981) - all characterized by having a short rostrum, carapace without transverse ridges covered by spinules and granules, the antennal peduncle with a well-developed anterior prolongation of article 1 , and male gonopods absent from first abdominal somite. All these peculiarities suggested that the scabra group represented an independent lineage from Munida, but further investigations were recommended. Later work confirmed the taxonomic significance of this group and the genus Paramunida Baba, 1988 was formally described in a report on the chirostylid and galatheid crustaceans from the "Albatross" Philippine Expedition (Baba 1988). This new genus accommodated the species belonging to the scabra group plus two new described species $P$. longior and $P$. setigera. Paramunida was substantially enlarged through the MUSORSTOMTDSB expeditions in waters around the Philippines, Indonesia and New Caledonia (Macpherson 1993; Baba 2005), Wallis and Futuna (Macpherson 1996), eastern Australia (Ahyong and Poore 2004), Fiji and Tonga (Macpherson 2004), French Polynesia (Macpherson 2006), New Zealand (Ahyong 2007), Taiwan and Japan (Baba et al. 2009; Macpherson and Baba 2009), and the Solomon Islands (Cabezas et al. 2009). Most recently, the taxonomic revision of the genus resulted in the description of 11 new species (Cabezas et al. 2010), and examination of material collected during the PANGLAO expeditions added three new ones namely $P$. akaina, $P$. aspera and $P$. aurora (Cabezas \& Chan, 2014). After the taxonomic rearrangements proposed in the present study the genus Paramunida comprises 40 species (see below).

## Paramunida baigae sp. n.

http://zoobank.org/5ECE748F-15AA-4AF9-9767-866162CB3B58
Figs 1, 2, 3

Material examined. Holotype: Christmas (Kiritimati) Island, Line Islands, Kiribati, $01^{\circ} 51.3^{\prime} \mathrm{N}, 157^{\circ} 30.4^{\prime} \mathrm{W}$, February-March 1973, 183 m (NOAA RV Townsend Cromwell Cruise): male, 16.6 mm (LACM-CR1973-3312). Paratypes: collected with holotype: 9 males $11.4-17.2 \mathrm{~mm}$ ( 2 broken), 3 females, $13.5-14.1 \mathrm{~mm}, 2$ ovigerous females, 11.6-14.2 mm (LACM-CR1973-3313).

Description. Carapace: As long as broad, dorsal surface covered with spinules; each spinule usually on short arcuate striae, with few short uniramous setae. Epigastric region with 2 spines, each behind supraocular spine; with median row of spinules behind rostral spine. Mesogastric region with median row of 3 small spines. Anterior branch of cervical groove with short setae. Cervical groove distinct. Cardiac and anterior branchial regions slightly circumscribed. Cardiac region with a median row of 3 small spines, first thicker than others. Each branchial region with row of spines near cardiac region. Frontal margin slightly concave. Lateral margins convex, with some


Figure I. Paramunida haigae sp. n. male holotype, 16.6 mm (LACM-CR1973-3312). Christmas (Kiritimati) Island. A carapace and abdomen, dorsal view $\mathbf{B}$ carapace, lateral profile $\mathbf{C}$ sternum $\mathbf{D}$ left antennule and antenna, ventral view $\mathbf{E}$ right maxilliped 3, lateral view. Scale: 5 mm (scale 1 for $\mathbf{A - C}, \mathbf{E}$; scale 2 for $\mathbf{D}$ ).
spines and iridescent setae on anterior half. Anterolateral spine well developed, reaching sinus between rostral and supraocular spines. Rostral spine spiniform, with thin dorsal longitudinal carina; supraocular spines well developed and slender and shorter than rostrum (Figs 1A, B, 3).

Sternum: Thoracic sternite 4 with few arcuate striae; sternites $5-7$ smooth (Fig. 1C).
Abdomen: Abdominal somites 2-3 each with 4 well-developed spines on anterior ridge, posterior ridge with 2 median spines. Abdominal somite 4 with 4 spines on an-
terior ridge; posterior ridge with distinct single median spine. Ridges with numerous spinules and a few small spines (Fig. 1A).

Eyes: Maximum corneal diameter more than one-third distance between bases of anterolateral spines.

Antennule: Article 1 slightly exceeding corneae, with distomesial spine small and as long as distolateral; about twice longer than wide and with fringe of long setae along lateral margin; lateral margin with distal slender portion about half as long as proximal convex portion (Fig. 1D).

Antenna: Anterior prolongation of article 1 overreaching antennular peduncle by about one-third of its length. Article 2 about twice length of article 3 and twice longer than wide, ventral surface with scales; distomesial spine spiniform without tuff of setae, overreaching end of article 3, not reaching end of antennal peduncle, reaching midlength of anterior prolongation of article 1 , and clearly not reaching end of basal article of antennule, distolateral spine not reaching end of article 3; article 3 about 1.5 times longer than wide and unarmed (Fig. 1D).

Maxilliped 3: Ischium about twice length of merus measured along extensor margin, flexor margin bearing long distal spine; merus with well-developed median spine on flexor margin; extensor margin unarmed (Fig. 1E).

Pereopod 1 (cheliped): Long and slender, squamate, between 6.5-7.5 times carapace length; carpus about as long as palm, and $7-10$ times longer than high; palm 1.1-1.5 times fingers length. Base of carpus without bundle of setae (Fig. 2A-C).

Pereopods 2-4 (P2 lacking in holotype): Long and slender, with scales on lateral sides of meri, carpi and propodi; scales with short setae. P2 2.5-3.5 times carapace length, merus 1.1-1.6 times longer than carapace, about $8-10$ times as long as high, 4 times as long as carpus and 1.5 times as long as propodus; propodus about $7-10$ times as long as high, and 1.4-1.7 times dactylus length. Merus with well-developed spines on extensor margin, increasing in size distally; flexor margin with few spines and one well developed distal spine; row of small spines along flexolateral margin. Carpus with few small extensor spines, small distal spine on extensor and flexor margin. Propodus with small movable flexor spines. Dactylus compressed, slightly curved, with longitudinal carinae along mesial and lateral sides, flexor border unarmed. End of P2 carpus not reaching end of P1 merus. P3 with similar spination and article proportions as P2; propodus slightly longer than P 2 propodus, merus and dactylus as long as those of P 2 . P4 as long as P2; merus 1.1-1.3 times carapace length; propodus and dactylus slightly longer than those of P 3 ; merocarpal articulation clearly exceeding end of anterior prolongation of article1 of antennal peduncle (Fig. 2D-G).

Etymology. This species is dedicated to the renowned carcinologist Janet Haig (1925-1995) who first classified the material examined.

Remarks. Paramunida haigae sp. n. closely resembles $P$. antares Cabezas, Macpherson \& Machordom, 2010 from New Caledonia. The new species is readily separated from $P$. antares in having the rostrum spiniform rather than triangular. Moreover, the mesogastric region in $P$. antares has 3 well-developed spines, but these spines are very small in $P$. haigae sp. $n$. The two species also differ in the article 2 of


Figure 2. Paramunida haigae sp. n. male holotype, 16.6 mm (LACM-CR1973-3312). Christmas (Kiritimati) Island. A left merus P1, dorsal view B left carpus P1, dorsal view $\mathbf{C}$ left P1, palm and fingers, dorsal view $\mathbf{D}$ right P 3, lateral view $\mathbf{E}$ right P 3 dactylus $\mathbf{F}$ male paratype, 11.5 mm (LACM-CR1973-3313), right P2, lateral view. G left P4, lateral view. Scale: 5 mm (scale 1 for $\mathbf{A}-\mathbf{C}, \mathbf{D}, \mathbf{G}$; scale 2 for $\mathbf{F}$; scale 3 for $\mathbf{E}$ ).
the antennal peduncle: twice as long as wide in the new species but only 1.5 times in $P$. antares. Finally, the distomesial spine of antennal article 2 clearly overreaches the end of article 3 in the new species, but this spine only reaches the end of the article 3 in $P$. antares.

The new species is also very close to $P$. achernar Cabezas, Macpherson \& Machordom, 2010 from Tonga. Paramunida haigae sp. n. can be distinguished from $P$. achernar by having 3 small mesogastric spines (vs. 3 well-developed spines in $P$. achernar). Furthermore, the anterior prolongation of antennal article 1 is clearly longer in $P$. haigae sp. n., overreaching the antennular peduncle by about one-third of its length but only by one-fourth in $P$. achernar, and the distomesial spine of antennal article 2 overreaching the end of article 3 in the new species (vs. only reaching the end of the article 3 in $P$. achernar). Finally, the merocarpal articulation of P3 clearly exceeds the anterior prolongation of the antennal article 1 in the new species, only slightly exceeding the anterior prolongation in $P$. achernar.

Of the regional Central Pacific Paramunida species, P. haigae sp. n. can be easily distinguished from P. hawaiiensis Baba, 1981 from Hawaii in having the rostral spine larger than supraocular spines instead of smaller or at most equal to supraocular spines. Furthermore, the distomesial spine of article 2 reaches end of antennal peduncle in $P$. hawaiiensis but never reaches it in the new species. The new species can also be easily distinguished from P. echinata Macpherson, 1999 from Marquesas Islands in having the rostral spine spiniform instead of triangular. Finally, $P$. haigae sp. n. is also easily distinguishable from P. spatula Macpherson, 2006 from the Austral archipelago by the shape of the anterior prolongation of antennal article 1.

Distribution. Christmas (Kiritimati) Island, Kiribati, at 183 m depth.

## Genus Hendersonida gen. n.

http://zoobank.org/C149D702-673C-48D8-BD97-DD6F20A8B59B
Type species. Munida granulata Henderson, 1885; here designated and by monotypy.
Diagnosis. Carapace as long as wide; dorsal surface granulose, with some scattered spines and small spinules with short uniramous setae and without transverse ridges; few and short setae along anterior branch of cervical groove; posterior margin with some spines; rostrum spiniform, upturned distally, larger and thicker than supraocular spines; supraocular spines small, clearly not reaching midlength of rostrum and falling short the end of corneae; margin between rostral and supraocular spines straight or slightly concave; anterolateral spines well developed situated at front near anterolateral angles, reaching the level between rostrum and supraocular spines; lateral margins with some spines. Eyes large, maximum corneal diameter about half distance between bases of anterolateral spines. Lateral margin of antennular article 1 with distal slender portion about half as long as proximal inflated portion, with 2 distal spines. Antennal peduncle with anterior prolongation of article 1 spiniform; article 2 with distomesial spine long, almost reaching end of anterior prolongation of article 1. P1-P4 long and
slender, squamate; P2-P4 dactyli slender, curved and unarmed along flexor margin. Male gonopods only present on the second abdominal somites.

Etymology. The generic name Hendersonida acknowledges the meaningful contributions of John Robertson Henderson (1863-1925) to the field of crustacean taxonomy. Gender: feminine.

Remarks. The carapace dorsal surface devoid of distinct transverse ridges or striae, the rostral spine broad at base, the antennal peduncle with a well-developed anterior prolongation of article 1 and the male gonopods absent from the first abdominal link this new genus to Paramunida Baba, 1988. This close relationship has been confirmed by molecular evidence that have rendered this new genus as the sister group of Paramunida (Cabezas et al. 2012, Cabezas and Chan 2014). Hendersonida gen. n. may be easily differentiated from Paramunida by having the dorsal surface of the carapace covered by granules and the distomesial spine of the antennal article 2 almost reaching the end of anterior prolongation of article 1 . The genus contains one species.

## Hendersonida granulata (Henderson, 1885)

Fig. 3
Munida granulata Henderson, 1885: 409 (S of the Fiji Islands, 549 m). - Henderson 1888: 133, pl. 14, figs 3, 3a, 3b (off Matuku, Fiji, 576 m).
Paramunida granulata. - Baba, 1988: 176, fig. 72 (Moluccas off W coast of Halmahera, 545 m). - Macpherson 1993: 452, figs 3, 13 (New Caledonia, Loyalty Islands and Indonesia; reexamination of type material; 439-650 m). - Macpherson 1996: 412 (SW Pacific (Futuna Island, Wallis Islands, Bayonnaise Bank), 400-450 m). Macpherson 2004: 287 (Fiji and Tonga, 395-592 m). - Ahyong and Poore 2004: 68 (Queensland, 548 m). - Baba 2005: 302 (key, synonymies). - Baba et al. 2008: 172 (list of occurrences). - Macpherson and Baba 2011: 60. - Cabezas et al. 2010: 23, fig 13C, 16I (Tonga, Vanuatu, Loyalty Islands, 550-600 m).

Diagnosis. (modified from Cabezas et al. 2010) Rostrum clearly triangular, larger than supraocular spines, with thin dorsal carina; margin between rostral and supraocular spines straight or slightly concave. Minute spinules on gastric and hepatic regions forming groups arising from scale-like striae and with few short uniramous setae. Mesogastric region with 1 well-developed spine. Median cardiac region with 3 or 4 well-developed spines. Few and short setae along anterior branch of cervical groove. Sternal plastron squamate, with numerous striae on sternites 4-7. Lateral margin of antennular article 1 with distal slender portion about half as long as proximal inflated portion. Antennal peduncle with anterior prolongation of article 1 spiniform; article 2 twice longer than broad, with distomesial spine long, almost reaching end of anterior prolongation of article 1, distolateral spine nearly reaching end of article 3; article 31.5 times longer than broad. Base of P1 carpus without bundle of setae. P2 propodus 7-8 times as long as wide, and 1.2-1.3 times longer than dactylus.


Figure 3. Dorsal surface of the carapace, dorsal view. A Paramunida haigae sp. n., NOAA Townsend Cromwell Cruise, holotype, male 16.6 mm . B Hendersonida granulata, BATHUS 2, Stn CP735, female, 13.7 mm .

Distribution. Philippines, Indonesia, Queensland, New Caledonia, Loyalty Islands, Fiji, Tonga, Futuna Island, Vanuatu, Wallis Islands and Bayonnaise Bank, between 395 and 650 m .

Remarks. Detailed illustrations for H. granulata are included in Baba (1988), Macpherson (1993) and the antennule, antenna and dorsal surface of the carapace were newly illustrated in Cabezas et al. (2010).

## Discussion

The present study updates the taxonomy of the genus Paramunida Baba, 1988 by describing a new species from the Central Pacific Ocean and transferring one species to a new genus. Deep waters in the Central Pacific Ocean have been poorly sampled and our knowledge on diversity of squat lobster fauna is scarce (Baba 2011; Schnabel et al. 2009). The new species herein described, P. haigae sp. n., represents the fourth record of the genus for Central Pacific waters.

The new genus here described contains only Hendersonida granulata. Although morphologically very similar to Paramunida, recent studies revealed that this species was phylogenetically and genetically very different from the other species of the genus (Cabezas et al. 2010; Cabezas et al. 2012; Cabezas and Chan 2014). This new lineage


Figure 4. Bayesian tree of the combined dataset ( $16 \mathrm{~S}+\mathrm{ND} 1$ ) as modified from Cabezas and Chan (2014). One asterisk represents $\mathrm{Pp}=1-0.90$ and two asterisks $\mathrm{Pp}=0.70-0.85$. Hendersonida gen. n . is presented in bold.
possesses two conspicuous diagnostic characteristics that make it easy to differentiate from species of Paramunida: (1) the armature of the dorsal surface of the carapace, and (2) the length of the distomesial spine of antennal article 2 . Hendersonida is unique in having a granulated carapace and the distomesial spine of antennal article 2 almost reaching the end of anterior prolongation of article 1 (Cabezas et al. 2010). All other characters present a certain degree of variation among species and they are not useful to distinguish genera. At a molecular level, divergence values between Paramunida and Hendersonida are within the range cited for other squat lobster genera (Cabezas et al. 2008; Machordom and Macpherson 2004), with a mean divergence of $8.05 \%$ for the 16 S gene, $18.5 \%$ for the ND1 gene and $15.3 \%$ for the COI gene. Furthermore, recent phylogenetic studies including mitochondrial and nuclear markers confirmed $H$. granulata as a highly supported monophyletic clade separated by a long branch from Paramunida s.s. and originated at least 10 mya before the radiation of Paramunida between 21-17 million years ago (Cabezas and Chan 2014; Cabezas et al. 2012). Based on these findings, our decision to designate a new genus is well supported (Fig. 4).

Hendersonida granulata is a widespread species distributed from the Philippines to to Northern Australia and the South Western Pacific, including New Caledonia, Vanuatu, Fiji, Tonga and Wallis and Futuna, between 395 and 650 m . This is unusual, since most deep-sea squat lobsters are characterized by having reduced geographic ranges confined to a single archipelago or a biogeographic area (Schnabel et al. 2011). Previous studies have reported how widely distributed species within deep-sea squat lobsters are most likely to be complexes of species with more restricted distributions (Cabezas et al. 2012; Poore and Andreakis 2011, 2012). For H. granulata only specimens from the South West Pacific (New Caledonia, Vanuatu and Tonga) have been studied in an integrative phylogenetic framework (Cabezas et al. 2012), so future surveys collecting new material through its entire range will help to infer genealogical relationships among geographically distinct populations. The taxonomic rearrangements in this study bring current diversity within Paramunida up to 40 species and up to 21 genera within the family Munididae.

## Key to species of Paramunida

1 Anterior prolongation of antennal article 1 spatulate
P. spatula Macpherson, 2006

- Anterior prolongation of antennal article 1 spiniform................................. 2

2 Rostral spine smaller or at most equal to supraocular spines........................ 3

- Rostral spine larger than supraocular spines ................................................ 8

3 Margin between rostral and supraocular spines clearly convex....................... P. curvata Macpherson, 2004

- Margin between rostral and supraocular spines straight or slightly concave.... 4

4 Antennal article 2 with minute distomesial spine.

- Antennal article 2 with well-developed distomesial spine ..... 5
5 Mesogastric region with 3 well-developed spines in midline
P. hawaiiensis (Baba, 1981)
Mesogastric region with minute spines ..... 6
6 Rostrum triangular ..... 7
- Rostrum spiniform P. aurora Cabezas \& Chan, 2014
7 present P. setigera Baba, 1988
Sternal plastron with few striae on each side of sternites 5-7. Bundle of setae at base of carpus of P1 absentP. tenera Cabezas, Macpherson \& Machordom, 20108P2-P4 propodi slender, about 20 times as long as broad
P. longior Baba, 1988
- P2-P4 propodi 7-14 times as long as broad ..... 9
9 Distomesial spine of antennal article 2 mucronated or bluntly produced ..... 10
- Distomesial spine of antennal article 2 spiniform ..... 23
10
Mesogastric region with 1 (rarely 2) spine ..... 11
Mesogastric region with a median row of 3 or 4 distinct spines ..... 14
11 Sternal plastron with numerous striae P. proxima (Henderson, 1885)
Sternal plastron with few striae on each side of sternites 5-7 ..... 12
12
Distomesial spine of antennal article 2 clearly overreaching antennal pe-duncle13
- Distomesial spine of antennal article 2 nearly reaching end of antennal pe-duncle
P. antipodes Ahyong \& Poore, 2004
Distolateral spine of antennal article 2 not reaching end of article 3P. akaina Cabezas \& Chan, 2014
Distolateral spine of antennal article 2 overreaching end of article 3
P. belone Macpherson, 1993
14
Distomesial spine of antennal article 2 slightly or clearly overreaching anten-nal peduncle15
- Distom ..... 2015 Lateral margin of antennular article 1 with distal slender portion as long as proxi-mal inflated portion....... P. spica Cabezas, Macpherson \& Machordom, 2010- Lateral margin of antennular article 1 with distal slender portion about halfas long as proximal inflated portion16
16 Distolateral spine of antennal article 2 exceeding antennal article 3P. salai Cabezas, Macpherson \& Machordom, 2009
- Distolateral spine of antennal article 2 not reaching end of antennal article 3 ..... 17
17 Mesial margin of antennal article 2, including distal spine, straight. Rostrumtriangular or spiniform18
- spiniform ..... 19
18 Rostrum triangular .. P. ascella Cabezas, Macpherson \& Machordom, 2010
$\qquad$
P. parvispina Cabezas, Macpherson \& Machordom, 2010 Mesogastric region with a row of 3 or 4 distinct spines21Sternal plastron with nu
produced distomesiallyP. evexa Macpherson, 1993
Sternal plastron with few striae, sternites $5-7$ with few striae on each side. Article2 of antennal peduncle produced distomesially ending in distinct spine22
22
Rostrum with thin dorsal carina. ..... 24
Distomesial spine of antennal article 2 clearly exceeding antennal peduncle ...P. leptotes Macpherson \& Baba, 2009
Distomesial spine of antennal article 2 at most reaching end of antennal pe- duncle ..... 25
25
Mesogastric region with 1 (rarely 2) spine ..... 26
Mesogastric region with a row of 3 or 4 distinct spines ..... 29
26
Median cardiac region with 1 spine P. pronoe Macpherson, 1993
Median cardiac region with a row of 3 or 4 spines ..... 27
27
Tufts of long and dense setae along anterior branch of cervical grooveP. crinita Cabezas, Macpherson \& Machordom, 2010
- Few and short setae along anterior branch of cervical groove ..... 28
28 lateral side P. polita Macpherson, 1993
- Sternal plastron with numerous striae P. scabra (Henderson, 1885)
29 Sternal plastron with numerous striae ..... 30
Sternal plastron with few striae, sternites 5-7 only with few striae on eachlateral side31
30 Antennal article 3 twice as long as broad. Few and short setae along anteriorbranch of cervical grooveP. thalie Macpherson, 1993
- Antennal article 3 slightly longer than broad. Tufts of long and densesetae along anterior branch of cervical groove

| 31 | Distomesial spine of antennal article 2 reaching or slightly exceeding end of antennal peduncle. Distolateral spine of antennal article 2 reaching or slightly exceeding end of antennal article 3 |
| :---: | :---: |
| - | Distomesial spine of antennal article 2 not reaching end of antennal peduncle. Distolateral spine of antennal article 2 not reaching end of antennal article 3 $\qquad$ |
| 32 | Antennal article 3 as long as wide .......... P. aspera Cabezas \& Chan, 2014 |
| - | Antennal article 3 about 1.5 times longer than wide $\qquad$ .........................P. marionis Cabezas, Macpherson \& Machordom, 2010 |
| 33 | Antennal article 3 more than twice longer than broad. $\qquad$ ............................................................ P. amphitrita Macpherson, 1996 |
| - | Antennal article 3 as long as broad or at most 1.5 times longer than broad.... 34 |
| 34 | Antennal article 2 as long as or more than 3 times longer than broad ....... 35 |
| - | Antennal article 2 at most twice longer than broad ............................... 36 |
| 35 | Distomesial spine of antennal article 2 reaching or slightly overreaching end of antennal article 3. Spinules on gastric and hepatic regions mostly forming groups arising from scale-like striae.. $\qquad$ P. pictura Macpherson, 1993 |
| - | Distomesial spine of antennal article 2 not reaching end of antennal article <br> 3. Spinules on gastric and hepatic regions mostly not in groups, lacking scaly striae. $\qquad$ P. poorei Cabezas, Macpherson \& Machordom, 2010 |
| 36 | Antennal article 2 slightly longer than broad... P. cretata Macpherson, 1996 |
|  | Antennal article 2 twice longer than broad.......................................... 37 |
| 37 | Row of small epigastric spines behind rostral spine absent $\qquad$ P. luminata Macpherson, 1996 |
| - | Row of small epigastric spines behind rostral spine present ..................... 38 |
| 38 | Rostrum triangular... P. antares Cabezas, Macpherson \& Machordom, 2010 |
| - | Rostrum spiniform ......................................................................... 39 |
| 39 | Mesogastric region with 3 small spines. Merocarpal articulation of P3 clearly exceeding end of anterior prolongation of antennal article $1 \ldots \boldsymbol{P}$. haigae $\mathbf{~ s p . ~} \mathbf{n}$. |
| - | Mesogastric region with 3 well-developed spines. Merocarpal articulation of P3 slightly exceeding end of anterior prolongation of antennal article. $\qquad$ P. achernar Cabezas, Macpherson \& Machordom, 2010 |

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# Morphology and identification of the final instar nymphs of three cicadas (Hemiptera, Cicadidae) in Guanzhong Plain, China based on comparative morphometrics 

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#### Abstract

The present investigation provides comparative morphometrics of the final instar nymphs of three dominant cicada species, i.e., Cryptotympana atrata (Fabricius), Meimuna mongolica (Distant) and Platypleura kaempferi (Fabricius), in Guanzhong Plain, China. Particularly, characters on the antennae, legs, and apex of abdomen of both males and females of these three species were investigated and analyzed. In addition, the numbers of hind tibial spines of the final instar nymphs of 21 representatives of Cicadoidea were compared. The results provide useful characteristics for nymph identification of related species and for further taxonomic and phylogenetic analysis of Cicadoidea.


## Keywords

Cicadoidea, immature stage, underground pest

## Introduction

Cicadas (Hemiptera, Cicadidae) are well known for their loud calling songs produced by male adults during summer (Young and Bennet-Clark 1995) and their long-term immature stage which is much longer than the adult stage, lasting several
years underground (Boulard 1965, Pachas 1966, Logan 2006). During their subterranean lives from the first to final instars, cicada nymphs, burrowing through soil and feeding on xylem roots (White and Strehl 1978), may occasionally cause damage to their host plants. They have powerful forelegs modified for digging, and related morphological characters have been recognized for nymph identification in a few species by several authors (Boulard 1965, Pachas 1966, Hayashi 1976, Williams and Simon 1995, Ellingson et al. 2002). However, little information is available for nymph morphology or identification of most cicada species.

The cicadas Cryptotympana atrata (Fabricius), Meimuna mongolica (Distant) and Platypleura kaempferi (Fabricius), all belonging to the subfamily Cicadinae, are the three most dominant species in Guanzhong Plain of Shaanxi Province, China, which lies north of Qinling Mountains, the convergence zone of the Palaearctic and the Oriental regions and the natural boundary between northern and southern China. Like other cicadas, these three species, particularly Cr. atrata (a dominant pest in apple orchards of northen China), can cause great harm including twig dieback in host plants when large numbers of females insert eggs into the stems of trees and, furthermore, injuries caused by the feeding of them usually go undetected since their nymphs are long-lived underground (Zhu et al. 2012). Previous studies on these cicadas were mainly focused on adult morphology and taxonomy (Chou et al. 1997), and the morphology or morphometrics of the final instar exuviae (Kato 1931, Hayashi 1974, 1975, 1987; Lee et al. 2012). Herein, we investigate the morphometrics of the final instar nymphs of these three cicadas, aiming to give a detailed description of the final instar nymphs, compare the gross morphology among different species, and provide more information for nymph identification and future investigation about their biology, ontogeny and ethology.

## Materials and methods

## Materials

All nymphs of the final instar were collected by digging beneath the woods, i.e., Cr. atrata beneath Populus tomentosa Carr., P. kaempferi beneath Metasequoia glyptostroboides Hu \& Cheng, and M. mongolica beneath Pyrus xerophila Yü on the campus of Northwest A\&F University, Yangling, Shaanxi Province, China, from October to December, 2013. All captured nymphs were transferred alive to a beaker and anesthetized by chilling in a $4{ }^{\circ} \mathrm{C}$ refrigerator for morphological investigation. Exuviae and adult cicadas of the above three species were also collected on their host plants from June to July, 2013, respectively, aiming to confirm the identification of the final instar nymphs of each related species based on morphology. In addition, the nymphs of the final instar, exuviae and adult cicadas of Subpsaltria yangi Chen (belonging to the subfamily Tettigadinae) and Karenia caelatata Distant (belonging to the subfamily Cicadettinae) were also collected in the same way in Mts Helan, Ningxia Hui Autonomous Region,

China, in June, 2012, and at Ningshan County in Mts Qinling, Shaanxi Province, China, in July and August, 2012, respectively. All the above mentioned materials and the exuviae of Cicadetta shansiensis (Esaki \& Ishihara) deposited in the Entomological Museum of Northwest A\&F University, China were examined, aiming to make a comparative morphological study on the hind tibial spines among these species and also other related species which have been investigated by some authors (Hayashi 1999; Maccagnan and Martinelli 2004, 2011; Logan and Connolly 2005).

## Methods

Nymphs were classified to sex by the developing genitalia at the apex of abdomen. For males, the several terminal abdominal segments of part materials were slightly extracted to show the $9^{\text {th }}$ abdominal sternite if necessary, which was partly concealed by the $10^{\text {th }}$ abdominal sternite. Observations of the morphological features were carried out using a Motic SMZ168 Stereoscopic Zoom Microscope. Photographs were taken with a scientific digital micrography system equipped with an Auto-montage imaging system and a Qimaging Retiga 2000R digital camera (CCD). Drawings were made with the aid of a camera lucida attached to the microscope.

Twenty individuals ( 10 males and 10 females, respectively) of each species were measured. The measurements are as follows: (1) crown length (CL) measured on dorsal view along its median line from frontoclypeal suture to posterior margin of head (Fig. 1A); (2) pronotum length (PL) measured on dorsal view along its median line (Fig. 1A); (3) pro-mesonotum length (PML) measured on dorsal view from pronotum to mesonotum along its median line (Fig. 1A); (4) head width (HW) measured on dorsal view from the outside of one compound eye to the other (Fig. 1B); (5) pronotum width (PW) measured on dorsal view at the posterior margin (Fig. 1B); (6) body length (BL) measured on lateral view from the apex of postclypeus to the distal margin of abdomen (Fig. 1C); (7) wing length (WL) measured on lateral view from rim of pronotum to apex of wing pocket (Fig. 1C); (8) postclypeus length (PCL) measured on front view from its suture with the anteclypeus to the frontoclypeal suture (Fig. 1D); (9) postclypeus width (PCW) measured on front view from one side of the outermost edge of frontoclypeal to the other (Fig. 1D); (10) fore femur length (FL) measured along the median line of its external side (Fig. 1E); (11) fore tibiae length (TL) measured along the median line of its external side (Fig. 1E); (12) femoral tooth angle (FA) measured between the longitudinal axis of femur and its posterior tooth (Fig. 1E).

Among these measurements, FA was measured using the Image Lab version 2.2.4.0 software (MCM Design, Hillerød, Denmark). The remaining measurements were taken using a vernier caliper with the accuracy of 0.02 mm .

Multivariate and univariate general linear model (GLM) analyses were conducted to determine whether morphological characters differed by species or sex. Statistical analyses were performed using SPSS 17.0.


Figure I. Morphological characters of final instar nymphs used for measurements. BL body length; CL crown length; FA femoral tooth angle; FL fore femur length; HW head width; PCL postclypeus length; PCW postclypeus width; PML pro-mesonotum length; PL pronotum length; PW pronotum width; TL fore tibiae length; WL wing length.

The subfamily and tribal classification follows that of Moulds (2005). The terminology adopted to describe the structures of the foreleg was based on Duffels and Ewart (1988), and the femoral formula used to indicate the number and sequence of the teeth of fore femur was based on Maccagnan and Martinelli (2004).

## Results

Morphology of the final instar nymph of Cryptotympana atrata (Fabricius, 1775)
Cryptotympana atrata (Fabricius, 1775)
Figs 2-5
Tettigonia atrata Fabricius, 1775: 681
Cicada atrata (Fabricius): Goeze 1778: 149
Tettigonia pustulata Fabricius, 1787: 266
Cicada nigra Olivier, 1790: 750
Fidicina bubo Walker, 1850: 82
Fidicina atrata (Fabricius): Walker 1850: 89
Cryptotympana atrata (Fabricius): Stål 1861: 613

Cryptotympana sinensis Distant, 1887: 415
Cryptotympana dubia Haupt, 1917: 229
Cryptotympana coreanus Kato, 1925: 13
Cryptotympana santoshonis Matsumura, 1927: 49
Cryptotympana wenchewensis Ouchi, 1938: 82
Cryptotypmana pustulata castanea Liu, 1940: 82
Cryptotympana pustulata fukienensis Liu, 1940: 82
Measurements (mm or degree). Male ( $\mathrm{n}=10$ ): BL 26.8 (23.5-31.5), PCL 5.2 (4.75.5), PCW 5.2 (4.9-5.7), CL 3.1 (2.9-3.3), HW 11.7 (10.9-12.1), PL 8.3 (7.7-8.7), PW 14.3 (13.6-15.2), PML 16.5 (15.5-17.3), WL 12.6 (11.9-13.1), FL 6.2 (5.96.5), TL 6.6 (6.1-7.1), FA 71.4 (69.5-73.2).

Female ( $\mathrm{n}=10$ ): BL: 26.3 (23.3-29.7), PCL 5.2 (4.9-5.4), PCW 5.2 (5.0-5.3), CL 3.0 (2.8-3.2), HW 11.4 (10.5-12.1), PL 8.1 (7.3-8.7), PW 14.2 (13.6-15.1), PML 16.2 (15.2-17.0), WL 12.5 (11.5-13.1), FL 6.2 (5.7-6.5), TL 6.5 (6.0-7.0), FA 71.6 (69.4-73.4).

Description. Body (Fig. 2A, B) dark brown, curved in lateral view, with sparse setae mainly on venter.

Head (Fig. 2B). Somewhat triangular in dorsal view; crown including white compound eyes about four times wider than long and about the same width as anterior margin of pronotum. Antenna brown, filiform. Postclypeus prominently swollen, covered with dense brown pile. Rostrum reaching to posterior coxae.

Thorax (Fig. 2A, B). Pronotum broad, paramedian and lateral fissures distinct, pronotal collar undeveloped, posterior margin distinctively concave medially in dorsal view. Mesonotum slightly wider than pronotum, with two small scutal depressions on disc. Metanotum very small. Fore wing bud developed, reaching to middle of $3^{\text {rd }}$ abdominal segment laterally; hind wing bud slightly developed.

Leg (Figs 5A, B, 6A, D). Generally dark brown. Fore femur with femoral formula 2-1-7: posterior tooth long and sharp, accessory tooth robust and sharp, intermediate tooth with projection in one of its sides; femoral comb usually with seven teeth, the first tooth about as large as the second tooth. Fore tibia arched, flattened laterally; apical tooth long; point of blade of tibia large and long, tooth-like, separated from apical tooth of blade by a strong incision. Apex of tibia with five spines in both mid and hind legs. Pretarsi of all legs well developed into a pair of claws of unequal sizes.

Abdomen (Fig. 4A-C). Size varying depends on the development of the nymph. In female, $8^{\text {th }}$ and $9^{\text {th }}$ sternites with two sharp posterior marginal protrusions. In male, $9^{\text {th }}$ sternite almost entirely concealed by $10^{\text {th }}$ sternite, four protrutions present on surface: a large triangular protrusion near lateral margins, respectively, and a pair of very small rounded protrusions on posterior margin; $10^{\text {th }}$ sternite smooth.

Variations of femoral comb. Ten percent $(2 / 20)$ of the individuals investigated with femoral comb with six teeth, instead of seven teeth.


Figure 2. Final instar nymphs. A Cryptotympana atrata, lateral view of body B C. atrata, dorsal view of body C Meimuna mongolica, lateral view of body D M. mongolica, dorsal view of body E Platypleura kaempferi, lateral view of body $\mathbf{F}$ P. kaempferi, dorsal view of body. Scale bars $=5.0 \mathrm{~mm}$.


Figure 3. Antennae of final instar nymphs. A Cryptotympana atrata B Meimuna mongolica C Platypleura kaempferi. Scale bars $=0.5 \mathrm{~mm}$.


Figure 4. Abdominal apex in ventral view of final instar nymphs. A Cryptotympana atrata, female $\mathbf{B}$ and C C. atrata, male D Meimuna mongolica, female E and $\mathbf{F}$ M. mongolica, male $\mathbf{G}$ Platypleura kaempferi, female $\mathbf{H}$ and $\mathbf{I}$. . Kaempferi, male. Scale bars $=1.0 \mathrm{~mm}$.

## Morphology of the final instar nymph of Meimuna mongolica (Distant, 1881)

## Meimuna mongolica (Distant, 1881)

Figs 2-5
Cosmopsaltria mongolica Distant, 1881: 638
Meimuna mongolica (Distant): Distant 1906: 66
Meimuna suigensis Matsumura, 1927: 1
Meimuna chosensis Matsumura, 1927: 52
Meimuna heijonis Matsumura, 1927: 52
Meimuna santoshonis Matsumura, 1927: 52
Meimuna gallosi Matsumura, 1927: 52

Measurements (mm or degree). Male ( $\mathrm{n}=10$ ): BL 19.8 (18.0-21.3), PCL 3.5 (3.23.7), PCW 3.2 (2.9-3.5), CL 2.4 (2.2-2.7), HW 7.2 (6.8-7.7), PL 4.6 (4.4-4.8), PW 7.8 (7.3-8.2), PML 9.4 (8.7-9.8), WL 8.7 (8.0-9.4), FL 4.8 (4.5-5.0), TL 4.6 (4.3-4.8), FA 61.4 (60.5-62.8).

Female ( $\mathrm{n}=10$ ): BL 19.1 (17.9-20.9), PCL 3.3 (3.1-3.6), PCW 3.0 (2.7-3.2), CL 2.2 (2.0-2.6), HW 6.9 (6.8-7.2), PL 4.4 (4.2-4.7), PW 7.5 (7.2-8.3), PML 8.9 (8.4-9.7), WL 8.4 (8.0-8.9), FL 4.7 (4.5-4.9), TL 4.5 (4.2-4.7), FA 61.4 (60.1-63.2).

Description. Body (Fig. 2C, D) pale brown, narrow and elongated, with setae scattered mainly on venter.

Head (Fig. 2D). Somewhat triangular in dorsal view; crown including white compound eyes about three times wider than long and slightly wider than anterior margin of pronotum. Antenna brown, filiform. Postclypeus prominently swollen, covered with dense brown pile. Rostrum reaching to posterior coxae.

Thorax (Fig. 2C, D). Pronotum broad, paramedian and lateral fissures distinct, pronotal collar undeveloped, posterior margin distinctively concave medially in dorsal view. Mesonotum slightly wider than pronotum, with two small scutal depressions on disc. Metanotum very small. Fore wing bud developed, reaching to middle of $4^{\text {th }}$ abdominal segment laterally, hind wing bud slightly developed.

Leg (Figs 5C, 6B, E). Generally dark brown. Fore femur with femoral formula 2-1-7 or 2-1-8: posterior tooth long and sharp, accessory tooth small, with apex somewhat blunt, intermediary tooth robust; femoral comb usually with seven or eight teeth, the first tooth about as large as the second tooth. Fore tibia arched, flattened laterally; apical tooth long; point of blade of tibia very small, toothlet-like, separated from apical tooth of blade by a very weak incision. Tibia with five apical spines in both mid and hind legs. Pretarsi of all legs well developed into a pair of claws of unequal sizes.

Abdomen (Fig. 4D-F). Size varying depending on the development of the nymph. In female, $8^{\text {th }}$ and $9^{\text {th }}$ sternites with two sharp posterior marginal protrusions. In male, $9^{\text {th }}$ sternite totally concealed by $10^{\text {th }}$ sternite, three protrusions present on surface: a smaller medial, coniform protrusion near anterior margin, and two larger rounded
protrusions on posterior margin; $10^{\text {th }}$ sternite with two distinct projections adjacent to anterior margin.

Variations of femoral comb. Forty percent $(8 / 20)$ and $60 \%(12 / 20)$ of the individuals observed with seven and eight teeth on the femoral comb, respectively.

## Morphology of the final instar nymph of Platypleura kaempferi (Fabricius, 1794)

Platypleura kaempferi (Fabricius, 1794)
Figs 2-5
Tettigonia kaempferi Fabricius, 1794: 23
Cicada kaempferi (Fabricius): Walker 1850: 117
Platypleura kaempferi (Fabricius): Butler 1874: 189
Platypleura fuscangulis Butler, 1874: 189
Platypleura hyalino-limbata Signoret, 1881: 62
Platypleura repanda Uhler, 1896: 276 (nec Linnaeus)
Platypleura tsuchidai Kato, 1936: 758
Platypleura retracta Liu, 1940: 74

Measurements (mm or degree). Male ( $\mathrm{n}=10$ ): BL 19.0 (18.1-19.6), PCL 3.2 (3.13.5), PCW 3.3 (3.2-3.5), CL 2.2 (2.1-2.4), HW 7.5 (7.3-7.8), PL 4.7 (4.3-5.0), PW 9.2 (8.9-9.6), PML 10.0 (9.4-10.5), WL 7.4 (7.0-7.8), FL 4.1 (3.9-4.3), TL 4.5 (4.2-4.6), FA 77.1 (76.1-78.1).

Female ( $\mathrm{n}=10$ ): BL 18.3 (17.7-18.6), PCL 3.1 (2.9-3.4), PCW 3.2 (3.1-3.4), CL 2.1 (2.0-2.2), HW 7.3 (7.1-7.7), PL 4.6 (4.2-4.9), PW 8.9 (8.6-9.2), PML 9.8 (8.9-10.2), WL 7.2 (6.9-7.6), FL 4.0 (3.8-4.2), TL 4.3 (4.1-4.5), FA 77.2 (76.4-78.8).

Description. Body (Fig. 2E, F) brown, well curved in lateral view, with sparse setae mainly on venter.

Head (Fig. 2F). Somewhat triangular in dorsal view; crown including white compound eyes about three times wider than long and slightly wider than the anterior margin of the pronotum. Antenna brown, filiform. Postclypeus prominently swollen, covered with dense brown pile. Rostrum extending beyond posterior coxae.

Thorax (Fig. 2E, F). Pronotum broad, paramedian and lateral fissures distinct, pronotal collar developed, posterior margin distinctively concave medially in dorsal view. Mesonotum about as wide as pronotum, with two small scutal depressions on disc. Metanotum very small. Fore wing bud developed, reaching to middle of $3^{\text {rd }}$ abdominal segment laterally, hind wing bud slightly developed.

Leg (Figs 5D, 6C, F). Generally dark brown. Fore femur with femoral formula 2-1-7: posterior tooth long and sharp, accessory tooth robust and sharp, intermediate tooth with projection in one of its sides; femoral comb usually with seven teeth, the


Figure 5. Left foreleg of final instar nymphs. A Cryptotympana atrata, inner view B $C$. atrata, outer view C Meimuna mongolica, outer view D Platypleura kaempferi, outer view. acf, accessory tooth of femur; apt, apical tooth of tibia; bt, blade of tibia; f, femur; fc, femoral comb; itf, intermediate tooth of femur; pbt, point of blade of tibia; ptf, posterior tooth of femur; $t$, trochanter; ta, tarsus; ti, tibia. Scale bars $=1.0 \mathrm{~mm}$


Figure 6. Spines at the apex of mid and hind tibiae of final instar nymphs. A mid tibia of Cryptotympana atrata B mid tibia of Meimuna mongolica $\mathbf{C}$ mid tibia of Platypleura kaempferi $\mathbf{D}$ hind tibia of $C$. atrata $\mathbf{E}$ hind tibia of $M$. mongolica $\mathbf{F}$ hind tibia of $P$. Kaempferi. Scale bars $=1.0 \mathrm{~mm}$.
first tooth distinctly larger than the second tooth. Fore tibia arched, flattened laterally; apical tooth long; point of blade of tibia large and long, tooth-like, separated from apical tooth of blade by a strong incision. Apex of tibia usually with four spines in both mid and hind legs, but sometimes with a very small accessory spine. Pretarsi of all legs well developed into a pair of claws of unequal sizes.

Abdomen (Fig. 4G-I). Size varying depending on the development of the nymph. In female, $8^{\text {th }}$ and $9^{\text {th }}$ sternites with two sharp posterior marginal protrusions. In male, $9^{\text {th }}$ sternite almost entirely concealed by $10^{\text {th }}$ sternite, six protrusions present on surface: two triangular protrusions adjacent to posterior margin, and four rounded protrusions on posterior margin; $10^{\text {th }}$ sternite with a very large, medial, globular protrusion adjacent to anterior margin.

Variations of femoral comb. Twenty percent (4/20) of the individuals studied with femoral comb with eight teeth, instead of seven teeth.

## Morphometrics and comparative morphology of antennae and forelegs among the three cicadas

The results showed that the species (Wilks' $\lambda=0, \mathrm{~F}=817.078$, hypothesis $\mathrm{df}=24$, error $\mathrm{df}=90.000, \mathrm{P}=0$ ) was a significant factor for all morphological characters, and that sex (Wilks' $\lambda=0.469, \mathrm{~F}=4.241$, hypothesis $\mathrm{df}=12$, error $\mathrm{df}=45.000, \mathrm{P}=0$ ) was also significant for all characters, except for BL, FL and FA (Table 1).

There are great similarities in the gross morphology of antennae among the final instar nymphs of these three cicadas, i.e., the scape inserts in an antennal fovea of the cranium at the side of the postclypeus near an anterior tentorial pit, which is partially concealed in the antennal fovea and dorsally covered by the overhanging ridge of the vertex. However, differences also exist and are mainly shown in two aspects of these species: i) the shape of antennae (tapering apically in Cr. atrata and $P$. kaempferi (Fig. 3A, C), but apical segment of flagellum in M. mongolica with full length in similar diameter (Fig. 3B)); and ii) the number of flagellar segments (seven in M. mongolica (Fig. 3B), eight in Cr. atrata (Fig. 3A) and nine in P. kaempferi (Fig. 3C)).

Similarly, though there are many similarities in the morphology of forelegs among the final instar nymphs of these three cicadas, differences also exist and are mainly shown in four aspects: i) the shape of the base of posterior tooth on femur (extraordinarily broadened in M. mongolica (Fig. 5C), but moderately broadened in Cr. atrata and P. kaempferi (Fig. 5B, D)), ii) the shape of the base of intermediate tooth (extraordinarily broadened in Cr. atrata and P. kaempferi (Fig. 5B, D), but moderately broadened in M. mongolica (Fig. 5C)); iii) the shape of blade of tibia (with a large and long tooth-like point of blade of tibia in Cr. atrata and P. kaempferi (Fig. 5B, D), but with a very small, reduced apical toothlet-like point of blade of tibia in M. mongolica (Fig. 5C)); and iv) the femoral tooth angle (about $61^{\circ}, 71^{\circ}$ and $77^{\circ}$ in M. mongolica, Cr. atrata and $P$. kaempferi, respectively).

Table I. Results of univariate general linear model (GLM) for the morphological characters measured in cicada nymphs.

| Source | Variable | df | M.S. | F | P |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Species | BL | 2 | 376.178 | 199.226 | 0.000 |
|  | PCL | 2 | 25.226 | 887.718 | 0.000 |
|  | PCW | 2 | 27.038 | 1002.519 | 0.000 |
|  | CL | 2 | 4.289 | 219.129 | 0.000 |
|  | HW | 2 | 121.916 | 1349.714 | 0.000 |
|  | PL | 2 | 87.079 | 1291.314 | 0.000 |
|  | PW | 2 | 245.548 | 1809.702 | 0.000 |
|  | PML | 2 | 314.792 | 1488.336 | 0.000 |
|  | WL | 2 | 150.091 | 963.221 | 0.000 |
|  | FL | 2 | 24.449 | 782.357 | 0.000 |
|  | TL | 2 | 27.888 | 631.693 | 0.000 |
|  | FA | 2 | 1271.572 | 1386.341 | 0.000 |
| Sex | BL | 1 | 6.144 | 3.254 | 0.077 |
|  | PCL | 1 | 0.131 | 4.598 | 0.036 |
|  | PCW | 1 | 0.171 | 6.328 | 0.015 |
|  | CL | 1 | 0.216 | 11.036 | 0.002 |
|  | HW | I | 0.963 | 10.658 | 0.002 |
|  | PL | 1 | 0.353 | 5.230 | 0.026 |
|  | PW | 1 | 0.561 | 4.132 | 0.047 |
|  | PML | 1 | 1.700 | 8.038 | 0.006 |
|  | WL | 1 | 0.662 | 4.245 | 0.044 |
|  | FL | 1 | 0.096 | 3.072 | 0.085 |
|  | TL | 1 | 0.241 | 5.451 | 0.023 |
|  | FA | 1 | 0.122 | 0.132 | 0.717 |
| Error | BL | 56 | 1.888 |  |  |
|  | PCL | 56 | 0.028 |  |  |
|  | PCW | 56 | 0.027 |  |  |
|  | CL | 56 | 0.020 |  |  |
|  | HW | 56 | 0.090 |  |  |
|  | PL | 56 | 0.067 |  |  |
|  | PW | 56 | 0.136 |  |  |
|  | PML | 56 | 0.212 |  |  |
|  | WL | 56 | 0.156 |  |  |
|  | FL | 56 | 0.031 |  |  |
|  | TL | 56 | 0.044 |  |  |
|  | FA | 56 | 0.917 |  |  |

Comparison of the number of hind tibial spines and condition of intermediate tooth in 21 representatives of Cicadoidea

In Tettigarctidae, three hind tibial spines were found in Tettigarcta crinita Distant. In Cicadidae, the numbers of hind tibial spines of the final instar nymphs of different species are usually the same within a subfamily, but vary among different subfamilies (Table 2). In Tettigadinae, three hind tibial spines were observed in Subpsaltria yangi.

In Cicadettinae, except for Cicadetta shansiensis with three and Karenia caelatata with five hind tibial spines, four hind tibial spines were observed in all other species: Amphipsalta cingulata (Fabricius), A. zelandica (Boisduval), Kikihia ochrina (Walker), Ki. scutellaris (Walker), Notopsalta sericea (Walker), Rhodopsalta cruentata (Fabricius), and Carineta fasciculata (Germar). In Cicadinae, three hind tibial spines were found in the genus Mogannia Amyot \& Audinet-Serville, four hind tibial spines in the genus Nipponosemia Kato, five hind tibial spines in the four investigated species (Cr. atrata (Fig. 6D), M. mongolica (Fig. 6E), Quesada gigas (Olivier) and Fidicina mannifera (Fabricius)), and four hind tibial spines with an additional small accessory spine internally were observed in the other four species, i.e., Dorisiana drewseni (Stål), D. viridis (Olivier), Fidicinoides pronoe (Walker) and P. kaempferi (Fig. 6F).

There are some differences in the situation of an intermediate tooth on fore femur from the femoral comb, e.g., continuous from the femoral comb, or well separated from the comb. For example, the intermediate tooth is continuous from the femoral comb in M. mongolica (Fig. 5C), Ci. shansiensis and Ka. caelatata; however, it is well separated from the comb in Cr. atrata (Fig. 5B), P. kaempferi (Fig. 5D) and S. yangi.

## Discussion

The present study is the first to focus on the comparative morphology of the three cicadas in Guanzhong Plain. Cicadas usually have a long immature stage underground, which causes difficulties in their nymphal instar determination. A few species were reported to have five nymphal instars by some authors, e.g., Mogannia minuta Matsumura, Magicicada septendecim (Linnaeus), and Diceroprocta apache (Davis) (Hayashi 1976; Maier 1980; Ellingson et al. 2002); while a few other species were reported to have four instars by several authors, e.g., Cryptotympana atrata and Leptopsalta yamashitai (Esaki \& Ishihara) (Hu et al. 1990; Kang et al. 2005). The number of nymphal instars in cicadas needs to be readdressed based on more investigations. Though major similarities shared by nymphs of different instars within a species, the nymphs of final instar can be easily distinguished from the remaining instar nymphs by the well developed and rounded eye-capsule, the developed wing buds, and the apex of abdomen. In addition, some characteristics of the final instar nymphs are preserved in their exuviae, e.g., the femoral tooth angle, the shapes of foreleg and postclypeus, etc., which are informative for the recognition of the final instar nymphs from the remaining instar nymphs within a species. However, nymphs and adults of cicadas within a species have significant morphological differences which are closely related to their different ecological niches (Li and Wei 2013), and cause difficulties in species identification of most cicada nymphs. In the present study we show that the final instar nymphs of cicadas can be distinguished from each other according to their morphology. Among which, the differences of antennae and the $9^{\text {th }}$ and $10^{\text {th }}$ sternites in males may be important characters for taxonomic and phylogenetic analysis. In addition, the number of the hind tibial spines of the final instar nymphs may be an
Table 2. Number of hind tibial spines of the 21 representatives of Cicadoidea.

| Species or genera | Tribes | Subfamilies | Families | Numbers | Sources |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Tettigarcta crinita Distant, 1883 | Tettigarctini | Tettigarctinae | Tettigarctidae | 3 | This study |
| Subpsaltria yangi Chen, 1943 | Tibicinini | Tettigadinae | Cicadidae | 3 | This study |
| Cicadetta shansiensis (Esaki \& Ishihara, 1950) | Cicadettini | Cicadettinae |  | 3 | This study |
| Amphipsalta cingulata (Fabricius, 1775) | Cicadettini |  |  | 4 | Logan and Connolly (2005) |
| A. zelandica (Boisduval, 1835) | Cicadettini |  |  | 4 | Logan and Connolly (2005) |
| Kikihia ochrina (Walker, 1858) | Cicadettini |  |  | 4 | Logan and Connolly (2005) |
| K. scutellaris (Walker, 1850) | Cicadettini |  |  | 4 | Logan and Connolly (2005) |
| Notopsalta sericea (Walker, 1850) | Cicadettini |  |  | 4 | Logan and Connolly (2005) |
| Rhodopsalta cruentata (Fabricius, 1775) | Cicadettini |  |  | 4 | Logan and Connolly (2005) |
| Carineta fasciculata (Germar, 1821) | Carinetini (= Sinosenini Boulard) |  |  | 4 | Maccagnan and Martinelli (2011) |
| Karenia caelatata Distant, 1888 | Carinetini (= Sinosenini Boulard) |  |  | 5 | This study |
| Mogannia Amyot \& Audinet-Serville, 1843 | Cicadatrini (=Moganniini) | Cicadinae |  | 3 | Hayashi (1999) |
| Nipponosemia Kato, 1925 | Cicadatrini (=Moganniini) |  |  | 4 | Hayashi (1999) |
| Cryptotympana atrata (Fabricius, 1775) | Cryptotympanini |  |  | 5 | This study |
| Meimuna mongolica (Distant, 1881) | Dundubiini |  |  | 5 | This study |
| Quesada gigas (Olivier, 1790) | Hyantiini |  |  | 5 | Maccagnan and Martinelli (2004) |
| Fidicina mannifera (Fabricius, 1803) | Fidicinini |  |  | 5 | Maccagnan and Martinelli (2011) |
| Dorisiana drewseni (Stål, 1854) | Fidicinini |  |  | 4 with an additional small spine internally | Maccagnan and Martinelli (2011) |
| D. viridis (Olivier, 1790) | Fidicinini |  |  |  | Maccagnan and Martinelli (2011) |
| Fidicinoides pronoe (Walker, 1850) | Fidicinini |  |  |  | Maccagnan and Martinelli (2011) |
| Platypleura kaempferi (Fabricius, 1794) | Platypleurini |  |  |  | This study |

important morphological characteristic in phylogenetic analysis, although the number of hind tibial spines may be variable within a group, e.g., number of hind tibial spines of the genera Nipponosemia and Mogannia in the tribe Cicadatrini (=Moganniini) are 4 and 3, respectively (Hayashi 1999). The number of hind tibial spines of the final instar nymphs of more taxa need to be investigated when more materials become available. Surprisingly, the number of the hind tibial spines of the final instar nymphs of Karenia caelatata is 5, which is different with that of other investigated members of Cicadettinae, but is consistent with that of some members belonging to the Cicadinae, e.g., Cr. atrata, Meimuna mongolica, Fidicina mannifera, and Quesada gigas. Moulds (2005) and Boulard (2008) attributed the Karenia in the Cicadettinae (=Tibicininae auct.). However, the number of the hind tibial spines of the final instar nymphs together with some other characteristics of adults (e.g., metanotum distinctly concealed by the cruciform elevation on dorsal midline, uncus well developed with uncal lobes elongated apically, etc.) suggest that it seems more plausible to place this genus in the Cicadinae. The systematic placement of this genus needs further investigation.

Cicada nymphs could extend to 120 cm soil layer underground (Hugie and Passey 1963), though nymphs are most abundant within the well-rooted soil A horizon (eluvial horizon) and B horizon (illuvial horizon), typically between 10 and 30 cm from the ground surface (Luken and Kalisz 1989, O'Geen and Busacca 2001). However, burrowing depth of nymphal cicadas varies depending on the species. For example, our investigation showed that Platypleura kaempferi nymphs were located at $10-30 \mathrm{~cm}$ from the soil surface, being consistent with the results of Uematsu and Onogi (1980), and M. mongolica nymphs were mainly distributed in $21-30 \mathrm{~cm}$ soil layer and could extend to 60 cm . Interestingly, significant differences were also observed in the forelegs of these two species. This suggests that the different burrowing depth in soil of the nymphs of different cicada species should be closely related to the development of their forelegs, and that the forelegs may provide promising characters for taxonomy and for future investigation about biology, ontogeny and ethology of related species.

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# A new species of Dendrocerus (Hymenoptera, Megaspilidae) from southern Brazil 

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#### Abstract

A new species of the megaspiline genus Dendrocerus Ratzeburg is described and figured. Dendrocerus riograndensis sp. n., is known from a series of males from the central region of Rio Grande do Sul, Brazil, and were captured with Malaise traps from an agricultural tobacco field.


## Keywords

Ceraphronoidea, systematic, new species

## Introduction

The parasitoid wasp family Megaspilidae includes more than 450 species in 11 genera worldwide (Dessart 2006), but the fauna of South America remains little known. Johnson and Musetti (2004) noted the genus Dendrocerus Ratzeburg to be cosmopolitan, with 96 described species, but only 11 of them are recorded from the Neotropical Region. Four species of Dendrocerus are known from Brazil: Dendrocerus sylviae

Dessart, collected in Pará State, D. carpenteri (Curtis), collected in many regions of Brazil, D. aphidum (Rondani), recordered in Rio Grande do Sul State and D. phallocrates Dessart, collected in São Paulo State (Dessart and Cancemi 1986; Dessart 1987; Tavares 1996; Dessart 1996; Martinez 2003).

Among species in Dendrocerus males of the halidayi species group are characterized by their flabellate antennae, with long branches on the proximal 4, 5, or 6 flagellomeres. There are 21 species of this group distributed on all continents except Antarctica, with the following four species from the New World tropics: D. araucanus Dessart (Chile), D. mexicali Dessart (Mexico), D. sylviae Dessart (Brazil) and D. ranquel Martinez (Argentina) (Dessart 1999; Martinez 2003). The objective of this paper is to describe a new species of Dendrocerus belonging to the halidayi group.

## Material and methods

Specimens were collected with a Malaise trap in an organically managed cultivation of tobacco (Nicotinana tabacum L.) during the 2011-2012 crop, in Santa Cruz do Sul, Rio Grande do Sul, Brazil. The material was studied using a stereoscopic microscope trinocular Motic Quimis Q764ZT and is deposited in the Coleção Entomológica de Santa Cruz (CESC).

The morphological nomenclature, format for the description, and measurements employed follow that of Dessart (1999), while Martinez (2003) is followed for features of the flagellomeres and side branches. The relative measures, except the entire body length, are expressed in millimeters.

## Results

## Dendrocerus riograndensis Pezzini \& Köhler, sp. n.

http://zoobank.org/F2786CA1-6E1C-4860-8A4D-E34368737B16
Etymology. The specific epithet is based on the state of Rio Grande do Sul from where the type series was captured.

Diagnosis. Male head moderately transverse; antenna with five rami on first to fifth flagellomeres (antennomeres three through seven), not articulated, remaining flagellomeres without rami. Mesoscutellum without grooves or lateral carina; propodeum without armature. Metasoma smooth and shining, without punctures.

Description. Male: Total body length 1.32 mm , forewing length 0.85 mm . Coloration: Brownish throughout; Metasoma, mouthparts, and legs light brown; trochanters, femoral apices, tarsi, and tibial bases lighter. Wings hyaline with brown venation.

Head: Coriaceous, pubescent; moderately transverse. Ocelli forming an isosceles triangle with wider base, median ocellus bordered anteriorly by an obvious depres-
sion；preoccipital depression bounded by only a groove posterior to lateral ocelli and separated by a distance shorter than diameter；preoccipital suture visible until base of ocellar triangle．Compound eyes subcircular，pubescent，setae distinctly shorter than those on remainder of head．Supraclypeal depression conspicuous，with adjacent areas glabrous；intertorular carina present．Antenna flabellate，with coarse bristles（Fig．6）， with elongate rami laterally on proximal five flagellomeres（flagellomeres I－V，or an－ tennomeres III－VII），A8－11 cylindrical and without lateral rami；scape： 18 （5）；pedi－ cel： 1 （1）； $\mathrm{A}_{3}: 2, \mathrm{R}_{1}: 15 ; \mathrm{A}_{4}: 2, \mathrm{R}_{2}: 12, \mathrm{~A}_{5}: 2, \mathrm{R}_{3}: 8, \mathrm{~A}_{6}: 2, \mathrm{R}_{4}: 7,5 ; \mathrm{A}_{7}: 3, \mathrm{R}_{5}: 7 ; \mathrm{A}_{8}: 5(2)$ ； $\mathrm{A}_{9}: 2$（1）； $\mathrm{A}_{10}: 7$（5）； $\mathrm{A}_{11}: 3$（1）．

Mesosoma：Mesoscutum，axillae，and mesoscutellum pubescent，coriaceous（Fig． 7）．Anterior border of mesoscutum in dorsal view strongly inclined，notauli com－ plete，weakly crenulate；sulci between axillae and mesoscutellar disc roughly crenulate． Mesoscutellum transversely convex and longitudinally simple，not carinate laterally． Mesopleuron slightly coriaceous，with some bristles anteriorly，and separated ventrally from metapleuron by crenulate suture．Propodeum without armature．Metanotal sulci extending posteriorly，convergent medially；propodeal carina transverse，forming X－ shaped structure，posterior propodeal area smooth．

Wings：Forewing without cells，marginal vein long and pubescent，membrane with numerous microtrichia，pterostigma well developed（as in all species of the family）， 0.12 mm diameter，poststigmal veins well defined and long（Fig．5）；no other veins present，hind wing without venation．

Metasoma：Nine visible segments，integument smooth and shiny；fusiform，more convex ventrally than dorsally，ovoid in dorsal view．

Female：Unknown．

## Biology．Unknown．

Comments．The number of lateral rami on the antenna，the dimensions of each flagel－ lomere（length $\times$ width），and the form of the forewing veins serve to distinguish $D$ ．riogran－ densis sp．n．from the other species of the halidayi species group．Dendrocerus sylviae belongs to the halidayi group，but only the female is known，separated from the other species by the possession of longitudinal grooves on $\mathrm{T}_{\text {III }}$ ．

Distribution．Dendrocerus riograndensis sp．n．is known presently only from the central region of Rio Grande do Sul，Brazil．Future surveys will be needed in order to better ascertain its total potential distribution．

Material examined．Holotype：BRAZIL．Rio Grande do Sul：Santa Cruz do Sul， （Agronomy，Development，Extension and Training Center＂ADET＂，2948＇22．92＂S， $52^{\circ} 19^{\prime} 42.00^{\prime \prime} \mathrm{W}, 102 \mathrm{~m}$ ），Malaise trap，Köhler A．Leg．， $10^{\top}, 16 . X I I .2011$ ，（CESC 43389／17）．Paratypes：Eight males with the same data as the holotype：BRAZIL．Rio Grande do Sul：Santa Cruz do Sul，（Agronomy，Development，Extension and Train－ ing Center＇$A D E T$＂， $29^{\circ} 48^{\prime} 22.92$＂S， $52^{\circ} 19^{\prime} 42.00$＂W，102m），Malaise trap，Köhler A． Leg．，1 ${ }^{\lambda}$ ，16．XII．2011，（CESC 43364／20）；1才，20．XII．2011，（CESC 44067／11）；2才， 27．XII．2011，（CESC 45553／14）；1才，03．I．2012，（CESC 46152／13）；1ठ，10．I．2012， （CESC 46511／10）；2才，17．I．2012，（CESC 46916／16）．


Figure I. Detail of male antenna of $D$. mexicali (redrawn from Dessart 1991).


Figure 2. Detail of male antenna of D. araucanus (redrawn from Dessart 1991).


Figure 3. Detail of male antenna of $D$. ranquel (redrawn from Martinez 2003).


Figure 4. Detail of male antenna of $D$. riograndensis sp. n.


Figure 5. Detail of forewing with pterostigma, of $D$. riograndensis sp. n. Scale: 0.1 mm .


Figure 6. Detail of the antenna with coarse bristles, of $D$. riograndensis sp. n. Scale: 0.1 mm .


Figure 7. Head, mesoscutum, axilla, and mesoscutellum (dorsal view), of $D$. riograndensis sp. n.. Scale: 0.1 mm .
Key to males of the halidayi species group in the Neotropical region

1 Antenna with five unarticulated branches; $\mathrm{T}_{\text {III }}$ smooth, without punctures.... 2

- Antenna with five articulated branches basally (Fig. 1); $\mathrm{T}_{\text {III }}$ with punctures ...
D. mexicali
$2 \quad \mathrm{~A}_{8}$ same length as $\mathrm{A}_{7} ; \mathrm{A}_{8}$ approximately same length as $\mathrm{A}_{11} \ldots \ldots . . . . . . . . . . . . . . . . . . . . .3$
- $\quad A_{8}$ twice length of $A_{7} ; A_{8}$ longer than $A_{11}$ (Fig. 2) ..................... D. araucanus

3 Antennal branches relatively thin; $A_{7}$ and $A_{8}$ of similar width; $R_{1}$ smaller than $\mathrm{R}_{2} ; \mathrm{R}_{5}$ length approximately similar to $\mathrm{A}_{7}$ (Fig. 3) ...................... D. ranquel

- Antennal branches relatively thick; $A_{7}$ thinner than $A_{8} ; R_{1}$ and $R_{2}$ of same length; $\mathrm{R}_{5}$ much longer than $\mathrm{A}_{7}$ (Fig. 4) .................. D. riograndensis sp. $\mathbf{n}$.


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# An interactive multi-entry key to the species of Megalostomis Chevrolat, with description of a new species from Paraguay (Chrysomelidae, Cryptocephalinae) 

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#### Abstract

The main goal of this contribution is to release an interactive multi-entry key to all known species of the genus Megalostomis Chevrolat. This key constitutes a new tool created to aid the identification of the species of this diverse genus, which occasionally may be difficult to identify to the species-level, due to the lack of reference collections for most countries within its distribution range, and to the presence of intraspecific variation and secondary sexual characters. It is expected that this on-line key will facilitate future periodic updates, and will benefit all those persons interested in identifying these taxa. The present paper also includes the description of Megalostomis juanenrique sp. n., a new species from Paraguay. In addition, Megalostomis gigas Lacordaire, and Megalostomis robustipes Monrós are newly cited for the fauna of Paraguay. The online interactive Lucid key is available at http://keys.lucidcentral.org/keys/v3/megalostomis. Offline Lucid data files in LIF and SDD formats are also available at doi: 10.3897/zookeys.425.7631. app1 and doi: 10.3897/zookeys.425.7631.app2.


## Keywords

Lucid interactive key, Clytrini, Megalostomina, new species, new records

[^0]
## Introduction

South America harbors a high diversity of Clytrini. Megalostomis Chevrolat, 1836 is the largest genus within the Neotropical subtribe Megalostomina, its species are phytophagous in the adult stage, primarily leaf and flower feeders. As larvae, they are case-bearers, some species are known to live in ant nests (Riley et al. 2002). Agrain and Roig-Juñent (2011) recently tested the phylogenetic relationships among its species, proposing two species groups with distinct morphological and biogeographical characteristics. More recently, Agrain (2013) provided a taxonomic review of the genus including all its species. Both former works anticipated that the study of specimens from certain countries would surely yield some taxonomic novelties. This was confirmed when studying a small lot of specimens collected by J. M. Viana, over thirty years ago, in Paraguay. The latter not only corroborates the need for further field and curatorial work for this group, but also inspired the need for having an online key that can be easily improved, corrected and expanded in short time and in one place, as our knowledge of these taxa is progressively enriched. Therefore, the main goal of this contribution is to release an interactive multi-entry key to all known species of the genus Megalostomis Chevrolat ( 43 including the new one). It is expected that this type of key will be more valued by the community than traditional printed keys, granting perpetuity of the key's life, acknowledging the original and subsequent authors (Penev et al. 2009). For more information concerning Lucid keys visit http://www.lucidcentral.org.

## Materials and methods

## Specimens studied

The material examined during this work came from Agr. Eng. Juan Enrique BarrigaTuñon's personal Collection (JEBC).

## Depository of types

IADIZA Instituto Argentino de Investigaciones de las Zonas Áridas, Mendoza, Argentina.
INBP Museo Nacional de la Historia Natural del Paraguay, San Lorenzo, Paraguay.
JEBC Juan E. Barriga T. Collection, Curicó, Chile.
MACN Museo Argentina de Ciencias Naturales, Buenos Aires, Argentina.

## Dissections of adults

Specimens were soaked in $\mathrm{NH}_{3}$ for ten minutes to relax their anatomical structures. To study the male genitalia, the aedeagus was removed through the pygidium and cleared
in $10 \%$ hot KOH for five minutes (for the female the whole abdomen was removed). After the KOH clearing process, the aedeagus was washed in $80 \%$ ethanol and then glycerin was injected through the basal foramen, using a needle and the help of curved dissecting forceps, in order to evert the internal sac. Upon completion of the examination, the genitalia and other dissected parts were placed in a plastic microvial with glycerin, and pinned directly beneath each specimen.

## Character nomenclature

Terminology follows Agrain 2013 (and references therein).

## Illustrations and photography

All photographs were taken by the author. The source photos were taken at multiple focal planes (at least five) and then combined into a single image (with all features in focus) with the free software CombineZP (http://www.hadleyweb.pwp.blueyonder. co.uk/CZP/News.htm). Images for this study were taken with a Canon PowerShot S50 digital camera, mounted on a Leica S6E stereomicroscope. For the matrix-based key, all illustrations of characters and their states were taken from Agrain and RoigJuñent (2011) supplementary material, while all species plates were modified from Agrain (2013), except for the illustration of M. subnitida Guérin [modified from Guérin (1952)], and M. monrosi Medvedev [modified from Medvedev 1998].

## Creation of the multi-entry key

The multi-entry interactive key was created by exporting the nexus data matrix used in Agrain and Roig-Juñent (2011), to DELTA software (http://delta-intkey.com). All taxa, and characters with their respective states were imported from .dlt to .lif file format into Lucid builder 3.3 (http://www.lucidcentral.org), by use of Lucid translator (http://www.lucidcentral.com/en-us/software/lucid3/lucidbuilder/lucidtranslator. aspx). Within Lucid builder 3.3 (free edition), pictures for most character states, species plates, country level occurrence data (taken from Agrain 2013 checklist), and a few autapomorphies that were not in the original data matrix were added in order to maximize the probability of precise identification. Online interactive Lucid key is available at http://keys.lucidcentral.org/keys/v3/megalostomis. Lucid data files are provided in appendices as follows: Suppl. material 1: Lucid Interchange Format version 3 (LIF3) file, and Suppl. material 2: SDD (Structure of Descriptive Data) Schema. Also a copy of the deployed key (LKC4), containing high resolution pictures can be obtained from the author`s website: http://fedeagrain.wordpress.com/lucid-keys/ as a zip file, this is the equivalent to a CD-ROM version, for off-line (local) use, once decompressed you
will need Lucid Player 3.3 installed on your system to visualize it. This player is multiplatform and is freely available at http://www.lucidcentral.com/en-us/software/lucid3/ lucidplayer.aspx.

## Results

## Taxonomy

Megalostomis is easily diagnosable from Cryptocephalinae by the following characters: body length 6-14 mm, width $4-10 \mathrm{~mm}$; prothorax without lateral antennal grooves; tarsal claws simple; prosternum evident between procoxae; procoxae globose; eyes strongly emarginate; and dorsal plate of aedeagus with straight margin. The latter two characteristics were found to be synapomorphies for the genus according to Agrain and Roig-Juñent 2011.

## Megalostomis juanenrique sp. n.

http://zoobank.org/A5DD6649-D388-41FE-BDF4-102FECBF81FD
Figs 1-3

Type locality. PARAGUAY, San Pedro: Cororō (Lat. $23.439011^{\circ}$ S, Long. $56.501807^{\circ} \mathrm{W}$ ).
Type specimens. Holotype: male, pinned. Original labels: "White label (handwritten): Paraguay-San Pe /dro-Córoro/ M. Viana XI_1983_, Blue label (printed): ex Coleccion/ M. VIANA/ ARG 006244, White label (printed): Coleccion / J. E. BARRIGA / CHILE 067786". Red label (printed): Megalostomis juanenrique sp. n. / Holotype/ Des. Agrain F. A. 2014. IADIZA. Allotype: female, pinned, with genitalia in a separate microvial. Original labels: "White label (handwritten): Paraguay.S.P./ Córoro/ M. Viana 1979, Blue label (printed): ex Coleccion/ M. VIANA/ ARG 006337, White label (printed): Coleccion / J. E. BARRIGA / CHILE 067005". Red label (printed): Megalostomis juanenrique sp. n. / Allotype/ Des. Agrain F. A. 2014. IADIZA. Paratype: male, pinned, with genitalia in a separate microvial. Original labels: "White label (printed): PARAGUAY/ Córoro/ dic 1983/ leg. M. Viana, White label (printed): Coleccion / J. E. BARRIGA / CHILE 138773". INBP. Paratype: male, pinned. Original labels: "White label (handwritten): Paraguay-S P/ Córoro/ M. Viana _1976, Blue label (printed): ex Coleccion/ M. VIANA/ ARG 006362, White label (printed): Coleccion / J. E. BARRIGA / CHILE 063063". MACN. Paratype: male, pinned. Original labels: "White label (handwritten): Paraguay-San/ Pedro-Córoro/ XI_1985_ M. Viana, Blue label (printed): ex Coleccion/ M. VIANA/ ARG 006238, White label (printed): Coleccion / J. E. BARRIGA / CHILE 069889". JEBC. Paratype: male, pinned, with genitalia in a separate microvial. Original labels: "White label (handwritten): Paraguay. S.P./ Córoro/ M. Viana 1976, Blue label (printed): ex Coleccion/ M. VI-


Figure I. Megalostomis juanenrique sp. n. A Habitus dorsal (male) B Habitus lateral (male) C Habitus dorsal (female) $\mathbf{D}$ Head of male (frontal view) $\mathbf{E}$ Head of female (frontal view).

ANA/ ARG 006406, White label (printed): Coleccion / J. E. BARRIGA / CHILE 067050 ". JEBC. Paratype: male, pinned, with genitalia in a separate microvial. Original labels: "White label (handwritten): Paraguay. S.P./ Córoro/ M. Viana 1976, Blue label (printed): ex Coleccion/ M. VIANA/ ARG 006270, White label (printed): Coleccion / J. E. BARRIGA / CHILE 057918". JEBC. All paratypes with my label: Red label (printed): Megalostomis juanenrique sp. n./ Paratype/ Des. Agrain F. A. 2014.

Diagnosis. This new species belongs to the Megalostomis grossa species group as defined by Agrain and Roig-Juñent (2011). Characters of this species group exhibited by this species are: uniform antennal coloration, presence of a longitudinal carina at interocular space, and pronotal disc pilosity limited to or denser at its margins; even if this species shares some characters with its relatives (e.g. the hypertrophied eye stalk with M. grandis (Forsberg), it can be easily separated from this and other related species by several characters: male mandibles exceeding length of clypeus, head wider than long, subrectangular pronotal disc longer than high, with its lateral margins visible from above, central dorsal plate of kotpresse (female) with three arms, apical margin


Figure 2. Megalostomis juanenrique sp. n. A-C Male aedeagus and internal sac: A Dorsal view B Lateral view $\mathbf{C}$ Ventral view.
of dorsal plate of aedeagus straight, among others. As a rule, for this genus, the best way to identify the females (without dissecting them) is by comparing the coloration pattern with their respective males.

Body length. $11.3-12.5 \mathrm{~mm}$, width: $6.3-7.2 \mathrm{~mm}$.
Male (Figs 1A-B, D, 2). Coloration pattern: elytra reddish, with three interspaced sub-transverse black bands, all reaching lateral margin of elytra, basal band reaching humeral carina, and apical band reaching elytral apex. Head: anterior surface smooth, with central protuberance at upper region of clypeus, mostly glabrous with disperse pubescence; inter-ocular space with thin longitudinal carina, with a pair of slightly depressed sub-adjacent areas on either side; internal margin of eyes with strongly marked carina, posterior side of eye with salient post-ocular protuberance, next to a marked furrow. Mandibles asymmetric; apex of mandibles with a series of peaks (molariform area) surrounding a depressed area that fits into the right mandible; right mandible curved inward, mandibular apex with a very sharp tooth on its underside; left mandible straight, with sharp bifurcated teeth; with common intra-specific variation in size among examined series. Clypeal margin, straight, without auricular appendix. Antennae: as long as pronotum, black; scape robust; serrate beyond fourth antennomere, eleventh antennomere with two marginal excavations that delimit central lobe. Thorax: pronotal disc: longer than high, sub-rectangular, lateral margins visible from above; posterior projection short; median lateral region with slight transverse constriction; punctation weak and regular; pronotal disc pilosity limited to its margins; scutellum wider than long, with posterior margins curved, covered with white dense pubescence. Elytra: apical margin projected, surpassing pygidium; elytral humeral region wider than pronotal base, gradually narrowing toward apex; diffuse punctation, denser than that of pronotum; elytral margin narrow, enlarged in humeral region; humeral carina rounded, apex with transverse mark. Genitalia: (Fig. 2A-C) apex of dorsal plate of median lobe as wide as its base, with anterior margin straight; lateral arms of median lobe short, with setae; dorsal sclerite of internal sac (not everted) upward-directed, forming a wing-shaped structure in dorsal view; ejaculatory guide with ventral keel; sperm transfer structure with campanulate sclerite.

Female (Figs 1 C, E, 3). Coloration pattern: same as male. Head: smaller than that of male; mandibles compact and short. Antennae: same as male, but shorter than pronotum (with smaller antennomeres). Thorax: pronotum with sides slightly more curved than in male. Abdomen: sternites same as in male, fifth sternite excavate. Pygidium: with apical excavation and, apical transverse depressed area. Rectal sclerites: dorsal rectal sclerites (Fig. 3A) represented only by dorsal (subquadrate) apodemes, and central dorsal plate. Central dorsal plate (Fig. 3B) subquadrangular with three arms; ventral rectal sclerites with very short apodemes. Spermathecal capsule: (Fig. 3C) U-shaped, distal part more than $2 \times$ longer than proximal part, proximal part longer than base; angle formed between basal and apical regions of spermathecal capsule less than $45^{\circ}$; apex of spermathecal capsule long, with sharp tip. Eighth sternite with central tooth (Fig. 3D).

Etymology. Specific name is treated as a noun in apposition (ICZN 1999, Art. 34.2.1), it is dedicated to the distinguished Chilean coleopterist Agr. Eng. Juan Enrique Barriga-Tuñon, who generously received me in his outstanding insect collection.


Figure 3. Megalostomis juanenrique sp. n. A Kotpresse ventral sclerite B Kotpresse central dorsal plate and dorsal apodemes $\mathbf{C}$ Spermathecal capsule $\mathbf{D}$ Sternite VIII.

## Species newly recorded for Paraguay

## Megalostomis gigas Lacordaire

One specimen from: PARAGUAY, San Pedro: Cororō (Lat. $23.439011^{\circ} \mathrm{S}$, Long. $56.501807^{\circ}$ W). February 1979, Leg. J. M. Viana. JEBC.

## Megalostomis robustipes Monrós

One specimen from: PARAGUAY, Dep. Cordillera: Pirareta, (Lat. 25.483333³, Long. $\left.56.933333^{\circ} \mathrm{W}\right) .26-31$ August, 2011. Leg. U. Drechsel. IADIZA.

## An interactive multi-entry key to the species of Megalostomis

The main benefit of the interactive multi-entry key is that the user needs to follow fewer options compared with traditional dichotomous keys, and these options can be freely chosen; this generally leads to more effective identification. More important, both experienced and inexperienced users are more likely to succeed when identifying problematic species (Drinkwater 2009). The software used to visualizes the key allows "filtered" and "ranked" options when determining a specimen. The first option will eliminate taxa that do not coincide with the chosen character states; while the latter option will keep all taxa, but listing them in priority order in accordance with their agreement (expressed in percentages) with the chosen character states. This latter option is likely to reduce erroneous identifications. According to the score analyzer of the software, the key presented in this paper has at most 23 state differences to diagnose the species of Megalostomis. Therefore, when using the key, it is worthwhile to keep adding character states even if the identification is finished, or to start the identification over again, selecting a different set of characters in order to ensure accurate identification. Users of this key should keep in mind that this was created directly from a phylogenetic dataset, so it has more character to choose from than a traditional key which is focused on diagnostic characters. As a final advice, even if the key includes country level occurrences for each species, it is better to start the identification by using morphological characters, including country level occurrences only to confirm identity, since distribution data is lacking for several countries.

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## Supplementary material I

Key to the species of Megalostomis v.1.0, LIF3 file
Federico A. Agrain
Data type: Lucid Interchange Format version 3 (LIF3), file: Key to the species of Megalostomis v.1.0.
Explanation note: This XML-based format allows exchange of the key with other key developers.
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## Supplementary material 2

## Key to the species of Megalostomis v.1.0, SDD file

Federico A. Agrain
Data type: Lucid SDD file: Key to the species of Megalostomis v.1.0.
Explanation note: This is a XML-based file structured using the internationally agreed SDD (Structure of Descriptive Data) Schema. This SDD file may be used to exchange the Lucid key with other SDD-compliant applications.
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# A new and aberrant species of Dugesia (Platyhelminthes, Tricladida, Dugesiidae) from Madagascar 

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#### Abstract

In this paper we report a new species of Dugesia of the family Dugesiidae from Madagascar, representing the fourth species of freshwater planarian known from this global biodiversity hotspot. In some respects the new species is aberrant, when compared with its congeners, being characterized by a head with smoothly rounded auricles, a peculiar course of the oviducts, including the presence of a common posterior extension, and by the asymmetrical openings of the vasa deferentia at about halfway along the seminal vesicle. Further, it is characterized by a ventral course of the ejaculatory duct with a terminal opening, very long spermiducal vesicles and unstalked cocoons. Its diploid chromosome complement with 18 chromosomes represents an uncommon feature among fissiparous species of Dugesia.


## Keywords

Tricladida, Dugesia, Madagascar, morphology, karyology, reproduction, new species

## Introduction

Madagascar, the fourth largest island in the world, is one of the priority global hotspots for biodiversity conservation (Mittermeier et al. 2004, Vieites et al. 2009) with $5 \%$ of the total species in the world and a very high percentage of endemism (70\%).

Unfortunately, this outstanding species richness is greatly endangered, due to rapidly increasing deforestation, soil erosion, and habitat destruction in large areas of the island (Harper et al. 2007, Andreone et al. 2008). This is particularly true for freshwater environments and their communities of benthic invertebrates.

It has been shown that Madagascar represents a global biodiversity hotspot for land planarians (Sluys 1998, 1999) but, unfortunately, freshwater triclads have received much less attention. With respect to freshwater planarians only representatives of the genus Dugesia Girard, 1850 of the family Dugesiidae Ball, 1974 have been reported from Madagascar. This genus is present with three species, two of which are endemic, viz. Dugesia debeauchampi De Vries, 1988, D. myopa De Vries, 1988, and D. milloti De Beauchamp, 1952 (cf. De Beauchamp 1952, De Vries 1988, Stocchino et al. 2002) (Table 1). The first species is known only from the type locality, Nosy Bé Island, north-west of Madagascar. Dugesia myopa is reported from two localities, viz. Andranoboka Cave, northwest of Mahajanga, and from Andringitra, in the southeast of the island. In contrast, $D$. milloti is recorded from widely scattered Madagascan localities, viz. Montagne d'Ambre, Ivohibe, Morafenobé, and occurs also on the island of Anjouan in the Comoros Archipelago (De Beauchamp 1952, De Vries 1988).

A recent field survey performed by one of us (R. Manconi) on the presence and distribution of Madagascan aquatic invertebrates in some unexplored lentic or lotic freshwaters of the High Plateau and the oriental slope (31 sites) yielded several new records of planarians, suggesting that species richness of this taxon in the island is underestimated.

In this paper we describe a new, and in some respects aberrant, species of Dugesia that was identified on the basis of morphological and karyological data. This contribution represents a first step of a more comprehensive faunistic and taxonomic study of planarian populations on Madagascar. An integrative taxonomic analysis, including molecular and morphological data, is in progress in order to analyze in more detail the problematic position of Madagascan freshwater triclads in an historical biogeographic scenario involving the splitting of Gondwana (cf. Sluys et al. 1998).

## Materials and methods

The collected specimens were transferred to the laboratory and were reared in glass bowls under semi-dark conditions at $18+/-2^{\circ} \mathrm{C}$; the worms were fed with fresh beef liver.

For morphological study specimens were fixed for 24 hours in Bouin's fluid, dehydrated in an ascending ethanol series, cleared in clove oil, and embedded in synthetic paraffin. Serial sections were made at intervals of $6-8 \mu \mathrm{~m}$ and were stained with Mallory-Cason. Reconstructions of the copulatory apparatus were obtained by using a camera lucida attached to a compound microscope.

For karyological analyses metaphasic plates were obtained by the squashing method and also air drying (splashing), following Vacca et al. (1993). The squashing method

Table I. Annotated checklist of Madagascan freshwater triclads.

| Taxa | Geographic Distribution and Habitat | References |
| :---: | :---: | :---: |
| Dugesiidae Ball, 1974 |  |  |
| Dugesia Girard, 1850 |  |  |
| Dugesia bifida Stocchino \& Sluys, 2014 | small unnamed tributary of the Mania River (type locality), southern branch of the High Tsiribihina hydrographic basin, western slope, south-east Madagascar, coll. R. Manconi, September 2011 | present paper |
| Dugesia debeauchampi <br> De Vries, 1988 | Teyiamarango Stream (type locality), Nosy Bé Island, north Madagascar, coll. F. Starmühlner, 1958 | De Vries 1988 |
| Dugesia milloti <br> De Beauchamp, 1952 | Morafenobé, Mahajeby Forest (type locality), Manambaho hydrographic basin, western slope, central-west Madagascar, coll. M. Paulian; Ivohibe, eastern slope, south-east Madagascar, coll. J. Millot, 1950; Mutsamudu Fall, Anjouan, Comoros Archipelago, coll. J. Millot, October 1953; Roussette Stream, Montagne d'Ambre area, north-eastern slope and Ambre, north Madagascar, 17 September 1957 | De Beauchamp 1952, <br> De Vries 1988 |
| Dugesia myopa <br> De Vries, 1988 | Zanadoa, Andringitra, south-east Madagascar, 1949; Andranoboka Cave (type locality), northwest of Mahajanga, Betsiboka hydrographic basin, western slope, north-west Madagascar, coll. M. Paulin, December 1951 | De Beauchamp 1952, <br> De Vries 1988 |

was performed on single caudal regenerative blastemas of 10 specimens in order to verify the uniformity of the chromosome complement of the strain. The air drying method was performed on 15 intact specimens, thus yielding good metaphasic plates for karyometrical analysis. The chromosome complement was characterized on the basis of 6 metaphasic plates. Karyometric values were calculated after first arranging the chromosomes according to their gradually decreasing lengths. Relative length was calculated as chromosome length $\times 100 /$ total length of the haploid genome. Centromeric index was calculated as length of short arm $\times 100 /$ total length of the chromosome. Chromosomal nomenclature follows Levan et al. (1964).

The histological material is deposited at Naturalis Biodiversity Center, Leiden, The Netherlands (ZMA collection code), and in the Giacinta A. Stocchino collection (CGAS), University of Sassari.

## Abbreviations used in the figures

bc: bursal canal; ca: common atrium; cb: copulatory bursa; cg: cement glands; cm: circular muscle; cpe: common posterior oviducal extension; d: diaphragm; du: ductule; e: eye; ed: ejaculatory duct; epg: extra bulbar penial glands; g: gonopore; Im: longitudinal muscle; lob: left oviducal branch; lod: left oviduct; lvd: left vas deferens; ma: male atrium; o: ovary; od: oviduct; ov: oviducal vesicle; pb: penis bulb; pg: penial glands; ph: pharynx; pp: penis papilla; rod: right oviduct; rob: right oviducal branch; rvd: right vas deferens; $s$ : sperm; sg: shell glands; sp: spermatophore; sv: seminal vesicle; spv: spermiducal vesicle; t: testes; tu: tuba; v: vitellarium.

## Results

## Systematic Account

Order Tricladida Lang, 1884
Suborder Continenticola Carranza, Littlewood, Clough, Ruiz-Trillo, Baguñà \& Riutort, 1998
Family Dugesiidae Ball, 1974
Genus Dugesia Girard, 1850

## Dugesia bifida Stocchino \& Sluys, sp. n.

http://zoobank.org/E72381C7-6FC3-422F-A8DA-1D4D0856B521
Figs 1-7; Tables 1-2

Material examined. Holotype: ZMA V.Pl. 7189.1, one set of sagittal sections on 8 slides, Central High Plateau, between Antsirabe ( $19^{\circ} 86^{\prime} 32^{\prime \prime} \mathrm{S}, 47^{\circ} 03^{\prime} 36^{\prime \prime} \mathrm{E}$ ) and Ambositra ( $20^{\circ} 53^{\prime} 14^{\prime \prime} \mathrm{S}, 47^{\circ} 24^{\prime} 61^{\prime \prime} \mathrm{E}$ ), near the small village of Antsariboti, Madagascar, 16 September 2011, coll. R. Manconi.

Paratypes: CGAS Pla 7.1, ibid., sagittal sections on 7 slides; CGAS Pla 7.2, ibid., sagittal sections on 4 slides; CGAS Pla 7.3, ibid., transverse sections on 20 slides. CGAS Pla 7.4-5, ibid., horizontal sections on 4, 7, slides respectively; ZMA V.Pl. 7189.2, ibid., horizontal sections on 6 slides; ZMA V.Pl. 7189.3, ibid., horizontal sections on 3 slides; ZMA V.Pl. 7189.4 , ibid., horizontal sections on 4 slides; ZMA V.Pl. 7189.5, ibid., sagittal sections on 7 slides; ZMA V.Pl. 7189.6, ibid., horizontal sections on 5 slides.

Diagnosis. Dugesia bifida is characterized by the presence of the following features: body slender; head with smooth, rounded auricles; oviducts that recurve before opening into the bursal canal and provided with a common posterior extension; slightly asymmetrical openings of the oviducts into the bursal canal; absence of ectal reinforcement; large seminal vesicle; asymmetrical openings of the vasa deferentia into the seminal vesicle, the openings situated at halfway along the vesicle; very long spermiducal vesicles; large diaphragm; ventral course of the ejaculatory duct; terminal opening of the ejaculatory duct; unstalked cocoons; chromosomal number $2 \mathrm{n}=18$.

Etymology. The specific epithet is derived from the Latin adjective bifidus, split into two parts, and alludes to the fact that the peculiar long common oviduct splits into two branches, each branch subsequently opening into the bursal canal.

Geographical distribution. Known only from the type locality in the High Tsiribihina hydrographic basin, Madagascar.

Habitat. Planarians were found in running water in a paddy field area at an altitude ca. 1300 m asl in the Central High Plateau, along Route Nationale 7, between Antsirabe and Ambositra, near the small village of Antsariboti (Fig. 1). The small, unnamed stream is a tributary of the Mania River in the southern branch of the High Tsiribihina hydrographic basin. The animals, scattered and not abundant,


Figure I. Geographic distribution of Dugesia species recorded from Madagascar and adjacent islands. Type locality of D. bifida in the High Tsiribihina hydrographic basin indicated by an asterisk.
were collected from running clear water, under small pebbles on coarse sand at a depth of $3-10 \mathrm{~cm}$. A survey of ca. 50 pebbles, performed at the end of the dry season (September), revealed complete absence of planarian cocoons, as well as other invertebrates, excepting very small larvae of mayflies.

Description. Body of living specimens slender, ranging from 6 to 7 mm in length and $0.4-0.6 \mathrm{~mm}$ in width in fissiparous specimens and from $11-15 \mathrm{~mm} \times 1.5-2 \mathrm{~mm}$


Figure 2. Dugesia bifida. Habitus of living specimens, $\mathbf{A}$ fissiparous individual $\mathbf{B}$ ex-fissiparous individual.
in sexualized specimens. Two eyes present in the centre of the head; unpigmented auricular grooves marginally placed just posteriorly to the eyes. Head with smooth, rounded auricles and with five sensory fossae on either side of its anterior margin.

The dorsal surface light grey-brown, with two darker lateral stripes running from the central part of the pharynx to its posterior part, where they form a single median stripe that runs to the tail. In sexualized specimens the pigmentation is darker than fissiparous animals (Fig. 2). The ventral surface is paler than the dorsal body surface.

The pharynx is positioned in the posterior half of the body and measures about $1 / 9$ th of the body length. Inner and outer pharyngeal musculature bilayered, i.e. without an extra, third, outer longitudinal muscle layer in the inner sheath of muscles.

The ovaries, localized just behind the brain, are weakly hyperplasic. They occupy half of the dorso-ventral space of the body and are particularly expanded in horizontal direction. The anterior portion of the infranucleated oviducts is expanded into


Figure 3. Dugesia bifida. Holotype ZMA V.Pl. 7189.1, sagittal reconstructions of the copulatory apparatus (anterior to the right), A male copulatory apparatus $\mathbf{B}$ female copulatory apparatus.
a tuba that may communicate, at a poorly defined position, with the dorsal side of the ovaries or with the center of the ovarian masses, dependent upon the hyperplasic condition of the ovaries (Fig. 6A). The oviducts run ventrally in a caudal direction to beyond the level of the genital pore and, subsequently, recurve anteriad to open at the same level into a long, posterior duct with an histology similar to that of an oviduct. For descriptive purposes we consider this to be a common posterior oviducal extension. The right oviduct opens dorsally into this long common duct while the left oviduct opens ventrally. From this point the common posterior duct divides into two branches, which open separately and asymmetrically through the posterior wall of the bursal canal. The left branch opens slightly dorsally to the right one. The openings of these two branches into the vertically running section of the bursal canal are situated close together (Figs 3A, B, 4). The lumen of the common posterior oviducal extension, and also that of the two branches contains ample sperm. In CGAS Pla 7.1 specimen the most posterior part of the common posterior oviducal extension communicates through a thin ductule with the ventral part of an adjacent vitellarium (Fig. 5C). In the holotype ZMA V.Pl. 7189.1 and in specimens CGAS Pla 7.1 and CGAS Pla 7.2 the


Figure 4. Dugesia bifida. Schematic horizontal reconstruction of the copulatory apparatus.
lumen of the oviducts has an irregular diameter and is generally quite spacious, thus in some parts forming a kind of vesicle (Fig. 6B).

The numerous mature, fully developed testes are situated dorsally and extend from the level of the ovaries to the posterior end of the body. Spermatogenesis appears to proceed in a regular fashion, in that no anomalies, such as irregularly shaped spermatids and spermatozoa, were observed (Fig. 6C). Vitellaria are located between the testes and the intestinal branches and extend to some distance posteriorly to the copulatory apparatus.

The large copulatory bursa is lined by a columnar, glandular epithelium bearing basal nuclei and it is surrounded by a layer of muscles. In the holotype ZMA V.Pl. 7189.1 a spermatophore full of sperm is present in the lumen of the bursa. From the postero-dorsal wall of the bursa the bursal canal runs in a caudal direction to the left


Figure 5. Dugesia bifida. Microphotographs of the copulatory apparatus. A Holotype ZMA V.Pl. 7189.1, sagittal section showing the penis bulb ( pb ) and the penis papilla ( pp ) with the seminal vesicle (sv) and the ejaculatory duct (ed) B Holotype ZMA V.Pl. 7189.1, sagittal section showing the opening of the right oviducal branch (rob) through the posterior wall of the bursal canal (bc), and the common posterior oviducal extension (cpe) full of sperm (s) C Paratype CGAS Pla 7.1, sagittal section showing the caudal part of the common posterior oviducal extension (cpe) and the ductule (du) communicating with the ventral part of an adjacent vitellarium (v).


Figure 6. Dugesia bifida. A Holotype ZMA V.Pl. 7189.1, microphotograph of the right hyperplasic ovary (o) with the tuba (tu) B Paratype CGAS Pla 7.1, microphotograph of the oviduct (od) with an expansion (ov) C Holotype ZMA V.Pl. 7189.1, microphotograph of mature testes ( t ) with sperm (s).
of the copulatory apparatus and, after a narrowing, communicates with the common atrium. The bursal canal is lined with cylindrical, infranucleated, ciliated cells and is surrounded by a thin subepithelial layer of longitudinal muscles, followed by a layer of circular muscles. Ectal reinforcement is absent. The very abundant shell glands open into the vaginal section of the bursal canal, at the level of the oviducal openings (Figs 3A, 5B).

The scarcely developed penis bulb, rich in glands, consists of intermingled longitudinal and circular muscle fibres. Extra-bulbar penial glands (staining yellow with Mallory-Cason) penetrate the penis bulb at its dorsal and ventral side. The penis bulb houses a very large, flask-shaped seminal vesicle, lined with a nucleated epithelium (Figs 3A, 5A). The vasa deferentia penetrate the proximal, anterior section of the penis bulb and open separately and asymmetrically into the seminal vesicle at a position about halfway along the vesicle, at the point where it narrows. The right vas deferens opens dorsally to the left one. The seminal vesicle opens into the ejaculatory duct via a large, valve-like diaphragm. In all specimens examined the sperm ducts form welldeveloped spermiducal vesicles, packed with sperm. These vesicles are very long and extend over a large distance, viz. from the root of the pharynx to the penis bulb. The diaphragm, located approximately at the base of the penis papilla, receives the openings of penis glands. The stubby, asymmetrical penis papilla is covered by an infranucleated epithelium that is underlain with a subepithelial layer of longitudinal muscles. The ejaculatory duct follows a ventral course and has a terminal opening. A ventrally displaced course of the ejaculatory duct is present in all specimens examined, albeit that this condition is more clearly expressed in some specimens as compared to others, depending on the state of contraction of the penis papilla. For example, in paratype V.Pl. 7189.5 the penis papilla is cone-shaped and shows a distinctly ventrally displaced ejaculatory duct, with a terminal opening. A similar situation is present in paratype CGAS Pla 7.2. In contrast, in the holotype and in paratype CGAS Pla 7.1 the penis papilla is much more stubby, due to contraction, with the result that the ventral course of the ejaculatory duct is much less pronounced. The ejaculatory duct, which in most of the specimens examined contained an empty spermatophore, is lined by a cuboidal, infranucleated epithelium (Figs 3, 5A).

The genital atrium is divided into a common atrium and a male atrium and is lined by an infranucleated epithelium that is underlain by a subepithelial layer of circular muscles, followed by a layer of longitudinal muscle fibres. The common atrium opens ventrally through the gonopore, which receives the openings of the cement glands (Figs 3A, B, 5A, B).

## Karyology

Metaphasic plates revealed that the specimens constantly showed a set of 18 chromosomes. Chromosomes from six metaphasic plates could be arranged, according to their length, into nine groups of two chromosomes with a diploid chromosome set


Figure 7. Dugesia bifida. A metaphasic plate $\mathbf{B}$ karyogram $\mathbf{C}$ idiogram.

Table 2. Dugesia bifida. Mean values and standard deviations of the relative length (r. l.) and centromeric indices (c. i.).

| Chromosome |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | $\mathbf{1}$ | $\mathbf{2}$ | $\mathbf{3}$ | $\mathbf{4}$ | $\mathbf{5}$ | $\mathbf{6}$ | $\mathbf{7}$ | $\mathbf{8}$ | $\mathbf{9}$ |
| r.l. | $13.84 \pm$ | $13.12 \pm$ | $12.14 \pm$ | $11.38 \pm$ | $11.06 \pm$ | $10.65 \pm$ | $9.98 \pm$ | $9.27 \pm$ | $8.49 \pm$ |
|  | 0.68 | 0.41 | 0.45 | 0.16 | 0.34 | 0.40 | 0.37 | 0.20 | 0.57 |
|  | $41.55 \pm$ | $47.04 \pm$ | $45.46 \pm$ | $47.04 \pm$ | $47.54 \pm$ | $48.36 \pm$ | $48.23 \pm$ | $49.50 \pm$ | $48.06 \pm$ |
| c.i. | 3.40 | 1.73 | 3.35 | 1.04 | 2.72 | 2.68 | 1.57 | 1.20 | 1.59 |

of $2 \mathrm{n}=18 ; \mathrm{n}=9$. Analysis within each group of chromosomes revealed uniformity in both length and centromeric position. Chromosomal length decreases gradually, with low standard deviation values. Centromeric indices showed great variation, in particular for some chromosomes that exhibit high standard deviations, such as numbers 1 and 3 (Fig. 7, Table 2).

The karyometric data indicate that the diploid chromosome complement is characterized by metacentric heterobrachial chromosomes, with the exception of chromosome 8 , which is metacentric, bordering on metacentric isobrachial (Table 2).

## Life cycle

The life cycle was monitored for two years under laboratory conditions. All 22 individuals were asexual at collection at the end of the dry season of a particularly arid year (September, 2011). In the laboratory the strain notably increased in numbers due to asexual reproduction by fission. After having been kept in the laboratory for about eight months fissiparous specimens displayed a sexualization process (ca. 4\%) producing ex-fissiparous individuals characterized by hyperplasic ovaries, large body size and development of the copulatory apparatus. This sexualization process, as well as mating, occurred from spring to summer (May-September), followed by fertile cocoon deposition from June to November. After a growth phase of ca. three months, the juveniles divided repeatedly, thus producing new fissiparous clones. After cocoon deposition, the ex-fissiparous individuals resorbed the copulatory apparatus and returned to the fissiparous mode. All cocoons are characterized by the absence of a pedicel, and were cemented firmly to the substratum, i.e. to the wall and bottom of the bowls in which the animals were reared.

## Discussion

Dugesia bifida differs from its congeners in its external morphology, in particular the head shape, the peculiar course of the oviducts at the level of the copulatory apparatus, including the presence of a common posterior extension, and in the asymmetrical openings of the vasa deferentia at about halfway along the seminal vesicle. Further, it is characterized also by a ventral course of the ejaculatory duct with a terminal opening, a large seminal vesicle, and unstalked cocoons.

Almost all known species of Dugesia are characterized by a distinctly triangular head with pointed auricles, whereas in D. bifida the latter are much more smoothly rounded. The only other species of Dugesia with a peculiar external morphology is $D$. milloti, which is characterized by a head with a high triangular shape and prominent, pointed auricles. The latter species is known only from Madagascar and from the island of Anjouan in the Comoros Archipelago (De Beauchamp 1952, De Vries 1988). It is remarkable that these two Dugesia species with an aberrant external morphology both occur on Madagascar.

The peculiar condition of the oviducts in $D$. bifida lies in the fact that the ducts open into a common posterior extension, which anteriorly divides into two branches before opening into the vaginal section of the bursal canal. Such a condition was never reported before for the genus Dugesia. Usually, in this genus the oviducts run in caudal direction and at the level of the copulatory apparatus open symmetrically or asymmetrically into the vaginal part of the bursal canal. Exceptions to this rule are D. myopa
and D. congolensis De Beauchamp, 1951 from the Afrotropical Region, D. mertoni (Steinmann, 1914) from the Australasian region, $D$. deharvengi Kawakatsu \& Mitchell, 1989, and D. andamanensis (Kaburaki, 1925) from the Oriental Region, in which the oviducts fuse to form a short common oviduct before opening into the bursal canal. Two other species, viz. D. lindbergi De Beauchamp, 1959 from the Palaearctic and Oriental regions and D. uenorum Kawakatsu \& Mitchell, 1995 from the Australasian Region are characterized by a polymorphism concerning symmetrical openings and a common oviducal condition (cf. Sluys et al. 1998).

That in D. bifida the common posterior extension belongs to the oviducts is demonstrated by the fact that the histological architecture of this common duct is the same as the rest of the oviducts. The continuation of the oviducts caudally to the copulatory apparatus may be functionally related to the presence of vitellaria in the tail region.

This common posterior extension of the oviducts in D. bifida reminds one of the caudally branched oviducts reported for some other genera of the Dugesiidae viz. Spathula Nurse, 1950 from New Zealand and Australia, Reynoldsonia Ball, 1974, and Eviella Ball, 1977 from Australia, and three species of the genus Romankenkius Ball, 1974 from Tasmania (cf. De Vries and Sluys 1991, Sluys 2001). According to Sluys' (2001) phylogenetic analysis, presence of caudally branched oviducts could be considered as a synapomorphy for a group comprising the genera Spathula, Eviella, and Reynoldsonia, albeit that there are also three cases of parallelism, viz. Romankenkius sinuosus Sluys \& Kawakatsu, 2001, R. libidinosus Sluys \& Rohde, 1991, and R. pedderensis Ball, 1974 [note that this interpretation differs somewhat from that discussed by Sluys (2001, p. 70)].

Caudally branched oviducts occur also in other species belonging to different freshwater families, such as the dendrocoelid Macrocotyla glandulosa Hyman, 1956 and the cavernicolan Rhodax evelinae, Marcus, 1946 (cf. Sluys 2001).

Among African and Madagascan species of Dugesia absence of ectal reinforcement in D. bifida is shared only with D. aethiopica Stocchino et al., 2002 and D. afromontana Stocchino \& Sluys, 2012 (Stocchino et al. 2002, 2012).

From Madagascar only three species of Dugesia have been reported up to this moment, viz. Dugesia debeauchampi, D. milloti, and D. myopa (De Beauchamp 1952, De Vries 1988) (Table 1). In contrast to D. bifida, a central course of the ejaculatory duct is displayed by these three species, while $D$. myopa also has a short common oviduct and reduced eyes. Dugesia bifida shares with D. milloti and D. myopa a large diaphragm, whereas $D$. debeauchampi has a small diaphragm.

As for life history, the life cycle of D. bifida under laboratory conditions is comparable to that of other African species, such as D. aethiopica and D. afromontana, in which post-pharyngeal transverse fissioning occur continuously, while sexual reproduction followed by fertile cocoon deposition is less frequent, involving only a small percentage of individuals. In contrast to D. aethiopica, ex-fissiparous specimens of D. bifida do not retain the fissioning ability during their sexual state (cf. Stocchino and Manconi 2013).

In those species of Dugesia in which ex-fissiparous specimens develop from fissiparous strains, these sexualized individuals are characterized by the presence of hyperpla-
sic ovaries and underdeveloped testes, in which germ cells show degenerative processes (cf. Stocchino et al. 2012). In contrast, in D. bifida the testes are well developed in all specimens examined, without anomalies of the germ cells. Moreover, the hyperplasic ovaries in $D$. bifida are not visible through the dorsal body wall of living animals, in contrast with other species, such as D. sicula Lepori, 1948, D. maghrebiana Stocchino et al., 2009, D. aethiopica, D. afromontana, and D. arabica Harrath \& Sluys, 2013 (Stocchino et al. 2012, Stocchino and Manconi 2013, Harrath et al. 2013). This may be due to the fact that in $D$. bifida the hyperplasic ovaries are more weakly developed and more expanded in horizontal direction than in vertical direction.

The cocoons of $D$. bifida are unstalked but firmly attached by cement to the substratum. The production of unstalked cocoons is an uncommon condition in the genus Dugesia, for which they are generally reported as being provided with a pedicel and a terminal plate (Gourbault 1972, Ball and Reynoldson 1981). Among dugesiids, stalked cocoons are known also from Schmidtea Ball, 1974 (cf. Gourbault 1972), Cura Strand, 1942 (Ball 1974, Grant et al. 2006), and Girardia dorotocephala (Woodworth, 1897) (cf. Gourbault 1972). In Spathula ochyra Ball \& Tran, 1979 the cocoons are enclosed in a jelly-like dome, attached to the substratum (Grant et al. 2006). Unstalked cocoons that can be fastened to the substratum very lightly or attached firmly to it by cement, are reported for the dugesiid genus Neppia Ball, 1974, the planariid genera Phagocata Leidy, 1847 and Seidlia Zabusov, 1911, and the dendrocoelid genus Dendrocoelopsis Kenk, 1930. Specifically, unstalked cocoons have been reported for the planariid species Planaria torva (Müller, 1774), Polycelis nigra (Müller, 1774), P. tenuis Ijima, 1884, P. felina (Dalyell, 1814), and Crenobia alpina (Dana, 1776), and for the dendrocoelids Bdellocephala punctata (Pallas, 1774), Dendrocoelum lacteum (Müller, 1774), D. album (Steinmann, 1910), D. romanodanubialis (Codreanu, 1949), and D. vesiculosus Stocchino \& Sluys, 2013, (Gourbault 1972, Ball 1974, Ball and Reynoldson 1981, Stocchino et al. 2013, Kawakatsu pers. comm., Stocchino pers. obs.).

As for karyology, D. bifida shows a diploid chromosome complement of 18 chromosomes with basic number $\mathrm{n}=9$. Among Dugesia species this basic number is shared by only six other species: D. sicula, D. maghrebiana and D. biblica Benazzi \& Banchetti, 1972, from the Mediterranean region; D. arabica from Yemen; D. aethiopica and D. afromontana from the Afrotropical region (cf. Stocchino et al. 2004, Stocchino et al. 2012, Harrath et al. 2013).

Dugesia bifida represents the easternmost record of a species with a basic chromosomal number $\mathrm{n}=9$. However, $D$. bifida differs from all of these other species in that it is the only species in which fissiparous specimens exhibit a diploid chromosome complement. Dugesia sicula and D. biblica have sexual and fissiparous populations with diploid and triploid chromosome complements, respectively (Pala et al. 1995, Stocchino et al. 2012). Dugesia afromontana is known only from two fissiparous populations with a triploid chromosome complement (Stocchino et al. 2012). Dugesia maghrebiana is represented by a fissiparous population, characterized by a condition of diffuse mosaicism or mixoploidy, with each individual having triploid and tetraploid cells (Stocchino et al. 2009). The only fissiparous population of D. aethiopica shares
with $D$. maghrebiana the condition of mixoploidy, but chromosome complements of the former species are diploid and triploid. Dugesia arabica shows a combination of the above-mentioned conditions, in that it has diploid populations that reproduce sexually, triploid fissiparous populations, and mixoploid (diploid and triploid) populations reproducing sexually as well as by fission (Harrath et al. 2013).

With respect to this group of six species discussed above, it turns out that fissiparous populations are always triploid or mixoploid, while sexual populations are diploid.

The presence of well developed testes with very abundant sperm and weakly hyperplasic ovaries in ex-fissiparous specimens of D. bifida may be related to its diploid condition, in that it allows a more regular meiosis. A similar condition was reported for $D$. colapha Dahm, 1967 from Ghana, in which the fissiparous populations that produced ex-fissiparous individuals under laboratory conditions were characterized by regular diploid chromosome complements ( $2 \mathrm{n}=16 ; \mathrm{n}=8$ ). However, in this case, ex-fissiparous individuals were always sterile, despite their regular gametogenesis (Dahm 1967).

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# A new species of Litarachna (Acari, Hydrachnidia, Pontarachnidae) from a Caribbean mesophotic coral ecosystem 

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#### Abstract

New records of pontarachnid mites (Acari: Hydrachnidia) from the Caribbean island of Puerto Rico are presented. Litarachna lopezae sp. n., from substrata collected from Bajo de Sico, a mesophotic coral reef ecosystem in Mona Passage off Puerto Rico, is described as new to science. The new species was collected from nearly 70 m depth, the greatest depth from which pontarachnid mites have been found until now. In addition, a Litarachna sp. was also found in association with the tube of the polychaete Sabellastarte magnifica (Shaw, 1800) at the shallow waters of north Puerto Rico.


## Keywords

Water mites, taxonomy, marine environment, Puerto Rico, biodiversity

## Introduction

The water mite family Pontarachnidae Koenike, 1910, the only family of the Hydrachnidia occurring in the marine environment, represents a well-defined monophyletic clade. Most species are reported from the littoral zone of marine waters in tropical

[^1]and subtropical areas. In Australia and South Africa species have been recorded from estuarine freshwaters, and two species in South Korea are known to live only in marine interstitial environments (Pešić 2013). Nothing is known about the life cycle of the Pontarachnidae. So far, three species are known from the Caribbean Sea, i.e. Litarachna degiustii Cook, 1958 (Bimini, Bahamas - Cook 1958, Netherlands Antilles - Pešić et al. 2008), L. caribica Pešić, Chatterjee \& Schizas, 2008 (Netherlands Antilles - Pešić et al. 2008) and Pontarachna nemethi Pešić, Chatterjee \& Schizas, 2012 (Vieques Island of Puerto Rico - Pešić et al. 2012b).

Mesophotic coral ecosystems (MCEs) are light-dependent habitats dominated by macroalgae, sponges and scleractinian corals and are found on the insular and continental slopes of Caribbean islands between 30 and 100 m (Locker et al. 2010). Even at the lower depth range $(70-100 \mathrm{~m})$, there is enough light for photosynthesis to take place enhancing the growth of several scleractinian coral species (e.g. Agaricia spp., Montastraea spp.) and algae. The MCEs of Puerto Rico represent a potential biodiversity hotspot for marine arthropods and so far 2 mites (Pešić et al. 2012b, current paper), 1 harpacticoid copepod (Corgosinho and Schizas 2013) and 9 cumaceans (Petrescu et al. 2012, 2013, 2014a, b) new to science have been described.

In this paper we describe a new species, Litarachna lopezae sp. n. collected during the second of 3 mesophotic cruises (2010-2012) organized by the University of Puerto Rico at Mayagüez (UPRM), the Caribbean Coral Reef Institute (CCRI) and the Department of Marine Sciences (DMS) of UPRM (Sherman et al. 2013). We also describe the female specimen of a tentative new species which was found in association with the polychaete Sabellastarte magnifica (Shaw, 1800) from a shallow water habitat in north Puerto Rico.

## Materials and methods

Material examined in the present study was collected from Bajo de Sico ( $18^{\circ} 14^{\prime} 41.676^{\prime \prime} \mathrm{N}, 67^{\circ} 24^{\prime} 45.791^{\prime \prime} \mathrm{W}$ ), a mesophotic reef formation located in Mona Passage off Puerto Rico. During the 2011 Mesophotic Cruise of DMS-UPRM, divers equipped with Tri-Mix Rebreathers collected substrata (loose rubble, corals, sponges, algae) from 69.5 m depth and placed them in sealed plastic bags. As soon as the samples returned to the surface they were placed on a 1 mm and 0.125 mm sieves. Samples were washed with filtered seawater and the portion of fauna retained on the 0.125 mm sieve was preserved in $95 \%$ ethanol. One specimen was collected from Rio Grande, Puerto Rico ( $18^{\circ} 25^{\prime} 11.86^{\prime \prime N}, 65^{\circ} 47^{\prime} 40.43$ "W) from marine littoral. This specimen was found while tubes of the polychaete Sabellastarte magnifica were washed into a 0.063 mm sieve. Mites and other fauna were extracted by Alexandra Galindo and the fourth author with the aid of a stereomicroscope and placed back in 95\% ethanol. Slide-mounting was done in Hoyer's fluid and water mites were treated in laboratory as decribed by Gerecke et al. (2007). All drawings were prepared using a drawing tube attached to a Olympus BX43 brightfield microscope. The holotype
and paratypes are planned to be deposited in the Museum of Natural History of Montenegro in Podgorica.

All measurements are given in $\mu \mathrm{m}$. The following abbreviations are used: $\mathrm{Cx}-\mathrm{I}=$ first coxae, $\mathrm{dL}=$ dorsal length, $\mathrm{H}=$ height, $\mathrm{L}=$ length, $\mathrm{I} / \mathrm{II} / \mathrm{III} / \mathrm{IV}-\mathrm{L}-1-6=$ first to sixth segments of the first to fourth legs, $\mathrm{P}-1$ to $\mathrm{P}-5=$ palp segments 1 to 5 , vL $=$ ventral length, $\mathrm{W}=$ width.

## Systematics

## Genus Litarachna Walter, 1925

## Litarachna lopezae sp. n.

http://zoobank.org/4C577A5A-9287-476A-A611-12DD3667687C
Figs 1A-B, 2, 3A-E
Type series. Holotype male, dissected and slide mounted, Puerto Rico, Bajo de Sico, $18^{\circ} 14^{\prime} 41.676^{\prime \prime} \mathrm{N}, 67^{\circ} 24^{\prime} 45.791^{\prime \prime} \mathrm{W}$, depth $69.5 \mathrm{~m}, 20 . \mathrm{iv} .2011$. Paratypes: three males, two females, one deutonymph, same data as holotype, one male and one female dissected and slide mounted.

Diagnosis. Adults. Idiosoma small (L 250-300 $\mu \mathrm{m}$ ); first coxal plates fused; glan-dularium-like structure fused with Cx-IV, a pair of small platelets with (according to Wiles et al. 2002) coxoglandularia 4 and associated setae free in the integument near the lateral posterior apodemes of Cx-IV; ventral margin of P-4 with a setal tubercle and a small peg-like seta.

Description. General features - First coxal plates fused medially; suture lines Cx-I/ II complete, suture line Cx-II/III and Cx-III/IV incomplete. Posterior margin of CxIV with two pairs of apodemes of moderate length, the medial longer than lateral ones, extending beyond anterior margin of genital field; glandularium-like structure on the outer side of lateral posterior apodemes of Cx-IV, fused with the fourth coxal plates; a pair of small platelets with coxoglandularia 4 and associated setae free in the integument near the lateral posterior apodemes of Cx-IV; posterior to the genital field a pair of platelets with three pores, and three pairs of small wheel-like acetabula, with relatively few radiating spokes. Excretory pore unsclerotized, near posterior end of idiosoma. Palp: ventral margin of P-2 concave without extension; ventral margin of P-4 with a setal tubercle and a small peg-like seta. Legs (Fig. 3C): swimming seta numbers: III-L-5, 1; IV-L-4, 1; IV-L-5, 1. Male: genital field consisting of a sclerotized ring with four pairs of setae; four pairs of perigenital setae free in integument around genital field. Female: pre and postgenital sclerites bowed.

Deutonymph. As in adults but lacking genital field; glandularium-like structure free in the integument on the outer side of lateral posterior apodemes of $\mathrm{Cx}-\mathrm{IV}$.

Measurements. Male (holotype, in parentheses paratype, $\mathrm{n}=1$ ) - Idiosoma (ventral view, Figs 1A, 2) L 258 (268), W 234 (230); coxal field L 116 (106), Cx-III W 154


Figure I. Litarachna lopezae sp. n., Bajo de Sico (A male, B female): idiosoma, ventral view. Scale bar $=100 \mu \mathrm{~m}$.
(158); ring-shaped genital plate L 29 (31), W 25 (24); chelicera total L (116). Palp (Figs 3A-B): total L 183 (182), dL/H, L/H ratio: P-1, 17/12, 1.38 (16/12, 1.3); P-2, 52/28, 1.89 (53/29, 1.86); P-3, 22/23, 0.97 (23/23, 1.0); P-4, 67/17, 4.0 (67/17, 4.0); P-5, 25/10, 2.5 (23/10. 2.4); dL P-2/P-4 ratio 0.78 (0.79); dL of I-L-3-6: 35, 34, 53 (52), 71 (73); I-L-6 H 17 (15), I-L-6 dL/H ratio 4.3 (4.7); dL of IV-L-2-6: 35, 42, 68, 88, 88.

Female - Idiosoma (ventral view, Fig. 1B) L 295, W 237; coxal field L 118, Cx-III W 160; genital field L 44, pregenital sclerite W 34, postgenital sclerite W 35; chelicera total L 134. Palp (Fig. 3D): total L 190, dL/H, L/H ratio: P-1, 16/12, 1.3; P-2, 59/31, 1.9; P-3, 19/24, 0.78; P-4, 71/17, 4.2; P-5, 25/11, 2.35; dL P-2/P-4 ratio 0.83; dL of IV-L-4-6: 74, 91, 92.

Deutonymph - Idiosoma (ventral view, Fig. 3E) L 220, W 190; coxal field L 90, Cx-III W 119; palp: total L 129, dL/H, L/H ratio: P-1, 13/9, 1.4; P-2, 34/20, 1.7; P-3, 17/17, 1.0; P-4, 48/13, 3.7; P-5, 17/7, 2.46 dL P-2/P-4 ratio 0.71.

Etymology. This species is named after the famous Puerto Rican singer Jennifer Lopez.

Remarks. Six Litarachna species have their first coxal plates fused, i.e., L. degiustii Cook, 1958 (Caribbean Sea - Cook 1958, Pešić et al. 2008), L. amnicola Cook, 1986 (Tasmania - Cook 1986, Pešić and Smit 2009), L. brasiliensis Smit, 2007 (Brazil - Smit 2007), L. caribica Pešić, Chatterjee \& Schizas, 2008 (Caribbean Sea - Pešić et al. 2008),


Figure 2. Photograph of Litarachna lopezae sp. n., male, Bajo de Sico: ventral view.
L. indica Pešić, Chatterjee \& Ingole, 2012 (West Indian coast - Pešić et al. 2012a) and L. minuta Pešić, Chatterjee \& Marshall, 2013 (Brunei Darussalam - Pešić et al. 2013).

Due to the glandularium-like structure fused with posterior margin of $\mathrm{Cx}-\mathrm{IV}, L$. lopezae sp. n. most closely resembles to $L$. minuta, a species known from a single female from Brunei Bay, but differs by a pair of small platelets with coxoglandularia 4 and associated setae lying free in the integument, not fused with Cx-IV (fused in L. minuta).

Moreover, peg-like seta at the base of P-4 ventral projection separates new species from $L$. minuta and other species with fused first coxal plates.

Habitat. The mites were collected from 69.5 m depth. The greatest depth at which pontarachnid mites have been recorded previously was reported by Pešić et al. (2012b) who found Pontarachna nemethi in a mesophotic coral ecosystem near Vieques Island of Puerto Rico at 67 m depth.

Distribution. Only known from the type locality.


Figure 3. Litarachna lopezae sp. n., Bajo de Sico ( $\mathrm{A}-\mathrm{C}=$ male, $\mathrm{D}=$ female, $\mathrm{E}=$ deutonymph ): $\mathbf{A}-\mathbf{B}, \mathbf{D}$ palp C I-L-5-6 E idiosoma, ventral view. Scale bar $=100 \mu \mathrm{~m}$.

## Litarachna sp.

Fig. 4A-D
Material examined. Puerto Rico, Rio Grande, $18^{\circ} 25^{\prime} 11.86^{\prime \prime} \mathrm{N}, 65^{\circ} 47^{\prime} 40.43^{\prime \prime} \mathrm{W}$, depth $0.5 \mathrm{~m}, 15 . \mathrm{ii} .2014$, one female, dissected and slide mounted.

Description. Female. General features - Cx-I separated medially; suture lines Cx-I/ II and Cx -III/IV complete, suture line Cx-II/III incomplete; posterior margin of CxIV with two pairs of apodemes of moderate length, the medial broad and longer than lateral ones, extending beyond posterior margin of genital field; pair of small platelets with coxoglandularia 4 and associated setae free in the integument between the posterior apodemes of Cx -IV; pre- and post-genital sclerites strongly bowed, almost touching each other, pregenital sclerite arrow-shaped (Fig. 4C); posterior to the genital field a pair of platelets with three pores and a glandularium-like structure, and three pairs of small wheel-like acetabula, with relatively few radiating spokes; excretory pore


Figure 4. Litarachna sp., female, Rio Grande: A idiosoma, ventral view B palp C genital field D I-L-5 and -6 . Scale bars $=100 \mu \mathrm{~m}$.
unsclerotized, near posterior end of idiosoma. Palp: P-2 ventral margin concave, P-5 longer than $1 / 2$ of P-4. Legs (Fig. 4D): swimming seta numbers: IV-L-4, 1; IV-L-5, 1.

Measurements - Idiosoma (ventral view, Fig. 4A) L 323, W 269; coxal field L 197, Cx-III W 219; genital field L 71, pregenital sclerite W 50, postgenital sclerite W 46; chelicera total L. Palp (Fig. 4B): total L 307, dL/H, L/H ratio: P-1, 18/19, 0.97; P-2, 87/44, 1.98; P-3, 46/34, 1-36; P-4, 100/24, 4.1; P-5, 56/14, 4.0, dL P-2/P-4 ratio 0.87; dL of I-L-2-6: 42, 48, 55, 81, 90; I-L-6 H 23, I-L-6 dL/H ratio 3.9; dL of IV-L-2-6: 49, 58, 91, 106, 108.

Remarks. The single female from Puerto Rico closely resembles Litarachna communis Walter, 1925, a species widespread in Mediterranean (Pešić et al. 2012a), but clearly differs in having more bowed, arrow-shaped pregenital sclerite. Most probably we are dealing with an undescribed species, but since male specimens were not available, a final decision cannot be made.

Habitat. The single specimen was collected from a tube of a live Sabellastarte magnifica (Shaw, 1800), a large, tubiforous shallow water polychaete of the Caribbean. The polychaete specimens were collected together with their tubes and were washed in a 0.063 mm sieve to examine the associated fauna. Our sampling strategy limits our conclusions whether the female mite was collected from inside or outside the polychaete tube.

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# Onychiurid species from Wanda Mountains in China, with descriptions of two new species (Collembola, Onychiuridae) 

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#### Abstract

A checklist of onychiurid species from the Wanda Mountains in China is presented. Eighteen species belonging to twelve genera have been found, including two new species. Bionychiurus qinglongensis sp. n. can be easily distinguished from other known species of the genus by the absence of pseudocelli on Th. I tergum and fewer number of vesicles in postantennal organ. Onychiurus heilongjiangensis $\mathbf{s p}$. n. is diagnosed by pseudocellar formulae as $32 / 133 / 33352$ dorsally and $3 / 011 / 31120$ ventrally, parapseudocellar formula as $0 / 000 / 111001+1^{\mathrm{m}}$, ratio of anal spine/unguis as 0.6 , unguiculus without inner basal lamella, and male ventral organ absent.


## Keywords

Taxonomy, new species, Bionychiurus qinglongensis sp. n., Onychiurus heilongjiangensis sp. n.

## Introduction

Heilongjiang Province is located in the northeast China, at the highest latitudes and the northernmost end of the country. It neighbors Russia across the Heilongjiang and Wusuli rivers running in its north and east respectively; in the west, it adjoins the Inner Mongolian Autonomous Region; and to its south is Jilin Prov-
ince. It covers an area of $454,000 \mathrm{~km}^{2}$, accounting for $4.7 \%$ of the nation's total. However, the fauna of Onychiuridae has been ignored in this area for many years. From 2011, we have investigated this group in Heilongjiang Province and reported several species gradually, including eleven species new to science and five new record species (Sun and Wu 2011; Sun et al. 2013a, b, c, d; Sun 2014). The Wanda Mountains are located in the eastern Heilongjiang Province of China, as a northern extension of the Changbai Mountain Range. During the investigations of the Collembola collected in this area, eighteen onychiurid species belonging to twelve genera have been found, including two new species, Bionychiurus qinglongensis sp. n. and Onychiurus heilongjiangensis sp. n. In the present work, a checklist of the onychiurid species from Wanda Mountains is presented and descriptions of the two new species are given.

## Material and methods

Eight sampling sites of Wanda Mountains have been studied, which are all planted forest with Quercus mongolica Fisch. ex Ledeb., Pinus koraiensis Sieb. et Zucc., Betula costata Trautv. or Populus davidiana Dode (Table 1, Fig. 5). Specimens were collected by Berlese extraction, cleared in lactic acid and then mounted in Marc André II solution. They were studied using a Nikon Eclipse 80i microscope. The material is deposited in the Key Laboratory of Wetland Ecology and Environment, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun.

Labial types are determined after Fjellberg (1999). Areas and chaetal nomenclature of labium follow Massoud (1967) and D'Haese (2003). Chaetae on anal valves are identified after Yoshii (1996). Chaetae on the furcal area are classified in accordance with Weiner (1996). Tibiotarsus chaetotaxy formula follows Deharveng (1983), and is expressed as: total number of chaetae (number of chaetae in row $C$, number of chaetae in row B , number of chaetae in distal rows $\mathrm{A}+\mathrm{T})$, for example $22(3,8,11)$.

## Abbreviations used in descriptions

Ant. - antennal segments, PAO - postantennal organ, Th. - thoracic segments, Abd. - abdominal segments, ms - microsensillum, pso - pseudocellus, psx - parapseudocellus, Sp - posterior $S$-chaeta on Abd. V tergum, ${ }^{m}$ - unpaired pseudopore or parapseudocellus.

The pseudocelli, parapseudocelli and pseudopores formula are the number of pseudocelli, parapseudocelli, or pseudopores by half-tergum (dorsally) or half-sternum (ventrally) as follows: head anterior, head posterior/Th . I, II, III/Abd. I, II, III, IV, V (for instance: 32/033/33343).

## Systematics

## Checklist of onychiurid species from Wanda Mountains in China (see Table 1 for details on sample sites)

Allonychiurus songi Sun \& Wu, 2012
Locality. Bawu'er, Hulin, Jiejinshan, Qiyuan, Qinglongshan, Shendingshan, Zhenbaodao.

Bionychiurus qinglongensis sp. n.
Locality. Qinglongshan.
Bionychiurus changbaiensis Sun \& Wu, 2012 Locality. Zhenbaodao.

Dimorphaphorura sanjiangensis Sun \& Wu, 2012
Locality. Bawu'er, Qinglongshan, Shendingshan.
Heteraphorura seolagensis (Lee, 1974)
Locality. Hulin, Jiejinshan, Qiyuan, Qinglongshan, Zhenbaodao.

Hymenaphorura maoerensis Sun, 2014
Locality. Jiejinshan.
Hymenaphorura wusuliensis Sun \& Wu, 2011
Locality. Wulindong.
Oligaphorura chankaensis Sun \& Wu, 2012
Locality. Shendingshan.

Oligaphorura koreana (Weiner, 1994)
Locality. Jiejinshan, Qiyuan, Zhenbaodao.

Oligaphorura ursi Fjellberg, 1984
Locality. Hulin, Shendingshan.

Onychiurus heilongjiangensis sp. n.
Locality. Qinglongshan, Jiejinshan.
Protaphorura bicampata (Gsin, 1956)
Locality. Hulin.

Protaphorura minima Sun, Zhang \& Wu, 2013
Locality. Jiejinshan, Qiyuan, Qinglongshan.

Protaphorura nutak (Yosii, 1972)
Locality. Zhenbaodao.
Psyllaphorura raoheensis Sun \& Wu, 2011
Locality. Bawu'er, Qinglongshan, Shendingshan, Wulindong, Zhenbaodao.

Sensillonychiurus changchunensis Sun \& Wu, 2012
Locality. Shendingshan.
Supraphorura furcifera (Börner, 1901) new record Locality. Zhenbaodao.

Thalassaphorura macrospinata Sun \& Wu, 2012
Locality. Hulin, Jiejinshan, Qinglongshan, Zhenbaodao.
Thalassaphorura problematica Sun, Deharveng \& Wu, 2013
Locality. Wulindong, Zhenbaodao.

## Bionychiurus qinglongensis sp. n.

http://zoobank.org/2811C6C2-4DCD-43EA-8A52-CF3210DB03F0
Figures 1-2

Type material. Holotype male, two female paratypes on slides - China, Heilongjiang, Shuangyashan, Baoqing, Qinglongshan ( $46.1504^{\circ} \mathrm{N}, 131.9591^{\circ} \mathrm{E}$ ), 14.Aug.2010, litter and soil under Betula costata Trautv., Berlese extraction, Wu Donghui et al. leg. (LD-10-344).

Diagnosis. Pso formulae as $32 / 033 / 33343$ dorsally and 11/000/01(0)010 ventrally. Subcoxae 1 of legs I, II and III with 1,1 and 1 pso respectively. Psx formula as $00 / 000 / 100001^{\mathrm{m}}$ ventrally. PAO with $14-17$ compound vesicles. Th. I-III sterna with $0+0,1+1,1+1$ chaetae respectively. Ventral tube with $1+1$ basal chaetae. Subcoxae 1 of legs I, II and III with 5, 7 and 6 chaetae respectively.

Description. Body length: females $1.30-1.37 \mathrm{~mm}$, male holotype 1.08 mm . Shape of body typical of the genus: cylindrical with strong anal spines on papillae. Color in alcohol white.

Pso formulae: 32/033/33343 dorsally and 11/000/01(0)010 ventrally (Figs 1A, B, C). Subcoxae 1 of legs I-III with 1 pso each. Parapseudocellar formulae: $00 / 000 / 100001^{\mathrm{m}}$ ventrally, dorsally psx absent (Figs 1A, B, C). Pseudopore formulae: $00 / 011 / 11110$ dorsally and $00 / 111 / 001^{\mathrm{m}} 1^{\mathrm{m}} 0$ ventrally (Figs $1 A, B, C$ ).

Head. Antennae short, 1.2 times as long as head. Length ratio of antennal segments I: II: III: IV = 1: 1.5: 1.5: 2. Subapical organite on Ant. IV with globular
Table I. List of the sampling sites of Wanda Mountains in China.

| No. | Name of the site | City | County | Coordinates | Altitude | Habitats | Collector |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| 1 | Jiejinshan | Jiamusi | Tongjiang | $47.9185^{\circ} \mathrm{N}, 132.8503^{\circ} \mathrm{E}$ | 95 m | Litter and soil under Quercus mongolica Fisch. <br> ex Ledeb. | 7 Aug. 2010 | Donghui Wu et al. |
| 2 | Qiyuan | Shuangyashan | Raohe | $46.6295^{\circ} \mathrm{N}, 133.4319^{\circ} \mathrm{E}$ | 155 m | Litter and soil under Pinus koraiensis Sieb. et <br> Zucc. | 16 Aug. 2010 | Donghui Wu et al. |
| 3 | Wulindong | Shuangyashan | Raohe | $46.5650^{\circ} \mathrm{N}, 133.6690^{\circ} \mathrm{E}$ | 207 m | Litter and soil under Pinus koraiensis Sieb. et <br> Zucc. | 18 Sept. 2011 | Haitao Wu and <br> Lihong Song |
| 4 | Zhenbaodao | Shuangyashan | Raohe | $46.4882^{\circ} \mathrm{N}, 133.8454^{\circ} \mathrm{E}$ | 75 m | Litter and soil under Pinus koraiensis Sieb. et <br> Zucc. | 16 Aug. 2010 | Donghui Wu et al. |
| 5 | Shendingshan | Shuangyashan | Raohe | $46.4760^{\circ} \mathrm{N}, 133.3031^{\circ} \mathrm{E}$ | 159 m | Litter and soil under Pinus koraiensis Sieb. et <br> Zucc. | 16 Aug. 2010 | Donghui Wu et al. |
| 6 | Qinglongshan | Shuangyashan | Baoqing | $46.1504^{\circ} \mathrm{N}, 131.9591^{\circ} \mathrm{E}$ | 259 m | Litter and soil under Betula costata Trautv. | 14 Aug. 2010 | Donghui Wu et al. |
| 7 | Bawu'er | Shuangyashan | Baoqing | $46.1370^{\circ} \mathrm{N}, 132.8580^{\circ} \mathrm{E}$ | 149 m | Litter and soil under Populus davidiana Dode | 15 Aug. 2010 | Donghui Wu et al. |
| 8 | Hulin | Jixi | Hulin | $45.7633^{\circ} \mathrm{N}, 133.0453^{\circ} \mathrm{E}$ | 91 m | Litter and soil under Quercus mongolica Fisch. <br> ex Ledeb. | 15 Aug. 2010 | Donghui Wu et al. |

apex; basolateral ms above the second proximal row of chaetae. Ant. III sensory organ consisting of 5 papillae, 5 guard chaetae, 2 small sensory rods, 2 granulated sensory clubs and a later ms (Fig. 2A). Ant. II with 15 chaetae. Ant. I with 10 chaetae. Antennal base weakly marked. PAO with $14-17$ compound vesicles arranged in two rows along axis of organ (Fig. 2B). Dorsal cephalic chaeta d0 present. $4+4$ p-chaetae between posterior a-pso on head, p1 in line with others (Fig. 1A). Mandible with strong molar plate and 4 apical teeth. Maxilla bearing 3 teeth and 6 lamellae. Maxillary palp simple with 1 basal chaeta and 2 sublobal hairs. Labral chaetal formula 4/342. Labium with 6 proximal, 4 basomedian (E, F, G and f), and 5 basolateral chaetae (b, c, d, e, e') (Fig. 1B); labial palp of AC type, labial papillae A-E with 1, 4, 0, 3, and 3 guard chaetae respectively. $5+5$ postlabial chaetae along ventral groove (Fig. 1B).

Body chaetotaxy. S-chaetae formulae as 11/022/222120 dorsally and 11/000/000100 ventrally (Figs 1A, B, C). Tiny and blunt ms, present on Th. II-III dorsal-laterally (Fig. 1A). Dorsal ordinary chaetae differentiated, Sp: m1: p1 ratio on Abd. $V=1: 0.8: 2.5$ (Fig. 1A). Th. I tergum with 9-12 chaetae. Th. II-III terga with $5+5$ and Abd. I-III terga with 3-4+3-4 chaetae on both side of axial line respectively and no unpaired axial chaetae (Fig. 1A). On each other abdominal terga from Abd. IV to Abd. VI tergum axial chaetae m0 present (Fig. 1A). Th. I-III sterna with $0+0,1+1$, $1+1$ chaetae between legs respectively.

Appendages. Subcoxae 1 of legs I, II and III with 5, 7 and 6 chaetae, subcoxae 2 with 1,5 and 5 chaetae respectively. Coxae of legs I, II and III with 4,11 and 14 chaetae, trochanters with 10 chaetae each and femora with 15,17 and 17 chaetae. Tibiotarsi of legs I, II and III with $22(3,8,11), 20(1,8,11)$ and $21(2,8,11)$ chaetae (Fig. 2C). Unguis without teeth. Unguiculus slender and pointed, 0.8-0.9 times as long as inner edge of unguis, without inner basal lamella (Fig. 2C). Ventral tube with $6-9+6-9$ distal chaetae and $1+1$ basal chaetae, without anterior chaetae (Fig. 1C). Furca reduced to cuticular fold with 4 small dental chaetae posteriorly and two manubrial rows of chaetae (Fig. 2B)

Genital plate with 16-25 chaetae in females (Fig. 2D), 26 in male. Male ventral organ absent. Anal valves with numerous acuminate chaetae; each lateral valve with a0, 2a1 and 2a2; upper valve with chaetae a0, 2a1, 2b1, 2b2, c0, 2c1 and 2c2 (Fig. 1C). Anal spines set on distinct papillae, as long as inner edge of hind unguis (Fig. 1A).

Derivatio nominis. Named for the name of mountain (Qinglongshan, and -shan means mountain in Chinese) where the species was found.

Discussion. Until now, there are four known species belonging to the genus Bionychiurus: B. changbaiensis Sun \& Wu, 2012 from China, B. normalis (Gisin, 1949) from Europe, B. oblongatus (Lee \& Park, 1986), and B. yongyeonensis (Yosii, 1966) from South Korea (Bellinger et al. 1996-2014; Sun and Wu 2012). The new species can be easily distinguished from all the above mentioned congeners by the absence of pso on Th. I tergum (present in other species) and fewer number of vesicles in PAO (more than 17 in other species).


Figure I. Bionychiurus qinglongensis sp. n. A dorsal side of body $\mathbf{B}$ ventral side of head $\mathbf{C}$ general view of Abd. I-VI sterna. Scale bars: $0.1 \mathrm{~mm}(\mathbf{A}-\mathbf{C})$.


Figure 2. Bionychiurus qinglongensis sp. n. A Ant. III sensory organ B PAO C Abd. IV sternum D distal part of leg III E female genital plate. Scale bars: $0.1 \mathrm{~mm}(\mathbf{C}-\mathbf{E}), 0.01 \mathrm{~mm}(\mathbf{A}-\mathbf{B})$.

## Onychiurus heilongjiangensis sp. n.

http://zoobank.org/540034F0-0A0F-4599-9E38-91BDC042C420
Figures 3-4
Type material. Holotype male, 7 female and 6 male paratypes on slides - China, Heilongjiang, Shuangyashan, Baoqing, Qinglongshan ( $46.1504^{\circ} \mathrm{N}, 131.9591^{\circ} \mathrm{E}$ ), 14.Aug.2010, litter and soil under Betula costata Trautv., Berlese extraction, Wu Donghui et al. leg. (LD-10-444, LD-10-445); 12 female and 3 male paratypes on slides - China, Heilongjiang, Jiamusi, Tongjiang, Jiejinshan ( $47.9185^{\circ} \mathrm{N}, 132.8503^{\circ} \mathrm{E}$ ), 7.Aug.2010, litter and soil under Quercus mongolica Fisch. ex Ledeb., Berlese extraction, Wu Donghui et al. leg. (LD-10-484, LD-10-485, LD-10-486, LD-10-488).

Diagnosis. Pso formulae as $32 / 133 / 33352$ dorsally and $3 / 011 / 31120$ ventrally. Psx formula as $0 / 000 / 111001+1^{\mathrm{m}}$ ventrally.. Ratio of AS/unguis as 0.6 . Unguiculus without inner basal lamella. Male ventral organ absent.

Description. Body length: females $1.6-1.8 \mathrm{~mm}$, males $1.4-1.6 \mathrm{~mm}$; holotype 1.6 mm . Shape of body typical of the genus: cylindrical with anal spines on papillae, Abd. III-IV more or less broadened. Color in alcohol white.


Figure 3. Onychiurus heilongjiangensis sp. n. A dorsal side of body B PAO C Ant. I-IV D Ant. III sensory organ $\mathbf{E}$ antennal tip $\mathbf{F}$ labium $\mathbf{G}$ distal part of leg III $\mathbf{H}$ Abd. II-VI sterna. Scale bars: 0.1 mm (A, C, G-H), $0.01 \mathrm{~mm}(\mathbf{B}, \mathbf{D}-\mathbf{F})$.


Figure 4. Onychiurus heilongjiangensis sp. n. A ventral side of head $\mathbf{B}$ anal valves $\mathbf{C}$ Abd. IV sternum. Scale bars: $0.1 \mathrm{~mm}(\mathbf{A}-\mathbf{C})$.

Pso formulae: 32/133/33352 dorsally and 3/011/31120 ventrally (Figs 3A, H, 4A). Subcoxae 1 of legs I-III with 2 pso each. Parapseudocellar formulae: $0 / 000 / 111001+1^{\mathrm{m}}$ ventrally, dorsally psx absent (Figs 3A, H, 4A). Pseudopore formulae as 00/011/11110 dorsally and $00 / 111 / 0001^{\mathrm{m}} 00$ ventrally (Figs 3A, H, 4A).

Head. Antennae short, as long as head. Length ratio of antennal segments I: II: III: IV = 1: 1.5: 1.5: 2.0. Subapical organite on Ant. IV with globular apex (Fig. 3C); invaginated apical bulb present (Fig. 3E); basolateral ms above the first proximal row of chaetae (Fig. 3C). Ant. III sensory organ composed of 5 papillae, 5 guard chaetae, 2 small rods, and 2 smooth sensory clubs (Fig. 3D); lateral ms just behind sensory organ (Fig. 3C). Ant. II with 14 chaetae. Ant. I with 8 chaetae. Antennal base well marked. PAO with 14-16 compound vesicles arranged in two rows along axis of organ (Fig. 3B). Dorsal cephalic chaeta d0 present. 3+3 p-chaetae between two inner posterior pso


Figure 5. Sampling localities in Wanda Mountains.
on head, pl anterior to others (Fig. 3A). Mandible with strong molar plate and 4 apical teeth. Maxilla bearing 3 teeth and 6 lamellae. Maxillary palp simple with 1 basal chaeta and 2 sublobal hairs. Labral chaetae $4 / 142$. Labium with 6 proximal, 4 basomedian ( E , F, G, and f) and 5 basolateral (b, c, d, e, e') chaetae (Fig. 4A). Labial palp of AB type, labial papillae A-E with 1, 4, 0, 3 and 3 guard chaetae respectively (Fig. 3F). Postlabial chaetae $4-5+4-5$ along ventral groove (Fig. 4A).

Body chaetotaxy. S-chaetae not distinguishable from ordinary chaetae. Tiny and blunt ms, present on Th. II and III dorso-laterally (Fig. 3A). Dorsal ordinary chaetae poorly differentiated, usually coarse and short. Th. I tergum with 8-14+8-14 chaetae. Th. II-Abd. III terga with $4-5+4-5$ chaetae along axial line respectively, usually with
asymmetrical chaetae (Fig. 3A). Abd. IV tergum with p0 chaeta; Abd. V tergum without axial chaeta; Abd. VI tergum with m0 chaeta (Fig. 3A). Th. I-III sterna without chaetae between legs.

Appendages. Subcoxae 1 of legs I-III with 5, 5 and 5 chaetae, subcoxae 2 with 1 , 4 and 4 chaetae respectively. Coxae of legs I, II and III with 3,9-11 and 9-12 chaetae, trochanters with 9 chaetae each and femora with 15,16 and 16 chaetae. Tibiotarsi of legs I, II and III with $17(9,7,1), 18(9,7,2)$ and $17(9,7,1)$ chaetae, M-chaeta absent. Unguis without teeth. Unguiculus slender and pointed, 0.8 times as long as inner edge of unguis, without inner basal lamella (Fig. 3G). Ventral tube with 6-8+6-8 distal chaetae, without anterior or basal chaetae. Furca reduced to finely granulated area, with 4 small dental chaetae in one row posteriorly; three manubrial rows of chaetae present (Figs 3H, 4C).

Genital plate with 17-23 chaetae in females, 40-63 chaetae in males. Male ventral organ absent. Anal valves with numerous acuminate chaetae; each lateral valve with chaetae a 0 and 2 a 1 ; upper valve with chaetae a0, 2b1, 2b2, c0, 2c1 and 2c2 (Fig. 4B). Anal spines set on distinct papillae, 0.6 times as long as inner edge of hind unguis (Fig. 3A).

Derivatio nominis. Named for the province of the type locality.
Discussion. The new specie shares the same dorsal pso formula from head to Abd. IV (32/133/3335), ventral pso formula from head to Abd. I $(3 / 011 / 3)$ and number of pso on subcoxae 1 of legs I-III (2, 2, 2 respectively) with a number of the known European species, i.e. O. ambulans (Linnaeus, 1758) sensu Pomorski, 1998, O. arans Gisin, 1952, O. circulans Gisin, 1952, O. insinuans Gisin, 1952, O. subcirculans Gisin, 1962, and O. sublegans Gisin, 1960, but it can be distinguished easily from all these species as having only 2 pso on each side of Abd. V (3 or 4 in other species) and no male ventral organ (present in other species).

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# Two more new species of Aphidura (Hemiptera, Aphididae), and a note on variation in Aphidura bozhkoae Narzikulov 

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#### Abstract

Two new species of Aphidura Hille Ris Lambers, 1956 (Hemiptera, Aphididae, Macrosiphini) are described; Aphidura libanensis sp. n. from Prunus prostrata in Lebanon, and Aphidura corsicensis sp. n. from Cerastium soleirolii in Corsica (France). Studies of Aphidura bozhkoae specimens from different localities have revealed that this species varies in its pattern of dorsal sclerotisation and other morphological characters, within and between populations. An updated key for identifying the world's species of Aphidura is presented.


## Keywords

Aphidura, new taxa, descriptions, intraspecific variation, key of species

## Introduction

Aphidura Hille Ris Lambers, 1956 (Hemiptera, Aphididae, Macrosiphini) currently includes 22-25 subjective valid species, exhibiting a Mediterranean-Pontian-Turanian distribution with extensions to neighbouring areas and exceptionally (one species) to the Russian Far East. Many of the species are associated with Caryophyllaceae, and some feed on Prunus and related genera, indicating that there may be host alternation
between Amygdaloideae (Rosaceae) and Caryophyllaceae, but their biology and life cycles are mostly unknown (Blackman and Eastop 2014). Twelve of the species were described in 2013 (Kadyrbekov 2013; Nieto Nafría et al. 2013), adding greatly to the knowledge of the genus. Two more species were present among specimens conserved in the collection of the Natural History Museum in London (BMNH), and they are described in this paper.

Aphidura bozhkoae Narzikulov, 1958 was described from Prunus (= Cerasus) verrucosa in Tajikistan, and has subsequently been found on various species of Prunus in the neighbouring countries Kyrgyzstan and Uzbekistan, and also in Kazakhstan, Iran and Georgia (Nieto Nafría et al. 2013). The variation in several morphological features exhibited by this species, both within and between populations, is remarkable. In particular, Iranian specimens (illustrated by Nieto Nafría et al. 2013: 10, fig 3A) differ from the Tajik type series (illustrated by Narzikulov 1958: 16, fig. 1) to such an extent that D. Hille Ris Lambers gave them the specific epithet "nitens", which was however never published. The variable characters include those that have been used to discriminate between other species of Aphidura, some of which are described from only single collections, so for future work on this genus it is important that the variation in $A$. bozhkoae should be reviewed and discussed.

## Material and methods

Specimens of the following five samples have been studied:
(1) 9 apterous viviparous females and 1 alate viviparous female: TAJIKISTAN: Cerasus verrucosa, Iskandirkul, 14-VII-1959 (3 apterae, 1 alate); same host-plant, Ziddy [Siddi in slide label by Hille Ris Lambers], 28-VI-1954 (6 apterae), N. Narzikulov leg.; Aphidura bozhkoae, Narzikulov det.; BMNH collection. Both Tajik localities are type-localities of this species (Narzikulov 1958: page 20).
(2) 54 apterous viviparous females and 13 alate viviparous females: TAJIKISTAN: Cerasus verrucosa, Varzov ( 1 km above Kondara), 24-V-1981; Cerasus erythrocarpa, Ziddy, 9-VI-1981, G. Kh. Shaposhnikov leg. (samples 7524 and 7551); Aphidura bozhkoae, G. Kh. Shaposhnikov det.; Zoologicheskiy Institut collection. Ziddy is a type-locality of this species and Varzov is near Kondara, which is also a typelocality. These specimens were dry (alcohol had completely evaporated); they were hydrated and later mounted on microscopic slides.
(3) 115 apterous viviparous females and 63 alatae viviparous females: IRAN: several host plants, localities and dates (data in Nieto Nafría et al. 2013: table I); G. Remaudière leg. (samples i196, i2961, i3028, i4092 and i4347); Aphidura "nitens", manuscript name, D. Hille Ris Lambers or G. Remaudière det.; Aphidura bozhkoae, J. M. Nieto Nafría det.. Muséum national d'Histoire naturelle and BMNH collections.
(4) 5 apterous females: LEBANON, Prunus prostrata, Jabal el Barouk, 22-V-1973, D. Hille Ris Lambers leg. (sample 754); Aphidura "nitens" manuscript name, D. Hille Ris Lambers det.; BMNH collection.
(5) 3 apterous viviparous females: CORSICA (France), Cerastium soleirolii, North slopes of Mount Cinto, 08-VII-1980, J. H. Martin leg. (sample number 3061); Aphidura "cerastii", manuscript name, D. Hille-Ris-Lambers det.; BMNH collection.

Morphological measurements were made according to Nieto Nafría and Mier Durante (1998). Measurements in text and table are lengths except when indicated otherwise as width or diameter. Tables have a distribution of data like tables in Nieto Nafría et al. (2013) in order to facilitate the comparisons.

A Leica DC digital 96 camera with IM 1000 version 1.10 software was used for the photomicrographs, which have been taken and mounted by L. M. Fernández Blanco.

## Results and discussions

## Descriptions of new species

## Aphidura libanensis sp. n.

http://zoobank.org/ECE4A97D-D03F-45CA-BEB5-57C64853ED30

Diagnosis. Siphunculi tapering to apex or slightly swollen in a distal portion. Dorsum of metathorax to abdominal segment 6 with a discal plate (apterae). Mesosternal mammariform processes pale, flat and with spinules (apterae). Abdominal marginal tubercles usually present. Tarsal formula 3.3.3. (sometimes one tarsus with 4 setae)

Apterous viviparous female, from 5 specimens (Figs 1A-E). Unknown colour in life, possibly shiny black with antennae, legs and siphunculi pale brown. In mounted specimens, antennae, mesosternal mammariform processes, legs, siphunculi, abdominal segment 8 sclerotized band, cauda and genital and anal plates are yellowish brown, with very proximal and distal portions of siphunculi, antennal segment VI and very apical portion of antennal segment V somewhat darker than aforementioned structures, all in contrast with head and dorsum of thorax and most part of abdomen, which are very dark brown. Frons undulated. Head with rugosity lines and spinules (both more abundant on dorsum). Antennal cuticle imbricated. Setae on body dorsum, antennae, and most of those on legs thick, with apices blunt or slightly capitate. Mesosternal mammariform processes pale, flat and with spinules. A discal plate present (from metathorax to abdominal segment 6); prothorax, mesothorax and abdominal segment 7 with wide transverse bands, which are darker than the discal plate, abdominal segment 8 with a band paler than the others. Marginal tubercles usually present on both sides of prothorax and several abdominal segments; they are small, but the abdominal ones are relatively tall. Siphunculi tapering to apex or slightly swollen in the distal


Figure I. Aphidura libanensis sp. n., apterous viviparous females. A Habitus B Part of prothorax with a marginal tubercle (signalled with an arrow) C Marginal zone of abdominal segments 3 and 4, with marginal tubercles (signalled with arrows) D Mesosternum with mammariform processes (signalled with arrows) E Mesosternum with mammariform processes (signalled with arrows) of another specimen.
quarter, spinulose imbrication, distinct preapical incision and flange. Cauda broadly triangular. Metric and meristic features in Table 1.

Types. Holotype: apterous viviparous female, collected on Prunus prostrata Labill. (Rosaceae), Jabal el Barouk (Lebanon), 2-V-1973, Hille Ris Lambers leg.; paratypes: four apterous females collected at the same time as the holotype.

Etymology. The specific name of the new species is an adjective that means inhabitant of Lebanon, in feminine.

Table I. Metric and meristic features of apterous viviparous females of A. libanensis $\mathrm{sp} . \mathrm{n}$. and A. corsicensis $\mathrm{sp} . \mathrm{n}$.

|  | A. libanensis | A. corsicensis |
| :--- | :---: | :---: |
|  | $\mathbf{n}=\mathbf{5}$ | $\mathbf{n}=\mathbf{3}$ |
| Body [mm] | $1.805-1.938$ | $2.025-2.088$ |
| Antenna [mm] | $1.605-1.698$ | $1.590-1.655$ |
| Antenna / Body [times] | $0.87-0.93$ | $0.79-081$ |
| Ant. segm. III [mm] | $0.43-0.51$ | $0.48-0.52$ |
| Ant. segm. IV [mm] | $0.27-0.31$ | $0.27-0.29$ |
| Ant. segm. V [mm] | $0.20-0.24$ | $0.22-0.24$ |
| Ant. segm. VI base [mm] | $0.09-0.13$ | $0.11-0.12$ |
| Ant. segm. VI processus terminalis | $0.39-0.46$ | $0.31-0.34$ |
| Ant. segm. VI processus terminalis / Ant. segm. III [times] | $0.78-1.03$ | $0.60-0.67$ |
| Ant. segm. VI processus terminalis / base [times] | $3.56-4.89$ | $2.63-3.00$ |
| Ultimate rostral segm. [mm] | $0.12-0.13$ | $0.15-0.16$ |
| Ultimate rostral segm. / its basal width [times] | $2.00-2.27$ | $2.58-2.73$ |
| Ultimate rostral segm. / Ant. segm. VI base [times] | $0.96-1.39$ | $1.25-1.36$ |
| Hind femur [mm] | $0.52-0.56$ | $0.61-0.63$ |
| Hind tibia [mm] | $0.96-1.03$ | $1.06-1.09$ |
| Hind tibia / Body [times] | $0.52-0.56$ | $0.52-0.53$ |
| Hind tarsus, 2nd segm. [mm] | $0.11-0.12$ | $0.10-0.11$ |
| Hind tarsus, 2nd segm. / Ultimate rostral segm. [times] | $0.84-1.00$ | $0.65-0.75$ |
| Abdominal (segm. 2-6) marginal tubercles [number] | $0-3$ | 0 |
| Siphunculus [mm] | $0.30-0.35$ | $0.42-0.45$ |
| Siphunculus / Body [times] | $0.17-0.18$ | $0.20-0.22$ |
| Siphunculus / Ant. segm. III [times] | $0.65-0.74$ | $0.82-0.89$ |
| Siphunculus / its basal width [times] | $3.50-4.27$ | $4.83-5.60$ |
| Siphuncular width, maximal / basal [times] | $0.56-0.70$ | $0.61-0.82$ |
| Siphuncular width, maximal / minimal [times] | $1.05-1.11$ | $1.04-1.10$ |
| Siphuncular minimal width / Hind tibiae, diameter at middle [times] | $1.13-1.80$ | $1.00-1.18$ |
| Cauda [mm] | $0.13-0.16$ | $0.23-0.24$ |
| Cauda / siphunculus [times] | $0.40-0.49$ | $0.51-0.57$ |
| Cauda / its basal width [times] | $0.87-0.97$ | $1.41-1.50$ |
| sin |  |  |
|  |  |  |

Setae on ...

| ... Frons [ $\mu \mathrm{m}$ ] | 25-35 | 32-37 |
| :---: | :---: | :---: |
| ... Frons / b. d. Ant. segm. III [times] | 1.1-1.6 | 1.3-1.5 |
| ... Vertex [ $\mu \mathrm{m}$ ] | 15-30 | 32-35 |
| ... Vertex / b. d. Ant. segm. III [times] | 0.7-1.4 | 1.2-1.4 |
| ... Antennal segment III [ $\mu \mathrm{m}$ ] | 12-15 | 10-14 |
| ... Antennal segment III / b. d. Ant. segm. III [times] | 0.6-0.7 | 0.4-0.6 |
| ... Ultimate rostral segm, [number] | 6-9 | 17-19 |
| ... Hind femur, dorsal [ $\mu \mathrm{m}$ ] | 12-18 | 17-25 |
| ... Hind femur, ventral [ $\mu \mathrm{m}$ ] | 22-28 | 25-38 |
| $\ldots$... Hind tibia, dorsal, at middle [ $\mu \mathrm{m}$ ] | 22-33 | 27-33 |
| ... Hind tibia, dorsal / Tibial diameter (at middle) [times] | 0.5-0.8 | 0.5-0.7 |
| ... Hind tarsus, first segm. [number] | 3 (4) | 3 |


|  | A. libanensis | A. corsicensis |
| :--- | :---: | :---: |
|  | $\mathbf{n}=\mathbf{5}$ | $\mathbf{n = 3}$ |
| $\ldots$ Abdominal segm. 2-4 [ $\mu \mathrm{m}$ ] | $10-18$ | $25-30$ |
| ... Abdominal segm. 2-4 / b. d. Ant. segm. III [times] | $0.5-0.8$ | $1.0-1.1$ |
| .. Abdominal segm. 8 [ $\mu \mathrm{m}]$ | $27-33$ | $32-38$ |
| .. Abdominal segm. 8 / b. d. Ant. segm. III [times] | $1.2-1.5$ | $1.3-1.4$ |
| .. Abdominal segm. 8 [number] | 4 | $3-4$ |
| .. Genital plate, discal [number] | 2 | $5-7$ |
| ... Genital plate, marginal [number] | $10-15$ | $13-20$ |
| ... Cauda [number] | $6-8$ | $6-9$ |

Note: Used abbreviations: Ant., Antennal; b. d., basal diameter; n. number of measured specimens; segm., segment or segments.

Discussion. D. Hille Ris Lambers thought that these Lebanese specimens were conspecific with others found on Prunus in Iran, which were being studied by him and G. Remaudière, and that they all belonged to an undescribed species, which was named in draft "nitens" (Remaudière's epistolary archive). Certainly these Lebanese aphids do not belong to any previously described species, but they are also not conspecific with the Iranian ones, which we believe to be $A$. bozhkoae (see next section).

Aphidura libanensis sp. n. resembles Iranian specimens of $A$. bozhkoae and the East Asian species $A$. mordvilkoi in the shape of the siphunculi and the extensive and solid discal plate, but there is an important difference in the number of first tarsal setae: four setae on each first tarsal segment in A. bozhkoae and $A$. mordvilkoi, and three in A. libanensis. A. libanensis and $A$. mordvilkoi can also be distinguished from one another by the number of accessory setae on the ultimate rostral segment (6-9 in A. libanensis, 2-4 in A. mordvilkoi) and the relative length of the processus terminalis (3.6-4.9 times base of antennal segment VI in A. libanensis, 2.2-2.7 times in A. mordvilkoi). A. libanensis and A. bozhkoae are very similar in absolute and relative lengths of most body parts, the setae of $A$. libanensis (Table 1) are all generally longer than those of $A$. bozhkoae (Table 2).

## Aphidura corsicensis sp. n.

http://zoobank.org/EA353AA4-B640-4294-A9B0-8A7C58C546CB

Diagnosis. Siphunculi slightly swollen. Dorsum of thorax and abdomen with setiferous sclerites (apterae). Mesosternal mammariform processes pale, flat and smooth (apterae). Abdominal marginal tubercles absent. Tarsal formula 3.3.3

Apterous viviparous female, from 3 specimens (Fig. 2). Shiny dark green in life. In mounted specimens, head, antennae, legs, siphunculi, cauda, genital and anal plates, and thoracic and abdominal sclerites yellowish brown, with apex of antennal segments III, IV and V, antennal segment VI, very apex of tibiae, tarsi and apex of siphunculi something darker than aforementioned structures. Frons undulated. Head mostly smooth, some rugosity lines are present on dorsum. Proximal section of anten-


Figure 2. Aphidura corsicensis sp. n., apterous viviparous female. A Habitus B Mesosternum with mammariform processes (signalled with arrows).
nal segment III and segments IV-VI imbricated. Setae on body dorsum, antennae and most of those on legs thick with apices blunt or slightly capitate. Mesosternal mammariform processes pale, flat and smooth. Setiferous sclerites present on dorsum of thorax and abdomen, mostly with one setae and similar in size to spiracular sclerites; marginal sclerites on abdominal segments 2-4 often coalescent; those on segment 6 forming postsiphuncular sclerites; and those on segment 7 and 8 partially coalescent;

Table 2. Metric and meristic features of apterous viviparous females of $A$. bozhkoae from Tajikistan, collected by Narzikulov (column Tajik. [Na.]) and by Shaposhnikov (column Tajik. [Sh.], and from Iran, collected by Remaudière. In parenthesis exceptional data.

|  | Tajik. [Na.] | Tajik. [Sh.] | Iran [Re.] |
| :---: | :---: | :---: | :---: |
|  | $\mathrm{n}=7$ | $\mathrm{n}=13$ | $\mathrm{n}=31$ |
| Body [mm] | 1.950-2.125 | 1.925-2.250 | 1.413-2.125 |
| Antenna [mm] | 1.408-1.748 | 1.475-1.785 | 1.290-1.765 |
| Antenna / Body [times] | 0.70-0.83 | 0.70-0.88 | 0.73-0.92(1.07) |
| Ant. segm. III [mm] | 0.36-0.49 | 0.40-0.48 | 0.35-0.52 |
| Ant. segm. IV [mm] | 0.22-0.28 | 0.22-0.28 | 0.19-0.34 |
| Ant. segm. V [mm] | 0.19-0.22 | 0.18-0.24 | 0.18-0.25 |
| Ant. segm. VI base [mm] | 0.10-0.12 | 0.10-0.12 | 0.09-0.12 |
| Ant. segm. VI processus terminalis | 0.39-0.52 | 0.40-0.55 | 0.33-0.48 |
| Ant. segm. VI processus terminalis / Ant. segm. III [times] | 0.98-1.10 | 0.98-1.18 | (0.72)0.85-1.15 |
| Ant. segm. VI processus terminalis / base [times] | 3.58-4.76 | 3.86-5.00 | 2.89-4.55 |
| Ultimate rostral segm. [mm] | 0.12-0.13 | 0.13-0.15 | 0.11-0.14 |
| Ultimate rostral segm. / its basal width [times] | 1.56-2.36 | 1.92-2.64 | 1.92-2.89 |
| Hind tibia / Body [times] | 0.44-0.52 | 0.42-0.55 | 0.47-0.62 |
| Hind tarsus, 2nd segm. [mm] | 0.12-0.14 | 0.12-0.14 | 0.10-0.13 |
| Hind tarsus, 2nd segm. / Ultimate rostral segm. [times] | 0.92-1.04 | 0.89-1.00 | 0.77-1.00 |
| Abdominal marginal (segm. 2-6) tubercles [number] | 0 | 0 | 0-6 |
| Abdominal (segm. 8) spinal tubercles [number] | 0 | 0 | $0(2-3)$ |
| Siphunculus [mm] | 0.35-0.48 | 0.36-0.45 | 0.28-0.44 |
| Siphunculus / Body [times] | 0.17-0.22 | 0.18-0.22 | 0.17-0.25 |
| Siphunculus / Antennal segm. III [times] | 0.90-1.04 | 0.85-1.10 | 0.69-0.92 |
| Siphunculus / its basal width [times] | 3.68-5.67 | 3.60-5.25 | 3.42-6.15 |
| Siphuncular width, maximal / basal [times] | 0.60-0.73 | 0.50-0.75 | 0.53-0.85 |
| Siphuncular width, maximal / minimal [times] | 1.00-1.04 | 1.00-1.10 | 1.00-1.11 |
| Siphuncular minimal width / Hind tibiae, diameter at middle [times] | 1.14-1.50 | 1.10-1.60 | 1.06-1.64(2.00) |
| Cauda [mm] | 0.17-0.18 | 0.17-0.22 | 0.12-0.18 |
| Cauda / siphunculus [times] | 0.37-0.51 | 0.39-0.51 | 0.35-0.48 |
| Cauda / its basal width [times] | 1.08-1.21 | 1.05-1.33 | 0.87-1.33 |
| Setae on ... |  |  |  |
| ... Frons [ $\mu \mathrm{m}$ ] | 10-14 | 7-20 | 8-13 |
| ... Frons / b. d. Ant. segm. III [times] | 0.3-0.5 | 0.3-1.0 | 0.3-0.6 |
| ... Vertex [ $\mu \mathrm{m}$ ] | 10-15 | 10-22 | 8-10 |
| ... Vertex / b. d. Ant. segm. III [times] | 0.5-0.6 | 0.4-1.0 | 0.3-0.6 |
| ... Antennal segment III [ $\mu \mathrm{m}$ ] | 7-10 | 7-10 | 6-10 |
| ... Antennal segment III / b. d. Ant. segm. III [times] | 0.3-0.4 | 0.3-0.4 | 0.3-0.5 |
| ... Ultimate rostral segm, [number] | 9-13 | 7-18 | 6-11 |
| $\ldots$... Hind femur, dorsal [ $\mu \mathrm{m}$ ] | 10-13 | 9-18(28) | 8-13 |
| ... Hind femur, ventral [ $\mu \mathrm{m}$ ] | 12-20 | 12-20(35) | 10-23 |
| $\ldots$... Hind tibia, dorsal, at middle [ $\mu \mathrm{m}$ ] | 15-24 | 17-30 | 10-23 |
| ... Hind tibia, dorsal / Tibial diameter (at middle) [times] | 0.4-0.6 | 0.4-0.8 | $0.2-0.8$ |
| ... Hind tarsus, first segm. [number] | 4 | 4 | 4 |
| ... Abdominal segm. 2-4 [ $\mu \mathrm{m}$ ] | 9-13 | 10-15 | (5)8-10 |
| ... Abdominal segm. 2-4 / b. d. Ant. segm. III [times] | 0.3-0.5 | 0.4-0.7 | $0.2-0.5$ |


|  | Tajik. [Na.] | Tajik. [Sh.] | Iran [Re.] |
| :--- | :---: | :---: | :---: |
|  | $\mathbf{n = 7}$ | $\mathbf{n = 1 3}$ | $\mathbf{n = 3 1}$ |
| .. Abdominal segm. $8[\mu \mathrm{~m}]$ | $10-15$ | $12-20(25)$ | $8-15$ |
| .. Abdominal segm. 8 / b. d. Ant. segm. III [times] | $0.4-0.6$ | $0.6-0.9(1.1)$ | $0.3-0.6$ |
| .. Abdominal segm. 8 [number] | $2-4$ | $(2) 4(5)$ | 4 |
| .. Genital plate, discal [number] | $2(4)$ | $2(3)-$ | 2 |
| .. Genital plate, marginal [number] | $12-20$ | $12-24$ | $12-19$ |
| .. Cauda [number] | $8-12$ | $7-10$ | $6-10$ |

Note: Used abbreviations: Ant., Antennal; b. d., basal diameter; n. number of measured specimens; segm., segment or segments.
sclerites on abdominal segments 6-8 with spinules. Intersegmental sclerites small. Siphunculi slightly swollen (maximal width 1.04-1.10 times minimal width of the stem), with small scales and distinct preapical incision and flange. Cauda tongue shaped. Metric and meristic features in Table 1.

Types. Holotype: apterous viviparous female, collected on Cerastium soleirolii Ser. ex Duby. (Caryophyllaceae), North slope of Mount Cinto, (Corsica, France), noted as more than 1100m, 08-VII-1980, J. H. Martin leg.. Paratypes: two apterous females from the same colony as the holotype.

Etymology. The specific name of the new species is an adjective that means inhabitant of Corsica, in feminine.

Discussion. This species was included as Aphidura sp. in the host lists and key to aphids on Cerastium by Blackman and Eastop (2006), but has not previously been formally described.

Aphidura corsicensis can be easily distinguished from other Aphidura species by the abundant dorsal setiferous sclerites, which are not present in any other species of the genus. The host plant is not normally found below 1900m (Arthur Chater, Kew Gardens, pers. comm.) but the plant hosting $A$. corsicensis was estimated to have been collected at around 1100-1200m (Martin).

## The variability of Aphidura bozhkoae Narzikulov

Narzikulov (1958) described Aphidura bozhkoae as a shiny black aphid with black siphunculi feeding on Cerasus verrucosa (currently in Prunus) in Tajikistan. His illustration (Fig. 1 on p.16) depicts an aptera with extensive dorsal sclerotisation, but the metanotal sclerite is separate from the abdominal discal plate and has a mesial gap. In specimens from the type series in the BMNH collection this gap is quite wide and coincides with an indentation of the anterior part of the abdominal discal plate, to form a conspicuous irregularly-shaped window. All these specimens have siphunculi as dark as or darker than the discal plate. In contrast, specimens from Iran in the BMNH collection all have the metanotal sclerite forming a broad band fused with the abdominal discal plate. so that the sclerotisation extends as a solid shield from metonotum to abdominal tergite 6 .


Figure 3. Aphidura bozhkoae apterous viviparous females from Ziddy (Tajikistan). A-C habitus, anterior part D-E Mesosternum with mammariform processes. A specimen collected in 1954 by Narzikulov B-E Two specimens collected in 1981 by Shaposhnikov. Note the different dorsal pattern and the different size and form of processes.

Furthermore, these apterae have pale siphunculi. Another difference between Tajik and Iranian specimens in the BMNH collection concerns the mesosternal processes, which are well developed and well separated from each other in Tajik apterae, but much lower and more rounded in the specimens from Iran. These last features, all of them, are also shown by the specimen in Fig. 2A of Nieto Nafría et al. 2013).

In addition, the Tajik specimens all lack marginal or spinal tubercles, whereas in specimens from Iran small marginal tubercles may be present on some of abdominal tergites 2-6, and there may also be spinal tubercles on abdominal tergite 8 (Table 1).

When morphometric data are compared, however, the ranges of measurements and ratios for all characters agree well between countries, any slight differences being


Figure 4. Aphidura bozhkoae mesosternal processes of five apterous viviparous females collected in Ziddy (Tajikistan) in 1981 by G. Shaposhnikov, same sample as the two specimens in Figure 3.
within the expected range of intraspecific variation (Table 2). Furthermore, examination of more Tajik material from the Zoologicheskiy Institut collection (Figs 3, 4), and of more apterae from Iran in the collection of the Muséum national d'Histoire naturelle, showed that the BMNH specimens constituted the extremes of a range of variation in a single species, with Iranian apterae in particular varying in the dorsal pattern of sclerotisation, pigmentation of siphunculi and shape of mesosternal processes. Further confirmation came from photomicrographs of A. bozbkoae from Prunus incana in Georgia kindly provided by S. Bardjadze, which showed a similar range of variation in these characters within a single sample.

Intraspecific variation in the extent of dorsal sclerotisation is evident in other species of Aphidura, especially in those that are widely collected such as A. picta. However this usually involves varying degrees of fragmentation or reduction of the entire discal plate rather than any specific part of it; in this respect the variation shown by A. bozhkoae is unusual. Siphuncular pigmentation and the extent of development of
the mesosternal processes are characters that have been used to discriminate between Aphidura species, but experience with $A$. bozhkoae shows that they need to be applied carefully, especially when describing new species from small samples.

## Key to apterous viviparous females of Aphidura species of the world (updated from the key of Nieto Nafría et al. 2013).

In brackets are: (1) when necessary morphological characters are included that do not have correspondence in the other proposition of the disjunctive, but which are useful to confirm identification; (2) illustration reference, from literature or in this paper; (3) host plants; and (4) distribution, with countries listed in geographical order from West to East.
$1 \quad$ Siphunculi markedly swollen (maximal swollen width at least 1.2 times minimal stem width) 2

- $\quad$ Siphunculi of different form (cylindrical, subcylindrical, tapering or slightly swollen, see Nieto Nafría et al. (2013): "generic characters" section) .9
2 Most of dorsal setae placed on conical tubercles. [Dorsum without segmental pigmented sclerotisation; Nieto Nafría et al. (2013): fig. 6. On Acanthophyllum sp. Iran] A. acanthophylli
- Dorsal setae not placed on tubercles............................................................ 3

3 Mesosternal processes and cauda pale4

- Mesosternal processes and cauda more or less pigmented, light brown to brown 6

4 Siphunculi dark brown, 2.3-2.7 times cauda which has 7-11 setae. Abdominal dorsum with spino-pleural patch, postsiphuncular sclerites pigmented and marginal sclerites. [Ultimate rostral segment 1.0-1.2 times second segment of hind tarsi; cauda 1.1-1.2 times its basal width; Kadyrbekov (2013): fig. 8. On Silene suffrutescens and $S$. sp. Kazakhstan] A. nomadica

- Siphunculi pale, sometimes with smoky apex, 1.6-2.2 times cauda, which has $6-7$ setae. If a spino-pleural patch present then ultimate rostral segment is 1.2-1.5 times second segment of hind tarsi .5

5 Antennal segment VI processus terminalis at least 1.4 times antennal segment III and approximately 4 times antennal segment VI base. Longest dorsal setae on abdominal segment 2-4 approximately $3 \mu \mathrm{~m}$. Cauda tongue-shaped. Dorsum pale with dark intersegmental sclerites. [Nieto Nafría et al. (2013): fig. 5C. On Gypsophila sp. Grece] A. graeca

- Antennal segment VI processus terminalis at most 1.1 times antennal segment III and at most 3.1 times antennal segment VI base. Longest dorsal setae on abdominal segment $2-4$ are $7-13 \mu \mathrm{~m}$. Cauda triangular, sometimes slight constricted. Dorsum with variable sclerotisation and pigmentation,
sometimes mostly pale. [Nieto Nafría et al. (2013): fig. 5D. On Silene sp., and an unidentified caryophyllaceous species. Iran] ........ A. amphorosiphon
- Dorsum of thorax and abdomen never with setiferous sclerites; other sclerotization can be present

12 Siphunculus slightly swollen with a maximal width close to 1.2 times minimal stem width and 1.6-2.0 times cauda, which is $1.5-1.8$ times its basal width and has $7-11$ setae; both as dark as head dorsum and thoracic and abdominal sclerotisation (a discal plate can be present). Longest dorsal setae on abdominal segment $2-4$ are $10-11 \mu \mathrm{~m}$ and approximately 0.5 times basal diameter of antennal segment III. [Kadyrbekov (2013): fig. 6. On Melandrium album. Kazakhstan] A. melandrii - Characters not in above combination......................................................... 13 13 Siphunculus at most 1.95 times cauda (which is short triangular), pale or uniformly dusky and slight swollen. Dorsum of head and mesosternal processes pale. Segmental thoracic and abdominal sclerotisation and pigmentation absent. 14

- $\quad$ Siphunculus at least 1.90 times cauda, both diversely shaped and coloured. Dorsum of head and mesosternal processes pale or pigmented. Thoracic and abdominal segmental sclerotisation and pigmentation rare completely absent ... 15 Siphunculus at least $0.26 \mathrm{~mm}, 0.6-0.95$ times antennal segment III, and $1.7-1.95$ times cauda, which is longer than its basal width. Mesosternal processes conspicuous. [Nieto Nafría et al. (2013): fig. 3A. On Dianthus carthusianorum, $D$. caryophyllus, $D$. commutatus, $D$. monspessulanus, $D$. rupicola, $D$. sp. and Silene borysthenica. Portugal, Spain, France, Switzerland, Italy and Ukraine] A. pujoli and 1.7-1.9 times cauda, which is not longer than its basal width. Mesosternal processes sometimes inconspicuous. [Nieto Nafría et al. (2013): fig. 5B. On Dianthus sp. Pakistan]


## A. pakistanensis

Antennal segment I at least 1.25 times its maximal width. Longest dorsal setae on abdominal segments $2-4$ are $35-55 \mu \mathrm{~m}$ and 1.5-2.0 times basal diameter of antennal segment III. [Discal plate oval and dark; siphunculi weakly ornamented, smooth distad; Nieto Nafría et al. (2013): fig. 2B. On Silene italica, S. nutans, perhaps S. viscosa, and S. sp.; France, Italy, Greece] .......A. delmasi

- Antennal segment I at most 1.2 times its maximal width. Longest dorsal setae on abdominal segments $2-4$ at most $25 \mu \mathrm{~m}$ and 1.2 times basal diameter of antennal segment III. 16
Abdomen usually with spinopleural patch and separate marginal sclerites; if a discal plate is present then it has irregular margins and frequently there are windows in spinal areas of the thoracic, if integrated, and anterior abdominal segments. Dorsal patch or plate smooth and reticulated. Siphunculi dark brown to black, subcylindrical and usually straight, 1.8-2.0 times cauda, which is broad triangular and has $10-16$ setae. Ultimate rostral segment with $6-10$ accessory setae. [Nieto Nafría et al. (2013): fig. 2C. On Silene inaperta, S. italica, S. nutans, S. saxifraga, S. otites, S. vulgaris, S. wolgensis and S. sp. France, Switzerland, Italy, Hungary, Romania, Ukraine and Russia]
A. ornata Characters not in above combination. 17

22 Antennal segment VI processus terminalis 5.0-5.5 times antennal segment VI base. Cauda triangular or tongue-shaped with slight proximal constriction. Ultimate rostral segment shorter than second segment of hind tarsi. [Nieto Nafría et al. (2013): fig. 6C. On Gypsophila arenaria, G. paniculata, G. perfoliata, G. sp. Slovakia, Hungary, Ukraine, Kazakhstan, Russia: Western Siberia]

- Antennal segment VI processus terminalis 2.8-4.0 times antennal segment VI base. Cauda tongue-shaped. Ultimate rostral segment 1.23-1.45 times second segment of hind tarsi. [Clypeus swollen both forward and laterally; Nieto Nafría et al. (2013): fig. 5D. On Spergularia marina. Iran]
A. urmiensis

Longest setae on abdominal segments $2-4$ (dorsum) and antennal segment III 3-8 $\mu \mathrm{m}$ and $0.15-0.50$ times basal diameter of antennal segment III .. 18 Longest setae on abdominal segments 2-4 (dorsum) and antennal segment III $8-25 \mu \mathrm{~m}$ and $0.30-1.60$ times basal diameter of antennal segment III; if they are $8 \mu \mathrm{~m}$ long then marginal abdominal tubercles present or ultimate rostral segment shorter than second segment of hind tarsi.19

Siphunculi dark brown, head dorsum, mesosternal processes and cauda brown to dark brown. Ultimate rostral segment 1.15-1.25 times second segment of hind tarsi. Cauda 1.4-1.8 times its basal width. [Nieto Nafría et al. (2013): fig. 2D. On Gypsophila paniculata, Silene borysthenica, S. moldavica, S. otites, S. wolgensis and S. sp. Slovakia, Hungary, Greece, Ukraine, and Moldova]
A. pannonica

- $\quad$ Siphunculi (with smoked apex, head dorsum, mesosternal processes and cauda pale. Ultimate rostral segment as long as second segment of hind tarsi. Cauda 1.0-1.1 times its basal width. [Kadyrbekov (2013): fig. 1. On Gypsophila perfoliata. Kazakhstan] A. togaica Marginal tubercles on prothorax and abdominal segments 2-4 usually present and spinal tubercle on abdominal segment VIII sometimes present ... 20 Cauda tongue-shaped, 1.40-1.80 times its basal width. Mesosternal processes more or less pigmented, usually darker than tibiae. [Thoracic and abdominal sclerotisation variable, usually a spinopleural abdominal patch with irregular edges and windows in several segments, including the posterior ones; siphunculi pig-
mented, but usually pale than abdominal sclerotized dorsum; Nieto Nafría et al. (2013): fig. 1C. On Dianthus barbatus, D. caryophyllus, D. crinitus, D. sp., Silene conoidea, S. fruticosa, S. italica, S. otites, S. paradoxa, S. thymifolia, S. vulgaris, and S. sp. Spain, France, Italy, Slovenia, Hungary, Greece, Bulgaria, Turkey, Israel, Iran, Afghanistan, Pakistan, Tajikistan, and Russia: Asiatic] $\qquad$ A. picta
- Cauda triangular, although sometimes with a slight proximal constriction, 1.05-1.40 times its basal width. Siphunculi and mesosternal processes as pale as tibiae.


24 Ultimate rostral segment $0.90-1.00$ times second segment of hind tarsus, with $8-10$ accessory setae. Cauda approximately $1.30-1.40$ times its basal width. Longest dorsal setae on abdominal segment $2-3$ are $8-11 \mu \mathrm{~m}$ and $0.3-0.5$ basal diameter of antennal segment III. [Kadyrbekov (2013): fig. 2. On Silene lithophila. Kazakhstan]. A. massagetica

- Ultimate rostral segment 1.05-1.45 times second segment of hind tarsus, with $10-16$ accessory setae. Cauda approximately $1.05-1.35$ times its basal width. Longest dorsal setae on abdominal segment $2-3$ are 13-23 $\mu \mathrm{m}$ and $0.6-1.0$ basal diameter of antennal segment III. [Nieto Nafría et al. (2013): fig. 4A, B. On Silene gallica and S. paradoxa. France].....................A. gallica


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