

# First records of *Cotylea* (Polycladida, Platyhelminthes) for the Atlantic coast of the Iberian Peninsula

Carolina Noreña<sup>1,†</sup>, Daniel Marquina<sup>1,‡</sup>, Jacinto Perez<sup>2,§</sup>, Bruno Almon<sup>3,|</sup>

**1** Dept. Biodiversidad y Biología Evolutiva. Museo Nacional de Ciencias Naturales (CSIC). Calle Jose Gutierrez Abascal 2, 28006 Madrid. Spain **2** Grupo de Estudios do Medio Mariño (GEMM), Puerto deportivo s/n 15960 Ribeira, A Coruña, Spain **3** Instituto Español de Oceanografía, Canary Islands, Centro Oceanográfico de Canarias, Vía Espaldón, parcela 8, 38180 Santa Cruz de Tenerife, Spain

† <http://zoobank.org/DD03B71F-B45E-402B-BA32-BB30343E0D95>

‡ <http://zoobank.org/DFD934A4-AF1E-4A7E-A8F8-05C1F75887F3>

§ <http://zoobank.org/1B36DC0B-C294-4FC7-85CE-1B0C7C658129>

| <http://zoobank.org/7C752276-FBC7-4B16-9203-936B1BC46224>

Corresponding author: Carolina Noreña (norena@mncn.csic.es)

---

Academic editor: D. Gibson | Received 21 January 2014 | Accepted 1 April 2014 | Published 22 April 2014

---

<http://zoobank.org/D73FC0CA-E824-41CD-A18C-553BE2471DFE>

---

**Citation:** Noreña C, Marquina D, Pérez J, Almon B (2014) First records of *Cotylea* (Polycladida, Platyhelminthes) for the Atlantic coast of the Iberian Peninsula. ZooKeys 404: 1–22. doi: 10.3897/zookeys.404.7122

---

## Abstract

A study of polyclad fauna of the Atlantic coast of the Iberian Peninsula was carried out from 2010 to 2013. The paper reports nine new records belonging to three Cotylean families: the family Euryleptidae Lang, 1884, Pseudocerotidae Lang, 1884 and the family Prosthlostomidae Lang, 1884, and describes one new species, *Euryleptodes galikias* sp. n.

## Keywords

Euryleptidae, Prosthlostomidae, Pseudocerotidae, new species, Spain

## Introduction

As a result of studies by Bock (1913), Faubel (1983/1984), Faubel and Warwick (2005), Prudhoe (1985) and Hyman (1940), the North European and North American polyclad fauna of the Atlantic coast is relatively well known. Therefore, sufficient information is available to establish, or recognize, zoogeographical distribution patterns for polyclads in the Atlantic region. The five regions proposed by Prudhoe (1985) are: 1. the Eastern North Atlantic cold-temperate or eastern boreal province, which extends from northern Norway to the English Channel; 2. the Lusitanian province, or North Atlantic warm-temperate region, which comprises the area from the southern part of the English Channel to the Black Sea coasts, including the Mediterranean Sea and the Cape Verde Islands; 3. the Western boreal province, comprising the northern west coast of the Atlantic Ocean, the coasts of Canada and the United States from Bat Baffin (Canada, Arctic Circle) to Cape Cod (New England, United States); 4. the Carolinian province, extending from south of Cape Cod to North Carolina; 5. the West Indian province encompasses the Gulf of Mexico to southern Brazil, including the Antilles and Bermuda, characterized by warm tropical waters.

Some Mediterranean species, such as *Prostheceraeus rubropunctatus* Lang, 1884; *P. vittatus* (Montagu, 1815) Lang, 1884; *P. roseus* Lang, 1884; *Anonymus virilis* Lang, 1884; or *Oligocladus sanguinolentus* (Quatrefages, 1845), are also found in the Cape Verde archipelago or along the European coast, but there are no records for these species for the American Atlantic coast. Except for species that have a nearly worldwide distribution, such as *Thysanozoon brocchii* (Risso, 1818) Grube, 1840) or *Stylostomum ellipse* (Dalyell, 1853) Lang, 1884, there are no species shared between the eastern and western coast of the Atlantic Ocean.

Temperature seems to form a natural barrier along the longitudinal axis of the Atlantic Ocean. The distribution of polyclads follows a north-south pattern for both the east and west coasts. Therefore, there are species common to both the Mediterranean and Scandinavian coast, but not to the American and European coasts.

In this paper, we present nine new records belonging to three Cotylean families: the family Euryleptidae Lang, 1884, Pseudocerotidae Lang, 1884 and the family Prosthlostomidae Lang, 1884, and we describe one new species, *Euryleptodes galikias* sp. n.

## Material and methods

Polyclads were found off the coast of northwestern Spain (Galicia) by scuba diving, while cataloguing the marine fauna of Ria de Arosa (NW Spain) under the auspices of the Grupo de Estudo do Medio Mariño ("Study Group of the Marine Environment" GEMM), which has been operating for the past 10 years. These surveys cover a bathymetric range from the intertidal zone to a maximum accessible depth for scuba diving of approximately 40 meters. Most species were found in habitats typical for polyclads e.g., empty mollusc shells, over Bryozoa, algae, etc.



Specimens were first photographed in the field, then collected by hand using a brush or net and stored in containers according to specimen size. Once in the laboratory, a small piece of tissue was carefully removed for DNA analysis, and the rest of the animal was fixed with Bouin's fluid for a complete histological study of its anatomy. The specimens were dehydrated in alcohol, cleared in xylene and subsequently photographed to record details about the eyes, pigmentation, body shape and tentacles, as well as the location of the pharynx and reproductive organs. Afterwards, they were embedded in paraplast, sagittally sectioned at 7  $\mu\text{m}$  and stained with Azan trichrome. Reconstructions of internal morphologies were derived from serial sagittal sections. Measurements were determined from fixed material.

The material was deposited in the Invertebrate Collection of the Museo Nacional de Ciencias Naturales (MNCN; Spain).

Abbreviations used in the figures

<b>b</b>	brain	<b>p</b>	papillae
<b>ce</b>	cerebral eyes	<b>ph</b>	pharynx
<b>cg</b>	cement glands	<b>pv</b>	prostatic vesicle
<b>cp</b>	cement pouch	<b>rh</b>	rhabdites
<b>ed</b>	ejaculatory duct	<b>s</b>	stylet
<b>esv</b>	external seminal vesicle	<b>spb</b>	spermiducal bulbs
<b>fa</b>	female atrium	<b>su</b>	ucker
<b>fe</b>	frontal eyes	<b>sv</b>	seminal vesicle
<b>fp</b>	female pore	<b>t</b>	tentacle
<b>i</b>	intestine	<b>te</b>	tentacular eyes
<b>ib</b>	intestinal branches	<b>ts</b>	testes
<b>ma</b>	male atrium	<b>uv</b>	uterine vesicles
<b>mp</b>	male pore	<b>v</b>	vagina
<b>o</b>	ovaries	<b>vd</b>	vas deferens
<b>op</b>	oral pore		

## Systematics

**Suborder COTYLEA** Lang, 1884

**Family EURYLEPTIDAE** Lang, 1884

**Genus *Cycloporus*** Lang, 1884

***Cycloporus papillosus* (Sars, 1878) Lang, 1884**

[http://species-id.net/wiki/Cycloporus\\_papillosus](http://species-id.net/wiki/Cycloporus_papillosus)

Figure 1

**Material examined.** Four individuals captured during summer, autumn and winter between 2010 and 2012 (11/10/2010; 03/08/2011; 27/01/2011; 09/01/2012). Voucher:

one specimen sectioned sagittally, stained with azan and deposited in the Invertebrate Collections of the MNCN; Cat. Nr: MNCN 4.01/573 to 4.01/599 (27 slides). Further material: one specimen cat. Nr. MNCN 4.01/600 to 4.01/625 (26 slides).

**Description.** Elongated worms 11 mm long and 6 mm wide. Body shape elongated with light undulating margins and rounded anterior and posterior ends. Dorsal surface with numerous papillae. Colouration orange, yellowish orange or translucent grey with white patches at the mid-dorsal line (Figure 1A, B, C, D). Ventral side smooth and pale. Short inconspicuous marginal tentacles. Sucker located approximately in the middle of the body (Figure 1E). Tentacular eyes scattered over dorsal margin of tentacles (Figure 1A), cerebral eyes in two elongated, anteriorly anastomosing clusters. Plicate cylindrical or tubular pharynx near anterior end, frontally oriented; oral pore posterior to brain. Male and female genital pores clearly separated and posterior to pharynx (Figure 1E).

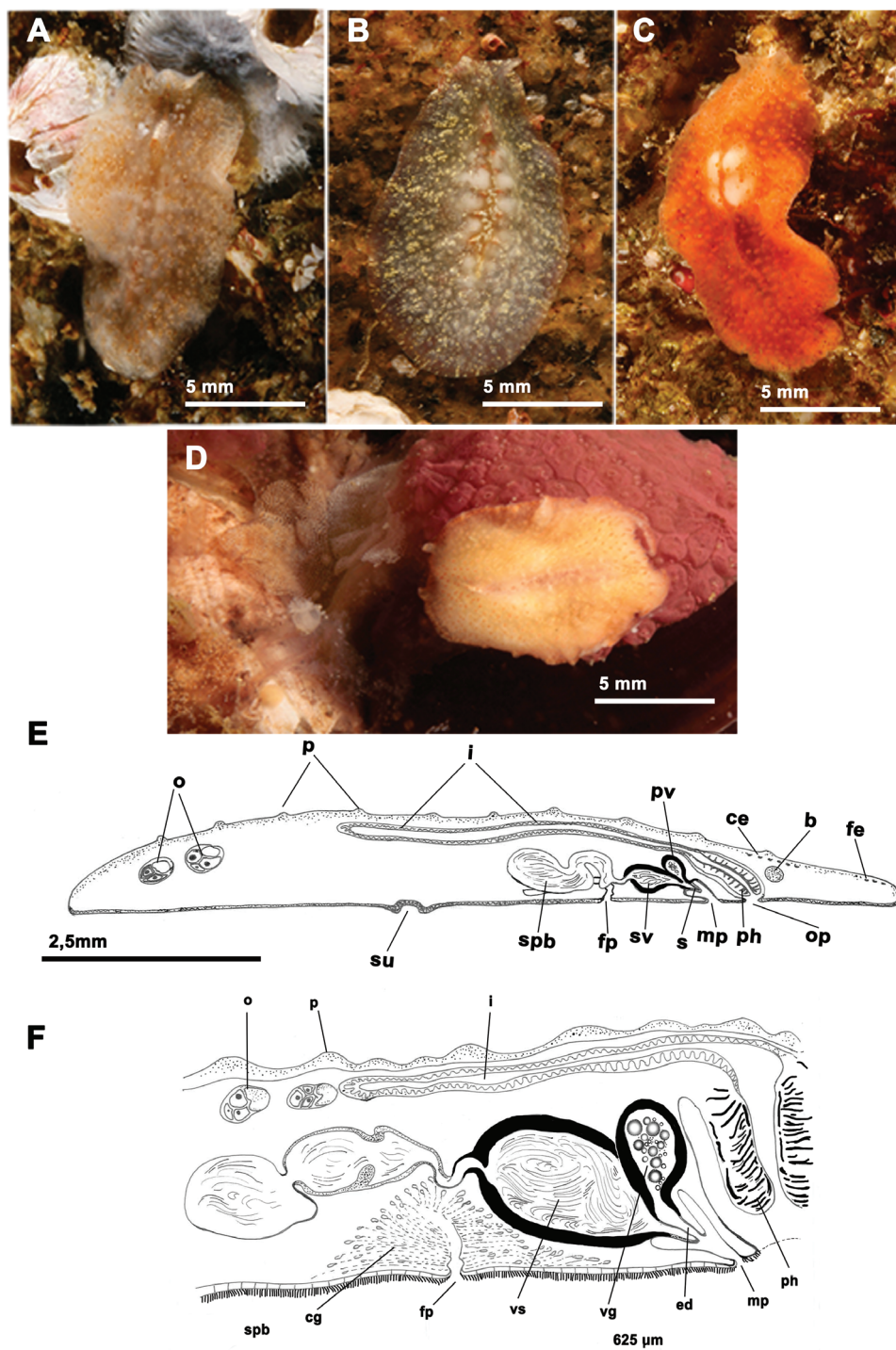
Male copulatory apparatus located posterior to male pore and oriented mainly dorso-ventrally, but also directed frontally (Figure 1E, F). Male system consists of a short, armed (stylet) penis papilla, a true prostatic vesicle with a smooth glandular epithelium, and a well developed, muscularized seminal vesicle. Prostatic vesicle opens directly into penis papilla, and seminal vesicle empties directly into distal end of prostatic vesicle. Vasa deferentia, sometimes very dilated, open proximally through a common duct (common vas deferens) into seminal vesicle.

Inconspicuous female system lies posterior to the male pore and is characterized by a short female atrium, female duct (vagina), and characteristic uterine vesicles. Abundant cement glands are located around female pore and distal part of vagina.

**Remarks on biology.** *Cycloporus papillosus* is a natural predator of *Botrylloides violaceus* Oka, 1927 (Ascidacea), which is a clear example of an invasive species. *B. violaceus* grows on all types of substrates, including other living animals such as mussels, small sea cucumbers or other ascidians, covering them completely and killing them. *B. violaceus* has completely replaced *Botrylloides leachii*, the autochthonous ascidian in this area. Both species of ascidians compete for the same substrate. *C. papillosus* preys on *B. violaceus* and places its egg plates (Fig. 1D) in the folds of this species (in the area where new zooids grow and extend the colony) or under the unattached colony, thereby ensuring larval protection and the availability of food after hatching.

**Distribution.** In Galicia, three specimens of *C. papillosus* were captured from mussels collected on *Botrylloides violaceus* on the docks of the Yacht Club Ribeira (Ria de Arosa, Galicia, Spain). Depth varied between 0.5 and 1 metres (42°33.7770N / 008°59.2970W; 42°33.7850N / 008°59.3140W; 42°33.7930N / 008°59.3290W). Another specimen (Figure 1B) was collected on a colony of *Botryllus schlosseri* (Ascidacea) growing on a rock of the island of Rua (Ria de Arosa, Galicia, Spain), at a depth of 14 metres (42°32.9650N / 008°56.4590W).

This is the southernmost European record for *C. papillosus*, and the first for the Atlantic coast of the Iberian Peninsula. Other localities from where this polyclad has been reported are: Bergen, Norway (Jensen 1878); Rovigno, Croatia (Vátova 1928); Susaki near Simoda, Japan (Kato 1937); Porto Praia, Cape Verde (Laidlaw 1906); Plymouth, United Kingdom (Gamble 1893).



**Figure 1.** *Cycloporus papillosus*. **A–D** dorsal views of living animals **E** sagittal reconstruction of a whole specimen **F** sagittal reconstruction of the copulatory apparatus. Anterior to the right in **E**, **F**.

## Genus *Eurylepta* Ehrenberg, 1831

### *Eurylepta cornuta* (O.F. Müller, 1776) Ehrenberg, 1831

[http://species-id.net/wiki/Eurylepta\\_cornuta](http://species-id.net/wiki/Eurylepta_cornuta)

Figure 2

**Material examined.** Two mature specimens captured during winter 2012 (15/01/2012). Voucher: one specimen sectioned sagittally, stained with Azan and deposited in the Invertebrate Collections of the MNCN; Cat. Nr: MNCN 4.01/626 to 4.01/647 (22 slides).

**Description.** Captured worms 10 mm long and 5 mm wide. Body shape elongated, with straight margins. Dorsal surface smooth. Background coloration of the dorsal surface pale brown, translucent, with dark branched bands, red or brown, depending on intestinal contents, (Figure 2A). Ventral side pale yellow without bands. With narrow conical marginal tentacles; sucker slightly posterior to the middle of the bodies. Tentacular eyes at the base of the tentacles (Figure 2C) and cerebral eyes in two elongated clusters, sometimes extending over the pharynx. Tubular, whitish pharynx is visible at the anterior end (Figure 2C).

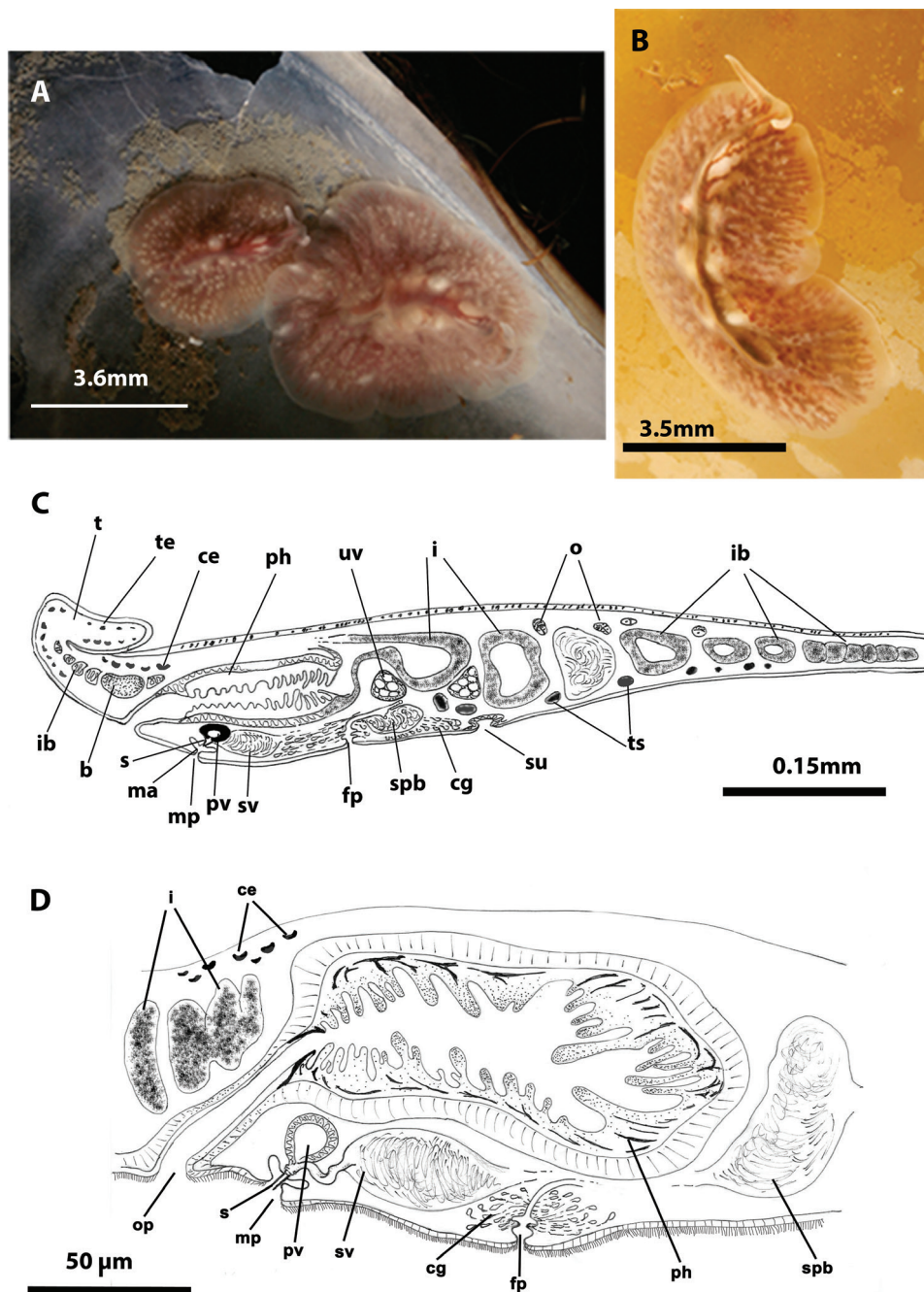
Male copulatory apparatus located posterior to male pore and directed forwards (Figure 2D). Prostatic vesicle oriented antero-dorsally, with a smooth glandular epithelium, and directly connected to tubular stylet of penis papilla. Seminal vesicle empties through a narrow short duct into distal end of prostatic vesicle. Female pore lies closely behind male pore, but is clearly separated. Female atrium elongated. Cement pouch rounded and followed by a short vagina and opening of uteri. A pair of uterine vesicles is present.

**Remarks.** *Eurylepta cornuta* shows two varieties: 1. *E. cornuta* var. *lobianchi*, first described by Lang (1884) and known from the Mediterranean Sea, and 2. *E. cornuta* var. *melobesiarum*, first described by Schmittlein (1880) as *Proceros melobesiarum*. The main difference between these varieties is in the arrangement of the cerebral eyes (Lang 1884; Bock 1913). In *E. cornuta* var. *melobesiarum* the elongated patches are shorter than in *E. cornuta* var. *lobianchi*. Therefore, and in agreement with other authors (e.g., Micoletzky 1910; Bock 1913; Faubel 1984), we consider the difference not enough to maintain the two varieties and propose that should no longer be recognized.

**Distribution.** *Eurylepta cornuta* was found in empty shells of *Mytilus galloprovincialis* (Figure 2A), which were attached to mussel culture ropes suspended from specially designed rafts called “bateas”, located in La Palmeira (Ria de Arosa, Galicia, Spain) at a depth of 13 metres (42° 34.3910N / 008°56.6360W). Several specimens of *E. cornuta* (Figure 2B) were also captured for the first time within *Saccorhiza polyschides* stipes (macroalgae), at a depth of 8 metres in “Cuberto Camouco” (Ria de Arosa, Galicia, Spain) (42°33.4150N / 008°57.8390W). Another specimen was found under a stone on the island of Rua, at a depth of 14 metres (42°32.9200N / 008°56.4220W).

*Eurylepta cornuta* has been known since the 18<sup>th</sup> century from Kristiansand, Norway (O.F. Müller 1776) and since the 19<sup>th</sup> century from Belfast Bay, Ireland (Thompson 1845); Saint Malo, France (Keferstein 1868); Plymouth Sound, United Kingdom (Gamble 1893); and the Gulf of Naples, Italy (Lang 1884).





**Figure 2.** *Eurylepta cornuta*. **A, B** dorsal views of living animals **C** sagittal reconstruction of a whole specimen **D** sagittal reconstruction of the copulatory apparatus. Anterior to the left in **C, D**.

## Genus *Euryleptodes* Heath & McGregor, 1912

### *Euryleptodes galikias* sp. n.

<http://zoobank.org/7D732693-EB62-4A44-9958-FC589550BFEE>

[http://species-id.net/wiki/Euryleptodes\\_galikias](http://species-id.net/wiki/Euryleptodes_galikias)

Figure 3

**Material examined.** One specimen captured during winter 2012 (09/12/2012).

**Holotype.** One sagittally sectioned specimen, stained with Azan and deposited in the Invertebrate Collections of the MNCN; Cat. Nr. MNCN 4.01/502 to 4.01/572 (71 slides).

**Type locality.** Ribeira (Ria de Arosa, Galicia, Spain). Depth; 5 metres (42°33.7760N / 008°59.3390W).

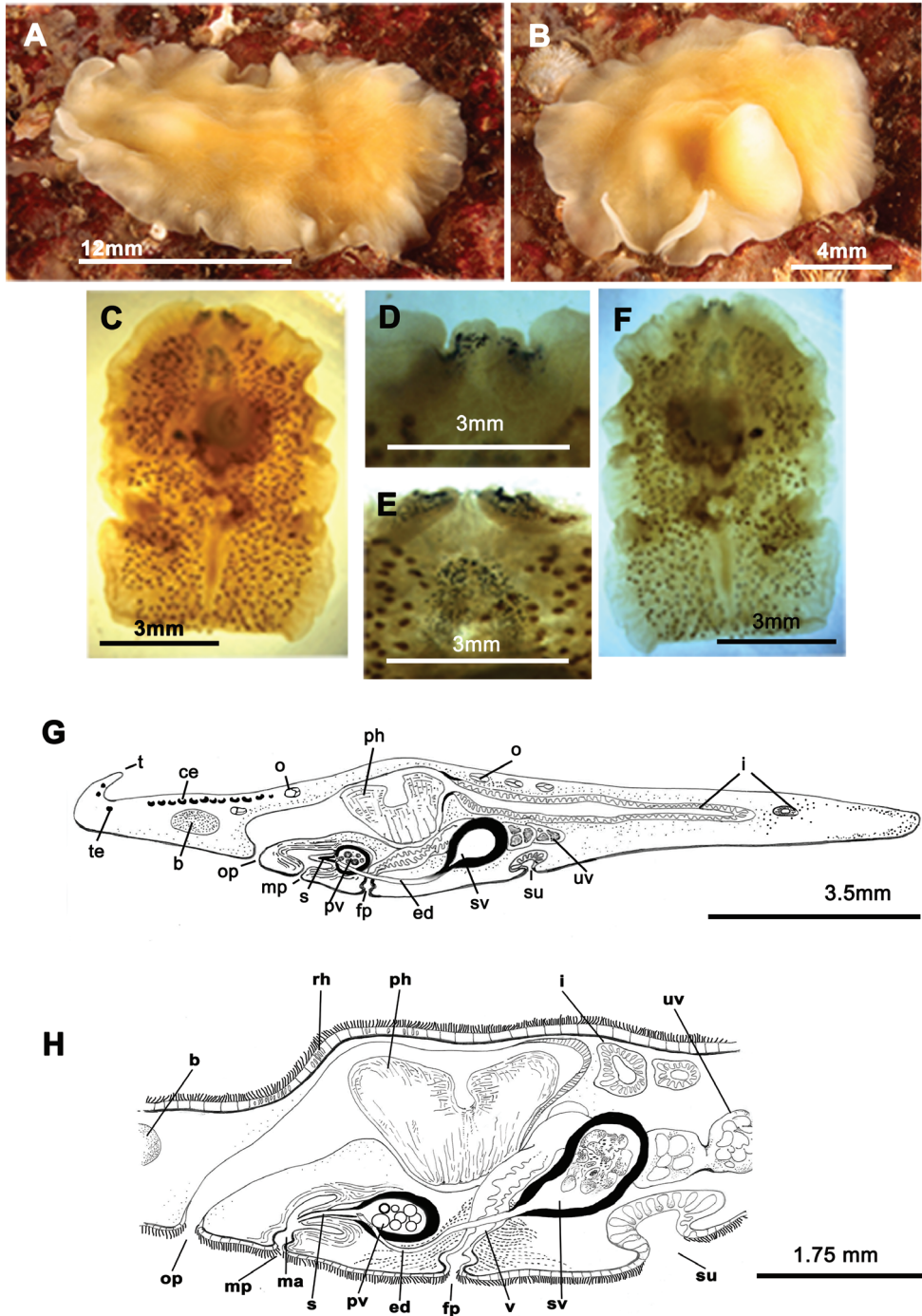
**Description.** Living elongated worms 25 mm long and 14.4 mm wide. Body shape broad, slightly oval and with undulating margins. Colouration in living animals yolk yellow (Figure 3A, B); fixed individuals have a transparent look; with small dark patches representing the uterine network over entire body surface (Figure 3C). Ventral side pale yellow. Marginal tentacles well-developed, conical. Sucker posterior to middle of the body. Tentacular eyes on the dorsal margin of tentacles (Figure 3D), marginal eyes in two small clusters on anterior margin (Figure 3E) and cerebral eyes in two elongated anteriorly anastomosing clusters (Figure 3E). Tubular pharynx near anterior end; oral pore closely posterior to brain. Male and female genital pores clearly separated, anterior and ventral to the pharynx, respectively (Figure 3F).

Male copulatory apparatus located posterior to the male pore and directed forwards (Figure 3G, H). Male system consists of a small penis papilla with a short stylet, a true prostatic vesicle with a smooth glandular epithelium and an elongated seminal vesicle. Prostatic vesicle is oval and small, and opens directly into the penis papilla, which projects into the male atrium. Seminal vesicle empties into distal end of prostatic vesicle through a long, narrow ejaculatory duct. Characteristic spermiducal vesicles open proximally into seminal vesicle.

Small female system is difficult to distinguish except for uterine network and well developed cement glands. Female pore lies behind male pore and is clearly separated from it. Vagina shows a small expansion (or cement pouch; cf Hyman 1953), ascends dorsally and subsequently splits into two uteri. Uteri extend as a uterine network with channels that connects to the ovaries. Uterine vesicles absent.

**Discussion.** Based on the presence of the conical marginal tentacles, the spermiducal vesicles, an armed penis and a uterine network, this new species belongs to the genus *Euryleptodes* Heath & McGregor, 1912 of the family Euryleptidae Lang, 1884. The genus *Euryleptodes* presently comprises of two species: *E. cavicola* Heath & McGregor, 1912 and *E. insularis* Hyman, 1953, both from California.

*Euryleptodes galikias* sp. n. differs from *E. insularis* in the long ejaculatory duct, the tentacular eyes over the tentacles, and the frontal marginal eyes.



**Figure 3.** *Euryleptodes galikias*. **A, B** dorsal view of a living animal **C** dorsal and **F**, ventral views of a fixed specimen **E** dorsal and **D** ventral details of the eyes **G** sagittal reconstruction of a whole specimen **H** sagittal reconstruction of the reproductive system **H**. Anterior to the left in **A**, **F** and **G**.

Compared to *E. cavicola*, which has a long stylet, the stylet of *E. galikias* is short. Furthermore, *E. cavicola* lacks spermiducal vesicles, thus distinguishing it from both *E. galikias* and *E. insularis*.

Differences in colouration are also apparent between the species: yolk yellow in *E. galikias* sp. n., greenish white in *E. cavicola*, and brown with dark spots in *E. insularis*. Lastly, the pattern of distribution is vastly different, with *E. galikias* off the eastern coast of the North Atlantic Ocean (Spain), while the other two species occur off the eastern coast of the North Pacific Ocean (California).

**Distribution.** *Euryleptodes galikias* sp. n. was found within empty shells of *Mytilus galloprovincialis*, which were attached to a boat anchor on the third dock of the Yacht Club Ribeira (Ria de Arosa, Galicia, Spain), of a depth of 5 metres (42°33.7760N / 008°59.3390W).

### Genus *Prostheceraeus* Schmarda, 1859

#### *Prostheceraeus vittatus* (Montagu, 1815) Lang, 1884

[http://species-id.net/wiki/Prostheceraeus\\_vittatus](http://species-id.net/wiki/Prostheceraeus_vittatus)

Figure 4

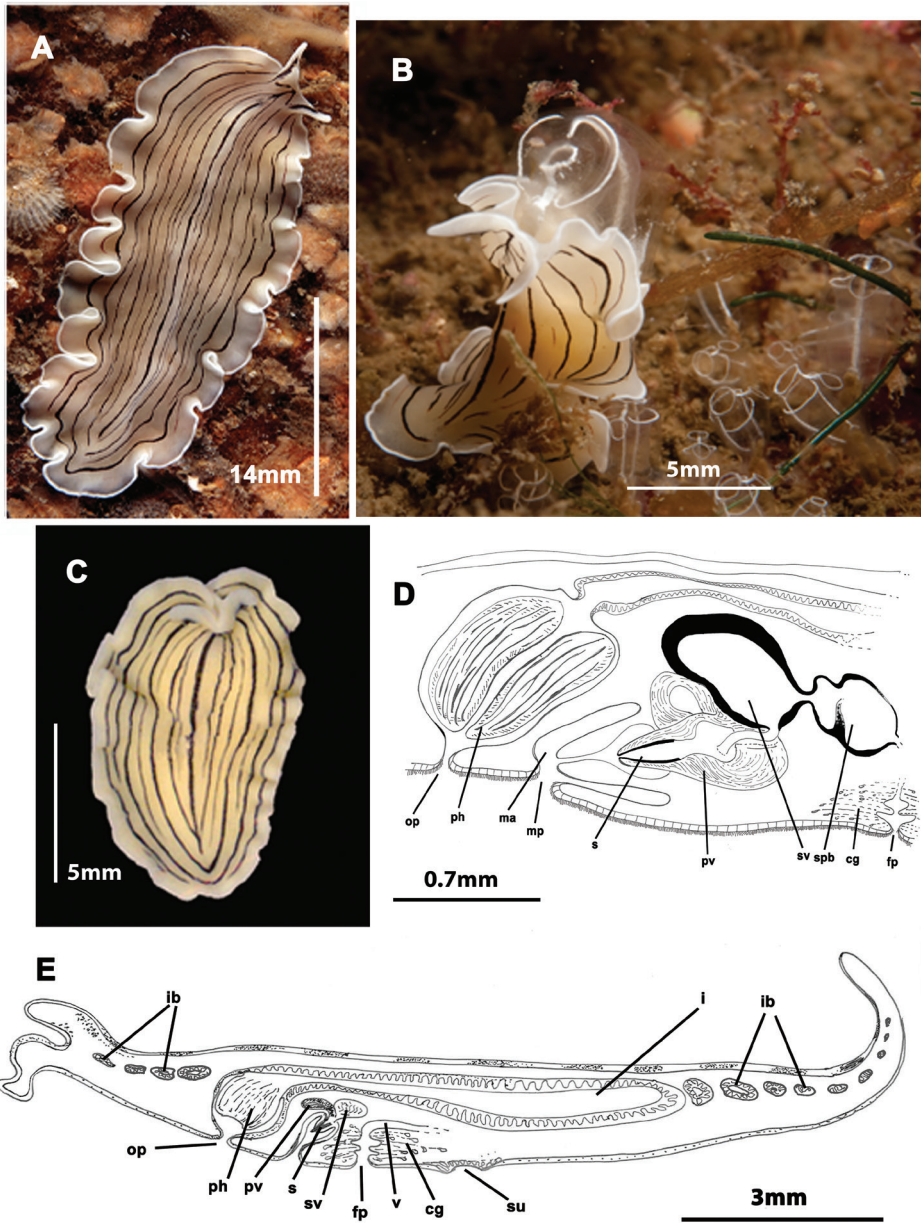
**Material examined.** Two specimens captured in winter 2010 and 2012 (15/03/2010 and 24/02/2012). Voucher: one specimen sectioned sagittally, stained with Azan and deposited in the Invertebrate Collections of the MNCN; Cat. Nr: MNCN 4.01/648 to 4.01/662 (15 slides).

**Description.** Elongated worms 10–30 mm long and 7–15 mm wide. Body shape elongated, leaf-shaped, with pointed anterior and posterior ends, and with light undulating margins. Marginal tentacles well developed with whitish edges and pointed ends. Dorsal surface smooth. Background coloration whitish or ivory, with black, continuous stripes; between the stripes, black discontinuous lines are present (Figure 4C). Faint white band runs along the entire body margin (Figure 4A, B). Ventral side smooth and pale. Sucker in middle of body or slightly more posterior (Figure 4E). Cerebral eyes form two compact elongated, frontally anastomosing groups (Figure 4A). Tentacular eyes scarce and mainly at base of tentacles. Tubular pharynx near anterior end, oral pore in posterior region first quarter of the body. Male and female genital pores clearly separated and located behind the pharynx (Figure 4D, E).

Male copulatory apparatus with antero-dorsally oriented prostatic vesicle (Figure 4D, E). Male system consists of a short penis papilla armed with a small conical stylet, a true prostatic vesicle with a smooth glandular epithelium and a seminal vesicle with a thick muscle layer. Vasa deferentia join a dilated common vas deferens that opens into seminal vesicle. Copulatory complex lies forwardly oriented, and seminal vesicle opens through a small duct directly into distal end of prostatic vesicle.

Female system lies posterior to male pore and is characterized by a short, rounded female atrium and a cement duct or pouch. In our specimen, a second dilatation





**Figure 4.** *Prostheceraeus vittatus*. **A** dorsal view of a living animal **B** living animal feeding on ascidians **C** dorsal view of a fixed specimen **D** sagittal reconstruction of the copulatory apparatus **E** sagittal reconstruction of a whole specimen. Anterior to the left in **D**, **E**.

(so-called shell gland pouch) follows the atrium, into which shell glands open. Posteriorly-orientated vagina and numerous uterine vesicles are situated medially to this pouch.

**Biology.** *Prostheceraeus vittatus* feeds mainly on *Clavelina* ascidians, as other *Prostheceraeus* species (Figure 4B).

**Remarks.** Lang (1884) mentions in the original description that specimens less than 1.5–2 cm in length were immature. However, sometimes our specimens from the Atlantic coast were mature, despite their small size (c. 1.5 cm in length).

**Distribution.** Two specimens of *Prostheceraeus vittatus* were captured during this study. The first animal was collected from “A Tiñosa” (Ria de Arosa, Galicia, Spain) on a rocky bottom between *Clavelina lepadiformis* colonies, at a depth of 24 metres (42°32.8240N / 008°57.9920W). The other worm was found on stones in “Petón Bajo” (Ria de Arosa, Galicia, Spain), at a depth of 16 metres (42°32.9880N / 008°57.9920W).

*Prostheceraeus vittatus* is known from the North Atlantic coasts of the United Kingdom, France, Ireland, Scandinavia, Norway, Denmark, from the Mediterranean shores in Italy (Gulf of Naples) (Faubel and Warwick 2005) and Spain (Catalonia) (Novell 2003). This is the first record for the species from the North Atlantic side of the Iberian Peninsula.

#### ***Prostheceraeus moseleyi* Lang, 1884**

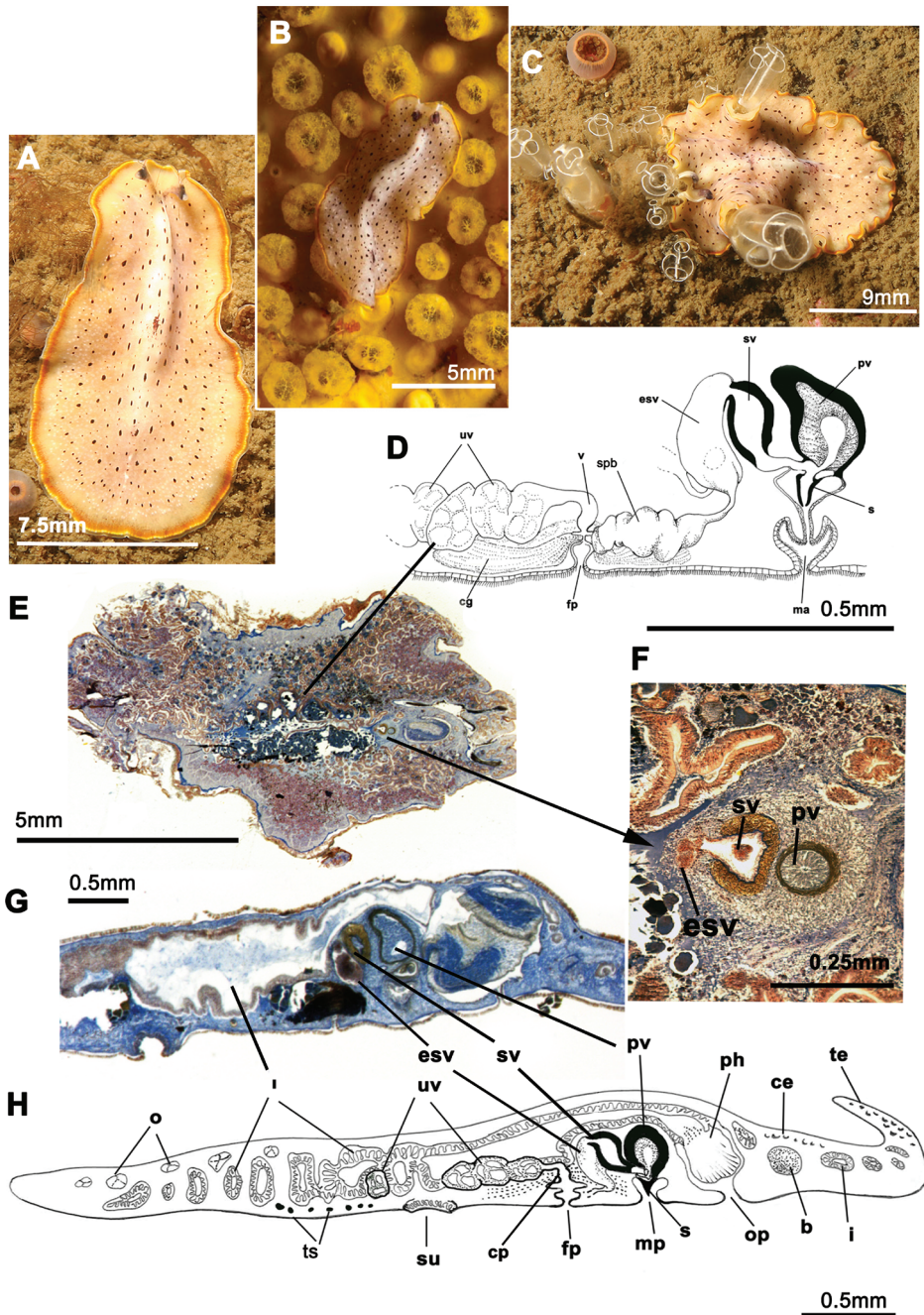
[http://species-id.net/wiki/Prostheceraeus\\_moseleyi](http://species-id.net/wiki/Prostheceraeus_moseleyi)

Figure 5

**Material examined.** Two specimens captured during the spring of 2010 (07/06/2010). Vouchers: one specimen sectioned sagittally, stained with Azan and deposited in the Invertebrate Collections of the MNCN: Cat. Nr: MNCN 4.01/663 to 4.01/688 (26 slides); one specimen sectioned sagittally, stained with Azan and deposited in the Invertebrate Collections of the MNCN: Cat. Nr: MNCN 4.01/689 to 4.01/731 (43 slides).

**Description.** Elongated worms 1–1.5 cm long and 0.5–0.7 mm wide (Figure 5A, B, C). Body shape elongated, wider at the posterior end, with light undulating margins and rounded anterior and posterior ends. Marginal tentacles with characteristic purple pigment. Dorsal surface smooth. Dorsal colouration whitish or yellowish, with dark, rounded spots and a yellow band along the body margin; at times background pigmentation faintly orange or pinkish (Figure 5A, B). Ventral side smooth and pale. Sucker approximately in middle of body. Tentacular eyes scarce and scattered over tentacles; cerebral eyes inconspicuous, in two elongated rows. Plicate cylindrical or tubular pharynx near anterior end, oriented frontally; oral pore behind brain. Male and female genital pores clearly separated and behind pharynx (Figure 5H).

Male copulatory apparatus perpendicular to male pore (Figure 5D, G, H). Male system consists of a short penis papilla, armed with a small conical stylet, a true prostatic vesicle with a smooth glandular epithelium, and a seminal vesicle with a thick muscle layer. Vasa deferentia, sometimes very dilated, join an expanded vas deferens (possibly an external seminal vesicle; Figure 5F, G) before opening directly into true seminal vesicle. Prostatic vesicle opens at tip of penis papilla and seminal vesicle opens through a duct into distal end of prostatic vesicle.



**Figure 5.** *Prostheceraeus moseleyi*. **A, B, C** dorsal views of living animals **D** sagittal reconstruction of the reproductive system **E** dorso-ventral histological sections of the whole animal **F** dorso-ventral histological sections of the copulatory apparatus **G** sagittal histological section in the region of the pharynx and copulatory apparatus **H** sagittal reconstruction of a whole specimen. Anterior to the right in **C, D, E, F, G** and **H**.



Female system (Figure 5D, E) lies posterior to male pore and is characterized by a short, rounded female atrium, a cement duct or pouch, followed by a second dilatation of atrium and a posteriorly-orientated vagina. Cement and shell glands empty into cement duct.

**Biology.** *Prostheceraeus moseleyi* feeds mainly on *Clavelina lepadiformis* (Ascidacea) (Figure 5B, C).

**Remarks.** Faubel (1984) considers *P. moseleyi* as a species *incertae sedis* because in the original description by Lang (1884) the uterine vesicles were not mentioned. In our specimens, multiple uterine vesicles (Figure 5D, E) run along both sides of the main body axis and therefore, *P. moseleyi* can now be considered as a valid species of the genus *Prostheceraeus*.

**Distribution.** Specimens of *P. moseleyi* were collected from the bottom of the pier at the Yacht Club Ribeira (Ria de Arosa, Galicia, Spain) at a depth of 5 to 10 metres (42°33.7410N / 008°59.3380W). This species was recorded from Italy (Gulf of Naples, Lang 1884) and Spain (in several localities of the coast of Catalonia, Novell 2003). Our sample represents the first record for the Atlantic coast and outside from the Mediterranean Sea.

## Family PROSTHIOSTOMIDAE Lang, 1884

### Genus *Prosthiostomum* Quatrefages, 1845

#### *Prosthiostomum siphunculus* (Delle Chiaje, 1822) Lang, 1884

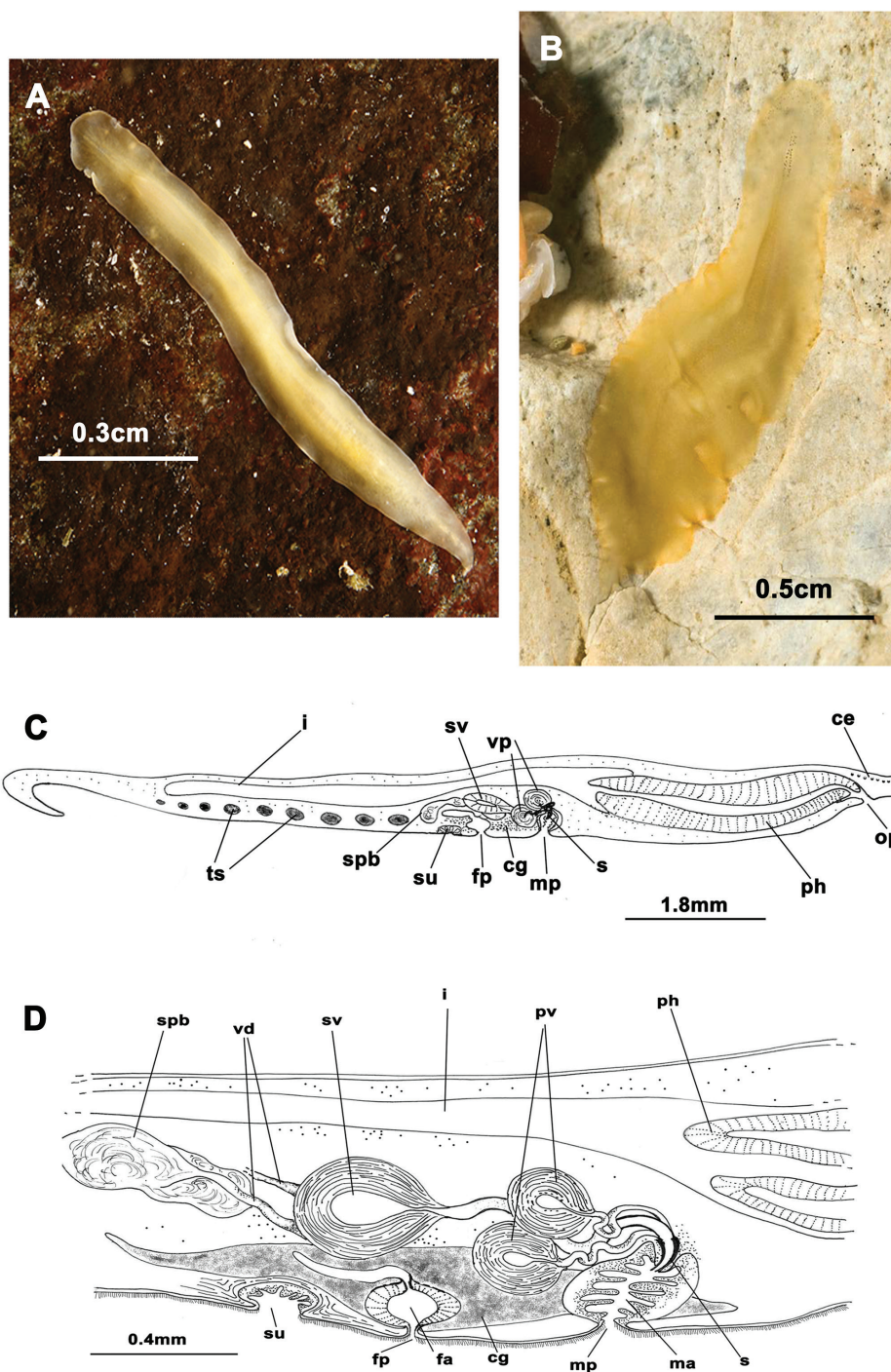
[http://species-id.net/wiki/Prosthiostomum\\_siphunculus](http://species-id.net/wiki/Prosthiostomum_siphunculus)

Figure 6

**Material examined.** Two specimens collected in winter 2012 (09/01/2012). Voucher: one specimen sectioned sagittally, stained with Azan and deposited in the Invertebrate Collections of the MNCN Cat. Nr: MNCN 4.01/732 to 4.01/744 (13 slides).

**Description.** Mature, elongated specimens 10–18 mm long, occasionally as long as 30 mm, and 4–6 mm wide. Body shape long and narrow, sometimes with a little fan-like expansion at the anterior end, straight margins, rounded anterior and tapered posterior end. Without tentacles. Dorsal surface smooth. Dorsal colouration beige to yellow, without spots or bands (Figure 6A, B). Ventral side smooth and pale. Sucker in the middle of the body or slightly anterior. Cerebral eyes arranged in two slightly curved rows; marginal eyes along the anterior edge; young individuals have an eye-free medial area. Well-developed plicate tubular pharynx located near the anterior end, oriented frontally; oral pore closely behind the brain. Male and female genital pores separated and located posterior to pharynx (Figure 6C).

Male system consists of a short penis papilla armed with a conical, Arabian dagger-like stylet an anterior orientated seminal vesicle and two spherical prostatic vesicles with a smooth glandular epithelium covered with a thick muscle layer. Vasa deferentia



**Figure 6.** *Prosthiostomum siphunculus*. **A, B** dorsal views of living animals **C** sagittal reconstruction of a whole specimen **D** sagittal reconstruction of the copulatory apparatus. Anterior to the right in figures C, D.

open separately into a seminal vesicle. Ejaculatory duct and prostatic ducts very long and spiral-shaped; prostatic ducts meet at proximal end of small penis papilla. Male atrium elongated and conical (Figure 6D).

The female system lies posterior to male pore and is characterized by a short, spindle-like female atrium, a cement gland pouch, followed by a backwards orientated vagina, and backwards directed uteri.

**Distribution.** One specimen was found in mussel samples collected from the floats of the third dock of the Yacht Club Ribeira at a depth of 1 metres (42°33.7700N / 008°59.3260W). The other animal was collected from under a rock in the area “A Ameixida” at a depth of 6 metres (42°32.2490N / 008°59.1640W). *Prosthiostomum siphunculus* is known from the western European Atlantic coasts, the Mediterranean and the Tyrrhenian Sea, and also from North and South Africa, Somalia and Vietnam (Prudhoe 1985).

### Other species of Polycladida found in the study area

The following four well-known species were identified on the basis of their characteristic external anatomy and were also photographed:

### Family EURYLEPTIDAE Lang, 1884

#### *Prostheceraeus roseus* Lang, 1884

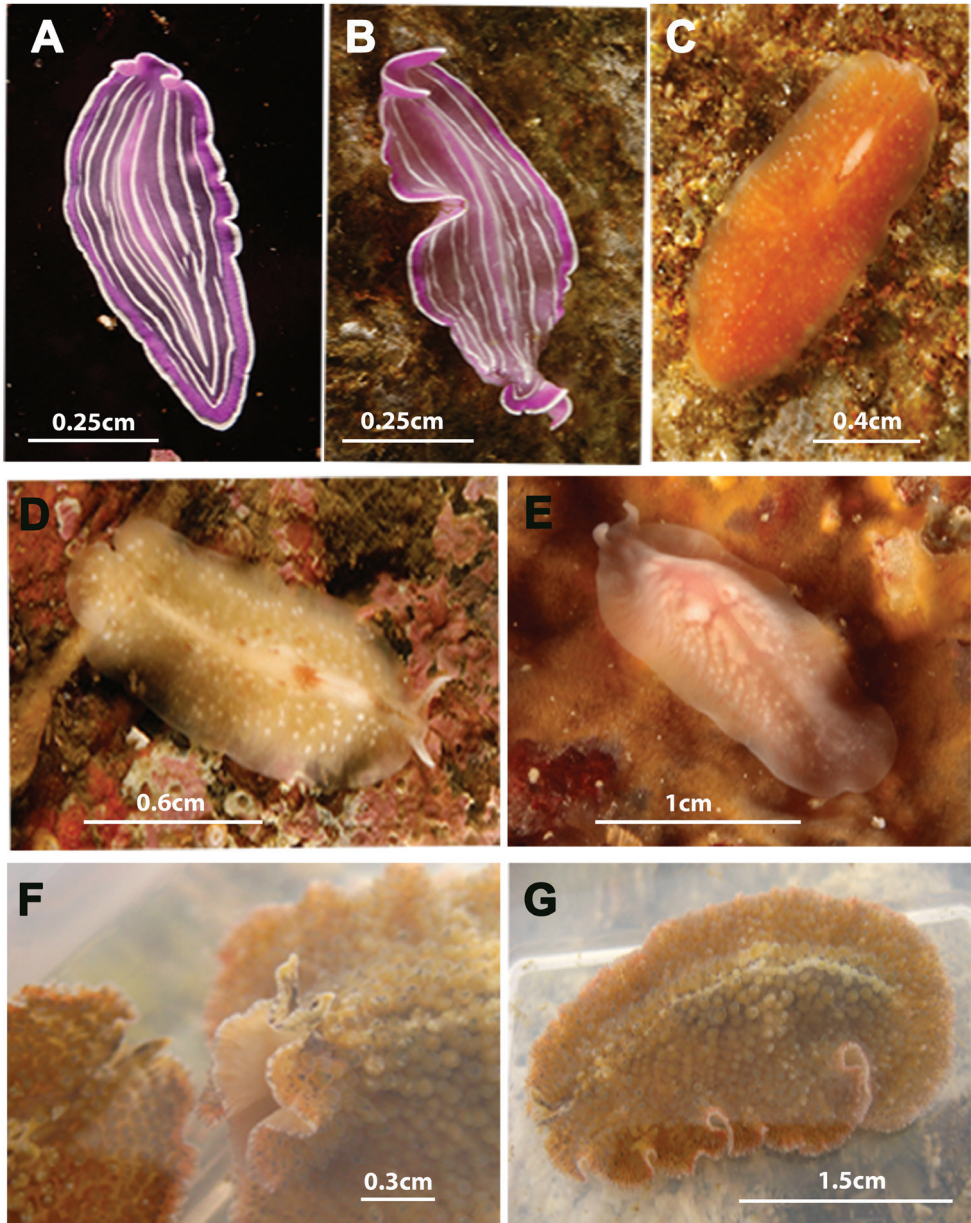
[http://species-id.net/wiki/Prostheceraeus\\_roseus](http://species-id.net/wiki/Prostheceraeus_roseus)

Figure 7A, B

**Remarks.** *Prostheceraeus roseus* belongs to the family Euryleptidae and is characterized by the following: an elongated, oval body shape and smooth surface; conspicuous marginal tentacles; cerebral eye clusters in two small, parallel rows that do not anastomose. This species is readily recognizable by its distinct pink to purple pigmentation with white longitudinal stripes and a white edge that runs along the entire body margin. The tubular, bell-shaped pharynx oriented frontally, and the digestive system well developed with numerous, anastomosing branches. The reproductive system shows the characteristics of the genus: male copulatory apparatus frontally oriented; penis armed with tubular pointed stylet and female complex with multiple uterine vesicles.

**Distribution.** *Prostheceraeus roseus* was one of the first species collected during this study, first in summer of 2010, then in autumn of 2013 (14/05/2010 and 10/10/2013). Two specimens were captured in the mouth of the Ria de Arosa in Corrubedo: one on a rocky wall in “Canteiro”, at a depth of 27 metres (42°30.5540N / 009°05.1260W), and the other on *Pycnoclavella producta* (Ascidacea) in “A Dianteira”, at a depth of 23 metres





**Figure 7.** Species photographed but not collected. **A, B** *Prostheceraeus roseus* **C** *Stylostomum ellipse* **D, E** *Oligocladus sanguinolentus* **F, G** *Thysanozoon brocchii*.

(42°30.9100N / 009°04.3750W). *P. roseus* is also known from the Gulf of Naples, Italy (Lang 1884). These findings represent the first record for the Atlantic coast outside the Mediterranean Sea.

***Stylostomum ellipse* (Dalyell, 1853) Lang, 1884**

[http://species-id.net/wiki/Stylostomum\\_ellipse](http://species-id.net/wiki/Stylostomum_ellipse)

Figure 7C

**Remarks.** *Stylostomum ellipse* belongs to the family Euryleptidae and is characterized by: a small to moderate size and oval outline, sometimes pear-shaped; dorsal surface smooth; marginal tentacles very reduced; small cerebral and marginal eye clusters. *Stylostomum ellipse* is also characterized by the common opening of the male and oral pore, located in front of the pharynx. In contrast, the female pore is situated behind the pharynx. The penis papilla is armed with a tubular pointed stylet and the female system has two uterine vesicles.

**Distribution.** Only one specimen of *Stylostomum ellipse* was captured (09/01/2012) from stones in the “O Rodal de Nuestra Señora”, at a depth of 32 metres (42°31.9010N / 008°58.5330W). *Stylostomum ellipse* is considered a cosmopolitan species. Records of *S. ellipse* are distributed around the world including the Mediterranean Sea, Atlantic coast of western Europe (Great Britain, France, Scandinavia), South America (Falkland Island, Tierra de Fuego), South Africa (Cape Town) (Prudhoe 1985) and Antarctica (Hallez 1905). Our samples represent the first record for the Iberian Peninsula.

***Oligocladus sanguinolentus* (Quatrefage, 1845) Lang, 1884**

[http://species-id.net/wiki/Oligocladus\\_sanguinolentus](http://species-id.net/wiki/Oligocladus_sanguinolentus)

Figure 7D, E

**Remarks.** *Oligocladus sanguinolentus* belongs to the family Euryleptidae and is characterized by an elongate body with a translucent appearance, thus making the intestinal contents visible and giving the animal a bloodied appearance. The marginal tentacles are long, narrow and clearly separated. The tentacular eyes scattered at the base of the tentacles; and the cerebral eyes in two elongated, diffuse clusters. The oral pore lies in front of the brain. An anal pore (Lang 1884) that opens at the dorsal surface was not observed. Male complex with a penis papilla armed with a tubular, pointed stylet. Female apparatus with multiple uterine vesicles.

**Distribution.** Two specimens were found: one on stones in the outer breakwater of the Yacht Club Ribeira at a depth of 8 metres (42°33.7760N / 008°59.2740W), and the other in “Torre de Abajo” at a depth of 28 metres (42°32.7150N / 008°57.0950W).

*Oligocladus sanguinolentus* is known from the coastal and littoral shores of Saint Malo (France), Isle of Man, Scilly Islands, various sites of Great Britain (United Kingdom), Gibstad (Norway) and Porto Grande de Sao Vicente (Cape Verde archipelago) (Faubel and Warwick 2005). The only record for the Mediterranean Sea is in the Gulf of Naples (Italy) (Lang 1884).



**Family PSEUDOCEROTIDAE Lang, 1884*****Thysanozoon brocchii* (Risso, 1818) Grube, 1840**

[http://species-id.net/wiki/Thysanozoon\\_brocchii](http://species-id.net/wiki/Thysanozoon_brocchii)

Figure 7F, G

**Remarks.** *Thysanozoon brocchii* belongs to the family Pseudocerotidae and is characterized by an oval, oblong body shape; marginal tentacles with eyes; a single pair of eye clusters which anastomose frontally (Figure 7F). The dorsal surface covered with characteristic acorn-like papillae, which may house small intestinal branches (Figure 7G). Reproductive system with the characteristics of the genus: i.e. a paired male apparatus with a seminal vesicle and armed penis papillae, a prostatic vesicle oriented antero- or medio-dorsally to the ejaculatory duct and a female apparatus with branched uteri.

**Distribution.** One specimen of *Thysanozoon brocchii* was found under a rock in the estuary of Muros in Punta Insuela (Galicia, Spain) (10/05/2010) at a depth of 5 metres (42°46.8550N / 009°00.2610W). *T. brocchii* is considered to be cosmopolitan with records from the Atlantic Ocean in South America, Peninsula Valdes (Brusa et al. 2009), Mar del Plata (Bulnes et al. 2011), Ilha de Sao Sebastiao, Ilha das Palmas; North America (Florida), Cape Verde Islands and South Africa; it is also recorded from the Pacific Ocean in Japan and Hawaii, and from the Mediterranean and Adriatic seas, including the Suez Canal (Prudhoe 1985). For the Atlantic coast of the Iberian Peninsula, our collections represent the first record.

**General discussion**

Previous polyclad studies of the North Atlantic Ocean coasts (Bock 1913, Prudhoe 1985, Faubel and Warwick 2005) suggested that the Iberian Peninsula may have a great diversity of species belonging to this group of flatworms, however the actual level of diversity in this region was unknown. This study describes, for the first time, species of the Order Polycladida from the Ibero-Atlantic environment. These records reconfirm the broad geographic range for some polyclad species like *Thysanozoon brocchii*, *Stylostomum ellipse*, *Prosthiostomum siphunculus* or *Oligocladus sanguinolentus*. The species collected can be divided into two groups according to their distribution.

Group 1: includes species with a cosmopolitan distribution that are known from the Atlantic Ocean and the Mediterranean Sea: *Prosthiostomum siphunculus*, *Thysanozoon brocchii* and *Stylostomum ellipse*.

Group 2: includes species with a distribution restricted to the European, North Atlantic and/or Mediterranean coasts and includes *Eurylepta cornuta*, *Oligocladus sanguinolentus*, *Prostheceraeus vittatus*, *P. roseus* and *P. moseleyi*. The two last-mentioned species had previously been known only from the Mediterranean Sea and are here reported for the first time from the Atlantic Ocean.

*Euryleptodes galikias* sp. n., is new to science, and its distribution is currently limited to the Galician Atlantic coast of the Iberian Peninsula.

In summary and based on the results, the distribution range of described polyclads (hitherto limited to the North Atlantic or the Mediterranean basin) has been expanded to the Atlantic shores of the Iberian Peninsula (Lusitanian Region).

## Acknowledgements

This study was supported by I+D Project grants CGL 2010–15786/BOS and CGL2011–29916, which are financed by the Spanish Ministry of Economy, and by the Spanish Agency for International Cooperation (AECI Project ref.: A/030038/10). We are grateful to Melinda Modrell for carefully linguistic review of the English text and deeply in debt to Magdalena and Bernhard Egger for their kindness and help in improving the text in general and English in particular. Thanks are due to the two anonymous reviewers for helpful comments and suggestions which substantially improved the manuscript.

## References

- Bock S (1913) Studien über Polycladen. Zoologiska bidrag fran Uppsala 2: 31–344.
- Brusa F, Damborenea C, Quiroga S (2009) First records of Pseudocerotidae (Platyhelminthes: Polycladida: Cotylea) from Patagonia, Argentina. Zootaxa 2283: 51–59
- Bulnes VN, Albano MJ, Obenat SM, Cazzaniga NJ (2011) Three Pseudocerotidae species (Platyhelminthes, Polycladida, Cotylea) from the Argentinean coast. Zootaxa 2990: 30–44.
- Dalyell JP (1853) Observations on some interesting phenomena in animal physiology, exhibited by several species of planaria. Archibald Constable, Edinburgh.
- Delle Chiaie S (1822–1829) Memorie sulla storia e notomia degli animali senza vertebre del regno di Napoli. Atlas of 109 Tables (1822); Vol. I (1823): 1–184; Vol. II (1825) 1–444; Vol. III (1828) 1–232; Vol. IV (1829) 1–214. Fratelli Fernandes, Napoli.
- Ehrenberg CG (1831) Phytozoa Turbellaria africana et asiatica. In: Hemprich PC, Ehrenberg CG (Eds) “Symbolae physicae.” Animalia evertebrata exclusis insectis recensuit Dr. CG Ehrenberg. Series prima cum tabularum decade prima. Berlin.
- Faubel A (1983) The Polycladida, Turbellaria; Proposal and establishment of a new system. Part I. The Acotylea. Mitteilungen aus dem Hamburgischen Zoologischen Museum und Institut 80: 17–121.
- Faubel A (1984) The Polycladida, Turbellaria; Proposal and establishment of a new system. Part II. The Cotylea. Mitteilungen aus dem Hamburgischen Zoologischen Museum und Institut 81: 189–259.
- Faubel A, Warwick RM (2005) The marine flora and fauna of the Isles of Scilly: free-living Plathelminthes (“Turbellaria”). Journal of Natural History 39: 1–47. doi: 10.1080/00222930310001613593

- Gamble FW (1893) The Turbellaria of Plymouth Sound and the neighbourhood. *Journal of the Marine Biological Associations (N S)* 3(1): 18, 30–47
- Grube AE (1840) Actinien, Echinodermen und Würmer des Adriatischen und Mittelmeeres nach eignen Sammlungen beschrieben. *Königsberg* 4: 51–56.
- Hallez P (1905) Note préliminaire sur les Polyclades recueillis dans l'Expédition antarctique du 'Français'. *Bulletin de la Société Zoologique de France, Année 1905*, 30. Paris 1905, 124–127.
- Heath H, McGregor EA (1912) New polyclads from Monterey Bay, California. *Proceedings of the Academy of Natural Sciences of Philadelphia* 64: 455–488.
- Hyman LH (1940) The polyclad flatworms of the Atlantic Coast of the United States and Canada. *Proceedings of the United States National Museum* 89: 449–492. doi: 10.5479/si.00963801.89-3101.449
- Hyman LH (1953) The polyclad flatworms of the Pacific coast of North America. *Bulletin of the American Museum of Natural History* 100: 269–391.
- Jensen OS (1878) Turbellaria ad litora Norvegiae occidentalia. *Turbellarier ved Norges Vestkyst*. J W Eided Bogtrykkeri, Bergen, 97 pp.
- Kato K (1937) Polyclads collected in Idu, Japan. *Japanese Journal of Zoology* 7: 211–232.
- Keferstein W (1868) Beiträge zur Anatomie und Entwicklungsgeschichte einiger Seeplanarien von St. Malo. *Abhandlungen der Königlichen Gesellschaft der Wissenschaften zu Göttingen* 14: 1–38, I–III.
- Laidlaw FF (1906) On the marine fauna of the Cape Verde Islands, from collections made in 1904 by Mr C. Crossland. The polyclad Turbellaria. *Proceedings of the Zoological Society of London*, 1906, 705–719.
- Lang A (1884) Die Polycladen (Seeplanarien) des Golfes von Neapel und der angrenzenden Meeresabschnitte. Eine Monographie. *Fauna und Flora des Golfes von Neapel und der angrenzenden Meeresabschnitte*, herausgegeben von der Zoologische Station in Neapel. Engelmann, Leipzig.
- Micoletzky H (1910) Die Turbellarienfauna des Golfes von Triest. *Arbeiten aus dem Zoologischen Institut der Universität Wien* 18: 167–182.
- Montagu G (1815) Description of several new or rare animals principally marine, discovered on the south coast of Devonshire. *Transactions of the Linnean Society of London* 11: 25–26, tab 5, fig 3.
- Müller OF (1776) *Zoologiae danicae prodromus, seu Animalium Daniae et Norvegiae indigenarum characteres, nomina, et synonyma imprimis popularium*. Havniae: typis Hallagerii.
- Novell C (2003) Tesis doctoral: Contribució al coneixement dels turbellaris policlàdides del litoral català. <http://hdl.handle.net/10803/781>
- Prudhoe S (1985) *A monograph on polyclad Turbellaria*. Oxford University Press, New York.
- Quatrefages J-LA de (1845) Études sur les types inférieurs de l'embranchement des annelés: mémoire sur quelques planairées marines appartenant aux genres *Tricelis* (Ehr.), *Polycelis* (Ehr.), *Prosthiostomum* (Nob.), *Proceros* (Nob.), *Eolidiceros* (Nob.), et *Stylochus* (Ehr.). *Annales des sciences naturelles. Zoologie* (1834) 4: 129–184.
- Risso A (1818) Mémoire sur quelques Gastéropodes nouveaux Nudibranches et Tectibranches observés dans la mer de Nice. *Journal de Physique, de Chimie, d'Histoire Naturelle et des Arts* 87: 272–273, 368–376.

- Sars GO (1878) Turbellaria ad litora Norvegiae occidentalia. In: Jensen OS (Ed) Turbellarier ved Norges Vestkyst. Eided Bogtrykkeri, Bergen.
- Schmidtlein R (1880) Vergleichende Übersicht über das Erscheinen grösser pelagischer Thiere und Bemerkungen über Fortpflanzungsverhältnisse einiger Seethiere im Aquarium. Mittheilungen aus der Zoologischen Station zu Neapel 2: 162–175.
- Thompson W (1845) Contributions to the Fauna of Ireland, including descriptions of some apparently new Species of Invertebrata. Ann. and Mag. of Nat. Hist. 15: 308–322. London 1845.
- Vatova A (1928) Compendis della flora e fauna del Mare Adriatico presso Rovigno con la distribuzione geografica delle species bentoniche. Memoria - Comitato talassografico italiano 143: 154–174.

# Two new species of the stonefly genus *Amphinemura* (Insecta, Plecoptera, Nemouridae) from China

Xiao-Yu Ji<sup>1,†</sup>, Yu-Zhou Du<sup>1,‡</sup>, Zhi-Jie Wang<sup>1,2,§</sup>

**1** School of Horticulture and Plant Protection & Institute of Applied Entomology, Yangzhou University, Yangzhou, 225009, China **2** Suqian College, Suqian, Jiangsu, 223800, China

† <http://zoobank.org/8D9CA829-4EE5-421D-B282-1F0A8CB3AAFA>

‡ <http://zoobank.org/1FDB5781-C8A0-4088-8D18-DCD5BB01C548>

§ <http://zoobank.org/DEEC63FC-CCE1-4485-BC5B-2D7F15845328>

Corresponding author: Yu-Zhou Du (yzdu@yzu.edu.cn)

---

Academic editor: M. Gottardo | Received 18 January 2014 | Accepted 31 March 2014 | Published 22 April 2014

---

<http://zoobank.org/F1856766-0CEE-4F38-A285-FEA7E0BF4F20>

---

**Citation:** Ji X-Y, Du Y-Z, Wang Z-J (2014) Two new species of the stonefly genus *Amphinemura* (Insecta, Plecoptera, Nemouridae) from China. ZooKeys 404: 23–30. doi: 10.3897/zookeys.404.7067

---

## Abstract

Two new species of the genus *Amphinemura* Ris from China are described and illustrated, i.e. *A. annulata* Du & Ji, **sp. n.** from Zhejiang, Shanxi, Shaanxi and Guizhou Province, and *A. lingulata* Du & Wang, **sp. n.** from Shaanxi and Sichuan Province. *A. annulata* is similar to *A. tricintusidens* Wang & Zhu in having an apical cavity of the epiproct, but the epiproct ventral sclerite and the median paraproct lobe of the two species are different. *A. lingulata* is related to *A. didyma* Zhu & Yang in having the similar epiproct, but they differ mostly in paraproct median and outer lobes.

## Keywords

*Amphinemura*, Nemouridae, Plecoptera, new species, China

## Introduction

The subfamily Amphinemurinae includes seven genera, i.e. *Amphinemura* Ris, *Indonemoura* Baumann, *Mesonemoura* Baumann, *Protonemura* Kempny, *Sphaeronemoura* Shimizu & Sivec, *Malenka* Ricker and *Tominemoura* Sivec & Stark, and the first five genera were

found in China. The genus *Amphinemura* is the largest genus of Amphinemurinae with more than 170 species from the Oriental and Holarctic Regions (Baumann 1975; DeWalt et al. 2013). The *Amphinemura* in China is represented by at least 70 species (Du et al. 2007; Du and Wang 2007; Li et al. 2005; Li and Yang 2005, 2006, 2007, 2008a, b, c, d, e, 2011, 2013; Wang et al. 2006; Wang et al. 2007; Wu 1926, 1935, 1938, 1949, 1962, 1973; Yang et al. 2005; Yang et al. 2005 and Zhu and Yang 2002, 2003). Herein we describe two new Chinese species of *Amphinemura* based on male specimens.

## Materials and methods

All type specimens are preserved in 75% or 99% ethanol and are deposited at the School of Horticulture and Plant Protection & Institute of Applied Entomology, Yangzhou University, China. Specimens were examined and illustrated using a Leica stereomicroscope-MZAPO. Abdomens were cut from the bodies, then treated in 5% NaOH, slowly heated to 40–50 °C for 1–3 minutes, and then the specimens were cleared rinsing in clean water. The morphological terminology follows that of Baumann (1975).

## Results

### *Amphinemura annulata* Du & Ji, sp. n.

<http://zoobank.org/F99B5974-7245-4B96-A61C-147FA0A25B4E>

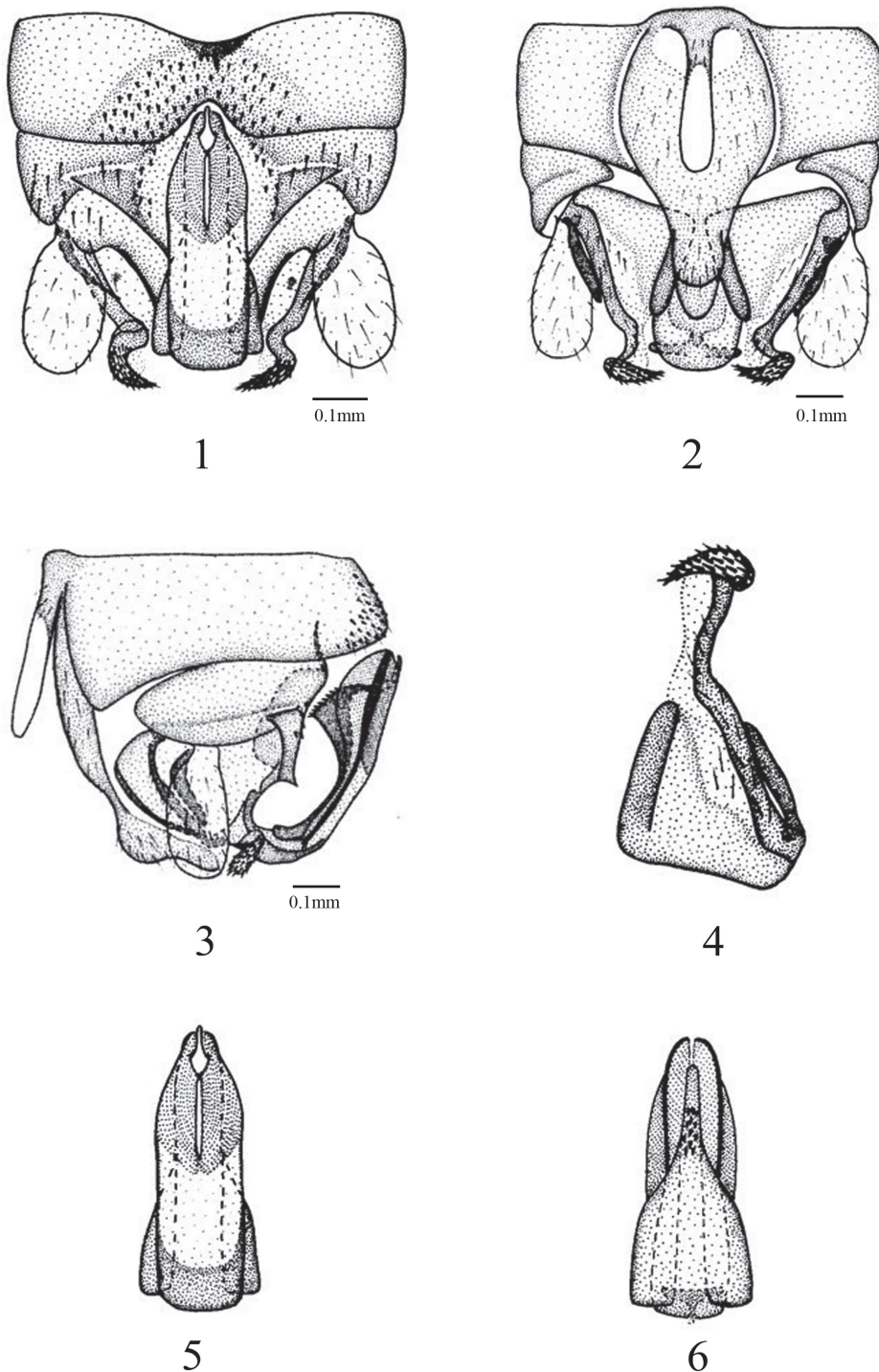
[http://species-id.net/wiki/Amphinemura\\_annulata](http://species-id.net/wiki/Amphinemura_annulata)

Figs 1–6

**Material examined.** Holotype ♂ from China, Zhejiang Province, Mt. Tianmu, 300m, 18–20 Mar. 2006, leg. Wang Zhi-Jie, Sun Yun. Paratypes 30♂♂, the same details as holotype; 1♂, Shanxi Province, Lishan Natural Reserve, 1222m, 24 Aug. 2012, leg. Shi Fu-Ming; 1♂, Shaanxi Province, Houzhenzi, Upriver of Hougou, 26 May 1995, leg. Du Yu-Zhou; 1♂, Shaanxi Province, Foping County, East River, 1240m, 25 Sept. 1996, leg. Xing Lian-Xi; 1♂, Guizhou Province, Maolan City, Dongkuang, 22 Oct. 1996, leg. Li Zi-Zhong.

**Adult habits.** Head and antennae brown, pronotum light brown, subquadrate, angles bluntly rounded, anterior margin wider than posterior margin, with lightly rugosities. Legs brown. Wings hyaline, light brown, veins brown.

**Male.** Forewing length 7.7–8.2 mm, hind wing length 6.6–6.8 mm. Tergum 9 weakly sclerotized, with a small mid-posterior indentation, and bearing a large bundle of tiny spines medially. Tergum 10 weakly sclerotized, with a rounded concavity below epiproct, bearing several spines on lateral margin, and a small triangular projections extruding from the base of the concavity, which is more distinct in lateral view (Figs 1 and 3). Hypoproct narrow basally, extending at midpoint and tapering with a blunt rounded tip, bulging before apex, which is more distinct in lateral view; vesicle slender, four times longer than



**Figures 1–6.** *Amphinemura annulata* male structures. **1** terminalia, dorsal aspect **2** terminalia, ventral aspect **3** terminalia, lateral aspect **4** paraproct (left) **5** epiproct, dorsal aspect **6** epiproct, ventral aspect.



wide (Figs 2 and 3). Paraproct divided into 3 lobes; inner lobe weakly sclerotized, thin and long, about half length of median lobe; median lobe broad basally, mostly membranous, with a long sclerotized band in its outer margin, apically curved inwards and forming an annular projection, bearing many tiny dense spines on the projection; outer lobe sclerotized, short, equal length to inner lobe (Fig. 4). Epiproct elongated, dorsal sclerite sclerotized basally, apical portion weakly sclerotized, taking up half of the dorsal sclerite, forming a small cavity at tip (Fig. 5); in lateral view, slender, well sclerotized pair of large lobes extending to ventral surface from midpoint (Fig. 3); ventral sclerite forming a triangular keel, which is more distinct in lateral view, bearing several black spines ventrally, tip of the ventral sclerite rising from the tip cavity of dorsal sclerite (Fig. 6).

**Female.** Unknown.

**Etymology.** The Latin “*annulata*” refers to median lobe of paraproct forming an annular projection pointing inwards.

**Diagnosis.** This new species is similar to *A. tricintusidens* Wang & Zhu, 2007 (in Wang et al. 2007). Both species having the dorsal sclerite of the epiproct with an apical cavity, but the new species can be separated from the latter by the form of the ventral sclerite of the epiproct and the median lobe of the paraproct. In *A. annulata* sp. n., the ventral sclerite forming a triangular keel, bearing several black spines ventrally, whereas in *A. tricintusidens*, the ventral sclerite of the epiproct forms two heavily sclerotized sclerites which fused at the apex, each bearing two rows of denticles on the surface of the sclerotized sclerites. The median paraproct lobe of *A. annulata* sp. n. is mostly membranous, its outer margin sclerotized to form a long sclerotized band, and forming an annular projection pointing inwards bearing many tiny dense spines on the projection, In *A. tricintusidens*, the median lobe is sclerotized, tapering medially, with a darkly sclerotized rounded tip at the apex, and 3 or 4 dentations surrounding one side of the tip.

***Amphinemura lingulata* Du & Wang, sp. n.**

<http://zoobank.org/DC92AC76-9A64-482E-887A-4C3D2F594D46>

[http://species-id.net/wiki/Amphinemura\\_lingulata](http://species-id.net/wiki/Amphinemura_lingulata)

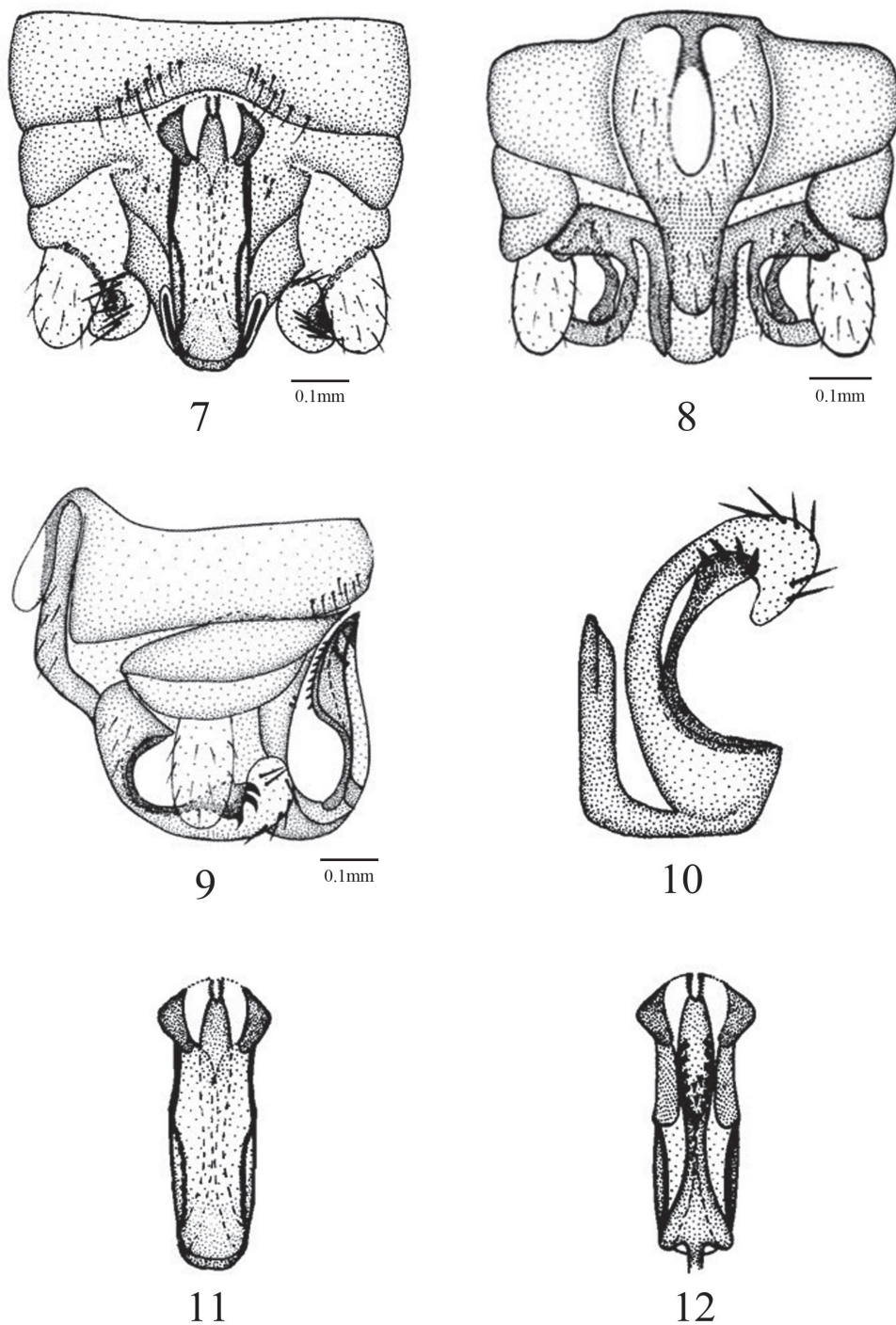
Figs 7–12

**Material examined.** Holotype ♂ China, Shaanxi Province, Houzhenzi, Upriver of Hougou, 26 May 1995, leg. Du Yu-Zhou. Paratypes 6♂♂, Sichuan Province, Laohegou Natural Reserve, 1700m, 25 May 2012, leg. Ji Xiao-Yu, Tang Xiao-Tian; 2♂♂, Shaanxi Province, Qinling Mountain Range, Railway Station of Qinling, 15 May 1995, leg. Wang Min.

**Adult habitus.** Head brown, antennae light brown, pronotum brown, with light rugosities. Legs brown. Wings subhyaline, light brown, veins brown.

**Male.** Forewing length 6.5–6.8 mm, hind wing 5.4–5.8 mm. Tergum 9 weakly sclerotized, bearing a row of long hairs at distal margin. Tergum 10 weakly sclerotized, with a large flat area below the epiproct, with few spines each side of the epiproct (Fig. 7). Hypoproct broad basally and tapering toward blunt rounded tip, vesicle slightly constricted





**Figures 7–12.** *Amphinemura lingulata* male structures. **7** terminalia, dorsal aspect **8** terminalia, ventral aspect **9** terminalia, lateral aspect **10** paraproct (left) **11** epiproct, dorsal aspect **12** epiproct, ventral aspect.

basally, three times longer than wide (Fig. 8). Paraproct divided into 3 lobes; inner lobe weakly sclerotized, slender, with a short darkly sclerotized line medially; median lobe weakly sclerotized basally, bearing several long strong spines on the large membranous and strongly curved tip; outer lobe darkly sclerotized, slender, with 4 or 5 strong spines at the triangular tip (Fig. 10). Epiproct slender in the dorsal aspect; dorsal sclerite side mostly membranous, with a pair of sclerotized small triangular tongue-shaped projections encasing each side of the bifurcated tip (Fig. 11); lateral arms slender, darkly sclerotized, a pair of sclerotized lateral bands convergent apically (Fig. 9); ventral sclerite forming a median keel-shaped ridge, bearing many black spines ventrally (Fig. 12).

**Female.** Unknown.

**Etymology.** The Latin “*lingulata*” refers to the pair of small triangular tongue-shaped projections encasing each side of the tip of epiproct.

**Diagnosis.** This new species is related to *A. didyma* Zhu & Yang (2002) in having the similar median and outer paraproct lobe. However, their epiprocts are markedly different. *A. lingulata* sp. n. is also similar to *A. zhoui* Li & Yang (2008b), *A. helan-shana* Li, Murányi & Yang (2013) and *A. tibetensis* Zhu & Yang (2003) in the epiproct with a pair of sclerotized small projections encasing each side of the tip, but their paraproct lobes are different obviously.

## Concluding remarks

Shimizu (1996), in a PhD study on East Asian Nemouridae, suggested seven species-groups for East Asian species of *Amphinemura*, i.e. the *flavicollis* group, the *clavigera* group, the *spinigera* group, the *sagittata* group, the *flavostigma* group, the *pentagona* group and the *megaloba* group, and arranged some Chinese species in these species-groups. However, only the *flavostigma* group and the *megaloba* group were subsequently published (Shimizu 1998a, b). A revision of the genus *Amphinemura* will be needed considering the high number of species included.

So far 74 species of *Amphinemura*, including the two new species documented in this paper, were recorded in China. It can be expected that more *Amphinemura* species will be found in the future because China is across the Palaearctic and Oriental Region and features not yet faunistically explored areas with suitable resources for stoneflies. Additionally, studies focused on the biogeography, phylogeny and evolution are also needed to further understand the species diversity of *Amphinemura*.

## Acknowledgements

We heartily thank the editor and reviewers for their critical review of an earlier version of this manuscript. We are very indebted to Qian Yu-Han for assistance with this paper. This research was supported by National Natural Science Foundation of China (No. 31071958).

## References

- Baumann RW (1975) Revision of the stonefly family Nemouridae (Plecoptera): A study of the world fauna at the generic level. *Smithsonian Contributions to Zoology* 211: 1–74. doi: 10.5479/si.00810282.211
- DeWalt RE, Neu-Becker U, Stueber G (2013) Plecoptera Species File Online. Version 5.0/5.0. [3.24.2013] <http://Plecoptera.SpeciesFile.org>
- Du Y-Z, Wang Z-J (2007) New species of the genus *Amphinemura* (Plecoptera: Nemouridae) from Yunnan, China. *Zootaxa* 1554: 57–62.
- Du Y-Z, Wang Z-J, Zhou P (2007) Four new species of the genus *Amphinemura* (Plecoptera: Nemouridae) from China. *Aquatic Insects* 29: 297–305. doi: 10.1080/01650420701552482
- Li W-H, Murányi D, Pan J-J, Yang D (2013) New and little known species of Nemouridae (Plecoptera) from Inner Mongolia of China. *Zootaxa* 3746: 473–480. doi: 10.11646/zootaxa.3746.3.6
- Li W-H, Yang D (2005) Two new species of *Amphinemura* (Plecoptera: Nemouridae) from Sichuan, China. *Zootaxa* 1083: 63–68.
- Li W-H, Yang D (2006) Three new species of *Amphinemura* (Plecoptera: Nemouridae) with a key to the species from Guizhou province, China. *Zootaxa* 1154: 41–48.
- Li W-H, Yang D (2007) Review of the genus *Amphinemura* (Plecoptera: Nemouridae) from Guangdong, China. *Zootaxa* 1511: 55–64.
- Li W-H, Yang D (2008a) Species of *Amphinemura* (Plecoptera: Nemouridae) from Tibet, China. *Zootaxa* 1688: 54–60.
- Li W-H, Yang D (2008b) New species of Nemouridae (Plecoptera) from China. *Aquatic Insects* 30: 205–221. doi: 10.1080/01650420802334038
- Li W-H, Yang D (2008c) A new species of *Amphinemura* (Plecoptera: Nemouridae) from China. *Zootaxa* 1892: 65–68.
- Li W-H, Yang D (2008d) Two new species of *Amphinemura* (Plecoptera: Nemouridae) from Yunnan, China, with the redescription of *A. triramia* (Wu, 1962). *Zootaxa* 1926: 61–67.
- Li W-H, Yang D (2008e) Two new species and two new records of stonefly family Nemouridae from Henan (Plecoptera: Nemouroidea). In: Shen X, Lu Ch (Ed) *The Fauna and Taxonomy of Insects in Henan*. China Agricultural Science and Technology Press, Beijing, 6: 11–16.
- Li W-H, Yang D (2011) Two new species of *Amphinemura* (Plecoptera: Nemouridae) from China. *Zootaxa* 2975: 29–34.
- Li W-H, Yang D, Sivec I (2005) A new species of *Amphinemura* (Plecoptera: Nemouridae) from China. *Entomological News* 116: 93–96.
- Shimizu T (1996) A Contribution to the Knowledge of the Family Nemouridae from East Asia (Insecta, Plecoptera). Ph.D thesis. Tokyo University of Agriculture, Tokyo, Japan.
- Shimizu T (1998a) The Group of *Amphinemura flavostigma* (Plecoptera, Nemouridae). *Aquatic Insects* 20: 203–208. doi: 10.1076/aqin.20.4.203.4469
- Shimizu T (1998b) The Group of *Amphinemura megaloba* (Plecoptera, Nemouridae). *Japanese Journal of Systematic Entomology* 42: 227–236.

- Wang Z-J, Du Y-Z, Sivec I, Li Z-Z (2006) Records and descriptions of some Nemouridae species (Order: Plecoptera) from Leigong Mountain, Guizhou province, China. *Illiesia* 2: 50–56.
- Wang Z-J, Zhang J-H, Zhu J-Y (2007) A new species of the genus *Amphinemura* (Plecoptera: Nemouridae) from Xinjiang, China. *Acta Zootaxonomica Sinica* 29: 13–16.
- Wu C.F. (1926) Two new species of stoneflies from Nanking. *The China Journal of Science and Arts* 5: 331–332.
- Wu C-F (1935) New species of stoneflies from East and South China. *Bulletin of the Peking Society of National History* 9: 227–243.
- Wu C-F (1938) *Plecopterorum sinensium*: A monograph of stoneflies of China (Order Plecoptera). Yenching University, 225 pp.
- Wu C.F. (1949) Sixth Supplement to the stoneflies of China (Order Plecoptera). *Bulletin of the Peking Natural History* 17: 251–256.
- Wu C-F (1962) Results of the Zoologico-Botanical expedition to Southwest China, 1955–1957 (Plecoptera). *Acta Entomologica Sinica* 11 (Supplement): 139–153.
- Wu C-F (1973) New species of Chinese stoneflies (Order Plecoptera). *Acta Entomologica Sinica* 16: 97–118.
- Yang D, Li W-H, Sivec I (2005) A new species of *Amphinemura* (Plecoptera: Nemouridae) from China. *Entomological News* 115: 226–228.
- Yang D, Li W-H, Zhu F (2005) A new species of *Amphinemura* from south China (Plecoptera: Nemouridae). *Zootaxa* 805: 1–4.
- Zhu F, Yang D (2002) Three new species of *Amphinemura* from China (Plecoptera: Nemouridae). *Acta Zootaxonomica Sinica* 27: 745–749.
- Zhu F, Yang D (2003) Three new species of *Amphinemura* (Plecoptera: Nemouridae) from Tibet. *Entomologia Sinica* 10: 51–56.

# Revision of the Australasian genus *Pseudidarnes* Girault, 1927 (Hymenoptera, Agaonidae, Sycophaginae)

Fernando Henrique Antonioli Farache<sup>1,†</sup>, Jean-Yves Rasplus<sup>2,‡</sup>

**1** PPG em Entomologia, Depto de Biologia/FFCLRP-USP Ribeirão Preto, SP, Brazil **2** INRA, UMR 1062 CBGP Montferrier-sur-Lez, France

† <http://zoobank.org/EA8F46AF-DCC9-47A0-8525-2AC43B22F063>

‡ <http://zoobank.org/981D4685-5DA3-44AD-B056-CA5877F06329>

Corresponding author: Fernando Henrique Antonioli Farache (fhafarache@gmail.com)

---

Academic editor: M. Engel | Received 11 February 2014 | Accepted 24 March 2014 | Published 22 April 2014

---

<http://zoobank.org/CC398EF9-0438-4889-8B86-5D2336FF2883>

---

**Citation:** Farache FHA, Rasplus J-Y (2014) Revision of the Australasian genus *Pseudidarnes* Girault, 1927 (Hymenoptera, Agaonidae, Sycophaginae). ZooKeys 404: 31–70. doi: 10.3897/zookeys.404.7204

---

## Abstract

The species of *Pseudidarnes* are revised, and six species are described: *P. acaudus* Farache & Rasplus, **sp. n.**; *P. astridae* Farache & Rasplus, **sp. n.**; *P. badiogeminus* Farache & Rasplus, **sp. n.**; *P. cooki* Farache & Rasplus, **sp. n.**; *P. kjellbergi* Farache & Rasplus, **sp. n.**; *P. laevis* Farache & Rasplus, **sp. n.** *Pseudidarnes minerva* Girault, 1927 and *P. flavicollis* Bouček, 1988 are redescribed. A key to the species is provided as well as illustrations for all females and all known males (except the wingless male of *P. minerva*). We also provided further discussion on ecology, morphological patterns, and host taxonomy. Online dichotomous and multi-access interactive LUCID keys to all *Pseudidarnes* species are available at <http://www.figweb.org/>.

## Keywords

Chalcidoidea, *Ficus*, Moraceae, non-pollinating fig wasp, gall maker

## Introduction

Sycophaginae is one of the six subfamilies of chalcid wasps that are strictly associated with *Ficus* syconia. A recent phylogenetic analysis of the Chalcidoidea recovered Agaoninae (fig pollinators) + Sycophaginae as a monophyletic group (Agaonidae),

whereas all other fig wasp subfamilies were included in Pteromalidae (Heraty et al. 2013). Sycophaginae are associated with only two subgenera of fig trees (*Urostigma* and *Sycomor*). Most of the species oviposit in figs (syconia) through the fig wall and are gall inducers or parasitoids of other fig wasps. However, some *Sycophaga* species are capable of entering the figs through a small pore (ostiole), as pollinating fig wasps do (Bouček 1988, 1993, Cruaud et al. 2011a, Cruaud et al. 2011b, Elias et al. 2008, Rasplus and Soldati 2005).

The Sycophaginae occur in all tropical regions of the world but are still poorly known, with about 60 described species and an estimated diversity of about 700 species (Cruaud et al. 2011b). There are five described genera of Sycophaginae, and at least one genus awaits description (Bouček 1988, Cruaud et al. 2011a, Cruaud et al. 2011b). Four of these genera occur in the Australasian region, namely *Pseudidarnes* Girault, 1927, *Eukoebelea* Ashmead, 1904, *Sycophaga* Westwood, 1840, and the undescribed genus (Ashmead 1904, Bouček 1988, Cruaud et al. 2011a, Cruaud et al. 2011b, Westwood 1840).

The phylogeny of the Sycophaginae recovered by the analysis of multiple genes (Cruaud et al. 2011a, Cruaud et al. 2011b) showed that the subfamily is subdivided into three main clades:

- 1) *Eukoebelea* (associated with Australasian *Malvanthera* fig trees) recovered as the sister lineage to all other genera.
- 2) A strongly supported clade of three genera, *Pseudidarnes* (*Malvanthera*) basal to *Anidarnes* Bouček, 1993 (associated with the New World *Americana* fig trees) plus the undescribed genus (associated with the Oriental *Conosycea* figs). A dichotomic key separating these three genera is provided by Farache et al. (2013).
- 3) A well-supported clade composed of two groups: *Sycophaga* (mostly associated with Australasian, oriental and afrotropical *Sycomor* fig trees) and *Idarnes* Walker, 1843 (associated with the New World *Americana* figs).

The biogeography of the subfamily has been discussed by Cruaud et al. (2011a). *Pseudidarnes* is only known from the Australasian region and includes two described species: *P. minerva* Girault, 1927 from Australia, associated with *Ficus rubiginosa* (Girault 1927), and *P. flavicollis* Bouček, 1988, described from Papua New Guinea and reared from *Ficus xylosyia* (Bouček 1988), but never collected again. *Pseudidarnes* species are strictly associated with syconia of *Ficus* section *Malvanthera* Corner (subg. *Urostigma*). This section includes 23 fig species mostly occurring in Australia and New Guinea (incl. Bismarck Archipelago), reaching Sulawesi, the Vanuatu islands and New Caledonia (Dixon 2003, Rønsted et al. 2008). *Malvanthera* fig trees are pollinated by *Pleistodontes* Saunders wasps (Lopez-Vaamonde et al. 2002).

Among the guilds that constitute fig wasp communities, *Pseudidarnes* species belong to the “large gall inducers” (Cruaud et al. 2011b, Segar and Cook 2012, Segar et al. 2013). The non-pollinating fig wasps belonging to this guild are larger than the co-occurring pollinators, exhibit short and thick ovipositors, and oviposit through the



syconium wall during the early development of the syconia before pollination (Cruaud et al. 2011b, Elias et al. 2008). Induction of large galls is usually correlated with small brood sizes, and their galls may occupy the whole syconium lumen (Bronstein 1999; Cook et al. 1997, West and Herre 1998). As other large gall inducers, *Pseudidarnes* species are usually rare in most host species and found in low abundance ( $0.1 \pm 0.5$ , mean  $\pm SE$ , insects per fig in *F. obliqua*) (Segar and Cook 2012).

*Pseudidarnes* males are usually winged, but wingless males of *P. minerva* were recorded with very low frequencies (Cook et al. 1997, Early 2000). The small wingless males mate with females inside the figs even before females leave their galls, while winged males usually (but not always) disperse before mating (Cook et al. 1997).

In this paper we describe and illustrate six previously unknown species (two from Australia and four from Papua New Guinea). Redescriptions are also provided for *P. minerva* and *P. flavicollis*. We finally elaborated both dichotomous and interactive on-line keys to the known species of *Pseudidarnes*.

## Methods

### Specimen sampling and morphological study

Maturing fig syconia were collected, opened, and transferred to tissue bags until the wasps emerge, which happens after a few hours - days. Wasps were killed using acetate and transferred to 70% ethanol. Most geographical coordinates and altitudes were estimated using label information. Field recorded coordinates were provided when available. Field-collected specimens were dehydrated through an ethanol and HMDS series (Heraty and Hawks 1998) and then mounted on cards following Noyes (1982). Morphological terminology follows Gibson (1997). Material examined sections of species descriptions were prepared using AUTOMATEX (Brown 2013), and posteriorly refined.

Type and specimen depositories, and their respective curators are:

- ASCI**     Australia, New South Wales, Orange, Orange Agricultural Institute, Agricultural Scientific Collections Unit (Peter Gillespie).
- BMNH**     United Kingdom, London, The Natural History Museum [formerly British Museum (Natural History)] (Natalie Dale-Skey Papilloud).
- CBGP**     France, Montpellier. Centre de Biologie pour la Gestion des Populations (Emmanuelle Artige).
- SAMC**     South Africa, Cape Town, Iziko South African Museum (Simon Van Noort).

### Illustration

To produce high quality images, some specimens were point-mounted on grey card in order to avoid loss of contrast caused by white background. Images were produced

with an EntoVision Premium Portable Imaging System, comprising a Leica M16 zoom lens, a JVC KY-75U 3CCD digital camera and a portable computer workstation running EntoVision Imaging Suite software (GT Vision, Hagerstown, MD U.S.A.). Cartograph v5.6.0 (Microvision, Evry, France) software was subsequently used to merge an image series (representing about ten to twenty focal planes), producing a single image with increased depth of field. Illumination was achieved using a “quadrant” setup, with four fibre optic light guides stemming from two individual light sources (Leica CLS 150 X), similar to the one described by Buffington and Gates (2008). Images were edited using Adobe Photoshop CS4© software.

## Results

### *Pseudidarnes* Girault, 1927

<http://species-id.net/wiki/Pseudidarnes>

[http://www.figweb.org/Fig\\_wasps/Agaonidae/Sycophaginae/Pseudidarnes/index.htm](http://www.figweb.org/Fig_wasps/Agaonidae/Sycophaginae/Pseudidarnes/index.htm)

**Type species.** *Pseudidarnes minerva* Girault, 1927, by monotypy.

**Diagnosis.** *Female*. Body length (excluding ovipositor) 2.3–3.7 mm. Body colour variable, yellow to dark brown, sometimes with green metallic tinge.

*Head*. Face sculpture smooth to reticulate or slightly engraved, sometimes punctate. Antennae inserted well above to slightly below the middle line of compound eyes, but never very close to the clypeal margin. Toruli separated by one torulus diameter or less. Clypeal margin bilobed. Maxillary palpi composed of four segments. Labial palpi composed of three segments. Supraclypeal area delimited by subantennal grooves. Antenna with 13 segments (two anelli), and a 14<sup>th</sup> segment very short and un conspicuous. Funicular segments 1–2 × as long as wide.

*Mesosoma*. Pronotum 1–2 × as long as high in lateral view. Notauli complete, deep and at least slightly crenulated. Mesoscutum as long as wide or longer than wide. Axilular sulcus straight to slightly concave. Propodeum transverse, smooth to slightly reticulate and sometimes with a median sulcus. Wings hyaline, sometimes slightly infusate medially. Marginal vein as long as stigmal vein, or longer. Postmarginal vein present (very short in *P. cooki* sp. n.). Marginal and postmarginal vein sometimes particularly widened (as Figs 4D, 6D, 8D, 10D & 16D).

*Metasoma*. First metasomal segment petiolate (petiole short in *P. acaudus* sp. n.). Ovipositor sheaths as long as body or shorter (very short in *P. acaudus* sp. n.).

*Male*. Very similar to female, usually slender and sometimes showing tinge variation. Male with very different colour patterns in *P. astridae* sp. n. (Figs 3–6).

### Key to species of *Pseudidarnes* Girault (based mainly on females)

Online dichotomous and multi-access interactive LUCID keys to *Pseudidarnes* species are available at: [http://www.figweb.org/Fig\\_wasps/Agaonidae/Keys/index.htm](http://www.figweb.org/Fig_wasps/Agaonidae/Keys/index.htm)



- 1 Ovipositor sheaths extremely short, only weakly protruding beyond metasoma apex (Figure 1A). Pedicel elongated, slightly shorter than the scape (Fig. 1D). Mesosoma entirely brown (Fig. 2A) ..... ***P. acaudus* sp. n.**
- Ovipositor sheaths long, distinctly protruding beyond metasoma apex. Pedicel clearly shorter than the scape, at most  $0.5 \times$  the scape length. Mesosoma colour different, metallic or, when brown, with at least the pronotum yellow in lateral view ..... **2**
- 2 Pronotum yellow, without metallic tinge. Mesoscutum with irregular transverse striae (Fig. 4A, 8A, 16A). Marginal and postmarginal veins widened (Figs 4F, 8F, 16F) ..... **3**
- Mesosoma metallic green, including pronotum. Mesoscutum smooth or reticulated. Marginal and postmarginal veins not widened ..... **5**
- 3 Head and mesosoma excluding pronotum with metallic tinge (Figs 15B, E, 16A). Propodeum with a well delimited and carinulated median sulcus, extending to the posterior margin of the sclerite (Fig. 16B). Metascutellum crenulated (Fig. 16B) ..... ***P. flavicollis* Bouček**
- Body without metallic tinge. Median sulcus of propodeum un conspicuous or absent. Metascutellum not crenulated ..... **4**
- 4 Mesosoma brown in dorsal view (Fig. 8A). Metascutellum with faint longitudinal striae (Figs 8B, 10B). Propodeum without median line (Figs 8B, 10B) ..... ***P. badiogeminus* sp. n.**
- Mesosoma yellow in dorsal view (Fig. 4A, brown in males, but at least mesoscutellum yellow, Fig. 6A). Metascutellum and median area of propodeum with irregular transverse rugae (Figs 4B, 6B) ..... ***P. astridae* sp. n.**
- 5 Mesosoma entirely smooth and shiny (Figs 19B, 20A, 21B, 22A). First funicular segment  $2 \times$  as long as wide (Figs 19D, 21D). Distal antennomeres not forming a definite clava (Figs 19C, 21C). Propodeum with a very short median line (Figs 20B, 22B) ..... ***P. laevis* sp. n.**
- Mesosoma sculpture mostly reticulate. First funicular segment ca.  $1\text{--}1.5 \times$  as long as wide. Distal antennomeres forming a definite clava. Propodeum medially with a deep carinulated sulcus, at least on the anterior half of the sclerite ..... **6**
- 6 Ovipositor sheaths short, about as long as the metasoma (Fig. 11A). Propodeum with a crenulated median sulcus extending to the posterior margin (Figs 12B, 14B) ..... ***P. cooki* sp. n.**
- Ovipositor sheaths longer than metasoma. Propodeal median sulcus not reaching the posterior margin ..... **7**
- 7 Petiole transverse in dorsal view (Fig. 18D). Median sulcus of the propodeum broad and extending over the anterior half of the sclerite (Fig. 18B). Postmarginal vein shorter than the stigmal (Fig. 18F) ..... ***P. kjellbergi* sp. n.**
- Petiole longer than wide in dorsal view (Figs 24D, 26D). Median sulcus of the propodeum extending over most of the propodeum length, not reaching the posterior margin (Figs 24B, 26B). Postmarginal vein as long as the stigmal (Figs 24F, 26F) ..... ***P. minerva* Girault**

## Species descriptions

### *Pseudidarnes acaudus* Farache & Rasplus, sp. n.

<http://zoobank.org/ECCB10FF-4783-45C9-A97E-02E1E27600CF>

[http://species-id.net/wiki/Pseudidarnes\\_acaudus](http://species-id.net/wiki/Pseudidarnes_acaudus)

Figures 1–2

**Material examined.** *Holotype*. ♀, **PAPUA NEW GUINEA:** Crater Mountain,  $-6.58^{\circ}$ ,  $145.08^{\circ}$ , 2000m, V.1990, McKee A., ex *Ficus* sp. (CBGP).

*Paratype*. ♀, same data as holotype, (CBGP).

**Diagnosis.** Pedicel elongated, slightly shorter than the scape. Mesosoma entirely brown. Petiole short, transverse in dorsal view. Ovipositor sheaths extremely short, only weakly protruding beyond metasoma apex.

**Description.** *Female*. Body length 2.6 mm. Metallic tinge absent or very feeble. Predominantly brown. Scape and pedicel yellow brown. Head darker than mesosoma. Petiole yellow. Legs predominantly yellow, coxae almost concolorous with mesosoma. Remaining leg segments predominantly yellow and brown.

*Head.* Antennae inserted far above the middle line of compound eyes. Scape slightly longer than pedicel. Pedicel very elongated (more than  $2 \times$  as long as wide), slender, and longer than first funicular segment. Anelli almost as long as wide, proximal anellus longer than wide. First funicular segment approximately  $1.5 \times$  as long as wide. Distal antennomeres not forming a distinct clava. Face sculpture slightly engraved. Face pilosity short and sparse. Supraclypeal area narrow, its delimiting sulci converging near epistomal groove, and its sculpture barely rugose. Lateral ocelli nearly  $1 \times$  its diameter far from the eye margin.

*Mesosoma.* Pronotum longer than high in lateral view. Mesoscutum slightly engraved reticulate. Frenal sulcus smooth. Mesepimeron sculpture mostly smooth, slightly engraved. Metascutellum very short and smooth, inconspicuous, and almost completely covered by frenalium. Propodeum with a well delimited and slightly carinated median sulcus, which extends to the posterior margin of the sclerite. Propodeum sculpture smooth, slightly rugose. Wings hyaline, with sparse pilosity. Marginal and postmarginal vein not particularly widened. Postmarginal vein as long as stigmal vein.

*Metasoma.* Petiole smooth and transverse in lateral view. Petiole dorsally without a longitudinal median sulcus. Ovipositor sheaths extremely short, only weakly protruding beyond metasomal apex.

*Male.* Unknown

**Etymology.** The specific name refers to the short ovipositor sheaths exhibited by this species.

**Biology.** Associated with an undetermined *Ficus* species collected in Papua New Guinea. Reared together with *Pseudidarnes laevis* sp. n., but less abundant than the later.



**Figure 1.** *Pseudidarnes acaudus* sp. n. female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

***Pseudidarnes astridae* Farache & Rasplus, sp. n.**

<http://zoobank.org/FBBF3AC7-E935-40E8-AF15-FA87970689EC>

[http://species-id.net/wiki/Pseudidarnes\\_astridae](http://species-id.net/wiki/Pseudidarnes_astridae)

Figures 3–6

**Material examined.** *Holotype*. ♀, **PAPUA NEW GUINEA: East New Britain:** Raunsepna, North Baining Mountains,  $-4.433^{\circ}$ ,  $151.783^{\circ}$ , 1000m, 26.II.1999, Vaamonde CL, ex *F. xylosyia* CLV11 (CBGP).





**Figure 2.** *Pseudidarnes acaudus* sp. n. female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

*Paratype.* ♂ Same data as holotype (CBGP).

**Diagnosis.** Body without metallic tinge. Pronotum long, nearly  $1.5\text{--}2\times$  as long as high in lateral view. Mesoscutum with faint irregular transverse striae. Median area of metascutellum and median area of propodeum with irregular transverse rugae. Marginal and postmarginal vein widened.

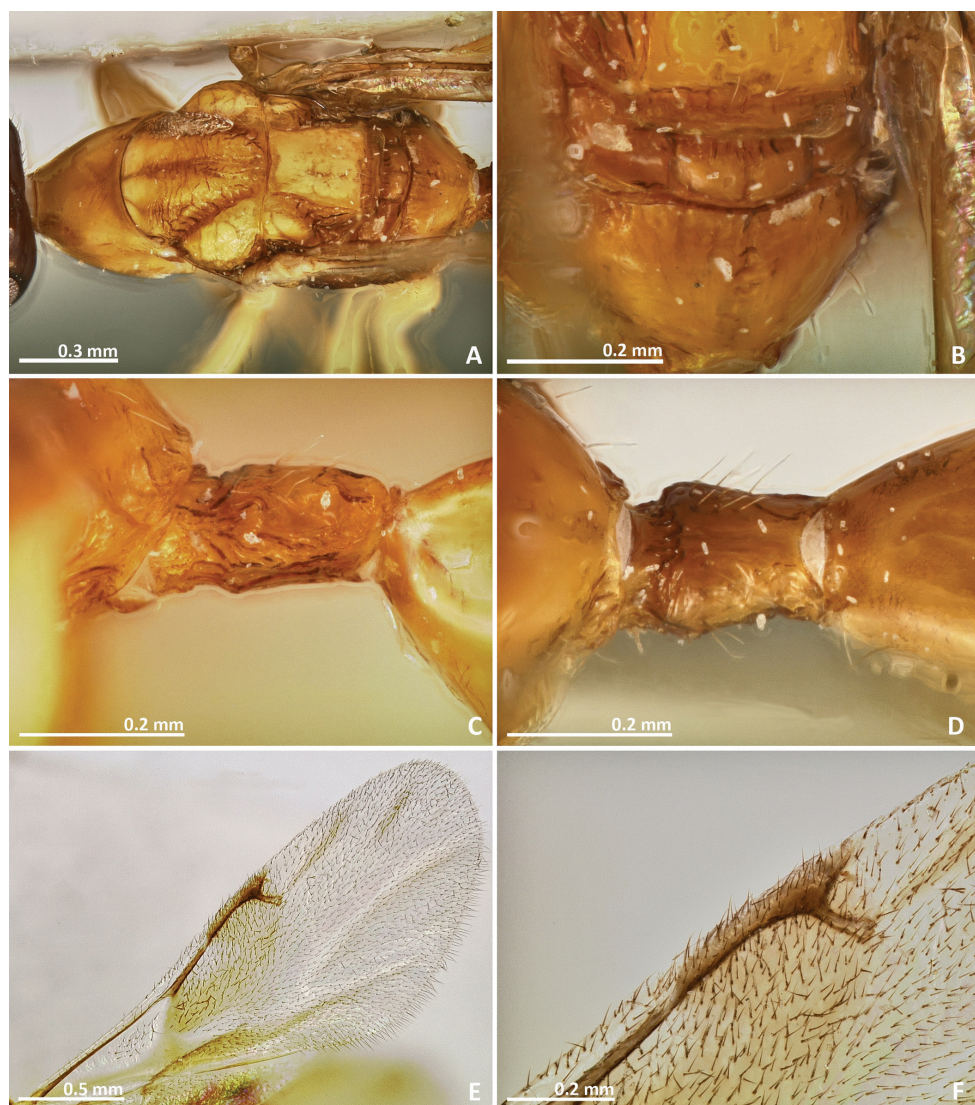
**Description.** *Female.* Body length 3.4 mm. Ovipositor sheaths length 1.8 mm. Metallic tinge absent. Predominantly yellow. Head dark brown. Mesepisternum, mesepimeron, and metapleuron predominantly brown.



**Figure 3.** *Pseudidarnes astridae* sp. n. female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

*Head.* Antennae inserted far above the middle line of compound eyes. Scape more than  $2 \times$  as long as pedicel. Pedicel elongated, slender, but shorter than first funicular segment. Anelli almost as long as wide. First funicular segment  $2 \times$  as long as wide. Distal antennomeres not forming a distinct clava. Face sculpture smooth with sparse punctures, lower face slightly engraved. Face pilosity long and dense. Supraclypeal area wide, its delimiting sulci not converging near epistomal groove, and its sculpture mostly smooth. Lateral ocelli  $0.5 \times$  its own diameter far from the eye margin.





**Figure 4.** *Pseudidarnes astridae* sp. n. female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

*Mesosoma.* Pronotum long, nearly  $1.5\text{--}2\times$  as long as high in lateral view. Mesoscutum with faint transversal striae. Mesoscutellum smooth. Frenal sulcus sparsely crenulated. Mesepimeron sculpture mostly smooth, slightly striate. Metascutellum longer than frenum and smooth, with faint and irregular transverse rugae at the median line. Propodeum sculpture mostly smooth. Median line of propodeum with irregular transverse rugae. Wings with rather dense pilosity, and medially infusate. Marginal and postmarginal vein widened. Postmarginal vein shorter than stigmal vein.



**Figure 5.** *Pseudidarnes astridae* sp. n. male. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

*Metasoma.* Petiole rugose,  $1.7 \times$  as long as high in lateral view. Petiole dorsally without a longitudinal median sulcus. Ovipositor sheaths long, distinctly protruding beyond metasoma apex. Ovipositor sheaths length  $2.8 \times$  hind tibia length and  $0.5 \times$  body length.

*Male.* Body length 4.2 mm. Characters agreeing with females, except the following. Body colour browner, mesoscutellum yellow in dorsal view. Posterior ocelli contiguous to the eye margin, and larger. Wing infuscation more pronounced.





**Figure 6.** *Pseudidarnes astridae* sp. n. male. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

**Etymology.** The specific name is dedicated to our friend Astrid Cruaud for the long long walks we share together in the jungles of the world, trying to find fig trees.

**Biology.** Reared from syconia of *Ficus xylosyca* Diels. *Ficus xylosyca* hosted three *Pseudidarnes* species. *Pseudidarnes badiogeminus* sp. n. was collected together with *P. astridae* sp. n. in New Britain, whereas *P. flavicollis* Bouček was collected in Bulolo. We are convinced that the host identification is correct for both samplings and that the guilds of non-pollinating fig wasps may vary with geography.



**Figure 7.** *Pseudidarnes badiogeminus* sp. n. female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

***Pseudidarnes badiogeminus* Farache & Rasplus, sp. n.**

<http://zoobank.org/0EA058A5-E760-4114-8CBD-72DF1D488803>

[http://species-id.net/wiki/Pseudidarnes\\_badiogeminus](http://species-id.net/wiki/Pseudidarnes_badiogeminus)

Figures 7–10

**Material examined.** *Holotype*. ♀, **PAPUA NEW GUINEA: East New Britain:** Raunsepna, North Baining Mountains,  $-4.433^{\circ}$ ,  $151.783^{\circ}$ , 1000m, 26.II.1999, Vaamonde CL, ex *F. xylosyia* CLV11 (CBGP).





**Figure 8.** *Pseudidarnes badiogeminus* sp. n. female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

*Paratype.* 1♀ 1♂ same data as holotype (CBGP).

**Diagnosis.** Pronotum long, nearly  $1.5\text{--}2 \times$  as long as high in lateral view. Mesosoma brown in dorsal view. Mesoscutum with irregular transverse rugae. Metascutellum with faint longitudinal striae. Marginal and postmarginal veins widened. Median line of propodeum absent

**Description.** *Female.* Body length 3 mm. Ovipositor sheaths length 1.6 mm. Metallic tinge absent. Body colour predominantly brown. Scape and pedicel yellow.



**Figure 9.** *Pseudidarnes badiogeminus* sp. n. male. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

Flagellomeres yellow brown. Head dark brown. Pronotum yellow brown laterally. Legs yellow, coxae browner.

*Head.* Antennae inserted far above the middle line of compound eyes. Scape nearly  $2 \times$  as long as pedicel. Pedicel elongated, slender, and as long as first funicular segment. Proximal anellus longer than wide. First funicular segment  $2 \times$  as long as wide. Distal antennomeres not forming a distinct clava. Face sculpture engraved, slightly rugose. Face pilosity short and sparse. Supraclypeal area narrow, its delimiting sulci converging





**Figure 10.** *Pseudidarnes badiogeminus* sp. n. male. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

near epistomal groove, and its sculpture barely rugose. Lateral ocelli  $0.5 \times$  its own diameter far from the eye margin.

**Mesosoma.** Pronotum long, nearly  $1.5\text{--}2 \times$  as long as high in lateral view. Mesoscutum transversally striate. Mesoscutellum mostly smooth. Frenal sulcus with shallow crenulation. Mesepimeron sculpture mostly smooth, slightly striate. Metascutellum longer than frenum, with faint longitudinal striae. Propodeum smooth, without median line. Wings with rather dense pilosity, and medially infusate. Marginal and postmarginal vein widened. Postmarginal vein longer than stigmal vein.

*Metasoma*. Petiole slightly rugose,  $1.7 \times$  as long as high in lateral view. Petiole dorsally without a longitudinal median sulcus. Ovipositor sheaths long, distinctly protruding beyond metasoma apex. Ovipositor sheaths length  $2.7 \times$  hind tibia length and  $0.5 \times$  body length.

*Male*. Body length 3.2 mm. Characters agreeing with females, except the following: ocelli larger and contiguous to the eye margin. Pedicel slightly shorter.

**Etymology.** The specific name refers to the similarity of *P. badiogeminus* with *P. astridae*, but showing different colour.

**Biology.** Collected from syconia of *F. xylosyca*. See *P. astridae* for further information.

***Pseudidarnes cooki* Farache & Rasplus, sp. n.**

<http://zoobank.org/2C54B54B-5925-45BD-A1CA-1440704749C8>

[http://species-id.net/wiki/Pseudidarnes\\_cooki](http://species-id.net/wiki/Pseudidarnes_cooki)

Figures 11–14

*Pseudidarnes* sp. ex *F. obliqua*; Cruaud et al. (2011a) BMC Evolutionary Biology, 11: 15pp. [phylogenetic position]

*Pseudidarnes* sp. ex *F. obliqua*; Cruaud et al. (2011b) Journal of Biogeography, 38: 209–225. [biogeography]

*Pseudidarnes* sp.; Segar ST, Cook JM (2012) Ecological Entomology, 37(5), 342–349. [ecology]

**Material examined.** *Holotype*. ♀, **AUSTRALIA: Queensland:** Cairns, Rex Lookout,  $-16.65^{\circ}$ ,  $145.56^{\circ}$ , 100m, 13.I.1999, Rasplus J.Y., ex *F. obliqua* (CBGP).

*Paratypes*. **AUSTRALIA: Queensland:** Cairns, Rex Lookout,  $-16.65^{\circ}$ ,  $145.56^{\circ}$ , 100m, 7♀, 1♂, 13.I.1999, Rasplus J.Y., ex *F. obliqua* (CBGP), North of Cairns, Coastal road,  $-16.65^{\circ}$ ,  $145.56^{\circ}$ , 100m, 1♀, 27.X.2005, Jousselin E. & Coeur d'Acier A., ex *F. obliqua*, n° JRAS01422 (CBGP), Port Douglas,  $-16.483230^{\circ}$ ,  $145.464058^{\circ}$ , 10m, 3♀, 28.X.2005, Jousselin E. & Coeur d'Acier A., ex *F. obliqua*, n° JRAS01429 (1 ♀ CBGP, 1 ♀ BMNH, 1 ♀ SAMC).

**Diagnosis.** Metallic tinge present at least in some body regions. Mesosoma sculpture mostly reticulate. Propodeum with a crenulated median sulcus extending to the posterior margin. Postmarginal vein shorter than stigmal vein. Ovipositor sheaths short, about as long as the metasoma.

**Description.** *Female*. Body length 2.3 mm. Ovipositor sheaths length 0.9 mm. Metallic tinge present at least in some body regions. Predominantly dark green. Antennae brown. Coxae almost concolorous with mesosoma. Femora brown. Tibiae and tarsi predominantly yellow. Metatibia proximally yellow brown. Metasoma predominantly brown.

*Head*. Antennae inserted just above the middle line of compound eyes. Scape nearly  $3 \times$  as long as pedicel. Pedicel almost as long as wide, pyriform, and shorter than first funicular segment. Anelli transverse. First funicular segment approximately  $1.5 \times$  as long as wide. Distal antennomeres forming a distinctive clava. Face sculpture reticu-



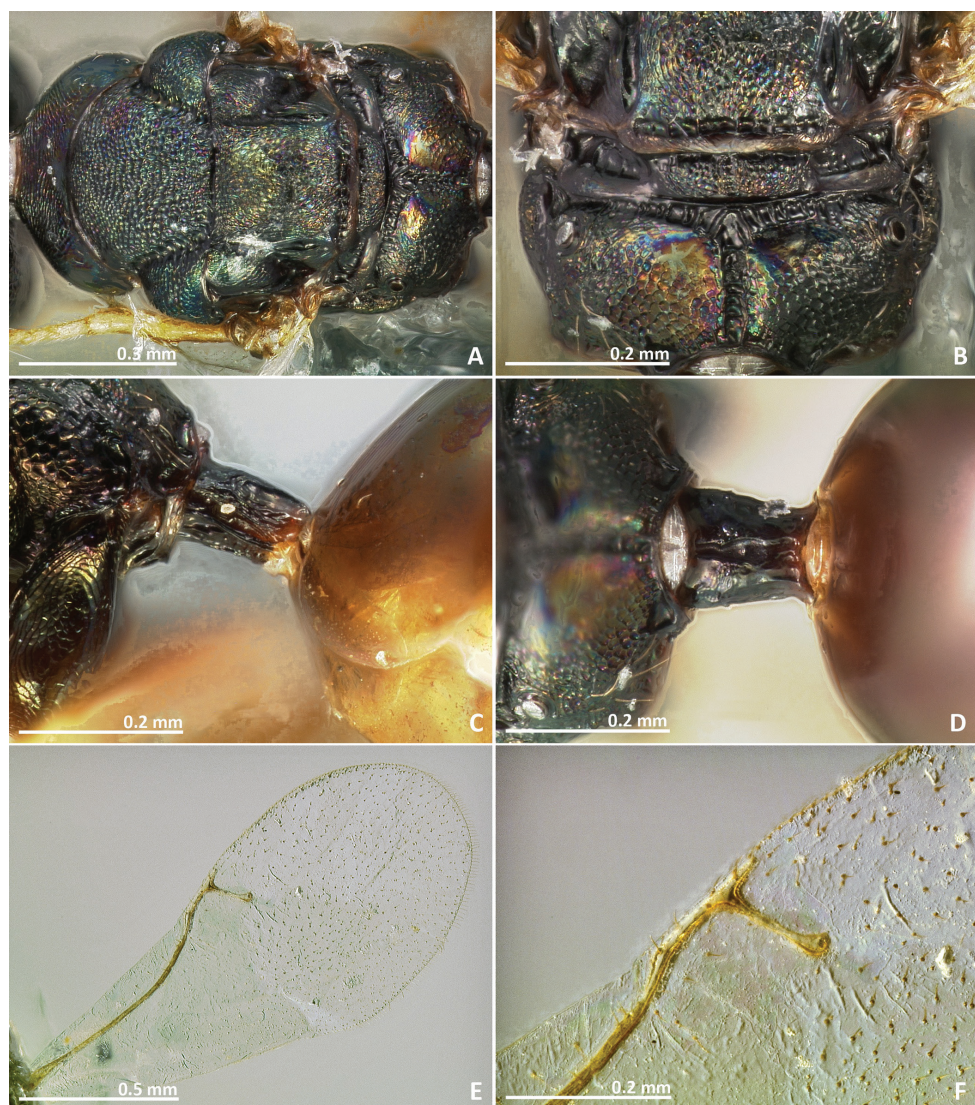


**Figure 11.** *Pseudidarnes cooki* sp. n. female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

late. Face pilosity short and sparse, becoming longer near oral margin and eyes. Supraclypeal area wide, its delimiting sulci not converging near epistomal groove, and its sculpture mostly smooth. Lateral ocelli  $1 \times$  its own diameter far from the eye margin.

*Mesosoma.* Pronotum short, nearly as long as high in lateral view. Mesoscutum strongly reticulate. Mesoscutellum reticulate. Frenal sulcus densely crenulated. Mesepimeron sculpture reticulate. Metascutellum longer than frenum, reticulate. Propodeum with a well delimited and carinated median sulcus, extending to the posterior margin of the sclerite. Propodeum sculpture reticulate, smooth near the





**Figure 12.** *Pseudidarnes cooki* sp. n. female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

proximal region of median line of propodeum. Wings hyaline, with sparse pilosity. Marginal and postmarginal vein not particularly widened. Postmarginal vein shorter than stigmal vein.

*Metasoma.* Petiole rugose,  $1.5 \times$  as long as high in lateral view. Petiole dorsally with a longitudinal median sulcus. Ovipositor sheaths long, distinctly protruding beyond metasoma apex. Ovipositor sheaths length  $2.25 \times$  hind tibia length,  $0.4 \times$  body length.

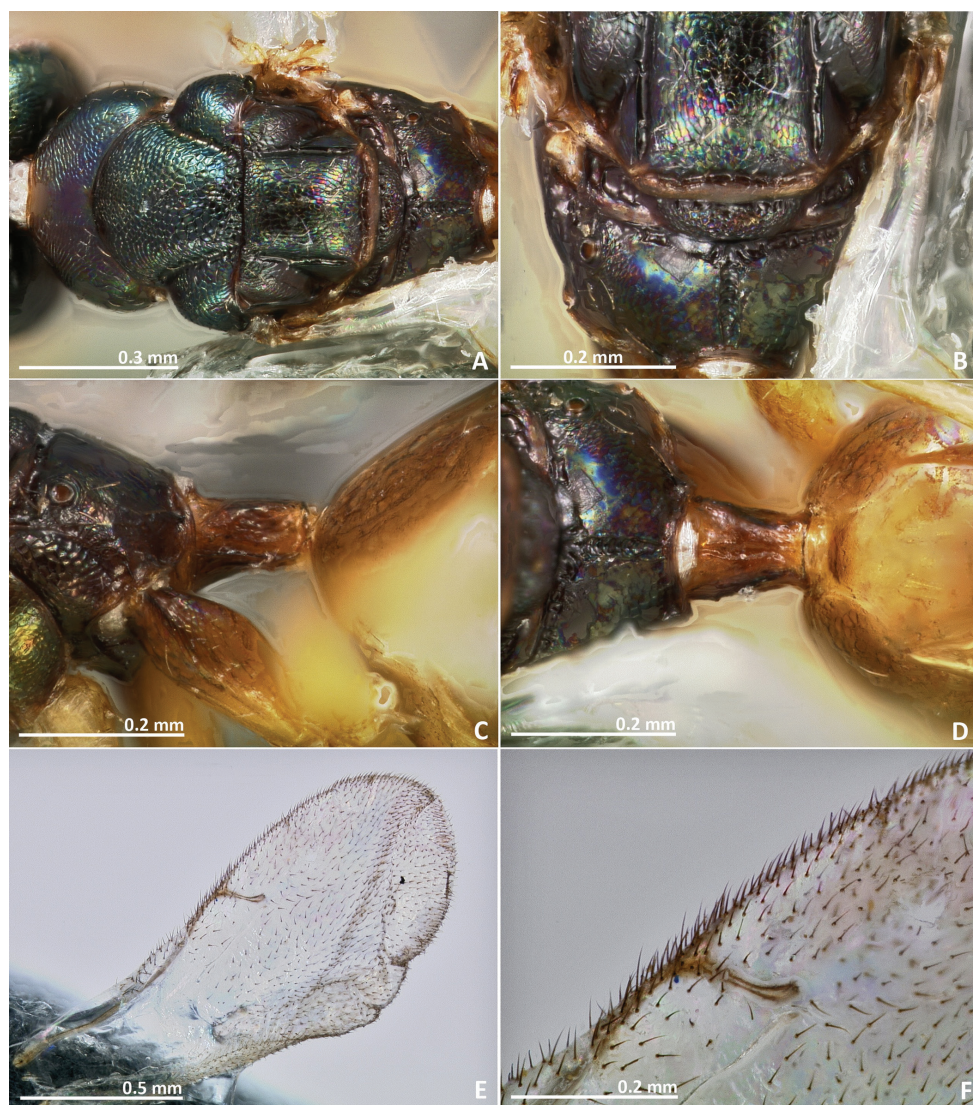




**Figure 13.** *Pseudidarnes cooki* sp. n. male. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

*Male.* Body length 2.4 mm. Characters agreeing with the females, except the following. Body slender. Antenna more yellow and inserted at the middle line of compound eyes or slightly below. Ocelli larger. Body sculpture fainter. Petiole more brown. Wings more pilose.

**Etymology.** The specific name is dedicated to our friend and colleague Dr. James Cook, in recognition of his amazing contribution to our knowledge of fig wasps.



**Figure 14.** *Pseudidarnes cooki* sp. n. male. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

**Biology.** This species is strictly associated with *Ficus obliqua* G. Forst. and was studied by Segar and Cook (2012) (referred as *Pseudidarnes* sp.). It was reared in low abundance ( $0.1 \pm 0.05$ , mean  $\pm SE$ ) and there were rarely more than four wasps in the same syconium.

**Molecular data.** GenBank sequences: COI HM770642; Cytb HM770596; EF-1a HM770545; rRNA 28S HM770704 (Cruaud et al. 2011a; Cruaud et al. 2011b).



***Pseudidarnes flavicollis* Bouček, 1988**

[http://species-id.net/wiki/Pseudidarnes\\_flavicollis](http://species-id.net/wiki/Pseudidarnes_flavicollis)

Figures 15–16

**Material examined.** *Holotype*. ♀, **PAPUA NEW GUINEA: Bulolo:** Manki area, –5.37°, 144.18°, 700m, 22.VII.1981, Roberts H., ex *F. xylosyia* (BMNH) [examined].

*Paratype*. ♀, same data as holotype (BMNH) [examined].

**Diagnosis.** Head and mesosoma excluding pronotum with metallic tinge. Pronotum long, nearly  $1.5\text{--}2 \times$  as long as high in lateral view. Mesoscutum with irregular transverse rugae. Marginal and postmarginal veins widened. Propodeum with a well delimited and carinulated median sulcus, extending to the posterior margin of the sclerite. Metascutellum as well as lateral panel of metanotum crenulated.

**Description.** *Female*. Body length 3.3 mm. Ovipositor sheaths length 1.6 mm. Metallic tinge present at least in some body regions. Predominantly yellow and green. Scape and pedicel yellow. Flagellomeres brown. Head green, green red near vertex. Pronotum yellow. Prepectus yellow brown. Remaining mesosoma green to dark green, propodeum green brown. Legs predominantly yellow, metacoxa proximally brown. Petiole dark brown. Metasoma predominantly yellow, dorsally brown.

*Head.* Antennae inserted far above the middle line of compound eyes. Scape nearly  $2 \times$  as long as pedicel. Pedicel elongated, slender, and shorter than first funicular segment. Anelli transverse. First funicular segment  $2 \times$  as long as wide. Distal antennomeres not forming a distinct clava. Face sculpture smooth with sparse punctures, lower face with engraved transverse striae. Face pilosity short and sparse, becoming longer near oral margin and eyes. Supraclypeal area wide, its delimiting sulci not converging near epistomal groove, and its sculpture mostly smooth. Lateral ocelli contiguous to the eye margin.

*Mesosoma.* Pronotum long, nearly  $1.5\text{--}2 \times$  as long as high in lateral view. Mesoscutum transversally striate. Mesoscutellum smooth. Frenal sulcus densely crenulated. Mesepimeron sculpture slightly striate. Metascutellum as well as lateral panel of metanotum crenulated, metascutellum longer than frenum. Propodeum with a well delimited and carinulated median sulcus, extending to the posterior margin of the sclerite. Propodeum sculpture mostly smooth, slightly rugose laterally. Wings hyaline, with rather dense pilosity. Marginal and postmarginal vein widened. Postmarginal vein longer than stigmal vein.

*Metasoma.* Petiole rugose,  $2 \times$  as long as high in lateral view. Petiole dorsally without a longitudinal median sulcus. Ovipositor sheaths long, distinctly protruding beyond metasoma apex. Ovipositor sheaths length  $2.7 \times$  hind tibia length,  $0.5 \times$  body length.

*Male.* Unknown

**Biology.** Collected from syconia of *F. xylosyia* Diels. See *P. astridae* biology section.





**Figure 15.** *Pseudidarnes flavicollis* Bouček, 1988, paratype, female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

***Pseudidarnes kjellbergi* Farache & Rasplus, sp. n.**

<http://zoobank.org/1DF43475-A3A3-431B-8B59-C3810F83970F>

[http://species-id.net/wiki/Pseudidarnes\\_kjellbergi](http://species-id.net/wiki/Pseudidarnes_kjellbergi)

Figures 17–18

**Material examined.** *Holotype*. ♀, **AUSTRALIA: Kununarra:** , –15.8319°, 128.8564°, 80m, 20.X.1997, Dixon, D., ex *F. platypoda*, n° PhD 455 (CBGP).

*Paratype*. ♀, same data as holotype (CBGP).



**Figure 16.** *Pseudidarnes flavicollis* Bouček, 1988, paratype, female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

**Diagnosis.** Mesosoma metallic green. Mesosoma sculpture mostly reticulate. Median sulcus of the propodeum extending over the anterior half of the sclerite. Postmarginal vein shorter than the stigmal. Petiole transverse in dorsal view. Ovipositor sheaths longer than metasoma.

**Description.** *Female.* Body length 3.1 mm. Ovipositor sheaths length 2.4 mm. Metallic tinge present at least in some body regions. Predominantly green. Antenna yellow brown. Coxae brown. Femora and tibiae predominantly brown. Tarsi yellow. Petiole brown. Metasoma green brown.

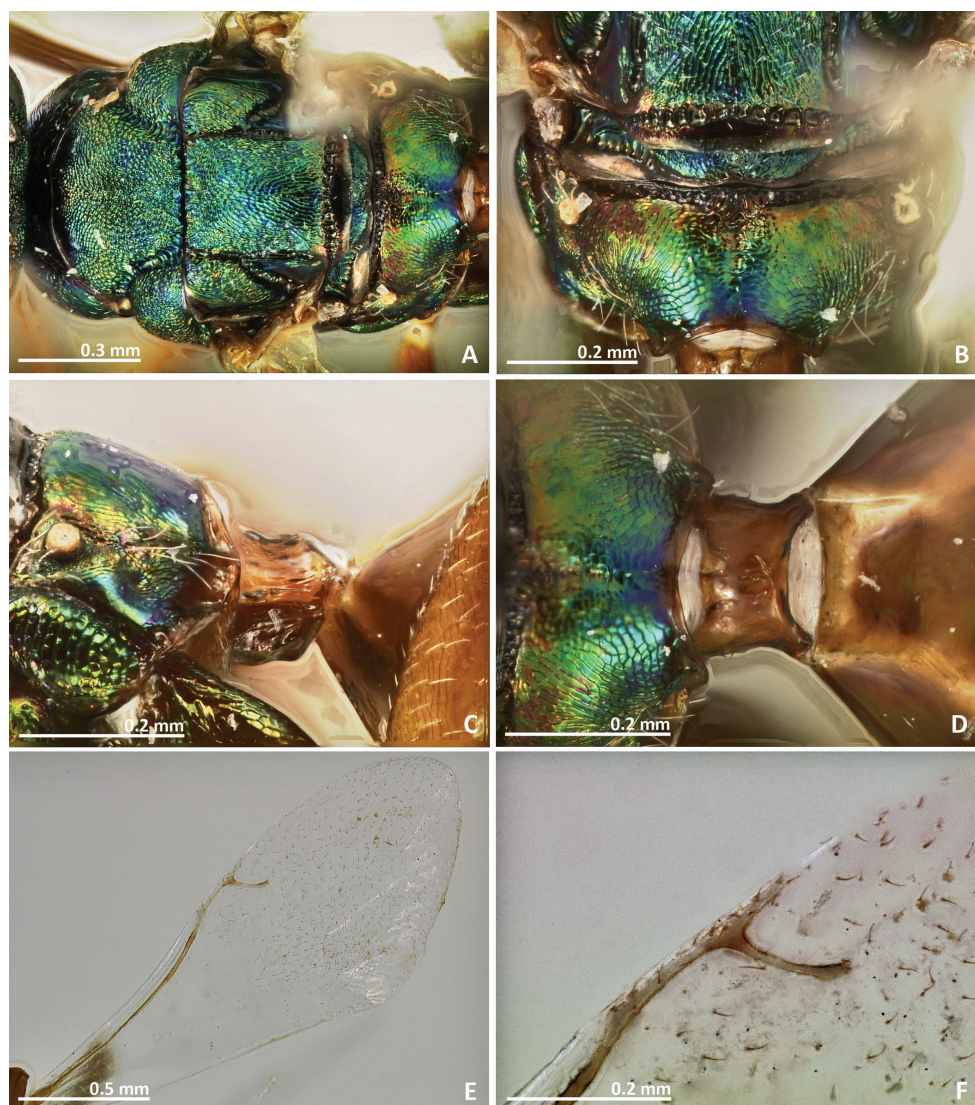




**Figure 17.** *Pseudidarnes kjellbergi* sp. n. female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

*Head.* Antennae inserted at the middle line of compound eyes. Scape nearly  $3 \times$  as long as pedicel. Pedicel almost as long as wide, pyriform, and as long as first funicular segment. Anelli transverse. First funicular segment approximately as long as wide. Distal antennomeres forming a distinctive clava. Face sculpture reticulate. Face pilosity short and sparse. Supraclypeal area wide, its delimiting sulci not converging near epistomal groove, and its sculpture mostly smooth. Lateral ocelli nearly  $1 \times$  its diameter far from the eye margin.





**Figure 18.** *Pseudidarnes kjellbergi* sp. n. female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

*Mesosoma.* Pronotum short, nearly as long as high in lateral view. Mesoscutum strongly reticulate. Mesoscutellum reticulate. Frenal sulcus densely crenulated. Mesepimeron sculpture slightly reticulate ventrally, becoming smooth in its medial and upper region. Metascutellum as long as frenalum, reticulate. Propodeum with a broad crenulated median line extending over the anterior half of the sclerite. Median line very faint or absent in the posterior half of the propodeum. Propodeum sculpture engraved reticulate. Wings hyaline, with sparse pilosity. Marginal and postmarginal vein not particularly widened. Postmarginal vein shorter than stigmal vein.

*Metasoma*. Petiole slightly rugose, and transverse in lateral view. Petiole dorsally without a longitudinal median sulcus. Ovipositor sheaths long, distinctly protruding beyond metasomal apex. Ovipositor sheaths length  $3.8 \times$  hind tibia length,  $0.7 \times$  body length.

*Male*. Unknown

**Etymology.** The specific name is dedicated to our friend and colleague Dr. Finn Kjellberg, in recognition of his excellent work in fig wasps.

**Biology.** Reared from syconia of *Ficus platypoda* (Miq.) A. Cunn. ex Miq.

***Pseudidarnes laevis* Farache & Rasplus, sp. n.**

<http://zoobank.org/475EA63C-EDE3-4BE9-819E-616FBEE94879>

[http://species-id.net/wiki/Pseudidarnes\\_laevis](http://species-id.net/wiki/Pseudidarnes_laevis)

Figures 19–22

**Material examined.** *Holotype*. ♀, **PAPUA NEW GUINEA**: Crater Mountain  $-6.58^{\circ}$ ,  $145.08^{\circ}$ , 2000m, V.1990, McKee A., ex *Ficus* sp., n° AM 451 (CBGP).

*Paratypes*. **PAPUA NEW GUINEA**: Crater Mountain  $-6.58^{\circ}$ ,  $145.08^{\circ}$ , 2000m, 20♀, 20♂, V.1990, McKee A., ex *Ficus* sp., n° AM 451 (17♀, 18♂ CBGP, 1♀, 1♂ BMNH, 1♀, 1♂ SAMC, 1♀ RPSP), 6♀, 1♂, V.1990, McKee A., ex *Ficus* sp., n° AM 550 (CBGP).

**Diagnosis.** Mesosoma metallic green, entirely smooth and shiny. First funicular segment  $2 \times$  as long as wide. Distal antennomeres not forming a definite clava.

**Description.** *Female*. Body length 3.7 mm. Ovipositor sheaths length 3.9 mm. Metallic tinge present at least in some body regions. Predominantly dark green. Antennae yellow brown. Legs yellow. Petiole dark brown. Metasoma predominantly brown, slightly green.

*Head*. Antennae inserted at the middle line of compound eyes or slightly above. Scape nearly  $3 \times$  as long as pedicel. Pedicel almost as long as wide, shorter than first funicular segment. Anelli transverse. First funicular segment  $2 \times$  as long as wide. Distal antennomeres not forming a distinct clava. Face sculpture smooth, with very sparse punctures. Face pilosity short and sparse. Supraclypeal area wide, its delimiting sulci not converging near epistomal groove, and its sculpture mostly smooth. Lateral ocelli nearly  $1 \times$  its diameter far from the eye margin.

*Mesosoma*. Pronotum longer than high in lateral view. Mesoscutum mostly smooth. Mesoscutellum smooth. Frenal sulcus densely crenulated. Mesepimeron sculpture mostly smooth. Metascutellum smooth, very short, inconspicuous, and almost completely covered by frenalum. Propodeum with a vestigial median line not extending from the beginning of the proximal region. Propodeum sculpture smooth. Wings hyaline, with rather dense pilosity. Marginal and postmarginal vein not particularly widened. Postmarginal vein as long as stigmal vein, or slightly longer.

*Metasoma*. Petiole  $2 \times$  as long as high in lateral view. Petiole sculpture in lateral view slightly rugose. Petiole dorsally with a longitudinal median sulcus. Ovipositor





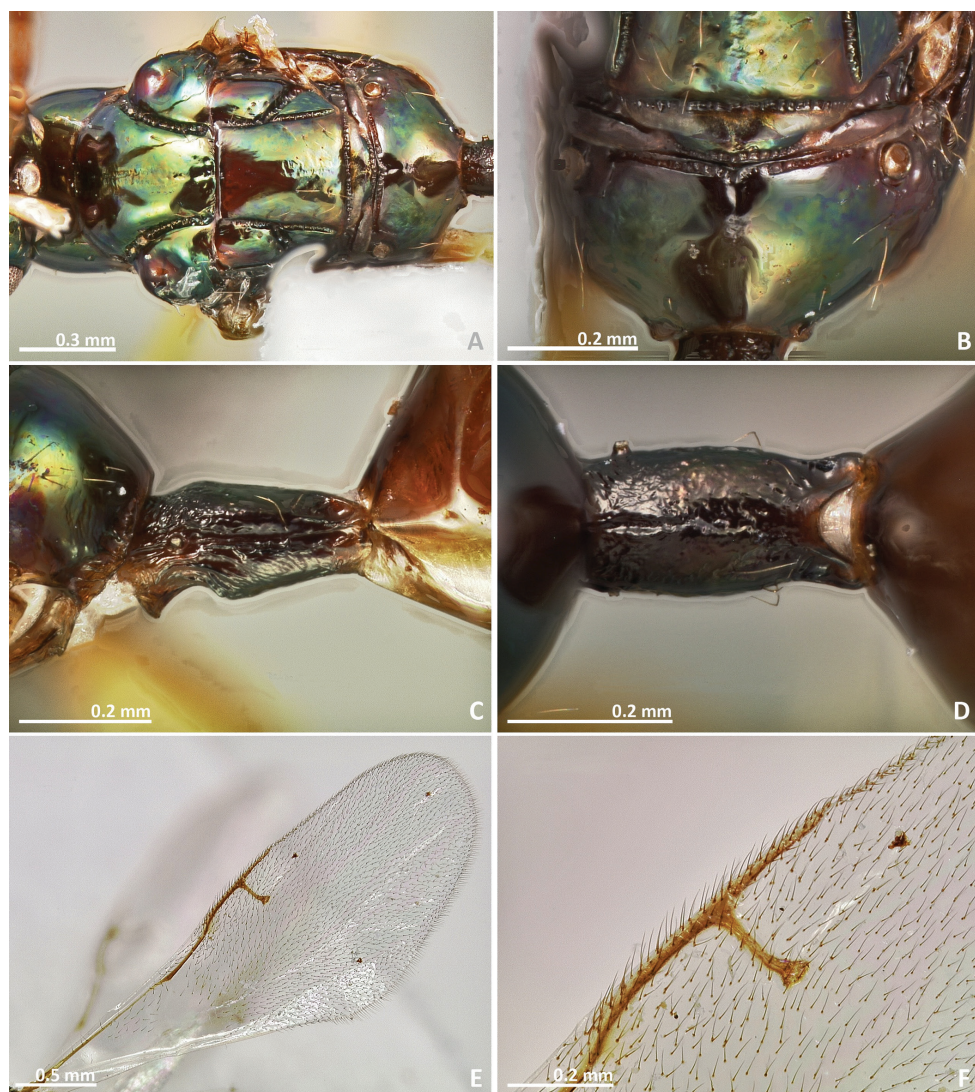
**Figure 19.** *Pseudidarnes laevis* sp. n. female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

sheaths long, distinctly protruding beyond metasoma apex. Ovipositor sheaths length  $4.3 \times$  hind tibia length, as long as body.

**Male.** Body length 3 mm. Characters agreeing with the females, except the following. Body slender. Anelli more transverse. Ocelli slightly larger than the female's ocelli. Wing more pilose.

**Etymology.** The specific name refers to the smooth body sculpturation exhibited by this species.

**Biology.** See *Pseudidarnes acaudus* biology section.



**Figure 20.** *Pseudidarnes laevis* sp. n. female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

***Pseudidarnes minerva* Girault, 1927**

[http://species-id.net/wiki/Pseudidarnes\\_minerva](http://species-id.net/wiki/Pseudidarnes_minerva)

Figures 23–27

*Pseudidarnes minerva*; Girault (1927) Records of the South Australian Museum. 3: 332.  
[description female]

*Pseudidarnes minerva*; Bouček (1988) Australasian Chalcidoidea, Cap. 6 p. 159 (key),  
187–188 pp. Figs 317–318. [New record, redescription]





**Figure 21.** *Pseudidarnes laevis* sp. n. male. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

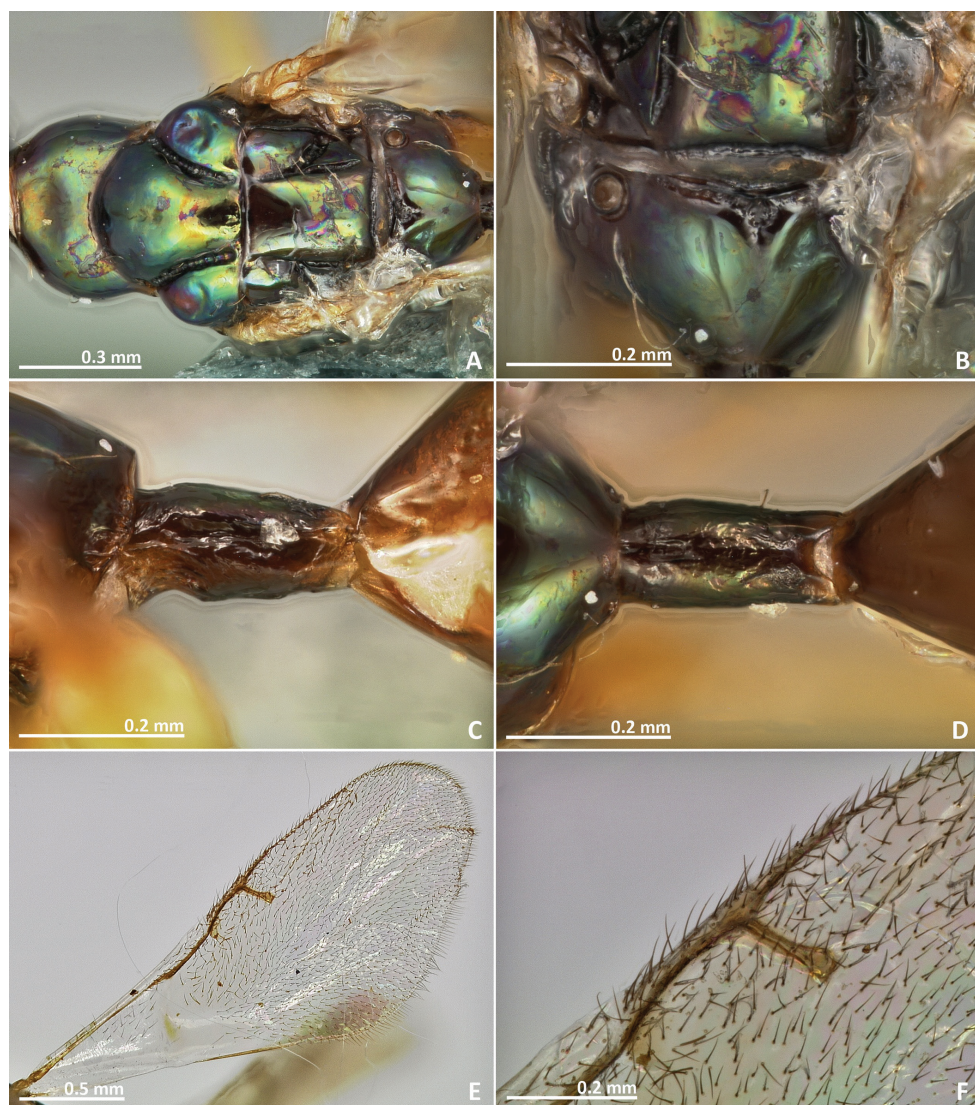
*Pseudidarnes minerva*; Fellowes et al. (1999) Behavioural Ecology and Sociobiology, 46: 95–102. [ecology]

*Pseudidarnes minerva*; Early JW (2000) New Zealand Entomologist, 23: 29–30. Fig. 1. [natural history, description of wingless male, new occurrence]

*Pseudidarnes minerva*; Cook (2005) In: Fellowes M, Holloway G, Rolff J (Eds), Insect Evolutionary Ecology, 83–110, Fig. 4.1. [ecology]

*Pseudidarnes minerva*; Cruaud et al. (2011a) BMC Evolutionary Biology, 11: 15pp. [phylogenetic position]

*Pseudidarnes minerva*; Cruaud et al. (2011b) Journal of Biogeography, 38: 209–225. [biogeography]



**Figure 22.** *Pseudidarnes laevis* sp. n. male. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

**Material examined.** *Holotype*. ♀, **AUSTRALIA: Queensland:** Brisbane and Lake Manchester,  $-27.48^{\circ}$ ,  $152.76^{\circ}$ , 67m, [no date], ex *F. rubiginosa* (ASCI) [examined].

**AUSTRALIA: Queensland:** Amity,  $-27.39^{\circ}$ ,  $153.44^{\circ}$ , 5m, 7♀, 23.I.1999, Rasplus J.Y. & Meusnier, S, ex *F. rubiginosa*, n° JRAS00722 (CBGP), Ballina,  $-28.89^{\circ}$ ,  $153.56^{\circ}$ , 5m, 7♀, 3♂, 25.I.1999, Rasplus J.Y. & Meusnier, S, ex *F. rubiginosa*, n° JRAS00726 (CBGP), 1♀, 25.I.1999, Rasplus J.Y., ex *F. rubiginosa*, n° JRAS00727\_01 (CBGP), Mount Molloy,  $-16.67^{\circ}$ ,  $145.33^{\circ}$ , 400m, 1♂, 25.X.2005, Joussetin E. & Coeur d'Acier A., ex *F. rubiginosa*, n° JRAS01418\_28 (CBGP), Yungaburra,  $-17.27^{\circ}$ ,  $145.58^{\circ}$ , 700m, 5♀, 15.I.1999, Rasplus J.Y. & Meusnier, S, ex *F. rubiginosa*, n°





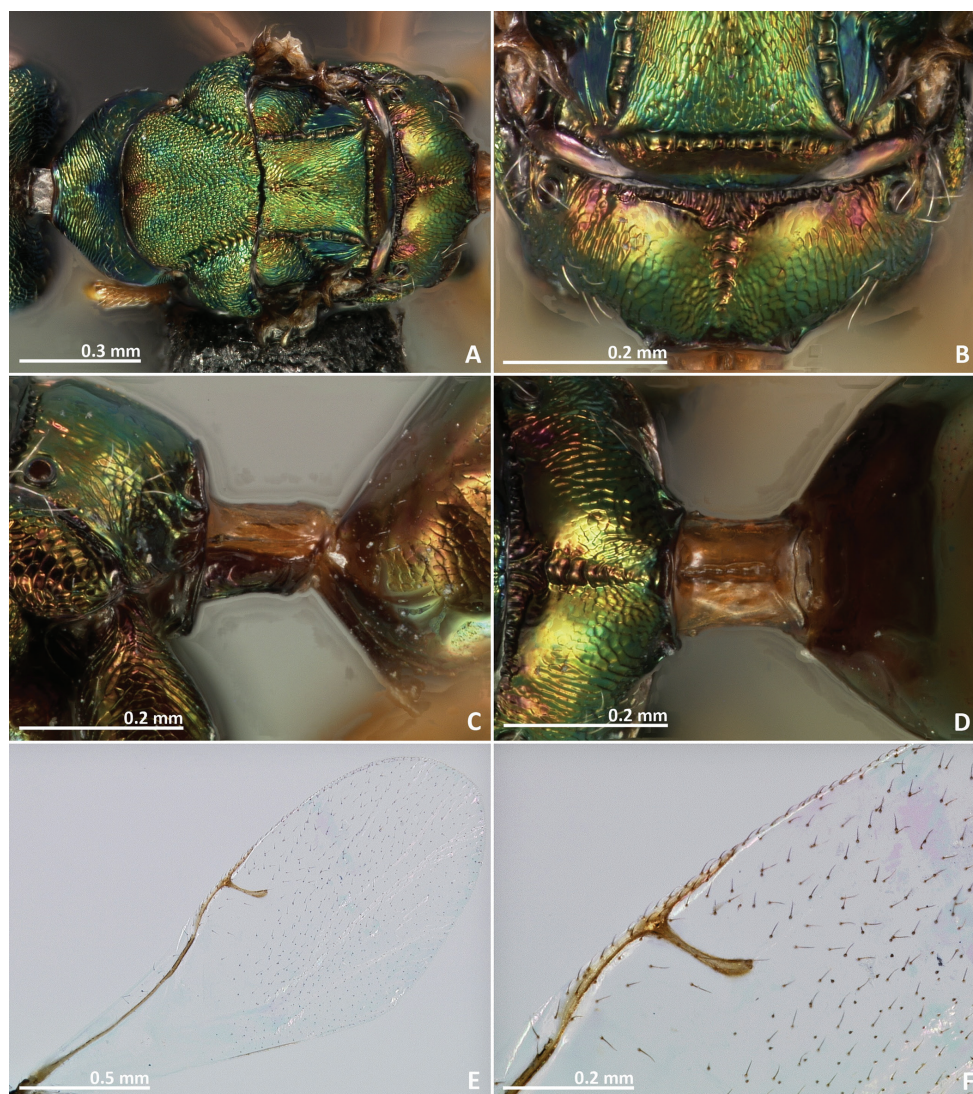
**Figure 23.** *Pseudidarnes minerva* Girault, 1927 female. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

JRAS00690 (CBGP); **Victoria:** Melbourne,  $-37.81^{\circ}$ ,  $144.96^{\circ}$ , 20m, 2♀, 2♂, I.1995, Cook J., ex *F. rubiginosa* (CBGP).

**Diagnosis.** Mesosoma metallic green, mostly reticulate. Median sulcus of the propodeum extending over most of the propodeum length, not reaching the posterior margin. Postmarginal vein as long as the stigmal. Petiole longer than wide in dorsal view. Ovipositor sheaths longer than metasoma.

**Description.** Body length 2.6 mm. Ovipositor sheaths length 2.33 mm.

**Coloration.** Metallic tinge present at least in some body regions. Predominantly green. Scape yellow. Petiole yellow brown. Flagellomeres brown. Coxae almost con-



**Figure 24.** *Pseudidarnes minerva* Girault, 1927 female. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

colourous with mesosoma. Femora brown. Tibiae and tarsi predominantly yellow. Metatibia proximally browner. Petiole brown. Metasoma browner dorsally.

*Head.* Antennae inserted at or slightly below the middle line of compound eyes. Scape nearly  $3 \times$  as long as pedicel. Pedicel almost as long as wide, pyriform, and as long as first funicular segment. Anelli transverse. First funicular segment longer than wide to approximately as long as wide. Distal antennomeres forming a distinctive clava. Face sculpture reticulate. Face pilosity short and sparse. Supraclypeal area wide, its delimiting sulci not converging near epistomal groove, and its sculpture mostly smooth. Lateral ocelli nearly one diameter far from the eye margin.

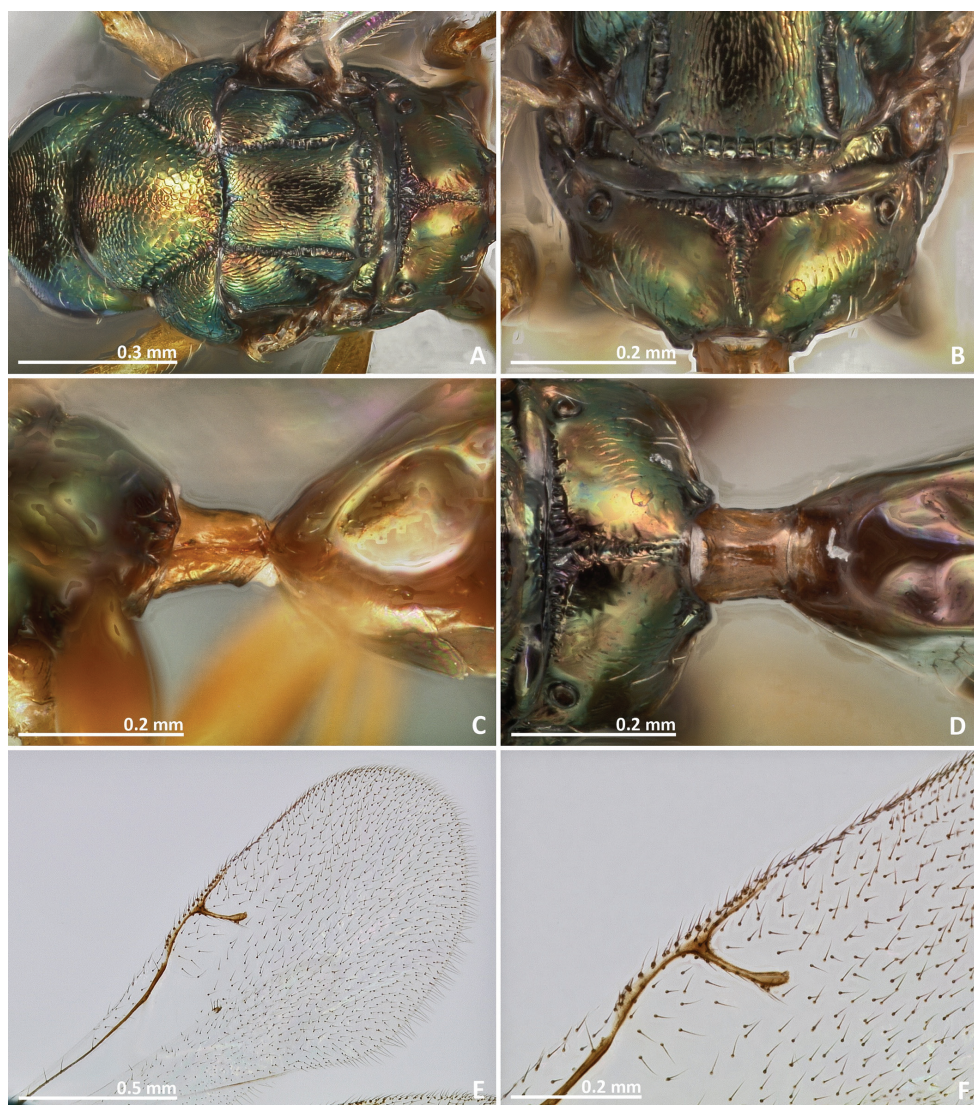




**Figure 25.** *Pseudidarnes minerva* Girault, 1927, male. **A** habitus lateral **B** mesosoma lateral **C** antenna **D** anelli **E** head, anterior view **F** vertex, dorsal view.

*Mesosoma.* Pronotum short, nearly as long as high in lateral view, or slightly longer than high. Mesoscutum reticulate. Mesoscutellum engraved. Frenal sulcus densely crenulated. Mesepimeron sculpture reticulate. Metascutellum very short, inconspicuous, and almost completely covered by frenum. Propodeum with a carinulated longitudinal median line, extending over most of the propodeum length, not reaching the posterior margin. Propodeum sculpture engraved reticulate, smooth near the proximal region of median line of propodeum. Petiole 1.5 × as long as high in lateral view.





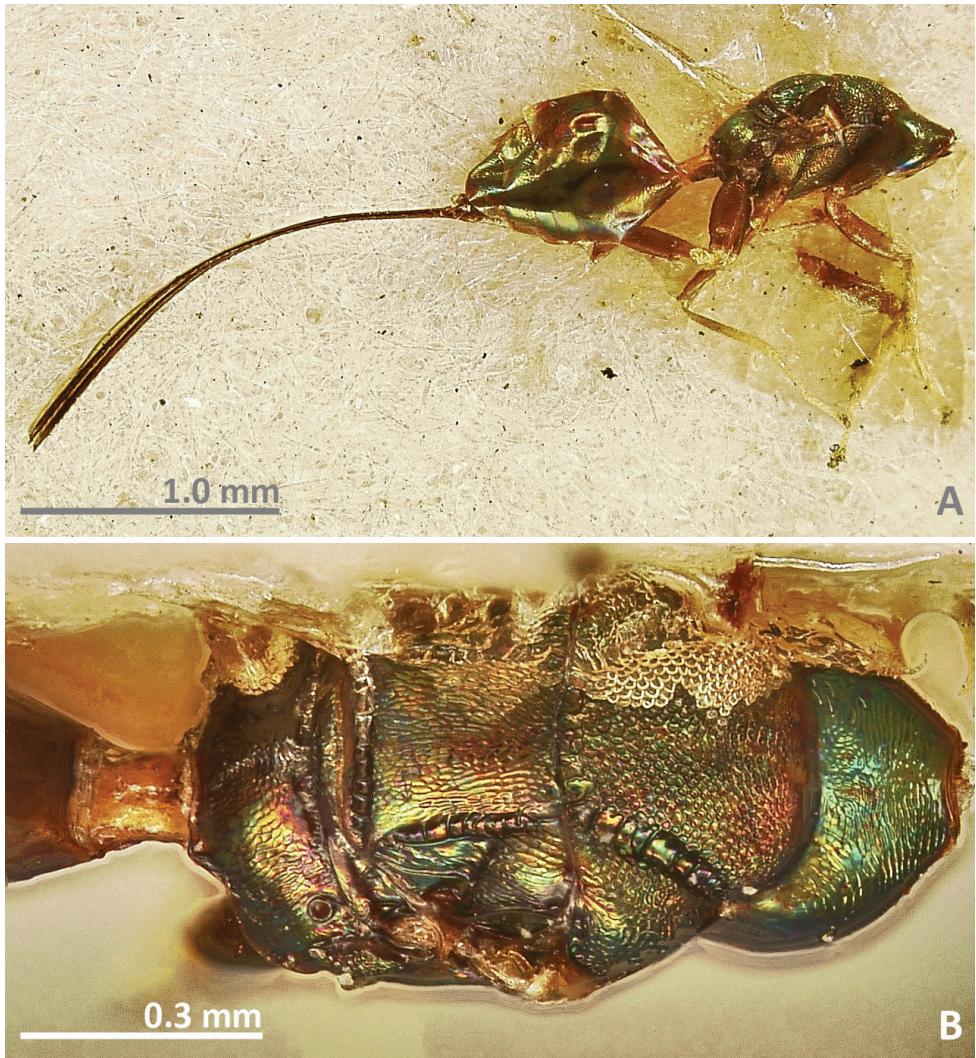
**Figure 26.** *Pseudidarnes minerva* Girault, 1927, male. **A** mesosoma dorsal **B** propodeum dorsal **C** petiole lateral view **D** petiole dorsal view **E** wing **F** detail of venation.

Wings hyaline, with sparse pilosity. Marginal and postmarginal vein not particularly widened. Postmarginal vein as long as stigmal vein, or slightly shorter.

*Metasoma.* Petiole sculpture in lateral view slightly rugose. Petiole dorsally with a longitudinal median sulcus. Ovipositor sheaths long, distinctly protruding beyond metasoma apex. Ovipositor sheaths length  $4.4 \times$  hind tibia length,  $0.9 \times$  body length.

*Male.* Body length 2.3 mm. Characters agreeing with the females, except the following. Body slender. Coxae brown, not concolorous with mesosoma. Anelli more transverse than





**Figure 27.** *Pseudidarnes minerva* Girault, 1927 holotype female. **A** habitus lateral **B** mesosoma in dorsal view.

female. Head sculpture engraved. Ocelli ca.  $2 \times$  the diameter of the female's. Body sculpture fainter. Wing more pilose.

Wingless males were described for this species (Early 2000), but they are uncommon.

**Biology.** Reared from *Ficus rubiginosa* Desf. ex Vent. syconia. *Ficus rubiginosa* is pollinated by *Pleistodontes imperialis* Saunders. Usually collected in low abundances, but sometimes they are quite abundant, at least in Eastern Australia and Auckland (as seen by Early 2000). Details about the biology of *Pseudidarnes minerva* and other wasps associated with *F. rubiginosa* in New Zealand were described by Early (2000).

**Molecular data.** GenBank sequences: COI HM770660; CytB HM770560; EF1a HM770504; rRNA 28S HM770665 (Cruaud et al. 2011a; Cruaud et al. 2011b).

***Pseudidarnes* sp. ex *Ficus baola***

*Pseudidarnes* sp. ex *F. baola*; Cruaud et al. (2011a) BMC Evolutionary Biology, 11: 15pp. [phylogenetic position]

*Pseudidarnes* sp. ex *F. baola*; Cruaud et al. (2011b) Journal of Biogeography, 38: 209–225. [biogeography]

**Material examined. SOLOMON ISLANDS: Gatokae:** Mbulo island,  $-8.76^{\circ}$ ,  $158.28^{\circ}$ , 100m, 1♂, 20.II.2019, Cruaud A & Rasplus JY, ex *F. baola*, n°JRAS02523\_02 (CBGP).

**Biology.** Collected in *Ficus baola* C. C. Berg.

**Comments.** Two specimens belonging to this species were included in the phylogenetic analysis by Cruaud et al. (2011a; 2011b). The remaining specimen is a male in poor state of conservation as it was removed from its gall before emergence, and cannot be described.

**Molecular data.** GenBank sequences: COI HM770640, JN001572; Cytb HM770594; EF-1a HM770543; rRNA 28S HM770702 (Cruaud et al. 2011a; Cruaud et al. 2011b).

## Discussion

Due to their low abundance and relative rarity, some species were described here from very small series. Nevertheless, many specimens of *P. laevis* were collected from the same sample of figs, which indicates that they can sometimes show high infestation rates. This pattern is also observed in *Anidarnes*, which are also large gallers, and are usually found at low abundances (Bronstein 1999) but are more abundant in a few samples (personal observation, Farache et al. 2013). The relative low abundance (compared to fig pollinators and some other Sycophaginae that induce smaller galls) shown by *Pseudidarnes* and *Anidarnes* may explain the difficulty experienced by earlier taxonomists when studying and describing species belonging to these genera.

Here we collected and analysed wasps from both sexes in five from the eight studied species. *Pseudidarnes* males were similar to the females, in contrast to many other wasps associated with fig inflorescences, which are sexually dimorphic and show wingless males. Nevertheless, wingless males occur in very low abundance for *P. minerva* (Cook et al. 1997, Early 2000), so the fact that we did not find them in other *Pseudidarnes* species may be due to sampling effect, and therefore new dimorphic males may be found in the future. Also, wingless males do not leave their natal figs and remain within flowers and bracts, which may hamper their sampling by unaware collectors.

The eight *Pseudidarnes* species were collected from five different hosts. *F. xylosyia* hosted three species, namely *P. astridae*, *P. badiogeminus*, and *P. flavicollis*. The former two species were reared together in the same sample. An undetermined fig species also hosted two species, *P. laevis* and *P. acaudus* (reared in a same sample). Despite the fact that more than one species may share the same fig, we did not find any



*Pseudidarnes* species occurring in more than one host, which possibly indicates that they are host specific.

*Pseudidarnes acaudus* is the most divergent of all collected species, and is easily recognizable by its extremely short ovipositor sheaths. The other species can be separated into two morphological groups that correspond well to their geography and to their host association. Papuan species show a slender mesosoma, long funicular segments, and body sculpture that is mostly smooth, while Australian species have a short and robust mesosoma, shorter funicular segments, and a reticulate body sculpture. The taxonomy of the section *Malvanthera* is also geographically consistent since two of its subsections (namely *Malvantherae* and *Platypodeae*) are primarily Australian, while subsection *Hesperidiiformes* has its diversity centre in New Guinea (Rønsted et al. 2008). All Australian hosts collected belong to subsection *Platypodeae*, whereas *F. xylosyia* from Papua belongs to subsect. *Hesperidiiformes*. The host of *P. laevis* is unknown, but probably it belongs to subsect. *Hesperidiiformes*, since the only species belonging to other *Malvanthera* subsections known to occur in Papua is *F. obliqua* (Rønsted et al. 2008).

This is the first revisionary treatment of *Pseudidarnes*. We believe that, due to the lack of previous careful sampling, several *Pseudidarnes* species remain to be discovered especially in New Guinea, but also in Australia. We hope that this work will encourage discovery and further studies on the biology of *Pseudidarnes* species.

## Acknowledgements

We are greatly indebted to Armelle Coeur d'acier, James Cook, Dale Dixon, Emmanuelle Jousselein, and Carlos L. Vaamonde for contributing with samples. For assistance in the development of online keys we express our gratitude to Simon van Noort. We also express our sincere gratitude to Astrid Cruaud and Serge Meusnier for kind sampling assistance to JYR. For the loan of specimens we thank the following curators: Natalie Dale-Skey Papilloud (BMNH, London, U.K.) and Peter Gillespie (ASCI, Australia). The Synthesys project. <http://www.synthesys.info/> funded the stay of JYR at the NMW and at NCB Naturalis. FHAF was funded by FAPESP (grants 2010/51158-5 and 2012/19815-1).

## References

- Ashmead WH (1904) Classification of the Chalcid flies or the superfamily Chalcidoidea, with descriptions of new species in the Carnegie Museum collected in S. America by Herbert H. Smith. *Memoirs of the Carnegie Museum* 1: 225–551.
- Bouček Z (1988) Australasian Chalcidoidea (Hymenoptera). A biosystematic revision of genera of fourteen families, with a reclassification of species. C.A.B. International, Wallingford, 1–832.
- Bouček Z (1993) The genera of chalcidoid wasps from *Ficus* fruit in the New World. *Journal of Natural History* 27: 173–217. doi: 10.1080/00222939300770071

- Bronstein J (1999) Natural history of *Anidarnes bicolor* (Hymenoptera: Agaonidae), a galler of the Florida strangling fig (*Ficus aurea*). Florida Entomologist 82: 454–461. doi: 10.2307/3496871
- Brown BV (2013) Automating the “Material examined” section of taxonomic papers to speed up species descriptions. Zootaxa 3683: 297–299. doi: 10.11646/zootaxa.3683.3.8
- Buffington M, Gates M (2008) Advanced imaging techniques II: using a compound microscope for photographing point-mounted specimens. American Entomologist 54: 222–224.
- Cook JM, Compton SG, Herre EA, West SA (1997) Alternative mating tactics and extreme male dimorphism in fig wasps. Proceedings of the Royal Society of London Series B Biological Sciences 264: 747–754. doi: 10.1098/rspb.1997.0106
- Cruaud A, Jabbour-Zahab R, Genson G, Couloux A, Peng Y-Q, Rong YD, Ubaidillah R, Pereira RAS, Kjellberg F, van Noort S, Kerdelhue C, Rasplus J-Y (2011a) Out of Australia and back again: the world-wide historical biogeography of non-pollinating fig wasps (Hymenoptera: Sycophaginae). Journal of Biogeography 38: 209–225. doi: 10.1111/j.1365-2699.2010.02429.x
- Cruaud A, Jabbour-Zahab R, Genson G, Kjellberg F, Kobmoo N, van Noort S, Da-Rong Y, Yan-Qiong P, Ubaidillah R, Hanson PE, Santos-Mattos O, Farache FHA, Pereira RAS, Kerdelhue C, Rasplus J-Y (2011b) Phylogeny and evolution of life-history strategies in the Sycophaginae non-pollinating fig wasps (Hymenoptera, Chalcidoidea). BMC Evolutionary Biology 11: 15 pp. doi: 10.1186/1471-2148-11-178
- Dixon DJ (2003) A taxonomic revision of the Australian *Ficus* species in the section *Malvanthera* (*Ficus* subg. *Urostigma*: Moraceae). Telopia 10: 125–153.
- Early JW (2000) Fig wasps (Hymenoptera: Agaonidae and Torymidae) in New Zealand. New Zealand Entomologist 23: 27–32. doi: 10.1080/00779962.2000.9722062
- Elias LG, Menezes AO Jr, Pereira RAS (2008) Colonization sequence of non-pollinating fig wasps associated with *Ficus citrifolia* in Brazil. Symbiosis 45: 107–111.
- Farache FHA, Cruaud A, Genson G, Pereira RAS, Rasplus J-Y (2013) Taxonomic revision and molecular phylogeny of the fig wasp genus *Anidarnes* Bouček, 1993 (Hymenoptera, Sycophaginae). Systematic Entomology 38: 14–34. doi: 10.1111/j.1365-3113.2012.00644.x
- Gibson GAP (1997) Morphology and terminology. In: Gibson GAP, Huber JT, Woolley JB (Eds) Annotated Keys to the Genera of Nearctic Chalcidoidea (Hymenoptera). National Research Council Press, Ottawa, 16–44.
- Girault AA (1927) Notes on and descriptions of Chalcid wasps (Chalcididae) in the South Australian Museum. Records of the South Australian Museum 3: 309–338.
- Heraty JM, Burks RA, A C, Gibson GAP, Liljeblad J, Munro JB, Rasplus J-Y, Delvare G, Jansta P, Gumovsky A, Huber JT, Woolley JB, Krogmann L, Heydon S, Polaszek A, Schmidt S, Darling DC, Gates M, Mottern J, Murray E, DalMolin A, Triapitsyn SV, Baur H, Pinto JD, van Noort S, Yoder MJ (2013) A phylogenetic analysis of the megadiverse Chalcidoidea (Hymenoptera). Cladistics 29: 466–542. doi: 10.1111/cld.12006
- Heraty JM, Hawks D (1998) Hexamethyldisilazane - A chemical alternative for drying insects. Entomological News 109: 369–374.
- Lopez-Vaamonde C, Dixon DJ, Cook JM, Rasplus J-Y (2002) Revision of the Australian species of *Pleistodontes* (Hymenoptera: Agaonidae) fig-pollinating wasps and their host-plant associations. Zoological Journal of the Linnean Society 136: 637–683. doi: 10.1046/j.1096-3642.2002.00040.x

- Noyes J (1982) Collecting and preserving chalcid wasps (Hymenoptera: Chalcidoidea). *Journal of Natural History* 16: 315–334. doi: 10.1080/00222938200770261
- Rasplus J-Y, Soldati L (2005) Familia Agaonidae. In: Fernández F, Sharkey MJ (Eds) *Introducción a los Hymenoptera de la Región Neotropical*. Sociedad Colombiana de Entomología & Universidad Nacional de Colombia, Bogotá, 683–698.
- Rønsted N, Weiblen GD, Savolainen V, Cook JM (2008) Phylogeny, biogeography, and ecology of *Ficus* section *Malvanthera* (Moraceae). *Molecular Phylogenetics and Evolution* 48: 12–22. doi: 10.1016/j.ympev.2008.04.005
- Segar S, Cook JM (2012) The dominant exploiters of the fig/pollinator mutualism vary across continents, but their costs fall consistently on the male reproductive function of figs. *Ecological Entomology* 37: 342–349. doi: 10.1111/j.1365-2311.2012.01370.x
- Segar ST, Dunn DW, Darwell CT, Cook JM (2013) How to be a fig wasp down under: The diversity and structure of an Australian fig wasp community. *Acta Oecologica* in press. doi: 10.1016/j.actao.2013.03.014
- Walker F (1843) Descriptions of Chalcidites discovered in St. Vincent's Isle by the Rev. Lansdown Guilding. *Annals and Magazine of Natural History* 12: 45–49. doi: 10.1080/03745484309442484
- West SA, Herre EA (1998) Partial local mate competition and the sex ratio: A study on non-pollinating fig wasps. *Journal of Evolutionary Biology* 11: 531–548.
- Westwood JO (1840) On caprification as practised upon the Figs in the south of Europe and the Levant, with descriptions of the insects employed for that purpose; and observations upon the *Agaon paradoxum* Dalman. *Transactions of the Entomological Society of London* 2: 214–224.



# The relevance, biases, and importance of digitising opportunistic non-standardised collections: A case study in Iberian harvestmen fauna with BOS Arthropod Collection datasets (Arachnida, Opiliones)

Izaskun Merino-Sáinz<sup>1</sup>, Antonio Torralba-Burrial<sup>2</sup>, Araceli Anadón<sup>1</sup>

**1** *Departamento de Biología de Organismos y Sistemas, Universidad de Oviedo. C/ Catedrático Rodrigo Uría s/n 33071 Oviedo, Asturias, España* **2** *Cluster de Energía, Medioambiente y Cambio Climático, Campus de Excelencia Internacional, Universidad de Oviedo, España*

Corresponding author: Antonio Torralba-Burrial (antoniotb@gmail.com)

---

Academic editor: V. Chavan | Received 29 October 2013 | Accepted 28 March 2014 | Published 24 April 2014

---

**Citation:** Merino-Sáinz I, Torralba-Burrial A, Anadón A (2014) The relevance, biases, and importance of digitising opportunistic non-standardised collections: A case study in Iberian harvestmen fauna with BOS Arthropod Collection datasets (Arachnida, Opiliones). ZooKeys 404: 71–89. doi: 10.3897/zookeys.404.6520 GBIF Key: <http://www.gbif.org/dataset/7ceb715-c3b0-4477-99e7-f6f3aca27bbe>

---

**Resource citation:** Universidad de Oviedo (2014). BOS Arthropod Collection of University of Oviedo (Spain): Opiliones unplanned collection events subset, as part of BOS Arthropod Collection Dataset: Opiliones (BOS-Opi). 472 data records in subset (3772 data records in original dataset). Contributed by: Merino-Sáinz I, Anadón A, Torralba-Burrial A. Online at [http://www.gbif.es:8080/ipt/resource.do?r=bos-opi\\_unplanned\\_collection\\_events](http://www.gbif.es:8080/ipt/resource.do?r=bos-opi_unplanned_collection_events) (subset description), <http://www.gbif.es:8080/archive.do?r=Bos-Opi> (original dataset), and <http://www.unioviedo.es/BOS/Zoologia/artropodos/opiliones> (original dataset), version 1.0 (last updated on 2013-06-30). GBIF key: <http://www.gbif.org/dataset/7ceb715-c3b0-4477-99e7-f6f3aca27bbe> (subset metadata), and <http://www.gbif.org/dataset/cc0e6535-6bb4-4703-a32c-077f5e1176cd> (original dataset), Data Paper ID: doi: 10.3897/zookeys.404.6520 and doi: 10.3897/zookeys.341.6130 (original dataset).

## Abstract

In this study, we analyse the relevance of harvestmen distribution data derived from opportunistic, unplanned, and non-standardised collection events in an area in the north of the Iberian Peninsula. Using specimens deposited in the BOS Arthropod Collection at the University of Oviedo, we compared these data with data from planned, standardised, and periodic collections with pitfall traps in several locations in the same area. The Arthropod Collection, begun in 1977, includes specimens derived from both sampling types, and its recent digitisation allows for this type of comparative analysis. Therefore, this is the first data-paper employing a hybrid approach, wherein subset metadata are described alongside a comparative

analysis. The full dataset can be accessed through Spanish GBIF IPT at <http://www.gbif.es:8080/ipt/archive.do?r=Bos-Opi>, and the metadata of the unplanned collection events at [http://www.gbif.es:8080/ipt/resource.do?r=bos-opi\\_unplanned\\_collection\\_events](http://www.gbif.es:8080/ipt/resource.do?r=bos-opi_unplanned_collection_events). We have mapped the data on the 18 harvestmen species included in the unplanned collections and provided records for some species in six provinces for the first time. We have also provided the locations of *Phalangium opilio* in eight provinces without published records. These results highlight the importance of digitising data from unplanned biodiversity collections, as well as those derived from planned collections, especially in scarcely studied groups and areas.

### Keywords

Biodiversity collections, entomological collections, digitisation priorities, sampling methodology, biases, Opiliones, distribution, Iberian Peninsula

## General description

**Purpose:** Existing knowledge on the distribution of harvestmen throughout the Iberian Peninsula is still highly fragmented (Prieto 2003). Several studies on particular genera (e.g., Prieto 2004, Prieto and Fernández 2007, Merino-Sáinz et al. 2013a), as well as studies with planned, repeated, and systematic samplings in some locations (Merino-Sáinz and Anadón 2008, 2013, Rosa García et al. 2009a,b, 2010a,b, Merino-Sáinz et al. 2013b) have contributed to improving this knowledge. Global or specific studies on biodiversity are also enabled by the review, digitisation, and data release of specimens housed in biodiversity collections at research centers, universities, museums, and in the possession of individuals. These practices facilitate the identification of gaps in our knowledge of taxa distribution across space and time.

Within this context, biodiversity data on specimens from the BOS Arthropod Collection (hosted at the Department of Organisms and Systems Biology, (Spanish acronym BOS), University of Oviedo) are being digitised and the data released through the Global Biodiversity Information Facility (GBIF) data-portal (Department information as data published and available datasets: <http://www.gbif.org/publisher/95cb537c-74c5-4c1e-ae24-32e7ea08f380>; general digitisation and data release workflow of the BOS Arthropod Collection: Torralba-Burrial and Ocharan 2013). However, there is a need to establish priorities in the digitisation of specimens data of biodiversity collections (see Berents et al. 2010 for different approaches), especially in situations where mass digitisation methods are not available (see Beaman and Cellinese 2012). As such, we evaluate whether the effort of reviewing and digitising (harvestmen) specimens from unplanned collection events can provide useful data on their biodiversity and distribution, or whether it is better to limit digitisation to only those specimens associated with standardised samplings (planned collection events), which provide quantitative data in each location and allow for comparisons between locations over time.

The BOS Arthropod Collection includes harvestmen from the northern part of the Iberian Peninsula that, since 1977, have been obtained through systematic repeated sampling in several locations, as well as through non-harvestmen-specific sampling and

accidental occurrences. Specimens were collected systematically from the Muniellos Biosphere Reserve between 2000 and 2002 (Merino-Sáinz and Anadón 2008), and from several locations in the provinces of Asturias, Cantabria, and Pontevedra between 2009 and 2011 (Merino-Sáinz et al. 2013c describe the harvestmen subcollection of BOS). Therefore, we decided to study and compare the data derived from unplanned collections events (untargeted sampling) with these data derived from planned, standardised, and periodic sampling. We have combined these analyses with the published results of similar studies using pitfall traps in western Asturias (Rosa García et al. 2009a,b, 2010a,b). In effect, this is the first data-paper to employ a hybrid approach, wherein the subset metadata from a large published dataset are described and a comparative analysis is carried out, in order to evaluate digitisation priorities. The aims of this paper are, thus, to 1) test whether the effort of reviewing and digitising (harvestmen) specimens from unplanned collection events can provide useful data about their distribution and biology, and 2) assess possible biases arising from the use of this type of data.

## Project details

**Project title:** Informatización de la Colección de Artrópodos BOS de la Universidad de Oviedo / Digitisation of the BOS Arthropod Collection of University of Oviedo

**Personnel digitisation and metadata creator:** A. Torralba-Burrial

**Administrative contact:** A. Anadón

**BOS-Opi determination specialist:** I. Merino-Sáinz

**Subset collectors:** Most of the collectors provided less than ten harvestmen records in this subset. Only Merino-Sáinz collected more than 10 specimens. All of the collectors are listed in Supplementary material 1 (<http://hdl.handle.net/10651/24734>) next to each specimen.

**Funding:** The digitisation of this biological collection was supported by the Spanish National R+D+i Plan (MICINN, Spanish Government, grant ref. PTA2010-4108-I) and PCTI Asturias (Asturias Regional Government, ref. COF11-38) through a contract with ATB.

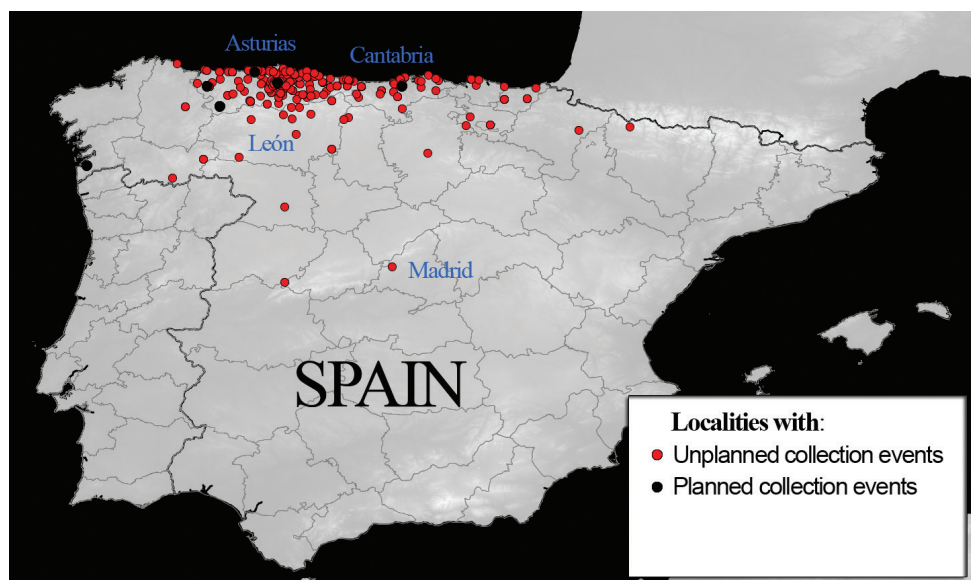
Specimens were identified by IMS, which was supported by a Severo Ochoa pre-doctoral grant (ref. BP08039, FICYC, Asturias Regional Government).

**Study area descriptions/descriptor:** Harvestmen data in the subset are from the same area as the full Opiliones of the BOS Arthropod Collection dataset. Specimens are mainly from the northern third of the Iberian Peninsula (chiefly the Spanish provinces of Asturias, Cantabria, and León, with a few records from other neighbouring provinces) (see Figure 1).

Data sources of harvestmen data from planned collection events with pitfall trapping: Merino-Sáinz and Anadón (2008, 2013); Merino-Sáinz et al. 2013c, Rosa García et al. (2009a,b, 2010a,b).

**Design description:** The data subset is part of the large dataset of Opiliones housed in the BOS Arthropod Collection (Universidad de Oviedo 2013-). Details of



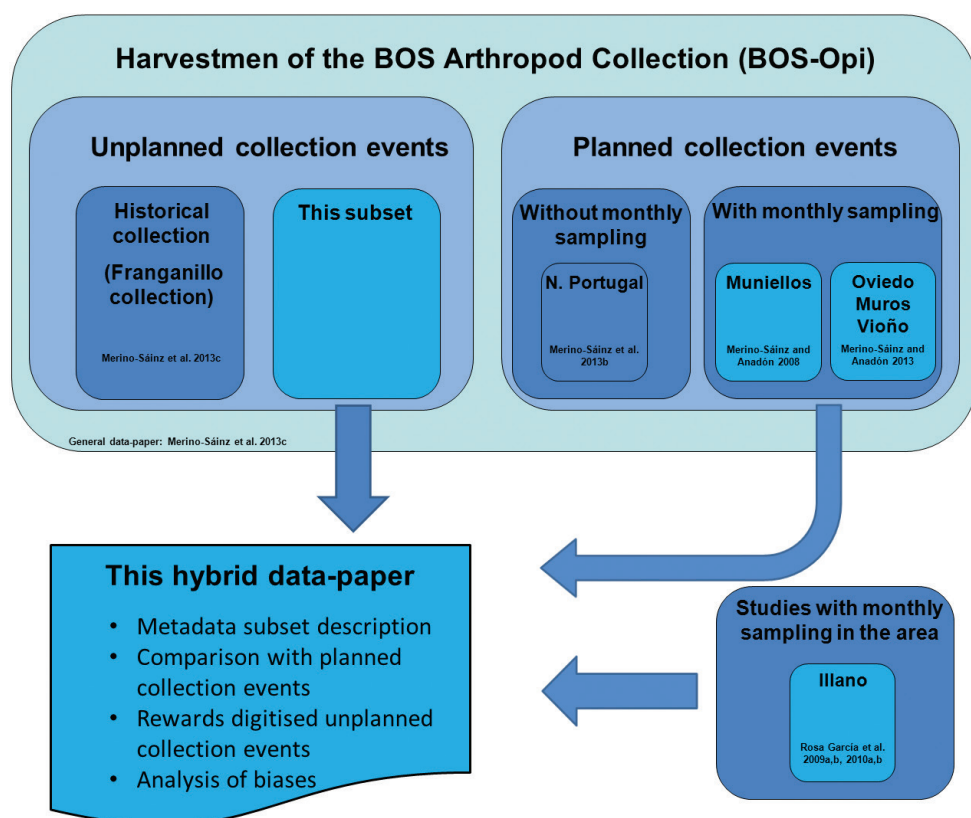


**Figure 1.** Distribution of specimens included in this subset.

the digitisation process are available in the description of the BOS Collection digitisation workflow (Torrallba-Burrial and Ocharan 2013) and in the data paper on the harvestmen subcollection (Merino-Sáinz et al. 2013c). In that data-paper, we argue that the large dataset could be used to assess, among other things, the importance of unplanned collection data in filling in knowledge gaps if planned (standardised sampling) collection data are not available or are incomplete. With this aim in mind, we chose a data subset from the harvestmen subcollection, which included data only from unplanned collection events. This subset was used to compare specimens data with published data obtained through planned, standardised, and periodic samplings using pitfall traps in several locations in the north of the Iberian Peninsula (see Merino-Sáinz and Anadón 2013 for a checklist of the species found in the studies). Moreover, we used all of the published data on Iberian harvestmen, not just the BOS Arthropod Collection harvestmen data, to analyse the distributional knowledge gained by digitising this subset, e.g., the first provincial records. Figure 2 shows a diagram depicting the methodological design of our analysis.

## Taxonomic coverage

**General taxonomic coverage description:** Seventeen taxa were identified to the species level. Due to the biological phase or sex of the specimens, or unresolved taxonomic issues, 39 records (8%) were assigned only to the genus level. Those specimens belonging to



**Figure 2.** A diagram depicting the methodological design of this hybrid data paper. Harvestmen in the BOS Arthropod Collection (Merino-Sáinz et al. 2013c) have come from several sources: some from unplanned collection events and some from planned collections. For this hybrid data-paper, we compared the data subset of unplanned collection events with the subsets of harvestmen from planned collection events using monthly sampling (Merino-Sáinz and Anadón 2008, 2013), and the harvestmen of similar planned events in the same area (Rosa García et al. 2009a,b, 2010a,b). All of the subsets compared appear in light blue in the diagram.

the genus *Paramiopsalis* represent species number 18. The numbers of records per species and per family (also including specimens identified to the genus level) are shown in Table 1.

The family Phalangidae comprised the largest number of identified species (seven), followed by Sclerosomatidae (five). However, when the number of records is considered, Sclerosomatidae was the most frequent family (around one hundred records for both *Leiobunum blackwalli* and *L. rotundum*), followed by Phalangidae, with only one species *Phalangium opilio* with a high number of records, similar to the *Leiobunum* species, and other species with only a few records. Five families (and the remaining species) had less than 30 records each.

**Table 1.** Harvestmen families and species included in the data subset.

Family	Species	Abundance	Chorology
Sclerosomatidae	<i>Leiobunum blackwalli</i> Meade	129	EU
	<i>Leiobunum rotundum</i> (Latreille)	94	EU
	<i>Homalenotus laranderas</i> Grasshoff	28	EI
	<i>Gyas titanus</i> Simon	19	EU
	<i>Leiobunum</i> spp.	5	
	<i>Homalenotus quadridentatus</i> (Cuvier)	3	EU
	<i>Homalenotus</i> spp.	3	
<b>Total Sclerosomatidae</b>	<b>5</b>	<b>281</b>	
Phalangidae	<i>Phalangium opilio</i> Linnaeus	109	HO
	<i>Odiellus</i> spp.	37	
	<i>Paroligolophus agrestis</i> (Meade)	16	HO
	<i>Dicranopalpus ramosus</i> (Simon)	13	EU
	<i>Odiellus simplicipes</i> (Simon)	10 ♂♂	EI
	<i>Odiellus seoanei</i> (Simon)	6 ♂♂	EI
	<i>Paroligolophus</i> spp.	5	
	<i>Odiellus spinosus</i> (Bosc)	2 ♂♂	EU
	<i>Megabunus diadema</i> (Fabricius)	4	EU
<b>Total Phalangidae</b>	<b>7</b>	<b>202</b>	
Ischyropsalididae	<i>Ischyropsalis hispanica</i> Roewer	10	EI
Nemastomatidae	<i>Nemastomella dentipatellae</i> (Dresco)	8	EI
	<i>Nemastoma hankiewiczii</i> (Kulczynski)	1	EI
<b>Total Nemastomatidae</b>	<b>2</b>	<b>9</b>	
Trogulidae	<i>Trogulus</i> sp. aff. <i>nepaeformis</i> (Scopoli)	21	
	<i>Anelasmaocephalus cambridgei</i> (Westwood)	1	EU
<b>Total Trogulidae</b>	<b>2</b>	<b>22</b>	
Sironidae	<i>Paramiopsalis</i> sp.	12	EI

EI: Iberian endemic, EU: European, HO: Holarctic (Merino-Sáinz and Anadón 2008)

**Taxonomic ranks**

**Kingdom:** Animalia  
**Phylum:** Arthropoda  
**Class:** Arachnida  
**Order:** Opiliones  
**Family:** Sclerosomatidae, Phalangidae, Ischyropsalididae, Nemastomatidae, Trogulidae, Sironidae  
**Common names:** Animals, Arthropods, Arachnids, Harvestmen

**Spatial coverage**

**General spatial coverage**

Harvestmen specimens of this subset are mainly from the northern third of Spain, similar to spatial coverage of the large dataset (see Merino-Sáinz et al. 2013c for a wider overview).



## Coordinates

40°21'36"N and 43°40'12"N Latitude; 7°26'24"W and 0°31'12"W Longitude.

## Temporal coverage

1977–2012.

## Natural collections description

**Parent collection identifier:** Colección de Artrópodos BOS

**Collection name:** Colección de Artrópodos BOS de la Universidad de Oviedo: Opiliones (BOS-Opi) subset recolecciones no planeadas

**Collection identifier:** <http://www.gbif.org/dataset/7cebf715-c3b0-4477-99e7-f6f3a-ca27bbe>

**Curatorial unit:** 472 with an uncertainty of 0 (Data records)

**Curatorial unit:** 536 with an uncertainty of 0 (Specimens)

## Methods

**Method step description:** This data subset was extracted from the large dataset of harvestmen in the BOS Arthropod Collection (University of Oviedo 2013-). Specimens data in the subset are listed in Supplementary material 1 - Appendix A (<http://hdl.handle.net/10651/24734>), which includes the municipality, location, date, sampling method, amount, sex, and collector of the 536 taxonomically identified specimens corresponding to BOS-Opi codes 493-960. Using these codes, most of the specimen data (including their georeferenced locations) are available in reusable format in the DarwinCore Archive of the data-paper describing the BOS-Opi subcollection (Merino-Sáinz et al. 2013c) and through the GBIF data-portal (Universidad de Oviedo 2013-, <http://data.gbif.org/datasets/resource/15038>).

Bibliographic records on each harvestmen taxon (except *O. spinosus*) are listed in Merino-Sáinz and Anadón (2008, 2013) and Merino-Sáinz et al. (2013a). In the faunistic analysis, each species was considered in accordance with its general distribution as Iberian endemic, European element, or Holarctic element (see Merino-Sáinz and Anadón 2008). Specimens identified as *Trogulus nepaeformis* belong to a related undescribed species probably endemic to the Iberian Peninsula, according to Schönhofer and Martens (2010). However, distribution data on this undescribed species are not available; thus, the chorological type (European element) is retained to compare data with previous articles (Merino-Sáinz and Anadón 2008, 2013) and to test whether biases exist.

We conducted a hierarchical cluster analysis (group average clustering algorithm, see algorithm choice discussion in Clarke and Gorley 2006) on similarity matrices, in order to compare this subset with the data obtained through planned, standardised, and periodic samplings using pitfall traps in several locations in the north of the Iberian Peninsula (Merino-Sáinz and Anadón 2013). The inventory of each locality included all of the harvestmen species sampled using the pitfall traps in that locality; subset inventory included all of the harvestmen species present in the unplanned events collection. Only qualitative data on species presence, rather than abundance data, were used in the analysis, and the similarity matrices were calculated using a species-presence Sørensen index (Sørensen 1948) (only positive results, i.e., species present in a pair of inventories, incremental similarity between inventories, species absent from both inventories -double negative- don't). Data were not standardised through sampling efforts, because the aims of the analysis were to compare the results of standardised sampling data with unplanned sampling data employing very different sampling and identification efforts. The analysis was carried out using the PRIMER V6 software (Clarke and Gorley 2006).

**Study extent description:** Harvestmen specimens included in the subset came from different localities in the Iberian Peninsula, at different distances from one another, and were collected at different dates between 1977 and 2011. Nonetheless, most of them came from the north of the Iberian Peninsula, and all of them came from the northern half (see Figure 1). The heterogeneity of the localities, most of which are only represented by a single sample or even only an isolated specimen, means that a general list of localities is not useful to short data exposition; rather, the locations are listed beside each specimen in Supplementary material 1 - Appendix A (supplementary file <http://hdl.handle.net/10651/24734>).

Harvestmen data obtained through planned collection using pitfall traps and deposited in the BOS Arthropod Collection included specimens collected from the Muniellos Biosphere Reserve between 2000 and 2002 (Merino-Sáinz and Anadón 2008) and from several locations in the provinces of Asturias (Muros, Oviedo, Villar), Cantabria (Vioño) and Pontevedra (Panjón) collected between 2009 and 2011 (Merino-Sáinz and Anadón 2013, Merino-Sáinz et al. 2013c). We also referred to published results of standardised pitfall samplings in western Asturias (Illano: Rosa García et al. 2009a,b, 2010a,b) for comparison purposes, as they were collected from the same areas (see Figure 1).

**Sampling description:** We studied a data subset of the harvestmen specimens in the BOS Arthropod Collection at the University of Oviedo that had been directly collected (by hand) on entomological field trips by students and lecturers from this department (listed in Supplementary material 1: <http://hdl.handle.net/10651/24734>, beside each specimen). This subset also included our own data obtained using diverse methods—collecting directly by hand, beating vegetation over an upturned umbrella, and using Berlese funnels, light traps, Malaise traps, and sieves; only 6% of collections used pitfall traps. Therefore, the specimens included in this study did not derive from harvestmen-targeted research projects, theses, or historical collections, but were collected at random with no prior sampling design.

## Quality control description

### Taxonomic identification

Specimens were identified by I. Merino-Sáinz using an Olympus SZX-ILLK200 stereoscopic microscope and the appropriate literature (Dresco 1948, 1954, Kraus 1961, Rambla 1959, 1967, 1973, 1976, 1980a, b, 1985, 1986, Sankey and Savory 1974, Martens 1978, 1982, Feliú 1981, Prieto 1990, 2004, Stol 2005, Pinto-Da-Rocha et al. 2007, Prieto and Fernández 2007, and Murienne and Giribet 2009).

### Digitisation quality control

The data quality control measures adopted throughout the digitisation process were described in the data-paper of the full dataset (Merino-Sáinz et al. 2013c) and in the digitisation workflow explication in Torralba-Burrial and Ocharan (2013). These controls included the validation and cleaning of geographic, taxonomic, and additional data associated with the harvestmen specimens (Merino-Sáinz et al. 2013c).

## Subset description

**Metadata language:** English

**Date of metadata creation:** 2014-02-05

**Hierarchy level:** Subset

**Metadata distribution:** [http://www.gbif.es:8080/ipt/resource.do?r=bos-opi\\_unplanned\\_collection\\_events](http://www.gbif.es:8080/ipt/resource.do?r=bos-opi_unplanned_collection_events)

**Format name metadata:** Ecological Metadata Language (EML) and HTML in web.

**Data distribution:** BOS-Opi dataset <http://www.gbif.es:8080/ipt/archive.do?r=Bos-Opi>

**Subset codes in dataset:** BOS-Opi codes 493-960.

**Publication date of data:** 2013-07-04

**Update police:** Subset will not be updated.

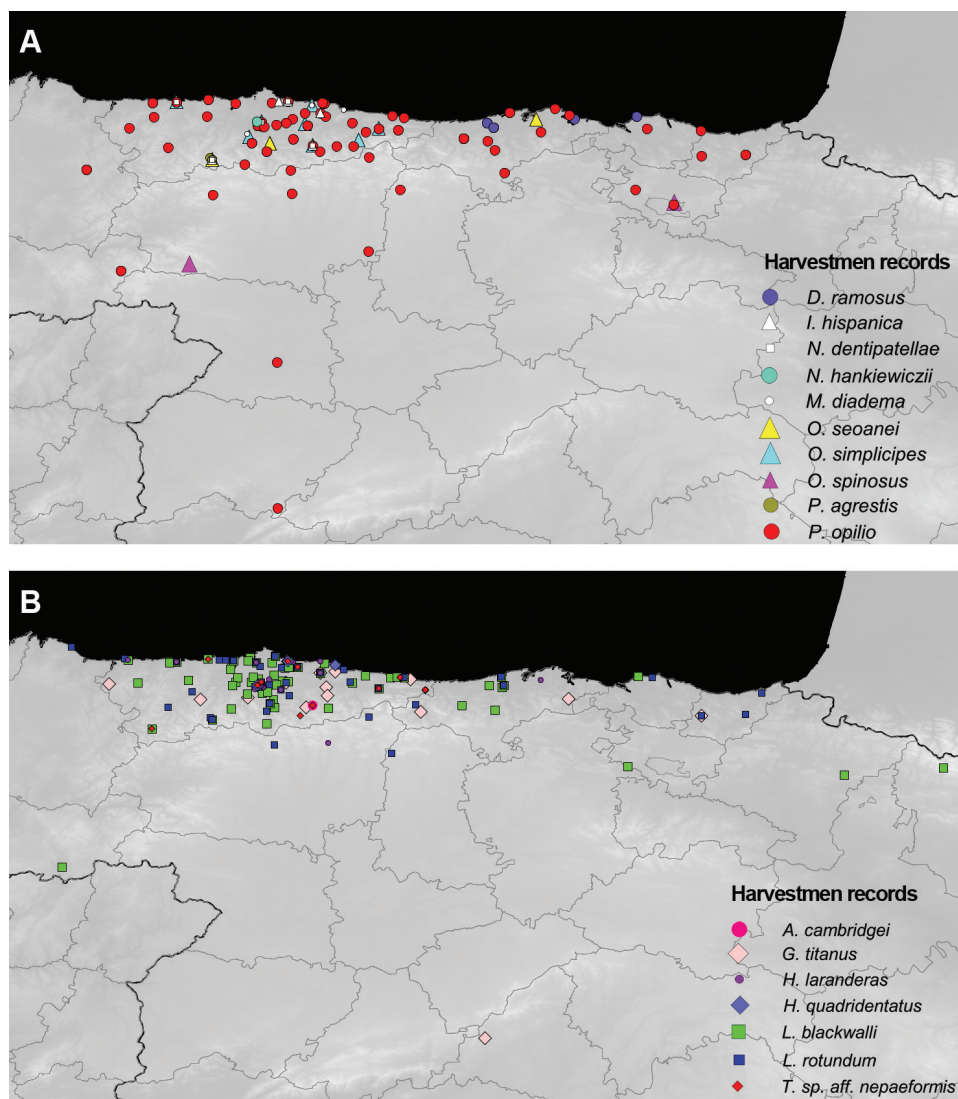
**Licences of use:** This BOS Arthropod Collection of University of Oviedo (Spain): Opiliones unplanned collection events subset, as part of BOS Arthropod Collection Dataset: Opiliones (BOS-Opi) dataset is made available under the Open Data Commons Attribution License: <http://www.opendatacommons.org/licenses/by/1.0/>.

## Data analysis

### Noteworthy records

In Figure 3, we have mapped the locations where each harvestmen species was found (listed in Supplementary material 1, <http://hdl.handle.net/10651/24734>), in order to facilitate rapid graphic assessment.





**Figure 3.** Distribution of harvestmen records in the unplanned collection events. **A** Ischyropsalididae, Nemastomatidae and Phalangidae **B** Scleromatidae and Trogulidae.

These records do not increase the harvestmen checklists for the provinces of Asturias and Cantabria (Merino-Sáinz and Anadón 2013), where planned, periodic, and standardised harvestmen-targeted sampling using pitfall traps have been carried out. However, the first records of five species are provided for other provinces that do not have this type of periodic targeted samplings. Table 2 lists the provinces with records for each species in this subset, indicating the first provincial records with an asterisk.

*Odiellus spinosus* is thus recorded for the first time in the provinces of León and Burgos. In Asturias, this species was found in Illano (Rosa García et al. 2009a,b,

**Table 2.** Presence of each harvestmen species by province according to data included in this data subset.

	Orense	Lugo	Asturias	León	Zamora	Salamanca	Cantabria	Palencia	Burgos	Vizcaya	Guipúzcoa	Álava	Navarra	Huesca	Madrid
<i>Ne. dentipatellae</i>			X												
<i>N. hankiewiczii</i>			X												
<i>T. nepaeformis</i>			X												
<i>A. cambridgei</i>			X												
<i>I. hispanica</i>			X												
<i>Ph. opilio</i>	X*	X*	X	X	X*	X*	X	X*	X	X*	X*	X*			
<i>M. diadema</i>			X												
<i>P. agrestis</i>			X												
<i>O. simplicipes</i>			X												
<i>O. seoanei</i>			X				X*								
<i>O. spinosus</i>				X*				X*							
<i>G. titanus</i>			X				X				X				X
<i>D. ramosus</i>							X			X*					
<i>L. blackwalli</i>	X*		X				X			X		X	X	X	
<i>L. rotundum</i>		X	X	X			X	X*	X	X	X				
<i>H. laranderas</i>			X	X			X								
<i>H. quadridentatus</i>			X												
<i>Paramiopsalis</i> sp.			X												

\* first records.

2010a,b), but was absent in the pitfall samplings from Muniellos (Merino-Sáinz and Anadón 2008) and Central Asturias (Merino-Sáinz and Anadón 2013). Endemic *Odiellus seoanei* was confirmed in Cantabria with the detection of a male specimen. Previously, there had only been a single, old data record by Fernández-Galiano (1910) based on one immature specimen; that record was questionable due to the variability in taxonomic characters of immature *Odiellus* (Santos et al. 2008).

The first finding of *Leiobunum rotundum* in Palencia was not surprising, as this European species is widespread throughout the north of the Iberian Peninsula (Prieto and Fernández 2007); its absence in this province can be attributed instead to the shortage of data on Iberian harvestmen. Similar circumstances explain the first record of *Leiobunum blackwalli* in Orense; this species is widespread in the north of the Iberian Peninsula, with records in the neighbouring Portuguese districts of Vila Real and Bragança (Prieto and Fernández 2007).

The fact that this data subset includes the first records of *Phalangium opilio* in eight provinces is another example of the scarcity of data on harvestmen throughout the Iberian Peninsula. *P. opilio* is a Holarctic species distributed throughout the peninsula from Galicia to Catalonia, with records in Portugal, Central Spain, and the Balearic Island (Kraus 1961), although specific information is lacking for several provinces.

Therefore, it is safe to state that the digitisation of such unplanned collections has the potential to address existing gaps in knowledge.

The data subset also includes several records older than the first published records of some species in Asturias and Cantabria provinces, confirm earlier studies. Thus, we provide older records for three harvestmen species (*M. diadema*, *H. laranderas*, and *P. agrestis*) first reported in Asturias in 2008 (Merino-Sáinz and Anadón 2008) and for another two species reported in the same paper, though erroneously identified (*O. simplicipes* specimens formerly identified as *O. ruentalis*, and *O. seoanei* specimens formerly identified as *O. spinosus*; see Merino-Sáinz and Anadón 2013). Moreover, one *H. laranderas* female from Cantabria with data collected in 1982 was included (the first record in this province dates from 2009: Merino-Sáinz and Anadón 2013).

### Are there biases?

In the area covered by this data subset of Opiliones, systematic sampling has been conducted in seven localities; therefore, this subset should include the species caught in these samples (see Merino-Sáinz and Anadón 2013). The composition and frequency of species in this unplanned, non-harvestmen-targeted subset with no sampling design show differences from the data derived from periodic pitfall sampling in the north of the Iberian Peninsula (Rambla 1985, Rosa García et al. 2009a, b, 2010a,b, Merino-Sáinz and Anadón 2013). Since specimens were collected directly by hand, it was possible to obtain information about the habitat choice and habitat use of several harvestmen species, for which there was scarce data from pitfall traps.

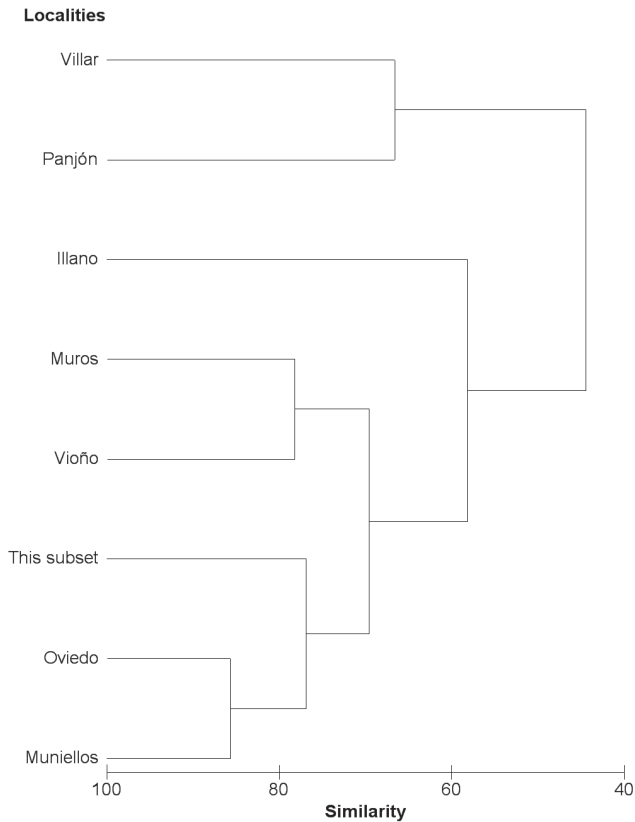
Table 3 shows that this subset of unplanned collection events, with a similar number of specimens, includes only one species fewer than the systematic study on Opiliones from the Muniellos Biosphere Reserve (Merino-Sáinz and Anadón 2008, 2009), the richest inventory of available studies on the area (Merino-Sáinz and Anadón 2013). Therefore, the unplanned collection events subset contains more species than any other listed study excluding Muniellos, even though the number of specimens is less than any of them by an order of magnitude. The next subset in the number of species, Oviedo, comprises 16 species with 15 times more studied specimens (Merino-Sáinz and Anadón 2013). Species richness and identity make this subset more similar to the inventories from Oviedo and Muniellos than to the remainder (see Figure 4). The differences with respect to the Muniellos Biosphere Reserve inventory are, on the one hand, the absence of three species – *Hadziana clavigera* (Simon), *Sabacon franzi* Roewer, and *Oligolophus hanseni* (Kraepelin) – from this subset and, on the other hand, the absence of *H. quadridentatus*, *D. ramosus*, and *O. spinosus* from Muniellos (specimens identified as *O. spinosus* in Muniellos are currently considered to belong to *O. seoanei*: Merino-Sáinz and Anadón 2013). Differences with respect to the Oviedo inventory are due to four species that were not located in planned collection events using pitfall traps (*Paramiopsalis* sp., *D. ramosus*, *O. spinosus*, and *M. diadema*); the last one might be present in the area according to its distribution and habitat preferences



**Table 3.** Number of harvestmen specimens and species with planned collection events (Oviedo, Muniellos, Illano, Muros and Vioño) and this subset.

	This subset	Oviedo	Muniellos	Illano	Muros	Vioño
Specimens	536	8452	770	1641	2687	2329
Species richness	18	16	19	14	13	12

Data sources of harvestmen data from planned collection events: Merino-Sáinz and Anadón (2008, 2013); Merino-Sáinz et al. 2013c, Rosa García et al. (2009a, b, 2010a, b).

**Figure 4.** Cluster hierarchical analysis with harvestmen presence data from seven locations with planned collection events and this general subset.

(see Merino-Sáinz et al. 2013b). *S. franzi* and *H. clavigera* were likewise located in planned collection events in Oviedo. *S. franzi* was also located with systematic standardised sampling in Muros and Illano; it coexists in the latter location with *Paroligolophus meadii* (O.P.-Cambridge), *O. hansenii*, and *Iberosiro* sp. Bivort and Giribert, without data in this digitised subset. All species collected in the other locations using standardised sampling protocols were also included in this general subset.

These results also show that some taxa are not usually found in non-harvestmen-targeted (or soil entomofauna-targeted) samplings; this was the case for small, inconspicuous species that occupy edaphic niches throughout their entire life cycle (see previous comments on absent species). Instead, other species were better represented and appeared more frequently in the present subset, for example, large species with long legs and arboreal or shrub habits, at least during the adult phase. Taxa with these features comprised almost 56% of the species in this data subset, including the three most frequently captured species. Thus, the major abundance of adult specimens of *Ph. opilio*, *L. rotundum* and *L. blackwalli* in the subset would be in line with observations in other geographic areas about vertical migration patterns throughout their life cycle (Todd 1949, Williams 1962, Allard and Yeargan 2005). The higher relative frequency of adults in these species is related to the use of active sampling methods, given that harvestmen spend more time in higher vegetation strata during their adult phase and are larger and more conspicuous than the immature specimens that predominate in soil and pitfall traps (e.g. Merino-Sáinz and Anadón 2013). However, *H. laranderas* and *T. sp. aff. nepaeformis* are linked to edaphic habitats throughout their entire life cycle and present a cryptic coloration; each one species represents 4-5% of the specimens in this subset, occupying the fourth and fifth positions in the list of species in terms of the number of specimens collected (Table 1).

These biases are due to the differences in body size and life history of each harvestmen species and should be considered in biogeographic analyses with accidental occurrences (unplanned samples). In the particular case of this digitised subset, European elements comprised 50% of specimens, 39% were Iberian endemics, and 11% were Holarctic taxa—percentages which are slightly different from those resulting from pitfall trapping in the same area (Merino-Sáinz and Anadón 2013: 44%, 44%, and 12%, respectively). In both cases, namely the use of unplanned, non-standardised collections and the use of pitfall trapping, several Iberian endemic taxa with narrow niches (e.g., subterranean/hypogeous taxa) were absent; thus, these methods are not suitable for obtaining information about those taxa. A summary of advantages and problems arising from the digitisation of this subcollection of unplanned sampling events is provided in Table 4.

**Table 4.** Main observations on the advantages and problems arising from the digitisation of unplanned collections in the case study of Iberian harvestmen in the BOS Arthropod Collection.

Advantages	Problems
Less effort (identification, digitisation) needed: lower number of specimens than planned, periodical, pitfall samples	Some biases detected in harvestmen present in the subcollection (body size, life history, phases of life cycle)
Similar species richness	Does not provide full phenological data
Justification of the investment made to collect/house/study such collections	Not suitable for taxa with very narrow niches (e.g., subterranean/hypogean taxa)
Bridges knowledge gaps	

## Conclusions

A small subcollection of harvestmen from the north of the Iberian Peninsula, gathered using non-Opiliones-targeted sampling methods and in many cases by non-specialist collectors, presented a high species richness similar to planned, periodic, and costlier studies. This subcollection enabled us to extend our knowledge on the distribution of 18 species. The 536 specimens in the subset showed very interesting faunistic results, while less effort was exerted on identification and digitisation than in planned, periodic collection events using pitfall traps. The data subset contained six first provincial records of various species; *Ph. opilio* locations in eight provinces without previous data were also provided. Nevertheless, we also detected some drawbacks to this type of data collection; collection was biased towards adults of larger species (with long legs or wide bodies) occupying shrubby or arboreal habitats, which may also affect the biogeographic analysis of the dataset. Nevertheless, this study highlights the importance of the general biodiversity collections in museums and at universities and the need to digitise their specimens, including the data from non-targeted, or unplanned, samplings, especially when poorly studied groups are involved. The digitisation of unplanned collections can help to justify the investments made to collect, house, and study such collections. Moreover, it is important to keep in mind that most of the collections at the university/museum, NGO, and amateur scientist levels are not comprised of data collected through planned events, but mainly through unplanned events. The digitisation of such unplanned collections has great potential to (1) bridge gaps in existing knowledge, and (2) strengthen existing understanding about the status of biodiversity.

## Acknowledgements

The authors wish to express their gratitude to all of the collectors (listed in Supplementary material 1: <http://hdl.handle.net/10651/24734>) who deposited harvestmen specimens in the BOS Arthropod Collection. Vishwas Chavan and anonymous reviewers enhanced the paper with their suggestions.

## References

### 1. References cited in the manuscript or metadata document

- Allard CM, Yeargan KV (2005) Diel activity patterns and microspatial distribution of the harvestman *Phalangium opilio* (Opiliones, Phalangiidae) in soybeans. *The Journal of Arachnology* 33: 745–752. doi: 10.1636/T04-17.1
- Chapman AD, Wiczorek J (Eds) (2006) Guide to Best Practices for Georeferencing. Global Biodiversity Information Facility, Copenhagen, 90 pp. [http://www.gbif.org/orc/?doc\\_id=1288](http://www.gbif.org/orc/?doc_id=1288)
- Clarke KR, Gorley RN (2006) PRIMER v6: User Manual/Tutorial. PRIMER-E Ltd. Plymouth.



- Beaman RS, Cellinese N (2012) Mass digitization of scientific collections: New opportunities to transform the use of biological specimens and underwrite biodiversity science. *ZooKeys* 209: 7–17. doi: 10.3897/zookeys.209.3313
- Berents P, Hamer M, Chavan V (2010) Towards demand-driven publishing: approaches to the prioritization of digitization of natural history collection data. *Biodiversity Informatics* 7: 113–119.
- Dresco E (1948) Remarques sur le genre *Dicranopalpus* Dol. et description de deux espèces nouvelles (Opiliones). *Bulletin du Muséum national d'histoire naturelle, Paris, 2e série* 20(4): 336–342.
- Dresco E (1954) Sur le genre *Gyas* (Opiliones). *Bulletin du Muséum national d'histoire naturelle, Paris, 2e série* 26(1): 85–92.
- Feliú J (1981) Contribución al conocimiento de los Opiliones de Navarra. Mc Thesis, Universidad de Navarra, Pamplona, Spain.
- Fernández-Galiano E (1910) Datos para el conocimiento de la distribución geográfica de los arácnidos en España. *Memorias de la Real Sociedad Española de la Historia Natural* 6(5): 343–424.
- Kraus VO (1961) Die Weberknechte der Iberischen Halbinsel (Arach., Opiliones). *Senckenbergiana Biologica* 42(4): 331–363.
- Martens J (1978) Weberknechte, Opiliones. *Gustav Fischer Verlag, Jena, Mainz*, 465 pp.
- Martens J (1982) Europäische Arten der Gattung *Sabacon* Simon 1879. *Senckenbergiana Biologica* 63(3\4): 265–296.
- Merino-Sáinz I, Anadón A (2008) La fauna de Opiliones (Arachnida) de la Reserva Integral Natural de Muniellos (Asturias) y del Noroeste de la Península Ibérica. *Boletín de la Sociedad Entomológica Aragonesa* 43: 199–210.
- Merino-Sáinz I, Anadón A (2009) Primera cita del género *Paramiopsalis* Juberthie, 1962 (Arachnida: Opiliones, Sironidae) para Asturias (España). *Boletín de la Sociedad Entomológica Aragonesa* 45: 556–558.
- Merino-Sáinz I, Anadón A (2013) La fauna de Opiliones (Arachnida) de Asturias y Cantabria (España): catálogos e importancia de las especies y de los endemismos. *Revista Ibérica de Aracnología* 23: 57–77.
- Merino-Sáinz I, Fernández-Álvarez FA, Prieto CE (2013a) Nuevos datos sobre *Megabunus diadema* (Fabricius, 1779) (Opiliones: Phalangidae). *Revista Ibérica de Aracnología* 22: 102–106.
- Merino-Sáinz I, Fernández-López J, Rodríguez-Martínez S, Da Silva G, Fernández González A, Fernández Menéndez D, Torralba-Burrial A (2013b) Opiliones forestales edáficos de la cuenca media-alta del río Támega (noreste de Portugal). *Revista Ibérica de Aracnología* 22: 117–120.
- Merino-Sáinz I, Anadón A, Torralba-Burrial A (2013c) Harvestmen of the BOS Arthropod Collection of the University of Oviedo (Spain) (Arachnida, Opiliones). *ZooKeys* 341: 21–36. doi: 10.3897/zookeys.341.6130
- Murienne J, Giribet G (2009) The Iberian Peninsula: ancient history of a hot spot of mite harvestmen (Arachnida: Opiliones: Cyphophthalmi: Sironidae) diversity. *Zoological Journal of the Linnean Society* 156: 785–800. doi: 10.1111/j.1096-3642.2008.00512.x

- Pinto Da Rocha R, Machado G, Giribet G (2007) *Harvestmen: The Biology of Opiliones*. Harvard University Press, Cambridge, Massachusetts, 597 pp.
- Prieto C (1990) The genus *Ischyropsalis* C. L. Koch (Opiliones, Ischyropsalididae) on the Iberian Península. I. Non-troglobitic species. *Acta Zoologica Fennica* 190: 315–320.
- Prieto C (2003) Primera actualización de la Check-List de los Opiliones de la Península Ibérica e Islas Baleares. *Revista Ibérica de Aracnología* 8: 125–141.
- Prieto C (2004) El género *Nemastomella* Mello-Leitao 1936 (Opiliones: Dyspnoi: Nemastomatidae) en la Península Ibérica, con descripción de la primera especie de Andalucía. *Revista Ibérica de Aracnología* 9: 107–121.
- Prieto CE, Fernández J (2007) El género *Leiobunum* C.L. Koch, 1839 (Opiliones: Eupnoi: Sclerosomatidae) en la Península Ibérica y el norte de África, con la descripción de tres nuevas especies. *Revista Ibérica de Aracnología* 14: 135–171.
- Rambla M (1959) Contribuciones al Estudio de los Opiliones de la Fauna Ibérica. Opiliones de la Sierra de Guadarrama. Publicaciones del Instituto de Biología Aplicada, Barcelona 29: 59–110.
- Rambla M (1967) Opiliones de Portugal. *Revista de Biología. Revista Brasileira e Portuguesa de Biología em geral* 6(1–2): 1–34.
- Rambla M (1973) Contribución al conocimiento de los Opiliones de la Fauna Ibérica. Estudio de las especies Ibéricas de los géneros *Odiellus* y *Lacinius* (Familia Phalangiidae). Publicaciones del Instituto de Biología Aplicada, Barcelona 54: 5–38.
- Rambla M (1976) La especie *Oligolophus agrestis* (Meade, 1855) en la Península Ibérica (Arachnida, Opiliones, Phalangiidae, Oligolophinae). Publicaciones del Departamento de Zoología. Universidad de Barcelona, Facultad de Biología, Barcelona 1: 23–27.
- Rambla M (1980a) Contribución al conocimiento de la fauna cavernícola del País Vasco. Arachnida, Opiliones. *Kobie, Grupo Espeleológico Vizcaíno, Bilbao* 10: 529–533.
- Rambla M (1980b) Les Nemastomatidae (Arachnida, Opilions) de la Péninsule Ibérique. V. *Nemastoma scabriculum* Simon 1879 et *Nemastoma hankiewiczii* Kulczynski 1909. *Colloque Arachnologique, Barcelona* 9: 195–202.
- Rambla M (1985) Artrópodos epigeos del Macizo de San Juan de la Peña (Jaca, Huesca). IV. Opiliones. *Pirineos* 124: 87–168.
- Rambla M (1986) Nuevos datos sobre *Dicranopalpus ramosus* (Simon, 1909) (Arachnida, Opiliones, Phalangiidae). In: Barrientos JA (Ed) *Actas del X Congreso Internacional de Aracnología* (Jaca, Spain, September 1986) 1: 373–382.
- Rosa García R, Jáuregui BM, García U, Osoro K, Celaya R (2009a) Effects of livestock breed and grazing pressure on ground-dwelling arthropods in Cantabrian heathlands. *Ecological Entomology* 34: 466–475. doi: 10.1111/j.1365-2311.2008.01072.x
- Rosa García R, Jáuregui BM, García U, Osoro K, Celaya R (2009b) Responses of arthropod fauna assemblages to goat grazing management in northern Spanish heathlands. *Environmental Entomology* 38(4): 985–995. doi: 10.1603/022.038.0405
- Rosa García R, Ocharan FJ, García U, Osoro K, Celaya R (2010a) Arthropod fauna on grassland-heathland associations under different grazing managements with domestic ruminants. *Comptes Rendus Biologies* 333: 226–234. doi: 10.1016/j.crv.2009.12.008

- Rosa García R, Ocharan FJ, Jáuregui BM, García U, Osoro K, Celaya R (2010b) Ground-dwelling arthropod communities present in three types of Cantabrian (NW Spain) heathland grazed by sheep or goats. *European Journal of Entomology* 107: 219–227. doi: 10.14411/eje.2010.029
- Schönhofer AL, Martens J (2010) Hidden Mediterranean diversity: Assessing species taxa by molecular phylogeny within the opilionid family Trogulidae (Arachnida, Opiliones). *Molecular Phylogenetics and Evolution* 54(1): 59–75. doi: 10.1016/j.ympev.2009.10.013
- Sankey JHP, Savory TH (1974) *British Harvestmen Arachnida: Opiliones*. Synopses of the British Fauna N°4. The Linnean Society of London by Academic Press London and New York. 76 pp.
- Santos L, Martín R, Prieto C (2008) Los *Odiellus* (Arachnida: Opiliones: Phalangiidae) de la Península Ibérica. IX Jornadas del Grupo Ibérico de Aracnología.
- Sørensen T (1948) A method of establishing groups of equal amplitude in plant sociology based on similarity of species and its application to analyses of the vegetation on Danish commons. *Biologiske Skrifter* 5(4): 1–34.
- Stol I (2005) Ecology and body size of the parthenogenetic *Megabunus diadema* (Fabricius, 1779) (Opiliones: Phalangiidae) at Karmøy, Western Norway. *Fauna Norvegica* 25: 39–44.
- Todd V (1949) The habits and ecology of the British Harvestmen (Arach., Opiliones) with special reference to those of the Oxford. *Journal of Animal Ecology* 18(2): 209–216. doi: 10.2307/1600
- Torralba-Burrial A, Ocharan FJ (2013) Iberian Odonata distribution: data of the BOS Arthropod Collection (University of Oviedo, Spain). *ZooKeys* 306: 37–58. doi: 10.3897/zookeys.306.5289
- Universidad de Oviedo (2013-) BOS Arthropod Collection Dataset: Opiliones (BOS-Opi). 3772 data records. Contributed by: Merino-Sáinz I, Anadón A, Torralba-Burrial A, Fernández-Álvarez FA, Melero Cimas VX, Monteserín Real S, Ocharan Ibarra R, Rosa García R, Vázquez Felechosa MT, Ocharan FJ. <http://www.gbif.es:8080/ipt/archive.do?r=Bos-Opi>, <http://www.unioviedo.es/BOS/Zoologia/artropodos/opiliones>, version 1.0 (last updated on 2013-06-30), GBIF key: <http://gbrds.gbif.org/browse/agent?uuid=cc0e6535-6bb4-4703-a32c-077f5e1176cd>, Data paper ID: doi: 10.3897/zookeys.341.6130
- Williams GC (1962) Seasonal and diurnal activity of harvestmen (Phalangida) and spiders (Araneida) in contrasted habitats. *Journal of Animal Ecology* 31: 23–42. doi: 10.2307/2330

## 2. References using data included in this data subset

- Merino-Sáinz I, Fernández-Álvarez FA, Prieto CE (2013a) Nuevos datos sobre *Megabunus diadema* (Fabricius, 1779) (Opiliones: Phalangiidae). *Revista Ibérica de Aracnología* 22: 102–106.
- Merino-Sáinz I, Anadón A, Torralba-Burrial A (2013c) Harvestmen of the BOS Arthropod Collection of the University of Oviedo (Spain) (Arachnida, Opiliones). *ZooKeys* 341: 21–36. doi: 10.3897/zookeys.341.6130



## **Supplementary material I**

### **Harvestmen specimens included in this unplanned collection events subset.**

Authors: Izaskun Merino-Sáinz, Antonio Torralba-Burrial, Araceli Anadón

Data type: Specimens data.

Explanation note: Alternative link for download: <http://hdl.handle.net/10651/24734>

Copyright notice: This dataset is made available under the Open Database License (<http://opendatacommons.org/licenses/odbl/1.0/>). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: doi: 10.3897/zookeys.404.6520.app1



# DNA barcoding as a screening tool for cryptic diversity: an example from *Caryocolum*, with description of a new species (Lepidoptera, Gelechiidae)

Peter Huemer<sup>1,†</sup>, Ole Karsholt<sup>2,‡</sup>, Marko Mutanen<sup>3,§</sup>

**1** Tiroler Landesmuseen Betriebsges.m.b.H., Naturwissenschaftliche Sammlungen, Feldstr. 11a, A-6020 Innsbruck, Austria **2** Zoological Museum, Natural History Museum of Denmark, Universitetsparken 15, DK-2100 Copenhagen **3** Biodiversity Unit, Department of Biology, University of Oulu, Oulu, Finland

† <http://zoobank.org/4E6F001E-DB9D-460C-AB39-0CE7CC773B59>

‡ <http://zoobank.org/BEE2C0B9-8156-411F-A1D6-79AA0ABA1B9D>

§ <http://zoobank.org/627B15F5-FFE5-40DE-9A78-FF2FD900A7B1>

Corresponding author: Peter Huemer ([p.huemer@tiroler-landesmuseen.at](mailto:p.huemer@tiroler-landesmuseen.at))

Academic editor: A. Zilli | Received 11 February 2014 | Accepted 7 April 2014 | Published 24 April 2014

<http://zoobank.org/E1B904C8-A4AE-42E9-90B0-60E0B1D83042>

**Citation:** Huemer P, Karsholt O, Mutanen M (2014) DNA barcoding as a screening tool for cryptic diversity: an example from *Caryocolum*, with description of a new species (Lepidoptera, Gelechiidae). ZooKeys 404: 91–111. doi: 10.3897/zookeys.404.7234

## Abstract

We explore the potential value of DNA barcode divergence for species delimitation in the genus *Caryocolum* Gregor & Povolný, 1954 (Lepidoptera, Gelechiidae), based on data from 44 European species (including 4 subspecies). Low intraspecific divergence of the DNA barcodes of the mtCOI (*cytochrome c oxidase 1*) gene and/or distinct barcode gaps to the nearest neighbor support species status for all examined nominal taxa. However, in 8 taxa we observed deep splits with a maximum intraspecific barcode divergence beyond a threshold of 3%, thus indicating possible cryptic diversity. The taxonomy of these taxa has to be re-assessed in the future. We investigated one such deep split in *Caryocolum amaurella* (Hering, 1924) and found it in congruence with yet unrecognized diagnostic morphological characters and specific host-plants. The integrative species delineation leads to the description of *Caryocolum crypticum* **sp. n.** from northern Italy, Switzerland and Greece. The new species and the hitherto intermixed closest relative *C. amaurella* are described in detail and adults and genitalia of both species are illustrated and a lectotype of *C. amaurella* is designated; a diagnostic comparison of the closely related *C. iranicum* Huemer, 1989, is added.



## Keywords

Lepidoptera, Gelechiidae, *Caryocolum*, species delineation, integrative taxonomy, DNA barcode, morphology, Europe

## Introduction

The genus *Caryocolum* Gregor & Povolný, 1954 is one of the most species-rich genera of European Gelechiidae (Huemer and Karsholt 2010). Having been revised in monographic papers (Klimesch 1953–1954, Huemer 1988), its taxonomy seemed well established. However, in the last decade new species were found in, e.g. Sicily, southern France and Greece (Bella 2008, Grange and Nel 2012, Huemer and Nel 2005, Huemer and Karsholt 2010) raising the number of described species to 51. Most of the species are considered indisputable based on their morphology and distinct biology – as far as known, these species are closely linked to Caryophyllaceae as their exclusive larval host-plant family. We investigate, for the first time in *Caryocolum*, the congruence of traditional morphological species delineation and molecular data from the COI barcode region for a vast majority of the European fauna, covering altogether 44 species, including four subspecies. Surprisingly, the potential for cryptic diversity proved extraordinarily high for a supposedly well-known genus and we newly describe one of the hitherto overlooked species.

## Material and methods

Extensive generic descriptions and diagnoses of European species of *Caryocolum* have been published in several reviews, particularly Huemer and Karsholt (2010) and Huemer (1988), and are thus not repeated here.

**Specimens.** Our study is based on about 50 specimens of the *Caryocolum amaurella* (Hering, 1924) species-group and an uncounted number of European *Caryocolum*, exceeding 1000 specimens, but only partially used for genetic analysis (see below). Most of the material was traditionally set and dried or alternatively spread; a few specimens are only pinned. Genitalia preparations followed standard techniques (Robinson 1976) adapted for male genitalia of Gelechiidae and (some) female genitalia of *Caryocolum* by the so-called “unrolling technique” (Pitkin 1986, Huemer 1987).

**DNA Barcodes.** Full-length lepidopteran DNA barcode sequences are a 648 base-pair long segment of the 5' terminus of the mitochondrial COI gene (*cytochrome c oxidase I*). DNA samples (dried leg) were prepared according to the accepted standards. Legs from 250 specimens of *Caryocolum* were processed at the Canadian Centre for DNA Barcoding (CCDB, Biodiversity Institute of Ontario, University of Guelph) to obtain DNA barcodes using the standard high-throughput protocol described in deWaard et al. (2008). Sequences longer than 500 bp were included in the analysis.

Successfully sequenced voucher specimens are listed in Suppl. material 1. Sequences were submitted to GenBank; further details including complete voucher data and images can be accessed in the public dataset “Lepidoptera of Europe *Caryocolum*” [dx.doi.org/10.5883/DS-LECARY](https://dx.doi.org/10.5883/DS-LECARY) in the Barcode of Life Data Systems (BOLD; Ratnasingham and Hebert 2007). Degrees of intra- and interspecific variation in the DNA barcode fragment were calculated under Kimura 2 parameter (K2P) model of nucleotide substitution using analytical tools in BOLD systems v3.0. (<http://www.boldsystems.org>). A neighbour-joining tree of DNA barcode data of European taxa was constructed using Mega 5 (Tamura et al. 2011) under the K2P model for nucleotide substitutions.

Photographic documentation. Photographs of the adults were taken with an Olympus SZX 10 binocular microscope and an Olympus E 3 digital camera and processed using the software Helicon Focus 4.3 and Adobe Photoshop CS4 and Lightroom 2.3. Genitalia photographs were taken with an Olympus E1 Digital Camera from Olympus BH2 microscope.

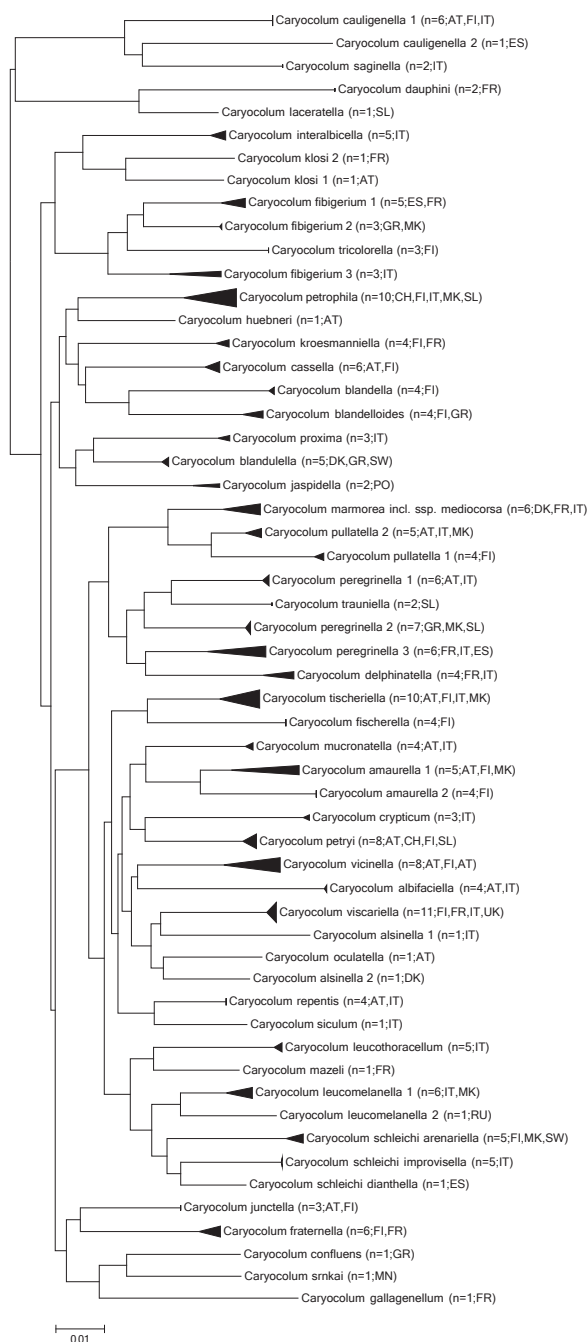
### Abbreviations of institutional collections

<b>BMNH</b>	The Natural History Museum (British Museum, Natural History) London (United Kingdom)
<b>TLMF</b>	Tiroler Landesmuseum Ferdinandeum, Innsbruck, Austria
<b>ZMUH</b>	Zoological Museum, University of Helsinki, Finland
<b>ZMUC</b>	Zoological Museum, Natural History Museum of Denmark, Copenhagen, Denmark
<b>ZMUO</b>	Zoological Museum, University of Oulu, Finland

## Results

### Molecular analysis

Forty-four of 51 European species were successfully sequenced, resulting in a full-length barcode fragment for 191 specimens and more than 500 bp for further 26 specimens (Fig. 1, Table 1, Suppl. material 1). Nine shorter sequences were not included in the analysis and sequencing of 24 specimens failed. The maximum intraspecific K2P distance varies from 0% in several species to 6.27% in *C. fibigerium*. Ten species have a high maximum intraspecific divergence greater than 2%. In six species (newly described species excluded) with a medium divergence greater than 3% potential cryptic diversity should be investigated. Furthermore, the intraspecific divergence of more than 3% in *C. schleichi*, a species separated into 3 allopatric subspecies, is beyond variation typically found within species, supporting their status as valid species. The only



**Figure 1.** Neighbour-joining tree (Kimura 2 parameter, built with MEGA 5; cf. Tamura et al. 2011), with only sequences longer than 500 bp considered. The width of the triangles represents the sample size, and the depth the genetic variation within the cluster. Currently recognized conspecific taxa with maximum divergence greater than 3% are shown as separate clades. Source: DNA Barcode data from BOLD (Barcode of Life Database, cf. Ratnasingham and Hebert 2007).



**Table 1.** Intraspecific mean K2P (Kimura 2 Parameter) divergences, maximum pairwise distances and distance to nearest neighbor.

Species	Mean Intra-Sp	Max Intra-Sp	Nearest Neighbour	Nearest Species	Distance to NN
<i>C. alsinella</i>	4.85	4.85	PHLAE427-11	<i>C. oculatella</i>	3.81
<i>C. amaurella</i>	3.05	4.76	LEATC402-13	<i>C. mucronatella</i>	5.21
<i>C. blandella</i>	0.16	0.3	LEFIK150-10	<i>C. blandelloides</i>	5.78
<i>C. blandelloides</i>	0.4	0.81	LEFIB755-10	<i>C. blandella</i>	5.78
<i>C. blandulella</i>	0.21	0.46	LEATD656-13	<i>C. proxima</i>	3.94
<i>C. cassella</i>	0.42	0.61	PHLAI019-12	<i>C. blandulella</i>	5.07
<i>C. cauligenella</i>	1.99	6.95	PHLAA069-09	<i>C. saginella</i>	6.61
<i>C. confluens</i>	N/A	N/A	PHLAF489-11	<i>C. srnkai</i>	4.54
<i>C. crypticum</i>	0.21	0.31	LEATC-402-13	<i>C. mucronatella</i>	5.41
<i>C. dauphini</i>	0	0	PHLAB900-10	<i>C. laceratella</i>	5.29
<i>C. delphinatella</i>	1.02	1.39	PHLAI203-13	<i>C. marmorea mediocorsa</i>	4.57
<i>C. fibigerium</i>	3.4	6.27	LEFIF467-10	<i>C. tricolorella</i>	4.67
<i>C. fischerella</i>	0	0	LEFIC281-10	<i>C. tischeriella</i>	4.5
<i>C. fraternella</i>	0.47	1.7	PHLAI156-12	<i>C. junctella</i>	4.55
<i>C. gallagenellum</i>	N/A	N/A	PHLAI019-12	<i>C. blandulella</i>	6.54
<i>C. huebneri</i>	N/A	N/A	LEFIJ1014-11	<i>C. petrophila</i>	4.88
<i>C. interalbicella</i>	0.4	0.77	PHLAI156-12	<i>C. junctella</i>	5.55
<i>C. jaspidella</i>	1.08	1.08	PHLAI019-12	<i>C. blandulella</i>	4.39
<i>C. junctella</i>	0	0	LEFIF480-10	<i>C. fraternella</i>	4.55
<i>C. klosi</i>	4.25	4.25	PHLAA055-09	<i>C. interalbicella</i>	5.56
<i>C. kroesmanniella</i>	0.31	0.61	LEEUA184-11	<i>C. blandulella</i>	4.9
<i>C. laceratella</i>	N/A	N/A	PHLAI447-13	<i>C. dauphini</i>	5.29
<i>C. leucomelanella</i>	1.47	3.79	PHLAG331-12	<i>C. mazeli</i>	3.76
<i>C. leucothoracellum</i>	0.12	0.3	PHLAG331-12	<i>C. mazeli</i>	4.24
<i>C. marmorea mediocorsa</i>	0	0	LEEUA182-11	<i>C. marmorea</i>	0.3
<i>C. marmorea</i>	1	1.54	PHLAI203-13	<i>C. marmorea mediocorsa</i>	0.3
<i>C. mazeli</i>	N/A	N/A	LEATE421-13	<i>C. leucomelanella</i>	3.76
<i>C. mucronatella</i>	0.3	0.46	PHLAE427-11	<i>C. oculatella</i>	4.87
<i>C. oculatella</i>	N/A	N/A	LEEUA388-11	<i>C. alsinella</i>	3.81
<i>C. peregrinella</i>	3.58	5.69	PHLAB899-10	<i>C. trauniella</i>	3.93
<i>C. petrophila</i>	0.97	2.26	PHLAH147-12	<i>C. huebneri</i>	4.88
<i>C. petryi</i>	0.23	0.61	PHLAD576-11	<i>C. repentis</i>	3.85
<i>C. proxima</i>	0.41	0.61	PHLAI019-12	<i>C. blandulella</i>	3.94
<i>C. pullatella</i>	2.07	3.61	LEATC292-13	<i>C. marmorea</i>	3.12
<i>C. repentis</i>	0	0	PHLAE429-11	<i>C. siculum</i>	3.33
<i>C. saginella</i>	0	0	LEFIJ778-10	<i>C. cauligenella</i>	6.61
<i>C. schleichi dianthella</i>	N/A	N/A	PHLAD573-11	<i>C. schleichi improvisella</i>	3.42
<i>C. schleichi improvisella</i>	0.06	0.15	PHLSA085-11	<i>C. schleichi dianthella</i>	3.42
<i>C. schleichi arenariella</i>	0.77	1.24	PHLSA085-11	<i>C. schleichi dianthella</i>	3.74
<i>C. siculum</i>	N/A	N/A	PHLAD576-11	<i>C. repentis</i>	3.33
<i>C. srnkai</i>	N/A	N/A	PHLAG580-12	<i>C. confluens</i>	4.54
<i>C. tischeriella</i>	1.09	2.02	PHLAD576-11	<i>C. repentis</i>	4.01
<i>C. trauniella</i>	0	0	PHLAB622-10	<i>C. peregrinella</i>	3.93

Species	Mean Intra-Sp	Max Intra-Sp	Nearest Neighbour	Nearest Species	Distance to NN
<i>C. tricolorella</i>	0	0	PHLAI014-12	<i>C. fibigerium</i>	4.67
<i>C. vicinella</i>	1.48	2.7	PHLAF105-11	<i>C. leucomelanella</i>	5.36
<i>C. viscariella</i>	0.22	0.47	LEEU388-11	<i>C. alsinella</i>	4.16

other subspecies we have examined are nominotypical *C. marmorea* and the recently separated *C. marmorea mediocorsa* with a very low divergence of 0.3%.

Sequences of the COI barcode region of all analysed morphospecies reveal significant interspecific genetic distances with barcode gaps ranging from a minimum of 3.11% to the nearest neighbour (*C. pullatella* – *C. marmorea*) to a maximum of 6.61% (*C. saginella* – *C. cauligenella*).

## Taxonomy

The *Caryocolum amaurella* species-group as defined by Huemer (1988) differs from other congeners mainly by the characteristic shape of the sacculus, which is unique in the genus. Until now it only included *C. amaurella* and *C. iranicum* (Huemer 1988, 1989b). Based on the DNA barcode divergence and diagnostic morphological characters combined with biological data we describe the new species *C. crypticum*. Due to the mix-up of *C. crypticum* with *C. amaurella* in recent identification guides the latter species is also re-described here in detail.

### *Caryocolum* Gregor & Povolný, 1954

*Caryocolum* Gregor & Povolný, 1954: 87.

**Type species.** *Gelechia leucomelanella* Zeller, 1839: 138.

### *Caryocolum crypticum* sp. n.

<http://zoobank.org/5E1FB9E5-3A65-49C6-80BF-A5CA7C4FFF99>

[http://species-id.net/wiki/Caryocolum\\_crypticum](http://species-id.net/wiki/Caryocolum_crypticum)

Figs 2–3, 6–7, 10–11, 14–15

**Type material. Holotype:** ♀ (Fig. 2), Italia sept., Teriolis merid., Laatsch, 1000 m, 29.6.1987 e.l. (*Silene otites* 10.5.), leg. Huemer, slide GEL 1234 ♀ (TLMF).

**Paratypes. Italy:** 1 ♂, South Tyrol, Vinschgau, Schleiser Leiten, 1350 m, 6.7.2013, leg. Huemer, slide GEL 1215, dna barcode id TLMF Lep 12313 (TLMF); 1 ♂ [without abdomen], same data (TLMF); 1 ♀, same data, but 18.8.2013, slide GEL 1232, dna barcode id TLMF Lep 11883 (TLMF); 1 ♀, same data, but dna



**Figures 2–5.** Adults. **2** *Caryocolum crypticum* sp. n., holotype **3** *C. crypticum* sp. n., paratype, female, Greece **4** *C. amaurella*, male, Finland **5** *C. amaurella*, male, Austria.

barcode id TLMF Lep 11882 (TLMF); 1 male [without abdomen], 8 ♀, same data, but 7.9.2013 (TLMF); 1 ♂, South Tyrol, Taufers, 1300 m, 22.8.1978, leg. Burmann, slide GU 86/041 P. Huemer (TLMF). **Switzerland:** 2 ♀, Wallis, Martigny-Rosel, 460 m, 28.6.–14.7.1983 e.l. (*Silene otites*), leg. Whitebread (Naturhistorisches Museum Basel, Switzerland). **Greece:** 1 ♀, Larisa, Ossa Oros, 1.5 km N Spilia, 940 m, 13.6.1988 e.l. (*Silene nutans*), leg. Huemer (TLMF).

**Diagnosis.** *Caryocolum crypticum* sp. n. is externally similar to several other species of the genus and can be best recognized by the largely unmarked forewings with cream costal and tornal spots. From its closest relatives *C. amaurella* and *C. iranicum* it differs by the rusty brown distal half of the thorax and the concolorous tegulae, the dark brown forewings with rusty brown scales, and the cream colours of the costal and tornal spots. The male genitalia of *C. crypticum* are very similar to those of *C. amaurella* but the valva is more slender and slightly longer (see Figs 6–7, 10–11 versus 8–9, 12–13). The similar *C. iranicum* differs by the shape of the sacculus with almost straight dorsal margin (see Huemer 1989b: Figs 14–16). However, the most striking diagnostic characters of the new species are found in the female genitalia which differ from *C. amaurella* particularly by the short lateral sclerites of the ductus bursae and the much longer and more slender signum hook (see Figs 14–15 versus 16–17). The



female genitalia furthermore differ from *C. iranicum* by the weakly cup-shaped rather than funnel-shaped antrum, shorter lateral sclerites of the ductus bursae, and the shorter apophysis anterior which is almost twice the length of segment VIII in *C. iranicum*.

**Description.** Adult (Figs 2–3). Wingspan 10.5–14 mm. Segment 2 of labial palpus with a few cream-coloured scales on inner and upper surface, blackish brown on outer and lower surface; segment 3 almost black with light tip. Antenna black, indistinctly lighter ringed. Head with light yellow frons and black neck; thorax blackish brown with rusty brown posterior part; tegulae rusty brown except for blackish brown base. Forewing blackish brown, mottled with some rusty brown, particularly in proximal half; supplementary black spots in fold and in cell obscure; costal and tornal spot small, cream, separated. Hindwing light grey.

Variation. No variation observed except for size, which differs considerably in two reared specimens from Italy and Greece.

**Male genitalia** (Figs 6–7, 10–11). Uncus subovate; tegumen stout; transtilla membranous; valva moderately short and slender, digitate, apex rounded; sacculus short, with angular ventral and weakly convex dorsal margin, apically pointed; posterior margin of vinculum with deep medial emargination and slight medial incision, two pairs of short processes developed; saccus long, comparatively broad at base, distal part gradually tapered; phallus long and slender, weakly curved, with some minute cornuti apically.

**Female genitalia** (Figs 14–15). Segment VIII without processes, subgenital plate sub-triangular, with numerous narrow folds, separated from sclerotized lateral plates by membranous zone; apophysis anterior about length of segment VIII; antrum short, about one quarter length of apophysis anterior, nearly cup-shaped; posterior part of ductus bursae with pair of short sclerites, extending to middle of apophysis anterior, and with two tiny sclerites anteriorly; signum with crescent-shaped base, long and slender, strongly bent hook.

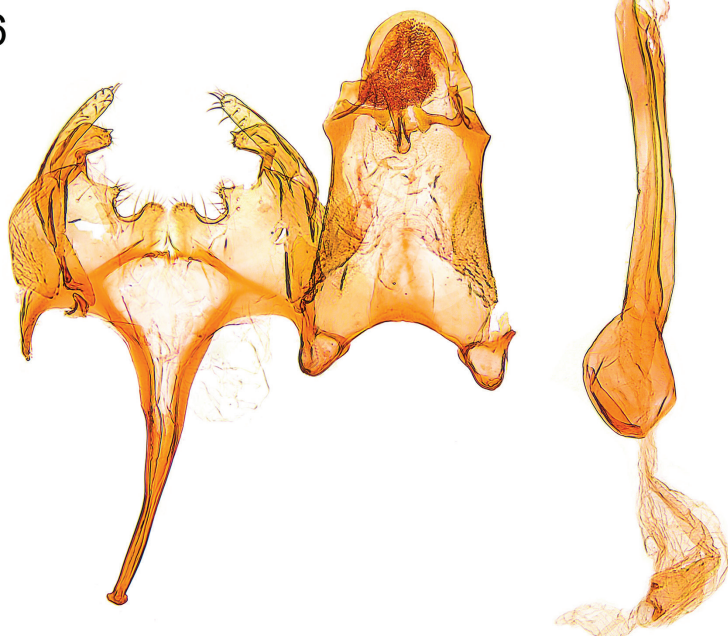
**Molecular data.** The intraspecific divergence of the barcode region is low with mean intraspecific divergence of 0.21% and maximum intraspecific divergence of 0.31% ( $n=3$ ). The distance to the nearest neighbour *C. mucronatella* is 5.41%, the divergence to the morphologically closest *C. amaurella* is 6.82%.

**Etymology.** The name “*crypticum*” refers to the cryptic morphology of the species and is derived from the latinized adjective *crypticus*.

**Distribution.** The species is known from widely separated localities in northern Italy, Switzerland and Greece, indicating a more widespread distribution in Sub-Mediterranean and Mediterranean Europe. However, the host-plants are much more widespread, ranging to northern Europe in the north and to Central Asia in the east. No sympatric occurrence with *C. amaurella* is reported though the two taxa can occur close to one another in the Alps.

**Bionomics.** The larva has been found in early spring, feeding in the stem of *Silene otites* (L.) Wibel (Caryophyllaceae) (Burmann 1990) and *Silene nutans* L. (Huemer 1989) but detailed descriptions of feeding habits and larval morphology are missing. The adult occurs from early July (reared material dates from mid-June to mid-July) to September and it is attracted to light. *C. crypticum* prefers xerophilous steppes

6



7



**Figures 6–7.** Male genitalia. **6** *Caryocolum crypticum* sp. n., paratype, Italy, slide GU 86/041 P.Huemer  
**7** *C. crypticum* sp. n., paratype, Italy, slide GEL 1215 P.Huemer.

and rocky habitats with sparse vegetation. Vertical distribution: from about 500 to 1300 m, restricted to mountainous areas.

**Remarks.** Huemer (1988) already examined females reared from *Silene otites* in Switzerland by Whitebread but in the absence of males considered them as deviating *C. amaurella*.

The majority of collected material belongs to females whereas *C. amaurella* is mainly known from the male sex. This may indicate differences in attraction to artificial lights or a female-biased sex ratio in *C. crypticum*.

### ***Caryocolum amaurella* (Hering, 1924)**

[http://species-id.net/wiki/Caryocolum\\_amaurella](http://species-id.net/wiki/Caryocolum_amaurella)

Figs 4–5, 8–9, 12–13, 16–17

*Lita amaurella* Hering 1924: 82, Figs 11–12.

*Lita viscariæ* Schütze 1926: 171.

**Material examined.** Lectotype ♂ [with nine labels]: ‘Fennia Ab Bromarf’ ‘R. F:tius’ ‘21.7.21’ [piece of celluloid where genitalia was mounted] ‘Type ♂’ [red] ‘Lita amaurella m. det. Mart. Hering ♂’ ‘Mus. Zool. H:fors spec. typ. No 7016 Lita amaurella Hering’ ‘Lita amaurella m. ♂ Sch.-Armatur Bromarf 21.7.21 Fabritius’ ‘LECTOTYPE O. Karsholt design.’

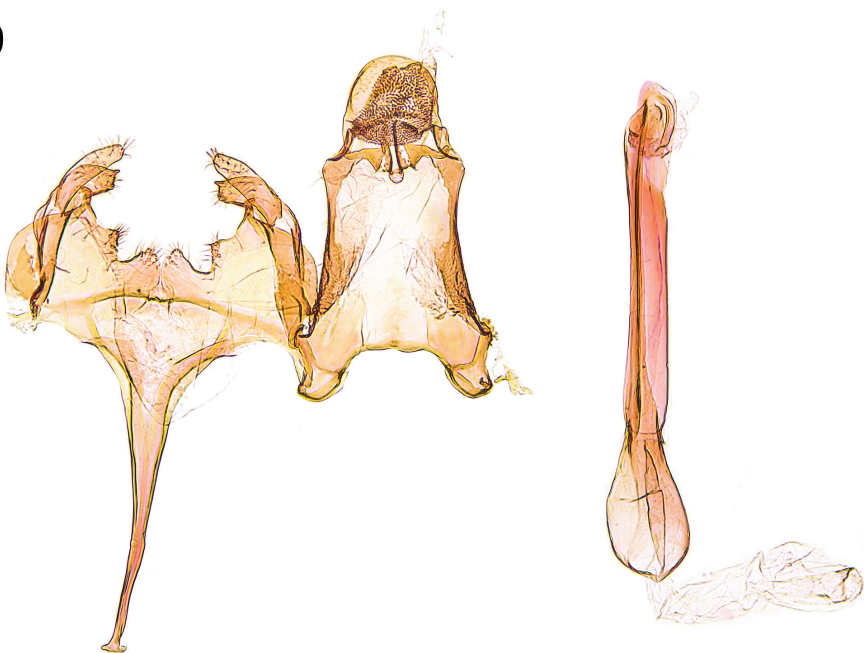
**Finland:** 1 ♂, Ab, Naantali, 25.8.1965, leg. Karvonen, slide Karsholt 2719; 2 ♂, N, Ekenäs, 17.–20.7.1980, leg. Fibiger; 1 ♂, N, Helsinki, 25.7.1982, leg. Schnack; 1 ♂, N, Borgå lk., Tirmo, 19–20.7.1980, leg. Fibiger; 5 ♂, same data, but 1.–2.8.1982, leg. Schnack; 2 ♂, U, Sluntle, 18.–31.7.1982, leg. Karsholt (all ZMUC); 6 ♂, 5 ♀, U, Porvoo, 6698:3426 Ånäs, e.l. 2012 (*Lychnis viscaria*), leg. Hirvonen (ZMUO); 4 ♂, V, Dragsfjärd, 664:3249, 2008, leg. Mutanen & Välimäki (ZMUO); 1 ♂, U, Hanko, 6642:3289, 2007, leg. Mutanen & Välimäki (ZMUO). **Sweden:** 2 ♂, Sk, Maglehen, 10.7.1965, leg. Svensson (TLMF, ZMUC); 1 ♂, Sm, Högsby, 13.7.1968, leg. Johansson; 1 ♂, Öl, Ödeshög, 17.7.1972, leg. Karsholt, slide Karsholt 1806; 1 ♂, St. Alvar, Tornrör, 25.7.1997, leg. Hendriksen, slide Hendriksen 1953; 2 ♂, Öl, Gårdby, 2.8.1999, leg. Hendriksen, slide Hendriksen 2411, 2415; 1 ♂, same data, but 22.7.2000; 1 ♂, Gtl., Hejnum Häller, 30.7.1977, leg. Hendriksen, slide Hendriksen 1944; 1 ♂, Ög, Ödeshög, 17.7.1972, leg. Karsholt; Upl., Film, 12.7.1995, leg. Hendriksen (all ZMUC). **Norway:** 2 ♂, On, Vinstra, 19.–29.7.1983, leg. Karsholt & Michelsen, slide Karsholt 4294, 4295; 2 ♀, same data, but 4.–5.7.1987, leg. Karsholt, slide Hendriksen 2099; 2 ♂, same data, but 9.8.1996, leg. Hendriksen (all ZMUC). **Denmark:** 1 ♂, Bornholm, Rø, 7.1892, leg. Gudmann, slide Wolff 2593; 5 ♂, 1 ♀, same data, but 28.7.1978, leg. Schnack, slide Schnack 1118; 1 ♂, Bornholm, Gudhjem, 1 ♂, 2 ♀, 29.6–3.7.1920, leg. Gudmann, slide Wolff 2625, 3682; 1 ♂, 1 ♀, same data, but e.l. 5.1921 (*Lychnis viscaria*), bred 21. & 28.6.1921, leg. Gudmann, slide Wolff 3681 (all ZMUC); 6 ♂, 8 ♀, Bornholm, Hammeren, 18.7.1977,



8



9



**Figures 8–9.** Male genitalia. **8** *Caryocolum amaurella* (Hering), Finland, slide GU 14/1373 P.Huemer; **9** *C. amaurella*, Finland, slide GU 14/1374 P.Huemer.

leg. Karsholt & Schnack, slide Hendriksen 1767, Karsholt 2948 TLMF, ZMUC); 2 ♂, same data, but 25.7.1977, leg. Schnack; 4 ♂, same data, but 16. –25.7.1978, leg. Schnack; 4 ♂, same data, but 19.–22.7.1979, leg. Hendriksen; 6 ♂, same data, but 29 –30.7.1981, leg. Hendriksen, slide Hendriksen 385, 561, 722; Bornholm, Randkløve, 1 ♂, 22.7.1977, leg. Schnack; Bornholm, 1 ♀, Ringe Bakker, 16.7.1978, leg. Schnack (all ZMUC). **Germany:** 1 ♂, 1 ♀, Lausitz, Umg. Bautzen, e.l. 1935 (*Lychnis viscaria*), leg. Starke (BMNH); 2 ♂, Thüringen, Bad Blankenburg, 14.7.1964, leg. Steuer (TLMF); 1 ♂, Thüringen, Bad Blankenburg, 8.7.1972, leg. Steuer (TLMF). **France:** 2 ♂, Alpes Maritimes, Col de la Cayolle, 2200–2300 m, 29.–30.7.2005, leg. Skou, slide Hendriksen 5364 (ZMUC). **Austria:** 1 ♂, Niederösterreich, Jauerling, 24.7.1935 (TLMF); 2 ♂, Oberösterreich, Windischgarsten, Veichtal, 23.7.1976, leg. Wimmer (TLMF); 1 ♂, Oberösterreich, Waldhausen, Schwarzenberg, 6.8.1997, leg. Wimmer (TLMF); 9 ♂, Kärnten, St. Jakob im Lesachtal, Mussen E, 1680–1800 m, 4.8.1999, leg. Huemer & Erlebach (TLMF). **Slovakia:** 1 ♀, Pol'ana, 28.7.1989, leg. Patocka (ZMUC). **Macedonia:** 4 ♂, NP Mavrovo, Korab, Korabska jezero, Kobilino pole, 2080–2180 m, 28.7.–1.8.2011, leg. Huemer & Tarmann (TLMF). **Turkey:** 2 ♂, 1 ♀, prov. Sivas, 10 km W Görün, 1650 m, 27.7.1989, leg. Esser & Fibiger, slide Huemer GU 90/130, GU 91/215; 4 ♂, prov. Erzerum, Kop Pass, 1750 m, 15.–16.9.1993, leg. Fibiger, slide Hendriksen 2889, 2894; 1 ♂, prov. Erzincan, Kizildağ, Geçidi, 2100 m, 19.8.1993, leg. Schepler, slide Hendriksen 2384 (all ZMUC).

**Diagnosis.** See above.

**Description.** Adult (Figs 4–5). Wingspan 10–14 mm. Segment 2 of labial palpus bone-white on inner and upper surface, blackish grey on outer and lower surface; segment 3 almost black with light tip. Antenna black, indistinctly lighter ringed. Head with light yellow frons and black neck; thorax and tegula black mottled with brown. Forewing blackish grey mottled with some light brown; base black; two indistinct black spots in fold; one oblique spot above it and one in cell; some white scales before and after these spots; costal and tornal spot small, white, rarely fused. Hindwing light grey.

Variation. The colour of the forewings varies from greyish to blackish. Worn specimens look lighter than fresh ones. Sometimes there are no white scales in the middle of the wing.

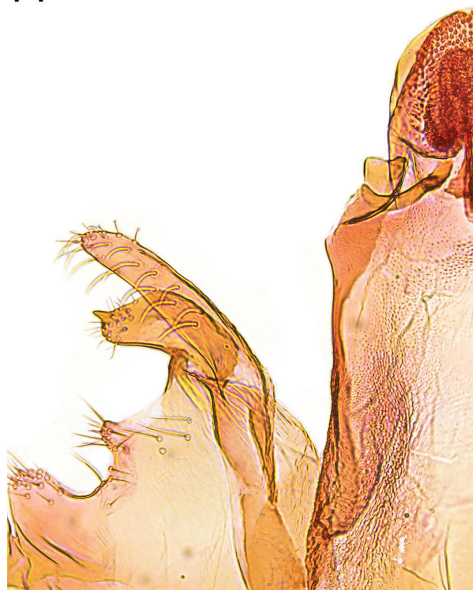
**Male genitalia** (Figs 8–9, 12–13). Uncus subovate; tegumen stout; transtilla membranous; valva short, moderately stout, apex rounded; sacculus short, with angular ventral and convex dorsal margin, apically pointed; posterior margin of vinculum with deep medial emargination and slight medial incision, two pairs of short processes developed; saccus long, comparatively broad at base, distal part gradually tapered; phallus long and slender, weakly curved, with some minute cornuti apically.

**Female genitalia** (Figs 16–17). Segment VIII without processes, subgenital plate sub-triangular, with numerous narrow folds, separated from sclerotized lateral plates by membranous zone; apophysis anterior slightly longer than segment VIII; antrum moderately short, about one-third to one-quarter length of apophysis anterior, broadly funnel-shaped; posterior part of ductus bursae with pair of lateral sclerites, extending to anterior third of apophysis anterior, and with two tiny sclerites anteriorly; signum with crescent-shaped base, short and stout, strongly bent hook.

10



11



12



13



**Figures 10–13.** Details of male genitalia (vinculum-valva-complex). **10** *Caryocolum crypticum* sp. n., paratype, Italy, slide GU 86/041 P.Huemer **11** *C. crypticum* sp. n., paratype, Italy, slide GEL 1215 P.Huemer **12** *Caryocolum amaurella*, Finland, slide GU 14/1373 P.Huemer **13** *C. amaurella*, Finland, slide GU 14/1374 P.Huemer.



**Molecular data.** The intraspecific divergence of the barcode region is high with mean intraspecific divergence of 3.01% and maximum intraspecific divergence of 4.62% (n=9). The distance to the nearest neighbour *C. mucronatella* is 5.21%, the divergence to the morphologically closest *C. crypticum* is 6.82%. The extraordinary high intraspecific divergence with 4 haplotypes is partially related to geographical pattern. However, we also found two haplotypes within one population in Finland and morphology does not support cryptic diversity.

**Distribution.** With certainty known from scattered records from northern and Central Europe and Turkey. All the specimens from north of the Alps that we have been able to cross-check are correctly attributed to *C. amaurella*. However, recent records from Ukraine (Bidzilya and Budashkin 2009) and Russia (southern Ural Mountains) (Junnilainen et al. 2010) have to be re-examined due to a possible mix-up with *C. crypticum*. Records from Switzerland are dubious, and at least in one instance refer to the new species, whereas those from France (Nel 2003) are confirmed (see Huemer and Karsholt 2010, Fig. 154c).

**Bionomics.** The larva has been recorded feeding on *Silene viscaria* (L.) Jess (= *Lychnis viscaria* L. (Caryophyllaceae) (Huemer and Karsholt 2010), while the other stated host-plants, namely *Silene otites* (L.) Wibel (Burmman 1990) and *S. nutans* L. (Huemer 1989a), refer to *C. crypticum*. Schütze (1926, 1931) gives a detailed account of the life-history. The larva feeds in April and May in the young terminal leaves which are – without spinning – attached to a tube where the larva is hidden. Dark frass is frequently extruded at the tip of the larval dwelling. Later it bores into the stem and the shoots often become swollen and stunted. Pupation takes place on the ground in a cocoon among debris. The adult occurs from late June to early September and it is attracted to light. *C. amaurella* is restricted to warm and sunny habitats such as dry meadows and pastures. Vertical distribution: from lowland localities to about 2200 m in the Alps.

**Remarks.** *Lita amaurella* was described from an unspecified number of specimens of both sexes (♂, ♀) from Finland (Bromarf) (Hering 1924). In order to stabilize nomenclature, a male, labelled as type, in ZMUH is here designated as lectotype (see data above). *Lita viscariae* was described from 67 specimens reared from *Silene viscaria* from Eastern Germany (near Rachlau) (Schütze 1926). No type material was traced during this and earlier studies (Huemer 1988), but the original descriptions and topotypical material leave no doubt about the identity.

Turkish specimens of *C. amaurella* examined by us differ from European specimens of this species by the thorax with rusty brown posterior part and the rusty brown tegulae with blackish brown base, similar to *C. crypticum*, and they are thus hardly separable from the latter on external characters. The genitalia of both sexes of *C. amaurella* from Turkey agree in all details with those of European *C. amaurella* and, because no contradicting genetic data is currently available, we consider them as belonging to that species.

One of the examined specimens of *C. amaurella* from Turkey was collected in the same locality (Kızıldağ Geçidi, prov. Erzincan) as a specimen *C. iranicum* in ZMUC. The latter species, which is only known from a few specimens, differs, as stated above, in characters of the male genitalia.

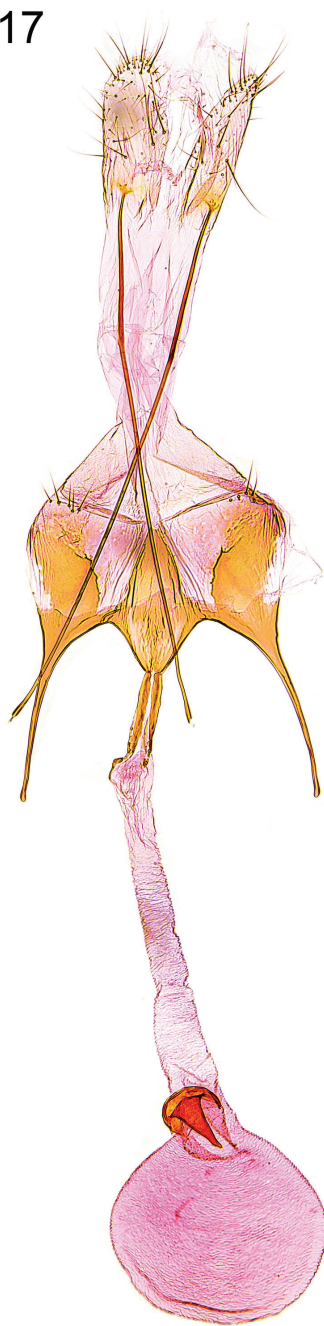


**Figures 14–15.** Female genitalia. **14** *Caryocolum crypticum* sp. n., holotype, slide GEL 1234 P.Huemer  
**15** *C. crypticum* sp. n., paratype, Italy, slide GEL 1232 P.Huemer.

16



17



**Figures 16–17.** Female genitalia. **16** *Caryocolum amaurella*, Finland, slide GU 14/1372 P.Huemer **17** *C. amaurella*, Finland, slide GU 14/1371 P.Huemer.



## Discussion

The genus *Caryocolum* is a rare example of European Microlepidoptera which has gained significant attention from specialists during the last decades. Several monographic papers, from Klimesch (1953–54) to Huemer and Karsholt (2010), are a sound base for a stable taxonomy and a pre-requisite to test congruence of classical morphologically-driven species delineation with that of molecular data. DNA barcoding has evolved as a widely accepted method for preliminary species delimitation (Monaghan et al. 2009, Hendrich et al. 2010, Kekkonen and Hebert 2014) and therefore the animal DNA barcode region seemed an appropriate genetic marker to be used for this purpose. Indeed, barcoding resulted in an excellent support for all of the 44 studied species with a distinct barcode gap to the nearest neighbour ranging from about 3% to nearly 7% interspecific divergence.

Intraspecific variation shows a different pattern. The majority of species has a low (<2%) maximum intraspecific divergence and thus seems taxonomically well defined. However, a remarkable number of species (8 species, nearly one quarter of all, 9 species with only one sample not considered) is characterized by maximum divergence exceeding 3% (Fig. 1). Such deep intraspecific splits often suggest the possibility of cryptic diversity (for examples in Lepidoptera, see Dinca et al. 2011, Hausmann et al. 2009, Huemer and Hebert 2011, Huemer et al. 2012, Huemer et al. 2013, Kaila and Mutanen 2012, Landry and Hebert 2013, Mutanen et al. 2012a, b, 2013, Segerer et al. 2011, Wilson et al. 2010). A morphological cross-check in one of these taxa, *Caryocolum amaurella*, proved the existence of a hitherto overlooked species with validity independently supported by morphology, biological data, and the DNA barcode. The potential of DNA barcoding for screening of cryptic diversity is obvious in this case, where morphological characters, particularly the normally well-separated male genitalia, are weak and thus have been neglected so far. Although deep intraspecific splits may alternatively refer to mitochondrial introgression, historical polymorphism or *Wolbachia* infection (Hurst and Jiggins 2005, Funk and Omland 2003), there is a considerable possibility of further cryptic diversity in the genus. In *C. schleichi* it seems most appropriate that the three sequenced subspecies should be considered as different species since host-plants and genitalia morphology differ as well (see i.e. Huemer and Karsholt 2010). The subspecies of *C. schleichi* are geographically isolated making their delimitation both rather artificial and very sensitive to the species concept applied (Mutanen et al. 2012c). An integrative revision of this group is in preparation by the authors. In contrast, the expected low divergence in subspecies is reflected by a very low divergence in *C. marmorea* and its subspecies *C. marmorea mediocorsa*. Diagnostic morphological characters seem present in further taxa from first examined samples, namely *C. fibigerium* and *C. peregrinella* with a maximum intraspecific divergence of 6.27% and 5.69% related to three deep phylogeographic splits in both species. Similar deep splits are observed in *C. alsinella* and in *C. cauligenella*. For all these taxa with subtle character differences a careful re-examination of morphology has to be undertaken in the future.

## Acknowledgments

We are particularly grateful to Paul Hebert and his team at the Canadian Centre for DNA Barcoding (Guelph, Canada), whose sequencing work was enabled by funding from the Government of Canada to Genome Canada through the Ontario Genomics Institute. We are also grateful to the Ontario Ministry of Research and Innovation and to NSERC for their support of the BOLD informatics platform.

Stefan Heim (TLMF) is acknowledged for his kind assistance with photographic work. We thank Robert J. Heckford (Plymouth, GB), Petri Hirvonen (Porvoo, Finland), Thierry Varenne (Nice, France), Christian Wieser (Klagenfurt, Austria) and Josef Wimmer (Steyr, Austria) for providing material for our examination, and Lauri Kaila (ZMUH) for access to the type of *Lita amaurella*.

We are particularly indebted to the Promotion of Educational Policies, University and Research Department of the Autonomous Province of Bolzano - South Tyrol for helping to fund the project “Genetic biodiversity archive - DNA barcoding of Lepidoptera of the central Alpine region (South, East and North Tyrol)”. Furthermore fundings from inatura Erlebnis Naturschau (Dornbirn, Austria) are acknowledged.

Last, but not least, we thank Martin Corley (Faringdon, UK) for linguistic improvement of the manuscript.

## References

- Bella S (2008) *Caryocolum siculum* sp. n. (Gelechiidae), feeding on *Gypsophila* (Caryophyllaceae) in Sicily. *Nota lepidopterologica* 31: 69–75.
- Bidzilya AV, Budashkin YI (2009) New records of Microlepidoptera from Ukraine. *Proceedings Zoological Museum Kiev Taras Shevchenko National University* 5: 14–28. [In Russian]
- Burmann K (1990) Beiträge zur Microlepidopteren-Fauna Tirols. XIV. *Caryocolum* Gregor & Povolný, 1954 (Insecta: Lepidoptera, Gelechiidae). *Berichte des naturwissenschaftlich-medizinischen Vereins Innsbruck* 77: 171–184.
- deWaard JR, Ivanova NV, Hajibabaei M, Hebert PDN (2008) Assembling DNA Barcodes: Analytical Protocols. In: Cristofre M (Ed) *Methods in Molecular Biology: Environmental Genetics*. Humana Press Inc., Totowa, USA, 275–293.
- Dinca V, Lukhtanov VA, Talavera G, Vila R (2011) Unexpected layers of cryptic diversity in wood white *Leptidea* butterflies. *Nature Communications* 2: 324. doi: 10.1038/ncomms1329
- Funk DJ, Omland KE (2003) Species-level parphyly and polyphyly: Frequency, causes and consequences, with insights from animal mitochondrial DNA. *Annual Review of Ecology, Evolution, and Systematics* 34: 397–423. doi: 10.1146/annurev.ecolsys.34.011802.132421
- Grange JC, Nel J (2012) *Caryocolum dauphini* n. sp., un endémique du Sud-Ouest alpin découvert dans le Parc national du Mercantour (Gelechiidae, Gnorimoschemini). *Oreina* 17: 22–23.
- Gregor F, Povolný D (1954) Systematische und zoogeographische Studie über die Gruppe der Arten *Gnorimoschema* Busck mit Rücksicht auf die richtige Diagnostik des Schädling *Gnorimoschema ocellatellum* Boyd. *Zoologické a Entomologické Listy* 3: 83–97, pl. 7, map.

- Hausmann A, Hebert PDN, Mitchell A, Rougerie R, Sommerer M, Edwards T, Young CJ (2009) Revision of the Australian *Oenochroma vinaria* Guenée, 1858 species-complex (Lepidoptera: Geometridae, Oenochrominae): DNA barcoding reveals cryptic diversity and assesses status of type specimen without dissection. *Zootaxa* 2239: 1–21.
- Hendrich L, Pons J, Ribera I, Balke M (2010) Mitochondrial Cox1 sequence data reliably uncover patterns of insect diversity but suffer from high lineage-idiosyncratic error rates. *PLoS ONE* 5: e14448. doi: 10.1371/journal.pone.0014448
- Hering EM (1924) Beitrag zur Kenntnis der Microlepidopteren-Fauna Finlands. *Notulae Entomologicae* 4: 75–84.
- Huemer P (1987) Eine modifizierte Genitalpräparationstechnik für die Gattung *Caryocolum*. *Mitteilungen der Schweizerischen Entomologischen Gesellschaft* 60: 207–211.
- Huemer P (1988) A taxonomic revision of *Caryocolum* (Lepidoptera: Gelechiidae). *Bulletin of the British Museum of Natural History (Entomology)* 57: 439–571.
- Huemer P (1989a) Bemerkenswerte Funde von *Caryocolum*-Arten aus den Südalpen und dem Mediterraneum (Lepidoptera, Gelechiidae). *Nachrichtenblatt der Bayerischen Entomologen* 38: 37–40.
- Huemer P (1989b) Neue und wenig bekannte Arten der Gattung *Caryocolum* Gregor & Povolný, 1954, aus Südwestasien. *Mitteilungen der Münchner Entomologischen Gesellschaft* 79: 127–142.
- Huemer P (2013) Die Schmetterlinge Österreichs (Lepidoptera). Systematische und faunistische Checkliste. *Studiohefte* 12, 304 pp.
- Huemer P, Karsholt O (2010) Gelechiidae II (Gelechiinae: Gnorimoschemini. In: Huemer P, Karsholt O, Nuss M. *Microlepidoptera of Europe*. Vol. 6. Apollo Books, Stenstrup, 586 pp.
- Huemer P, Nel J (2005) *Caryocolum mazeli* sp. n., a new species from southern France (Lepidoptera, Gelechiidae). *Bulletin de la Société Entomologique de France* 110: 125–127.
- Huemer P, Elsner G, Karsholt O (2013) Review of the *Eulamprotes wilkella* species-group based on morphology and DNA barcodes, with descriptions of new taxa (Lepidoptera, Gelechiidae). *Zootaxa* 3746: 069–100.
- Huemer P, Hebert PDN (2011) Cryptic diversity and phylogeography of high alpine *Sattleria* — a case study combining DNA barcodes and morphology (Lepidoptera: Gelechiidae). *Zootaxa* 2981: 1–22.
- Huemer P, Zlatkov B, Baixeras J (2012) *Dichrorampha dinarica*, new species, a century of confusion in European lepidopterology (Lepidoptera: Tortricidae) resolved by combining morphology and DNA barcoding. *Zootaxa* 3389: 41–50.
- Hurst GDD, Jiggins FM (2005) Problems with mitochondrial DNA as a marker in population, phylogeographic and phylogenetic studies: the effects of inherited symbionts. *Proceedings of the Royal Society Biological Sciences Series B* 272: 1525–1534. doi: 10.1098/rspb.2005.3056
- Junnilainen J, Karsholt O, Nupponen K, Kaitila JP, Nupponen T, Olschwang V (2010) The gelechiid fauna of the Southern Ural Mountains, part II: list of recorded species with taxonomical notes (Lepidoptera: Gelechiidae). *Zootaxa* 2367: 1–68.
- Kaila L, Mutanen M (2012) DNA barcoding and morphology support the division of *Elachista nuraghella* sensu auct. (Lepidoptera: Elachistidae: Elachistinae) into two vicariant species. *Zootaxa* 3343: 57–68.

- Kekkonen M, Hebert PDN (2014) DNA barcode-based delineation of putative species: efficient start for taxonomic workflows. *Molecular Ecology Resources*. doi: 10.1111/1755-0998.12233
- Klimesch J (1953–1954) Die an Caryophyllaceen lebenden europäischen *Gnorimoschema* Busck (= *Phthorimaea* Meyr.)-Arten. *Zeitschrift der Wiener Entomologischen Gesellschaft* 38 (1953): 225–239, 272–282, 311–319; 39 (1954): 273–288, 335–341, 357–362.
- Landry JF, Hebert PDN (2013) *Plutella australiana* (Lepidoptera: Plutellidae), an overlooked diamondback moth revealed by DNA barcodes. *Zookeys* 327: 43–63. doi: 10.3897/zookeys.327.5831
- Monaghan MT, Wild R, Elliot M, Fujisawa T, Balke M, Inward DJG, Lees DC, Ranaivosolo R, Eggleton P, Barraclough TG, Vogler AP (2009) Accelerated species inventory on Madagascar using coalescent-based models of species delineation. *Systematic Biology* 58: 298–311. doi: 10.1093/sysbio/syp027
- Mutanen M, Aarvik L, Landry J-F, Segerer A, Karsholt O (2012a) *Epinotia cinereana* (Haworth, 1811) bona sp., a Holarctic tortricid distinct from *E. nisella* (Clerck, 1759) (Lepidoptera: Tortricidae: Eucosmini) as evidenced by DNA barcodes, morphology and life history. *Zootaxa* 3318: 1–25.
- Mutanen M, Aarvik L, Huemer P, Kaila L, Karsholt O, Tuck K (2012b) DNA barcodes reveal that the widespread European tortricid moth *Phalonidia manniana* (Lepidoptera: Tortricidae) is a mixture of two species. *Zootaxa* 3262: 1–21.
- Mutanen M, Hausmann A, Hebert PDN, Landry J-F, deWaard J, Huemer P (2012c) Allopatry as a Gordian knot for taxonomists: patterns of barcode divergences in arctic-alpine Lepidoptera. *PLoS ONE* 7: e47214. doi: 10.1371/journal.pone.0047214
- Mutanen M, Kaila L, Tabell J (2013) Wide-ranging barcoding aids discovery of one-third increase of species richness in presumably well-investigated moths. *Scientific Reports* 3: 2901. doi: 10.1038/srep02901
- Nel J (2003) Microlépidoptères nouveaux ou rarement signalés de la faune de France (Lepidoptera). *Bulletin de la Société Entomologique de France* 108: 81–86.
- Ratnasingham S, Hebert PDN (2007) BOLD: The Barcode of Life Data System (<http://www.barcodinglife.org>). *Molecular Ecology Notes* 7: 355–364. doi: 10.1111/j.1471-8286.2007.01678.x
- Schütze KT (1926) *Lita viscaria* n. sp. *Deutsche entomologische Zeitschrift Iris* 40: 171–175.
- Schütze KT (1931) Die Biologie der Kleinschmetterlinge unter besonderer Berücksichtigung ihrer Nährpflanzen und Erscheinungszeiten. Frankfurt/Main, 235 pp.
- Segerer AH, Haslberger A, Grünwald T (2011 [“2010”]) *Olethreutes subtilana* (Falkovich, 1959): Unexpected occurrence of an ‘eastern’ leaf roller in Central Europe, uncovered by DNA barcoding (Tortricidae: Olethreutinae). *Nota lepidopterologica* 33: 197–206.
- Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S (2011) MEGA5: Molecular Evolutionary Genetics Analysis using Maximum Likelihood, Evolutionary Distance, and Maximum Parsimony Methods. *Molecular Biology and Evolution* 28: 2731–2739. doi: 10.1093/molbev/msr121
- Wilson JJ, Landry J-F, Janzen DH, Hallwachs W, Nazari V, Hajibabaei M, Hebert PDN (2010) Identity of the ailanthus webworm moth (Lepidoptera, Yponomeutidae), a complex of two



species: evidence from DNA barcoding, morphology and ecology. *ZooKeys* 46: 41–60. doi: 10.3897/zookeys.46.406

Zeller PC (1839) Versuch einer naturgemässen Eintheilung der Schaben. *Isis*, Leipzig 1839: 167–220.

## **Supplementary material I**

### **Sample information for specimens included in this study.**

Authors: Peter Huemer, Ole Karsholt, Marko Mutanen

Data type: species data

Explanation note: Process IDs are sequence identifiers in BOLD; Sample IDs are specimen identifiers; BINs are Barcode Identification Numbers in BOLD. Details of collecting data, images, sequences, and trace files for the barcoded specimens are available in the public BOLD dataset “DS-LECARY”, accessed at [dx.doi.org/10.5883/DS-LECARY](https://dx.doi.org/10.5883/DS-LECARY)

Copyright notice: This dataset is made available under the Open Database License (<http://opendatacommons.org/licenses/odbl/1.0/>). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: doi: 10.3897/zookeys.404.7234.app1



# A new species of genus *Tetrasticta* Kraatz (Coleoptera, Staphylinidae, Aleocharinae) from Xishuangbanna, Southwest China

Dan-Lin Zheng<sup>1,†</sup>, Mei-Jun Zhao<sup>1,‡</sup>

<sup>1</sup> Department of Biology, College of Life and Environmental Sciences, Shanghai Normal University, 100 Guilin Road, Xuhui District, Shanghai 200234, P. R. China

<sup>†</sup> <http://zoobank.org/664F20E5-091E-4673-81F1-02F596EF5A9D>

<sup>‡</sup> <http://zoobank.org/FBE4FA1F-5BCF-405E-98C1-4F86894643EA>

Corresponding author: Mei-Jun Zhao ([mjzhao@shnu.edu.cn](mailto:mjzhao@shnu.edu.cn))

---

Academic editor: J. Klimaszewski | Received 17 February 2013 | Accepted 21 March 2014 | Published 24 April 2014

<http://zoobank.org/C929E454-1B8D-492E-BFE7-25E67E8EA109>

---

**Citation:** Zheng D-L, Zhao M-J (2014) A new species of genus *Tetrasticta* Kraatz (Coleoptera, Staphylinidae, Aleocharinae) from Xishuangbanna, Southwest China. ZooKeys 404: 113–116. doi: 10.3897/zookeys.404.7276

---

## Abstract

*Tetrasticta bobbii* Zheng & Zhao, **sp. n.**, collected in Nangongshan, Xishuangbanna, Yunnan, is described and illustrated.

## Keywords

Aleocharini, first record, *Tetrasticta*, Yunnan, Xishuangbanna

## Introduction

The aleocharine genus *Tetrasticta* Kraatz, 1857 (Aleocharini) contains 13 species worldwide (Maruyama and Sugaya 2002; Maruyama 2004; Pace 2000, 2008, 2013b; Yamamoto and Maruyama 2013). Pace (2010) synonymized *Creochara* Cameron, 1931 with *Tetrasticta* and repeated this arrangement in his recent paper (Pace 2013a). According to Yamamoto and Maruyama (2013), the synonymization of *Tetrasticta* by Pace (2010) does not provide sufficient evidence and should not be consider as valid. Currently, no species of this genus has been reported from

Mainland China. In 2003, our colleagues Jia-Yao Hu and Liang Tang surveyed the staphylinid fauna of Nangongshan (Yunnan, Southwest China), and collected a small series of *Tetrasticta* specimens. A closer examination of this material revealed that the species was undescribed.

## Material and methods

All the types are deposited in the Insect Collection of Shanghai Normal University, Shanghai, China (SNUC).

Specimens were killed with ethyl acetate and preserved in 75% ethanol before dissection; photos of habitus were taken with a Canon EOS 7D with an MP-E 65mm macro photo lens.

The following abbreviations are applied in the text: **BL** – body length, from the anterior margin of the head to the posterior margin of the abdominal tergite VIII; **FBL** – forebody length, from the clypeal anterior margin to the posterior margin of elytra; **HD** – head length, from the clypeal anterior margin to the occipital constriction; **PL** – length of the pronotum along the midline; **HW** – width of the head across the eyes; **PW** – maximum width of the pronotum.

## Taxonomy

### *Tetrasticta bobbii* sp. n.

<http://zoobank.org/04ED9579-704E-4F46-9769-BD73955B5A4B>

[http://species-id.net/wiki/Tetrasticta\\_bobbii](http://species-id.net/wiki/Tetrasticta_bobbii)

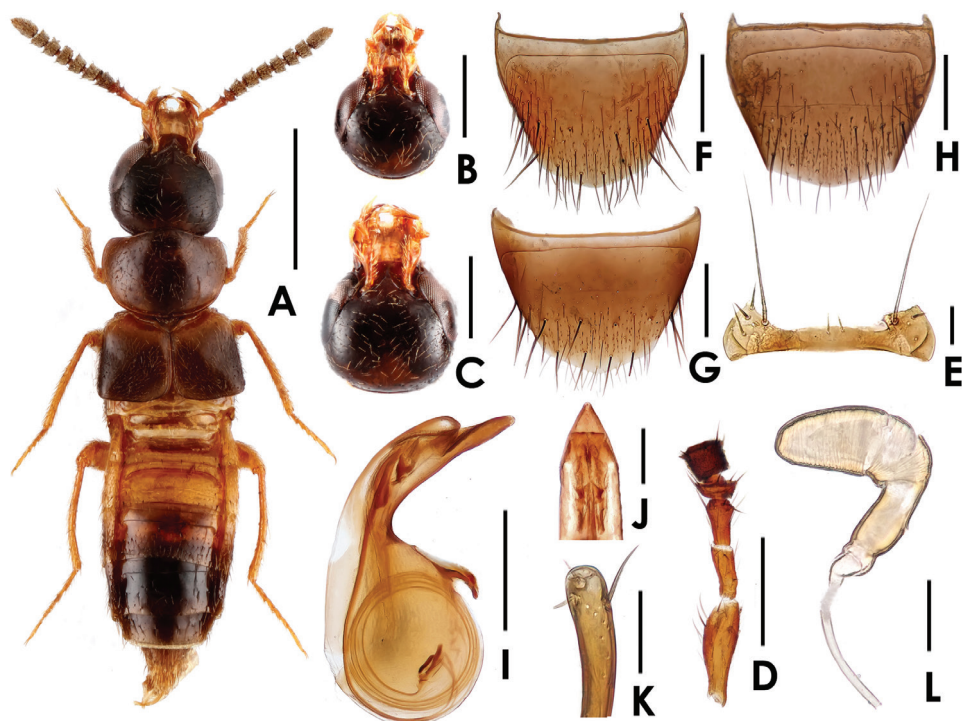
Fig. 1

**Type material. Holotype: China:** ♂, labelled ‘CHINA: Yunnan Prov., Xishuangbanna, Mengla County (勐腊县), Nangongshan (南贡山), alt. 800–1000 m, 7.VII.2003, Hu & Tang leg. / HOLOTYPE [red], *Tetrasticta bobbii* sp. n., Zheng & Zhao det. 2014, SNUC’. **Paratypes: China:** 1 ♂, 1 ♀ (preserved in a small tube filled with 75% ethanol), same data as holotype, both bearing the following label: ‘PARATYPE [yellow], *Tetrasticta bobbii* sp. n., Zheng & Zhao det. 2014, SNUC’.

**Description.** Body (Fig. 1A) shining. Coloration: head black; antennae and pronotum reddish brown; elytra reddish brown with anterior margin reddish yellow; legs reddish yellow; abdomen with tergites II–IV reddish yellow, tergites VI–VII black.

Head (Figs 1B, C) almost 1.05 times as wide as long; slightly narrower than pronotum; surface sparsely covered with yellow setae; eyes large. Antennae (Figs 1A, D) with segment I long, as long as combined length of segments II–III; segments II and III about one-half of segment I; segment IV extremely short, much wider than long; segments IV–V almost as wide as long; segments VI–X wider than long. Mandibles long, slender. Mentum distinctly transverse, about 3.62 times as wide as long; shaped





**Figure 1.** *Tetrasticta bobbii* sp. n. **A** male habitus, in dorsal view **B** female head, in dorsal view **C** male head, in dorsal view **D** antennomere I–V **E** mentum **F** male tergite VIII **G** male sternite VIII **H** female tergite VIII **I** median lobe of aedeagus, in lateral view **J** ditto, apical part, in ventral view **K** apical lobe of paramerite, in lateral view **L** spermatheca. Scales (mm): **A** = 1; **B**, **C** = 0.5; **D** = 0.3; **E** = 0.05; **F**, **G**, **H**, **I** = 0.2; **J**, **K**, **L** = 0.05.

as in Fig. 1E. Pronotum wider than long, about 1.38 times as wide as long; surface moderately covered with yellow setae; disc with three shallow depressions; shaped as in Fig. 1A. Elytra wider than long; surface moderately covered with yellow setae. Abdomen flattened, with subparallel lateral margins, widest at segments IV–V; tergite VIII with six pairs of macrosetae; sternite VIII (Fig. 1G) generalized in shape, posterior margin convex in the middle, with eight pairs of macrosetae.

Male: postocular margins straight for a short distance and then narrowed posteriorly (Fig. 1C); posterior margin of tergite VIII (Fig. 1F) broadly convex; median lobe of aedeagus (Figs 1I, J) slightly narrowed apically in lateral view; inner sac with flagellum coiled five times; apical lobe of paramerite (Fig. 1K) slightly dilated, apically with four setae.

Female: postocular margins immediately narrowed behind eyes (Fig. 1B); tergite VIII shaped as in Fig. 1H; spermatheca shaped as in Fig. 1L.

**Distribution.** Southwest China: Yunnan.

**Measurements. Male:** BL: 3.81–4.00; HL: 0.79–0.82; HW: 0.83–0.85; PL: 0.64–0.65; PW: 0.88–0.90; HW/HL: 1.05–1.06; PW/PL: 1.37–1.38; HW/PW:

0.94–0.95. **Female:** BL: 3.62; HL: 0.70; HW: 0.74; PL: 0.61; PW: 0.85; HW/HL: 1.06; PW/PL: 1.40; HW/PW: 0.87.

**Remarks.** *Tetrasticta bobbii* is most similar to *T. gnatha* in overall body shape, relatively long mandibles, but can be readily distinguished from it by the distinctly long antennal segments II–III, the different shape of abdominal tergite VIII and the form of aedeagal median lobe.

**Etymology.** Named after the Pomeranian dog of senior author.

## Acknowledgements

We thank Jia-Yao Hu and Liang Tang (both Shanghai, China) for collecting and providing the materials treated in this study. We thank Jan Klimaszewski (Quebec, Canada) for useful comments on a previous draft. We thank Munetoshi Maruyama (Fukuoka, Japan) and Roberto Pace (Verona, Italy) for sending us the reprints for our study. Two anonymous reviewers critically read the manuscript and provided helpful advice. The study is supported by the National Natural Science Foundation of China (No. 31101659 and No. 31172134, 31201734) and Shanghai Normal University (DZL125).

## References

- Kraatz G (1857) Beiträge zur Kenntniss der Termitophilen. *Linnaea Entomologica* 11: 44–56+pl. 1.
- Maruyama M (2004) Redescription of the genus *Creochara* (Coleoptera: Staphylinidae: Aleocharinae: Aleocharini) and its systematic position. *Canadian Entomologist* 136: 621–637.
- Maruyama M, Sugaya H (2002) A new species of *Tetrasticta* (Coleoptera, Staphylinidae, Aleocharinae) from Japan and Taiwan. *Japanese Journal of Systematic Entomology* 8(1): 17–21. doi: 10.4039/n03-091
- Pace R (2000) Aleocharinae della Thailandia (Coleoptera, Staphylinidae) (14 Contributo alla conoscenza delle Aleocharinae). *Bollettino del Museo Regionale di Scienze Naturali, Torino* 17(1): 39–86.
- Pace R (2008) Le specie di Thamiaracini, Oxypodini, Hoplandriini e Aleocharini del Borneo (Coleoptera, Staphylinidae). *Revue Suisse de Zoologie* 115(1): 157–183.
- Pace R (2010) Aleocharinae della regione orientale al museo di genova (Coleoptera, Staphylinidae). Estratto dagli Annali del Museo Civico di Storia Naturale “G. Doria” 102: 295–335.
- Pace R (2013a) Biodiversità della Aleocharinae della Cina: Hoplandriini, Aleocharini e Sinanarchusini (Coleoptera, Staphylinidae). *Contribution to Entomology* 63(1): 5–24.
- Pace R (2013b) New distributional data, new species and three new genera of Aleocharinae from Malaysia, Vietnam and Taiwan (Coleoptera: Staphylinidae). *Tropical Zoology* 26(1): 33–63. doi: 10.1080/03946975.2013.775832
- Yamamoto S, Maruyama M (2013) A peculiar new species of the genus *Tetrasticta* Kraatz (Coleoptera, Staphylinidae, Aleocharinae) from Peninsular Malaysia. *Zookeys* 336: 39–46. doi: 10.3897/zookeys.336.5382

# Three new species of *Melloleitaoina* Gerschman & Schiapelli, 1960 (Araneae, Mygalomorphae, Theraphosidae) from northern Argentina

Carlos Perafán<sup>1,†</sup>, Fernando Pérez-Miles<sup>1,‡</sup>

<sup>1</sup> Universidad de La República, Facultad de Ciencias, Sección Entomología, Iguá 4225, Montevideo, Uruguay

<sup>†</sup> <http://zoobank.org/F1E71689-C893-4F21-B756-224AA39A0BE5>

<sup>‡</sup> <http://zoobank.org/088FDA03-DD34-4D32-8342-7AAF91880EBC>

Corresponding author: Carlos Perafán (caperafanl@gmail.com)

Academic editor: I. Agnarsson | Received 13 September 2013 | Accepted 9 April 2014 | Published 24 April 2014

<http://zoobank.org/999F9E99-311D-4A28-A233-6D0984318757>

**Citation:** Perafán C, Pérez-Miles F (2014) Three new species of *Melloleitaoina* Gerschman & Schiapelli, 1960 (Araneae, Mygalomorphae, Theraphosidae) from northern Argentina. ZooKeys 404: 117–129. doi: 10.3897/zookeys.404.6243

## Abstract

Three new species of the monotypic genus *Melloleitaoina* Gerschman & Schiapelli, 1960 are described from northern Argentina: *M. mutquina* **sp. n.**, *M. uru* **sp. n.** and *M. yupanqui* **sp. n.** The female specimen originally described as *M. crassifemur* is not conspecific with the male holotype and thus is removed from this species and described as *M. uru* **sp. n.**; *M. crassifemur* is redescribed. All species are diagnosed, illustrated and a key to species is provided.

## Keywords

Tarantula, taxonomy, Theraphosinae

## Introduction

The subfamily Theraphosinae (Theraphosidae) is a speciose group of tarantulas distributed exclusively in the New World, whose greatest diversity is found in South America. Most tarantulas have cryptic habits, are predominantly nocturnal sit-and-wait hunters and have long lifespans. Females can live between 15 and 30 years while males live for significantly

shorter periods of time (Locht et al. 1999, Costa and Pérez-Miles 2002). Juveniles and adult females are sedentary while mature males disperse in search of females. Consequently several species have only been described on the basis of a single sex and the subsequent assignment of a specimen as a conspecific of the opposite sex is problematic.

The monotypic genus *Melloleitaoina* was established by Gerschman and Schiapelli (1960) on the basis of a single male specimen of the type species *Melloleitaoina crassifemur*, from Salta, Argentina. Later in 1973 the same authors illustrated the female spermathecae of a specimen from a location near the type locality in the same province. Raven (1985) considered *Melloleitaoina* a junior synonym of *Dryptopelma* Simon, 1889, but was restored by Pérez-Miles et al. (1996). This genus is morphologically similar and phylogenetically related to *Tmesiphantes*, *Plesiopelma* and *Homoeomma* (Pérez-Miles et al. 1996, Yamamoto et al. 2007).

*Melloleitaoina* is characterized by having a reduced number of labial cuspules, legs with few spines, all tarsal scopulae divided and lacking scopulae on metatarsal IV. Males have a thickened femur III, palpal organ with a long and curved embolus, and tibial apophysis with two very unequal branches. Females have spermathecae with two granulated seminal receptacles with a slight constriction near the apex (Gerschman and Schiapelli 1960, Gerschman and Schiapelli 1973, Pérez-Miles et al. 1996, Yamamoto et al. 2007).

Our study of the types and additional material deposited in the Museo Argentino de Ciencias Naturales “Bernardino Rivadavia”, led us to determine that the female of *M. crassifemur* is not conspecific with male holotype when contrasted with males from the same locality of the female. Also we found other individuals that fit with the diagnosis of the genus but are undescribed; they are described as new species herein.

## Material and methods

Urticating setae terminology follows Cooke et al. (1972) and Bertani (2002). Male palpal organ keel terminology follows Bertani (2000). All measurements were taken using an ocular micrometer and are given in millimeters (mm). We measured left legs and palps unless they were lost; measurements were taken in dorsal view along the central axis of the segments. Photographs were taken with a Lumenera Infinity Lite camera adapted to a stereoscopic microscope Olympus SZ 61. The geographic coordinates were determined using the Global Gazetteer ([www.fallingrain.com](http://www.fallingrain.com)). The distribution map was produced using DIVA-GIS 7.5 ([www.diva-gis.org](http://www.diva-gis.org)). The material examined is deposited in the Museo Argentino de Ciencias Naturales “Bernardino Rivadavia” (MACN).

The following abbreviations are used: ALE = anterior lateral eyes; AME = anterior median eyes; OQ = ocular quadrangle (including lateral eyes); p = prolateral; PB = prolateral branch of tibial apophysis; PI = prolateral inferior keel; PME = posterior median eyes; PMS = posteromedial spinnerets; PLE = posterior lateral eyes; PLS = posterolateral spinnerets; PS = prolateral superior keel; r = retrolateral; RB = retrolateral branch of tibial apophysis.



## Taxonomy

### Genus *Melloleिताoina* Gerschman & Schiapelli, 1960

<http://species-id.net/wiki/Melloleिताoina>

**Type species.** *Melloleिताoina crassifemur* Gerschman & Schiapelli, 1960

**Diagnosis.** Both sexes have a reduced number of labial cuspules (6–14), all tarsal scopulae divided and metatarsal IV scopulae absent. Males differ from other Theraphosinae by having a thickened femur III, palpal organ with a long and curved embolus with two prolateral keels (PI and PS) (Figs 3–4, 9–10, 16–17 and 26–27), and tibial apophysis with two very unequal branches (Figs 6, 11, 21 and 29). Females differ from other Theraphosinae by having spermathecae with two granulated seminal receptacles with a slight constriction near the apex (Figs 15 and 25) and spiniform setae on promargin of coxae III and IV (Figs 19 and 20). Females have type IV urticating setae while males have III–IV intermediated urticating setae.

**Affinities.** *Melloleिताoina* species share with *Plesiopelma* Pocock, 1901 and *Tmesiphantes* Simon, 1892, principally by the general morphology of the palpal bulb and tibial apophysis. *Melloleिताoina* males can be distinguished additionally from those of *Plesiopelma* by lacking nodule on metatarsi I and having only III–IV intermediated urticating setae. They can be distinguished from *Tmesiphantes* by having sigillas more rounded, male femur III incrassate and female spermathecae with granulated seminal receptacles and spiniform setae on promargin of coxae III and IV.

**Distribution.** Northern Argentina. Catamarca, Salta and Jujuy provinces (Fig. 1).

### *Melloleिताoina crassifemur* Gerschman & Schiapelli, 1960

[http://species-id.net/wiki/Melloleिताoina\\_crassifemur](http://species-id.net/wiki/Melloleिताoina_crassifemur)

Figs 2–6

**Material examined.** Only type material.

**Type material.** Holotype male from Argentina, Salta, Orán, Urundel, 335m above sea level, 23°33'0"S, 64°25'0"W, viii-1947, Misión Ricardo N. Orfila leg. (MACN-Ar 2285).

**Diagnosis.** Male differs from other *Melloleिताoina* species by the palpal bulb morphology with very curved embolus without triangular tooth, well-developed and subequal PI and PS (Figs 3 and 4), and apex widened (Fig. 5). Females unknown.

**Re-description.** Holotype male (MACN-Ar 2285): total length, not including chelicerae or spinnerets, 14.1, carapace length 5.9, width 5.3. Color (in alcohol): cephalothorax, legs light brown, abdomen grayish brown. Anterior eye row slightly procurved, posterior recurved. Eyes and interdistances: AME 0.18, ALE 0.30, PME 0.14, AME-ALE 0.06, PME-PME 0.38, PME-PL 0.04, ALE-PL 0.08, AME-PME 0.04, ALE-ALE 0.40. OQ length 0.50, width 0.96, clypeus 0.14. Fovea transverse, procurved, width 0.70. Chelicerae with 10/12 well-developed teeth on furrow promargin, few small teeth on the proximal area of furrow. Labium length 0.68, width 1.07 with

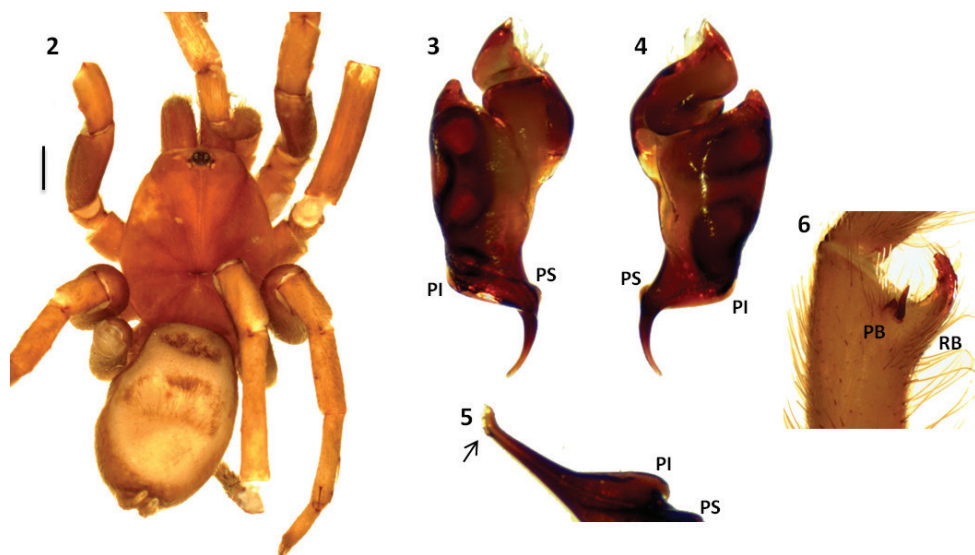


**Figure 1.** Map of Northern Argentina, geographical distribution of the *Melloleitaoina* species. *Melloleitaoina uru* (triangles); *Melloleitaoina crassifemur* (circle); *Melloleitaoina yupanqui* (cross); *Melloleitaoina mutquina* (square).

7 cuspules. Maxillae with 74 cuspules. Sternum length 2.70, width 2.70. Tarsi I-IV scopula widely divided, by conical setae thicker and longer. Tarsal claws with 2-3 small teeth on proximal half, near the inner edge. Sparse scopulae on metatarsi; metatarsi I-III apically scopulate, IV without scopula. Tibia I with prolatero-ventral distal apophysis with two very unequal branches (Fig. 6); PB subtriangular, small, with basal curved spine, much longer than branch, RB curved, much larger than PB with internal medial spine that exceeds length of branch. Metatarsus I slightly curved, flexion on RB. Femur III very thickened. Type III-IV intermediate urticating setae present. PMS well-developed, PLS normal, apical segment digitiform. Palpal organ piriform with the embolus very curved, two prolateral keels (PI and PS) present, subequal, apex widened (Figs 3–5).

Spination. Femora: palp, I-IV 0. Patellae: palp, I-IV 0. Tibiae: palp 0, I 0, II 1P, III 2V, 1P, 2R, IV 1R. Metatarsi: I 1V, II 1V, III 3V, 2P, IV 4V, 2P, 1R. Tarsi: palp, I-IV 0.

Legs and palpal segments lengths (femur/patella/tibia/metatarsus/tarsus). Palp: 3.1/2.0/2.5/1.2 total 8.8; I: 5.5/3.5/4.4/3.8/2.4 total 19.6; II: 4.9/1.9/3.6/4.4/2.3 total 17.1; III: 3.5/2.0/2.8/3.7/2.2 total 14.2; IV: 6.0/2.2/5.0/6.3/2.5 total 22.



**Figures 2–6.** *Melloleिताoina crassifemur*. **2** male holotype, dorsal view **3–5** left palpal bulb, **3** prolateral view **4** retrolateral view **5** detail of apex widened **6** left tibial apophysis (subapical spine on retrolateral branch RB lost). Scale bar = 1 mm.

**Remarks.** The female *M. crassifemur* was described thirteen years after the original description of the male holotype of the species. This female specimen was assigned as *M. crassifemur* because it was collected near the male type locality. We examined male specimens from the same locality of this female and found important morphological differences between these males and the holotype *M. crassifemur*, as the palpal bulb shape, the presence of a conspicuous triangular tooth on the embolus (Figs 16 and 17) and the spiniform setae on coxae III and IV (Figs 19–21), also it present on the female. Taking into account that these new males are sympatric with the female attributed to *M. crassifemur* we considered them as conspecific, and are here described as a new species.

***Melloleिताoina mutquina* sp. n.**

<http://zoobank.org/ECCA9985-CFAE-4CBE-98EE-30154F1CD4E6>

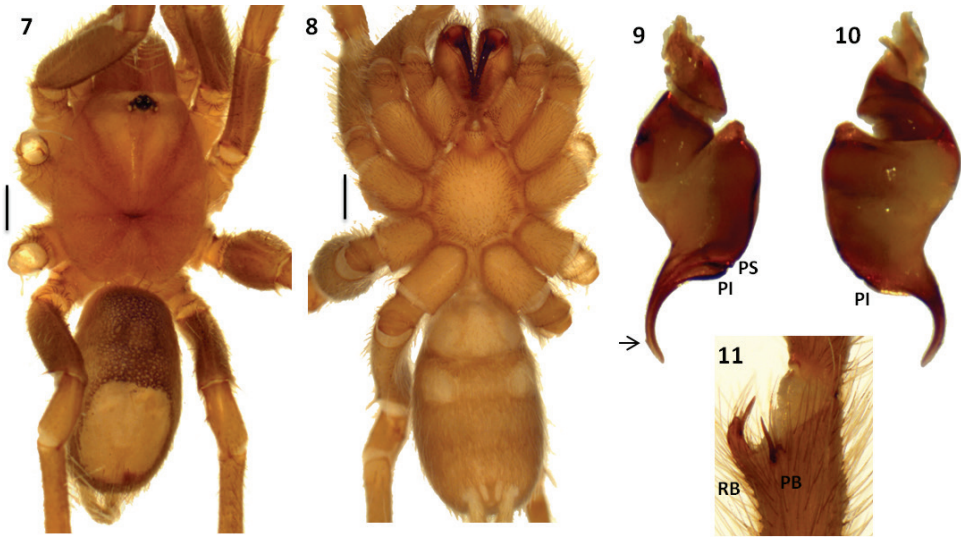
[http://species-id.net/wiki/Melloleिताoina\\_mutquina](http://species-id.net/wiki/Melloleिताoina_mutquina)

Figs 7–11

**Material examined.** Known only from types.

**Type material.** Holotype male from Argentina, Catamarca, Mutquín, 1500m above sea level, 28°19'0"S, 66°10'0"W, 2-ii-1981, E. Maury leg. (MACN-Ar 7737).

**Diagnosis.** Male differs from other *Melloleिताoina* species by the palpal bulb morphology with the embolus less curved, absence of triangular tooth, PS very flat and apex widened (Figs 9 and 10). Females unknown.



**Figures 7–11.** *Melloleitaoina mutquina*. 7–8 male holotype 7 dorsal view 8 ventral view 9–10 right palpal bulb 9 prolateral view 10 retrolateral view 11 right tibial apophysis. Arrow indicates apex widened. Scale bars = 1 mm.

**Description.** Holotype male (MACN-Ar 7737): total length, not including chelicerae or spinnerets, 9.4, carapace length 4.1, width 3.7. Color (in alcohol): cephalotorax, legs light reddish brown, cephalotorax with few brown and golden setae, abdomen brown with a patch of urticating setae golden brown. Anterior eye row procurved, posterior slightly recurved. Eyes and interdistances: AME 0.15, ALE 0.20, PME 0.11, PLE 0.15, AME-AME 0.11, AME-ALE 0.066, PME-PME 0.33, PME-PL 0.022, ALE-PL 0.077, AME-PME 0.055, ALE-ALE 0.40. OQ length 0.68, width 0.61, clypeus 0.022. Fovea transverse, procurved, width 0.66. Chelicerae with 10/9 well-developed teeth on furrow promargin, 5/3 small teeth on the proximal area of furrow. Labium length 0.48, width 0.78, with 6 cuspules. Maxillae with 38/39 cuspules. Sternum length 1.9, width 1.9. Tarsi I–IV scopula widely divided. Tarsal claws with 1–4 small teeth on proximal half, ventral midline. Sparse scopulae on metatarsi; metatarsus I scopulate on distal third, II apically, III and IV without scopula. Tibia I with prolate-ventral distal apophysis with two very unequal branches (Fig. 11); PB subtriangular, small, with basal curved spine, much longer than branch, RB curved, around ten times bigger than PB with internal medial spine that exceeds length of branch. Metatarsus I slightly curved, flexion on RB. Femur III thickened. Type III–IV intermediate urticating setae present. Palpal organ piriform with the embolus slightly curved, two prolateral keels (PI and PS) present, PS very flat, apex widened (Figs 9 and 10).

**Spination.** Femora: palp 0; I 0; II 0; III 0; IV 0. Patellae: palp 0; I 0; II 0; III 0; IV 0. Tibiae: palp 0; I 0; II 1P; III 1V, 1P; IV 1V, 1P, 1R. Metatarsi: I 1V; II 1V; III 3V, 2P; IV 4V, 1P. Tarsi: palp, I–IV 0.



Legs and palpal segments lengths (femur/patella/tibia/metatarsus/tarsus). Palp: 2.2/1.3/1.5/0.8 total 5.8; I: 3.5/2.0/2.9/2.1/1.4 total 11.9; II: 3.0/1.7/2.3/2.1/1.4 total 10.5; III: 2.7/1.4/1.8/2.3/1.4 total 9.6; IV: 3.8/1.7/3.0/4.0/1.8 total 14.3.

**Etymology.** The specific epithet *mutquina* is a noun taken in apposition and means place or thing to smell in Quichua language and refers to the locality of Mutquín, where this species is distributed. It denotes the aroma of the flora of the region that emerges especially after rains, perfuming the village of aromatics herbs.

***Melloleिताoina uru* sp. n.**

<http://zoobank.org/60708C57-F159-46CC-9EB3-3FFCEE3FE7A>

[http://species-id.net/wiki/Melloleिताoina\\_uru](http://species-id.net/wiki/Melloleिताoina_uru)

Figs 12–21

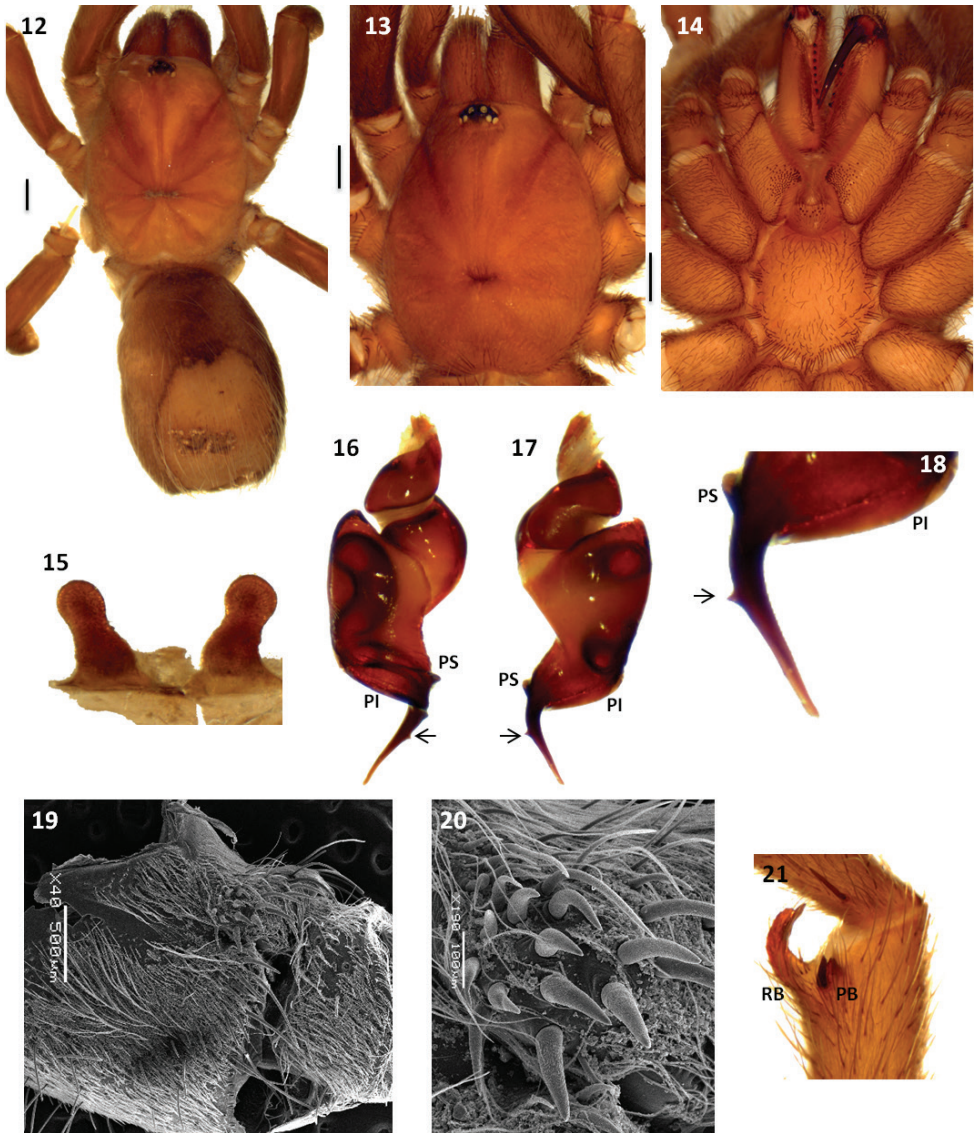
*Melloleिताoina crassifemur*: Gerschman and Schiapelli 1973: 87, Fig. 50 (in part, female only). **Syn n.**

**Material examined. Type material.** Holotype male from Argentina, Salta, 37,5 Km O. Hickmann, 235m above sea level, 23°11'60"S, 63°34'0"W, Goloboff, Coyle, Bennett leg. (MACN-Ar 26042). Paratypes: female from Argentina, Salta, Aguaray, Punilla, 570m above sea level, 22°16'0"S, 63°43'60"W, iv-1948, Biraben leg. (MACN-Ar 6542); 2 males and 1 female, with the same data (MACN-Ar 6543); male from Argentina, Salta, Campamento Vespucio, 450m above sea level, 22°36'0"S, 63°49'0"W, 10-13-v-1988, Goloboff leg. (MACN-Ar 26043); 2 females from Argentina, Salta, Pocitos (Salvador Mazza), 800m above sea level, 22°4'0"S, 63°43'0"W, xi-1951; Biraben leg. (MACN-Ar 6544).

**Other material.** Juvenile from Argentina, Salta, Aguaray, Punilla, 22°16'0"S, 63°43'60"W, iv-1948, Biraben leg. (MACN-Ar 6542); 3 juveniles from Argentina, Salta, Pocitos (Salvador Mazza), 22°4'0"S, 63°43'0"W, xi-1951; Biraben leg. (MACN-Ar 6544).

**Diagnosis.** Males differ from other *Melloleिताoina* species by the palpal bulb morphology with very curved embolus with a conspicuous subapical triangular tooth and well-developed PI and PS (Figs 16–18). Females differ from other *Melloleिताoina* species by the shape of the spermathecae with elongated seminal receptacles with small granules (Fig. 15).

**Description.** Holotype male (MACN-Ar 26042): total length, not including chelicerae or spinnerets, 13.8, carapace length 6.5, width 5.5. Color (in alcohol): cephalotorax, legs clear reddish brown, cephalotorax with few brown and golden setae, abdomen brown with a patch urticating setae golden brown. Anterior eye row slightly procurved, posterior slightly recurved. Eyes and interdistances: AME 0.17, ALE 0.27, PME 0.17, PLE 0.22, AME-AME 0.12, AME-ALE 0.06, PME-PME 0.43, PME-PLE 0.025, ALE-PLE 0.06, AME-PME 0.075, ALE-ALE 0.50. OQ length 0.97, width 0.72, clypeus 0.025. Fovea transverse, procurved, width 0.75. Chelicerae with 8 well-developed teeth



**Figures 12–21.** *Melloleitaoina uru*. **12** female, dorsal view **13–14** male holotype **13** cephalotorax **14** sternum, labium, maxillae and quelicerae **15** spermathecae **16–18** left palpal bulb **16** prolateral view **17** retrolateral view **18** detail of triangular tooth on embolus **19–20** coxa III **19** prolateral view **20** detail of spiniform setae **21** right tibial apophysis. Arrow indicates triangular tooth on embolus. Scale bars black = 1 mm.

on furrow promargin, 12/14 small teeth on the proximal area of furrow. Labium length 0.57, width 1.45, with 11 cuspules. Maxillae with 109/114 cuspules. Sternum length 2.6, width 2.6. Coxae III and IV with spiniform setae on promargin (Figs 19–20). Tarsi I–IV scopula widely divided. Tarsal claws with 3 small teeth on proximal half, ventral midline. Sparse scopulae on metatarsi; metatarsus I scopulate on distal half, II on distal third, III apically, IV without scopula. Tibia I with prolatero-ventral distal apophysis with two very unequal branches (Fig. 21); PB subtriangular, small, with basal spine, similar size to the branch, RB curved, at least five times bigger than PB with internal medial spine that exceeds length of branch. Metatarsus I slightly curved, flexion on RB. Femur III thickened. Type III–IV intermediate urticating setae present. Palpal organ piriform with the embolus very curved and with a conspicuous subapical triangular tooth, two prolateral keels (PI and PS) present, subequal (Figs 16–18).

Spination. Femora: palp 1P; I 1P; II 1P; III 1P, 1R; IV 1R. Patellae: palp 0; I 0; II 0; III 1P; IV 0. Tibiae: palp 2P; I 2V, 1P; II 3V, 1P; III 7V, 2P, 2R; IV 7V, 2P, 3R. Metatarsi: I 2V, 1P; II 2V, 1P; III 9V, 3P, 2R; IV 10V, 3P, 3R. Tarsi: palp, I–IV 0.

Legs and palpal segments lengths (femur/patella/tibia/metatarsus/tarsus). Palp: 3.2/2.0/2.7/1.1 total 9.0; I: 5.3/2.7/4.0/3.6/2.3 total 17.9; II: 4.7/2.4/3.3/3.2/2.1 total 15.7; III: 3.9/1.9/2.5/3.7/2.1 total 14.1; IV: 5.9/2.4/5.0/6.6/2.3 total 22.2.

Paratype female (MACN-Ar 6542): total length, not including chelicerae or spinnerets, 14.8, carapace length 6.6, width 5.7. Color (in alcohol): as in male. Anterior eye row slightly procurved, posterior slightly recurved. Eyes and interdistances: AME 0.17, ALE 0.31, PME 0.17, PLE 0.25, AME–AME 0.15, AME–ALE 0.06, PME–PME 0.4, PME–PLE 0.025, ALE–PLE 0.075, AME–PME 0.10, ALE–ALE 0.56. OQ length 1.02, width 0.95, clypeus 0.025. Fovea transverse, procurved, width 1.12. Chelicerae with 10 well-developed teeth on furrow promargin, 12 small teeth on the proximal area of furrow. Labium length 0.62, width 1.42, with 6 cuspules. Maxillae with 96/89 cuspules. Sternum length 2.9, width 2.9. Coxae III and IV with spiniform setae on promargin (Figs 19–20). Tarsi palp, I–IV scopula widely divided. Tarsal claws with 3 small teeth on proximal half, ventral midline. Sparse scopulae on metatarsi; metatarsus I scopulate on distal half, II on distal third, III and IV without scopula. Type IV urticating setae present. Spermathecae with two elongated seminal receptacles with small granules (Fig. 15).

Spination. Femora: palp 0; I 0; II 1P; III 1P, 1R; IV 0. Patellae: palp 0; I 0; II 0; III 1P; IV 0. Tibiae: palp 3V; I 0; II 0; III 4V, 3P, 1R; IV 7V, 3P, 2R. Metatarsi: I 2V; II 3V; III 6V, 3P, 2R; IV 10V, 3P, 3R. Tarsi: palp, I–IV 0.

Legs and palpal segments lengths (femur/patella/tibia/metatarsus/tarsus). Palp: 3.2/2.4/2.0/2.1 total 9.7; I: 4.0/2.5/3.0/2.4/1.5 total 13.4; II: 3.8/2.5/2.7/2.4/1.6 total 13.0; III: 3.2/2.4/2.0/3.0/1.6 total 12.2; IV: 4.7/2.6/4.0/5.0/2.0 total 18.3.

**Variation.** Males and females total length 9.2–16.5. Labium cuspules 6–14.

**Etymology.** The specific epithet is a noun taken in apposition and refers to an ancient legend Quichua, from the northern limit of Argentina, about the princess Inca Uru, who by their whims and bad government was transformed by the gods into a spider and forced to endlessly work weaving.

***Melloleिताoina yupanqui* sp. n.**

<http://zoobank.org/3BD376C4-CA74-4A6A-8D2F-99445C54587E>

[http://species-id.net/wiki/Melloleिताoina\\_yupanqui](http://species-id.net/wiki/Melloleिताoina_yupanqui)

Figs 22–29

**Material examined.** Known only from types.

**Type material.** Holotype male from Argentina, Jujuy, P. Nacional Calilegua, Seccional Aguas Negras, 605m above sea level (GPS), 23°45'43,3"S, 64°51'04,7"W ( $\pm$  10m, WGS84), 06-11-xii-2008, C. Grismado, M. Izquierdo, F. Labarque, G. Rubio, M. Burger, P. Michalik, P. Carrera, A. Ojanguren and C. Mattoni leg. (MAC-Ar 26041). Paratype female, same data as the holotype (MAC-Ar 26044).

**Diagnosis.** Male differs from other *Melloleिताoina* species by the palpal bulb morphology with a discontinuous PS, formed by two separate keels, very curved embolus without triangular tooth, well-developed PI and PS, and apex widened (Figs 26 and 28). Female differs from other *Melloleिताoina* species by the shape of the spermathecae with short seminal receptacles with large granules (Fig. 25).

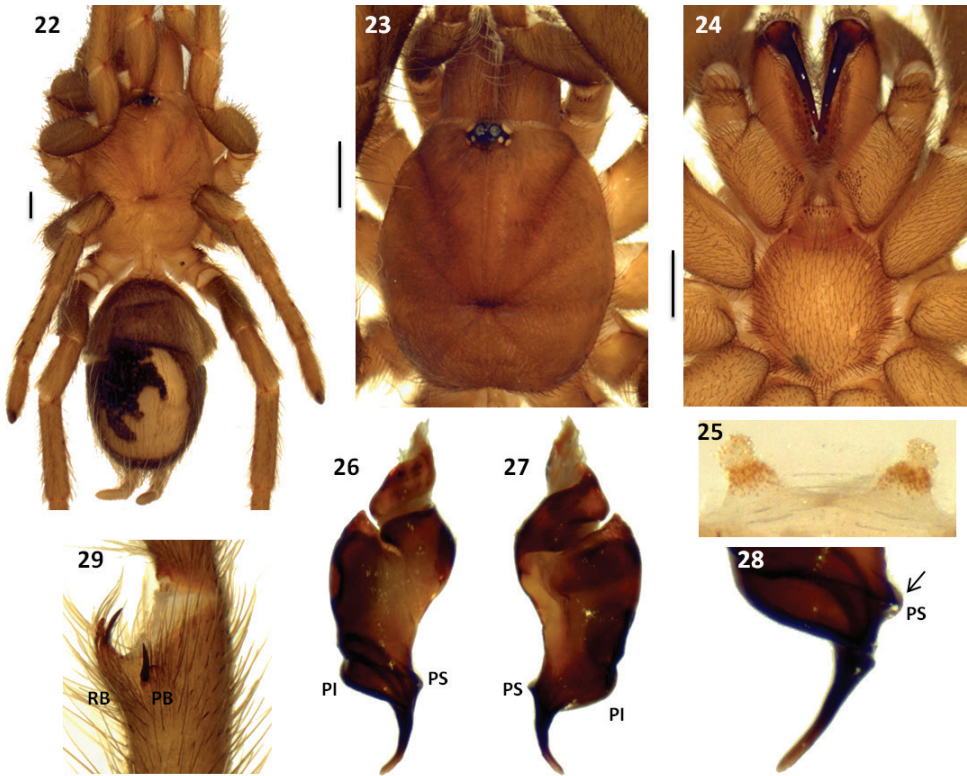
**Description.** Holotype male (MAC-Ar 26041): total length, not including chelicerae or spinnerets, 9.3, carapace length 4.3, width 4.0. Color (in alcohol): cephalotorax, legs light reddish brown, cephalotorax with few brown and golden setae, legs darker, mainly femora, abdomen brown with a patch of urticating setae golden brown. Anterior eye row slightly procurved, posterior slightly recurved. Eyes and interdistances: AME 0.17, ALE 0.22, PME 0.12, PLE 0.16, AME-AME 0.088, AME-ALE 0.033, PME-PME 0.36, PME-PL 0.022, ALE-PL 0.055, AME-PME 0.022, ALE-ALE 0.39. OQ length 0.75, width 0.55, clypeus 0.022. Fovea transverse, procurved, width 0.68. Chelicerae with 11 well-developed teeth on furrow promargin, 8/10 small teeth on the proximal area of furrow. Labium length 0.48, width 0.88, with 8 cuspules. Maxillae with 53/51 cuspules. Sternum length 2.0, width 2.0. Tarsi I-IV scopula widely divided. Tarsal claws with 2 small teeth on proximal half, near the inner edge. Sparse scopulae on metatarsi; metatarsus I scopulate on distal half, II scopulate on distal third, III only apically scopulate, IV without scopula. Tibia I with prolatero-ventral distal apophysis with two very unequal branches (Fig. 29); PB very short with basal spine, much longer than branch, RB curved, around ten times bigger than PB with internal medial spine that exceeds the length of branch. Metatarsus I slightly curved, flexion on RB. Femur III thickened. Type III-IV intermediate urticating setae present. Palpal organ piriform with the embolus very curved, two prolateral keels (PI and PS) present, discontinuous PS, formed by two keels, apex widened (Figs 26–28).

Spination. Femora: 0. Patellae: 0. Tibiae: palp 0; I 1P; II 0; III 2V, 2P, 1R; IV 4V, 2R. Metatarsi: I 1V; II 1V; III 6V, 3P, 1R; IV 8V, 1P, 2R. Tarsi: palp, I-IV 0.

Legs and palpal segments lengths (femur/patella/tibia/metatarsus/tarsus). Palp: 2.4/1.3/1.9/0.7 total 6.3; I: 3.9/2.2/3.3/2.7/1.7 total 13.8; II: 3.6/2.0/2.6/2.4/1.7 total 12.3; III: 2.9/1.4/1.8/2.5/1.6 total 10.2; IV: 4.6/1.7/3.6/4.9/2.2 total 17.0.

Paratype female (MAC-Ar 26044): total length, not including chelicerae or spinnerets, 10.6, carapace length 4.9, width 4.0. Color (in alcohol): as in male, but lighter.





**Figures 22–29.** *Melloleिताoina yupanqui*. **22** female, dorsal view **23–24** male holotype **23** cephalothorax **24** sternum, labium, maxillae and quelicerae **25** spermathecae **26–28** left palpal bulb **26** prolateral view **27** retrolateral view **28** detail of embolus showing PS discontinuous **29** right tibial apophysis. Scale bars = 1 mm

Anterior eye row slightly procurved, posterior slightly recurved. Eyes and interdistances: AME 0.17, ALE 0.31, PME 0.19, PLE 0.21, AME-AME 0.08, AME-ALE 0.04, PME-PME 0.31, PME-PLE 0.022, ALE-PLE 0.044, AME-PME 0.066, ALE-ALE 0.39. OQ length 0.81, width 0.66, clypeus 0.022. Fovea transverse, procurved, width 0.71. Chelicerae with 9 well-developed teeth on furrow promargin, 15/14 small teeth on the proximal area of furrow. Labium length 0.55, width 1.1, with 8 cuspules. Maxillae with 90/87 cuspules. Sternum length 2.1, width 2.1. Coxae III and IV with spiniform setae on promargin (as Figs 19 and 20). Tarsi palp, I-IV scopula widely divided. Tarsal claws with 2 small teeth on proximal half, near the inner edge. Sparse scopulae on metatarsi; metatarsus I scopulate on distal half, II on distal third, III and IV without scopula. Type IV urticating setae present. Spermathecae with two short seminal receptacles with large granules (Fig. 25).

Spination. Femora: palp 1P; I 1P; II 1P; III 1P, 1R; IV 0. Patellae: palp 0; I 0; II 0; III 1P; IV 0. Tibiae: palp 4V; IO; II 0; III 5V, 2P, 2R; IV 5V, 2R. Metatarsi: I 2V; II 2V; III 9V, 3P, 2R; IV 7V, 2P, 2R. Tarsi: palp, I-IV 0.

Legs and palpal segments lengths (femur/patella/tibia/metatarsus/tarsus). Palp: 2.5/1.6/1.7/1.6 total 7.4; I: 3.3/2.2/2.5/1.8/1.4 total length 11.2; II: 2.9/1.9/2.0/1.7/1.4 total 9.9; III: 2.6/1.5/1.7/2.3/1.4 total 9.5; IV: 3.7/1.7/2.8/3.3/1.7 total 13.2.

**Etymology.** The specific epithet is a patronym in honor to the most important Argentine musician of folklore Atahualpa Yupanqui, pseudonym of Héctor Roberto Chavero Aramburu (Juan A. de la Peña, Argentina, 1908 – Nimes, Francia, 1992).

### Key to males of *Melloleिताoina* species

- 1 Palpal bulb with a triangular tooth on the embolus (Figs 16–18).....*M. uru*
- Palpal bulb without a triangular tooth on the embolus .....2
- 2 Embolus slightly curved and very flat PS (Figs 9 and 10) ..... *M. mutquina*
- Embolus very curved and well developed PS.....3
- 3 Embolus with continues PS (Figs 3 and 4).....*M. crassifemur*
- Embolus with discontinuous PS (Figs 26–28).....*M. yupanqui*

### Acknowledgements

We would like to thank Martín Ramírez and Cristian Grismado from the Museo Argentino de Ciencias Naturales “Bernardino Rivadavia” for lending us all specimens reviewed and for his collaboration. Thanks to referees for the careful work and valuable comments. The author (C.P.) wants to thank CSIC, Universidad de la República, Uruguay, for the financial support (CSIC C 311/102). Open access to this paper was supported by the Encyclopedia of Life (EOL) Open Access Support Project (EOASP).

### References

- Bertani R (2000) Male palpal bulbs and homologous features in Theraphosinae (Araneae, Theraphosidae). J Arachnol. 28: 29–42. doi: 10.1636/0161-8202(2000)028[0029:MPB AHF]2.0.CO;2
- Bertani R (2002) Morfologia e evolução das cerdas urticantes em Theraphosidae (Araneae). PhD thesis, Instituto de Biociências, Universidade de São Paulo, São Paulo, Brasil.
- Cooke JA, Roth VD, Miller F (1972) The urticating hairs of Theraphosidae. Am Mus Novit. 2498: 1–43.
- Costa FG, Pérez-Miles F (2002) Reproductive biology of Uruguayan theraphosids (Araneae, Mygalomorphae). J Arachnol. 30: 571–587. doi: 10.1636/0161-8202(2002)030[0571:RB OUTA]2.0.CO;2
- Gerschman de Pikelin BS, Schiapelli RD (1973) La subfamilia Ischnocolinae (Araneae: Theraphosidae). Revta Mus. argent. Cien. Nat. Bernardino Rivadavia (Ent.) 4: 43–77.

- Gerschman de Pikelin BS, Schiapelli RD (1960) Un nuevo género con una nueva especie de Ischnocolinae (Araneae-Theraphosidae). *Physis* XXI (61): 200–206.
- Locht A, Yanez M, Vazquez I (1999) Distribution and natural history of Mexican species of *Brachypelma* and *Brachypelmides* (Theraphosidae, Theraphosinae) with morphological evidence for their synonymy. *J Arachnol.* 27: 196–200.
- Pérez-Miles F, Lucas SM, da Silva Jr PI, Bertani R (1996) Systematic revision and cladistic analysis of Theraphosinae (Araneae: Theraphosidae). *Mygalomorph.* 1: 33–68.
- Raven RJ (1985) The spider Infraorder Mygalomorphae (Araneae): cladistics and systematics. *Bull Am Mus Nat Hist.* 182: 1–180.
- Yamamoto FU, Lucas SM, Guadanucci JPL, Indicatti RP (2007) Revision of the genus *Tmesiphantes* Simon (Araneae, Mygalomorphae, Theraphosidae). *Revta bras. Zool.* 24: 971–980.

