RESEARCH ARTICLE



# A new species of parasitic copepod, Sarcotretes umitakae sp. n. (Siphonostomatoida, Pennellidae), on the rattail (Actinopterygii, Macrouridae) from the East China Sea, Japan

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Academic editor: D. Defaye | Received 19 August 2012 | Accepted 14 November 2012 | Published 29 November 2012 urn:lsid:zoobank.org:pub:B758624F-BFA9-4E73-8E87-800A3E049335

**Citation:** Uyeno D, Wakabayashi K, Nagasawa K (2012) A new species of parasitic copepod, *Sarcotretes umitakae* sp. n. (Siphonostomatoida, Pennellidae), on the rattail (Actinopterygii, Macrouridae) from the East China Sea, Japan. ZooKeys 246: 1–10. doi: 10.3897/zookeys.246.3872

## Abstract

A new species of copepod, *Sarcotretes umitakae* **sp. n.**, of the siphonostomatoid family Pennellidae is described based on female specimens from the rattail *Coelorinchus jordani* Smith and Pope (Actinopterygii: Gadiformes: Macrouridae) caught in the East China Sea. This species is characterized by exhibiting the following characters: the large proboscis projects strongly; the head bears paired lateral processes which are bulbous and taper into a slender horn; the twisting neck is significantly longer than the trunk; and the trunk bears an anterior constriction with a reduced abdomen.

#### **Keywords**

Mesoparasitic copepods, Sarcotretes umitakae sp. n., new species, East China Sea, rattail, mesopelagic fishes

## Introduction

Sarcotretes Jungersen, 1911, a pennellid genus, was originally established based on S. scopeli Jungersen, 1911 from Ireland, the eastern North Atlantic (Jungersen 1911). Wilson (1917) included six species in this genus: S. scopeli (type species), S. eristaliformis (Brian, 1908), S. gempyli (Horst, 1879), S. inflexus (Steenstrup and Lütken, 1861), S. nodicornis (Steenstrup and Lütken, 1861), and S. lobatus Wilson C.B., 1917. Subsequently, S. inflexus, S. nodicornis, S. gempyli, and S. lobatus were considered to be junior synonyms of S. scopeli by Hogans (1988). Recently, S. longirostris Ho, Nagasawa and Kim, 2007 was described, thus a total of three species are considered as valid in this genus at present (Ho et al. 2007). Members of the genus are parasitic on mesopelagic fishes (Cherel and Boxshall 2004), and they were often been found in food samples of penguins and whales (e.g. Cherel and Boxshall 2004; Ho et al. 2007). In this study, a new species of the genus is described based on females from the rattail Coelorin-chus jordani Smith and Pope (Actinopterygii: Gadiformes: Macrouridae) caught in the mesopelagic zone of the East China Sea.

### Materials and methods

Two specimens of the rattail C. jordani infected with copepods were caught in the East China Sea off the Tokara Islands, Kagoshima, Japan on 8 October 2011 during the cruise (UM-11-06) of the Umitaka-maru, a training and research vessel of Tokyo University of Marine Science and Technology (TUMSAT). The fishes were collected using an otter trawl towed for 30 minutes between two sites (29°58.02'N, 127°43.79'E to 29°59.57'N, 127°44.28'E) around 309 m in depth and, immediately after capture, they were preserved in 70% ethanol with copepods attached. In the laboratory, copepods were carefully removed from the tissues of fishes, and then soaked in lactophenol for a whole day before dissection. The appendages of the copepods were observed after dissecting with the method of Humes and Gooding (1964). Drawings were made with the aid of a drawing tube. Morphological terminology follows Huys and Boxshall (1991). Measurements (in millimeters) are shown as ranges, followed by means and standard deviations in parentheses. The flexed part was measured along the body axis. Type specimens are deposited in the crustacean collection at the National Museum of Nature and Science, Tsukuba (NSMT). The scientific name of fish follows Nakabo (2002).

#### Results

# Order Siphonostomatoida Burmeister, 1835 Family Pennellidae Burmeister, 1835 Genus *Sarcotretes* Jungersen, 1911

## Sarcotretes umitakae sp. n.

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**Type material.** Holotype female (NSMT–Cr 22253) and 2 paratypic females (NSMT–Cr 22254), ex *Coelorinchus jordani* Smith and Pope (Gadiformes: Macrouridae), taken off the Tokara Islands (29°58.02'N, 127°43.79'E to 29°59.57'N, 127°44.28'E), Kagoshima, East China Sea, Japan, 308.5–309.3 m depth, 8 October 2011, reg. K. Wakabayashi and Y. Tanaka.

**Type locality.** off the Tokara Islands (29°58.02'N, 127°43.79'E to 29°59.57'N, 127°44.28'E), Kagoshima, East China Sea, Japan.

**Description of postmetamorphic adult female.** Body (Fig. 2A) elongate, comprising head, neck, and trunk. Total length 43.42 (from tip of cephalothorax to end of abdomen). Head (holdfast) composed of cephalosome to third pediger (Fig. 2A, B), bearing elongate oral area projecting forward as cylindrical proboscis with multiple constrictions and mouth tube at its tip, with paired lateral processes (Fig. 2A, B) consisting of bulbous base drawn out into highly sclerotized horn-like process. Vestige of dorsal shield of cephalothorax and tergites of first to third visible on dorsal surface of head (Fig. 2D). Two paired small sclerites on ventral surface of basal region of oral cone (Fig. 2E). Neck (Fig. 2A) slender, longer than trunk, twisting and bearing bulge and constriction at posterior portion. Cylindrical trunk (Fig. 2A) 12.23 long (from enlarged end of neck to abdomen), 3.08 wide at widest part bearing paired hemispherical protrusions and reduced abdomen (Fig. 2F, G). Caudal rami absent.

Rostral area (Fig. 2H) triangular. Antennule (Fig. 3A) not segmented, located on sclerotized protrusion, bearing 10 blunt, long elements, and at least 17 short elements; 1 long distal seta with bifurcated tip. Antenna (Fig. 3B) subchelate, 3-segmented; proximal segment, unarmed; middle segment stout with a pointed process on innerdistal corner, hollowed out to receive terminal claw; terminal segment representing terminal claw with single basal seta. Mandible (Fig. 3C) located on lateral side of base of oral cone (Fig. 2E), represented by sclerotized process with unequal processes tip. Maxillule (Fig. 3D) bilobate; large inner lobe tipped with two naked setae; small outer lobe bearing one naked seta. Maxilla (Fig. 3E) 2-segmented; proximal segment rod-like, bearing round protrusion with pointed process in middle portion; terminal segment rod-like, tipped with curved spinulose process, small pointed process, and setulous lobe (Fig. 3F). Maxilliped absent.



**Figure 1.** *Sarcotretes umitakae* sp. n., female on *Coelorinchus jordani* Smith and Pope. **A** two specimens of *C. jordani* (181.5 mm TL and 142.8 mm TL) carrying the type series of *Sarcotretes* sp. n. (arrowheads) **B** coloration in life of paratype NSMT–Cr 22254 attached to host's body. Scale bars: **A**=20 mm; **B**=3 mm.

Legs 1 to 4 occurring tightly together and located between paired lateral processes of holdfast. Legs 1 and 2 (Figs. 3G, 4A) biramous, composed of inter coxal sclerites, protopods, and 2-segmented rami. Leg 3 (Fig. 4B) uniramous, without endopod; leg armature formula as follows:

	Protopod	Exopod	Endopod
Leg 1	1-0	I–1; I, I, 5	0–1;7
Leg 2	1-0	I–1; I, I, 5	0–1;7
Leg 3	1-0	0–0; I, I, 4	absent

Leg 4 (Fig. 4C) represented by vestigial intercoxal sclerite. Legs 5 and 6 absent.

*Variability of female.* The necks of all paratypes twist and turn in complex fashion (Fig. 4D). Measurements of the body parts of the specimens from the type series (n = 3) are as follows: body length (anterior margin of the head to distal end of the posterior processes on the trunk) 30.26-50.12 ( $41.27 \pm 10.10$ ); trunk length 11.27-13.48 ( $12.33 \pm 1.11$ ); trunk width 2.84-3.17 ( $3.03 \pm 0.17$ ).

Male. Unknown.

**Site.** The copepod attaches to various parts of the body surface of the host fishes (Fig. 1A). The head to the neck of the copepod was embedded in the host's musculature, and the trunk was protruded into the water (Fig. 1B)



**Figure 2.** *Sarcotretes umitakae* sp. n., female, holotype NSMT–Cr 22253. **A** habitus **B** anterior portion of body, dorsal, a1 = antennule, a2 = antenna **C** same, ventral, m1 = maxillule, m2 = maxilla, p1 = leg 1, p2 = leg 2, p3 = leg 3, p4 = vestige of leg 4 **D** vestige of dorsal cephalothoracic shield **E** tip of proboscis, lateral **F** posterior portion of body, ventral **G** same, lateral **H** rostral area and antennae, dorsal. Scale bars: **A**=3 mm; **B**, **C**, **F**, **G**=1 mm; **D**=500  $\mu$ m; **E**=300  $\mu$ m; **H**=150  $\mu$ m.



**Figure 3.** *Sarcotretes umitakae* sp. n., female, holotype NSMT–Cr 22253. **A** left antennule, anterior **B** left antenna, anterior **C** left mandible **D** left maxillule **E** left maxilla, lateral **F** distal part of left maxilla **G** right leg 1, anterior. Scale bars: **A**, **B**, **E**, **G**=100 μm; **C**, **D**=70 μm; **F**=50μm.



**Figure 4.** *Sarcotretes umitakae* sp. n., female, holotype NSMT–Cr 22253. **A** left leg 2, anterior **B** right leg 3, anterior **C** vestige of leg 4. *Sarcotretes umitakae* sp. n., female, paratype NSMT–Cr 22254 **D** habitus. Scale bars: **A**, **B**=100 μm; **C**=30 μm; **D**=3 mm.

**Etymology.** The specific name "*umitakae*" refers to the *Umitaka-maru*, a training and research vessel of TUMSAT.

Remarks. Currently, three species of Sacotretes: S. eristaliformis, S. longirostris, and S. scopeli, are considered to be valid (Ho et al. 2007). Sarcotretes umitakae sp. n. differs from S. eristaliformis and S. scopoli by having the holdfast with paired bulbous swellings drawn out into an elongate, horn-like process (vs. bulbous with or without a blunt tip) and the neck approximately 3 times as long as the trunk (vs. nearly as long as the trunk in S. eristaliformis and less than 3/4 of the trunk length in S. scopeli) (Brian 1912; Hogans 1988; Boxshall 1989; Cherel and Boxshall 2004; Ho et al. 2007). The two specimens described as S. eristaliformis by Kabata and Gusev (1966) were judged to be S. scopeli on the basis of a body length of 15.8–21.2 mm, the neck being shorter than the trunk, and relative length of the proboscis (Hogans 1988; Ho et al. 2007). In addition, S. scopeli differs from the new species by the absence of the vestige of leg 4 (vs. presence) (Boxshall 1989; Cherel and Boxshall 2004). Sarcotretes longirostris has the neck longer than the trunk like S. umitakae sp. n. and only these 2 species possess an extremely long proboscis among their congeners. Sarcotretes longirostris is, however, easily distinguishable from the new species by having the following characters: slender lateral head processes without swollen basal portions (vs. a bulbous swelling with a slender process); a not defined rostral area (vs. triangular); and a large and conical reduced abdomen, protruding to posterior (vs. small and non-conical and slightly protruded to posterodorsal) (Ho et al. 2007). In addition, the trunk has an anterior constriction in S. umitakae sp. n. not shared with any of the three known species (Figs. 2A, 4D).

### Discussion

Despite the fact that some morphological characters of Sarcotretes species (e.g. the shape of the holdfast, the length and flexure of the neck, and the length of the proboscis) show variability, they have been conventionally used to distinguish the species in this genus (Hogans 1988). Because these characters vary according to the site of attachment to the host and the age of the parasite, species identification using such characters may not make sense. Actually, based on those characters, Wilson (1917) had recognized 6 species as valid, but later, 4 species (S. gempyli, S. inflexus, S. lobatus, and S. nodicornis) were regarded as junior synonyms of S. scopeli by Hogans (1988). Nonetheless, we consider that such characters as the length of the proboscis and the shape of the lateral horns on the head, which were used in the key given by Wilson (1917), are useful identification characters. These characters do exhibit variability but there is no overlap between in S. umitakae sp. n. and existing species. Sarcotretes umitakae sp. n. and S. longirostris possess a strongly projecting proboscis which is not shared with other congeners. The lateral process of S. umitakae sp. n. comprising a bulge with a pointed tip is similar to that of S. scopeli and S. eristaliformis, but the greatly elongated tip in S. umitakae sp. n. is apparently distinguishable (see Brian 1912, pls. 9, 10; Hogans 1988, fig. 2b; Boxshall 1989, fig. 3; present study, Fig. 2).

Although a great care is required, it is considered that these characters can provide reliable evidence of species identity. On the other hand, all three specimens of *S. umitakae* sp. n. have a neck with a posterior protrusion and a constriction behind it. However, this character cannot be used to identify *S. umitakae* sp. n. because a similar character was observed in some specimens of *S. eristaliformis* and *S. scopeli* (see Brian 1912; Boxshall 1989).

The discovery of *S. umitakae* sp. n. in this study shows that there are at least 2 species of the genus in Japanese waters.

### Key to females of the species of Sarcotretes

1	Proboscis slightly projecting; holdfast composed of broad base with or
	without terminal process; neck shorter than or as long as trunk2
_	Proboscis elongate, strongly projecting; holdfast comprising slender processes;
	neck significantly longer than trunk
2	Body up to approximately 25 mm long; neck shorter than trunk; leg 4
	absent
_	Body approximately 45 mm or longer (about twice length of <i>S. scopeli</i> ); neck
	about as long as trunk; vestige of leg 4 (intercoxal sclerite) present
	S. eristaliformis
3	Slender, horn-like lateral processes on head (holdfast); trunk not constricted;
	reduced abdomen conical, projecting posteriorly
_	Lateral process bulbous tapering into slender, elongate horn; trunk with
	anterior constriction; abdomen reduced, vestigialS. umitakae sp. n.

### Acknowledgements

We would like to acknowledge the captain and crew of the *Umitaka-maru* (TUMSAT) for their support during the cruise. We also thank Dr. Tadashi Tokai (TUMSAT), an organizer of the cruise, for his courtesy and understanding of our study on the parasitic copepod fauna of the East China Sea. We are grateful to Dr. Yuji Tanaka (TUMSAT) for his assistance in collecting the specimens onboard the vessel. Part of this work received a financial support from Grants-in-Aid for JSPS Fellows (No. 23-4311 to D.U.).

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RESEARCH ARTICLE



# Morphology of the first-instar nymph and adult female of Kermes echinatus Balachowsky, with a comparison to K. vermilio Planchon (Hemiptera, Coccoidea, Kermesidae)

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Academic editor: Mike Wilson | Received 2 August 2012 | Accepted 6 November 2012 | Published 29 November 2012

**Citation:** Spodek M, Ben-Dov Y (2012) Morphology of the first-instar nymph and adult female of *Kermes echinatus* Balachowsky, with a comparison to *K. vermilio* Planchon (Hemiptera, Coccoidea, Kermesidae). ZooKeys 246: 11–26. doi: 10.3897/zooKeys.246.3766

### Abstract

The first-instar nymph and the adult female of *Kermes echinatus* Balachowsky (Hemiptera, Coccoidea, Kermesidae) are described and illustrated. This species is compared with *K. vermilio* Planchon, a morphologically similar species known in the Palaeractic region.

### **Keywords**

Scale insect, Quercus, evergreen oaks, Kermesidae, morphology, red dye

## Introduction

The scale insect family Kermesidae (Hemiptera, Coccoidea) develops and feeds exclusively on Fagaceae trees (Ben-Dov et al. 2012). This scale insect family is composed of one hundred species distributed among ten genera and they are currently known from the Nearctic, Oriental and Palaearctic regions of the world. *Kermes* Boitard is the principal genus in the European and Mediterranean regions. In these regions, twenty species of *Kermes* have been recorded, all off deciduous and evergreen oak trees (*Quercus*) (Ben-Dov et al. 2012). Individual insects develop mainly in bark crevices and on small twigs and branches (Sternlicht 1969, Bullington and Kosztarab 1985, Hu 1986, Podsiadlo 2005a). Most Kermesidae are not known to cause any visible injury to their host trees. There are however some reports of heavy infestations that can lead to branch dieback, flagging, reduced growth rates and occasionally tree death (Kozár 1974, Hamon 1977, Solomon et al. 1980, Viggiani 1991, Pellizzari et al. 2012).

Seven Kermesidae species, belonging to two genera, *Kermes* Boitard and *Nidularia* Targioni Tozzetti, have been described or recorded from Israel off *Quercus* sp. (Ben-Dov et al. 2012). Two species: *K. greeni* and *K. nahalali*, were described from the post-reproductive adult female (Bodenheimer 1931); three species: *K. echinatus, K. palestiniensis* and *K. spatulatus*, were described from first-instar nymphs (Balachowsky 1953); one species, *K. bytinskii*, was described from the adult female and all nymphal instars by Sternlicht (1969). The adult female of *Nidularia balachowskii* was described from Turkey (Bodenheimer 1941) and later was recovered in Israel (Bodenheimer 1944).

*Kermes echinatus* Balachowsky is one of six *Kermes* species found in Israel (Ben-Dov et al. 2012). To date, this species has only been reported off the evergreen oak, *Quercus coccifera* L., from one location in the Lower Galilee in Israel.

Some scale insects have been known as sources of red dye used in the textile, art and wine industries in the Mediterranean, Middle East and Central Asia regions (Leonardi 1920, Donkin 1977, Sarkisov 1984, Cardon 2007) for many centuries. Scholars have suggested that the red dye used for both secular and ritual purposes in Israel during ancient times apparently had been imported from neighboring countries where such dye producing scale insects as *Porphyrophora hameli* Brandt and *Kermes vermilio* Planchon are known (Sandberg 1997). However, Amar et al. (2005) extracted red dye from both the adult females and eggs of *K. echinatus* and chemically analyzed it and suggested that *K. echinatus* might be the "Tolaat Shani" (scarlet worm in Hebrew), an animal mentioned in the bible used for dye extraction during the period of the second Holy Temple Period (70 A.D.) in Israel.

Balachowsky (1953) described and illustrated the morphology of the first-instar nymph of *K. echinatus* from specimens that were collected off *Q. coccifera* from Nahalal forest located in the Lower Galilee in Israel. He also provided a brief description of the adult female, which included its color, shape and body dimensions. He compared the morphology of *K. echinatus* with the first-instar nymph of *K. vermilio*, another Palaearctic *Kermes* species not present in Israel, and concluded that "both species share general topography and structure of characters... therefore *K. echinatus* is the eastern neighbor of *K. vermilio*".

The first-instar of both *K. echinatus* and *K. vermilio* are easily distinguishable from other Mediterranean and European *Kermes* species due to the presence of conical, spine-like marginal setae (Balachowsky 1953, Pellizzari et al. 2012). The first-instars of other Palaearctic *Kermes* species possess hair-like, spatulate or club-shaped marginal se-

tae (Kuwana 1931, Balachowsky 1950, Sternlicht 1969, Hu 1986, Liu and Shi 1995, Podsiadlo 2005b). To date there has been no detailed taxonomic description of the adult female of *K. echinatus*. This study presents a description of the adult female and a redescription of the first-instar nymph of *K. echinatus*. We also compare the general appearance and morphology of both stages with *K. vermilio*.

# Materials and methods

## Specimen collections

Between 2010 and 2012, we collected specimens of *K. echinatus* off the evergreen oak, *Q. calliprinos* Webb, from forests in the Golan Heights, the Western, Upper and Lower Galilee regions and the Judean Mountains in Israel. The collection site at Timrat in the Lower Galilee is three km from Nahalal, the type locality of *K. echinatus*, and therefore we consider these specimens to be topotypic material. Some of the first-instar nymphs examined in this study emerged from females that were kept in sealed glass containers in the laboratory and other specimens were recovered from thin branches or from trunks of trees.

## Identification and morphological observations

Specimens were processed and mounted on microscope slides according to the methods outlined by Ben-Dov and Hodgson (1997). Illustrations of the adult female and the first-instar nymph of *K. echinatus* are generalizations of several specimens, showing the dorsum on the left and the venter on the right, with enlargements of important structures arranged around the central drawing. The structures are not drawn to the same scale between each other. Terms for morphological features follow, chiefly those of Bullington and Kosztarab (1985), Baer and Kosztarab (1985) and Hodgson (1994). Measurements of specimens and of morphological structures were made using an ocular micrometer on an Olympus BX51 phase contrast microscope. Measurements of structures are given in millimeters (mm) or microns ( $\mu$ m). Body length was measured from the farthest points of the head to the posterior end of the body and body width was the greatest width. Setal lengths were measured from the base of the seta to the apex, i.e. excluding the setal socket. Fresh topotypic specimens collected by us in Israel plus one syntypic first-instar nymph were used for the descriptions. The frequency of each structure is given for the entire body. The range is taken from twenty specimens.

Abbreviations of specimen depositories are as follows: **BMNH** - The British Museum (Natural History), London, U.K.; **ICVI** - Coccoidea Collection, Department of Entomology, Agricultural Research Organization, Bet Dagan, Israel; **MNHN** - Museum National d' Histoire Naturelle, Paris, France.

## Results

### Kermes echinatus Balachowsky

http://species-id.net/wiki/Kermes\_echinatus

Kermes echinatus Balachowsky, 1953: 181

**Note.** This species was originally described from the first-instar nymph collected from Israel, Nahalal forest, off *Q. coccifera*.

**Material examined. Adult female of** *K. echinatus.* Israel: All material was collected off *Q. calliprinos* by M. Spodek. At least twenty specimens were examined and all material is deposited in ICVI. Alonei Abba Reserve, 19.vi.2011, 26.vi.2011, 3.vi.2012 (MC:530, C:4999), MC:711); Eilon, 19.vi.2011, 22.vi.2011, 26.vi.2011, 3.vi.2012 (MC:533, MC:542, C:4998), MC:692); Nahal Dolev Reserve, 27.vi.2010, 17.vi.2011, 8.vi.2012, 15.vi.2012, 22.vi.2012 (MC:261, MC:699, MC:528, MC:695, MC:709); Hanita, 6.vi.2010 (MC:227); Mas'ada, 4.vii.2010 (MC:285); Nebi Hazuri, 4.vii.2010, 6.vii.2011 (MC:288, MC:556).

**First-instar nymph of** *K. echinatus.* Israel: Syntype (ICVI C:3691, MNHN 1065-8), Nahalal Forest, *Q. coccifera* 10.v.1950, Bytinski-Salz; All non-type material was collected off *Q. calliprinos* by M. Spodek, unless otherwise stated; at least twenty specimens were examined and all material is deposited in ICVI. Alonei Abba Reserve, 15.vii.2010, 26.vi.2011, 15.vii.2012 (MC:289, MC:559, MC:719); Eilon, 26.ix.2010, 21.iv.2011, 17.vi.2011, 1.vii.2011, 22.vii.2012 (MC:306, MC:486, MC:499, MC:550, MC:718); Hanita, 13.iii.2011 (MC:457); Nahal Dolev, 22.viii.2010, 8.viii.2011, 1.vii.2012 (MC:293, MC:562, MC:717); Nebi Hazuri, 17.viii.2000, Y. Ben-Dov (C:3409), 4.vii.2010, 6.vii.2011, 17.vii.2011, (C:4818, C:5003, MC:561); Neve Zuf, 10.vii.2000, 18.vii.2003, Y. Ben-Dov (C:4752, C:4751); Timrat, 21.vii.2011, 25.iii.2012 (MC:563, MC:651).

**Comparative material examined. Adult female of** *K. vermilio.* France: Corsica, *Q. ilex*, 7.vi.1999, J. Casevitz-Weal (2 specimens, ICVI C:3277); Le Vert Lasalle, *Q. coccifera*, 7.v.2007, D. Cardon (3 specimens, ICVI C:4257); Italy: Portofino, *Q.* ilex, 27.v.1971, D. Matile-Ferrero (2 specimens, MNHN 4594-2), Pistoia, *Q. ilex*, 13.viii.1986, A. Belcari (2 specimens, MNHN 10732-1), Bitonto (Bari), *Q. ilex*, 25.vii.2012, F. Porcelli (10 specimens, ICVI C-5132); Spain: Mieras (Gerona), *Q. coccifera*, 7.v.1987, A. Verhecken (1 specimen, MNHN 11526-1).

First-instar nymph of *K. vermilio*. France: Le Vert Lasalle, *Q. coccifera*, 24.vi.2007, D. Cardon (47 specimens, ICVI C:4272); Italy: Pistoia, *Q. ilex*, 13.viii.1986, A. Belcari (6 specimens, MNHN 10732-3), Bitonto (Bari), *Q. ilex*, 28.viii.2012, F. Porcelli (20 specimens, ICVI C-5133); Spain: Mieras (Gerona), *Q. coccifera*, 7.v.1987, A. Verhecken (1 specimen, MNHN 11526-2).



Figure 1. Kermes echinatus Balachowsky young adult female, general appearance.

**Description adult female. General appearance.** Young, pre-reproductive adult: Oval, soft and slightly convex; dorsum brownish-grey with 4 or 5 black longitudinal and 6–9 black transverse lines formed of dots and lines; 2.5–3.2 mm long and 2–3 mm wide (Fig. 1). Fully-mature reproductive female highly convex; dorsum brownish-grey with black, longitudinal and transverse lines; body tapering posteriorly (Figs 2, 3). Post-reproductive female oval and moderately convex, 2.9–4.4 mm long, 2.7–5.1 mm wide and 3.2–4.8 mm high; dorsum sclerotized; red with 6–9 black, transverse black lines represented as reticulated folds (Fig. 4).

Slide-mounted adult female. 2–3 mm long and 2–2.8 mm wide (Fig. 5).

**Margin. Marginal setae** conical,  $12-13 \mu m$  long and  $10-11 \mu m$  wide at base; arranged in a single row of 30-38 setae on each margin.

**Dorsum. Dorsal setae** hair-like, 7–9  $\mu$ m long, in submarginal band from cephalic tip of body to posterior end of body, about 28–33 on each side. **Conical setae**, similar in shape to marginal setae, randomly placed on dorsum, with 7–11 setae on each side; each seta 10–13  $\mu$ m long and 7–10 wide at base. **Bilocular pores** oval with a sclerotized rim, each 3  $\mu$ m long and 2  $\mu$ m wide; present throughout. **Tubular ducts** diffused throughout dorsum; each with outer ductule 12–17  $\mu$ m long, inner ductule 10–15  $\mu$ m long and with a sclerotized cup 5  $\mu$ m diameter.



Figure 2. Kermes echinatus Balachowsky mature reproductive female, general appearance.

Venter. Eyes circular, 20–25 µm diameter, each placed anterolaterally to each antenna. Legs absent. Antenna each 1-segmented; 26–35 µm long, 20–31 µm wide; each bearing 5–8 fleshy setae; each antennae is surrounded by a group of 40-45 multilocular pores; each pore 7-8 µm diameter with 10 loculi. Clypeolabral shield 235-250 µm long, 212–225 µm wide. Labium 3-segmented, triangular, 160–175µm long, 110– 135  $\mu$ m wide; labial setae as follow: basal segment with 2 setae, 5–8  $\mu$ m long; medial segment with 2 setae, 12-20 µm long; apical segment with 4 setae; 6 apical setae, 10-12 µm long and 2 subapical seta, 7-8 µm long. Mesothoracic and metathoracic spiracles subequal in size; peritreme 50–68  $\mu$ m long and 30–37  $\mu$ m wide; pores with 10 loculi and 8  $\mu$ m wide in a group of 15–22 locular pores laterad to each spiracle; also with 2 pores with 6 loculi, each 6 µm diameter, laterad to each anterior peritreme. Tu**bular ducts** present in a complete, dense submarginal band about 11 ducts wide and also sparsely throughout rest of venter; each duct with outer ductule 10-16 µm long; inner end of outer ductule with a sclerotized cup, 4-5 µm diameter, and inner ductule 11–15 µm long. Multilocular pores each diameter 10 µm with 10–12 loculi, arranged in 2-3 transverse rows on each abdominal segment; with a total of 114-120 pores on each segment; also with a group of 52-56 pores just posterior to vulva. Bilocular pores each 3 µm long and 2 µm wide, interspersed between tubular ducts in submar-



Figure 3. Kermes echinatus Balachowsky gravid females on tree trunk, general appearance.

ginal band. **Simple pores** 2  $\mu$ m diameter with a sclerotized rim, interspersed between multilocular pores on abdomen. **Ventral setae** 7–12  $\mu$ m long, distributed as follows: about 12 setae just anterior to clypeus between antennae; about 8 setae on median and submedian areas of thorax; about 11 setae mesad to each submarginal band of tubular ducts, in a line from antennae to anal ring; 6 or 8 setae, present in a band along each abdominal segment; plus 2 setae 20–25  $\mu$ m long, placed medially on each abdominal segment. **Microspines** each 1–2  $\mu$ m long, in groups of 3–5, in 3–8 rows on each abdominal segment. **Anal ring** ventral, forming a complete sclerotized circle; diameter 42–60  $\mu$ m; cells absent; with 6 setae, each 25–40  $\mu$ m long. **Other ventral setae** 1 pair of setae, each 10–12  $\mu$ m long, present just anterior to anal ring; 2 pairs of setae, each 10–12  $\mu$ m long, present just anterior to anal ring; 2 pairs of setae, each 10–12  $\mu$ m long, present posteriorly to anal ring; 1 pair of stout conical setae (similar in shape to marginal spinose setae but shorter), each 10–12  $\mu$ m long and wide, present on venter slightly above posterior margin; and 1 pair of apical setae, each 33–35  $\mu$ m long.



Figure 4. Kermes echinatus Balachowsky female with emerging first-instar nymphs.

**First-instar nymph. General appearance.** Dorsum and venter red, body oval and tapering posteriorly, 0.37–0.44 mm long and 0.14–0.2 mm wide. Each with a fringe of curly white wax on margins once first-instars settle on branch for feeding (Fig. 6).

Mounted specimen. Oval, 0.45–0.49 mm long and 0.20–0.25 mm wide (Fig. 7).

**Margin. Marginal setae** conical and slightly curved apically, each  $10-13 \mu m \log and 5 \mu m$  wide at base, in a complete line of 17-22 on each side, smaller conical setae, not-curved, each  $5-8 \mu m \log and 3-5 \mu m$  wide at base, in a submarginal row of 12-14 setae extending from mesothorax to anal lobe.

**Dorsum.** Derm membranous; intersegmental lines observable. **Dorsal setae** 8, each 6–8 µm long, placed in 2 submedian, longitudinal rows on thorax. **Simple pores** circular, 14, each about 1 µm diameter, placed in 2 submarginal, longitudinal rows on abdomen.

**Venter. Antennae** each 6-segmented; total length 102–110  $\mu$ m; with segment III and VI longest; setal distribution as follows: scape and pedicel each with 2 thin, hair-like setae; segment III with 1 long thin, hair-like seta; IV with 1 fleshy setae; V with 1 fleshy seta and 2 hair-like setae; apical segment with 2 fleshy setae and 5 hair-like setae. **Legs** well-developed; measurements of hind legs (length in  $\mu$ m); coxae 25–30, trochanter + femur 68–70, tibia 33–38, tarsus 45–50, claw 15–20; total leg length 187–200  $\mu$ m; trochanter with 2 oval, sensory pores on each side, each pore 3  $\mu$ m long and about 2  $\mu$ m wide; setae present on all leg segments; tarsal digitules each 25–30  $\mu$ m long and knobbed apically, extending beyond apex of claw; claw digitules



Figure 5. Kermes echinatus Balachowsky adult female.

knobbed apically, each 15–20  $\mu$ m long; each claw with a single denticle near tip. **Clypeolabral shield** well-developed; 68–75  $\mu$ m long and 63–75  $\mu$ m wide. **Labium** 3-segmented, triangular, 75–83  $\mu$ m long and 45–47  $\mu$ m wide; labial setae as follows: basal segment with 1 setae, rarely 2 setae, 5–8  $\mu$ m long, median segment with 2 hair-like setae on dorsal surface, 12–13  $\mu$ m long, apical segment with 6 subapical setae, each 16–20  $\mu$ m long and 2 apical setae 10–12  $\mu$ m long. **Spiracles** subequal in size; each peritreme 3–5  $\mu$ m diameter; apodeme crescent shaped, 13–15  $\mu$ m long;



Figure 6. Kermes echinatus Balachowsky first-instar nymph, general appearance.

each spiracle with 1 quinquelocular pore, 5  $\mu$ m diameter, placed anterolaterally. **Trilocular pores**, each about 3  $\mu$ m wide, distributed as follows: 2 pores between scape just anterior to clypeus; 1 mesad to each coxa, and 2 submedially on abdominal segments V–VII. **Bilocular pores** oval, 4 total, each 3  $\mu$ m long and 2  $\mu$ m wide, present between margin and each spiracle. **Ventral setae** interantennal setae 6, each 38–45  $\mu$ m long, in an longitudinal line medially between scapes; also 2 conical setae, about 14–16  $\mu$ m long and 5  $\mu$ m wide at base, on anterior apex of head; 1 seta 10–11  $\mu$ m long, mesad to each coxa associated with each trilocular pore; and 6 longitudinal lines of setae on abdomen; with 2 medial, 2 submedial and 2 submarginal setae per segment; medial and submedial setae each 10–15  $\mu$ m long, and submarginal setae 5–6  $\mu$ m long. **Microspines** each about 3  $\mu$ m long, arranged in groups of 3 or 4 in 2 transverse rows on each abdominal segment and sparsely on thorax. **Anal ring** located ventrally; composed of 2 semi-circles; diameter 20–25  $\mu$ m; each half circle with 4–6 cells and 3 pointed setae, each 13–18  $\mu$ m long. **Other setae** with a pair of



Figure 7. *Kermes echinatus* Balachowsky first-instar nymph.

setae, each 15–18  $\mu$ m long, anterior to anal ring, and a pair, each 15–20  $\mu$ m long, latero-posteriorly to anal ring. **Anal lobes** slightly developed; inner margin of each lobe with 1 pointed seta, 10–13  $\mu$ m long and 2–3  $\mu$ m wide, and 1 very long, flagellate seta apically 220–275  $\mu$ m long.

## Discussion

Prior to this study, *K. echinatus* had only been reported off the evergreen oak, *Q. coc-cifera*, from Nahalal forest, located in the Lower Galilee of Israel (Balachowsky 1953). During the 2010 to 2012 surveys of Kermesidae throughout the country, specimens were recovered off *Q. calliprinos* trees in the Golan Heights, Western, Upper and Lower Galilee regions, as well as the Judean Mountains. It is widely accepted by botanists that *Q. calliprinos* is probably an East Mediterranean subspecies of, or a vicariad species to, *Q. coccifera*, which is distributed in the Mediterranean territories of Europe (Zohary 1973, Jalas and Suominen 1976).

The present description of the first-instar nymph of *K. echinatus* agrees well with that of Balachowsky (1953). However our redescription includes several features that were not indicated in the original description but were observed by in us in fresh and type material. These features are the presence of: (i) dorsal submedial setae; (ii) ventral bilocular pores; (iii) a claw denticle on each leg; (iv) and microspines dispersed on thoracic and abdominal regions.

Balachowsky (1953) observed two main differences between the morphology of the first-instar nymph of *K. echinatus* and *K. vermilio*: (i) dorsal simple pores present on *K. echinatus* but absent on *K. vermilio* and (ii) the structure and arrangement of the marginal conical setae. In *K. echinatus*, the conical marginal setae are slightly longer and curved compared to those of *K. vermilio*. Balachowsky considered that the marginal setae of *K. vermilio* were of one length and arranged in two rows while those of *K. echinatus* were arranged in one row, but we found that first-instar *K. echinatus* also had two rows of conical marginal setae but that they differed in size and shape.

In addition, we observed other distinguishing features between the two species and these are summarized in Table 1. Pellizzari et al. (2012) added that the living specimens of *K. vermilio* are orange-red with yellow legs, whereas we noted that the first-instar nymphs of *K. echinatus* are red. We also noted a small denticle on each claw of *K. echinatus*. These were considered to be absent on *K. vermilio* by Balachowsky (1950) and Pellizzari et al. (2012).

The general appearance of young females and fully-grown reproductive females of *K. echinatus* differs from that of *K. vermilio*. The young female of *K. echinatus* is slightly convex, has a brownish-grey dorsum with 4 or 5 black longitudinal and 6–9 black transverse lines composed of dots and lines. The young female of *K. vermilio* is reddish without transverse and longitudinal lines. The fully-grown reproductive female of *K. vermilio* has been described as dark red or brown covered with a fine, white or pale grey

mealy wax (Pellizzari et al. 2012). In contrast, the fully-grown female of *K. echinatus* is not covered in wax and has transverse and longitudinal black lines on its dorsum. Both species at this stage are highly convex and subspherical.

The morphological features of the adult female of *K. echinatus* and *K. vermilio* are similar and are summarized in Table 2. Some of the shared features are the following; (i) dorsal and marginal conical setae; (ii) absence of legs; (iii) presence of numerous multilocular pores on abdominal segments as well as surrounding the antennae and spiracles; (iv) one-segmented antennae with fleshy setae; and (v) the anal ring located ventrally in both species. The most distinguishing feature of *K. echinatus* is the anal ring which has six setae and no cells whereas the anal ring of *K. vermilio* has cells but no setae. Some other differences between the two species are that *K. echinatus* has less conical setae on its margins and dorsum compared to 73–133 in *K. vermilio*. *Kermes echinatus* has 7–11 dorsal setae compared to about 70 dorsal setae in *K. vermilio*. Ventral loculate pores are only found on the abdominal segments and surrounding the spiracles in *K. echinatus* in contrast to *K. vermilio*, where they extend onto the metathorax from the abdomen.

Character	K. echinatus	K. vermilio
Dorsal simple pores	present	absent
Dorsal bilocular pores	absent	present
Locular pores associated with prothoracic spiracles	1 pore, 5 loculi	1 pore, 5 loculi and 7 loculi
Arrangement of marginal setae	2 rows	2 rows
Type of conical marginal setae	2 types	1 type
Denticle on claw of legs	present	absent

Table I. Comparison of some characters of the first-instar nymph of K. echinatus and K. vermilio.

Table 2. Comparison of some characters of the adult females of K. echinatus and K. vermilio.

Character	K. echinatus	K. vermilio
Marginal and dorsal conical setae	present	present
Hair-like setae in submarginal band on dorsum	present	absent
Conical setae in submarginal band on venter	absent	present
Legs	absent	absent
Position of anal ring	ventral	ventral
6 -locular pores associated with prothoracic spiracles	2 pores present	absent
Setae on anal ring	present	absent
Cells on anal ring	absent	present
Simple pores on abdomen	present	absent
Multilocular pores posterior to vulva	present	absent

## Conclusion

This paper describes the adult female of *K. echinatus* for the first time and redescribes the first-instar nymph. The general appearance and morphological features of *K. echinatus* and *K. vermilio*, two species that have been linked to sources of red dye in the Palaerarctic region, are compared. Distinguishing characters of the first-instar nymph and female of *K. echinatus* are presented. *K. echinatus* has only been recorded in Israel to-date and is one of seven species of Kermesidae occurring there.

## Acknowledgements

This study was partly funded to the senior author by The Karen Kayemeth LeIsrael (Project # 131-1621-11) and the Israel Taxonomy Initiative. This manuscript is part of the PhD thesis for the senior author. We thank Daniele Matile-Ferrero, Imre Foldi, (MNHN), Jon Martin (BMNH) and Francesco Porcelli (Department of Entomology, University of Bari, Italy) who helped us obtain specimens and slide-mounted material for this study. We would also like to thank Zvi Mendel and Murad Ghanim (Volcani Center, Bet Dagan, Israel) for their support throughout this project. Special gratitude is expressed to Alex Protasov for his photographic skills and technical support (Volcani Center, Bet Dagan, Israel). Collection permits at Nature Reserves in Israel, were kindly provided by the Israel Nature and Parks Authority.

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RESEARCH ARTICLE



# A new species of *Lesticus* Dejean, 1828 (Coleoptera, Carabidae) from the Finisterre Range, Papua New Guinea and a key to the genera of pterostichine-like Harpalinae of New Guinea

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Academic editor: T. Erwin | Received 12 October 2012 | Accepted 20 November 2012 | Published 29 November 2012

urn:lsid:zoobank.org:pub:6F65719D-D084-4B92-9A27-4180D1BC8E9C

**Citation:** Will K, Kavanaugh DH (2012) A new species of *Lesticus* Dejean, 1828 (Coleoptera, Carabidae) from the Finisterre Range, Papua New Guinea and a key to the genera of pterostichine-like Harpalinae of New Guinea. ZooKeys 246: 27–37. doi: 10.3897/zookeys.246.4112

### Abstract

*Lesticus finisterrae* (Carabidae: Pterostichini) **sp. n.** (type locality: Finisterre Range, Papua New Guinea), is described and characters to differentiate it from other "Trigonotomi" species are given. A key to the genera of pterostichine-like Harpalinae of the island, including all genera of Morionini, Cratocerini, Drimostomatini, Abacetini, Loxandrini and Pterostichini, is provided. The genus *Rhytisternus* (Pterostichini) is for the first time reported from New Guinea, represented by the likely adventive species *Rhytisternus laevis* (Macleay). The previously unknown male of *Stegazopteryx ivimkaensis* Will (Drimostomatini) is described.

## Keywords

New Guinea, Carabidae

## Introduction

Darlington reported 667 species of Carabidea from New Guinea in his treatment of the fauna (1971) and a search of the Zoological Record for new species and new records

suggests that the total is now easily greater than 700 species. Given the complexity of island's geological history (Baldwin et al. 2012) and the large areas that remain unsampled for beetles, the true diversity is probably at least double what is now known and some have estimated it could be as high as 2100 species (http://www.papua-insects.nl/insect%20orders/Coleoptera/Carabidae/Carabidae.htm)

One area poorly covered by Darlington's study was the Finisterre Range along the northeastern coast of Papua New Guinea, extending approximately from Madang in the north to Lae in the south. This area was previously recognized as a unique area of endemism (Liebherr 2008). During March and April of 1989, one of us [DHK] had the opportunity to visit this remote and physiographically isolated area and briefly sample the carabid beetle fauna near the highest part of the Finisterre Range, west of the village of Teptep. A challenging three-day uphill trek from Teptep brought our party, which also included George E. Ball of the University of Alberta, Edmonton, and Norman D. Penny of the California Academy of Sciences, to some of the highest forest in the region and to the ecotone between this upper montane moss forest and the grassland that replaces it at higher elevations (Fig. 1–2).

The forest canopy was between 10 and 20 meters tall, dense, with a lush coating of mosses and other epiphytes on tree trunks, branches, logs and stones on the forest floor. A low and fairly dense understory obscured most of the forest floor and made collecting difficult. Consequently, a few pitfall traps were installed in this cool and relatively dark habitat (Fig. 1). After two nights in the ground, the traps were collected and their contents examined. Only a single carabid beetle was found in the catch and the specimen was distinctly different from anything that has been described. Subsequent comparative microscopic study and dissection have confirmed this conclusion.

The genus *Lesticus* Dejean in New Guinea was treated by Darlington (1962, 1971) and again more recently by Dubault et al. (2008). The informal name "Trigonotomi" sensu Dubault et al. (2008) is used here as a working hypothesis for a presumably related group of species that includes members of *Lesticus, Trigonotoma* Dejean, *Euryaptus* Bates, *Pareuryaptus* Dubault, Lassalle & Roux and *Nesites* Andrewes. This complex of genera includes species that extend its range from northern Australia to Asia. Herein we describe this new species, provide a key to all of the pterostichine-like Harpalinae genera that have typically been associated with Pterostichini by Darlington and past authors that followed him. There is no evidence to support the monophyly of a group including all the taxa represented in the key, but their superficial similarity makes it convenient to treat them together for the purpose of identification until a satisfactory revision of the classification of Harpalinae is reached. Additionally, we add a new record for *Rhytisternus* Chaudoir and information on the male of *Stegazopteryx* Will.



**Figure 1.** View within upper montane moss forest habitat at 3050m elevation, Finisterre Range, Papua New Guinea. The pitfall trap in which the unique holotype of *Lesticus finisterrae* sp. n. was collected was located in the shaded area just below the middle of the figure.



**Figure 2.** Grassland/upper montane moss forest ecotone at 3050m elevation in the Finisterre Range, Papua New Guinea. The holotype of *Lesticus finisterrae* sp. n. was collected in a pitfall trap placed approximately 20m inside the forest edge.

# Methods

Methods and terms follow Will (2011).

## Specimens

Comparative material and specimens studied to develop the key are deposited in the following institutional collections: Australian National Insect Collection (ANIC), Canberra, ACT; California Academy of Science (CASC), San Francisco, CA; Essig Museum of Entomology (EMEC), Berkeley, CA; Institute Royal des Sciences Naturelles de Belgique, (IRSNB), Bruxelles; Museum of Comparative Zoology (MCZ), Harvard University, Cambridge, MA; The Natural History Museum, London (NHM); Nationaal Natuurhistorische Museum (NNM), Leiden, Netherlands; Muséum National d'Histoire Naturelle (MNHN), Paris; Queensland Museum (QM), Brisbane; Bohart Museum of Entomology (UCDC), Davis, CA; and Zoologische Staatssammlung (ZMS), München.

# Taxonomy

## Lesticus finisterrae Will & Kavanaugh, sp. n.

urn:lsid:zoobank.org:act:52A9C3D7-A304-4D05-83DC-D1E385182B6F http://species-id.net/wiki/Lesticus\_finisterrae

Holotype. Male. "PAPUA NEW GUINEA, Madang/Morobe Province border, Finisterre Range, Teptep area, 3.5 air km WNW of Kewieng No.4 village, 3050 m, 28 March 1989 stop #89-50// D. H. Kavanaugh, G. E. Ball & N. D. Penny collectors, Cal. Acad. Sci. Specimen//Papua New Guinea Expedition – 1989// California Academy of Sciences Type No. 18684//HOLOTYPE *Lesticus finisterrae* Will & Kavanaugh [red label]". Deposited CASC.

Type locality. Finisterre Range, Papua New Guinea. 5.99778°S, 52280°W.

**Diagnosis.** This species shares with other *Lesticus* species sharply hooked mandibular apices, relatively broad mentum, extremely wide gula (nearly the width of the mentum) and has the general form of a modified species of the *L. chloronotus* group. It is distinguished from all other species of *Lesticus*, including the New Guinea species covered in Darlington's keys and descriptions (1962, 1971) and Dubault et al.'s key (2008), by the combination of three or more setae in elytral intervals 3 and 5, the absence of transverse sulci of abdominal ventrites 4–5 and the presence of extremely prominent eyes.

Description. (Fig. 3), Size. Overall length (sbl) 20.0mm, greatest width over elytra 8.5mm. Color. Dorsal and ventral surfaces black to brunneous. Legs, mouthparts, and antennae slightly paler, lateral margins of pronotum and lateral and sutural areas of elytra margins piceous to rufopiceous. Luster. Dorsally and ventrally shiny. Iridescence. Elytra and ventral surface of body without spectral iridescence. Head. Dorsal microsculpture absent, entire surface with micropunctulae, clypeal-ocular sulci impression absent, shallowly rugose above eyes, with broad, shallow paramedial depressions, ocular ratio (greatest width over eyes/width between eyes at level of anterior supraorbital setae) 1.29, eyes moderately large size, very prominently "bulging". Labrum with anterior margin slightly emarginate, with six setae of which medial four setae equally distributed in medial half, distance from outermost medial seta to lateral seta about twice distance between medial setae. Antenna overall length moderately long, antennomere 11 just reaching beyond pronotal base. Thorax. Pronotum transverse, lateral margin sinuate near base, medial and basal setae touching lateral marginal bead; basal impressions obsolete; anterior angles scarcely produced; microsculpture not visible at 50× magnification; entire surface covered with minute punctulae. Elytral striae extremely shallow, scarcely impressed or absent; base of elytra not margined; humeri prominent but rounded; parascutellar punctures present at base of striae 1; interval 1 with two punctures at apex; interval 3 with three (right) or six (left) punctures; interval 5 with three (right) or four (left) punctures; interval 7 with one puncture near apex; interval 9



Figure 3. Dorsal habitus of Holotype of *Lesticus finisterrae*.



**Figure 4.** Male genitalia of holotype of *Lesticus finisterrae*. Median lobe. **A** left lateral **B** right lateral **E** ventral and **F** dorsal views. Parameres. **C** left **D** right. **ta** transverse apophysis.

with 20 evenly spaced punctures; elytral microsculpture visible at 50× as irregular isodiametric mesh of microlines; entire surface with micropunctulae. Male protaromeres 1–3 expanded with squamose setae ventrally. Tarsomere 5 on all legs ventral setose. Metatrochanter without setae. Metacoxa with single lateral seta. *Abdomen.* Abdominal ventrites smooth, glabrous except for very shallowly impressed transverse sulcus on last ventrite. Male with two setae on last ventrite. Aedeagus (Fig. 4) with small, dissimilar conchoid parameres; right paramere with large dorsal process (Fig. 4D); left paramere with a minute transverse apophysis (Fig. 4C). Median lobe with apex truncate; ostium dorsal; endophallus with small median sclerite.

**Etymology.** The specific epithet *finisterrae* refers to the type locality, the Finisterre Range. Additionally, Finisterre is a contraction that derives from the Latin *Finis Terrae*, meaning "End of the Earth", an appropriate metaphor for this remote and entomologically little-known region. Although *terrae* is the genitive form, the contraction is treated as a noun in apposition since the "end of the Earth" is used as the name for the location.

**Habitat.** Only a single specimen of *L. finisterrae* is known. It was collected in an unbaited pitfall trap placed in upper montane moss forest at an elevation of 3050 meters. The area was near the upper limit of forest growth and adjacent to open grassland (Fig. 2). Under the forest canopy, a fairly dense understory of low vegetation was present (Fig. 1).

**Identification and systematics.** "Trigonotomi" sensu Dubault et al. (2008) includes *Lesticus, Trigonotoma, Euryaptus* and *Pareuryaptus*. Although the group has not been rigorously tested for monophyly, it is reasonable to maintain its use as a working hypothesis based on similarity. Additionally, the genus *Nesites* Andrewes (cotype examined), which has the same combination of characters as *Trigonotoma*, should be included. It is likely that *Trigonotoma* and *Nesites* will prove to be synonymous. *Lesticus finisterrae* 

has the following states for characters given by Dubault et al. (2008): antennomere 1 less than combined length of antennomeres 2+3; setae at the anterior margin of the labrum more or less equally distributed; three or more setigerous punctures on interval 3 of the elytral disc. Females of *Lesticus* have four setae (two pairs) on the last ventrite, but the female for *L. finisterrae* is unknown. The parascutellar striae are generally present and long in *Lesticus* species; however in *L. finisterrae* all striae are very shallow or not impressed. There is no indication of the parascutellar striae. *Lesticus finisterrae* deviates from other species of *Lesticus* by the extremely prominent eyes, depressed form and lack of sulci on the abdominal ventrites. Based on general similarity and adjacencies of ranges, it seems probable that *L. finisterrae* is an isolated and derived member of the *L. chloronotus* group.

## Key to pterostichine-like Harpalinae

Below is a revised key to the genera of "pterostichites" of New Guinea based on the key by Darlington (1962:499–500). We have excluded *Mecyclothorax* Sharp (Moriomorphini, Liebherr 2011), which was included in Darlington's key, as it is not a member of the Harpalinae and only distantly related. It is easily distinguished from all the included taxa by the presence of a fixed seta in the mandibular scrobe.

1	Front tibia fossorial, outer apical angle strongly produced; bodyform parallel- sided: antennae moniliform
_	Front tibia with outer anical angle not produced: (other characters variable) 2
2	Small compact: antennae moniliform: elytron with basal pore (if present) at
2	base of 3rd stria
_	Size and form variable; antennae usually not moniliform; elytron with basal
	pore (if present) near or inside base of 2nd stria5
3	Elytron without basal pore; anteriolateral prothoracic setae almost on ante-
	rior angles
_	Elytron with basal pore at base of 3rd stria; anteriolateral prothoracic setae
	about 2/5 prothoracic length behind anterior angles
4	Elytra ovoid or elongate-ovoid, apex broadly rounded
_	Elytra elongate-rectangular, bluntly truncate at the apex
5	Antennomere 2 attached to 1 more eccentrically than usual; mentum trans-
	verse; metacoxal anterior sulcus sinuate; (small, 4.7–6.8mm, in New Guinea
	hydrophilic species)
_	Antennomere 2 attached to 1 less eccentrically; (other characters variable, but
	never in combination as above)6
6	Four basal antennomeres glabrous; size very large, length (in New Guinea)
	about 50mm or more

_	Three basal antennomeres glabrous; size much smaller7
7	Abdominal ventrites 4-6 with transversely impressed sulci or margined at
	base, at least toward sides
_	Abdomen with ventral segments not thus impressed or margined, or only
	ventrite 6 with a very shallowly impressed sulcus11
8	Elytron with 10th interval absent or not distinct from margin
	Prosopogmus Chaudoir [Pterostichini]
_	Elytron with a distinct 10th interval at least posteriorly9
9	Elytra with 3rd intervals impunctate; proepisterna longitudinally strigate 10
-	Elytra with 3rd intervals with setigerous punctures; parascutellar striae pre-
	sent (except when other striae obsolete); proepisterna not strigate (but often
	punctate) Lesticus (in part) [Pterostichini]
10	Metepisternum short, flight wing reduced
-	Metepisternum elongate, flight wing full (in New Guinea)
11	Small size, broad (prothoracic width/length c. 1.55-1.71), compact (super-
	ficially similar to Brachidius but with antennae less stout and basal pore of
	elytron present, at base stria 2)Cosmodiscus Sloane [Abacetini]
_	Size small to large, but never so broad and compact12
12	Elytra with 3rd interval punctate; parascutellar striae absent or nearly so 13
_	Elytra with 3rd interval impunctate; parascutellar striae variable17
13	Antennae subgeniculate, first antennomere moderately longer than 2 and 3
	together
_	Antennae not at all geniculate, first antennomere shorter than 2 and 3 to-
	gether14
14	Metepisterna scarcely longer than wide15
_	Metepisterna clearly longer than wide16
15	Elytra without plica. Mentum with prominent epilobes. Mentum tooth
	prominent, acuminate-entire Haploferonia Darlington [Loxandrini]
_	Elytra with plica. Mentum transverse, epilobes not prominent. Mentum
	tooth broad and emarginateLesticus (in part) [Pterostichini]
16	Prothorax not cordateLoxandrus Leconte [Loxandrini]
_	Prothorax cordate Nebrioferonia Straneo [Loxandrini]
17	Very small (4.0–4.5 mm.); parascutellar stria lacking
	<i>Tiferonia</i> Darlington [Abacetini]
-	Larger; parascutellar stria present18
18	Flight wings usually (not always) fully developed; body proportions normal,
	head not relatively very large Platycaelus Blanchard [Pterostichini]
_	Flight wings atrophied; head very large



**Figure 5.** Male genitalia of *Stegazopteryx ivimkaensis*. Median lobe. **A** left lateral **B** right lateral **C** ventral and **D** dorsal views.

### Additional records

#### Stegazopteryx ivimkaensis Will

A male specimen labeled "INDONESIA W-PAPUA 130km SE Kalmana, Omba (=Yamor) river 10–20km from coast, 4°05'49"S, 134°54'09"E, 10–20m, 09.–11. II.2011 leg. A. Skale (008)." Specimen deposited ZSM.

This specimen record extends the range of this species into eastern West-Papua. This specimen, the two female syntype specimens and the further records below, are all from elevations of 120m and below. This species is now known to range across nearly all of New Guinea. The examined male specimen is consistent with the description: of the genus based on females as given by Will (2004), with the following additions. sbl=5.4mm, greatest width of elytra 2.3mm. Secondary sexual characters- male protarsi not expanded but with spatulate setae ventrally on tarsomeres 1-3. Last ventrite with one pair of setae. Aedeagus right side up in repose. Right paramere larger and conchoid, left paramere small, peg-like. Median lobe of adeagus simple, no evident sclerotized structure on the endophallus (Fig. 5).

Additional locality records (not examined, Martin Baehr, in litt.). PAPUA NEW GUIN-EA Canopy Mission, Madang Province, Baiteta, Light, Leg. Olivier Missa: 1 male- AR 53, 30-V-1996; 1 male, 1 female- T 2, 31-V-1993; 1 female, X G, 24-IV-1996; 1 female, M 2, 30-IV-1996. One male and two females deposited IRSNB, one male and one female ZSM.
## Rhytisternus laevis (Macleay)

A female specimen labeled "New Guinea, Weam, Aug 1962// H. Olmus collector". Deposited ANIC. This species is distributed across northern Australia and it has not been previously reported from New Guinea. It is likely a recent accidental introduction or dispersal. We have only seen this single individual from New Guinea and it is unknown if the species is established.

## Acknowledgements

Fieldwork in Papua New Guinea in 1989 was generously supported by a ten-week fellowship provided by the Christensen Fund for work based at the Christensen Research Institute in Madang. We thank Martin Baehr for providing the records of *Stegazopteryx* and facilitating loans of other important specimens.

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RESEARCH ARTICLE



# A new species of Xorides Latreille (Hymenoptera, Ichneumonidae, Xoridinae) parasitizing Pterolophia alternata (Coleoptera, Cerambycidae) in Robinia pseudoacacia

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Academic editor: G. Broad | Received 30 August 2012 | Accepted 20 November 2012 | Published 29 November 2012

urn:lsid:zoobank.org:pub:6AF0D790-5EA6-46CF-A1E9-BF4E44C1A0F1

**Citation:** Sheng M-L, Zhao R-X, Sun S-P (2012) A new species of *Xorides* Latreille (Hymenoptera, Ichneumonidae, Xoridinae) parasitizing *Pterolophia alternata* (Coleoptera, Cerambycidae) in *Robinia pseudoacacia*. ZooKeys 246: 39–49. doi: 10.3897/zooKeys.246.3853

## Abstract

A new species is described, *Xorides benxicus* Sheng, **sp. n.**, reared from the cerambycid twig-boring pest of *Robinia pseudoacacia* Linnaeus, *Pterolophia alternata* Gressitt, 1938, in Benxi County, Liaoning Province, China. A key is given to the species similar to *Xorides benxicus* Sheng, namely *X. asiasius* Sheng & Hilszczański, 2009, *X. cinnabarius* Sheng & Hilszczański, 2009 and *X. sapporensis* (Uchida, 1928).

#### Keywords

Xorides, new species, key, parasitoid wasp, idiobiont, Pterolophia alternata, Cerambycidae, host plant, China

# Introduction

*Xorides* Latreille, 1809, belonging to the subfamily Xoridinae of Ichneumonidae (Hymenoptera), comprises 159 described species (Hilszczański and Plewa 2011, Yu et al. 2012). 43 species have been reported from China (Liu and Sheng 1998, Sheng et al.

1998, 1999, 2004, 2006, 2008, 2009, 2010, Zong and Sheng 2009). The hosts are larvae of wood-boring Coleoptera, mainly Cerambycidae and Buprestidae (Clément 1938, Hilszczański et al. 2005, Sheng et al. 2002, 2010, Yu et al. 1997). The status of the genus was defined by Townes (1969) and Wahl (1997).

In this article a new species of *Xorides* is described. The species was reared in Benxi County, Liaoning Province, at the southern border of the Eastern Palearctic part of China, as a parasitiod of *Pterolophia alternata* Gressitt, 1938 (Coleoptera: Cerambycidae), which bores twigs of *Robinia pseudoacacia* Linnaeus and is considered a pest.

The type locality is a forest composed of mixed deciduous angiosperms and evergreen conifers, mainly including *Robinia pseudoacacia*, *Castanea* spp., *Quercus* spp., *Larix* sp., *Rosa multiflora* var. *cathayensis* Rehd. & Wils., *Rubus* sp. and *Pinus tabulaeformis* Carr.

#### Materials and methods

Rearing parasitoids. Twigs of naturally heavily infested *R. pseudoacacia* trees were brought to the laboratory and maintained in a large nylon cage at room temperature. Water was sprayed over the trunks and twigs twice a week and emerged insects collected daily.

Rearing parasitoid larvae and pupae. Parasitoid larvae and cocoons were collected from galleries of wood-borers in infested twigs of *R. pseudoacacia* and stored individually in glass tubes with a piece of filter paper dipped in distilled water to maintain moisture and plugged tightly with absorbent cotton wool.

The host was identified by Professor Wen-Kai Wang, Changjiang University, Hubei Province, China.

Images of whole insects were taken using a CANON Power Shot A650 IS. Other images were taken using a Cool SNAP 3CCD attached to a Zeiss Discovery V8 Stereomicroscope and captured with QCapture Pro version 5.1.

The morphological terminology is mostly that of Gauld (1997). Wing vein nomenclature is based on Ross (1936) and the terminology on Mason (1986, 1990).

Type specimens and hosts are deposited in Insect Museum, General Station of Forest Pest Management, State Forestry Administration, P. R. China.

#### Xorides Latreille, 1809

http://species-id.net/wiki/Xorides

*Xorides* Latreille, 1809. Genera Crustaceorum et Insectorum secundum ordinem naturalem in familias disposita iconibus exemplisque plurimis explicata, 4:4. Type species: *Ichneumon indicatorius* Latreille.

**Diagnosis.** Apex of mandible chisel-shaped, unidentate. Subapical part of female flagella elbowed or bent, on the outer profile of the elbow or bend several peg-like bristles. Epomia present, usually strong, dorsally turning forward and sharply projecting. Front tibia usually thickened. Second tergum with an oblique basal groove on basolateral corner. Apical part of ovipositor cylindric or slightly depressed, the lower valve with several ridges.

#### Xorides benxicus Sheng, sp. n.

urn:lsid:zoobank.org:act:F9C857C8-B7D4-459C-A3F5-16B77FA18D26 http://species-id.net/wiki/Xorides\_benxicus Figures 1–12

Etymology. The name of the new species is based on the type locality.

**Types.** *Holotype*, Female, CHINA: Benxi County, Liaoning Province, 19 June 2012, leg. Mao-Ling Sheng. Paratypes: 3 females and 2 males, same data as holotype, except 18 to 19 June 2012.

**Diagnosis.** *Xorides benxicus* can be distinguished from the similar species of *Xorides*, possessing subapical terga with white apical spots in females, by the combination of the characters: head and mesosoma entirely black; face strongly convex centrally; apical part of lateral longitudinal carinae of area basalis combined; posterior part of second tergum with irregular longitudinal wrinkles; hind margins of terga 3 to 7 white, lateral parts of the white portions broken; last tergum with a smooth median longitudinal groove. Ovipositor sheath approximately 2.5 times as long as hind tibia. Flagella of male (Figure 5) slightly compressed, apex of each flagellomere swollen, lateral and ventral-lateral profiles with erect long setae, setae approximately 3.5 times as long as width of flagellomere and curved apically.

**Description. Female.** Body length 5.5 to 7.5 mm. Fore wing length 4.3 to 5.5 mm. Ovipositor sheath length 4.2 to 5.5 mm.

Head. Face (Figure 3) approximately 1.8 times as wide as long, strongly convex, with uneven, fine punctures; median portion shining, sparsely punctate; lower-lateral portion with indistinct oblique wrinkles; upper portion with median longitudinal groove; upper margin with strong median projection towards frons. Clypeal suture distinct. Clypeus with sub-basal transverse ridge; below ridge strongly inclined, weakly concave, with fine coriaceous texture. Mandible with fine median longitudinal groove; basal portion with fine longitudinal wrinkles; tooth shining. Subocular sulcus distinct. Malar space 0.6 to 0.7 times as long as basal width of mandible. Inner part of subocular sulcus shining with very sparse punctures, outer part with distinct oblique wrinkles and fine punctures. Gena in dorsal view approximately 0.7 times as long as width of eye; lower portion with longitudinal wrinkles, medially with dense punctures, upper portion smooth with very sparse and fine punctures. Vertex (Figure 4) smooth and shining, a few fine punctures. Interocellar area almost flat, with fine, dense punctures. Postocellar line about 1.7 times as long as ocularocellar line. Frons almost flat, with fine, dense, uneven punctures; median portion with fine longitudinal groove; lower portion deeply concave. Antenna relatively short, with 23 flagellomeres, each flagellomere longer than its diameter; penultimate



Figures 1–4. *Xorides benxicus* Sheng, sp. n. 1, 3–4 Holotype female 2 Paratype male 1,2 Body, lateral view 3 Head, anterior view 4 Head, dorsal view.

flagellomeres 4 to 6 strongly curved, each flagellomere at curve with 2 peg-like setae. Ratio of length from first to fifth flagellomeres: 2.9:3.5:3.7:3.9:3.9. Occipital carina complete.

Mesosoma. Anterior portion of pronotum with fine, dense punctures; lateral concavity smooth and shining, remaining portion with dense, coarse punctures; dorsal portion, neck, with three strong, forking longitudinal carinae (Figure 6). Epomia very strong, upper end reaching to upper margin of pronotum, projecting and turned inward to dorsal centre of neck. Mesoscutum with dense, fine punctures. Middle lobe of mesoscutum (Figure 7) and anterior portion of lateral lobes with dense, distinct punctures. Mesoscutum with longitudinal wrinkles postero-medially, postero-laterally smooth and shining. Notaulus shallow, reaching 0.6 to 0.7 × distance to posterior margin of mesoscutum. Scutoscutellar groove smooth with strong median longitudinal carina. Scutellum rough, with irregular wrinkles, medially convex, subapical-medially concave. Postscutellum semicircular ridge-shaped convexity, anteriorly deeply concave. Mesopleuron (Figure 8) shining, evenly convex, with fine dispersed punctures; ventro-posteriorly with distinct transverse groove; without speculum; mesopleural fovea consisting of short, shallow horizontal groove near mesopleural suture. Upper end of epicnemial carina reaching subalar prominence. Metapleuron rough, with irregular reticulate wrinkles. Submetapleural carina complete. Wings hyaline. Fore wing with vein 1cu-a distal to 1/M by 0.25 to 0.5 × length of 1cu-a. Vein 2rs-m almost disappeared, approximately 0.15 × distance between it and 2m-cu. Vein 2-Cu approximately as long as or slightly longer than 2cu-a. Hind wing vein 1-cu approximately as long as cu-a. Legs relatively slender. Fore and mid tibiae very thick, approximately columnar, subbasal part of ventral side angularly concave. Front side of fore tibia with short spines. Hind coxa elongate, medially distinctly expanded. Claws relatively small. Propodeum rough, completely areolated. Area basalis small, triangular, apical part of lateral longitudinal carinae combined. Area superomedia pentagonal, costula connecting approximately at its middle. Area externa with oblique longitudinal wrinkles. Area dentipara with irregular transverse wrinkles. Areas superomedia and posteroexterna with vague, irregular or reticulate wrinkles. Area petiolaris with distinct longitudinal wrinkles. Propodeal spiracle small, elongate.

**Metasoma**. First tergum 1.7 to  $1.8 \times as$  long as apical width, rough, with weak oblique median groove from median lateral margin extending backward to posterior median part; anterior to spiracle with transverse wrinkles; medially with irregular reticulate wrinkles; posteriorly with irregular longitudinal wrinkles. Median dorsal and dorsolateral carinae present from base to spiracle. Spiracle slightly convex, at anterior 0.35 of first tergum. Second tergum approximately  $0.76 \times as$  long as apical width; anteriorly with distinct punctures, posteriorly with irregular longitudinal wrinkles; with deep oblique groove cutting off basolateral corner. Third tergum 0.55 to  $0.6 \times as$  long as apical width, with weak punctures, posteriorly with weak and indistinct transverse wrinkles. Terga 4 to 6 very short, with fine leathery texture. Last tergum in dorsal



**Figures 5–9.** *Xorides benxicus* Sheng, sp. n. **5** Paratype male, basal portion of antenna **6–9** Holotype female **6** Pronotum and anterior portion of mesoscutum, dorsal view **7** Mesoscutum **8** Mesosoma, lateral view **9** Terga 2 & 3.



**Figures 10–12.** *Xorides benxicus* Sheng, sp. n. **10–11** Holotype female **10** Apical portion of metasoma, lateral view **11** Apical portion of ovipositor, lateral view **12** Cocoon.

view triangular, dorsally concave, with smooth median longitudinal groove. Ovipositor sheath approximately  $2.5 \times$  as long as hind tibia. Ovipositor relatively slender, apical portion depressed. Apical part of lower valve with 7 inclivous ridges, basal 4 or 5 distinct and strong; basal of the ridges with a roughened area, length of roughened area approximately as long as distance between basal ridge and end of lower valve.

**Color** (Figure 1). Black, except the following: anterior profile of scape and flagellomeres 10 to 13 white. Clypeus blackish brown, along ventral margin vaguely yellowish brown. Basal portion of mandible dark red. Posterior portion of malar space with small brown spot. Fore and mid coxae (except basal portions brownish black), apical spots of fore and mid femora, main portions of anterior and posterior profiles of fore and mid tibiae, hind coxa dorsoapically, hind margins of terga 3 to 7 except dorso-lateral sides, white. Fore and mid legs irregularly dark brown. Hind trochanter, femur basally, tibia medially, tarsomere 1 (to 2) blackish brown. Apex of hind femur, both apices of hind tibia irregularly brown. Hind tarsomeres (2) 3 and 4 brown to light brown. Apical margin of tergum 1 narrowly brown. Stigma dark brown. Veins brownish black.

**Male** (Figure 2). Body length 5.0 to 5.2 mm. Fore wing length 3.3 to 3.4 mm. Antenna length approximately 5.5 mm. Flagellum (Figure 5) slightly compressed, apex of each flagellomere swollen, lateral and ventral-lateral profiles with erect, long setae, setae approximately  $3.5 \times as$  long as width of flagellomere, curved apically. Stigma approximately  $3.2 \times as$  long as width. Antenna entirely black. Terga entirely black, or second tergum and apex of first tergum more or less blackish brown.

**Cocoon** (Figure 12). About 8 to 10 mm long, median width about 1.5 to 2.0 mm. yellowish grey.

Host. Pterolophia alternata Gressitt, 1938.

Host plant. Robinia pseudoacacia L.

This new species is similar to *X. asiasius* Sheng & Hilszczański, 2009, *X. cinnabarius* Sheng & Hilszczański, 2009 and *X. sapporensis* (Uchida, 1928), possessing subapical terga with white spots on apical part in females; flagellomeres with perpendicular hairs about as long as or longer than diameter of flagellomere, stigma short and wide, approximately or less 3x as long as wide, first tergum with oblique median groove running from median lateral margin extending backward to posterior median portion in male (*X. asiasius* unknown). It can be distinguished from them by the following key.

#### Key to the similar species to X. benxicus

1	Female
_	Male5
2	Median dorsal carinae of first tergum reaching to hind margin of first tergite3
_	Median dorsal carinae of first tergum at most reaching to median portion of
	first tergum
3	Terga 2 and 3 rough, with dense, indistinct punctures. Mesopleuron, propo-
	deum, femora and first tergum red. Scutellum with white spot
_	Apical portion of tergum 2 and entire tergum 3 transversely aciculate. Meso-
	pleuron, propodeum, scutellum, femora and first tergum entirely black
	X. sapporensis (Uchida)
4	Clypeus with fine transverse lines. Fore wing vein 1cu-a opposite 1/M. Ovi-
	positor sheath approximately 1.8 times as long as hind tibia. Ovipositor
	evenly and weakly down-curved, apically straight. Inner orbit, a large median
	spot on gena and apical spot on scutellum white. Terga 1 to 3 red. (Male
	unknown)

_	Clypeus slightly shagreened. Fore wing vein 1cu-a distal to 1/M. Ovipositor
	sheath approximately 2.5 times as long as hind tibia. Ovipositor straight, api-
	cally abruptly down-curved. Orbits, scutellum and terga 1 to 3 entirely black,
	except hind margin of tergum 1 narrowly reddish. X. benxicus Sheng, sp. n.
5	Terga 3 to 5, at least 4, transversely aciculate. Hind coxa and femur, me-
	tapleuron, propodeum and first tergum black
_	All terga entirely coarsely sculptured. Hind coxa and femur, at least parts of
	metapleuron, propodeum and first tergum red
	X. cinnabarius Sheng & Hilszczański
6	Antennal flagellum weakly compressed, apical portion of each flagellomere
	swollen and with erect, long setae, setae approximately 3.5 times as long as
	width of flagellomere
_	Antennal flagellum regular, apical portion of each flagellomere not swollen, se-
	tae approximately as long as width of flagellomereX. sapporensis (Uchida)

# Acknowledgements

The authors are deeply grateful to Dr. Gavin Broad, Department of Life Sciences, the Natural History Museum, London, UK, for reviewing this manuscript, and Prof. Wen-Kai Wang, Changjiang University, Hubei Province, China, for identifying the host. This project was supported by Liaoning Provincial Natural Science Foundation of China (No. 20102104) and the National Natural Science Foundation of China (NSFC, No. 31070585).

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CHECKLIST



# Reptiles from Lençóis Maranhenses National Park, Maranhão, northeastern Brazil

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Academic editor: N. Ananjeva | Received 23 December 2011 | Accepted 26 November 2012 | Published 29 November 2012

Citation: Miranda JP, Costa JCL, Rocha CFD (2012) Reptiles from Lençóis Maranhenses National Park, Maranhão, northeastern Brazil. ZooKeys 246: 51–68. doi: 10.3897/zookeys.246.2593

#### Abstract

We are presenting a list of the reptile species from Lençóis Maranhenses National Park (LMNP), Maranhão, Brazil, obtained during 235 days of field work. The study area is located in the contact zone between three major Neotropical ecosystems: Amazonia, Caatinga, and Cerrado. The PNLM encompasses the largest dune fields in Brazil, wide shrubby areas (restingas), lakes, mangroves, and many freshwater lagoons. We have recorded 42 species of reptiles in the area: 24 snakes, 12 lizards, two worm lizards, three turtles, and one alligator. About 81 % of the recorded species occurred only in restinga areas. Our data highlights the uniqueness of the PNLM in the context of the biomes that surround it and shows the importance of efforts to improve the conservation of reptiles living in the restinga, which currently comprise only about 20 % of the total area protected by the park, but which are the mesohabitat containing most of the reptile species in the Lençóis Maranhenses complex of habitats.

#### Resumo

No presente estudo apresentamos uma lista das espécies de répteis presentes no Parque Nacional dos Lençóis Maranhenses (LMNP), Maranhão, Brazil. A área do Parque Nacional dos Lençóis Maranhenses está localizada em um complexo ecótono na zona de contato entre a Amazônia, a Caatinga e o Cerrado. Além desta localização singular, a área inclui o maior campo de dunas do Brasil, extensas restingas, lagos, manguezais e uma grande quantidade de lagoas de água doce, formadas nos vales do campo de dunas. Registramos 42 espécies de répteis: 24 serpentes, 12 lagartos, duas cobras-cegas, três quelônios e um jacaré. Destas, cerca de 81 % foram encontrados apenas nos ambientes de restinga. Os resultados apresentados aqui enfatizam a singularidade do PNLM no contexto dos biomas que o cercam e ressaltam a importância

de ações para incrementar a conservação das áreas de restinga, as quais, atualmente, constituem apenas 20 % da área total protegida pelo parque, mas constituem o mesohábitat onde ocorre a maioria das espécies de répteis que vivem no complexo de ambientes que compõe os Lençóis Maranhenses.

#### **Keywords**

Richness, ecotone, lizards, snakes, turtles, worm lizards, dunes, restingas

#### Palabras clave

Riqueza, ecótono, lagartos, serpentes, tartarugas, cobras-cegas, dunas, restingas

#### Introduction

Brazil is a megadiverse country, including six biomes (Ab'Sáber 2003). One of its most distinguishable regions is the complex of ecotones adjacent to the Amazon forest of northern Brazil, the Caatinga in the northeastern portion of the country, and the Cerrado of central Brazil (Ab'Sáber 2003; Olson et al. 2005, Rodrigues 2005). In the very heart of this transitional region, one of the most remarkable environments is a region known as Lencóis Maranhenses in the state of Maranhão, located in the northeast of Brazil. The Lençóis Maranhenses comprises an unexpected and admirable landscape composed of the largest dunefields in Brazil (about 120,000 ha of continuous sand dunes), which is scattered by thousands of freshwater lagoons formed annually by rainfalls. In 1981, the area was converted into a park called Lençóis Maranhenses National Park due to its amazing scenery. Despite its relevance as a unique ecosystem in that transitional zone, animal and plant components are still poorly known. The few available studies just focused on individual species, usually only reporting its occurrence (e. g. Rêgo and Albuquerque 2006). There is no published inventory of the flora and fauna of Lençóis Maranhenses National Park. Regarding the fauna of reptiles in the park, the only study we are aware of is a description of a new turtle species by (Vanzolini 1995). The lack of a species list restricts the knowledge of the reptile species composition in the area. This hampers specific conservation efforts to protect local species diversity. Herein, as a result of almost two years of study conducted in the area, we are providing a report of reptile species composition for the Lençóis Maranhenses National Park, a detailed description of the main mesohabitats in the area, and some suggestions to improve the conservation of reptiles in the park.

#### Materials and methods

#### Study site

The Lençóis Maranhenses National Park (LMNP) is located in the Northeastern coast of Brazil (central coordinates: 02°31'02"S, 43°01'54"W, SAD69). The area of the park (about 155, 000 hectares) is composed of sand dunes, freshwater lagoons, restingas (local name for herbaceous and shrubby vegetation), lakes, mangroves, and 70 km of

beach. The dunefields arose from the varieties of sediments due to retrogradations from sedimentary deposits (Barreiras formation of Tertiary age), the correspondent widening of the continental shelf, successive marine transgressions since Pleistocene, and inputs of fluvial sediments from the main rivers in that region (Castro and Piorski 2002).

The climate in LMNP is warm (mean annual temperature: 28.5°C) with relatively little temperature variation throughout the year (about 1.1 °C, in average) and an annual rainfall between 1,600 and 2,400 millimeters (Nimer 1989, Castro and Piorski 2002). Most of the rain (about 70 %) occurs from January to May, when the level of underground water supply rises, and seasonally surfaces in the spaces between successive dunes, as lagoons.

We have distinguished the following seven mesohabitats in the LMNP: 1) Sand dunes "Morrarias" (vernacular expression): are sand dunes with no stabilizing vegetation, which is the most frequent and dynamic mesohabitat in the park (Figure 1A). The constant movement of sand dunes influences all other mesohabitats. The transportation of sand by wind constantly buries the vegetation in the area bordering the dunes (Figure 1B). Additionally, the migration of contiguous dunes spill water from one lagoon to the next closest one, resulting in a high interchange of water among lagoons. During the rainy season, however, the migration of dunes is slower because of the moisture that avoids sand transference (Parteli et al. 2006); 2) Freshwater lagoons: every year, thousands of freshwater lagoons appear in the dune fields in LMNP (Figure 1C). In the rainy season, lagoons can cover up to 41 % of the total area of the park (Levin et al. 2006), which represents about 64,000 ha. Most freshwater lagoons are shallow (less than 1m deep), and therefore are temporary. However, as reported to us by the native people, some rare lagoons can be as old as 16-20 years. Aquatic macrophytes like Utricularia sp. (Lentibulariaceae) and many species of algae can be found in freshwater lagoons; 3) Vargem (vernacular expression): are plain areas located in the depressions between successive dunes where there are herbaceous plants called "vassoura" (Figure 1D). The "vassoura" vegetation is composed mainly of plants of the genera Cassia (Fabaceae) and Borreria (Rubiaceae), which normally grow in areas where freshwater lagoons have dried up; 4) Restingas: are mosaics of open areas, freshwater lagoons, with herbaceous and shrubby vegetation (Figure 1E). The shrubby vegetation is composed mainly of grasses (Poaceae) and of "mirim" and "guajirú", which are plants belonging to the genera Humiria (Humiriaceae) and Chrysobalanus (Chrysobalanaceae) (Figure 1F). Restinga areas can be found within the park and in neighboring areas. These neighboring restinga areas are considered buffer zones around the park; 5) Innermost Isolated Restingas: In the middle of the dune field there are two "oases" (isolated restingas), one is called Queimada do Britos and the other is Baixa Grande. Queimada do Britos is the largest one, which is about 1,100 hectares. Baixa Grande has an area of 850 hectares and becomes largely flooded during the rainy season (Figure 2); 6) Rivers: There are many rivers and creeks in the region of the park. At least two of them connect to the fresh water lagoons during the rainy season: Rio Grande (in the region of Lagoa da Betânia) and Rio Negro (in the region of Lagoa da Esperança), which in the years of high precipitation, cross



Figure 1. Mesohabitats found at Lençóis Maranhenses National Park, Maranhão State, Northeastern Brazil: A Sand dunes "Morrarias" B Remains of restinga vegetation buried by sand transportation
C Freshwater lagoon D "Vargem" E Restinga mosaics in the boundary of sand dunes, including a lagoon
F Restinga mosaics far away from the sand dunes. Photos by J. P. Miranda.

the dunefield and reach the Atlantic Ocean; 7) Beaches: The coastal area of the park consists of 70 km of beaches. At these beaches it is common to find ropes, bottles and other human materials, which are constantly carried to the beach by the sea.

## Data collection

The species survey of reptiles at LMNP was conducted from September, 2004 to April, 2006 (IBAMA permit number 02001.004089/03-50). In that period, we made 47 trips, totaling 235 days of field sampling. At each site (see Figure 2), we sampled about three hectares. We used the time-constrained sample method, which is performed by walking slowly



**Figure 2.** Satellite image Landsat showing sample sites (light blue arrows) in the region of the Lençóis Maranhenses National Park, Maranháo State, Northeastern Brazil. The amount of sampling days is specified inside light blue squares. Two arrows linked to the same squares and pointing to different places indicate that the sampling effort in the square is divided between these sites. White areas with blue spots are sand dunes and freshwater lagoons, respectively. The greenish areas flecked by the orange color, are restingas habitats (green represents shrubby areas and orange most opening areas). QB and BG indicate the position of the isolated restingas called Queimada dos Britos and Baixa Grande, respectively. The violet line indicates the territory of LMNP. The red line is the road that accesses the park (MA-402). The location of the provinces Barreirinhas and Humberto de Campos are provided in the yellow rectangles. The position of the park in South America is provided upper right. Satellite image modified after Castro & Piorski (2002).

and searching for specimens in all visually accessible microhabitats (Heyer et al. 1994). The sampling effort was calculated during several days of sampling. Each day of sampling equaled the efforts of two collectors. These two collectors searched for specimens from 09:00 to 15:00 h (6 hours during the day) and from 19:00 to 23:00 h (4 hours during the night), totaling 1,410 hours of diurnal sampling and 940 hours of nocturnal sampling. Occasionally, additional specimens were obtained by random encounters, or were provided by the local people. Voucher specimens were deposited in the Coleção Herpetológica "Claude d'Abbeville" (CHMA) at Universidade Federal do Maranhão, Chapadinha, Maranhão, Brazil. The nomenclature of species follows the proposed taxonomy of Zaher et al. (2009) and Grazziotin et al. (2012) for families Colubridae and Dipsadidae, Hedges and Conn (2012) for family Mabuyidae, and Harvey et al. (2012) for Teiidae. Other families follow the nomenclature of the Brazilian List of Reptiles Species (SBH 2012).

#### Data analysis

We constructed species-accumulation curves that were generated using the nonparametric binomial mixture model of Mao et al. (2005). Additionally, to estimate the overall predicted species richness (extrapolation) for each reptile grouping (lizards or snakes), we used the first order Jackknife estimator (Heltshe and Forrester 1983). All analyses were made with EstimateS 8.0 (Colwell 2007). The results of Jackknife estimator appear within ± 1 standard deviation.

#### Results

We recorded 42 reptile species in the LMNP: 12 species of lizards, belonging to 11 genera and eight families (Gekkonidae, Sphaerodactylidae, Mabuyidae, Gymnophthalmidae, Iguanidae, Polychrotidae, Teiidae and Tropiduridae); two species of worm lizards belonging to the genus *Amphisbaena*, in the family Amphisbaenidae; 24 species of snakes, belonging to 20 genera and four families (Boidae, Colubridae, Dipsadidae and Elapidae); three species of turtles, belonging to three genera and three families (Cheloniidae, Dermochelyidae, and Emydidae); and one species of alligator (Alligatoridae) (Table 1).

The species-accumulation curves for snakes and lizards have different slopes and confidence intervals according to the reptile group studied. Nevertheless, both curves predicted more species than currently recorded. Richness for each reptile group was quite close to the predicted values, especially for the lizards (Figures 3 and 4). The first order Jackknife estimator predicted that 13 to 15 species of lizards  $[N(J1) = 13,99 \pm 1,40]$ , and 28 to 34 species of snakes  $[N(J1) = 30,97 \pm 2,95]$ might be recorded in LMNP. Overall, most reptile species that we recorded at LMNP were found in the restingas (Table 1).

**Table 1.** Reptile species recorded at Lençóis Maranhenses National Park, Maranhão State, Northeastern Brazil, respective environments of occurrence in the park, and figure numbers (when available). In the column mesohabitat, the numbers correspond to the following mesohabitats: 1) Sand dunes "Morrarias"; 2) Freshwater lagoons; 3) Vargem; 4) Restingas; 5) Innermost isolated restingas; 6) Rivers; 7) Beaches.

Reptilia	Mesohabitat	Figure
Order SAURIA		
Family Sphaerodactylidae		
Gonatodes humeralis (Guichenot, 1855)	4	5A
Family Gekkonidae		
Hemidactylus mabouia (Moreau de Jonnès, 1818)	4, 5	5B
Family Gymnophthalmidae		
Colobosaura modesta (Reinhardt and Lütken, 1862)	4	
Family Iguanidae		
Iguana iguana (Linnaeus, 1758)	4, 5	5C

Reptilia	Mesohabitat	Figure
Family Mabuyidae		
Varzea bistriata (Spix, 1825)	4, 5	5D
Brasiliscincus heathi (Schmidt & Inger, 1951)		5E
Family Polychrotidae		
Polychrus acutirostris Spix, 1825	4	5F
Family Teiidae		
Ameiva ameiva (Linnaeus, 1758)	4, 5	5G
Ameivula ocellifera (Spix, 1825)	1, 4, 5	5H
Kentropyx calcarata Spix, 1825	4	6A
Tupinambis teguixin (Linnaeus, 1758)	1, 4, 5	
Family Tropiduridae		
Tropidurus hispidus (Spix, 1825)	1, 4, 5	6B
Order AMPHISBAENIA	1	
Family Amphisbaenidae		
Amphisbaena ibijara Rodrigues, Andrade & Lima, 2003	4	
Amphisbaena vermicularis Wagler, 1824	4	6C
Order SERPENTES		
Family Boidae		
Boa constrictor Linnaeus, 1758	4	6D
Eunectes murinus (Linnaeus, 1758)	4, 5	
Family Colubridae		
Chironius flavolineatus (Jan, 1863)	4	
Drymarchon corais (Boie, 1827)	4, 5	6E
Leptophis ahaetulla (Linnaeus, 1758)	4, 5	6G
Mastigodryas bifossatus (Raddi, 1820)	4, 5, 6	
Oxybelis aeneus (Wagler, 1824)	4	6H
Oxybelis fulgidus (Daudin, 1803)	4	7A
Spilotes pullatus (Linnaeus, 1758)	4	
Tantilla melanocephala (Linnaeus, 1758)	4	
Family Dipsadidae		
Helicops angulatus (Linnaeus, 1758)	4, 5, 6	6F
Hydrodynastes gigas (Duméril, Bribon & Duméril, 1854)	4, 6	
Erythrolamprus poecilogyrus (Wied-Neuwied, 1825)	1, 4, 5	7B
Erythrolamprus taeniogaster (Jan,1866)	4	
Leptodeira annulata (Linnaeus, 1758)	4	
Lygophis meridionalis (Schenkel, 1902)	4	
Oxyrhopus trigeminus Duméril, Bibron & Duméril, 1854	4, 5	7C
Philodryas nattereri Steindachner, 1870	4	7D
Philodryas olfersii (Lichtenstein, 1823)	4	
Psomophis joberti (Sauvage, 1884)	4	7E
Taeniophallus occipitalis (Jan, 1863)	4	

Reptilia	Mesohabitat	Figure
Thamnodynastes hypoconia (Cope, 1860)	4	7F
Xenodon merremii (Wagler, 1854)	4	7G
Family Elapidae		
Micrurus ibiboboca (Merrem, 1820)	4, 5	
Order TESTUDINES		
Family Cheloniidae		
Chelonia mydas (Linnaeus, 1758)	7	
Family Dermochelyidae		
Dermochelys coriacea (Vandelli, 1761)	7	
Family Emydidae		
Trachemys adiutrix Vanzolini, 1995	1, 2, 3	7H
Order CROCODYLIA		
Family Alligatoridae		
Caiman crocodilus (Linnaeus, 1758)	6	



**Figure 3.** Accumulation curve for lizards recorded at the region of the Lençóis Maranhenses National Park (LMNP), Maranhão State, Northeastern Brazil (solid line). Dashed lines are confidence intervals at 95 %. The total number of sampling days is 235. A sample is equal to the search effort of two people looking for reptile species from 09:00 to 15:00 h and from 19:00 to 23:00 h.

## Discussion

In the management plans of the LMNP, there is no list of Herpetofauna's species (Castro and Piorski 2002). However, there is mention about the occurrence of the Brazilian slider turtle, *Trachemys adiutrix*, in the park. Thus, our study added 41



**Figure 4.** Accumulation curve for snakes recorded at the region of the Lençóis Maranhenses National Park (LMNP), Maranhão State, Northeastern Brazil (solid line). Dashed lines are confidence intervals at 95 %. The total number of sampling days is 235. A sample is equal to the search effort of two people looking for reptile species from 19:00 to 23:00 h and from 09:00 to 15:00 h.

species to the number of reptiles known in the LMNP. Despite the uniqueness of the environment, and conditions in the LMNP, the reptile taxocenosis recorded there includes species normally found in the biomes that surround it. For instance, *Gonatodes humeralis* and *Varzea bistriata* which are associated with the Amazon, and *Philodryas nattereri, Polychrus acutirostris* and *Brasiliscincus heathi* which are typical Cerrado and Caatinga inhabitants. One remarkable exception is *Trachemys adiutrix*, which is endemic to a small area at the coast of Maranhão and Piauí (see Avila-Pires 1995, Rodrigues and Prudente 2011, Mesquita et al. 2006, Vanzolini 1995 and references therein). Therefore, LMNP has a significant importance for conservation of reptiles in Brazil, as it includes in one relatively small area (155,000 ha), a single taxocenosis of reptiles that combine species from various Brazilian biomes, all in a very unique landscape.

In LMNP there are only three species of lizards: (*Tropidurus hispidus, Ameivula ocellifera*, and *Tupinambis teguixin*), one snake (*Erythrolamprus poecilogyrus*), and *Trachemys adiutrix* which were recorded in sand dune areas. Additionally, two sea turtles were recorded at the coastal area of the park (*Chelonia mydas* and *Dermochelys coriacea*). Thus, about 81 % (34 species) of reptile species recorded at LMNP were only found in restingas. In addition, the management plans of the LMNP emphasize the innermost isolated restingas for their actions in conservation, as opposed to restinga areas located at the southern LMNP (Castro and Piorski 2002). The impor-



**Figure 5. A** *Gonatodes humeralis* (female) **B** *Hemidactylus mabouia* **C** *Iguana iguana* (juvenile) **D** *Varzea bistriata* **E** *Brasiliscincus heathi* **F** *Polychrus acutirostris* **G** *Ameiva ameiva* **H** *Ameivula ocellifera* from Lençóis Maranhenses National Park, Maranhão State, Northeastern Brazil. Photos by J. P. Miranda.

tance of actions to protect the innermost isolated restingas is justified due to the high diversity of plant species found in those isolated areas (Castro and Piorski 2002). However, the reptile taxocenosis from the park did not follow that pattern of rich-



Figure 6. A Kentropyx calcarata B Tropidurus hispidus C Amphisbaena ibijara D Boa constrictor E Drymarchon corais F Helicops angulatus G Leptophis ahaetulla H Oxybelis aeneus from Lençóis Maranhenses National Park, Maranhão State, Northeastern Brazil. Photos by J. P. Miranda.

ness. The species recorded at the isolated restingas were only a subset of the reptile species registered at the restingas which are adjacent to sand dunes in the southern LMNP (see Table 1 and Figure 2).



Figure 7. A Oxybelis fulgidus B Erythrolamprus poecilogyrus C Oxyrhopus trigeminus D Philodryas nattereri E Psomophis joberti F Thamnodynastes hypoconia G Xenodon merremii H Trachemys adiutrix (two individuals in a freshwater lagoon) from Lençóis Maranhenses National Park, Maranhão State, Northeastern Brazil. Photos by J. P. Miranda.

During our field work, we observed that the restingas in the south area of the park (both inside the park and in the buffer zones) have been strongly disturbed by the clandestine openings of paths created to transport tourists to the dunes and lagoons in the park, using off-road vehicles. This problem is more severe during the rainy season (from January from June) when paths become muddy quickly, and new paths are continuously opening. Restinga are extremely sensitive to clearing because the poor soils hinder habitat recomposition (Hay et al. 1981). In fact, the restinga areas in the south of the park (outside LMNP) are included in an environmental protection area called APA Upaon Açu-Miritiba-Alto Preguiças, which is a type of protected area in the Brazilian system of conservation units which ensures sustainable use. However, the surveillance of this use is still very limited.

On the beach and sand dune areas, we recorded three species of turtles which are included in the IUCN Red List of Threatened Species (IUCN 2012): *Dermochelys coriacea* (critically endangered), *Chelonia mydas* (endangered), and *Trachemys adiutrix* (endangered). During our field work, we observed the *T. adiutrix* (locally called "Pininga") being used as a food source; eaten by the extremely poor human population which live in the park area and surroundings. These turtles cannot be seen very easily in LMNP during most of the year. However, as the lagoons begin to diminish (both in number and surface) in the dry season, these turtles concentrate in the few remaining lagoons, making it easy to capture a large number of individuals at the same time. Some people capture the turtles and keep them alive, in order to eat them during the dry season. This is the time when fish and other food items are scarce for the local human population.

In the sand dunes at LMNP, which is an extremely open area, the ground temperature can easily exceed 70° C during the warmest period of the day (JPM Pers. Obs.). This particular characteristic of sand dune areas reinforces the importance of shelters (dead branches and patches of vegetation) and burrows for the species that live there. For lizards, shelters and burrows are important for thermoregulation, as this is one of the few options to decrease exposure to the sun (Rocha et al. 2009). Furthermore, in sand dune areas at LMNP, there are some species of predatory birds like "carcará" (Polyborus plancus, Falconidae) and "caburé" (Athene cunicularia, Strigidae) (JPM Pers. Obs), known to prey on reptiles (Andrade et al. 2010, Vargas et al. 2007). Thus, shelters and burrows may also be important for the protection against predators. Whiptail lizards (Ameivula ocellifera) have a great ability to dig (see Eifler and Eifler 1998), which might have been important for their successful establishment in sand dunes. The other lizard species found in sand dunes, Tropidurus hispidus, is not able to dig as well, but is known as a species with great flexibility in habitat use (Avila-Pires 1995). In sand dunes at LMNP, T. hispidus is often sheltered in dead branches of the shrubs buried by sand. During our fieldwork, we often observed the clandestine traffic of off-road vehicles in the dunes, which can be harmful to the reptiles that live in that mesohabitat due to the fact that heavy vehicles destroy a large number of burrows and shelters used by those species. This would be similar to the adverse effects of off-road vehicles on lizard populations observed by Busack and Bury (1974) in the Mojave desert, USA.

The only exotic invasive reptile species at LMNP was the gecko *Hemidactylus mabouia*, which was found on different occasions in natural habitats and microhabitats within the study area. This lizard, which is native from Africa, is one of the five invasive reptile species presently known to occur in Brazil (Rocha et al. 2011) and has continuously invaded natural environments in Brazil during the past 70 years (Rocha et al. *in press*).

Our data highlight the singularity of the LMNP in the context of the biomes that surround it, and also demonstrate the importance of actions to improve conservation of reptiles that live in both sand dunes and restingas in LMNP. Currently, restingas comprise only about 20% of the total area protected by the park, but most reptile species live in restinga mesohabitats. Thus, we suggest the addition of restinga areas adjacent to the park (buffer zones) to be incorporated into the national park, which is a fully protected conservation unit in the Brazilian system of conservation units. This would be the most effective way to protect the biodiversity of reptiles in the restinga areas in that region. Moreover, regarding the sand dunes areas; we suggest an improvement in the security at the LMNP to prevent the illegal use of off-road vehicles inside the park territory, the promotion of actions to monitor the activities of sea turtles at the coast of LMNP, and the implementation of an effective strategy to protect the Brazilian slider turtle. This strategy could be in the form of awareness campaigns, or even the promotion of training courses (e. g. tourist guides, waiters, cooks, or hotel maids) for those living in the park region. This would place local people into the tourism business, which would not only improve their economical capacity, but also reduce their need to use *Trachemys adiutrix* as a food item.

## Acknowledgements

We thank Antonio Pereira, Maria Grossa, Arnaldo Oliveira Silva, Edmilson Godé, Adriano Kid Azambuja and Thiare Fortes for field assistance; Romário Ferreira de Matos, Raiana Cristina Araújo for help in laboratory, and Kristen Hammer for English review of the manuscript. JPM thanks Conselho Nacional de Desenvolvimento Científico e Tecnológico – CNPq for granting his PhD Scholarship and Fundação de Amparo à Pesquisa e ao Desenvolvimento Científico e Tecnológico do Maranhão – FAPEMA (INFRA 00563/10) for research grants. JCLC was supported by a scholarship from CNPq. CFDR received grants from CNPq (304791/2010-5 and 470265/2010-8) and from Fundação Carlos Chagas Filho de Amparo à Pesquisa do Estado do Rio de Janeiro – FAPERJ (Process E-26/102.404.2009) through Programa Cientistas do Nosso Estado. Valdir Germano, Francisco Franco, Daniel Fernandes and Miguel T. Rodrigues helped us with the identification of the specimens. This study was supported by Instituto Biomas and by a research grant from Fundação "O Boticário" de Proteção à Natureza (Process 0612/2004-1). Three anonymous reviewers provided valuable suggestions on the manuscript.

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## Appendix

#### Specimens examined

Voucher specimens of the present study were deposited in the Coleção Herpetológica "Claude d'Abbeville" (CHMA) at Universidade Federal do Maranhão, Chapadinha, Maranhão, Brazil.

## Order SAURIA

#### Family Sphaerodactylidae

Gonatodes humeralis – CHMA 554.

#### Family Gekkonidae

Hemidactylus mabouia - CHMA 538, 542, 545, 555, 557, 561.

#### Family Mabuyidae

Varzea bistriata – CHMA 503.

Brasiliscincus heathi – CHMA 504, 505, 549.

## Family Polychrotidae

Polychrus acutirostris – CHMA 551, 556.

#### Family Teiidae

Ameiva ameiva – CHMA 507, 509, 511, 535, 548, 569. Ameivula ocellifera – CHMA 506, 508, 510, 515, 517, 520-524, 571, 574. Kentropyx calcarata – CHMA 512. Tupinambis teguixin – CHMA 567.

## Family Tropiduridae

Tropidurus hispidus – CHMA 558-560, 562-564.

## Order AMPHISBAENIA

#### Family Amphisbaenidae

Amphisbaena ibijara – CHMA 516, 518. Amphisbaena vermicularis – CHMA 513, 519.

## **Order SERPENTES**

## Family Colubridae

Chironius flavolineatus – CHMA 537, 572. Drymarchon corais – CHMA 576. Mastigodryas bifossatus – CHMA 566. Oxybelis aeneus – CHMA 541. Spilotes pullatus – CHMA 570. Tantilla melanocephala – CHMA 575.

#### Family Dipsadidae

Helicops angulatus – CHMA 529, 530, 573. Hydrodinastes gigas – CHMA 568. Erythrolamprus poecilogyrus – CHMA 536, 539, 543, 552. Leptodeira annulata – CHMA 528 Oxyrhopus trigeminus – CHMA 525-527. Philodryas nattereri – CHMA 534, 544. Philodryas olfersii – CHMA 540. Psomophis joberti – CHMA 500, 501, 553. Taeniophallus occipitalis – CHMA 550. Thamnodynastes hypoconia – CHMA 531-533. Xenodon merremii – CHMA 546.

## Family Elapidae

Micrurus ibiboboca – CHMA 547.

## **Order TESTUDINES**

#### Family Emydidae

Trachemys adiutrix - CHMA 565.

RESEARCH ARTICLE



# Two new species of Neozoanthus (Cnidaria, Hexacorallia, Zoantharia) from the Pacific

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Academic editor: Bert W. Hoeksema | Received 22 August 2012 | Accepted 15 November 2012 | Published 29 November 2012

urn:lsid:zoobank.org:pub:C829F5F6-81F6-4424-9DE7-F330DF5A23D5

**Citation:** Reimer JD, Irei Y, Fujii T (2012) Two new species of *Neozoanthus* (Cnidaria, Hexacorallia, Zoantharia) from the Pacific. ZooKeys 246: 69–87. doi: 10.3897/zooKeys.246.3886

#### Abstract

The zoanthid genus *Neozoanthus* was originally described in 1972 from a single species in Madagascar. This monotypic genus was placed within its own family, Neozoanthidae, given its unusual characters of only partial sand encrustation, and an endodermal sphincter muscle combined with a brachycnemic mesenterial arrangement. Recently, undescribed specimens of *Neozoanthus* were discovered thousands of kilometers away in both Australia and Japan. While the phylogenetic and evolutionary aspects of *Neozoanthus* spp. are now somewhat well understood, the new specimens remained undescribed. Here we describe the specimens as two new species, *N. uchina* **sp. n.** from the Middle Ryukyu Islands of southern Japan, and *N. caleyi* **sp. n.** from the waters around Heron Island, on the Great Barrier Reef in Australia. Both species can be distinguished from each other and the type species, *N. tulearensis*, by their distributions, oral disk colors, and average numbers of tentacles. Additionally, each species appears to have subtle differences in their cnidae. The division of Japanese and Australian specimens into two species is strongly supported by recently reported phylogenetic data. The discovery and description of these two species highlights how little is known of zoanthid species diversity in the Indo-Pacific.

#### Keywords

Zoanthid, Great Barrier Reef, Okinawa, Neozoanthus, new species

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#### Introduction

Zoanthids are a hexacorallian order (Zoantharia =Zoanthidea) of benthic anthozoans with gross morphological characteristics partially reminiscent of both hard corals and sea anemones. Similar to many Scleractinia, most zoanthid species are colonial, with individual polyps connected by common tissue (=coenenchyme). However, like actiniarians, most zoanthids do not secrete hard skeletons. Instead most zoanthids incorporate sand and/or detritus into their body walls to help strengthen their structure. There are, however, exceptions to these general characters within Zoantharia, with some solitary (e.g. the genus *Sphenopus*), non-encrusting (Zoanthidae; *Zoanthus, Acrozoanthus, Isaurus*), and skeleton-secreting (*Savalia*) taxa.

While zoanthids can be found in a wide variety of marine environments from shallow waters to the deep sea, much of their diversity is found in subtropical and tropical coral reef ecosystems, particularly within the suborder Brachycnemina (Swain 2010). The suborder Brachycnemina includes both encrusting (Sphenopidae) and non-encrusting (Zoanthidae) families, grouped together by possession of a mesodermal sphincter, unlike species in suborder Macrocnemina, which generally possess an endodermal sphincter (but see Swain 2010). Additionally, most coral reef brachycnemic zoanthids are in symbiosis with endosymbiotic dinoflagellate zooxanthellae (=*Symbiodinium* spp.). The most common genera are *Zoanthus* (Zoanthidae) and *Palythoa* (Sphenopidae), which are a major component of coral reef fauna in both the Atlantic and Indo-Pacific. The other coral reef zoanthid genera in Brachycnemina have been little studied due in part to their rarity or cryptic nature, and include the genera *Acrozoanthus* (Ryland 1997; Reimer et al. 2011c) and *Isaurus* (Reimer et al. 2008) in the family Zoanthidae, and the genus *Sphenopus* (Soong et al. 1999; Reimer et al. 2012) in the Sphenopidae.

There is one additional family of brachycnemic zoanthids, the monotypic Neozoanthidae. Neozoanthidae was erected by Herberts in 1972 to contain the genus and species *Neozoanthus tulearensis*, described from unusual zoanthid specimens found in coral reefs of Madagascar. The specimens were notable for zoanthids in that they had an endodermal sphincter (Herberts 1972: 139, fig. 11) but brachycnemic mesentery arrangement, unlike all other Brachycnemina. Furthermore, specimens were only partially encrusted with sand by having no encrustations around the oral ends (=tops) of polyps (Herberts 1972: 139, fig. 10). This genus has remained monotypic, and until the recent rediscovery of undescribed *Neozoanthus* specimens from the Indo-Pacific (Reimer et al. 2011a), no additional specimens had been noted in the literature.

*Neozoanthus* represents a unique evolutionary step in the zoanthid phylogeny as the only partially encrusted group of zoanthids (Reimer et al. 2011a) (Figure 1) (Table 1). Surprisingly, from mitochondrial 16S ribosomal DNA phylogenetic analyses, this group appears to be very closely related to the genus *Isaurus*, yet also has an indel unique to tropical macrocnemic Hydrozoanthidae (Reimer et al. 2011a), indicating *Neozoanthus* has a unique and perhaps complex evolutionary history.

In this study, utilizing both morphological and molecular techniques, we formally describe two new *Neozoanthus* species from subtropical regions of the Great Barrier



**Figure 1.** Internal structure of *Neozoanthus uchina* sp. n. showing encrustation in outer mesoglea and ectoderm, characteristic of *Neozoanthus* spp., with irregularly-sized encrustation **A** light microscope histological cross-section, and **B** scanning electron microscope image. Both images of specimen RMNH Coel 40098 (Table 2). Abbreviations: **cm**=complete mesenteries, **e**= sand/detritus encrustation (in **B**) or where encrustation existed before decalcification (in **A**), **im**=incomplete mesenteries, **e**=ectoderm, **e**=endoderm, **m**=mesoglea, **ss**=encrusted sponge spicules. Scales: **A**=100 µm, **B**=200 µm.

Genus	Encrusta- tion?	Sphincter complexity	Sphincter position	Lacunae?	Mesogleal canals?	Endodermal invagination?
Palythoa	Yes	Simple	Mesogleal	No	Yes	No
Zoanthus	No	Double	Mesogleal	Yes	Yes	No
Isaurus	No	Simple	Mesogleal	No	No	Yes
Neozoanthus	Partial	Simple	Endodermal	No	No	No
Specimens in this study	Partial	Simple	Endodermal	No	No	No

**Table 1.** Summary of morphological characters of major brachycnemic zoanthid genera compared with specimens examined in this study (adapted from Reimer et al. 2011a) to show placement of specimens within *Neozoanthus* Herberts, 1972.

Reef, Australia and the Ryukyu Archipelago, Japan by examining specimens recently reported in Reimer et al. (2011a). It is hoped these formal descriptions will provide a basis for future research into this enigmatic genus of zoanthids.

## Methods

## Sample collection

Specimens were collected as detailed in Reimer et al. (2011a) from locations in the Heron Island region of the Great Barrier Reef, Australia, and from the Ryukyu Islands in southern Japan (Table 2). In situ observations were also performed as detailed in Reimer et al. (2011a). Specimens were initially preserved in 70-99% ethanol.

## Specimen examination/decalcification/histology

Specimens were examined, decalcified, and sectioned as detailed in Reimer et al. (2011a), with additional analyses as detailed below. As detailed in Reimer et al. (2011a), two polyps from two specimens each (Australia and Japan) were examined (total n=8).

## Morphological analyses

External morphology of specimens was examined using both preserved specimens and in situ images. Polyp dimensions (oral disk diameter, polyp height) for both in situ and preserved specimens were obtained, as were the following data: tentacle number, color of polyp, color(s) of oral disk, relative amount of sand encrustation, associated/substrate species. Additionally, the relative development of the coenenchyme was examined.

For internal examinations, the following data were obtained: mesentery form (brachycnemic or macrocnemic arrangement), mesentery numbers, presence/absence
Table 2. Neozoanthus specimens examined in this study with cytochrome oxidase subunit I (COI) and mitochondrial 16S ribosomal DNA (mt 16S rDNA) Gen-Bank Accession Numbers. Data based on similar table in Reimer et al. (2011a).

Specimen number	Species	Collection location	Latitude and longitude	Depth (m)	Collection date	Collector(s)	COI	mt 16S rDNA
NSMT Co1554	N. caleyi	North West Reef, GBR, Australia	23.3180°S, 151.7170°E	10	Nov. 17, 2009	JD Reimer	NA	NA
HI141	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.034°E	21	Nov. 18, 2009	JD Reimer	HM991247	HM991230
HI142	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	21	Nov. 18, 2009	JD Reimer	HM991248	HM991231
HI143	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	21	Nov. 18, 2009	JD Reimer	NA	HM991232
HI144	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	20	Nov. 18, 2009	JD Reimer	NA	HM991233
HI145	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	18	Nov. 18, 2009	JD Reimer	HM991249	HM991234
HI199	N. caleyi	Heron Channel, GBR, Australia	23.4448°S, 151.9504°E	22	Nov. 22, 2009	JD Reimer	NA	NA
HI200	N. caleyi	Heron Channel, GBR, Australia	23.4448°S, 151.9504°E	23	Nov. 22, 2009	JD Reimer	HM991250	HM991235
HI209	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	28	Nov. 23, 2009	JD Reimer	HM991251	HM991236
HI214	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	6	Nov. 23, 2009	JD Reimer	HM991252	HM991237
MTQ G65793	N. caleyi	Sykes Reef, GBR, Australia	23.4322°S, 152.0338°E	4	Nov. 23, 2009	JD Reimer	HM991253	HM991238
HI224	N. caleyi	Heron Channel, GBR, Australia	23.4532°S, 151.9005°E	26	Nov. 24, 2009	JD Reimer	HM991254	HM991239
HI225	N. caleyi	Heron Channel, GBR, Australia	23.4532°S, 151.9005°E	25	Nov. 24, 2009	JD Reimer	HM991255	HM991240
HI227	N. caleyi	Heron Channel, GBR, Australia	23.4532°S, 151.9005°E	25	Nov. 24, 2009	JD Reimer	HM991256	HM991241
HI231	N. caleyi	Heron Channel, GBR, Australia	23.4530°S, 151.9171°E	23	Nov. 24, 2009	JD Reimer	HM991257	HM991242
HI101114-13	N. caleyi	Sykes Reef, GBR, Australia	23.4316°S, 152.0493°E	29	Nov. 14, 2010	JD Reimer	NA	NA
RMNH Coel 40098	N. uchina	Manza, Okinawa, Japan	26.5047°N, 127.8450°E	25	Sept. 1, 2008	JD Reimer et al.	NA	NA
MISE 545	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	1 to 2	Sept. 5, 2008	JD Reimer et al.	NA	NA
MISE 546	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	1 to 2	Sept. 5, 2008	JD Reimer et al.	NA	NA
USNM 1194728	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	1 to 2	Sept. 5, 2008	JD Reimer et al.	HM991246	HM991227
MISE 549	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	1 to 2	Sept. 5, 2008	JD Reimer et al.	NA	NA
NSMT Co1553	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	1 to 2	Sept. 5, 2008	JD Reimer et al.	HM991243	NA

Specimen number	Species	Collection location	Latitude and longitude	Depth (m)	Collection date	Collector(s)	COI	mt 16S rDNA
MISE 560	N. uchina	Yona, Okinawa, Japan	26.7684°N, 128.1976°E	13	Sept. 24, 2008	JD Reimer, T Fujii	NA	NA
MISE 1092	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	1	July 2008	JD Reimer	NA	NA
MISE 1093	N. uchina	Teniya, Okinawa, Japan	26.5638°N, 128.1408°E	Inter- tidal	July 2008	JD Reimer	NA	NA
MISE 1115	N. uchina	Tinyuhama, Korijima, Okinawa, Japan	26.7149°N, 128.0127°E	24	Dec. 28, 2008	JD Reimer	HM991245	HM991228
MISE 1116	N. uchina	Tinyuhama, Korijima, Okinawa, Japan	26.7149°N, 128.0127°E	24	Dec. 28, 2008	JD Reimer	HM991244	HM991229
MISE 1400	N. uchina	Omonawa, Tokunoshima, Kagoshima, Japan	27.6669°N, 128.9685°E	6	March 9, 2010	JD Reimer	NA	NA
MISE 1401	N. uchina	San, Tokunoshima, Kagoshima, Japan	27.8693°N, 128.9699°E	10	March 10, 2010	JD Reimer	NA	NA
<b>MISE 1402</b>	N. uchina	San, Tokunoshima, Kagoshima, Japan	27.8693°N, 128.9699°E	12	March 10, 2010	JD Reimer	NA	NA
MISE 1403	N. uchina	Zampa, Okinawa, Japan	26.4414°N, 127.7119°E	NA	August 29, 2008	JD Reimer	NA	NA
MISE MO-100	N. uchina	Tebiro, Amami-Oshima, Kagoshima, Japan	28.4013°N, 129.6178°E	10	March 16, 2011	M Obuchi	NA	NA

Abbreviations: GBR=Great Barrier Reef; NA=not acquired. Sample number abbreviations as in Methods.

of encrustations, location of encrustations, location and development of the sphincter muscle, presence/absence of gonads. Decalcification, histology and electron microscopy were performed as described in Reimer et al. (2011a).

### Nematocyst observation

Undischarged nematocysts were measured from tentacles, column, actinopharynx, and mesenterial filaments of polyps (specimens examined n=2-4 colonies/species) for both new species. 400x images of the nematocysts were obtained by optical microscope, and measured using the software ImageJ (National Institutes of Health, USA). Nematocyst nomenclature generally followed England (1991), however both Schmidt (1974) and Hidaka and co-workers (1987; 1992) have previously suggested basitrichs and mastigophores are same type of the nematocyst, and thus in this study, these two types were dealt with as the same type (basitrichs and *b*-mastigophores), unless they could be clearly distinguished from one another (basitrichs and *p*-mastigophores), in which case they were analyzed separately. Both holotypes and all paratypes of both newly described species were examined.

# DNA extraction and PCR amplification/Phylogenetic analyses

Phylogenetic analyses are detailed and were performed on both species in Reimer et al. (2011a). No new genetic analyses were performed in this study. DNA from specimens were extracted, DNA target regions (cytochrome oxidase subunit I; mitochondrial 16S ribosomal DNA) amplified by PCR, and sequences analysed as detailed in Reimer et al. (2011a). DNA alignments from Reimer et al. (2011a) are available from the corresponding author.

## Abbreviations used

MTQ	Museum of Tropical Queensland, Townsville, Australia.			
USNM	Smithsonian National Museum of History, Washington D.C., USA			
NSMT	National Museum of Nature and Science, Tokyo, Japan			
RMNH	Naturalis Biodiversity Center, Leiden, the Netherlands			
MISE	Molecular Invertebrate Systematics and Ecology Laboratory, University of			
	the Ryukyus, Nishihara, Okinawa, Japan			

# **Systematics**

Additional data related to both species, including tables, phylogenetic trees, and histological images, are reported in Reimer et al. (2011a). The specimens in Reimer et al. (2011a) were placed into the genus *Neozoanthus* Herberts, 1972 based on the summary of morphological characters (Table 1).

### Family Neozoanthidae Herberts, 1972

**Diagnosis.** Brachycnemic zoanthids with a simple endodermal sphincter muscle (Figure 2) that are only partially sand-encrusted (Figure 1).

#### Genus Neozoanthus Herberts, 1972

**Type species.** *Neozoanthus tulearensis* Herberts, 1972 **Diagnosis.** As for the family above.

Neozoanthus caleyi sp. n.

urn:lsid:zoobank.org:act:3BD527A8-F3CC-4A09-A933-313EBAE6A45C http://species-id.net/wiki/Neozoanthus\_caleyi Figures 2, 3, 5A, Tables 2, 3, S1

Synonymy: "GBR clade" of *Neozoanthus* Reimer et al. 2011a: fig. 2. "*Neozoanthus* sp. Australia" - Reimer et al. 2011a: 986, 989, fig. 4.

### Material examined.

**Type specimens.** Holotype, specimen number MTQ G65793. Colony in two pieces, 5 polyps on a  $2.5 \times 2.0$  cm stone and 4 polyps on a  $2.0 \times 1.0$  cm stone (originally one colony). Polyps approximately 2.3-5.0 mm in diameter, and approximately 2.5-3.0 mm in height from stoloniferous coenenchyme. Polyps and coenenchyme encrusted with irregularly sized and colored sand grains. There was no noticeable variation between holotype and other specimens. Preserved in 99.5% ethanol.

*Paratype* (from Australia): Paratype 1. Specimen number NSMT Co1554. North West Reef, Queensland, at 10 m by JDR, November 17, 2009.

**Type locality.** Australia, Queensland: Great Barrier Reef, Sykes Reef, 23.4322°S, 152.0338° E, reef with coral rubble, at 4 m, 23 November 2009, JDR leg.

**Other material** (all from Great Barrier Reef, Queensland, Australia; coll. JDR): Sykes Reef MISE HI-141 to 145 (n=5), 18-21 m, 18 November 2009; MISE HI-209, 28 m, 23 November 2009; MISE HI-214, 9 m, 23 November 2009; MISE HI101114-13, 29 m, 14 November 2010. Heron Island Channel, MISE HI-199 to 200 (n=2), 22-23 m, 22 November 2009; MISE HI-224 to 225, 227 (n=3), 25-26 m, 24 November 24 2009; MISE HI-231, 23 m, 24 November 2009 (see also Table 2).



**Figure 2.** Longitudinal section of *Neozoanthus caleyi* sp. n. specimen HI225 showing endodermal sphincter muscle (=sm). **A** Light microscope **B** scanning electron microscope. Both scales =200  $\mu$ m.



**Figure 3.** *Neozoanthus caleyi* sp. n. in situ around Heron Island on the Great Barrier Reef, Queensland, Australia. **A** Specimen HI214 at Sykes Reef, depth=9 m, November 23, 2009 **B** Close-up of a single polyp showing yellow coloration at base of tentacles; specimen HI145 at Sykes Reef, depth=18 m, November 18, 2009 **C** Specimen HI231 at Heron Channel, depth=23 m, November 24, 2009 **D** Uncollected specimen at Heron Channel, depth=approximately 20 m, November 2011. Scales approximately 1 cm. **A**, **B** taken by JD Reimer, **C**, **D** taken by Gary Cranitch.

**Description.** *Size:* Polyps in situ approximately 2–5 mm in diameter when open, and approximately 2–3 mm in height.

*Morphology: Neozoanthus caleyi* sp. n. has 28 to 40 (average 33±3.9, n=18 polyps on 8 colonies) conical tentacles. Tentacles are usually shorter than the expanded oral disk diameter (e.g. 50-80% of oral disk width). Tentacles may be grayish-blue, yellow, or transparent, often with black, white, or fluorescent blue bands or patterning (Figure 3). Well-developed, simple endodermal sphincter. No bractae are visible. All specimens are zooxanthellate. Polyps are externally heavily encrusted with sand and other particles of irregular sizes, excepting the oral end, which is free of encrustation and appears a bluish-gray similar to as seen in some *Zoanthus* species. When fully contracted, the sand-free oral end is often not visible, and polyps resemble small balls of sand. Polyps extend well clear of reduced or stoloniferous coenenchyme (Figure 3). Oral disks may

Morphological character	N. tulearensis	<i>N. uchina</i> sp. n.	<i>N. caleyi</i> sp. n.
Distribution	NE Madagascar	Middle Ryukyu Is- lands, Okinawa, Japan	Heron Island, Great Barrier Reef, Australia
Depth	No data	Intertidal to 25 m	4 to 29 m
Oral disk color	Greenish-beige to yellow	Light gray-blue, white, rust or deep wine red	Light gray-blue, white, or deep wine red
Polyp diameter (mm)	1.5 to 5.0	2.2 to 5.1	2.3 to 5.0
Polyp height (mm)	2.0 to 12.0	2.0 to 8.5	2.5 to 3.0
Number of ten- tacles (avg. ± SE)	38–44	32-42 (38±3.0)	28-40 (33±3.9)
Cnidae			
Column	Microbasic mastigophores	Holotrichs	Holotrichs
Pharynx	Microbasic mastigophores	Holotrichs, basitrichs, spirocysts	Holotrichs, basitrichs
Tentacles	Holotrichs, spirocysts	Holotrichs, basitrichs, spirocysts	Holotrichs, basitrichs, spirocysts
Filaments	Holotrichs, microbasic mastigophores	Holotrichs, <i>p</i> -mastigo- phores	Holotrichs, basitrichs, <i>p</i> -mastigophores

**Table 3.** Comparison of various features of *Neozoanthus tulearensis* Herberts, 1972, *N. caleyi* sp.n. and *N. uchina* sp. n.

be a variety of colors, including light gray-blue, white, or deep wine red. Occasionally, white, yellow, or light blue dots may be seen on the oral disk in regular circular patterns, and the oral opening (mouth) is often white in color. A "skirt" of different coloration (usually white or lighter coloration than remainder of oral disk) covering up to approximately 90 degrees of the oral disk is often seen in the area of the dorsal directive. Colonies consist of tens to <100 polyps, connected by stolons with no welldeveloped coenenchyme.

*Cnidae:* Basitrichs and microbases (often difficult to distinguish), holotrichs (large and small), spirocysts (see Table S1, Figure 5).

**Differential diagnosis.** Differs from *Neozoanthus tulearensis* Herberts, 1972 and *Neozoanthus uchina* sp. n. with regards to distribution (southern Great Barrier Reef as opposed to Madagascar and Ryukyu Archipelago, respectively), coloration (no yellow observed in any *N. uchina* sp. n.), and tentacle count (*N. tulearensis* = 38 to 44 tentacles (n= 8 colonies; 18 polyps), *N. uchina* sp. n. = average 38±3.0 tentacles, n= 9 colonies; 24 polyps). The two new *Neozoanthus* species' tentacle counts are statistically significant (t-test, p<0.001). The two new *Neozoanthus* species mt 16S rDNA sequences differ by three base pairs (Reimer et al. 2011a).

**Etymology.** Named for Dr. Julian Caley, the leader of the Australian Census of Coral Reef Ecosystems (CReefs) project. Dr. Caley's acceptance of the first author's participation in CReefs led to the discovery of this species. Noun in genitive.

**Habitat, ecology and distribution.** Specimens from the Great Barrier Reef were found at depths from 4 to 29 m. Despite repeated surveys, no *N. caleyi* sp. n. have been found further north around Lizard Island despite zoanthid-focused surveys (Burnett et al. 1997; J.D. Reimer & T. Fujii, unpublished data), and it may be that this species is limited to a subtropical distribution in the Great Barrier Reef.

*Neozoanthus caleyi* sp. n., although not found at many locations surveyed, was locally common, particularly at locations that were characterized by strong currents and some sedimentation, with large coarse sand particles scattered over the bottom or rocks, for example on the bottom of Heron Channel. Preference for such environments may be related to its encrustation patterns. Colonies were never found in locations completely exposed to light, yet all colonies were zooxanthellate. Most colonies were relatively small, consisting of tens (not hundreds) of polyps, with polyps spread out and connected by thin stolons (Figure 3).

**Notes.** This species can close its polyps much more rapidly than those of other zooxanthellate zoanthid genera (Reimer pers. obs).

**DNA Sequences.** Originally listed in Table S1 in Reimer et al. (2011a). Cytochrome oxidase subunit I: HM991247-HM991257 Mitochondrial 16S ribosomal DNA: HM991230-HM991242

#### Neozoanthus uchina sp. n.

urn:lsid:zoobank.org:act:BBAB21D0-275E-4287-90F4-4928CE1BF05E http://species-id.net/wiki/Neozoanthus\_uchina Figures 1, 4, 5B, Tables 2, 3, S1

Synonymy: "*Neozoanthus* sp. okinawa" – Reimer 2010: 25, 27, fig. 8. Reimer et al. 2011a: 986, 989, fig. 4. "Japan clade" of *Neozoanthus* - Reimer et al. 2011a: fig. 2.

### Material examined.

**Type specimens.** Holotype, specimen number NSMT-Co1553. Colony of 17 polyps connected by stoloniferous coenenchyme on a rock approximately 4.5 × 3.0 cm. Polyps approximately 2.0–4.4 mm in diameter, and approximately 2.0–5.4 mm in height from coenenchyme. Polyps and coenenchyme encrusted with irregularly sized and colored sand grains. There was no noticeable variation between holotype and other specimens. Preserved in 99.5% ethanol. Original label.

*Paratypes* (all from Japan): Paratype 1. Specimen number USNM 1194728. Collected from Teniya, Nago, Okinawa, at 1 to 2 m by JDR, September 5, 2008. Paratype 2. Specimen number RMNH Coel 40098. Collected from Manza, Onna, Okinawa I., Japan, at 25 m by JDR, 1 September, 2008.

**Type locality**. Japan, Okinawa Prefecture, Okinawa Island: Nago City, Teniya, 26.563832°N, 128.140822°E, in small cracks on reef flat at 1 to 2 m depth, 5 September 2008, J.D. Reimer (JDR) leg.



**Figure 4.** *Neozoanthus uchina* sp. n. in situ. **A** Partially closed polyps showing lack of encrustation at oral end **B** Colonies of two different color morphotypes **C** Close-up of polyps of the same color morphotype as on the left in **B**) **D** Polyps showing variation in oral disk color where the dorsal directive is located. Scales approximately 1 cm. **A** to **D** images taken by Masaru Mizuyama, September 20, 2010, in the lower intertidal zone at Kamomine, Tokunoshima, Kagoshima, Japan, specimens uncollected **E** Colony MISE MO-100 in situ on March 16, 2011 at Tebiro Beach, Amami-oshima, Kagoshima, Japan. Image taken by Masami Obuchi.

Tentacles	Pharynx	Filaments	Column
1. Sp 2. LH 3. SH	4. B 5. Sp 6. LH 7. SH?	8. pM 9. LH 10. SH?	11.H
		8. DM 9. LH 10. SH?	11.H - 10 - 20 - 30 - 40 - 50

# A Neozoanthus caleyi sp. n.

## **B** Neozoanthus uchina **sp. n.**



**Figure 5.** Cnidae of *Neozoanthus caleyi* sp. n. and *N. uchina* sp. n. from the tentacles, pharynx, and filaments showing their relative size. Type abbreviations: **Sp**=spirocysts, **H**=holotrichs, **LH**=large holotrichs, **SH**=small holotrichs, **B**=basitrichs, **SH**?=potential small holotrichs, **pM**=*p*-mastigophores. Size and frequency data are given in Table S1.

**Other material** (all from Japan, coll. JDR unless noted): Teniya, Okinawa I., Okinawa, MISE 545, 546, 549 (n=3), 1-2 m 5 September 2008; Yona, Okinawa I., Okinawa, MISE 560, 13 m, coll. JDR and Takuma Fujii (TF), 24 September 2008; Teniya, Okinawa I., Okinawa, MISE 1092, 1093 (n=2), intertidal - 1 m, 1 July 2008; Tinyuhama, Korijima I., Okinawa, MISE 1115, 1116 (n=2), 24 m, 28 December 2008; Omonawa, Tokunoshima I., Kagoshima, MISE 1400, 9 m, 9 March 2010; San, Tokunoshima I., Kagoshima, MISE 1400, 9 m, 9 March 2010; Zampa, Okinawa I., Okinawa, MISE 1403, at unknown depth, 29 August 2008 (see also Table 2); Tebiro Beach, Amami-Oshima I., Kagoshima, MISE MO-100, 10 m, coll. Masami Obuchi, 16 March 2011.

**Description.** *Size:* Polyps in situ approximately 2.2-5.1 mm in diameter when open, and approximately 2-8.5 mm in height.

*Morphology: Neozoanthus uchina* sp. n. has 32 to 42 (average 38±3.0, n=24 polyps on 9 colonies) conical tentacles. Tentacles are usually shorter than the expanded oral

disk diameter (e.g. 50-80% of oral disk width). Tentacles may be grayish-blue, rust red, or transparent, often with black, white, or fluorescent blue bands or patterning (Figure 4). No bractae are visible, and all specimens were zooxanthellate. Polyps are externally heavily encrusted with sand and other particles of irregular sizes, excepting the oral end, which is free of encrustation and appears a bluish-gray similar to as seen in some *Zoanthus* species. When fully contracted, the sand free oral end is often not visible, and polyps resemble small balls of sand. Polyps extend well clear of reduced or stoloniferous coenenchyme (Figure 4). Oral disks may be a variety of colors, such as light gray-blue, white, rust or deep wine red. Occasionally, white or light blue dots may be seen on the oral disk in regular circular patterns, and the oral opening (mouth) is often white or cream in color. A "skirt" of different coloration (usually white or lighter coloration than remainder of oral disk) covering up to approximately 90 degrees of the oral disk is often seen in the area of the dorsal directive. Colonies consisted of tens to <100 polyps, connected by stolons with no well-developed coenenchyme.

*Cnidae:* Basitrichs and microbasic *p*-mastigophores (often difficult to distinguish), holotrichs (large and small), spirocysts (see Table S1, Figure 5).

**Differential diagnosis.** Differs from *Neozoanthus tulearensis* Herberts, 1972 and *Neozoanthus caleyi* sp. n. with regards to distribution (Ryukyu Archipelago as opposed to Madagascar and southern Great Barrier Reef, respectively), coloration (yellow observed in some *N. caleyi* sp. n.), and tentacle count (*N. tulearensis* = 38 to 44 tentacles, *N. caleyi* sp. n. = average  $33\pm3.9$  tentacles). The two new *Neozoanthus* species' tentacle counts are statistically significant (t-test, p<0.001). Often polyps are much taller (to 8.5 mm) than *N. caleyi* sp. n. (to 3.0 mm), although height ranges overlap (Table 3). The two new *Neozoanthus* species mt 16S rDNA sequences differ by three base pairs (Reimer et al. 2011a).

*Neozoanthus uchina* sp. n. is currently the only partially encrusted zoanthid described from the Ryukyu Archipelago.

**Etymology.** Named for the Okinawan dialect word for Okinawa, "uchina", the prefecture where this species was first found. Noun in apposition.

Habitat, ecology, and distribution. Specimens from the Ryukyu Archipelago were found at depths from the intertidal zone to 25 m. Despite repeated surveys focused on zoanthids, no *N. uchina* sp. n. have been found further north on Yakushima Island or mainland Japan, nor further south in the Miyako and Yaeyama Islands of southern Okinawa, and it may be that this species is limited to a subtropical distribution in the Middle Ryukyu Islands. Additionally, despite surveys, thus far no specimens have been reported from neighboring Taiwan (Reimer et al. 2011d) or the Ogasawara Islands (Reimer et al. 2011b).

*Neozoanthus uchina* sp. n., although not found at many locations surveyed, was locally common, particularly at locations that were characterized by strong currents and some sedimentation, with large coarse sand particles scattered over the bottom or rocks. Preference for such environments may be related to its encrustation patterns. Colonies were almost always found in cracks and holes in rocks partially exposed to light, and usually not in locations completely exposed to light. Most colonies were

relatively small, consisting of tens (not hundreds) of polyps, with polyps spread out and connected by thin stolons (Figure 4).

**Notes.** This species can close its polyps much more rapidly than species of other zooxanthellate zoanthid genera (Reimer pers. obs).

**DNA Sequences.** Originally listed in Table S1 in Reimer et al. (2011a). Cytochrome oxidase subunit I: HM991243-HM991246 Mitochondrial 16S ribosomal DNA: HM991227-HM991229

### Discussion

### Neozoanthidae a valid family?

As stated previously (Reimer et al. 2011a), given *Neozoanthus*' close phylogenetic affiliation with *Isaurus*, a genus of the family Zoanthidae, it is unlikely that Neozoanthidae is a valid family grouping. However, given the unique morphological characters of *Neozoanthus*, as well as a unique mt 16S rDNA indel shared with Hydrozoanthidae, more molecular data from other markers are needed before Neozoanthidae is formally merged into Zoanthidae. These results clearly indicate that at least one of the diagnostic characters for erecting Neozoanthidae by Herberts (1972), sphincter muscle position, does not have utility, as the family was erected based in a large part on an endodermal (macrocnemic) sphincter muscle. As shown by Swain (2010), sphincter muscle position is apparently not diagnostic for higher level (genus, family, suborder) taxonomy in zoanthids, and furthermore, as mentioned in Reimer et al. (2011a), many morphological features in zoanthids, including the presence or absence of sand encrustation and sphincter muscle position can evolve or change relatively rapidly.

#### Ecology of Neozoanthus

Both new species in this study were found in areas notable for their strong currents. Both species, although zooxanthellate, were found in areas somewhat sheltered from direct sunlight, unlike many *Zoanthus* and *Palythoa* spp. The sand encrustation plus *Neozoanthus* species' preference for cracks and overhangs may have led to their lack of discovery in both Australia and Japan until 2008-2009 (Reimer et al. 2011a). Certainly, the species are not "cryptic" in the classic sense, as they have colorful oral disk patterns, but the fact that they had been overlooked until recently demonstrates the lack of attention paid to zoanthids in field surveys.

*Neozoanthus caleyi* sp. n. possesses *Symbiodinium* (=zooxanthellae) of subclade C1 sensu LaJeunesse (2002) (Reimer and Irei unpubl. data), a "generalist" type of *Symbiodinium* known to be sensitive to thermal stress, and the distribution patterns of *N. uchina* sp. n. and *N. caleyi* sp. n. fit well with this symbiont's physiology, as both species were usually not found at depths of <10 m exposed directly to sunlight and colonies in shallower waters were in cracks or crevices that provided shading. Despite the fact that no studies have yet been conducted on their ecology, the presence of both species from shallow to deeper waters in areas of strong currents combined with somewhat long tentacles (*e.g.* compared to *P. heliodiscus* with tentacles only 10% length of oral disk – Ryland and Lancaster 2003) indicates both *Neozoanthus* species may be mixotrophic, obtaining energy from both prey capture and photosymbiotic *Symbiodinium*.

As mentioned in the species' descriptions, for now it appears that both *N. uchina* sp. n. and *N. caleyi* sp. n. have subtropical distributions, as no colonies were found to regions directly north (both species) or directly south (*N. uchina* sp. n.) of their distribution. However, due to their small size and preference for semi-cryptic microhabitats, we cannot discount the possibility that there are further populations of both species that await discovery. Furthermore, it is known there are unidentified *Neozoanthus* species in Indonesia (Reimer & Hoeksema, unpublished data), and specimens are needed to complete work on these.

## Conclusions

Two new species of *Neozoanthus* from the Pacific are formally described, one from the Great Barrier Reef and one from the Middle Ryukyu Islands.

The discovery of these two species (detailed in Reimer et al. 2011a) and their relative commonness at some sampling sites indicates that much work remains to be performed in order to properly understand zoanthid diversity in the Indo-Pacific.

We recommend the utilization of the combination of both molecular results (Reimer et al. 2011a) with the morphological descriptions given in this study for zoanthid identification and description, as seen in many recent studies.

### Acknowledgements

The author thanks the following people at the University of the Ryukyus (UR): Dr. Mamiko Hirose (now Ochanomizu U.) for histology help, and Dr. Frederic Sinniger (now JAMSTEC), Masami Obuchi (now Biological Institute on Kuroshio) and Masaru Mizuyama for specimens, distribution information, and in situ images. On the Great Barrier Reef, the Census of Coral Reef Ecosystems (CReefs) Australia Project, and in particular Dr. Julian Caley and Shawn Smith (both AIMS) are thanked for sampling help, and Gary Cranitch (AIMS) for images. CReefs was a field project of the Census of Marine Life. Dr. Bert Hoeksema (Naturalis Biodiversity Center) and Julian Sprung are thanked for information on *Neozoanthus* in the central Indo-Pacific. This study was funded in part by the Japan Society for the Promotion of Science, and the Rising Star Program and the International Research Hub Project for Climate Change and Coral Reef/Island Dynamics at UR. The third author was supported in part by the Japan Society for the Promotion of Science for Australia were collected under Great Barrier Reef Marine Park Authority permit #G32313.1 and Queensland Fisheries permit # 95152. Two anonymous reviewers' comments greatly improved the manuscript.

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# Appendix

Types, relative abundance and sizes of cnidae in *Neozoanthus uchina* sp. n. and *N. caleyi* sp. n. (doi: 10.3886/zookeys.246.3886.app) File format: Microsoft Word Document (doc).

**Explanation note:** Table S1 Types, relative abundance and sizes of cnidae in *Neozoanthus uchina* sp. n. and *N. caleyi* sp. n.

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**Citation:** Reimer JD, Irei Y, Fujii T (2012) Two new species of *Neozoanthus* (Cnidaria, Hexacorallia, Zoantharia) from the Pacific. ZooKeys 146: 69–87. doi: 10.3897/zookeys.146.3886.app