ZooKeys 337: 1–18 (2013) doi: 10.3897/zookeys.337.5770 www.zookeys.org

RESEARCH ARTICLE



Three new species of cockroach genus Symploce Hebard, 1916 (Blattodea, Ectobiidae, Blattellinae) with redescriptions of two known species based on types from Mainland China

Zongqing Wang^{1,†}, Yanli Che^{1,‡}

I Institute of Entomology, College of Plant Protection, Southwest University, Beibei, Chongqing 400716, China

† http://zoobank.org/B29AEB84-9DB0-4C98-9CE8-FD5BC875771B‡ http://zoobank.org/8ED9AE03-E0EB-4DCE-BE08-658582983CC4

Corresponding author: Zongqing Wang (zqwang2006@126.com)

Academic editor: Jes Rust | Received 11 June 2013 | Accepted 9 September 2013 | Published 30 September 2013 http://zoobank.org/CABA56B3-5F25-45F1-B355-3B28D0032012

Citation: Wang Z, Che Y (2013) Three new species of cockroach genus *Symploce* Hebard, 1916 (Blattodea, Ectobiidae, Blattellinae) with redescriptions of two known species based on types from Mainland China. ZooKeys 337: 1–18. doi: 10.3897/zooKeys.337.5770

Abstract

Three new species of *Symploce* Hebard from China are described: *Symploce sphaerica* **sp. n.**, *Symploce para-marginata* **sp. n.** and *Symploce evidens* **sp. n.** Two known species are redescribed and illustrated based on types. A key is given to identify all species of *Symploce* from mainland China.

Keywords

Insecta, Dictyoptera, Ectobiidae, Episymploce, new species, cockroaches

Introduction

The genus *Symploce* was erected by Hebard in 1916, and it is the most primitive genus of the family Ectobiidae, for related to the earliest fossil species *Pinblattella citimica* according to Vršanský (1997). Princis (1969) lists 109 species of this genus in *Orthop-terorum Catalogus*, among which 23 species were known from China. Asahina (1974) added 2 species named *S. miyakoensis* and *S. okinoerabuensis* from Japan, and in 1979,

he described 2 species S. yayeyamana, S. gigas and 2 subspecies S. gigas okinawana, S. striata wulai; transferred Parasymploce accuminata (Shiraki, 1931) and Ischnoptera furcata Shiraki, 1931 to Symploce as well. Kumar (1975) reported 3 species, S. medleri, S. marshallae, S. distincta, from Africa. Feng and Woo (1988, 1993, 1999) reported S. wulingensis, S. mamillatus, S. guizhouensis species from Southern China; Woo and Feng published two species S. bispota and S. quadripinis, respectively in 1988 and 1992. In 2002, Feng described a species of Symploce, named jianfengensis from China. Izquierdo and Medina (1992) reported one species from Gran Canaria, Spain. Roth (1984a) redefined the generic characters of Symploce and redescribed 9 species from West Indies and a key was given for the males. In the same year, Roth (1984b, 1984c) described 13 species from New Guinea, 5 species and 3 known species from Borneo, Flores, India and Philippines, and defined groups for New Guinea species; in addition, keys were given based on examined males. In 1985a he described 6 species, redescribed 2 known species from Borneo, Sumatra and West Malaysia, a key was given to the males; In 1985b he described four new species and redescribed five species, and brief descriptions of three known species were given from Bey-Bienko's original description; In 1986a, b he described 8 species and redescribed 14 known species from African and divided them into four groups, a key to males is given. In 1987a Roth redescribed 6 species and 4 subspecies from Taiwan and Japanese Islands. A key to males and a worldwide distribution checklist of all species of the genus were given. Roth (1991) transferred Symploce triramosa (Saussure, 1869) and Symploce tchadiana Roth, 1987 to Carbrunneria Princis and Supella Rehn respectively. Roth (1997) synonymized 2 species, S. bicolorata Roth, 1985 with Haplosymploce montis (Shelford, 1906) and S. ferruginea Roth, 1985 with Haplosymploce nigra (Hanitsch, 1928) and transferred 12 species to Episymploce. And in 1999, Roth described a species, Symploce stupida. During the period of revising the genus Symploce, Roth reassigned many species; among them, most Chinese species listed in Princis' Catalogue (except S. striata) were related and transferred to Episymploce Bey-Bienko. Subsequently Roth (2003) transferred 2 species, namely, S. guizhouensis and S. mamillatus, described by Feng and Woo (1988) to Episymploce based on the drawings of the supra-anal and subgenital plates. Up to now, only 65 species were included in this genus worldwide (Beccaloni 2007), of which 8 species are from China (including Taiwan) but just 5 species are distributed in mainland China, one of which, Symploce dimorpha has been transferred into Episymploce by Roth (1987b).

In the present paper, after checking the specimens deposited in the Insect Collection of Southwest University (SWU), 3 species new to science are described and illustrated and 2 known species based on types are redescribed.

Materials and methods

Terminology used in this paper is mainly according to Roth (2003). Measurements are based on specimens examined. The genital segments of the examined specimens were macerated in 10% NaOH and observed in glycerin jelly using a Motic K400

stereomicroscope. All the drawings were made with the aid of a Motic K400 stereomicroscope. Photographs of the specimens were made using a Canon 50D plus a Canon EF 100mm f/2.8L IS USM Macro lens with the aid of the Helicon Focus software. The type specimens are deposited in the Insect Collection of Southwest University, Beibei, China.

Taxonomy

Symploce Hebard, 1916 http://species-id.net/wiki/Symploce

Symploce Hebard, 1916: 355; Roth 1984a: 26.

Type species. Blatta capitata Saussure, 1862.

Diagnosis. Tegmina and wings fully developed or more or less reduced. Radius vein of hind wing unbranched or branched near the middle; cubitus vein straight to distinctly curved with 1-6 complete and 0-6 incomplete branches; apical triangle absent or present, small or large. Specialization on the male abdominal tergum varies considerably in position and shape or is completely unmodified. Supra-anal plate symmetrical, rarely asymmetrical. Subgenital plate weakly to strongly asymmetrical, rarely symmetrical, with various highly specialized styli. Anteroventral margin of front femur usually Type A₃, rarely Type B₃ or between Type A and B. Tarsal claws symmetrical, unspecialized.

Note. It is proposed that *S. quadrispinis* Feng & Woo, *S. stellatus* Feng & Woo, 1999 and *S. wulingensis* Feng & Woo should be transferred to *Episymploce*. Plus one known species, *S. jianfengensis* Feng, 2002, which was not recorded in the catalogue by Beccaloni (2007), also should be transferred to *Episymploce* (Wang at al. in press).

Distribution. Oriental, Australian, African and Palaearctic Regions.

Key to species of Symploce from Mainland China (Males)

The first abdominal tergum unmodified2
The first abdominal tergum modified
Pronotum with two black longitudinal, irregular maculae centrally, supra- anal plate linguiform
Pronotum with two small V- shaped brown spots centrally, supra-anal plate trapeziform
Seventh abdominal tergum modified with a pair of large depressions where some setae are situated
Seventh abdominal tergum unmodified4
Pronotum pitch black, lateral board brown S. paramarginata sp. n.
Pronotum yellowish brown with shallow U-shaped dark brown macula near base

1. Symploce torchaceus Feng & Woo, 1999

http://species-id.net/wiki/Symploce_torchaceus Figs 1–2, 11–19

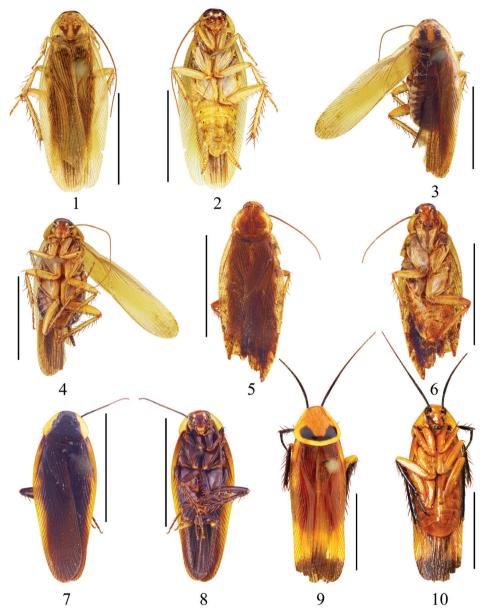
Symploce torchaceus Feng & Woo, 1999: 51.

Description. Length, male, pronotum: length × width: 3.2×3.8 mm, tegmen: 15mm, overall length: 18mm. Body brown (Fig. 1). Head brown with a dark brown band on disc, which is wide and short. Face brown with a dark brown band. Maxillary palpomus brown and apex dark brown (Fig. 2). Pronotum pale brown with a pair of V-shaped rufous maculae in centre (Figs 1, 11). Tergum (except T₁) with a dark brown spot on each side, and a dark brown stripe on disk.

Vertex with interocular width slightly less than ocellus width, distinctly narrower than distance between antennal sockets. Fourth and fifth maxillary palpomus of same length, both distinctly shorter than third (Fig. 2). Pronotum elliptical and width longer than length, with anterior margin nearly truncate and hind margin slightly produced in the middle (Fig. 11). Tegmen and hind wing well developed, entirely covering abdomen (Fig. 1). Tegmen narrow and long; radius vein with apical posterior branch, which terminates at the apical margin, and with 2 small branches, one of them branched; median vein with 2 branches (Fig. 12). Radius vein of hind wing branched beyond the middle and the branches bifurcated again near apex, median vein slightly curved and simple; cubitus slightly curved with 3 complete and 3-4 incomplete branches, triangular apical area reduced and small (Fig. 13). Anteroventral margin of front femur type A₃, pulvilli present on 4 proximal tarsomeres, tarsal claws symmetrical and unspecialized, and arolia present. The 1st abdominal tergum (T₁) unmodified, T₇ specialized with some setae on disc; lateral plates of T₉ similar with hind margin rounded and unspecialized (Fig. 14).

Male genitalia. Supra-anal plate (Fig. 15) in ventral view symmetrical and nearly trapeziform, hind margin nearly straight, left side with 3 small spines and right side with 2 small spines. Right and left paraprocts (Fig. 15) obviously asymmetrical, left one dendritic and apices tapering, right one with apex scattered with many fine spines and 1 branch near base, which resembles an antler. Subgenital plate (Fig. 16) weakly asymmetrical and hind margin slightly produced in the middle, left side concave at apical half and right side curved; two styli dissimilar and lying at apex, left style large which is similar to one torch directed laterad, and with 3 small teeth at outer margin near base, right style smaller and with 3 acute spines at apex. Hook of left phallomere with sclerotized portion very small, on left side, slender and with V-shaped incision (Fig. 17). Median phallomere (Fig. 18) long and lanciform with apex tapering, right phallomere (Fig. 19) skilletlike with a twist of sclerite.

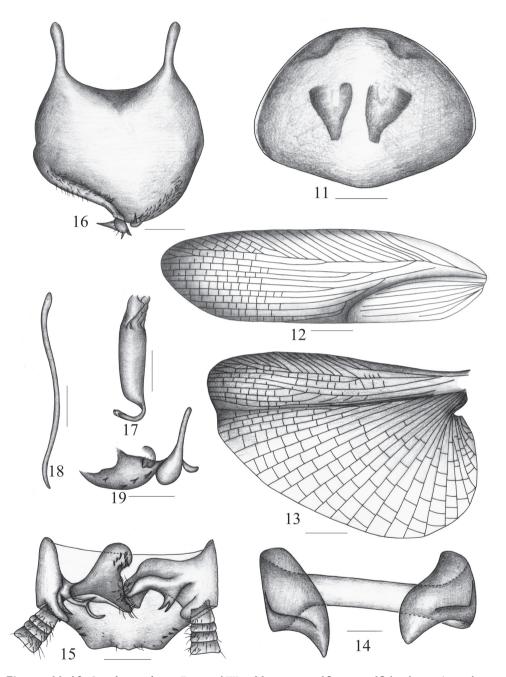
Materials examined. One male (holotype), China: Fujian Prov., Mt. Wuyishan, 10 July 1982, coll. Feng Xia; one male (paratype), China: Hainan Prov., Mt. Jian-fengling, Tianchi, 21 March 1983, coll. Shaoying Liang; one male, China: Hainan



Figures 1–10. 1–2. *Symploce torchaceus* Feng & Woo, male: 1 holotype, dorsal view 2 same, ventral view 3–4 *Symploce bispot* Feng and Woo, male: 3 holotype, dorsal view 4 same, ventral view 5–6 *Symploce sphaerica* sp. n., male: 5 holotype, dorsal view 6 same, ventral view 7–8 *Symploce paramarginata* sp. n., male: 7 holotype, dorsal view 8 same, ventral view 9–10 *Symploce evidens* sp. n., male: 9 holotype, dorsal view 10 same, ventral view. Scale bars = 1.0 cm.

Prov., Mt. Jianfeng, 12 March 1982, coll. Maobin Gu; four males, China: Hainan Prov., Mt. Jianfengling, 25 March 1985, coll. Zhiqing Chen.

Distribution. China (Fujian, Hainan).



Figures 11–19. *Symploce torchaceus* Feng and Woo **11** pronotum **12** tegmen **13** hind wing (vannal region damaged) **14** abdominal tergum 9 and lateral plates, ventral view **15** supra-anal plate and paraprocts, ventral view **16** subgenital plate, dorsal view **17** hook-like phallomere **18** median phallomere **19** right phallomere. Scale bars = 1.0 mm (Fig. 11), 2.0 mm (Figs 12–13), 0.5 mm (Figs 14–19).

2. Symploce bispot Feng & Woo, 1988

http://species-id.net/wiki/Symploce_bispot Figs 3–4, 20–27

Symploce bispot Feng & Woo, 1988: 31.

Description. Length, male, pronotum: length × width: 4.0 × 5.0mm, tegmen: 19.0mm, overall length: 19.5–20.0mm. Body brown (Fig. 3). Head brown with area between and beyond ocellus reddish brown. Occiput region pale yellow. Antenna brown with base yellowish brown and apex dark brown. Maxillary palpomus yellowish brown and apical segment blackish brown (Fig. 4). Pronotum brown with dark brown maculae on disc (Figs 3, 20). Tergum reddish brown and both sides pale brown.

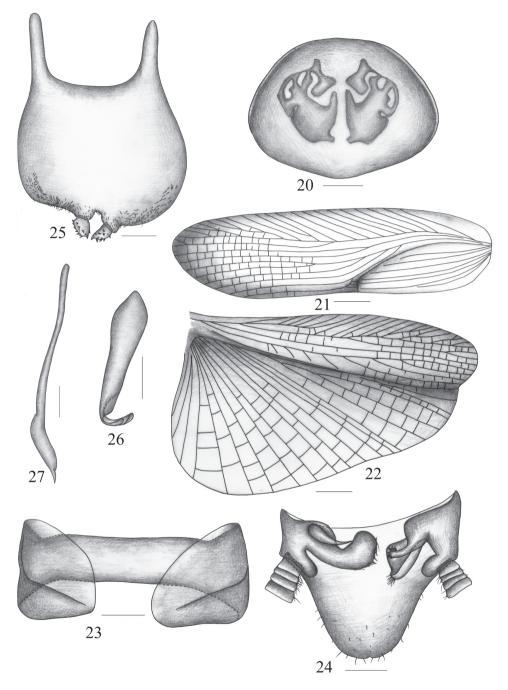
Vertex with interocular width slightly narrower than ocellus width, distinctly less than distance between antennal sockets. Fourth and fifth maxillary palpomus about same length, both distinctly shorter than the third (Fig. 4). Pronotum more or less elliptical, with anterior margin nearly truncate and hind margin obviously produced in the middle; pronotum with irregular maculae as Fig. 20 on disc. Tegmen and hind wing well developed, entirely covering abdomen. Tegmen narrow and long; radius vein with apical posterior branch, which bifurcated at apical part; median vein branched near the middle (Fig. 21). Radius vein of hind wing branched around the middle and apex of branch bifurcated, median vein simple; cubitus vein with 3 complete and 4 incomplete branches, triangular apical area small (Fig. 22). Anteroventral margin of front femur Type A₃, pulvilli present on 4 proximal tarsomeres, tarsal claws symmetrical and unspecialized, and arolia present. T₁, T₇ unmodified, 1 semitransparent spot present on disc of T₃, T₄, T₅, T₆. Lateral plate of T₉ about similar and with hind margin unproduced and unspecialized (Fig. 23).

Male genitalia. Supra-anal plate in ventral view symmetrical and about linguiform (Fig. 22). Right and left paraprocts (Fig. 24) evidently asymmetrical, left one dendritic and apices tapering, right one with apex scattered with lots of fine spines and 1 branch near base, which resembles an antler. Subgenital plate (Fig. 25) slightly asymmetrical, hind margin slightly concave in the middle and produced, where with two styles lying at both sides; style nearly elliptical and with small spines at outer margin. Hook of left phallomere with sclerotized portion very small, on left side, slender and with V-shaped incision (Fig. 26). Median phallomere long and lanciform, with apex tapering and rarely curved near apex (Fig. 27).

Female similar to male. Supra-anal plate triangular, subgenital plate broad and round.

Materials examined. One male (holotype), China: Xizang Prov., 23 November 1983, coll. Yinheng Han; one female (paratype), same data as holotype; one male, China: Xizang Prov., Beibeng, 13 December 1977, coll. Jianshe Wu.

Distribution. China (Xizang).



Figures 20–27. *Symploce bispot* Feng & Woo (right phallomere missing) **20** pronotum **21** tegmen **22** hind wing **23** abdominal tergum 9 and lateral plates, ventral view **24** supra-anal plate and paraprocts, ventral view **25** subgenital plate, dorsal view **26** hook-like phallomere **27** median phallomere. Scale bars = 1.0 mm (Fig. 20), 2.0 mm (Figs 21–22), 0.5 mm (Figs 23–27).

3. *Symploce sphaerica* sp. n.

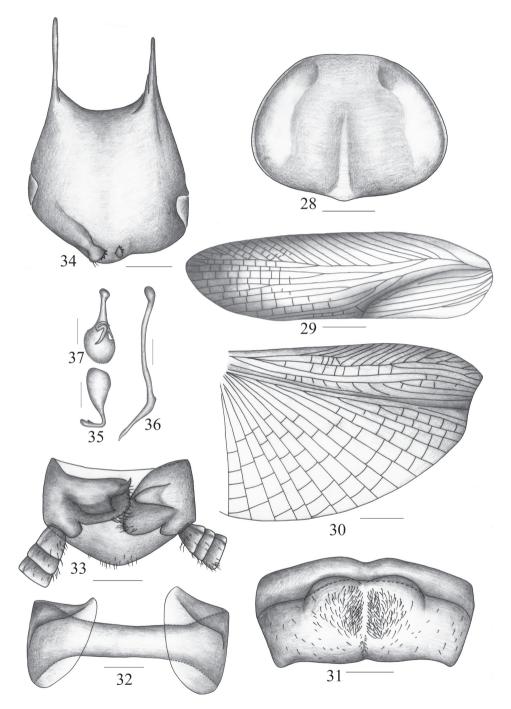
http://zoobank.org/54B15C64-8C73-4A8B-A6AA-185AB2107546 http://species-id.net/wiki/Symploce_sphaerica Figs 5–6, 28–37

Description. Length, male, pronotum: length \times width: 3.0 \times 4.0mm, tegmen: 13.0mm, overall length (including tegmen): 15.5mm. Body yellowish brown (Fig. 5). Vertex brown. Ocellar spot pale yellow. Antenna with base yellowish brown and apex dark brown. Maxillary palpomus with fourth and fifth segments dark brown, others yellowish brown (Fig. 6). Pronotum with reddish brown maculae at disk, lateral borders and anterior margin yellowish brown (Figs 5, 28).

Vertex with interocular space slightly less than ocellus width and distinctly narrower than distance between antennal sockets. Third and fifth maxillary palpomus about same length, both distinctly longer than the fourth (Fig. 6). Pronotum with hind margin slightly produced in the middle. Tegmen with apical posterior branch behind the radius vein, which bifurcated at apical part; median vein with two branches, the longer one further bifurcated (Fig. 29). Hind wing with radius vein branched not over the middle, median vein simple, cubitus veins slightly curved with 2 complete and 2 incomplete branches, and triangular apical area reduced and small (Fig. 30). Anteroventral margin of front femur type B_3 , pulvilli present on 4 proximal tarsomeres, tarsal claws symmetrical and unspecialized, and arolia present. First abdominal tergum (T_1) specialized with a tuft of agglutinated hair directed caudad; seventh abdominal tergum (T_7) specialized with a pair of large depressions where some setae are situated (Fig. 31); and ninth abdominal tergum (T_9) with lateral plates similar, not produced and unspecialized, but the right one slightly narrower than the left (Fig. 32).

Male genitalia. Male supra-anal plate (Fig. 33) symmetrical with some setae scattered in ventral view, hind margin nearly triangular and slightly concave in the middle. Right and left paraproct dissimilar and unspecialized (Fig. 33). Subgenital plate (Fig. 34) asymmetrical, apex of lateral borders thickened, middle of hind margin slightly produced where two preapical styli are lying; the styli dissimilar, the left one larger, nearly spherical and apex with 3–4 teeth, the right smaller, nearly cylindrical and apex with 2 processes (Fig. 34). Hook of left phallomere with sclerotized portion very small, on left side, hook portion slender and with V-shaped and subapical incision (Fig. 35); median phallomere slightly curved at apical half and apex spine-like and acute (Fig. 36); right phallomere irregular sclerite (Fig. 37).

Materials examined. *Holotype*, male, China: Guangxi Prov., Jinxiu, Mt. Shengtangshan, 18 October 1999, coll. Mingai Gao. *Paratypes*, one male, China: Guangxi Prov., Jinxiu, Luoxiang, 18 October 1999, coll. Xingke Yang; one male, China: Guangxi Prov., Jinxiu, Mt. Shengtangshan, 18 October 1999, coll. Mingai Gao; one female, China: Guangxi Prov., Jinxiu, Mt. Shengtangshan, 18 October 1999, coll. Xuezhong Zhang.



Figures 28–37. *Symploce sphaerica* sp. n. **28** pronotum **29** tegmen **30** hind wing **31** abdominal tergum 7, dorsal view **32** abdominal tergum 9 and lateral plates, ventral view **33** supra-anal plate and paraprocts, ventral view **34** subgenital plate, dorsal view **35** hook-like phallomere **36** median phallomere **37** right phallomere. Scale bars = 1.0 mm (Fig. 28), 2.0 mm (Figs 29–30), 0.5 mm (Figs 31–37).

11

Remarks. The species is similar to *Episymploce mamillatus* (Feng & Woo), but can be distinguished as follows: 1) hind margin of anal plate with indistinct incision (hind margin of anal plate with distinct V-shaped concavity in *E. mamillatus*); 2) right and left paraprocts unspecialized (paraprocts specialized in *E. mamillatus*); 3) subgenital plate with two nearly spherical styli (styli spine-like in *E. mamillatus*). And the species differs from all other *Symploce* spp. by the special styli.

Etymology. The specific epithet "sphaericus" is derived from Latin, which refers to the left style being nearly spherical (dissimilar from the right style).

4. Symploce paramarginata sp. n.

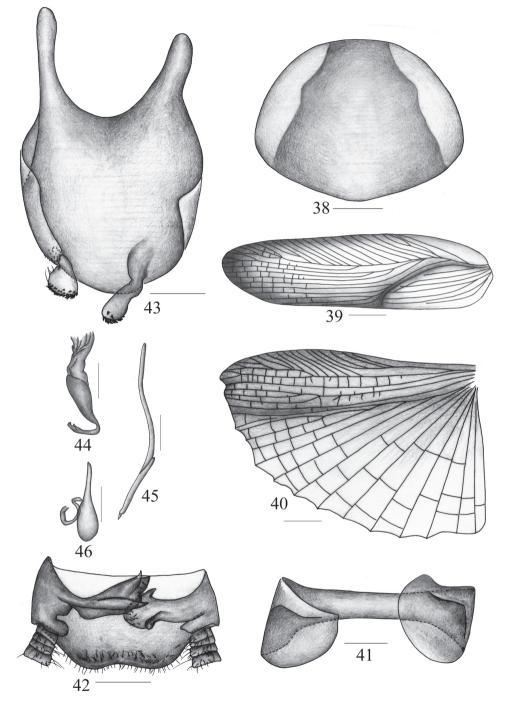
http://zoobank.org/66C616E4-5D66-428F-9F29-6C94E374B0EF http://species-id.net/wiki/Symploce_paramarginata Figs 7–8, 38–46

Description. Length, male, pronotum: length \times width: 3.5 \times 4.0mm; tegmen 13.0mm; overall length including tegmen 14.5–16.5 mm. Body dark brown (Fig. 7), vertex and ocellar spot reddish brown and face black. Antenna and maxillary palpomus dark brown (Fig. 8). Pronotum with disc black and lateral borders orange (Figs 7, 38). Tegmina brown, legs reddish brown and abdomen reddish or blackish brown (Fig. 7).

Vertex with interocular space distinctly wider than distance between antennal sockets, and ocellus width slightly less than distance between antennal sockets. Third and fourth maxillary palpomus about same length, both distinctly longer than the fifth. Tegmen with apical posterior branch behind radius vein, and the branch bifurcated, one of the second division further branched (Fig. 39). Hind wing with radius vein branched over the middle and the branches bifurcated at apex; both median and cubitus veins slightly curved, median vein simple, but cubitus veins with 2-3 complete and 1-4 incomplete branches, and triangular apical area reduced and small (Fig. 40). Anteroventral margin of front femur type B_3 , pulvilli present on 4 proximal tarsomeres, tarsal claws symmetrical and unspecialized, and arolia present. First abdominal tergum (T_1) specialized with a tuft of hair; seventh abdominal tergum (T_7) unmodified; and ninth abdominal tergum (T_9) with lateral plates similar, not produced and unspecialized (Fig. 41).

Male genitalia. Supra-anal plate (Fig. 42) symmetrical and trapeziform, hind margin slightly concave. Right and left paraproct (Fig. 42) dissimilar, right paraproct with spine-like process on the base. Subgenital plate asymmetrical, left side thickened and upturned, right side slightly upturned; two styli similar and padlike, both apices with minute spines (Fig. 43). Hook of left phallomere large and robust at apex and slender at hook portion, on left side, with V-shaped and subapical incision (Fig. 44); median phallomere distinctly curved at middle and lanciform, and apex spine-like and acute (Fig. 45). right phallomere (Fig. 46) same to that of *Symploce torchaceus*.

Female is similar to male; supra-anal plate symmetrical and trapeziform; subgenital plate simple and hind margin round.



Figures 38–46. *Symploce paramarginata* sp. n. **38** pronotum **39** tegmen **40** hind wing **41** abdominal tergum 9 and lateral plates, ventral view **42** supra-anal plate and paraprocts, ventral view **43** subgenital plate, dorsal view **44** hook-like phallomere **45** median phallomere **46** right phallomere. Scale bars = 1.0 mm (Fig. 38), 2.0 mm (Figs 39–40), 0.5 mm (Figs 41–46).

Materials examined. *Holotype*, male, China: Guizhou Prov., Maolan, 24 May 1998, coll. Zizhong Li. *Paratypes*, two males, China: Guangxi Prov., Huaping, 6 November 1963, coll. Jikun Yang; one male, China: Hainan Prov., 25 October 1997, coll. Maofa Yang; one male, China: Guangxi Prov., Jinxiu, 13 May 1999, coll. Xingke Yang; two females, China: Guangxi Prov., Napo, 19 October 2000, coll. Wenzhu Li & Jun Chen; two females, China: Guangxi Prov., Cheng Fang, 25 November 1999, coll. Yanzhou Zhang.

Remarks. The new species superficially resembles *Episymploce marginata* Bey-Bienko, but can be distinguished from the latter by: 1) seventh abdominal tergum unmodified, the latter with two broad sockets covered with hair, 2) subgenital plate with two appendages which is not bifurcated, the latter with apex of appendage bifurcated.

Based on type of vein, the unmodified seventh tergum and symmetrical supra-anal plate, this species should be placed in *Symploce*, and the species is different from other species in this genus for it's dark brown colors and special macula on pronotum.

Etymology. The Latin word "paramarginata" refers to the superficial resemblance of this species to *Episymploce marginata* Bey-Bienko.

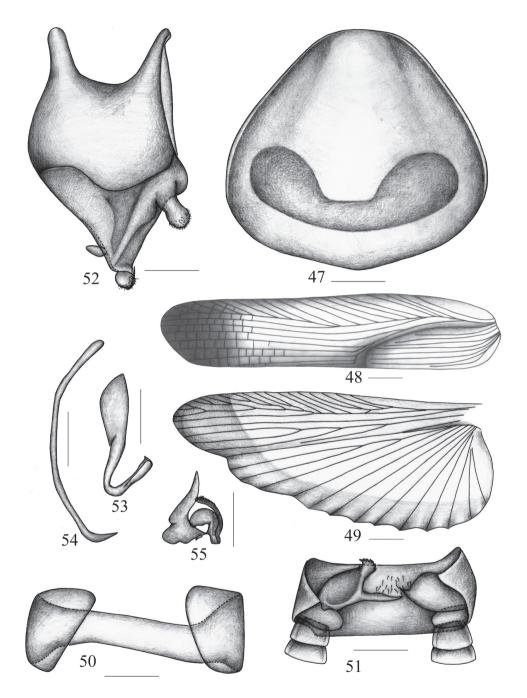
5. Symploce evidens sp. n.

http://zoobank.org/CFD981C4-9590-4294-BC9A-C73EDABF8DDF http://species-id.net/wiki/Symploce_evidens Figs 9–10, 47–58

Description. Length, male, pronotum: length × width: 5.5×6.0 mm, tegmen 18.5–19.0mm; overall length (including tegmen) 24.0mm. Body yellowish brown (Figs 9, 56–58). Antenna pitch-black except apex of flagellum reddish brown. Labrum, maxillary palpus and labial palpus pitch-black. Pronotum yellowish brown with dark brown maculae at disc (Figs 9, 47). Tegmen pale reddish brown and apex blackish brown (Figs 9, 56–58); wing pale brown with inner and apical margin blackish brown (Fig. 49). Legs yellowish brown, tibiae and tarsi blackish brown (Fig. 10).

Vertex with interocular space wider than distance between antennal sockets, and ocellus width about same as distance between antennal sockets. Third and fourth maxillary palpomus about same length, both distinctly longer than the fifth. Pronotum nearly trapezoid with shallow U-shaped macula near base, and hind margin distinctly curved (Figs 9, 47, 56–58). Tegmen with apical posterior branch of radius vein unbranched; median vein also unbifurcated (Fig. 48). Hind wing with radius vein branched beyond the middle, median vein simple, cubitus with 2 complete and 3 incomplete branches, and triangular apical area reduced and small (Fig. 49). Middle of the first abdominal tergite elevated, with a tuft of hair directed forward, T7 and T9 unmodified (Fig. 50).

Male genitalia. Supra-anal plate (Fig. 51) symmetrical, hind margin truncate and turning ventrad. Paraprocts dissimilar, right one bearing a long spine-like process and a hand-like process on distad; left one with a spine-like process distad (Fig. 51). Sub-



Figures 47–55. *Symplace evidens* sp. n. **47** pronotum **48** tegmen **49** hind wing **50** abdominal tergum 9 and lateral plates, ventral view **51** supra-anal plate and paraprocts, ventral view **52** subgenital plate, dorsal view **53** hook-like phallomere **54** median phallomere **55** right phallomere. Scale bars = 1.0 mm (Fig. 47), 2.0 mm (Figs 48–49), 0.5 mm (Figs 50–55).



Figures 56–58. *Symploce evidens* sp. n. in Mountain Qixianling, Baoting County, Hainan Province, 2 May 2013 (photographs by Keliang Wu).

genital plate (Fig. 52) asymmetrical, hind margin produced in the middle, right stylus arising at apex and left stylus spine-like directed downwards near base of right one, right side with an irregular sclerite with apex is serrated. Hook-like phallomere on the left (Fig. 53), median phallomere with tapering apex (Fig. 54); right phallomere with lots of irregular sclerites and one of them with slim setae (Fig. 55).

Female is similar to male. Supra-anal plate symmetrical, triangular; subgenital plate broad, hind margin slightly arced, near lateral sides concaved.

Materials examined. *Holotype*, male, China: Hainan Prov., Mt. Jianfengling, Tianchi, 8-10 October 1964, coll. Hui Ren. *Paratypes*, one male, China: Hainan Prov., Mt. Jianfengling, 22 December 1982, coll. Zhiqing Chen; one female, China: Hainan Prov., Mt. Diaoluoshan, 12 October 1965, coll. Sikong Liu; one female, China: Fujian Prov., Huangken, 17 November 1980, coll. Bangkan Huang; one female, China: Hainan Prov., Mt. Jianfengling, Tianchi, 28 December 1983, coll. Tianyuan Luo; one male and one female, China: Hainan Prov., Mt. Jianfengling, China: Hainan Prov., Mt. Jianfengling, 27 April 2013, coll. Shunhua Gui; one male, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll. Yan Shi; two females, China: Hainan Prov., Mt. Qixianling, 2 May 2013, coll.

Remarks. This species resembles *Symploce striata*, but can be differed by the following characters: 1) pronotum with shallow U-shaped macula, without macula in *S. striata*; 2) apex of tegmen with blackish brown macula, without macula in *S. striata*; 3) terminal half of subgenital plate distinctly triangular, trapezoidal in *S. striata*.

Etymology. The specific name is derived from the Latin adjective "evidens", referring to the pronotum with an evident shallow U-shaped macula.

Discussion

The small order- Blattodea has been investigated and researched for more than two hundred years (Wang and Che 2010). Why are there still more unknown species? The main reason might relate to diversity of habitats, and the methods we used in investigation. In the past time we usually collected cockroaches by searching the habitat for Blattodea by day or at night, especially under dry branches and fallen leaves, or rotting logs, and before obtaining specimens periods of observation may be preceded. This collection tends to be more specific and less ineffective. Passive collecting, such as light traps, also tends to be ineffective. For instance, the most cockroach specimens were obtained by light traps, but species concerned was few; occasionally some were sampled by sweep net, similarly, a narrow variety of cockroaches could be collected.

Traditional sweep at ground litter by day well known by most of cockroach researchers, few collectors can search for cockroaches at night with the aid of highlight torches or cap-lamp, and fewer has a wide knowledge that cockroaches also live on the trees. To everyone's surprise, Blattodea represented most of the biomass in the canopy (Basset 2001). We have obtained a great number of cockroach specimens by night searching and canopy fogging in recent years. One new species of this paper, *Symploce evidens* sp. n., only several specimens were acquired by traditional collection after years of effort; but by night searching, we have got a large number of specimens of this species (some soaked in alcohol are excluded) and ecological photos (Figs 56–58) have also been taken successfully. They are mostly secretive and typically ground-dwelling insects that hide by day in cracks and crevices, or under stones (Wang ZQ, pers. obs.). Although having tegmina and well-developed wings, *Symploce evidens* sp. n., usually like to crawl on the leaves and fly slowly at night; even frightened by light or sound, they will not flee in panic like *Periplaneta americana* Linnaeus (Wu KL, pers. obs.).

Adult cockroaches usually have two sets of wings. The tegmina are somewhat sclerotized; while the hindwings are membranous, and generally wider than tegmina. It is generally believed that cross veins play an important role in supporting and reinforcement of the hindwings. Cross veins are normally present throughout most of the wings of cockroaches, and it is only in certain of the more specialized forms such as the Ectobiinae, Anaplectinae and *Oulopteryx* in Corydiidae that they are reduced to a number that can be used in classification (Rehn 1951).

Acknowledgements

We are sincerely grateful to Prof. J. R. Schrock (Department of Biological Sciences, Emporia State University, USA) for revising the manuscript. This study is supported by the Natural Sciences Foundation of Project of CQ CSTC (2010BB5140), the National Natural Sciences Foundation of China (30900146, 31093430) and also partly by the Fundamental Research Funds for the Central Universities (XDJK2012B025, XDJK2013B013).

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RESEARCH ARTICLE



On the occurrence of the fireworm *Eurythoe complanata* complex (Annelida, Amphinomidae) in the Mediterranean Sea with an updated revision of the alien Mediterranean amphinomids

Andrés Arias¹, Rômulo Barroso^{2,3}, Nuria Anadón¹, Paulo C. Paiva⁴

l Departamento de Biología de Organismos y Sistemas (Zoología), Universidad de Oviedo, Oviedo 33071, Spain 2 Pontificia Universidade Católica do Rio de Janeiro, Rio de Janeiro, Brazil 3 Museu de Zoologia da Unicamp, Campinas, SP, Brazil 4 Departamento de Zoologia, Instituto de Biologia, Universidade Federal do Rio de Janeiro (UFRJ), Rio de Janeiro, RJ, Brasil

Corresponding author: Andrés Arias (ariasandres.uo@uniovi.es)

Academic editor: C. Glasby | Received 17 June 2013 | Accepted 19 September 2013 | Published 30 September 2013

Citation: Arias A, Barroso R, Anadón N, Paiva PC (2013) On the occurrence of the fireworm *Eurythoe complanata* complex (Annelida, Amphinomidae) in the Mediterranean Sea with an updated revision of the alien Mediterranean amphinomids. ZooKeys 337: 19–33. doi: 10.3897/zookeys.337.5811

Abstract

The presence of two species within the *Eurythoe complanata* complex in the Mediterranean Sea is reported, as well as their geographical distributions. One species, *Eurythoe laevisetis*, occurs in the eastern and central Mediterranean, likely constituting the first historical introduction to the Mediterranean Sea and the other, *Eurythoe complanata*, in both eastern and Levantine basins. Brief notes on their taxonomy are also provided and their potential pathways for introduction to the Mediterranean are discussed. A simplified key to the Mediterranean amphinomid genera and species of *Eurythoe* and *Linopherus* is presented plus an updated revision of the alien amphinomid species reported previously from the Mediterranean Sea. A total of five exotic species have been included; information on their location, habitat, date of introduction and other relevant features is also provided.

Keywords

Alien polychaetes, cryptic species, Gibraltar Strait, Lessepsian migrant

Introduction

Introductions of alien species are threatening the economic and ecological well-being of marine ecosystems worldwide. The impacts of alien species on their new environments include alterations of established food webs, importation of new diseases or parasites, competition with native species for food and space, and even changing gene pools (Occhipinti-Ambrogi et al. 2011; Cosentino and Giacobbe 2011; Arias et al. 2013a, 2013b; Çinar 2013). Invaders are able to modify the receiving ecosystems, restructuring the ecological relations within communities, altering evolutionary processes and causing dramatic changes in native populations. Over 80% of alien polychaete species recorded to date in the Mediterranean Sea come from the Red Sea and the Indo-Pacific (Çinar 2013), presumably reaching the Mediterranean through the Suez Canal and being considered as Lesseptian migrants or Erythrean species (Por 1978). The remaining ~20% originate from the Atlantic Ocean and were introduced to this region mainly via "shipping" (Çinar 2013). In recent decades, the rate of polychaete invasions has exponentially increased and currently in the Mediterranean Sea the number of alien polychaete species is roughly 100 (Zenetos et al. 2012; Çinar 2013).

Amphinomidae is a well-known family of polychaetes that is globally distributed, reaching its highest diversity in shallow tropical and subtropical waters (Kudenov 1995) and occurring at all depths, including abyssal areas (Kudenov 1993). Large tropical species of amphinomids are normally colourful and commonly referred as "fireworms" with hollow calcareous harpoon-type chaetae containing complanine, a trimethylamine compound that cause intense irritation on skin after penetrating the skin of anyone handling them roughly (Kudenov 1993, 1995; Nakamura et al. 2008). The parapodia are biramous with dense bundles of chaetae. The notopodium bears a single true dorsal cirrus (lateral cirrus) and some species may have a second accessory dorsal cirrus (branchial cirrus). The neuropodium has a single ventral cirrus. Besides having calcareous instead of chitinous chaetae, as present in other polychaetes, most amphinomids and other members of the Amphinomida have well-developed nuchal organs known as caruncles, which extend back mid-dorsally for several segments (Kudenov 1995, Rouse and Pleijel 2001).

Shallow water forms play an important ecological role mainly in rocky and coral reef environments, where species such as *Hermodice carunculata* (Pallas, 1766) are major predators of both soft corals (Alcyonacea) and hard corals (Scleractinia) (Ott and Lewis 1972, Vreeland and Lasker 1989). Furthermore, *H. caranculata* is known to act as reservoir and vector of pathogens associated with coral bleaching (Sussman et al. 2003). Another common shallow-water species is *Eurythoe complanata* (Pallas, 1766), which has been traditionally considered as having a wide circumtropical distribution. Nevertheless, recently it was demonstrated that *E. complanata* is actually a species complex. The phylogeographic analysis performed by Barroso et al. (2010) identified three closely related species forming a species complex: two species (one from eastern Pacific and the other from the Atlantic) are morphologically identical and fit the description

of *E. complanata*; and the third one, slightly morphologically different from the others, corresponds to the species *E. laevisetis*. Thereby, we are here proposing the term 'morphospecies' to refer to *E. complanata* and *E. laevisetis*, a concept that will be explored in the discussion below. Recently *E. cf. complanata* was also reported from the eastern and central Mediterranean (Barroso et al. 2010, Arias et al. 2013a respectively) but its presence in the Mediterranean Sea was questioned (Zenetos et al. 2010, 2012). Therefore, in order to elucidate the current status of this species complex in the Mediterranean and update its taxonomy, specimens previously identified as *E. complanata* collected from the central and eastern Mediterranean were morphologically re-examined, taking into account the new data for this species complex. Additionally, an updated key to currently known genera and five alien species in Mediterranean Amphinomidae is included.

Methods

Field collections were made along the Maltese Islands, Central Mediterranean, on hard substrata from the shallow subtidal rocky areas at Cirkewwa Harbour (35°59'N, 14°19'E) and St. Julian's Bay (35°55'N, 14°29'E) in March 2011 (Figure 1). Large specimens were randomly removed by a swift hand motion. Small specimens were collected using grabs and screened using a 1 mm mesh sieve. The worms were removed from the residue under a stereomicroscope. Then, all specimens were relaxed in MgCl₂ isotonic with seawater, fixed in 10% formaldehyde solution, rinsed in fresh water and finally transferred to 70% ethanol. Photographs were taken using a stereomicroscope Nikon SMZ-1000 equipped with a digital camera; before photography, specimens were stained with lithic carmine solution. Lithic carmine staining increased the contrast of some morphological structures, such as caruncle, branchiae, parapodial lobes and cirri. Glycerol slides of parapodial sections, examined under a compound light microscope Leica DM 2500, were used for the detailed examination of chaetal morphology and distribution.

The examined material was deposited at the Invertebrate Collection of the Department of Biology of Organisms and Systems (BOS) of University of Oviedo. Detailed location data is given below in the 'Material examined' sections of the respective species. The number of specimens in each sample is given in parentheses after the museum abbreviation and registration number. Furthermore, preserved specimens identified as *E. complanata* from the Gibraltar Strait, eastern Mediterranean (deposited in the MNCN), and the coasts of Atlit, Israel (deposited in the BMNH), were re-examined.

Additionally, comparative material was also studied: *Eurythoe laevisetis* Fauvel, 1914: São Tomé Island: IBUFRJ 0545; *Eurythoe* cf. *laevisetis*: Sal Island (Cape Verde): BOS-Amp1; Gran Canary (Canary Islands): BOS-Amp2; *Eurythoe complanata*: Bocas del Toro, Panamá (Caribbean): IBUFRJ 0542. Red Sea (unknown locality): BMNH 1923.3.20.8.

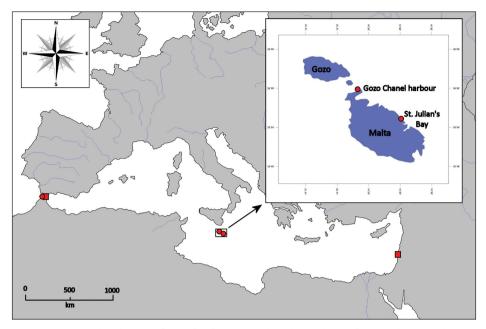


Figure 1. Current distribution of *Eurythoe laevisetis* (red circles) and *Eurythoe complanata* (red squares) along the Mediterranean Sea.

Abbreviations

BMNH	The Natural History Museum, London, U.K.
BOS	Biology of Organisms and Systems, University of Oviedo, Spain
IBUFRJ	Instituto de Biologia, Universidade Federal do Rio de Janeiro, Brazil
MNCN	Museo Nacional de Ciencias Naturales, Madrid, Spain

An updated check-list of the alien amphinomid species is provided based on an exhaustive review of the species records in the literature. The species data were mainly extracted from the regional reviews on alien species and compilations of polychaete species. We have also included data on their ecology, distribution and other relevant features.

Results

The revision of the literature along with our results (observations on 28 Mediterranean specimens belonging to *E. complanata* complex) revealed that five amphinomid species belonging to three genera were determined to be alien species in the Mediterranean Sea: *E. laevisetis, E. complanta, Linopherus acarunculatus* (Monro, 1937), *Linopherus canariensis* Langerhans, 1881 and *Notopygos crinita* Grube, 1855. The diagnostic differences between these species are summarised in the key provided. Furthermore, information about location, habitat, date of introduction and other relevant features are provided in Table 1.

Species	Locality	Year	Mediterranean area	Habitat	Others features	Reference
	Atlit (Israel)	1937	Eastern			Monro 1937 Current work
	Gulf of Eilat (Israel)	1976	Eastern	intertidal reefs of <i>Dendropoma</i> spp	Occurring together with another amphinomid <i>Linopherus acarunculatus</i>	Ben-Eliahu 1976
Eurythoe complanata	Isabel II Island Gibraltar Strait (Spain)	September 1992	Western	On rocks, 3–6 m depth	Occurring sympatrically with	
(Pallas, 1766)	Isabel II Island Gibraltar Strait (Spain)	July 1993	Western	On rocks, 3–6 m depth	Eurythoe laevisetis	Current work
	Congreso Island Gibraltar Strait (Spain)	July 1993	Western	On rocks, 3 m depth		Current work
	Chafarinas Islands Gibraltar Strait (Spain)	1995	Western	Rocky substrate		López 1995
	Isabel II Island Gibraltar Strait (Spain)	September 1992	Western	On rocks, 3–6 m depth	Occurring sympatrically with	
<i>Eurythoe laevisetis</i> Fauvel, 1914	Isabel II Island Gibraltar Strait (Spain)	July 1993	Western	On rocks, 3–6 m depth	Eurythoe complanata	Current work
	Gozo Harbour (Malta)	March 2011	Central	Rocky bottom 0.5–1 m depth	Associated with the invasive Branchiomma bairdi	Current work
	Lebanon	1966	Eastern	Shallow waters	Referred to as <i>Pseudeurythoe</i>	Laubier 1966
Linopherus acarunculatus (Monro, 1937)	Gulf of Elat (Israel)	1976	Eastern	Intertidal reefs of <i>Dendropoma</i> spp	acarunculata Monro, 1937. Çinar (2009) suggest that these records could be <i>L. canariensis</i> Langerhans, 1881	Ben-Eliahu 1976
	Kemer (Turkey)	July, 1993	Eastern	5 m depth on algae		
Linopherus canariensis	Cyprus	May 1997	Eastern	35 m depth on sandy substrate	Associated with <i>Brachiomma</i> lanceolatum	Çinar 2009
Laugenians, 1001	Antalya Bay (Turkey)	1997	Eastern		Referred to as <i>Pseudeurythoe</i>	Ergen and Çinar 1997
	Cyprus	2005	Eastern		acarunculata Monro, 1937	Çinar 2005

Table 1. Summary of current knowledge on exotic Mediterranean Amphinomidae.

23

On the occurrence of the fireworm Eurythoe complanata complex...

Species	Locality	Year	Mediterranean area	Habitat	Others features	Reference
	Turkey	September- October 2005	Eastern	On rocks between 0.1-5 m Mainly in C. <i>officinalis</i> substrate		Çinar 2009
	Italy	2005	Central			Occhipinti-Ambrogi et al. 2011
	Lake of Faro (Italy)	May 2008	Central	Artificial modules with a neighboring sandy bottom, 1.2 m depth	Showed an invasive behaviour, reaching densities of 41.86 ind / m ²	Cosentino and Giacobbe 2011
Notopygos crinita Grube, Italy 1855	Italy	1983	Central		Currently this species is considered as not established in the Mediterranean (Zenetos et al. 2010; Occhipinti- Ambrogi 2011)	Zenetos et al. 2010

Family Amphinomidae Lamarck, 1818

Genus Eurythoe Kinberg, 1857

Type species. *Eurythoe capensis* Kinberg, 1857, subsequent designation: *Eurythoe complanata* (Pallas, 1766).

Eurythoe laevisetis Fauvel, 1914

http://species-id.net/wiki/Eurythoe_laevisetis Fig. 2A–F

Eurythoe laevisetis Fauvel, 1914: 116, pl VIII fig. 28-30, 33-37. Type locality: São Tomé Island, Gulf of Guinea.

Material examined. *Eurythoe* cf. *complanata*: Gozo Harbour (Malta), 35°50'N, 14°35'E (Mar. 2011): BOS-Amp3 (2 specimens), BOS-Amp4 (9 specimens).

Eurythoe complanata: Isabel II Island (Chafarinas Islands, Spain), 35°11'N, 2°26'W (Sep. 1992): MNCN 16.01/3340 (1 specimen); (Jul. 1993) MNCN 16.01/33394 (1 specimen).

Diagnosis and description. Body depressed elongated, rectangular in cross section. Specimens from Malta ranged in length from 14 to 52 mm with a mean of 39 mm (N=11, SD=12.09). Live specimens have a uniform orange-pinkish colour (Fig. 2A–C), on which the gills and a bright red caruncle stand out, and white chaeta fascicles forming two longitudinal bands along the body (Fig 2A, B). Prostomium rounded with 2 pairs of inconspicuous eyes arranged in a square and three antennae, two lateral ones in an anterior position and one slightly behind the others. The anterior end has a bilobed prebuccal lobe where are inserted a pair of cirriform palps (Fig. 2D). The caruncle is elongated and extends until the third chaetiger (Fig. 2C, D). Each segment is provided with a pair of arborescent gills that are present from the second chaetiger to the posterior region (Fig. 2C, D). Biramous parapodia with digitiform dorsal and ventral cirri, similar in size. Notochaetae of two types: very fine with a small spur that continues in a capillary-like thorn; and thicker with a marked spur (spurred capillary notochaeta) (Fig. 2E).

Remarks. Several Maltese specimens present evidence of regeneration of the anterior and posterior end. All preserved specimens have whitish colour and lack the characteristic harpoon notochaetae. The two pairs of eyes are extremely inconspicuous, the anteriormost being similar in size to the posterior one. Specimens from Malta and Chafarinas Islands were morphologically identical to the Atlantic *E. laevisetis* from the Canary Islands and Cape Verde and *E. laevisetis* from São Tomé Island.

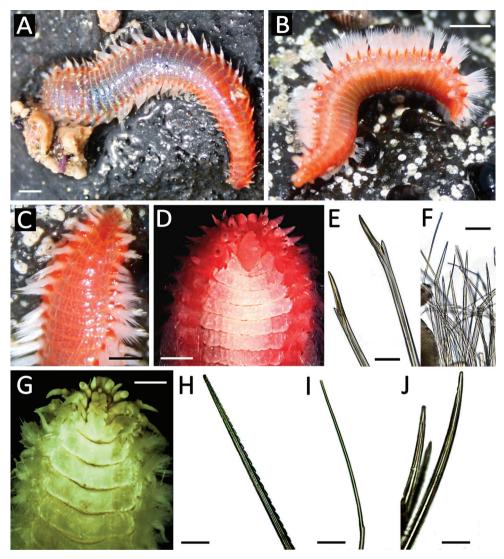


Figure 2. *Eurythoe laevisetis* from Malta. A live specimen, general view B live specimen, lateral view
C live specimen anterior end, dorsal view D detailed view of anterior end, dorsal view E neurochaetae
F spurred capillary notochaetae. *Eurythoe complanata* from Israel G detailed anterior end, dorsal view
H harpoon notochaeta I notopodial spurred capillar notochaeta J notoacicular spines.

Eurythoe complanata (Pallas, 1766)

http://species-id.net/wiki/Eurythoe_complanata Fig. 2G–J

Aphrodita complanata Pallas, 1766: 109, pl. 8, fig. 19-26. Type locality: Antigua Island, Caribbean Sea.

Eurythoe brasiliensis Hansen, 1882: 4, fig. 5-9. *Lycaretus neocephalicus* Kinberg, 1867: 55-56. *Eurythoe kamechameha* Kinberg, 1857: 14; 1910,36, pl. 12, fig. 13. *Eurythoe pacifica* Kinberg, 1857: 14; 1910: 36, pl. 12, fig. 11. *Eurythoe indica* Kinberg, 1867: 90. *Eurythoe ehlersi* Kinberg, 1867: 90. *Eurythoe havaiva* Kinberg 1867: 90. *Eurythoe corallina* Kinberg 1857:14; 1910:36, pl. 12, fig. 12. *Eurythoe alcyonaria* Gravier, 1902: 83, fig. 38, b-m.

Material examined. *Eurythoe complanata*: Isabel II Island (Chafarinas Islands, Spain), 35°11'N, 2°26'W (Sep. 1992): MNCN 16.01/3337 (2 specimens), MNCN 16.01/3338 (2 specimens), MNCN 16.01/3340 (1 specimen); Congreso Island (Chafarinas Islands, Spain), 35°11'N, 2°26'W (Jul. 1993): MNCN 16.01/3336 (1 specimen); Isabel II Island (Chafarinas Islands, Spain), 35°11'N, 2°26'W: MNCN 16.01/33394 (2 specimens). Atlit (Israel), 32°41'N, 34°56'E (1937): BMNH 1937.4.7.1-5 (7 specimens).

Diagnosis and description. Israeli specimens ranged from 20 to 45 mm in length with a mean of 31 mm (N=7, SD=9.77). Prostomium rounded with 2 pairs of eyes arranged in a square, the first being larger (Fig. 2G), and with three antennae, lateral ones in an anterior position and the single one slightly posterior. Anterior end with a bilobed prebuccal lobe, carrying a pair of cirriform palps. The caruncle is elongated and extends until the third chaetiger (Fig. 2G). Each segment is provided with a pair of arborescent branchiae that are present from the second chaetiger to the posterior end. Biramous parapodia with dorsal and ventral cirri digitiform, similar in size. Notochaetae of three types: harpoon-like (Fig. 2H); spurred capillaries with small spurs (Fig. 2I) and thicker smooth notochaetal spines (Fig. 2J). Notoacicula are very small, hastate, limited in number and always form an arc immediately in front of the dorsal cirrus. Neurochaetae are bifurcate, with prongs of different lengths.

Remarks. One specimen regenerating the posterior end. Pairs of eyes inconspicuous in some specimens, but always with the anterior pair larger than posterior pair. Specimens from Chafarinas Islands had a mean size of 37 mm (N= 8, SD = 7.24). All preserved specimens had a brownish colour.

Key to genera of Amphinomidae and species of *Eurythoe* and *Linopherus* of the Mediterranean Sea (modified from Borda et al. 2012)

1	Caruncle absent	Hipponoa
_	Caruncle present, variably developed	2
2	Oval body	
_	Elongated body; subcylindrical or quadrangular cross section	

3	Dorsal accessory (branchial) cirri plus dorsal cirri on anteriormost abrancl	hi-
	ate chaetigers; in branchiate chaetigers, one dorsal cirri per notopodium;	bi-
	pinnate branchiae Chloe	eia
_	Dorsal accessory (branchial) cirri plus dorsal cirri on all chaetigers; palma	
	branchiae	
4	First chaetiger dorsally continuous, complete	-
_	First chaetiger dorsally discontinuous, not complete	
5	Hooks present in the first chaetiger; caruncle roundParamphinon	ne
_	Hooks not present in the first chaetiger	6
6	Branchiae limited to anterior segments Linopherus	10
_	Branchiae on all segments after the chaetiger 2 or 3 Amphinon	
7	Caruncle large and conspicuous, extending beyond one chaetiger posteriorly	
_	Caruncle small and inconspicuous, not extending beyond one chaetiger po	os-
	teriorlyCryptonon	
8	Caruncle without a median lobe, with folds obliquely arranged Hermod	ice
_	Caruncle with a smooth median lobe	9
9	Caruncle not sinusoidalEurythoe	11
_	Caruncle sinusoidalPareuryth	oe
10	First branchiae present on chaetiger 3 L. canarien	sis
_	First branchiae present on chaetiger 4	us
11	Three types of notochaetae present: spurred capillary, notoacicular spine a	
	harpoon E. complana	ıta
_	Two types of notochaetae present: spurred capillary and notochaetal spin	
	harpoon absen	

Discussion

Members of the family Amphinomidae have a number of characteristics that gives the group high invasive potential. They show high biological plasticity and reproductive habits that include both sexual and asexual reproduction; possess a great capacity of regeneration and a large dispersal capability due to their long-term rostraria larvae (Kudenov 1995, Cosentino and Giacobbe 2011). Four amphinomid species are currently considered to be established in the Mediterranean Sea: *E. laevisetis, E. complanata, L. canariensis* and *L. acarunculatus* (Table 1). *Notopygos crinita* is presumably no longer present in the Mediterranean Sea, having been a case of accidental introduction that failed to establish (Zenetos et al. 2010, 2012, Occhipinti-Ambrogi et al. 2011). However, the recently introduced *L. canariensis* has displayed a highly invasive capacity and great potential for colonization, which are particularly favoured in stressed and degraded habitats where populations reach densities over 42 individuals/m² (Cosentino and Giacobbe 2011).

The use of the term 'morphospecies' for referring to *E. complanata* has been proposed as an alternative to overcome the identification difficulties associated with this

species complex, which includes two cryptic species along with *E. laevisetis.* Here, we have an example of two species that are genetically distinct but morphologically identical under the same 'morph', named as *E. complanata*. So, the *E. complanata* complex erected by Barroso et al. (2010) is actually formed by two morphospecies, *E. complanata* and *E. laevisetis.* The former includes two cryptic species which occur natively, one in the eastern Pacific and one in the Atlantic.

The E. complanata complex represents one more case of species group that is likely to be introduced in the Mediterranean, but which has been underestimated and misidentified. Re-examination of specimens from Malta, Chafarinas Islands and Israel demonstrates the existence of two morphospecies belonging to the E. complanata complex in the Mediterranean Sea: E. laevisetis in the western and central Mediterranean and E. complanata in the western and Levantine basins. Moreover, the Israeli E. complanata is not a recently introduced species, but one that had been present since, at least 1937. All examined specimens from Malta and two from Chafarinas Islands belong to the species E. laevisetis, characterized by the absence of the harpoon notochaetae. According to Barroso et al. (2010), the 'Atlantic-island-restricted species', differentiated by DNA sequences and morphology from E. complanata is, actually, E. laevisetis. This species was erroneously considered the junior synonym of E. complanata by several authors (e.g. Fauvel 1947, Ebbs 1966). According to Fauvel (1914), the main diagnostic feature distinguishing E. laevisetis from the related E. complanata is its lack of harpoon notochaetae (Barroso et al. 2010). After the examination of the E. laevisetis specimens (without harpoon notochaetae) from different localities (Malta, Chafarinas Islands, Canary Islands, Cape Verde and São Tomé Island), we observed that both anteriormost and posterior pairs of prostomial eyes were similar in size in all studied specimens, being always very inconspicuous. By contrast, all examined specimens belonging to *E. complanata* exhibited, besides the characteristic harpoon chaetae, anterior eyes larger than posterior ones.

On the other hand, all examined specimens from Israel and nine from Chafarinas Islands were morphologically identical to *E. complanata* from the Atlantic and Pacific *sensu* Barroso et al. (2010), including the characteristic harpoon notochaetae, length of caruncle, prostomial appendages, branchial distribution pattern and other types of notopodial and neuropodial chaetae. These specimens differ from *E. laevisetis* by the presence of the harpoon notochaetae and size differences between the two pairs of eyes, with the anterior pair always larger than the posterior ones.

Kinberg (1857) first described the genus *Eurythoe* in the Mediterranean Sea based on *Eurythoe syriaca* from the Syrian coasts and *Eurythoe hedenborgi* from Dr. Hedenborg's collection. Later, Monro (1937) reported *E. complanata* for the first time from the Mediterranean, considering *E. syriaca* as its junior synonym. Nevertheless, Hartman (1948) when reviewing the species described by Kinberg considered *E. syriaca* as a valid species. In the same review, as well as in her later world catalogue Hartman (1959) regarded *E. hedenborgi* as a questionable species, even though no justification was provided. More recently, Çinar (2008) described *Eurythoe turcica* from the Levantine coast of Turkey and differentiated this species from the related Indo-Pacific *Eurythoe parvecarunculata* Horst, 1912. Nevertheless, Borda et al. (2012) transferred these latter two species to the genus *Cryptonome* based on a phylogenetic analysis. Therefore, based upon a comprehensive review of the literature descriptions we propose that currently only two species can be validly assigned to the genus *Eurythoe* in the Mediterranean Sea, *E. complanata* and *E. laevisetis*.

The origins, plausible pathways and introduction vectors of these related amphinomids into the Mediterranean may be discerned by focusing on populations of the central (E. laevisetis), western (E. laevisetis and E. complanata) and Levantine (E. complanata) regions. For example, Maltese and Chafarinas populations of E. laevisetis may have originated from Atlantic islands through the Gibraltar Strait. Such a scenario is wholly consistent with arrivals of other Atlantic species of marine invertebrates into the Mediterranean such as the gastropod Marginella glabella (Linnaeus, 1758), which is presently colonizing the coasts of Málaga (SE Spain, western Mediterranean) from the Canary Islands and West Africa (Luque et al. 2012). The Gibraltar Strait was also suggested to be the main pathway of introduction for other polychaetes such as the invasive sabellid Branchiomma bairdi (McIntosh, 1885), which is associated with E. laevisetis in Maltese Islands (Arias et al. 2013a) and for other conspicuous amphinomids, such as *H. carunculata*. The Mediterranean populations of the latter also seem to have descended from Atlantic ones (Ahrens et al. 2013) as well as L. canariensis populations from the Italian coasts (Cosentino and Giacobbe 2011). Two different plausible hypotheses concerning *E. complanata* populations must be considered in relation to their present geographical distributions. For example, Israeli populations could be Lessepsian migrants due to their proximity to the Suez Canal. On the other hand, E. complanata from the Chafarinas islands and also localized in the Strait of Gibraltar, could be Atlantic migrants from the Canaries or other Atlantic archipelagos. However, multiple routes and times of introduction for all studied populations (Chafarinas, Malta and Israel) seem tenable and cannot be excluded. Further research mainly using molecular markers of Maltese and Israeli populations, as well as Red Sea and Canary Island ones, is needed to give more information concerning their origins and dispersion in the Mediterranean Sea. Finally, it is essential to emphasize that the great dispersive capacity of *E. complanata* (Barroso et al. 2010) is likely due to the inferred high longevity of its planktotrophic rostraria larvae (Bhaud 1972); additionally, the combination of asexual and sexual reproduction (Kudenov 1974) may promote the invasive potential of this species. Therefore, a detailed monitoring of the dynamics of Maltese and Israeli populations, as well as setting up a current distribution map should be undertaken in order to establish and understand the evolution of E. complanata complex across the Mediterranean Sea.

Acknowledgements

We are grateful to C. Glasby, J. Kudenov and one anonymous reviewer for valuable comments and suggestions that greatly improved the manuscript. The first author (AA)

is supported by a Severo Ochoa fellowship from the FICYT Foundation (Principado de Asturias, Spain). CNPq (Conselho Nacional de Desenvolvimento Científico e Tecnológico) provided support to RB. CNPq and FAPERJ (Fundação Carlos Chagas Filho de Apoio à Pesquisa do Estado do Rio de Janeiro) provided financial support to PCP.

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RESEARCH ARTICLE



A new species of Numbakullidae Guțu & Heard, 2002 (Tanaidacea, Peracarida, Crustacea) from the Great Barrier Reef, Australia

Anna Stępień^{1,†}

I Department of Invertebrate Zoology and Hydrobiology, University of Łódź, Banacha 12/16, Łódź 90-237, Poland

+ http://zoobank.org/A1A1CBED-2F4D-4968-8321-EFB42765EF1E

Corresponding author: Anna Stepień (stepie.anna@gmail.com)

Academic editor: Niel Bruce Received 2 July 2013 Accepted 7 August 2013 Published 1 October 201	3
http://zoobank.org/FB76AC3B-D8F6-4D4E-B738-C43DA62199B1	_

Citation: Stępień A (2013) A new species of Numbakullidae Guțu & Heard, 2002 (Tanaidacea, Peracarida, Crustacea) from the Great Barrier Reef, Australia. ZooKeys 337: 35–47. doi: 10.3897/zookeys.337.5903

Abstract

A new species of *Numbakulla* Guțu & Heard, 2002 (Tanaidacea) is described from Heron Island (southern Great Barrier Reef, Queensland) collected during the Census of Coral Reefs Ecosystem (CReefs) program. The new species is the third member of the family and can be recognized by the combination of characters as: length/width ratio of the body, which is 6:7, pereonite 4 longer than the rest, the presence of eyes, a blunt rostrum, antenna article 2 elongated, cheliped carpus with row of inner setae, pereopod 6 carpus with spines, pleopod endopod with denticles.

Keywords

Tanaidacea, Numbakullidae, Numbakulla, Australia, CReefs, coral reefs

Introduction

To date 176 species of Tanaidacea have been described from Australia. The best studied parts of the continent are the Bass Strait (Victoria) with 66 species (Błażewicz-Paszkowycz and Bamber 2007a, b, 2012; Bamber and Błażewicz-Paszkowycz 2013), Moreton Bay (Queensland) with 35 species (Boesch 1973; Guțu 2006a; Błażewicz-Paszkowycz and Bamber 2007a, b; Bamber 2008, 2013), and Esperance Bay (Western Australia) with 24

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species (Bamber 2005). In contrast, the tropical coast of Australia with its coral reefs is comparatively poorly studied, with only 14 species of Tanaidacea described.

This paper describes a new species of *Numbakulla* Guțu & Heard, 2002 from the Great Barrier Reef (eastern Australia). The material was collected in the vicinity of Heron Island during the Census of Coral Reefs Ecosystem (CReefs) program in 2010.

Numbakulla Guțu & Heard, 2002 was erected for two species *N. pygmaeus* Guțu & Heard, 2002 (type species, continental shelf of north-western Australia) and *N. srilank-ensis* Guțu, 2006 (coral reefs of Sri Lanka), and was defined principally by the presence of filtering setae on the merus and carpus of pereopod 1 and the longer third article of the maxilliped. These characters, together with the large carapace and pleotelson, pereopod 6 differing from pereopods 4–5, and the presence of only four pairs of pleopods were considered of sufficient significance to create the new family, the Numbakullidae.

Material and methods

Material was collected during the CReefs expeditions organized by AIMS (Australian Institute of Marine Science) to Heron Island (Great Barrier Reef) in 2010. Pieces of coral rubble were collected by hand while SCUBA diving and placed into buckets (20 l) with a few drops of formaldehyde to encourage animals to leave their microhabitats (tubes and crevices). The samples, with the animals still alive, were then washed over a fine mesh (0.3 mm), the residue sorted under the microscope, and tanaidacean specimens were collected and preserved in 80% ethanol (see Stępień and Błażewicz-Paszkowycz 2009). Material is held at the Museum of Tropical Queensland, Australia (MTQ).

Morphological terminology follows Błażewicz-Paszkowycz and Bamber (2012).

Results

Systematics

Suborder Apseudomorpha Sieg, 1980

Family Numbakullidae Guțu & Heard, 2002

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

Remarks. Guțu and Heard (2002) mentioned some morphological similarities between Numbakullidae and two other families: Kalliapseudidae Lang, 1956 and Metapseudidae Lang, 1970. The presence of filtering setae suggests a relationship to Kalliapseudidae. Guțu and Heard (2002) also listed number of characters between the two families, which imply that present of setae is a parallelism.

Numbakullidae resembles the Metapseudidae in the appearance of pereopods 2–5 (elongated propodus and well-developed dactylus). Guțu (2006) later suggested that

Numbakulla is most closely similar to three genera from subfamily Chondropodinae Guțu, 2008 (Metapseudidae): *Calozodion* Gardiner, 1973, *Zaraza* Guțu, 2006c and *Chondropodus* Guțu, 2006a based on configuration of terminal lobe of labium.

Despite this similarities there are many characters which distinguish the Chondropodinae from the Numbakullidae: short pleotelson, usually with lateral incision, spines on antennule, labium with lateral spines, pereopod 1 digging type, with row of spines and plumose setae on basis, and propodus longer than carpus. At this point, therefore the relationships of the Numbakullidae remained unclear.

Undoubtedly *Numbakulla* represents a separate family. According to a phylogenetic analysis based on morphological characters, the Numbakullidae is not closely related to either the Kalliapseudidae nor the Metapseudidae (personal observations, study in progress).

Genus Numbakulla Guțu & Heard, 2002

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

Diagnosis. (amended after Guțu and Heard 2002): Body dorso-ventrally flattened, elongated, more than four times as long as wide. Cephalothorax and pleotelson very large in relation to pereon and pleon (cephalothorax at least as long as first three pereonites combined, pleotelson as long as all pleonites combined). Pereonites and pleonites wide and short, all similar in length or fourth pereonite longer than the rest, pleonites all similar in length. Eyes present or absent. Maxilliped palp with numerous inner plumose setae. Epignath elongate, with long terminal spine. Cheliped with exopodite, carpus and propodus very large. Pereopod 1 with exopodite bearing distally 4 to 5 plumose setae, propodus very short, at least two times shorter than carpus. Pereopod 6 with long carpus and propodus and with long plumose setae on basis, merus and carpus. Four pairs of pleopods. Uropod with elongated basis and multisegmented endopod, inserted medially on lateral margin of pleotelson. Sexual dimorphism very pronounced in large size of male cheliped.

Type species. N. pygameus Guțu & Heard, 2002, by monotypy.

Species included. *N. pygameus* Guțu & Heard, 2002, *N. srilankensis* Guțu, 2006, *N. pii* sp. n.

Numbakulla pii sp. n.

http://zoobank.org/34385B01-7446-45DA-863D-A22C97B1B808 http://species-id.net/wiki/Numbakulla_pii Figs 1–4, Photo 1

Material examined. *Holotype*: female (MTQ W34252), Stn HI 10-009E, 23°25'53"S, 152°2'57"E, Sykes Reef, reef slope, coral rubble on sand, 12m depth, 14 November 2010, coll. C. Buxton.

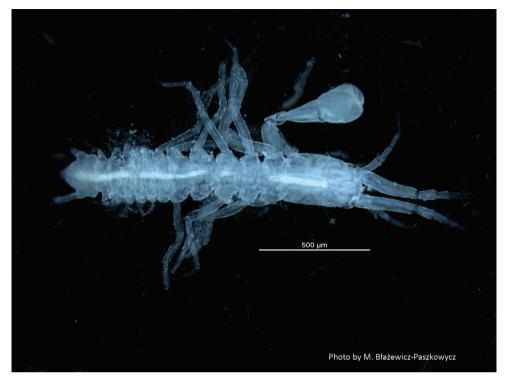


Photo I. Numbakulla pii sp. n., paratype female. Body dorsal view.

Paratypes: 14 females (MTQ W34253), Stn HI 10-009E, same locality as holotype; eleven females (MTQ W34254), Stn HI 10-009D, 23°25'53"S, 152°2'57"E, Sykes Reef, reef flat, small coral rubble on sand, 12 m depth, 14 November 2010, coll. M. Błażewicz-Paszkowycz, C. Buxton; one female (MTQ W34256) Stn HI 10-009B, 23°25'53"S, 152°2'57"E, Sykes Reef, reef slope, coral rubble at base of the wall, 27 m depth, 14 November 2010, coll. S. Smith, C. Buxton; four females (MTQ W34255) Stn HI 10-013A, 23°35'12"S, 152°3'44"E, Lamont Reef, reef slope, coral rubble on sand, 21 m depth, 15 November 2010, coll. C. Buxton.

Etymology. Named after Pi Patel, the central character from the novel Life of Pi, written by Yann Martel, one of favourite authors novel.

Diagnosis. Body dorsoventrally flattened, 6.7 times longer than wide, blunt rostrum, eyes present, pereonite 4 clearly longer than the rest, mandible palp articles 2 with spines on outer margin, lacina mobilis narrow, one-denticled, antennule outer flagellum with six segments, antenna peduncle article 2 elongated, cheliped carpus with inner row of seven setae, pereopod 1 basis with row of setae, pereopod 6 carpus with row of ventral spines, pleopod endopod with proximal acute denticles, uropod endopod with 13 segments.

Description of female. Body (Figs 1A, B, photo 1) 1.7 mm long, 6.7 times as long as wide. Cephalothorax 22% of total body length, with blunt rostrum; ocular

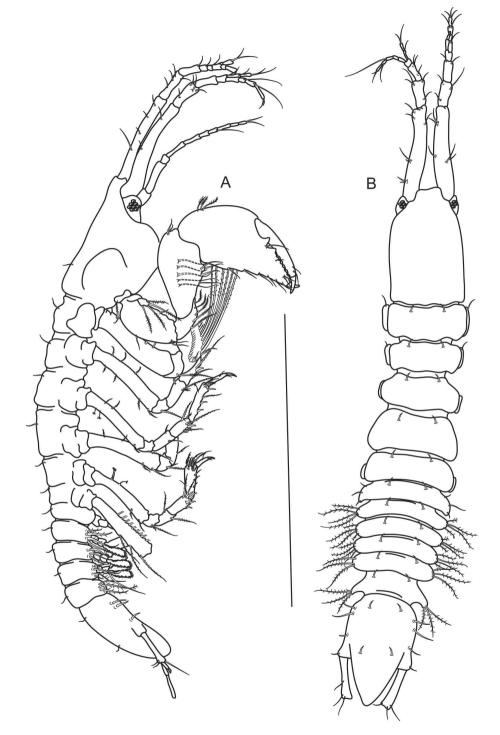


Figure 1. *Numbakulla pii* sp. n., holotype female. **A** body dorsal view **B** body lateral view. Scale line = 1 mm.

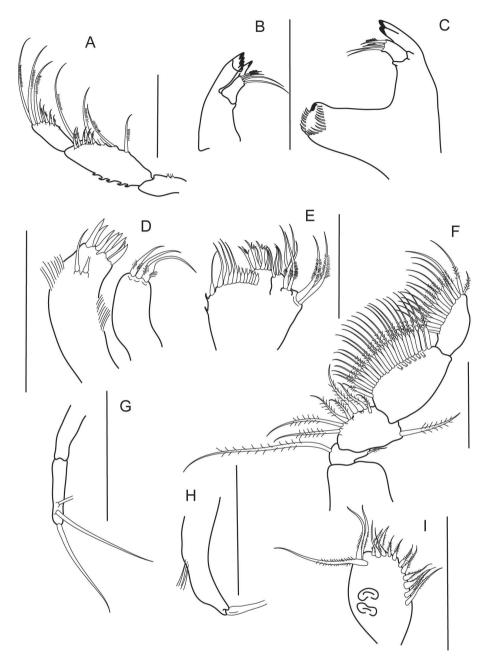


Figure 2. *Numbakulla pii* sp. n., **A** palp of left mandible **B** left mandible **C** right mandible **D** maxillule **E** maxilla **F** maxilliped **G** palp of maxillule **H** epignath **I** maxillipedal endite. Scale line = 0.1mm.

lobes present, with visual elements. Pereon: pereonites 1, 2 and 3 similar in length, 0.3 times as long as wide, pereonite 4 longest, 0.4 times as long as wide; pereonites 5 and 6 shorter, about 0.2 times as long as wide; all pereonites with pair of setae dorsally;

last pereonite with three setae laterally. Pleon 19% of total body length, first four pleonites similar in length, 0.1 times as long as wide, with three or four long plumose setae laterally and pair of setae dorsally, and with rounded lateral margin; last pleonite 0.2 times as long as wide, with three plumose setae laterally and acute lateral margin. Pleotelson acute posteriorly, 1.6 times as long, with four setae laterally and two pairs of setae dorsally.

Antennule (Fig. 3A): peduncle article 1 7.5 times as long as wide, with one penicillate seta on outer margin, and with 13 simple setae on whole surface; article 2 0.2 times as long as article 1, with one simple seta on inner margin and with five simple setae and two penicillate setae on distal margin; article 3 2.4 times as long as wide, with four distal simple setae; article 4 with four distal simple setae; outer flagellum with six segments, segments 1 and 3 with aesthetasc, last segment with four simple setae; inner flagellum with two segments, last segment with four simple setae.

Antenna (Fig. 3B): peduncle article 1 with rounded apophysis bearing distal plumose seta; article 2 elongate, five times as long as wide, with three inner plumose setae; squama narrow, bearing two simple setae; article 3 0.2 times as long as article 2, with two distal simple setae; article 4 0.6 times as long as article 2, with one penicillate seta; article 5 similar in length to article 4, with two mid-length simple setae, and distally with two simple, and three penicillate setae; flagellum with four segments, distal segment with three simple setae.

Mouthparts: Left mandible (Fig. 2B) bearing denticulate pars incisiva; lacinia mobilis with one blunt tooth; setiferous lobe with two simple and two plumose setae; palp (Fig. 2A) article 1 1.7 times as long as wide, with two inner setae; article 2 2.5 times as long as wide, with five long and five short plumose setae on inner margin, and with denticulate outer margin; article 3 2.2 times as long as wide, with seven inner plumose setae. Right mandible (Fig. 2C) with two teeth on pars incisiva; pars molaris robust, margin with row of simple setae. Maxillule (Fig. 2D) outer endite with eight distal spines and two subdistal setae; inner and outer margin finely setose; inner endite with three distal plumose setae; palp (Fig. 2G) two-articled, with two subdistal and one distal setae. Maxilla (Fig. 2E) outer lobe of moveable endite with plumose setae, two subdistally and four on distal margin; inner lobe with four distal simple setae; outer lobe of fixed endite with some simple setae and three trifurcated setae; inner endite with rostral row of twelve setae, inner margin with two denticles. Maxilliped (Fig. 2F) basis 0.8 times as long as wide, naked; palp article 1 0.4 times as long as wide, with one outer plumose seta and with one long inner plumose setae; palp article 2 1.3 times as long as wide, with one outer plumose seta, and with inner row of seven plumose setae; palp article 3 1.7 times as long as wide, with two inner rows of plumose setae (marginal row of 23 setae, submarginal row of six setae); palp article 4 2.7 times as long as wide, with inner row of 15 plumose setae. Endite (Fig. 2I) with 15 plumose setae along outer and distal margin, and with two coupling hooks. Epignath (Fig. 2H) narrow, with strong distal seta.

Cheliped (Fig. 3C) basis 1.8 times as long as wide, ventrally with two proximal setae, and two distal setae, and dorsally with one seta; exopodite three-articled, last arti-

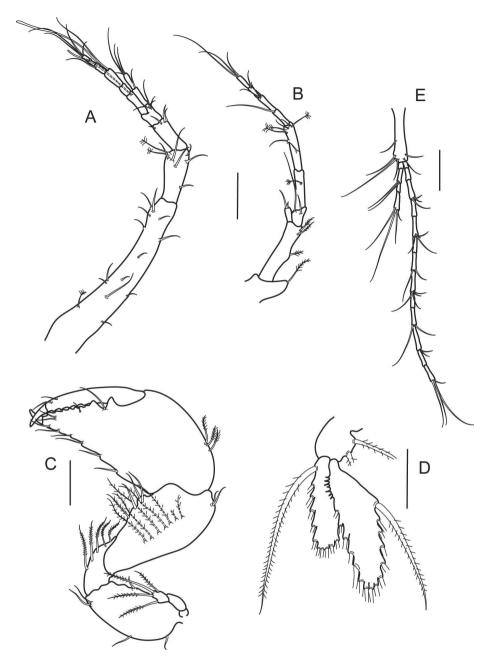


Figure 3. *Numbakulla pii* sp. n., **A** antennule **B** antenna **C** left cheliped **D** pleopod **E** uropod. Scale line = 0.1 mm.

cle with four plumose setae; merus 1.5 times as long as wide, with five ventral plumose setae; carpus 1.7 times as long as wide, with four ventrodistal setae, two dorsodistal setae, and with row of seven inner plumose setae; palm of propodus as long as wide,

with three dorsal plumose setae and with six simple setae along ventral margin of palm and fixed finger; fixed finger as long as palm, cutting edge crenulated, with four simple setae, and two proximal tooth-like apophysis; dactylus with unguis 1.2 times as long as propodus, with two dorsal setae, cutting edge crenulated.

Pereopod 1 (Fig. 4A) basis four times as long as wide, dorsally with six plumose setae; exopodite three-articled, distal article with four plumose setae; ischium 0.3 times as long as wide, with one ventrodistal seta; merus 1.8 times as long as wide, with middorsal plumose seta and two dorsodistal plumose setae, ventral margin with two rows of numerous, long plumose setae; carpus 0.7 times as long as merus, dorsally with three plumose setae; propodus 1.3 times as long as wide, half as long as carpus, with one penicillate and one plumose setae, and one spine dorsally, ventral margin with three plumose setae and four spines; dactylus together with unguis as long as propodus, with one dorsal, and two ventral setae.

Pereopod 2 (Fig. 4B) basis six times as long as wide, with four ventral plumose setae and three dorsal setae, ischium as long as wide, with plumose ventral seta; merus three times as long as wide, ventral margin with one plumose seta and thin spine, dorsal margin with one plumose seta; carpus 0.7 times as long as merus, with two plumose setae and two spines ventrally, and with three dorsodistal plumose setae; propodus 5.3 times as long as wide, 2.1 times as long as carpus, with one dorsal penicillate setae, ventral margin with one plumose seta and three spines, three finely denticulate setae near dactylus insertion; dactylus together with unguis 0.7 times as long as propodus, with seta on dorsal and ventral margin.

Pereopod 3 (Fig. 4C) similar to pereopod 2, but basis with two ventral penicillate setae.

Pereopod 4 (Fig. 4D) basis 5.6 times as long as wide, with three dorsal plumose setae and with two ventral plumose setae; ischium 0.6 times as long as wide, with two ventrodistal plumose setae; merus 1.6 times as long as wide, with dorsal plumose seta and with three dorsodistal and one mid-dorsal plumose setae; carpus 1.1 times as long as merus, with two dorsal plumose setae, and with four spines and two plumose setae on ventrodistal and distal margin; propodus 4.8 times as long as wide, 1.7 times as long as carpus, with dorsal penicillate seta, ventral margin with three thin spines and one simple seta, and three finely denticulated setae near dactylus insertion; dactylus together with unguis 0.8 times as long as propodus, with simple seta on ventral and dorsal margin.

Pereopod 5 (Fig. 4E) basis 5.5 times as long as wide, with three proximal penicillate setae on dorsal margin and with two ventral plumose setae; ischium 0.9 times as long as wide, with two plumose ventrodistal setae; merus 1.3 times as long as wide, with plumose seta dorso- and ventrodistally; carpus 1.6 times as long as merus, with five spines and two plumose setae on distal margin; propodus (Fig. 4F) three times as long as wide, 1.3 times as long as carpus, with three thin ventral spines, and with eleven finely denticulate plumose setae near dactylus insertion; dactylus together with unguis 0.7 times as long as propodus, with simple setae on dorsal and ventral margin.

Pereopod 6 (Fig. 4G) basis 6.8 times as long as wide, with ten dorsal plumose setae, and with three ventral plumose setae; ischium 0.6 times as long as wide, with two ventrodistal plumose setae; merus 1.7 times as long as wide, with six dorsal plumose

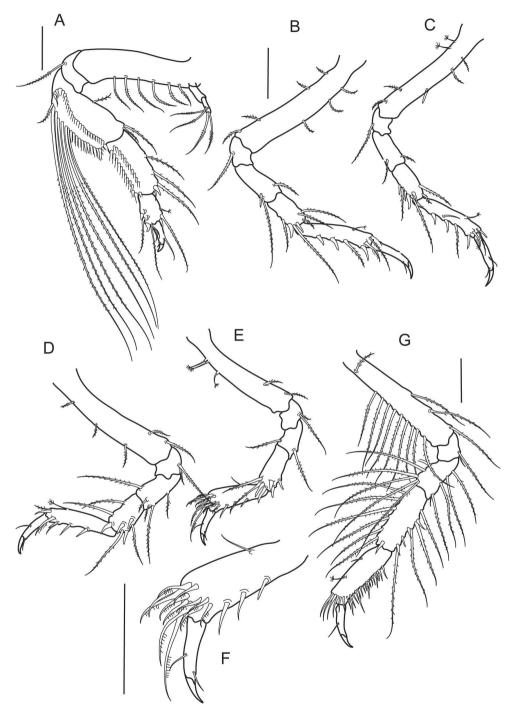


Figure 4. *Numbakulla pii* sp. n., **A** percopod 1 **B** percopod 2 **C** percopod 3 **D** percopod 4 **E** percopod 5 **F** propodus of percopod 5 **G** percopod 6. Scale line = 0.1 mm.

setae, ventral margin with four plumose setae, and one short, simple seta; carpus 1.7 times as long as merus, with six plumose setae and one spine on dorsal margin, and with four plumose setae and four spines on ventral margin; propodus 3.3 times as long as wide, similar in length to carpus, dorsally with one simple and one penicillate seta, and with row of numerous simple setae along ventral and distal margin; dactylus together with unguis 0.7 times as long as propodus, with one dorsal setae.

Pleopods (Fig. 3D) in four pairs, basis with two inner marginal plumose setae; exopod with 13 marginal plumose setae; endopod with 15 marginal plumose setae and proximal acute denticles on inner margin.

Uropod (Fig. 3E) basis 6.5 times as long as wide, with six distal and one marginal setae; exopod of four-segments; segment 2 with two distal setae; last segment with four distal setae; endopod of 13 segments, most with simple setae, last segment with four distal setae.

Remarks. The new species differs from the two previously described species by the length/width ratio of the body, which is 6.7 in *Numbakulla pii* sp. n., 8.0 in *N. srilankensis* and 4.5 in *N. pygmaeus. N. pii* also differs in lacking "glandular formations" on the pereopods, present in the other species, having ventral spines on pereopod 6 carpus and acute proximal denticles on endopod of pleopod. *Numbakulla pii* can be distinguished by the appearance of the mandible with a narrow, one-denticled lacina mobilis and palp article 2 with denticles.

The new species is similar to *N. srilankensis* in having a flat rostrum, well-developed eyes and pereonite 4 clearly longer than the rest, while in *N. pygmaeus* rostrum is rounded, eyes are absent and all pereonites are similar in length. *N. pii* resembles *N. srilankensis* also in bearing row of inner setae on cheliped carpus, but in first species the row contain seven setae and in the second species 24. Both species have also a row of setae on basis of pereopod 1 (absent in Australian species)

Numbakulla pii shares with N. pygameus an elongated peduncle article 2 of the antenna, which is short in N. srilankensis.

Distribution. The species is known from Lamont Reef and Sykes Reef, the Capricorn Group, southern Great Barrier Reef, eastern Australia and was recorded from a depth range of 12–27m in coral rubble.

Key to the species of genus Numbakulla

1	Eyes present, pereonite 4 longer than the rest pereonites, cheliped carpu
	with inner row of setae
_	Eyes absent, all pereonites similar in length, cheliped carpus without inne
	row of setae
2	Antennule outer flagellum with six segments, antenna article 2 elongated
	pereopod 6 carpus with spine
_	Antennule outer flagellum with five segments, antenna article 2 short, pereo
	pod 6 carpus without spines

Acknowledgements

The material for this paper was collected during CReefs Australia node Project (Field program of Census of Marine Life) that was generously sponsored by BHP Billiton in partnership with the Great Barrier Reef Foundation and the Australian Institute of Marine Science (AIMS). I am grateful to Magdalena Błażewicz-Paszkowycz (University of Łódź) and Chad Buxton for collecting and sorting material and making it available for my research, and to Dr. Roger N. Bamber for improving the English throughout the manuscript. The research has been financed through the Polish Ministry of Science and Higher Education Grant No. 7984/B/P01/2011/40.

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RESEARCH ARTICLE



Morphology, bioacoustics, and ecology of Tibicen neomexicensis sp. n., a new species of cicada from the Sacramento Mountains in New Mexico, U.S.A. (Hemiptera, Cicadidae, Tibicen)

Brian J. Stucky^{1,†}

I Department of Ecology and Evolutionary Biology, University of Colorado, Boulder, Colorado, USA

+ http://zoobank.org/81035622-5CF4-443E-9959-817BA3A37339

Corresponding author: Brian J. Stucky (stuckyb@colorado.edu)

Academic editor: Allen Sanborn | Received 10 July 2013 | Accepted 12 September 2013 | Published 1 October 2013

http://zoobank.org/FB4CCB9C-1AF9-4D0E-B8ED-D24B521CD901

Citation: Stucky BJ (2013) Morphology, bioacoustics, and ecology of *Tibicen neomexicensis* sp. n., a new species of cicada from the Sacramento Mountains in New Mexico, U.S.A. (Hemiptera, Cicadidae, *Tibicen*). ZooKeys 337: 49–71. doi: 10.3897/zooKeys.337.5950

Abstract

Tibicen neomexicensis **sp. n.**, a new species of cicada found in the Sacramento Mountains of southcentral New Mexico, is described. *T. neomexicensis* closely resembles *T. chiricahua* Davis morphologically, but males of the two species have highly distinct calling songs that differ in phrasal structure, amplitude burst rates, and pulse structure. Unlike *T. chiricahua*, male *T. neomexicensis* use conspicuous dorso-ventral abdominal movements to modulate the amplitude and frequency of their calls. *T. neomexicensis* is also smaller on average than *T. chiricahua*, and differences in the color patterns of the wing venation identify these two species morphologically. Both species are dependent on pinyon-juniper woodlands and have similar emergence phenologies. These species appear to be allopatric, with *T. chiricahua* found west of the Rio Grande in New Mexico, Arizona, and Mexico, and *T. neomexicensis* so far known only from New Mexico, east of the Rio Grande. *T. chiricahua* and *T. neomexicensis* males share a common genitalic structure that separates them from all other species of *Tibicen*, and the possible evolutionary and biogeographic history of these likely sister species is also discussed.

Keywords

Cicadidae, Tibicen, bioacoustics, cicada, cryptic species

Introduction

Cicadas, crickets, katydids, and many other insects produce airborne acoustic signals that play an essential role in reproduction (Alexander 1960, 1967, Capinera et al. 2004). For cicadas, acoustic communication is the single most important factor in mate recognition, pair formation, and premating reproductive isolation (Alexander and Moore 1958, Boulard 2006). As such, the calling songs of male cicadas have become an essential part of cicada taxonomy. Acoustic studies have led to the discovery of numerous "cryptic" cicada species that are morphologically nearly identical to other species but can be readily identified by their unique mating calls (e.g., Davis 1922, Alexander and Moore 1962, Popov 1989, Marshall and Cooley 2000, Quartau and Simoes 2005, Sueur and Puissant 2007, Cole 2008, Gogala et al. 2008). In some cases, acoustic analyses provide the only means to identify a species with certainty (e.g., Sueur et al. 2007, Gogala et al. 2008).

During fieldwork in New Mexico in 2012, I observed cicadas that fit the morphological description of *Tibicen chiricahua* Davis (Davis 1923) in both the Magdalena Mountains of west-central New Mexico and the Sacramento Mountains of southcentral New Mexico. However, the populations from these two mountain ranges had completely different calling songs, suggesting the presence of two species and rendering the taxonomic identities of both populations uncertain. To help resolve this problem, I traveled to the type locality of *T. chiricahua*, Pinery Canyon in the Chiricahua Mountains of southeastern Arizona (Davis 1923), to record the calls of true *T. chiricahua*. The calling songs recorded in the Chiricahua Mountains were the same as those recorded in the Magdalena Mountains in New Mexico, revealing that the cicadas in the Sacramento Mountains were a previously unrecognized species, described here as *Tibicen neomexicensis*. Upon closer inspection, it became clear that these two species exhibited subtle morphological differences, as well.

In this paper, I describe *Tibicen neomexicensis* and compare its morphology to *T. chiricahua*, describe and compare the calling songs and calling behaviors of *T. neomexicensis* and *T. chiricahua*, and compare the geographic distributions of the two species. Finally, I discuss the general ecology, phenology, and daily activity patterns of *T. neomexicensis* and consider its possible evolutionary relationship with *T. chiricahua*.

Methods

Field sites and specimens examined

All field work was conducted during May and June of 2012. Cicadas identified as *Tibicen chiricahua* were observed and audio recorded in the Magdalena Mountains west of Socorro, New Mexico, and at the type locality for *T. chiricahua*, Pinery Canyon in the Chiricahua Mountains of southeastern Arizona (Davis 1923). Specimens of the new species were observed and recorded at its type locality.

To estimate the geographic ranges of the two species and better understand morphological variation across these ranges, I examined a total of 202 specimens previously identified as *T. chiricahua* from the collections of the Arthropod Museum at New Mexico State University (NMSU), the C. P. Gillette Museum of Arthropod Diversity at Colorado State University (CSUC), the Frank M. Hasbrouck Insect Collection at Arizona State University (ASUT), the Snow Entomological Museum at the University of Kansas (SEMC), the Texas A&M University Insect Collection (TAMU), the University of Arizona Insect Collection (UAIC), and the University of Colorado Museum of Natural History (UCMC). The SEMC specimens included a male paratype of *T. chiricahua* from Davis's original type series. I also examined high-resolution digital photographs of the holotype male and allotype female of *T. chiricahua*, which are currently housed in the collection of the Academy of Natural Sciences of Drexel University (ANSP).

Morphology

Morphological terminology follows Moulds (2005, 2012). Morphometric measurements were made with a digital caliper. Fore wing width was measured from the node to the posterior edge, head width was measured between the eyes, and pronotum width was taken at the widest point between the lateral angles.

Audio recordings and analysis

Cicada calling songs were recorded in the field using a Sennheiser ME 66 shotgun microphone with an MZW 66 PRO windscreen connected to a Sony PCMM10 digital audio recorder. All recordings were made as uncompressed, 16-bit PCM audio at a sampling rate of 44.1 kHz. For each recording, the microphone was held between 0.5 and 2 meters away from the calling cicada. This was close enough to minimize background noise, but far enough away to avoid any near-field acoustic effects in the frequency range of the calling songs (Michelsen and Nocke 1974, Peterson 1980).

Cicada calls were analyzed to determine peak frequencies, amplitude burst rates, and the number of sound pulses per amplitude burst. In this paper, I use the term "pulse" in the sense of Broughton (1963) and "amplitude burst" to mean a single group of high-amplitude pulses in an amplitude-modulated pulse train (see Figures 4 and 5). I elected to use "amplitude burst" rather than "syllable," which has been used inconsistently in cicada bioacoustics and usually with disregard to the precise definitions of Broughton (1963) and Ragge and Reynolds (1998).

Analyses were conducted using Audacity[®] (Audacity Team 2012) and customwritten software. Peak frequency was estimated by identifying the highest peak in a power spectral density plot generated by a 512-sample Fast Fourier Transform with the Hamming window function. If there were two or more peak frequencies that differed by less than 0.5 dB, their average was taken as the overall peak frequency. The amplitude burst rate (i.e., pulse amplitude modulation rate) was calculated by first estimating the call's amplitude envelope, then using a gate function to identify the amplitude peaks in the signal (Beeman 1998). To estimate the number of sound pulses per amplitude burst, a sequence of 12 bursts was selected from the middle of each call, the audio data were normalized so that the maximum signal amplitude was at 0 dBFS, and the beginning and ending pulses of each amplitude burst were determined by identifying the first and last pulses with absolute sample values that exceeded 50% of the maximum sample value (that is, -6.02 dBFS).

The calls of both *T. chiricahua* and *T. neomexicensis* can be divided into three phrases (see results below), but the boundaries between phrases are often indistinct. To avoid the non-repeatability and potential bias of estimating the phrase durations by simple visual or aural inspection of the call oscillograms, I used objective criteria based directly on the audio data. All audio data were first normalized so that the peak amplitude was at 0 dBFS. For both *T. chiricahua* and *T. neomexicensis*, the first phrase began at the start of the call, and the end of the first phrase and beginning of the second phrase was defined by the first amplitude burst that reached -3 dBFS. For *T. chiricahua*, the end of the second phrase was defined by the last amplitude burst to reach -3 dBFS, while for *T. neomexicensis*, the end of the second phrase was defined as the end of the second phrase was defined as the end of the second phrase was defined as the end of the second phrase was defined by the second phrase was defined as the end of the modulated portion of the call. For both species, the third phrase consisted of all audio from the end of the second phrase to call termination.

I did not include ambient air temperatures in the acoustic analyses. North American cicadas utilize a variety of behavioral and physiological thermoregulation tactics, so ambient temperature is often a poor indicator of a calling cicada's body temperature (Toolson 1987, Hastings 1989, Sanborn et al. 1992, Sanborn 2000, 2004).

Biogeography

The locations of field sites that I personally visited were determined using a Garmin nüvi 260 GPS receiver. Specimen label data lacking latitude and longitude information were georeferenced primarily using data from the Geographic Names Information System of the United States Geological Survey (http://geonames.usgs.gov/), and in some cases using Google Earth (http://earth.google.com/). Landcover data were from the Southwest Regional Gap Analysis Project (USGS National Gap Analysis Program 2004). The distribution of pinyon-juniper woodlands was estimated by mapping all land cover types that included both pinyon pines (*Pinus edulis, P. monophylla*) and junipers (*Juniperus* spp.) as dominant tree or shrub species (codes S038, S039, S040, S052, and S112). Landcover types with junipers but not pinyon pines and sparsely vegetated types (< 10% plant cover) were excluded. QuantumGIS (Quantum GIS Development Team 2012) was used to produce the distribution map.

Statistical analysis

Acoustic and morphometric data were analyzed in R (R Core Team 2013) using univariate multiple linear regression with categorical predictor variables (i.e., ANOVA models). Morphometric data were modeled with species and sex as predictors, while acoustic data were modeled with species as the sole predictor. For all analyses, preliminary *F*-tests were used to compare models with locality as a predictor (two localities for *T. chiricahua* and the type locality for *T. neomexicensis*) to models that grouped all *T. chiricahua* data together (i.e., used species as a predictor). In all cases, there was not a significant difference between the models (all *p*-values > 0.0788), so the data from the two locations for *T. chiricahua* were grouped together for both the morphometric and acoustic analyses. Plots of the standardized residuals were examined to verify that the data met the model assumptions. Because *T. neomexicensis* is most easily separated from *T. chiricahua* by its distinctive calling song, the statistical analyses only included specimens from localities that had been acoustically surveyed.

Results

Tibicen neomexicensis sp. n.

http://zoobank.org/4847B3E5-22BF-4262-868B-FA06341DB9B4 http://species-id.net/wiki/Tibicen_neomexicensis

Type locality. USA, New Mexico, Lincoln County, Lincoln National Forest, near the junction of Forest Road 105 and State Highway 37, 33.5287°N, 105.6939°W (datum: WGS84), elevation 2188 m, pinyon-juniper forest.

Holotype male. Pinned specimen (Figures 1–3). Original label: "NM: Lincoln Co. | Lincoln NF, FR 105 | 33.5287°N, 105.6939°W | May 31, 2012 7178 feet | Brian and Erin Stucky". UCMC, specimen identifier UCMC 0046172.

Paratypes. 8 males and 3 females, same label data as holotype; 2 males and 2 females, same label data as holotype except collected on May 30, 2012. The paratypes are currently housed in the UCMC and the author's collection. Upon publication, paratypes will also be transferred to the ANSP, the Smithsonian National Museum of Natural History (NMNH), NMSU, and the SEMC.

Description. *Head.* Slightly wider than anterior margin of pronotum. Vertex and frons black, marked with orange-brown on the posterior margin near the eyes and immediately lateral of the lateral ocelli. Supra-antennal plates black dorsally with an orange-brown mark adjacent to the postclypeus, orange-brown ventrally marked with black immediately above the antennae, and orange-brown along the anterior margin except for immediately adjacent to the postclypeus. Antennae mostly black with distal margin of scape yellowish, proximal half of pedicel dark brown in some specimens. Dorsal surface of head sparsely covered with short golden hairs and with longer, silvery-

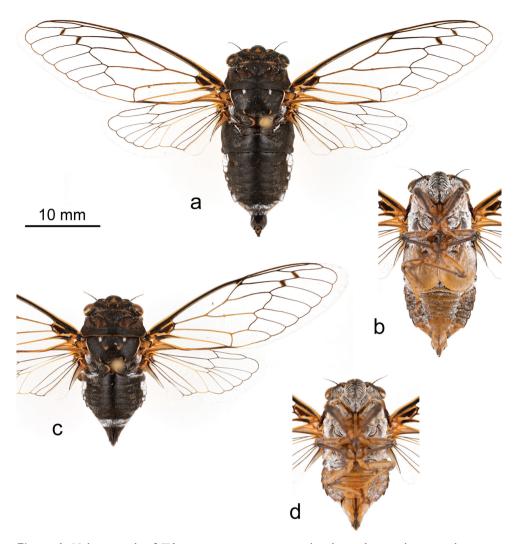


Figure 1. Holotype male of *Tibicen neomexicensis* sp. n.: **a** dorsal view **b** ventral view; and paratype female: **c** dorsal view **d** ventral view.

white hairs behind the eyes. Ventral surface mostly covered with dense, silvery-white hairs. Postclypeus black, marked with orange-brown on the anterior-medial margin and with a triangular orange-brown mark adjacent to the frontoclypeal suture. Transverse grooves of postclypeus lined with pruinosity and silvery-white hairs. Anteclypeus black, yellowish posterolaterally, with a medial brown spot at the junction with the postclypeus. Lora mostly black, marked with yellow along the lateral margins. Genae black anteriorly, yellowish posteriorly where they border the lora. Proximal two thirds of rostrum yellowish, labrum and distal one third of rostrum black, with the apex extending posteriorly to the hind coxae.

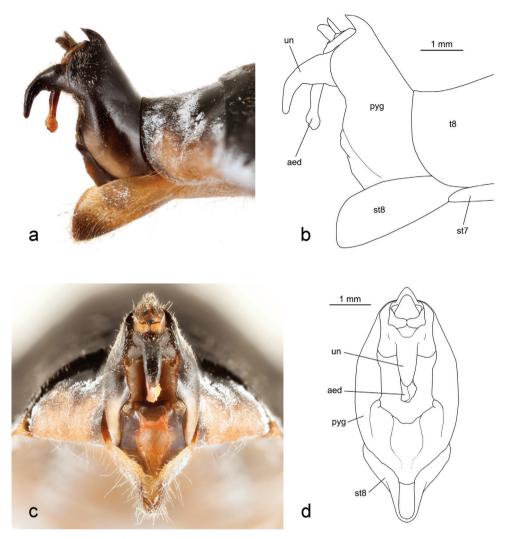


Figure 2. Terminalia of holotype male *Tibicen neomexicensis* sp. n.: **a** and **b** lateral view; and **c** and **d** posterior view. Abbreviations: **aed**–aedeagus, **pyg**–pygofer, **st**–sternite, **t**–tergite, **un**–uncus.

Thorax. Pronotum black, marked faintly with dark brown between the paramedian and lateral fissures and between the lateral fissures and pronotal collar, brown markings often more extensive in females. Pronotal collar black, lined with orange along the anterior margin between the eyes and along the lateral margins, extending to the posterior margin and fading to black medially. Some specimens have the entire posterior margin lined with orange. Pronotum sparsely covered with fine golden hairs. Mesonotum black marked with orange as follows: two J-shaped lines following the parapsidal suture, a small spot at the terminal end of each anterior arm of the cruciform elevation, two C-shaped marks starting at the origin of the anterior arms of the cruciform

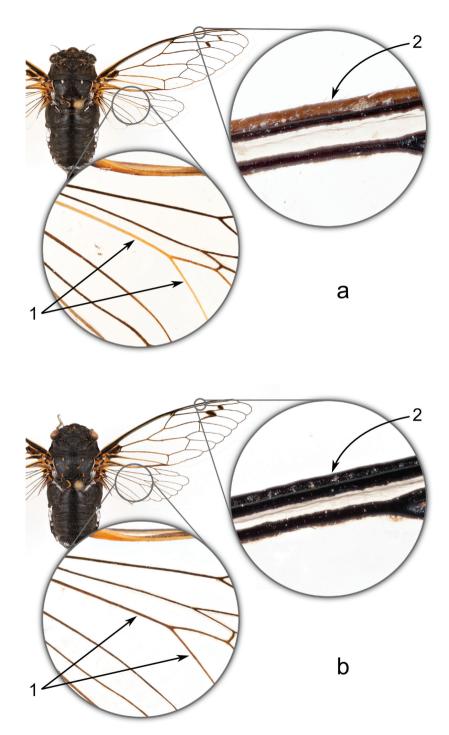


Figure 3. Key morphological features separating *T. neomexicensis* sp. n. (**a**) and *T. chiricahua* (**b**): **1**) the color of the cubitus anterior vein (CuA) and its second branch (CuA₂) in the hind wing, and **2**) the color of the anterior margin of the subcostal vein (Sc) of the fore wing.

elevation and curving medially then laterally towards the posterior arms, and a large mark near the base of each fore wing. Mesonotum with two small pruinose spots on the anterior margin just lateral of the parapsidal sutures, lateral margin also pruinose. Mesonotum sparsely covered with fine golden hairs, with longer silvery-white hairs in the depressions of the cruciform elevation and along the posterolateral margins. Visible portion of metanotum black, covered with silvery-white hairs laterally. Ventral surface of thorax often heavily pruinose and covered with silvery-white hairs, yellowish except for katepisternum 2, anterior portion of basisternum 2, anepimeron 2, central part of katepimeron 2, meron 2, anterior portions of trochantins 2 and 3, episternum 3, and basisternum 3, all of which are black.

Legs. Fore coxae orange marked with brown apically and with the anterolateral surface dark brown except along the margins. Middle and hind coxae orange marked with dark brown laterally. Coxae covered with silvery-white hairs and often pruinose. Trochanters orange, variably marked with brown. Femora orange, apex mostly yellow, brown ventrally, with longitudinal brown stripes that often merge apically and basally. Silvery-white hairs on femora mostly confined to brown markings. Femoral spines brown basally with dark brown apices. Tibiae orange ventrally, brown dorsally with brown markings expanded at the base, covered with silvery-white hairs. Tibial spurs and comb dark brown. Tarsi variable in color but usually dark brown dorsally and light brown to orange ventrally. Claws brown basally with dark brown apices.

Wings. Fore wings hyaline with 8 apical cells, crossveins r and r-m usually strongly infuscated. Costal margin yellow, C vein black, R+Sc vein black with posterior margin pale along the radial cell. Sc vein black beyond the node, subcostal margin brown to dark yellow. Basal cell mostly black, anterior and posterior borders yellow. M vein yellowish-black from its base to the junction with M₁₊₂, black beyond. M₃₊₄ yellowish-black. M₁₊₂ yellowish-black becoming black apically. CuA vein yellow from its base to the junction with CuA₂ yellowish-black. CuP+1A and 2A+3A veins mostly yellow, ambient vein dark yellowish-black, remaining venation black. Hind wings hyaline with 6 apical cells. Sc+RA, RA, CuA between base and CuA₂, and CuA₂ veins mostly yellow to yellowish-black. Ambient vein black marked with yellow along 1st cubital cell and 6th apical cell. Remaining venation mostly black or dark brown. 3rd anal cell gray marked with reddish-orange basally.

Opercula. Male opercula yellowish marked with black on the anterolateral and anteromedial margins, overlapping medially. Posterior margins smoothly rounded, not quite reaching the posterior margin of sternite II. Female opercula yellowish, becoming black anterolaterally. Posterior margin sinuate, reaching the anterior margin of sternite II. Meracanthus black basally with a yellow apex.

Abdomen. Dorsal surface of abdomen almost entirely black, sparsely covered with short golden and silvery hairs. Tergite 8 orange-brown laterally. Tergites 3-7 often marked with orange-brown laterally, markings usually strongest on tergite 3. Timbal covers black, sometimes dark brown centrally, completely concealing timbal. Timbal with 3 long ribs, 4 intercalary ribs, and an incomplete 4th long rib. Dorsal abdomen pruinose at the fol-

lowing locations: along the anteromedial margins of the timbal covers in males; along the anterolateral margins of tergite 2 in females; the lateral margins of tergites 3-7, most prominently on tergite 3; the lateral margins of tergite 8, often extending medially to cover most of the tergite. Sternites orange to yellowish, usually dark brown laterally and anterolaterally. Epipleurites orange to yellowish, indistinctly marked with dark brown or black. Ventroposterior portion of male sternite VIII dark brown.

Male terminalia. Pygofer black, becoming brown or yellowish laterally along the lobes, and with a small brown spot dorsally at the base of the dorsal beak. Dorsal beak not quite as long as anal styles. Anal styles black. Median lobe of uncus slender, black, strongly bent ventrally and terminating in a rounded point. Aedeagus reddish-brown.

Female terminalia. Abdominal segment 9 yellowish-orange ventrally, black dorsally starting at about the lateral mid-line. Dorsal beak about as long as anal styles. Sternite VII yellowish-orange, usually brown laterally, deeply notched at the middle of the posterior margin. Visible portion of gonocoxite IX yellowish-orange, indistinctly marked with brown near the posterior end. Ovipositor sheath black, ventromedial margins partially lined with orange. Ovipositor sheath extends posteriorly about as far as anal styles.

Measurements. All measurements are reported in mm as mean (range, standard deviation). Males (*n* = 11): head width: 8.3 (8.1–8.5, 0.13); pronotum width: 8.9 (8.5–9.3, 0.25); fore wing length: 28.1 (26.8–29.6, 0.76); fore wing width: 9.8 (9.3–10.5, 0.41); body length: 24.9 (23.1–26.3, 1.05). Females (*n* = 5): head width: 7.8 (7.7–7.9, 0.07); pronotum width: 8.4 (8.2–8.5, 0.14); fore wing length: 26.7 (26.2–27.4, 0.48); fore wing width: 9.1 (8.9–9.3, 0.20); body length: 20.0 (19.3–20.9, 0.64).

Etymology. The specific epithet refers to the U.S. state of New Mexico. As far as is currently known, *Tibicen neomexicensis* is endemic to this state.

Morphometric comparison of T. neomexicensis and T. chiricahua

Five morphometric measurements were taken for both *T. chiricahua* and *T. neomexicensis*: fore wing length, fore wing width, head width, pronotum width, and total body length. The correlation coefficient matrix for these five variables revealed that all five measurements were very strongly correlated with one another. All pairwise correlation coefficients excluding body length were > 0.91, and all pairwise correlation coefficients including body length were > 0.80. Body length in adult cicadas is not constant and instead varies according to a cicada's abdominal posture, so the lower correlation coefficients for body length were not surprising. Given the high correlation among the five variables, analyzing each separately would have been largely redundant, so comparative analysis focused on fore wing length (Table 1). Fore wing length is invariant in adult cicadas and easily measured for either live or preserved specimens.

Analysis of the linear model including both species and sex as predictors of fore wing length revealed that this simple model explained much of the variation in size among the cicadas, and that the effects of both predictors were highly significant (R^2

variable	species	mean	95% CI	range	std. dev.	n
	<i>T. chiricahua</i> , M	31.3	30.6-32.0	29.4-33.1	1.06	11
f	T. neomexicanus, M	28.1	27.6–28.6	26.8–29.6	0.76	11
fore wing length (mm)	<i>T. chiricahua</i> , F	29.1	28.0-30.2	28.0-30.1	0.88	5
	T. neomexicanus, F	26.7	26.1–27.3	26.2–27.4	0.48	5
	T. chiricahua	7.12	6.56–7.67	5.73-8.47	0.82	11
peak frequency (kHz)	T. neomexicanus	7.27	7.02–7.52	6.07-7.81	0.45	15
	T. chiricahua	54	52.4–55.6	50.5-59.3	2.32	11
amp. burst rate (bursts/s)	T. neomexicanus	27.8	27.4-28.3	26.5-29.5	0.86	15
. 1 1. 1 1	T. chiricahua	5.02	4.54–5.51	3.92-6.42	0.723	11
pulses per amplitude burst	T. neomexicanus	8.34	7.70-8.98	7.25–11.33	1.066	13
	T. chiricahua	1.72	1.21-2.23	0.49-2.80	0.758	11
phrase 1 length (s)	T. neomexicanus	2.04	1.66-2.42	1.25-3.24	0.6	12
	T. chiricahua	7.82	6.95-8.68	6.02-10.83	1.286	11
phrase 2 length (s)	T. neomexicanus	6.68	5.77-7.59	4.86-10.96	1.575	14
	T. chiricahua	3.75	3.10-4.39	2.37-5.82	0.963	11
phrase 3 length (s)	T. neomexicanus	1.65	1.36-1.94	0.75-2.33	0.479	13

Table 1. Summary statistics for fore wing length and six acoustic variables for *T. neomexicensis* sp. n. and *T. chiricahua* (M = male, F = female).

= 0.805, p < 0.000001 for both variables). After adjusting for the size differences between males and females, the fore wings of *T. neomexicensis* are, on average, about 2.9 mm shorter than those of *T. chiricahua* (95% CI: 2.3–3.5). An *F*-test comparing this simple two-factor model to a model that included a (species sex) interaction term revealed that the two models were not significantly different (F = 1.202, p = 0.282). Therefore, the data show that for the morphometric measurements used in this study, *T. neomexicensis* is significantly smaller than *T. chiricahua*, and that for both species, females are significantly smaller than males. It must be noted, though, that this analysis was limited to localities for which acoustic data were available, and it is possible that these species exhibit greater variation in size across their full ranges.

Description and comparison of the calling songs of T. neomexicensis and T. chiricahua

Calling song of *T. neomexicensis.* The calling song of *T. neomexicensis* can be divided into three phrases, each of which consists of a continuous train of pulses (Figure 4). The first phrase represents the initial increase in amplitude as the cicada begins calling and lasts an average of 2.04 seconds (95% CI: 1.66–2.42; full descriptive statistics for all acoustic parameters are given in Table 1). The second phrase is the main phrase of the call and is produced at or near maximum amplitude. This phrase lasts an average of 6.68 seconds (95% CI: 5.77–7.59) and has a mean peak frequency of 7.27 kHz (95% CI: 7.02–7.52). The first two phrases are characterized by distinctive amplitude and frequency modulations that group pulses into regular

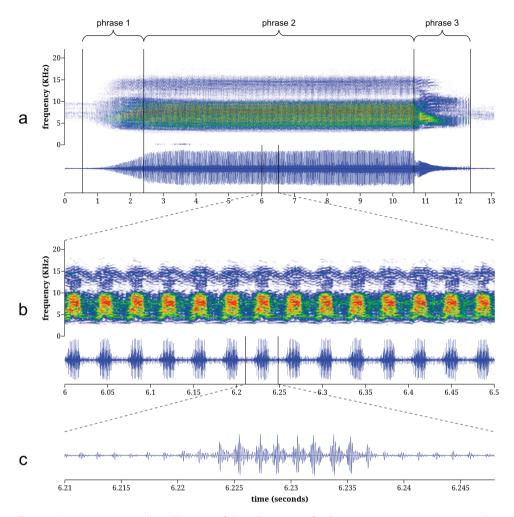


Figure 4. Spectrograms and oscillograms of the calling song of *Tibicen neomexicensis* sp. n.: **a** complete call **b** 0.5 seconds from the middle of phrase 2, illustrating 14 amplitude bursts **c** a single amplitude burst from **b**, illustrating the pulse structure. Spectrograms were generated using a 256-sample Fast Fourier Transform with the Hamming window function.

"bursts" of high amplitude. During the main phrase, these amplitude bursts are delivered at a mean rate of 27.8 bursts/s (95% CI: 27.4–28.3) and each amplitude burst consists of 8.34 pulses on average (95% CI: 7.70–8.98). The amplitude and frequency modulations are accompanied by rapid dorso-ventral movements of the cicada's abdomen. These movements modulate frequency and amplitude by changing the acoustic properties of the sound-producing system (Pringle 1954). The third and final phrase of the call lasts an average of 1.65 seconds (95% CI: 1.36–1.94) and begins with a rapid initial drop in overall amplitude followed by a gradual decrease

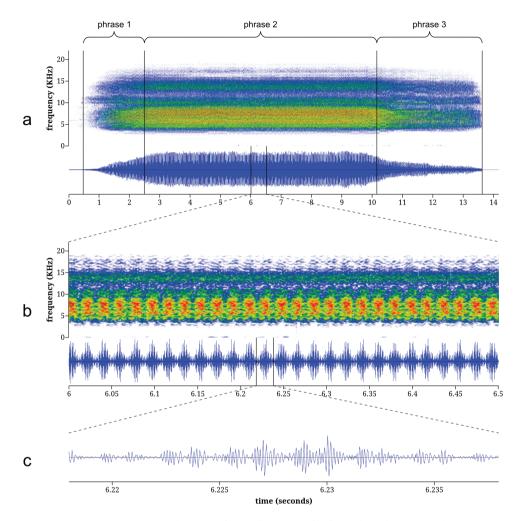


Figure 5. Spectrograms and oscillograms of the calling song of *Tibicen chiricahua*: **a** complete call **b** 0.5 seconds from the middle of phrase 2, illustrating 27 amplitude bursts **c** a single amplitude burst from **b**, illustrating the pulse structure. Spectrograms were generated using a 256-sample Fast Fourier Transform with the Hamming window function.

in amplitude until the calling song ends. During this final phrase, the amplitude and frequency modulations disappear, although the modulations sometimes briefly return as the call terminates.

Calling song of *T. chiricahua*. The calling song of *T. chiricahua* is also naturally divided into three phrases (Figure 5). The first phrase is the initial crescendo as the call begins and lasts an average of 1.72 seconds (95% CI: 1.21–2.23). The second, main phrase of the call has a mean duration of 7.82 seconds (95% CI: 6.95–8.68) with a peak frequency of 7.12 kHz (95% CI: 6.56–7.67). The third phrase is a gradual

decrescendo as the calling song terminates and lasts an average of 3.75 seconds (95% CI: 3.10–4.39). The entire call consists of an amplitude-modulated train of pulses. Pulses are grouped into high-amplitude bursts that, during the main phrase of the call, contain an average of 5.02 pulses per burst (95% CI: 4.54–5.51) and are delivered at a mean rate of 54.0 bursts/s (95% CI: 52.4–55.6).

Comparison of calling songs. Comparison of acoustic parameters, song structure, and physical behavior during call production verified that the calls of these two species are distinct. First, the underlying structures of the amplitude modulations of the calls differ. The mean amplitude burst rate of the call of T. chiricahua is nearly twice that of T. neomexicensis (54.0 and 27.8 bursts/s, respectively, t = 37.4, p < 0.000001), and the amplitude bursts of T. chiricahua contain about 3.3 fewer pulses per burst, on average, than those of *T. neomexicensis* (5.02 and 8.34 pulses/burst, respectively, t = 18.0, p < 1000.000001). There was no overlap in the ranges of observed values for either of these variables. Second, the phrasal structures of the calls also differ. The phrases in the call of T. chiricahua are defined merely by the overall pattern of amplitude changes in the call and have a relatively uniform sound quality throughout, while the third phrase of the call of T. neomexicensis is markedly different in quality from the other two phrases, lacking the characteristic modulations of phrases one and two. Furthermore, the beginning of the third phrase in T. neomexicensis is usually marked by an abrupt drop in amplitude, but the amplitude decreases gradually and smoothly from the second to the third phrases of T. chiricahua. Finally, the amplitude and frequency modulations in the call of T. neomexicensis are a result of rapid dorso-ventral movements of the abdomen during the calling song, but no such movements were apparent in the calling behavior of *T. chiricahua*.

The observed mean peak frequency of the main phrase of the call of *T. neomexicensis* was slightly higher than that of *T. chiricahua*, although the difference was not significant (7.27 and 7.12 kHz, respectively, t = 0.623, p = 0.539). Peak calling song frequency is constrained by body size for most cicadas, with larger cicadas having lower-frequency calls (Bennet-Clark and Young 1994). Thus, given that *T. neomexicensis* is smaller than *T. chiricahua* but the two cicadas are not grossly dissimilar in size, it is not surprising that their peak call frequencies are similar, and even though the difference was not significant, the observed higher pitch of the call of *T. neomexicensis* is consistent with the morphometric analysis.

Geographic distribution

T. chiricahua is more widely distributed than *T. neomexicensis*, ranging from central and southeastern Arizona to southwestern New Mexico (Figure 6). Although not depicted in Figure 6, *T. chiricahua* is also known from Chihuahua, Mexico (Sanborn 2007). *T. neomexicensis* is so far known only from the Sacramento Mountains in south-central New Mexico. All known localities for *T. chiricahua* are west of the Rio Grande, while *T. neomexicensis* has only been found east of the Rio Grande.

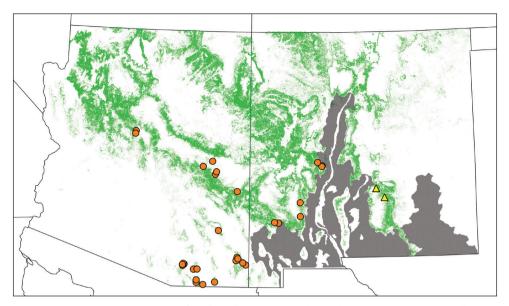


Figure 6. Geographic distribution of *T. chiricahua* (orange circles) and *T. neomexicensis* sp. n. (yellow triangles), estimated from field observations and museum specimens. Green regions indicate pinyon-juniper habitats. The gray region represents the Albuquerque Basin and Chihuahuan Desert. *T. chiricahua* is also found in Mexico (Sanborn 2007).

Four museum specimens, representing two unique collecting localities, could not be conclusively identified. One, a female collected June 15, 1937 in "Big Bend Park," Brewster Co, TX (TAMU), appeared to be *T. neomexicensis*. However, Phillips and Sanborn (2007) did not report any cicadas resembling *T. chiricahua* in their intensive surveys of Big Bend National Park, so this record is doubtful. The other three specimens were two males and one female collected June, 1966 "near" Ciudad Cuauhtémoc, Chihuahua, Mexico (UCMC). These specimens are similar to *T. neomexicensis* and *T. chiricahua*, but differ in that all three have abdomens strongly marked with orange dorsolaterally. More information is needed regarding cicadas from this locality to properly determine their taxonomic status.

Discussion

Tibicen is the second most diverse cicada genus in North America north of Mexico (Sanborn and Heath 2012), and recognition of *T. neomexicensis* increases the number of described species in this region to 32. *T. neomexicensis* belongs to the "southwestern *Tibicen* species," an informal subgroup of *Tibicen* species that differ morphologically from the *Tibicen* cicadas common in the eastern U.S (Davis 1930). These species are only found in the western United States and Mexico.

Diagnosis

Tibicen neomexicensis can be separated from all other North American species of *Tibicen* except for *T. chiricahua* by the combination of its size, almost entirely black dorsal color pattern (Figure 1), and the male's genitalia, particularly the shape of the uncus (Figure 2). Within *Tibicen*, this uncal structure is unique to *T. neomexicensis* and *T. chiricahua* (see Davis 1923 for a figure of the uncus of *T. chiricahua*).

In the field, *T. neomexicensis* and *T. chiricahua* are most easily distinguished by the unique calling songs of the males. Audio recordings of both species are available as online supplementary data for this paper. To human ears, the first and second phrases of the call of *T. neomexicensis* sound like a high-pitched, whiny buzz with easily discernible pulsations that correspond to the amplitude and frequency modulations. At the beginning of the third phrase, the abrupt transition to an unmodulated, uniform whine is perhaps the most aurally distinctive feature of the calling song.

In contrast, the call of *T. chiricahua* sounds like a monotonous, coarse buzz that rapidly increases in amplitude during the first phrase and then slowly fades away during the final phrase. Apart from the amplitude changes in the first and third phrases, there are no obvious changes in sound quality during the course of the call.

Both male and female specimens of *T. neomexicensis* and *T. chiricahua* can usually also be separated by the coloration of the wing venation. In *T. neomexicensis*, the anterior margin of the subcostal vein (Sc) of the fore wing is usually yellowish or at least noticeably lighter in color than the main part of the vein, which is dark black (Figure 3). In *T. chiricahua*, both the vein and its anterior margin are black (Figure 3). In addition, the cubitus anterior vein (CuA) in the hind wing of *T. neomexicensis* is yellow from its base to the junction with its second branch (CuA₂), and the basal two-thirds or more of CuA₂ is usually also yellow (Figure 3). In *T. chiricahua*, these two veins tend to be mostly or entirely black (Figure 3).

Although none of these morphological characters are 100% reliable, when used in combination, they identify nearly all specimens. The color of the margin of the Sc vein is the most reliable single morphological diagnostic character. Out of nearly 200 specimens examined, only 7 might have been misidentified by the color of the Sc vein alone. The color of CuA and CuA₂ is more variable, with some overlap between the two species, and the utility of this character seems to vary among populations of *T. chiricahua*. Unfortunately, in very old specimens, the colors of the wing veins sometimes fade, making identification difficult. Fore wing length can also be used to help confirm an identification, especially when the wing vein colors are ambiguous.

Ecology and behavior

Both *T. neomexicensis* and *T. chiricahua* are associated with pinyon-juniper woodlands, and neither species seems to occur in habitats where both pinyon pines (*Pinus edulis*, primarily) and junipers (*Juniperus* sp.) are absent (B. Stucky, pers. observation; Hast-

ings and Toolson 1991). Although Davis (1925), citing a correspondence from Douglas K. Duncan, reported specimens of *T. chiricahua* collected "on a high mountain plateau ... devoid of any vegetation except many clumps of a large heavy grass," he also noted that, "There is much timber around the edges of this plateau, pine, cedar, and juniper." Overall, records of these cicadas from Arizona and New Mexico closely overlap with the distribution of pinyon-juniper forests in those states (Figure 6).

Specimen label data and field observations indicate that adults of *T. neomexicensis* and *T. chiricahua* emerge in early summer and are mostly gone by the end of July. The earliest record for *T. chiricahua* is May 25 (in 1997) (specimen, CSUC). Although the UAIC has a specimen of *T. chiricahua* from Arizona with the date recorded as "September," the next latest collecting date is July 22 (in 1975) (UAIC), so the September date is either very unusual or in error. The majority of collecting events for *T. chiricahua* were in June, and Hastings and Toolson (1991) reported that June 1–8 was approximately the middle of the adult active season of *T. chiricahua* in the San Mateo Mountains of New Mexico in 1989.

Phenological data for *T. neomexicensis* are much more limited, but consistent with an annual pattern similar to *T. chiricahua*. The earliest record for *T. neomexicensis* is for May 30 (in 2012), at the type locality (B. Stucky, pers. observation), but at this time, there were already large numbers of females ovipositing, so the cicadas must have emerged some number of days earlier. The latest record is from June 7 (in 2005) (specimen, NMSU).

The daily activity patterns of these two cicada species are also similar. Once the sun warms them sufficiently, males of both species will sing throughout much of the day, with peak calling activity occurring from about mid-day through early afternoon (B. Stucky, pers. observation; Hastings and Toolson 1991). Calling activity greatly diminishes during the late afternoon and evening.

Although the nymphal host plants of *T. chiricahua* and *T. neomexicensis* are not known with certainty, there is anecdotal evidence that females of these species have different oviposition preferences. Hastings and Toolson (1991) reported *T. chiricahua* females ovipositing in dead pinyon pine and juniper branches. In contrast, numerous ovipositing females of *T. neomexicensis* were observed at the type locality, most of which were placing their eggs in the dead, dried stems of grasses and forbs, often quite near to the ground.

Both *T. chiricahua* and *T. neomexicensis* are commonly found with *T. duryi* Davis, another species that is specialized on pinyon-juniper habitats (Hastings et al. 1991, Kondratieff et al. 2002). At the type locality, *T. neomexicensis* was also syntopic with *Okanagana bella* Davis.

Relationship with T. chiricahua

T. neomexicensis and *T. chiricahua* are not only extremely similar morphologically, but the shared structure of the male genitalia separates them from all other species of *Tibi*-

cen. It therefore seems probable that *T. neomexicensis* and *T. chiricahua* are sister species, although a broader phylogenetic analysis of *Tibicen* is needed to confirm this.

Today, these species are apparently entirely allopatric, separated from one another by the uninhabitable Albuquerque Basin and Chihuahuan Desert. This might be a relatively recent phenomenon, though. At the time of the last glacial maximum, pinyons and junipers were widespread across much of what is today the Chihuahuan Desert (Betancourt et al. 1993, Lanner and Van Devender 1998, Thompson and Anderson 2001). As the climate changed at the end of the Pleistocene and beginning of the Holocene, pinyons and junipers retreated to higher elevations and became extirpated from modern desert regions (Betancourt et al. 1993). Moreover, climatological data and models suggest that this pattern probably repeated multiple times during the Pleistocene (Paillard 1997, Petit et al. 1999, Smith and Farrell 2005).

These habitat changes must have certainly affected the distributions of and interactions among the ancestors of modern *T. neomexicensis* and *T. chiricahua*. What impact, if any, this had on population divergence and speciation is unknown. However, theory predicts that secondary sexual traits can diverge rapidly in allopatry (Pomiankowski and Iwasa 1998), and if *T. neomexicensis* and *T. chiricahua* are sister species, it seems likely that geographic isolation caused by habitat shifts played at least some role in their evolution.

Conclusions

Acoustic, morphometric, and behavioral data all indicate that the cicadas resembling *Tibicen chiricahua* from New Mexico's Sacramento Mountains should be recognized as a distinct species, described here as *T. neomexicensis*. In particular, analysis of audio recordings confirms that the calls of these two species have significant, consistent structural and temporal differences, which provide the simplest means for identifying these cicadas in the field.

With the discovery of *T. neomexicensis*, the North American *Tibicen* are now known to encompass at least three complexes of morphologically cryptic species with distinct male calling songs: the *chiricahua* group [*T. chiricahua* and *T. neomexicensis*], the *dorsatus* group [*T. dorsatus* (Say) and *T. tremulus* Cole], and the *pruinosus* group [*T. linnei* (Smith & Grossbeck), *T. pruinosus* (Say), and *T. robinsonianus* Davis]. A phylogeographic and divergence-time analysis of the North American *Tibicen* species based on molecular data could not only help clarify the relationship between *T. neomexicensis* and *T. chiricahua*, but also shed light on the broader patterns of diversification for one of the most species-rich cicada genera in North America.

The geographic ranges of these species are still rather poorly documented, especially in Mexico, where *T. chiricahua* is currently known only from a single specimen (Sanborn 2007). Furthermore, compared to *T. chiricahua*, few localities are known for *T. neomexicensis*. Thus, additional field work is needed to clarify the distributions of these species. For *T. neomexicensis*, mountain ranges near the Sacramento Mountains that also have pinyon-juniper habitats, such as the Capitan Mountains to the northeast and Guadalupe Mountains to the southeast, are obvious targets for further exploration.

Acknowledgments

I am grateful to the following individuals for providing information about specimens, photographs, loans, or access to collections: Sandra Brantley (UNMC), Graeme Davis (NMSU), Boris Kondratieff (CSUC), Sangmi Lee (ASUT), Carl Olson (UAIC), Ed Riley (TAMU), Virginia Scott (UCMC), Jennifer Thomas (SEMC), and Jason Weintraub (ANSP). I thank the Bowers Lab at the University of Colorado, Rob Guralnick, Erin Stucky, and two anonymous reviewers for helpful feedback on the manuscript and figures. Erin Stucky also assisted with the fieldwork in NM and AZ. Publication of this article was generously funded by the University of Colorado Boulder Libraries Open Access Fund.

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Appendix I

Calling song of *Tibicen neomexicensis* sp. n. (doi: 10.3897/zookeys.337.5950.app1) File format: Waveform Audio File Format (wav).

Explanation note: Recorded on May 31, 2012, at the type locality for *T. neomexicensis*. Details of the recording methods are provided in the main text.

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Citation: Stucky BJ (2013) Morphology, bioacoustics, and ecology of *Tibicen neomexicensis* sp. n., a new species of cicada from the Sacramento Mountains in New Mexico, U.S.A. (Hemiptera, Cicadidae, *Tibicen*). ZooKeys 337: 65–87. doi: 10.3897/zookeys.337.5950 Calling song of *Tibicen neomexicensis* sp. n. doi: 10.3897/zookeys.337.5950.app1

Appendix II

Calling song of *Tibicen chiricahua* Davis. (doi: 10.3897/zookeys.337.5950.app2) File format: Waveform Audio File Format (wav).

Explanation note: Recorded on June 1, 2012, at the type locality for *T. chiricahua*. Details of the recording methods are provided in the main text.

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Citation: Stucky BJ (2013) Morphology, bioacoustics, and ecology of *Tibicen neomexicensis* sp. n., a new species of cicada from the Sacramento Mountains in New Mexico, U.S.A. (Hemiptera, Cicadidae, *Tibicen*). ZooKeys 337: 65–87. doi: 10.3897/zookeys.337.5950 Calling song of *Tibicen chiricahua* Davis. doi: 10.3897/zookeys.337.5950.app2

RESEARCH ARTICLE



Two new species of Chaco Tullgren from the Atlantic coast of Uruguay (Araneae, Mygalomorphae, Nemesiidae)

Laura Montes de Oca^{1,2,†}, Fernando Pérez-Miles^{1,‡}

I Sección Entomología, Facultad de Ciencias, Universidad de la República. Iguá 4225. CP 11400. Montevideo, Uruguay 2 Laboratorio de Etología, Ecología y Evolución, Instituto de Investigaciones Biológicas Clemente Estable. Av. Italia 3318. CP 11600. Montevideo, Uruguay

† http://zoobank.org/D7C8FE76-27B1-407D-94AB-956D19CB381E
‡ http://zoobank.org/088FDA03-DD34-4D32-8342-7AAF91880EBC

Corresponding author: Laura Montes De Oca (laumdeo@fcien.edu.uy)

Academic editor: *Matjaž Kuntner* | Received 12 June 2013 | Accepted 3 September 2013 | Published 1 October 2013 *http://zoobank.org/57B57379-3CF0-436B-9FD9-B5C3BC61D78E*

Citation: Montes de Oca L, Pérez-Miles F (2013) Two new species of *Chaco* Tullgren from the Atlantic coast of Uruguay (Araneae, Mygalomorphae, Nemesiidae). ZooKeys 337: 73–87. doi: 10.3897/zookeys.337.5779

Abstract

We describe two new species of the nemesiid spider genus *Chaco* from Rocha Province, Uruguay. These new species are diagnosed based on genital morphology, male tibial apophysis spination, and burrow entrance. We test cospecificity of one species, *C. costai*, via laboratory mating experiments. The new species are diagnosed and illustrated and habitat characteristics, and capture behavior are described. We conduct a cladistic analysis based on a previously published morphological character matrix that now includes the newly described species.

Resumen

En este trabajo se describen dos nuevas especies de arañas, nemésidas del género *Chaco* recolectadas en el departamento de Rocha, Uruguay. Las nuevas especies se distinguen de las conocidas por caracteres tomados de la morfología de la genitalia, número de espinas en la apófisis tibial de los machos y la tapa-cortina en la cueva. Se realizó un análisis filogenético de las especies del género *Chaco* incluyendo las nuevas especies en una matriz previa.

Keywords

Spiders, Taxonomy, Cladistics, *Chaco*, Nemesiidae, Natural history

Introduction

The family Nemesiidae Simon, 1889 comprises 43 genera (Platnick 2013) of medium sized spiders that have a transverse fovea, eyes grouped on a tubercle, 2-4 short spinnerets, anterior tarsi without spines, tarsi III and IV with light or absent scopula, without claw tufts and superior tarsal claws bipectinate with numerous teeth. Several authors have questioned nemesiid monophyly (Raven 1985, Goloboff 1993, 1995, Hedin and Bond 2006). Although morphological characters are somewhat equivocal, molecular data seem to support the group (Bond and Hedin 2006, Hedin and Bond 2006, Bond et al. 2012). Goloboff (1995) conducted the last major revision of the South American species of this family; since, only a few contributions to the systematics of the Nemesiidae family have been made (Indicatti and Lucas 2005; Indicatti et al. 2008; Lucas et al. 2008; Lucas and Indicatti 2010). Nemesiids live in silk tubes under stones or logs, or in burrows that are covered by a trapdoor (Goloboff 1995). In Uruguay the family is represented by four genera: Acanthogonatus Karsch, 1880; Pycnothele Chamberlin, 1917; Stenoterommata Homberg, 1881 and Chaco Tullgren, 1905. Due to a number of life history characteristics, they are difficult to collect and consequently little is known about their biology. Montes de Oca and Pérez-Miles (2003) reported the presence of Chaco Tullgren, 1905 in Uruguay but did not identify the species.

The genus *Chaco* was originally described on the basis of the type species *Chaco* obscura Tullgren, 1905 known from a female specimen. The species was characterized as having posterior lateral spinnerets with two apical segments of equal length and the absence of labial cuspules (Tullgren 1905). Goloboff (1987, 1995) diagnosed Chaco using the following character combination: four short spinnerets, eight eyes grouped on a tubercle, anterior legs with few spines, anterior tarsi scopulate, without spines and no claw tufts; tarsal claws with numerous teeth in two rows. Males of the genus can be distinguished by having a distal prolateral spur on tibia I comprising three or more spines, absence of inferior claw on all tarsi, patella III with 1-1-1 spines and anterior tibia without scopula. Females of Chaco are distinguished from other nemesiid genera by the absence of the following features: ITC from all legs; spigots with pumpkin-like socket; scopula on the anterior tibia (present in Prorachias and Pselligmus) and patella IV spination. Additionally Chaco females lack the autapomorphies indicated for the Diplothelopsini: anterior median eyes much larger than the minute posterior median eyes, posterior eye row slightly recurved, and short, wide caput (Goloboff 1995).

The characteristics of some individual specimens discovered along the coast of Uruguay have the diagnostic characteristics described for *Chaco* but differ from all the known species. In this article we describe, diagnose, and illustrate two new species: *Chaco castanea* sp. n. and *Chaco costai* sp. n. We present a cladistic reanalysis of the genus with newly described species and present some natural history data for the new taxa.

Material and methods

Specimens were examined using an Olympus SZH stereomicroscope. The description of color was based on live organisms when possible. Abbreviations: AME anterior median eyes, ALE anterior lateral eyes, PME posterior median eyes, PLE posterior lateral eyes, OQ ocular quadrangle, P prolateral, R retrolateral, D distal, STC superior tarsal claw, ITC inferior tarsal claw, FCE-MY Collection of Facultad de Ciencias, Entomología -Mygalomorphae. All measurements are in mm and were taken with an ocular micrometer. Total body length excludes chelicerae and spinnerets. Lengths were measured along a dorsal longitudinal line and widths were taken at maximum values. The OQ length was measured from the anterior edge of ALE to the posterior edge of PLE; the sternum length from the posterior angle to the labium edge (Goloboff 1995). Terminology for spination is modified from Pérez-Miles and Weinmann 2010. The formula gives the number of spines in the following order: dorsal-prolateral-retrolateral-ventral (p indicates a pair of spines that occur at this position). Spermathecae were cleared with clove oil and illustrated in dorsal view. Left male palpal bulb was removed from the cymbium and illustrated in prolateral and retrolateral views. Specimens were photographed using a Lumenera INFINITY 3 camera.

Cladistic analysis. We scored the newly described species (*Chaco castanea* and *C. costai*) for 32 characters (Table 1) from Goloboff (1995), removing invariant characters and adding five new ones (characters 27–31). Maxillary cuspules were codified separately for males and females (characters 9 and 10) to account for sexual dimorphism. The ingroup comprised 9 taxa: *Chaco castanea* sp. n.; *C. costai* sp. n.; *C. obscura* Tullgren, 1905; *C. patagonica* Goloboff, 1995; *C. sanjuanina* Goloboff, 1995; *C. tecka* Goloboff, 1995; *C. tigre* Goloboff, 1995 and *C. tucumana*

											1										2										3	\square
	0	1	2	3	4	5	6	7	8	9	0	1	2	3	4	5	6	7	8	9	0	1	2	3	4	5	6	7	8	9	0	1
L. longipes	0	1	1	2	0	0	0	0	0	0	0	0	2	1	0	1	1	1	1	0	0	0	1	1	0	0	0	0	0	1	0	?
D. bonariensis	0	1	1	2	0	0	0	0	0	0	0	1	2	1	?	1	0	1	1	0	0	0	1	0	0	0	0	?	0	0	0	?
C. calderoni	0	1	0	1	1	0	0	1	2	2	0	0	1	0	0	1	0	1	0	0	0	0	0	2	1	0	0	1	0	0	0	?
C. obscura	0	0	0	1	1	0	1	0	1	1	1	1	1	0	0	1	0	0	0	0	0	1	1	1	1	0	1	0	0	0	0	0
C. tucumana	0	0	0	1	1	0	1	0	2	1	1	1	1	0	0	1	0	0	0	0	0	1	1	1	1	0	1	0	0	0	0	0
C. socos	1	0	0	1	1	0	1	0	2	2	1	0	1	0	1	1	1	0	0	0	1	1	1	1	1	1	1	1	0	0	0	0
C. tigre	1	0	0	1	1	0	1	0	1	1	1	0	1	0	1	1	0	0	0	0	1	1	1	1	1	1	1	0	0	0	0	1
C. patagonica	0	0	0	0	1	1	0	1	0	?	1	0	0	?	1	1	?	?	0	1	1	1	?	?	?	1	0	0	0	?	?	?
C. tecka	0	0	1	1	1	1	1	1	1	?	1	0	0	?	1	1	?	?	0	0	1	1	?	?	?	1	0	1	0	?	?	?
C. sanjuanina	0	0	0	0	1	1	0	1	1	1	1	0	0	0	1	1	0	0	0	1	1	1	0	1	0	1	?	0	0	0	0	1
C. castanea	0	0	0	1	1	1	0	0	2	1	1	1	1	0	1	0	0	0	0	0	0	1	1	1	1	0	?	2	0	0	0	1
C. costai	0	0	0	1	1	0	0	0	2	2	1	0	1	0	0	0	0	0	0	0	1	1	1	1	1	0	0	0	1	1	1	0

Table 1. Data matrix for the genus *Chaco* and the outgroup.

Goloboff, 1995. The outgroup included: *Chilelopsis calderoni* Goloboff, 1995; *Diploth-elopsis bonariensis* Mello-Leitão, 1938 and *Lycinus longipes* Thorell, 1894. The outgroup taxa are considered close relatives of *Chaco* (Goloboff, 1995). The tree was rooted using *Lycinus longipes* Thorell, 1894. *Chaco melloleitaoi* (Bücherl, Timotheo & Lucas, 1971) is not included in the analysis because it in fact belongs to a different genus (Indicatti et al. 2011). It differs from *Chaco* by the presence of two prolateral megaspines on male first tibia and in the palpal organ morphology.

The data matrix (Table 1) was constructed using Nexus Data Editor ver 0.5.0 software (Page 2001). The cladistic analysis was carried out with the program TNT version 1.1 (Goloboff et al. 2003), using maximum parsimony as the optimality criterion. Tree searches were conducted using implicit enumeration and implied weighting (Goloboff 1993) with concavity indices (k) ranging from 1 to 6.

Characters scored. (0) Clypeus: 0, wide; 1, narrow (1) PE row: 0, recurved; 1 procurved (2) Eyes: 0, AME and PME subequal size; 1, AME much larger than PME (3) Pubescence: 0, absent; 1, light; 2, dense (4) Sternum: 0, wide; 1, normal; 2, narrow (5) Sternal sigilla: 0, conspicuous; 1, inconspicuous (6) Leg color: 0, uniform; 1, patterned (7) Setae on female posterior legs: 0, normal; 1, dense (8) Maxillary cuspules in females: 0, few (0-10); 1, medium (11-30); 2, many (over 30) (9) Maxillary cuspules in males: 0, few (0-10); 1, medium (11-30); 2, many (more than 30) (10) Rastellum: 0, weak; 1, strong (11) Female tarsi: 0, rigid; 1, flexible (12) Scopula IV: 0, absent/ very light; 1, light; 2, dense (13) Trichobothria on male cymbium: 0, medial third; 1, basal half (14) PMS spigot number: 0, many; 1, few (15) Male metatarsus IV: 0, 1-1-1P SUP; 1, 0-0-1P SUP (16) Dorsal spines in male palpal tibia: 0, absent; 1, present (17) Spines on male patella I-II: 0, 0/1P; 1, 1-1-1P (18) Female patella IV: 0, 0/1P; 1, 1-1-1P (19) Spines on female tarsi IV: 0, absent; 1, present (20) Spines on female tibia/ metatarsus I: 0, short; 1, long (21) Male tibial spur: 0, absent; 1, present (22) Male palpal tibia: 0, short; 1, long (23) Male bulb keels: 0, absent; 1, parallel keels or ridges along embolous base; 2, lateral keels or flanges (24) Male bulb duct: 0, basal portion evenly curved; 1, basal portion strongly sinuous (25) Female spermathecae: 0, no basal sphere; 1, with basal sphere (26) Habits: 0, flap door; 1, trap door. (27) Spermathecae fundus 0, subspherical; 1, reniphorm (28) female tibiae 0, normal; 1, short (29) Setae on male cymbium 0, thin hair like setae; 1, thickened setae (30) Two long dorsal setae on palpal tibiae setae 0, absent; 1, present (31) Spines on male tibial apophysis 0, five or less; 1, more than 5.

Results and discussion

Cladistic results

Analysis of the morphological data using implied weighting and implicit enumeration resulted in 2 most parsimonious trees. Topologies were stable across K values 1-6 (62 steps, CI= 0.61, RI= 0.65, K=1, fit= 8.75, K=6, fit= 2.79; Fig. 5A-B). The genus *Chaco* was recovered as monophyletic including the two new species, supported by 4 characters: PE row procurved (1), strong rastellum (10), spination of male patellae I-II (17) and the presence of male palpal tibial spur (21). The main difference between the topology of the two recovered trees was: *C. socos* + *C. tigre* is the sister group of *C. obscura* + *C. tucumana* in one tree, while in the other *C. socos* + *C. tigre* is the sister group of (*C. teka* (*C. patagonica*, *C. sanjuanina*)). The consensus tree (Fig. 5C) recovers a polytomy for *C. castanea*, *C. obscura*, *C. tucumana*, and the clades (*C. obscura*, *C. tucumana*) and (*C. teka* (*C. patagonica*, *C. sanjuanina*)). Based on these data the monophyly of the genus *Chaco* appears to be well supported with the inclusion of the new species; the addition of new characters in the future will be necessary to improve the resolution of relationships among several species. Regarding the biology of *C. costai*, the flap-like door of the burrow may be explained as an adaptation to sandy soil habitat.

Natural History

Chaco costai specimens are typically found in sandy soils of oceanic and river coastal areas associated with psammophyte vegetation (Fig. 2F). Individuals were collected from tubular vertical burrows of about 100mm length; the entrance diameter is about 10mm. The spider closes the burrow entrance with sand and silk when disturbed. *Chaco patagonica, C. costai* make a burrow that is covered with a thin, flaplike door (Fig. 2E). The door actually consists of a prolongation of the silk layer lining the interior of the burrow, covered by grains of sand; it is flexible and loosely articulated. According to field and laboratory observations, the spider begins foraging at night by standing at the top of the burrow with legs I-III extended lying in the substrate (Fig. 2E); similar to that reported by Bond and Coyle (1995) for *Ummidia*. After a prey item is captured, the spider returns to the interior of the burrow to feed; the burrow entrance remains open until later when the spider returns to the entrance to repair and close the flap-like- door.

A copulation event observed in the laboratory occurred over an eight minute time period at 18 °C. The male appeared to initiate courtship with body vibrations and pulling silk threads with his chelicerae. Body vibrations were caused by spasmodic contraction of legs I and II. The male approached the female burrow entrance and opened it with his chelicerae; the female then emerged from the burrow. Copulation took place at the burrow entrance; the male clasped his tibial apophysis with female chelicerae. The male performed 23 palpal insertions, alternating right and left palps. The mean duration of the insertions was 21.09±12.73 seconds. After copula the male retreated with legs I extended and female retreated in the burrow but maintained her first legs out towards the entrance. After 17 minutes the female closed the burrow flap-door.

Taxonomy

Family Nemesiidae Simon, 1889 Genus *Chaco* Tullgren, 1905

Chaco castanea sp. n.

http://zoobank.org/42AA4036-8DA2-4C0C-8AAB-92B5A15649DC http://species-id.net/wiki/Chaco_castanea Figs 1A–D, 3 A–E

Types. Male holotype (deposited in FCE-MY 0767) from, Rocha, Perla de Rocha, 34°25.0'S; 53°51.0'W, i.2001, coll. G. Calixto Female paratype (deposited in FCE-MY 0770) from Rocha, Cabo Polonio, 34°24.0'S; 53°47.0'W, 24.i–18.iii.2003, coll. F. Achaval. **Additional material examined.** Male from Rocha, Cabo Polonio, 19.xii.2003–18.iii.2004, coll. F. Achaval. 1m (deposited in FCE-MY 0769), Female from Rocha, Perla de Rocha, 34°25.0'S; 53°51.0'W, i.2001, coll. G. Calixto, 1f (FCE-MY 0766), female from Rocha, Cabo Polonio, 34°24.0'S; 53°47.0'W, 18.i–18. iii.2005, coll. F. Achaval, 1f (deposited in FCE-MY 0797).

Etymology. The specific epithet is a noun taken in apposition (chestnut) and is in reference to the brownish coloration of this species.

Diagnosis. Males (Fig. 1A) uniquely possess a tibial apophysis with 4 spines (Fig. 3D). *Chaco castanea* males differ from *C. tigre* and *C. socos* males by having a palpal organ with a sinuous spermophor and parallel longitudinal ridges (Figs 3A–C,) and by having a PME and AME that are subequal in diameter. Females of *C. castanea* (Fig. 1C) differ from the other known *Chaco* species by the presence of a large reniform spermathecal receptacle in combination with a short sinuous duct (Fig. 3E).

Description. Male (holotype). Total length 7.75; carapace 4.13 long, 3.27 wide; eight eyes grouped on a tubercle, AME 0.17; PME 0.13; ALE 0.16; PLE 0.16; OQ 0.86 long, 0.44 wide; clypeus 0.06; fovea 0.63; sternum oval, 1.93 long, 1.56 wide. Posterior sigillae sub-circular, sub-marginal. Labium sub-rectangular, 0.2 long, 0.56 wide; labial cuspules absent; 18/20 maxillary cuspules (Fig. 1B); chelicerae with 7 promarginal teeth of similar size; rastellum with 26 short, thick conical setae on promargin narrowing through retromargin. Leg, palpal measurements in Table 2; chaetotaxy in Table 3. Tarsus I-IV scopula entire; metatarsi I-II distal third, III-IV absent. Anterior tibiae without scopula. STC with numerous teeth in two lateral rows. ITC I-IV absent. Claw tufts absent. Tibial apophysis with 4 prolateral apical spines (Fig. 3D). Palpal organ spermophor strongly sinuous (Figs 3A–C). Four spinnerets, PMS short monoarticulated, PLS triarticulated apical article short, domed. Spigots without pumpkin-like socket. Body, legs light brown, abdomen with dark brown spots.

Female (paratype). Total length 18.2; carapace 7.2 long, 5.6 wide; caput raised; eight eyes grouped on a tubercle, AME 0.24; PME 0.25; ALE 0.36; PLE 0.31; OQ 0.75 long, 1.6 wide; clypeus 0.14; fovea 0.9; sternum oval, 2.3 long, 2.2 wide. Pos-



Figure 1. *Chaco castanea*. **A** Male holotype habitus, dorsal view **B** Male holotype maxillae and labium showing cuspules **C** Female paratype habitus, dorsal view **D** Female paratype maxillae and labium showing cuspules.

terior sigillae sub-circular, sub-marginal. Labium sub-rectangular, 0.7 long, 1.3 wide; 1 labial cuspule; maxillary cuspules 48/62 (Fig. 1D). Chelicerae with 6 promarginal teeth; first tooth smaller than second, decreasing thereafter; 10 retromarginal denticles; rastellum with 45 short, thick conical setae on promargin. Leg, palpal measure-

	Fe	Pa	Ti	Mt	Ta	Total
Palp	2	0.83	1.71	-	0.83	5.37
Ι	3.4	2	2.53	2.67	2.07	12.67
II	3.27	1.67	0.87	2.67	2.2	10.68
III	3	1.45	2.1	3.17	2.43	12.15
IV	3.93	1.6	3.73	4.33	2.93	16.52

Table 2. Length of legs palpal segments of the holotype male of *Chaco castanea*.

Table 3. Spination of legs and palps of holotype male Chaco castanea.	
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	Fe	Pa	Ti	Mt	Ta
Palp	2-1-1-0	0	0-3-1-0	_	0
Ι	4-1-1-0	0	0-2-0-6	0-1-1-3	0
II	5-2-3-0	0-1-0-0	0-2-0-3	0-1-1-3	0
III	0-0-0-6	0-3-1-0	6-2-2-6	3-4-4-6	0
IV	6-6-3-0	0-1-1-0	3-2-3-6	3-3-2-6	0

Table 4. Length of legs and palpal segments of the paratype female of Chaco castanea.

	Fe	Pa	Ti	Mt	Ta	Total
Palp	3.6	2.1	2.2	-	2.2	10.1
Ι	4.7	2.8	3.1	2.8	2.0	15.4
II	4.2	2.7	3.0	2.8	1.9	14.6
III	3.6	2.4	2.3	3.5	2.4	14.2
IV	5.0	3.2	4.2	5.1	2.9	20.4

Table 5. Spination of legs and palps of paratype female Chaco castanea.

	Fe	Pa	Ti	Mt	Ta
Palp	0-1-0-0	0-1-0-0	0-4-1-6	_	0
Ι	0-1-0-0	0-0-0-0	0-0-0-1	0-0-0-3	0
II	0	0-2-0-0	0-2-0-2	0-1-0-2	0
III	0-1-0-0	0-3-1-0	1-2-2-2	2-2-3-6	0
IV	1-0-1-0	0-0-0-0	0-2-3-6	0-2-3-8	0

ments in Table 4; chaetotaxy in Table 5. Tarsus I-IV scopula entire, metatarsus I-II entire, III–IV absent. Anterior tibiae without scopula. STC with numerous teeth in two lateral rows. ITC I-IV absent. Claw tufts absent. Palpal claw with 4 teeth in prolateral median line. Two spermathecal receptacles, single sinuous neck; reniform fundus (Fig. 3E). Four spinnerets, PMS short monoarticulated, PLS triarticulated apical article short, domed. Spigots without pumpkin-like socket. Coloration as in male.

Distribution. Uruguay, Rocha, Perla de Rocha and Cabo Polonio.

Chaco costai sp. n.

http://zoobank.org/65418A73-632C-4AEF-8CE9-2740E0F09EC4 http://species-id.net/wiki/Chaco_costai Figs 2A–F, 4A–E

Types. Male holotype (FCE-MY 1007) female paratype (FCE-MY 1006) and from Rocha, Perla de Rocha, 34°25.63'S, 53°52.27'W, 26-28.xii.2011, coll. A. Laborda, C. Castro, L. Montes de Oca.

Etymology. The specific epithet is a patronym in honor of Fernando G. Costa, a recognized Uruguayan arachnologist who greatly contributed to the knowledge of spiders and has inspired many colleagues and students.

Diagnosis. Males of *C. costai* (Fig. 2A) differ from the other species of the genus, except *C. obscura*, by the presence of numerous spines (7–10) on tibial apophysis (Fig. 4D); they can be distinguished from *C. obscura* by having a shorter embolous (Figs 4A–C). Female *C. costai* specimens (Fig. 2C) differ from most species of *Chaco* by having spermathecae with a sinuous neck (Fig. 4E). The species is distinguished from the geographically proximate species *C. castanea* by having a longer spermathecal neck and from *C. obscura* by the sinuous neck. *Chaco costai* differ from all other species of the genus (except *C. patagónica* and *C. tecka*) by having a flap door that covers the burrow (Fig. 2E).

Description. Male (holotype). Total length 14.9 ; carapace 7.7 long, 6.2 wide; eight eyes grouped on a tubercle, AME 0.28; PME 0.23; ALE 0.38; PLE 0.36; OQ 0.79 long, 1.04 wide; clypeus 0.22; fovea transverse, slightly procurve 1.5; sternum oval, 3.1 long, 2.8 wide. Posterior sigillae sub-circular, sub-marginal. Labium sub-rectangular, 1.2 long, 3.5 wide; 4 labial cuspules; 42/38 maxillary cuspules (Fig. 2B); chelicerae with 2 row of teeth, 6 promarginal, 5 retromarginal denticles; rastellum with 16 short, thick conical setae on promargin. ITC absent. Claw tufts absent. Leg, palpal measurements in Table 6; chaetotaxy in Table 7. Tarsus I-III scopula entire, IV divided; metatarsi I 4:5D, II 3:4 D, III 1:5 D, IV absent. Anterior tibiae without scopula. STC with numerous teeth in two lateral rows. Tibial apophysis with 7-10 prolateral apical spines (Fig. 4D). Palpal tibia with 2 dorsal long thin setae. Palpal organ spermophor very sinuous (Figs 4A–C). Four spinnerets, PMS short monoarticulated, PLS triarticulated apical article short, domed. Spigots without pumpkin-like socket. Cephalothorax, legs dorsally light brown, and ventrally dark brown; abdomen lighter with dark brown pattern.

	Fe	Pa	Ti	Mt	Ta	Total
Palp	3.9	1.9	2.4	_	1.2	9.4
Ι	6.7	3.8	4.7	5.5	3.8	24.5
II	5.9	3.5	4.4	5.3	3.8	22.9
III	5.0	3.2	3.8	5.7	4.4	22.1
IV	7.2	3.5	6.1	8.0	4.9	29.7

Table 6. Length of legs palpal segments of the holotype male of *Chaco costai*.



Figure 2. *Chaco costai*. **A** Male habitus, dorsal view **B** Male maxillae and labium showing cuspules **C** Female habitus, dorsal view **D** Female maxillae and labium showing cuspules **E** *Chaco costai* female ambushing in the burrow entrance, see the flap-like door **F** Habitat of *C. costai* showing psammophyte vegetation.

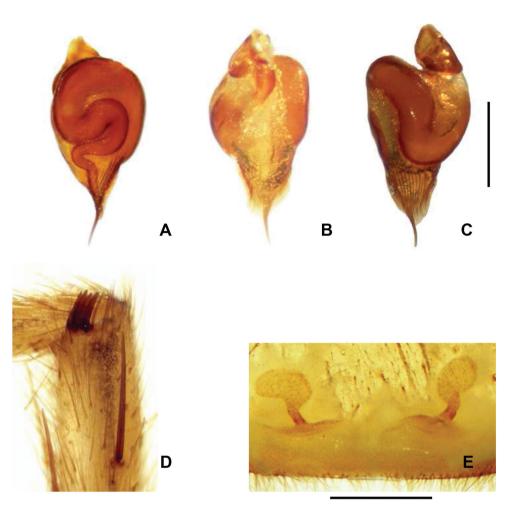


Figure 3. *Chaco castanea*. **A** Palpal organ, dorsal view **B** Palpal organ, retrolateral view **C** Palpal organ prolateral view **D** Male tibial apophysis with 4 spines, prolateral view **E** Spermathecae, dorsal view. Scale = 5mm.

Female (paratype). Total length 19.9; carapace 7.1 long, 5.7 wide; caput raised; eight eyes grouped on a tubercle, AME 0.2; PME 0.17; ALE 0.36; PLE 0.35; OQ 0.63 long, 1.16 wide; clypeus 0.27; fovea slightly procurved 1.10; sternum oval, 3.4 long, 2.9 wide. Posterior sigillae sub-circular, sub-marginal. Labium sub-rectangular, 0.47 long, 0.97 wide; 3 labial cuspules; 43/36 maxillary cuspules (Fig. 2D). Chelicer-ae with 8 promarginal teeth; 9 retromarginal denticles; rastellum with 18 short, thick conical setae on promargin. Leg, palpal measurements in Table 8; chaetotaxy in Table 9. Tarsus I-II scopula entire, III- IV divided by a wide band of longer conical setae, metatarsus I complete, II 2:3, III–IV absent. Anterior tibiae without scopula. STC with numerous teeth in two lateral rows. ITC I-IV absent. Claw tufts absent. Palpal claw with 4 teeth in median line. Two spermathecal receptacles, single sinuous long neck; sub-espheric fundus (Fig. 4E). Four spinnerets, PMS short monoarticulated,

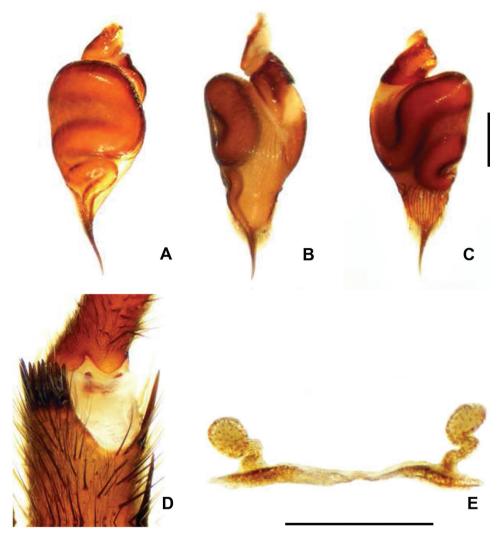


Figure 4. *Chaco costai*. A Palpal organ, dorsal view **B** Palpal organ retrolateral view **C** Palpal organ prolateral view **D** Male apophysis with 10 spines, prolateral view **E** Spermathecae, dorsal view. Scale = 5mm.

Table 7. Spination of legs and palps of male *Chaco costai*. The formula gives the number of spines in the following order: dorsal–prolateral–retrolateral–ventral.

	Fe	Pa	Ti	Mt	Ta
Palp	2-0-0-0	0-2-0-0	0-4-1-0	_	0
Ι	8-0-0-0	0-1-0-0	0-2-2-5	0-2-1-4	0
II	9-1-0-0	0-2-0-0	0-4-0-6	2-3-1-6	0
III	8-1-0-0	0-3-1-0	4-2-0-7	9-4-3-7	1-0-0-0
IV	9-0-0-0	0-1-1-0	4-1-4-6	7-3-3-8	1-0-0-0



Figure 5. Results from cladistics analyses. **A–B** Most parsimonious trees obtained by TNT (implied weighting). Length = 62, CI = 0.61, RI = 0.65, K =1 Fit = 10.33, K = 6 Fit = 3.48 **C** Strict consensus of cladograms **A** and **B**.

	Fe	Pa	Ti	Mt	Ta	Total
Palp	3.8	1.9	2.1	—	1.8	9.6
Ι	4.8	3.3	2.8	2.7	2.1	15.6
II	3.3	2.2	1.7	2.2	1.8	11.2
III	3.7	2.6	1.4	2.9	2.5	13.1
IV	3.8	2.3	2.8	3.1	2.0	14

Table 8. Length of legs and palpal segments of the paratype female of Chaco costai.

Table 9. Spination of legs and palps of female Chaco costai.

	Fe	Pa	Ti		Ta
Palp	0-1-0-0	0-4-0-0	0-1-0-9	—	0
Ι	2-0-0-0	0-1-0-0	0-2-0-4	0-0-0-4	0
II	1-0-0-0	0-1-0-0	2-0-0-3	1-1-0-5	0
III	2-0-0-0	0-3-1-0	1-2-2-5	1-4-3-7	0
IV	0-0-0-0	0-0-0-0	0-0-2-2	0-3-3-8	0

PLS triarticulated apical article short, domed. Spigots without pumpkin-like socket. Cephalothorax, legs brown, abdomen very light brown with darker dots.

Distribution. Only known from the type locality.

Acknowledgements

We thank F. Achaval, G. Calixto for collecting the first specimens and M.J. Albo, C. Castro, A. Laborda, L. Ziegler for their help in the fieldwork. We are also deeply indebted with Dr. J. Bond and Dr. P. Goloboff for their critical comments which help us to improve the manuscript.

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