RESEARCH ARTICLE



# Four new species of Desmiphorini (Coleoptera, Cerambycidae, Lamiinae)

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#### Abstract

Four new species of Desmiphorini are described: *Desmiphora (Desmiphora) orozcoi*, from Belize; *Estola wappesi*, from Bolivia; *Estola imitatrix*, from Bolivia; and *Gyrpanetes clarkei*, from Bolivia. A key to the species of *Gyrpanetes* is provided, and *E. wappesi* and *E. imitatrix* are included in an existing key.

## Keywords

Belize, Bolivia, Taxonomy

# Introduction

According to Monné (2014), Desmiphorini Thomson, 1860 included 73 genera and 462 species in the Neotropical Region. Recently, Monné and Wappes (2014) described a new genus and species of Desmiphorini. Thus, the total number of genera is now 74, with 463 species. *Desmiphora (Desmiphora)* Audinet-Serville, 1835, and *Estola* Fairmaire & Germain, 1859 are the genera with the largest number of species, at 67 and 95, respectively. Even so, new species are frequently described in these genera. In this report, we described one new species in *Desmiphora (Desmiphora*) and two in *Estola* genera.

*Gyrpanetes* Martins & Galileo, 1998, is a small genus with three described species, all of which are known only from Brazil. The new species described herein is the first record of this genus outside of Brazil.

## Material and methods

Photographs were taken with a Canon EOS Rebel T3i DSLR camera, Canon MP-E 65mm f/2.8 1–5× macro lens, controlled by Zerene Stacker AutoMontage software.

The acronyms used in the text are as follows:

ACMT	American Coleoptera Museum (James E. Wappes), San Antonio, Texas, USA
MNKM	Museo de Historia Natural Noel Kempff Mercado, Santa Cruz, Bolivia
MZSP	Museu de Zoologia, Universidade de São Paulo, São Paulo, Brazil
USNM	National Museum of Natural History, Washington, DC, USA.

#### Taxonomy

## Desmiphora (Desmiphora) orozcoi sp. n.

http://zoobank.org/4D71CB8E-CD03-4C5D-87B3-1FA9CDBC6C0B Figs 1, 2, 3

**Description.** Holotype female. Integument black; palpi reddish-brown; basal antennomeres dark-brown, gradually light-brown towards antennomere XI; tibiae and tarsi brown.

Head. Frons transverse; coarsely, abundantly, deeply punctate; pubescence short, yellowish-brown, distinctly not obliterating integument, mixed with long, moderately abundant setae. Area between antennal tubercles and vertex with punctures as on frons; yellowish-brown pubescence distinctly longer, denser than on frons, mixed with sparse small spots of white pubescence, and long, abundant setae. Coronal suture clearly distinct from clypeus to anterior edge of prothorax. Antennal tubercles with pubescence and setae as on frons, but smaller punctures. Area around eyes with narrow band of white pubescence. Area behind upper eye lobes with short, yellowish-brown, moderately sparse pubescence; region behind interconnection area of eyes lobes glabrous; area behind lower eye lobes with short, sparse yellowish-brown pubescence; along area closer to eyes, long, sparse setae. Genae partially striated, transversely with sparse yellowish-brown pubescence, more concentrated in some regions; with long, sparse setae. Gula shiny, glabrous, except for narrow band of yellowish-brown pubescence close to anterior margin. Distance between upper eye lobes equal to 0.75 times length of scape; distance between lower eye lobes equal to 1.25 times length of scape. Antennae as long as 1.2 times elytral length; reaching about distal fifth of elytra; scape, pedicel and antennomeres with long, yellowish-white setae throughout (longer ventrally; shorter on distal antennomeres); antennomeres IV-X with basal ring of whitish



Figures 1–6. 1–3 *Desmiphora (Desmiphora) orozcoi*, holotype female: 1 Dorsal habitus 2 Ventral habitus; 3 Lateral habitus. 4–6 *Estola wappesi*, holotype female: 4 Dorsal habitus 5 Ventral habitus 6 Lateral habitus.

pubescence; antennal formula based on antennomere III: scape = 0.77; pedicel = 0.20; IV = 0.92; V = 0.47; VI = 0.46; VII = 0.43; VIII = 0.38; IX = 0.33; X = 0.27; XI = 0.38.

Thorax. Pronotum moderately fine, sparsely punctate (punctures mostly obliterated by pubescence); yellowish-white dense pubescence, forming large "M-like" shape on disc; on each top of "M-like" there is distinct tuft; remaining surface with sparser yellowish pubescence; with long, moderately abundant brown and yellowish-white setae. Sides of prothorax with large, conical tubercle; pubescence yellowish, dense, mixed with long, abundant setae; coarsely, sparsely punctate. Prosternum and prosternal process with short, moderately sparse, yellowish pubescence, mixed by long, sparse setae. Metasternum laterally with yellowish pubescence (white depending on light), centrally glabrous. Scutellum glabrous on central base, laterally with yellowish-white pubescence. Elytra. Moderately coarsely, sparsely punctate on basal third (laterally denser), gradually finer towards apex; pubescence yellowish-brown (distinctly more abundant on basal third and near apex) and white (distinctly more abundant on basal two-thirds of distal half), not forming erect tufts; with long, abundant dark-brown and yellow setae; apex truncate, with outer angle rounded. Legs. Femora with yellowish-brown pubescence, absent on some areas; with long setae. Tibiae and tarsi with long setae.

Abdomen. Urosternites with yellowish pubescence, laterally denser (mainly on urosternite I); distal margin of urosternites I–IV with fringe of yellow pubescence; urosternites II–IV laterally with small spot of whitish pubescence; urosternite V deeply, triangularly depressed at apex.

**Dimensions in mm (female).** Total length, 9.6; length of prothorax at center, 2.2; greatest width of prothorax (between apices of tubercles), 2.6; anterior width of prothorax, 1.9; posterior width of prothorax, 2.0; humeral width, 2.8; elytral length, 6.8.

**Type material.** Holotype female, BELIZE, *Cayo District*: Las Cuevas Research Station (580m; 16°43.971'N, 88°59.196'W), 1–4.VI.2008, Ratcliffe, Cave, Jameson and Orozco col. (USNM).

**Etymology.** The species is named after Jesus Orozco (University of Nebraska State Museum, Nebraska, USA), one of the collectors of the holotype of the new species.

**Remarks.** Desmiphora (Desmiphora) orozcoi differs from all other species recorded from Central America and the Caribbean due to the absence of erect tufts of pubescence on elytra. The new species is somewhat similar to D. (D.) intonsa (Germar, 1824), but differs as follows: elytra do not have tufts of pubescence (present in D. (D.) intonsa); elytral apex without dark area, distinctly contrasting with all other regions of the surface (present in D. (D.) intonsa); elytral punctures on basal half not distinctly coarse (very coarse in D. (D.) intonsa). It can be separated from D. (D.) niveocincta (Lane, 1959), by the presence of light pubescence on center basal half of pronotum, by the absence of dense spots with white pubescence on the basal center half of the pronotum, the elytra have dense spots with white pubescence (the middle one forming a decumbent tuft), and the legs are pale.

The key by Breuning (1974) is based on many misidentifications, therefore it is not possible to modify it to include this species without a full revision.

#### Estola wappesi sp. n.

http://zoobank.org/B82CAF00-2F62-4ED8-8FA9-D11D592569D6 Figs 4, 5, 6

**Description.** Holotype female. Integument dark-brown, almost black; palpi brown; antennomeres III, V–VII, IX–XI with reddish-brown basal ring; basal two-thirds of antennomere IV reddish-brown, dorsally interrupted by incomplete brown ring; an-

tennomere VIII totally yellowish-white; tibiae brown on about basal half; metatarsi mostly reddish-brown.

Head. Frons transverse; coarsely, abundantly punctate; pubescence brown, moderately dense, not obliterating integument, mixed with small, sparse spots of white pubescence; with long, moderately abundant setae. Area between antennal tubercles and vertex with punctures, pubescence and setae as on frons. Coronal suture distinct from clypeus to level of posterior margin of eyes. Antennal tubercles elevated; finely punctate; pubescence and setae as on frons. Area behind eyes moderately coarsely punctate; pubescence white close to the eyes, brown towards prothorax; with some long setae. Genae with whitish pubescence, more brownish towards frons. Gula shiny, glabrous, except for narrow band of brown pubescence close to anterior margin. Lower eye lobes longer than twice length of genae; distance between upper eye lobes equal to 0.5 times length of scape; distance between lower eye lobes equal to length of scape. Antennae as long as 1.4 times elytral length; reaching elytral apex at about middle of antennomere X; light areas of antennomeres with yellowish-white pubescence; scape and pedicel with sparse, long setae throughout; antennomeres III-X ventrally with sparse, long setae; antennal formula based on antennomere III: scape = 1.20; pedicel = 0.40; IV = 1.52; V = 1.08; VI = 1.04; VII = 0.96; VIII = 1.12; IX = 0.64; X = 0.56; XI = 0.52.

Thorax. Pronotum coarsely, deeply, abundantly punctate; disc with three distinct gibbosities, two sub-rounded, placed antero-laterally, another elongate, placed centerbasally; pubescence brown, mixed with spots of white pubescence; with long, sparse setae. Sides of prothorax with acute, distinct tubercle about middle; pubescence brownish, mixed with whitish pubescence; coarsely, abundantly punctate. Prosternum, coarsely, moderately abundantly punctate; pubescence mostly brownish. Prosternal process with brownish pubescence. Metasternum laterally moderately coarsely, sparsely punctate; pubescence yellowish-brown, not obliterating integument. Scutellum centrally with brown pubescence, laterally with white pubescence. Elytra. Coarsely, densely, deeply punctate on basal two-thirds, sparser, finer on apical third; pubescence brown, mixed with spots of white pubescence, more abundant on distal third, forming irregular, transverse bands; with long, moderately abundant setae; apices individually rounded. Legs. Pubescence yellowish-brown, more whitish in some areas.

Abdomen. Urosternites with yellowish-brown pubescence, not obliterating integument.

Variability. Coronal suture inconspicuous between clypeus and antennal tubercles; pubescence behind eyes mostly whitish; antennae in male as long as 1.6 times elytral length; elytral spots of white pubescence somewhat variable in amount and distribution.

**Dimensions in mm (male/female holotype).** Total length, 4.90-5.20/5.50; length of prothorax at center, 1.00-1.10/1.10; greatest width of prothorax (between apices of tubercles), 1.20-1.40/1.40; anterior width of prothorax, 1.05-1.10/1.15; posterior width of prothorax, 1.05-1.10/1.15; humeral width, 1.60-1.60/1.85; elytral length, 3.60-3.65/4.10.

**Type material.** Holotype female, BOLIVIA, *Santa Cruz*: Refugio Los Volcanes (4 km N Bermejo; 18°06'S, 63°36'W; 1045–1350 m), 11–17.XII.2012, Wappes and

Skillman col. (MNKM). Paratypes – 2 males, same data as holotype (MZSP, ACMT); 1 male, same data as holotype, except for: 1000 m, 16–21.X.2007, J. Wappes & A. Cline col. (ACMT); Chaco above Achira (Florida province; Vicoquin Area; 18°07'S / 63°47'W; 1730 m), 1 male, 1 female, 22–25.I.2007, Wappes & Lingafelter col. (USNM, male; ACMT, female).

**Etymology.** The species is named after James E. Wappes (ACMT), for his friendship and constant help with the specimens studied.

**Remarks.** *Estola wappesi* differs from *E. boliviana* Breuning, 1940, as follows: lower eye lobes longer than twice the length of genae; antennomere III mostly dark; elytra with areas of white pubescence. In *E. boliviana* the lower eye lobes are shorter than twice the length of the genae, the antennomere III is whitish-yellow, and the elytra do not have areas with white pubescence. It differs from *E. strandiella* Breuning, 1942, mainly by the dark elytra, slightly attenuate towards apex (reddish-brown and more attenuated towards the apex in *E. strandiella*). *Estola wappesi* differs from *E. longeantennata* Breuning, 1940, by the dark elytra, by the light pubescence of elytra that is more abundant on the distal half, and by the antennomere VIII that is totally whitish-yellow. In *E. longeantennata* the elytra is reddish-brown, the light pubescence on elytra is abundant throughout, and the antennomere VIII is whitish-yellow only on basal third.

*Estola wappesi* can be included in the alternative of couplet "52", from Breuning (1974) (translated):

52	Lateral spine of prothorax very thin and sharp52'
_	Lateral spine of prothorax conical
52'	Light pubescence of elytra abundant throughout; antennomere VIII whitish-
	yellow only at basal third; elytra reddish-brown
	Estola longeantennata Breuning, 1940
_	Light pubescence of elytra more abundant on distal half; antennomere VIII
	totally whitish-yellow; elytra dark-brown

#### Estola imitatrix sp. n.

http://zoobank.org/9A488BEB-5972-4796-B8D3-2A1AC130CF05 Figs 7, 8, 9

**Description.** Holotype female. Integument black, except for: antennomere III reddish on basal third; antennomeres IV and VI reddish on basal two-thirds; antennomere V reddish on a narrow ring at base; tibiae with wide reddish ring on basal half, close to middle, and another covering approximately entire distal third; protarsomeres I–II mostly reddish; meso- and metatarsomeres I–III mostly reddish.

Head. Frons transverse; coarsely, moderately abundantly punctate; pubescence yellowish-brown, centrally mixed with white pubescence; with long, moderately abundant setae. Vertex moderately coarsely, abundantly punctate (punctures mostly obliterated by pubescence); pubescence yellowish-brown, mixed with whitish pubescence,



Figures 7–12. 7–9 *Estola imitatrix*, holotype female: 7 Dorsal habitus 8 Ventral habitus 9 Lateral habitus 10–12 *Gyrpanetes clarkei*, holotype male: 10 Dorsal habitus 11 Ventral habitus 12 Lateral habitus.

more distinctly on central tuft; with long, thick setae. Coronal suture marked from clypeus to anterior edge of prothorax, but mostly obliterated by pubescence. Antennal tubercles with yellowish-brown pubescence. Area behind eyes with yellowish-brown pubescence, more whitish, slightly denser closer to eyes; with long, yellowish setae behind lower eye lobes, mainly near eyes. Genae with yellowish-brown pubescence, mixed with sparse long setae. Gula glabrous, with very fine, sparse punctures towards prothorax; punctures slightly more distinct and sparse short setae present towards anterior edge; area close to anterior edge with band of yellowish-brown pubescence. Lower eye lobes about 2.5 times longer than genae; distance between upper eye lobes equal to 0.55 times length of scape; distance between lower eye lobes equal to length of scape. Length of antennae from base of scape to apex of antennomere VI equal

to 0.90 times elytral length (lacking antennomeres VII-XI); scape and pedicel with sparse, long setae throughout; antennomeres III–VI ventrally with sparse, long setae; antennal formula based on antennomere III: scape = 1.23; pedicel = 0.42; IV = 1.34; V = 1.11; VI = 1.00.

Thorax. Prothorax transverse. Pronotum coarsely, abundantly punctate (punctures partially obliterated by pubescence); pubescence yellowish-brown, moderately dense, on central triangular area, wide oblique band with white, dense pubescence on basal half, remaining surface with white pubescence, mixed with yellowish-brown pubescence; with long, thick, sparse setae (mainly anteriorly). Sides of prothorax with short, conical tubercle slightly before middle; coarsely, abundantly punctate; pubescence on tubercle white; remaining surface with yellowish-brown pubescence, denser towards ventral side of thorax, mixed with white pubescence on some areas. Ventral side of thorax with yellowish-brown pubescence, mixed with long setae (mainly centrally on prosternum and mesosternum). Scutellum centrally with brown pubescence, laterally with yellowish-brown pubescence. Elytra. Coarsely, densely, deeply punctate on basal half, sparser on distal half; pubescence, yellowish-brown, mixed with white pubescence; the latter forming a basal band, following along suture and connected with wide, transverse, somewhat distinct band slightly after middle, not reaching lateral margin; with long, thick, moderately abundant setae throughout; apex individually rounded. Legs. With long, yellow, moderately abundant setae.

Abdomen. Urosternites with yellowish-brown pubescence.

**Dimensions in mm (female).** Total length, 5.75; length of prothorax at center, 1.20; greatest width of prothorax (between apices of tubercles), 1.65; anterior width of prothorax, 1.40; posterior width of prothorax, 1.45; humeral width, 2.30; elytral length, 4.30.

**Type material.** Holotype female from BOLIVIA, *Tarija*: 300m S Palo Marcado (21°28'S, 63°08'W; *ca.* 300m; edge of Pilcomayo River; dry Chaco Forest; beaten dry *Acacia* tree), 12.XII.2007, R. Clarke & S. Zamalloa col. (MNKM).

**Etymology.** Latin, *imitatrix* = a female imitator; allusive to similar appearance to *E. basiflava* Breuning, 1943.

**Remarks.** Estola imitatrix differs from *E. densepunctata* Breuning, 1940, and *E. basiflava* as follows: body is wider; antennomere III, distinctly shorter than scape; antennomere III, light only on basal third; and pronotum has bands of white pubescence. In both species the body is slightly narrower, the antennomere III is about as long as the scape (sometimes slightly shorter) and dark, at most, on distal third (32 specimens of *E. densepunctata*, and 2 of *E. basiflava* were examined), and the pronotum has no bands with white pubescence. It differs from *E. compacta* Breuning, 1940, by the white pubescence on the base of elytra, prolonged along the suture (yellowish and not prolonged in *E. compacta*), and by the antennomere III being widely-ringed with black (entirely yellowish in *E. compacta*). *Estola imitatrix* can be separated from *E. fusco-marmorata* Breuning, 1940, by the body being shorter and wider (narrower and more elongated in *E. fuscomarmorata*), by the antennomere III being distinctly shorter than

*Estola imitatrix* can be included in the alternative of couplet "6", from Breuning (1974) (translated; modified):

6'	Pronotum and elytra distinctly spotted with white pubescence
_	Pronotum and elytra not spotted with white pubescence
6(6')	Only the extreme base of the elytra with pale yellow pubescence
	<i>E. basiflava</i> Breuning, 1943
_	Basal fourth of elytra with pale yellow pubescence
	<i>E. flavobasalis</i> Breuning, 1940
	<i>L. flavooasalis</i> breuning, 19

## Gyrpanetes clarkei sp. n.

http://zoobank.org/C2257CD2-E942-4922-8504-5B44E706849D Figs 10, 11, 12

**Description.** Holotype male. Integument dark-brown, almost black on some areas, except for: base of antennomere III–X (about basal half on antennomeres V–IX), gula, and peduncle of femora reddish-brown; one large, irregular, orangish area on each half of elytron.

Head. Frons transverse, fine, densely punctate; pubescence yellowish-brown, abundant, but not obliterating integument, denser, yellowish close to lower eye lobes. Antennal tubercles moderately elevate; sculpture as on frons; pubescence yellowish-brown towards inner side, more yellowish close to eye. Coronal suture distinct from clypeus to anterior edge of prothorax. Vertex with sculpture and pubescence as on frons. Area behind eyes with band of yellowish pubescence close to eyes, gradually glabrous towards anterior edge of prothorax. Genae with yellowish pubescence (more whitish depending on angle of light). Gula shiny, glabrous, except for band of yellowish pubescence close to anterior margin. Distance between upper eye lobes equal to 0.35 times length of scape; distance between lower eye lobes equal to 0.65 times length of scape. Antennae as long as 2.0 times elytral length; reaching elytral apex about base of antennomere VII; pedicel and antennomeres III–VI with long, dark setae on ventral side (sparser towards antennomere VI); light area of antennomeres with yellowish pubescence; antennal formula based on antennomere III: scape = 0.78; pedicel = 0.19; IV = 0.89; V = 0.73; VI = 0.65; VII = 0.56; VIII = 0.51; IX = 0.46; X = 0.40; XI = 0.38.

Thorax. Pronotum fine, densely punctate; disc with whitish-yellow pubescence (more whitish depending on angle of incision of light), shorter and slightly sparser on area forming "M-like shape" (less distinct depending on the viewing angle). Sides of prothorax with whitish-yellow pubescence, shorter and slightly sparser on central subrounded area; sculpture as on pronotum. Ventral side of thorax with yellowish-brown pubescence (more whitish depending on angle of light), not obliterating integument. Elytra. Center basal region without crest; sparse, coarsely punctate on basal half (denser around scutellum), distinctly sparser on distal half; with thick, moderately abundant, very long, dark setae throughout; pubescence whitish-yellow (more whitish depending on angle of incision of light), whitish and yellowish-brown, forming irregular drawing; around some punctures of distal half, pubescence forming ocellar macula; with two slightly distinct longitudinal dashed lines with whitish pubescence, more distinct around middle; apex obliquely truncate, with sutural angle rounded. Legs. Pubescence as on urosternites.

Abdomen. Pubescence on urosternites whitish-yellow (more whitish depending on angle of light), not obliterating integument

Variability. Gula and peduncle of femora brown.

**Dimensions in mm (male).** Total length, 4.10–4.35; length of prothorax at center, 0.80–0.85; greatest width of prothorax, 1.05–1.10; anterior width of prothorax, 0.85–0.90; posterior width of prothorax, 0.85–0.90; humeral width, 1.25–1.35; ely-tral length, 3.00–3.10. The smallest dimensions are those of the holotype.

**Type material.** Holotype and paratype male from BOLIVIA, *Tarija*: 24 km W Villamontes (21°21'S, 63°37'W; *ca.* 800 m; Valley Rio Isiri; Submontane Chaco Forest), 11.I.2008, R. Clarke & S. Zamalloa col. (holotype, MNKM; paratype, MZSP).

**Etymology.** The species is named after Robin O. S. Clarke, one of the collectors of the type specimens.

**Remarks.** *Gyrpanetes clarkei* differs from *G. oriba* Galileo & Martins, 2003, as follows: antennomere III is reddish only at base; pronotal pubescence forming "M-like" macula; elytral dashed lines with whitish pubescence partially distinct; elytral pubescence has areas distinctly exposing larger and more distinct integument (dark areas). In *G. oriba* the antennomere III is dark only at the apex, the pronotal pubescence does not form "M-like" macula, the elytral dashed lines are distinct from base to near apex, and the elytral pubescence is more distinctly whitish throughout.

Martins and Galileo (1998) included a key to separate the two species of the genus. It is necessary to provide a new key to include the four species of the genus:

1	Center basal crest of elytra distinctly elevated. Brazil (Paraná)
	<i>G. pukuaba</i> Martins & Galileo, 1998
_	Center basal crest of elytra not elevated 2
2(1)	Elytra with very sparse long setae. Brazil (Espírito Santo)
	G. cacapira Martins & Galileo, 1998
_	Elytra with distinct long setae throughout
3(2)	Annnomere III mostly reddish; pronotum without "M-like" macula; elytral
	dashed lines distinct from base to near apex. Brazil (Espírito Santo)
	G. oriba Galileo & Martins, 2003
_	Antennomere III mostly dark; pronotum with "M-like" macula; elytral
	dashed lines slightly distinct (more distinct around middle). Bolivia

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RESEARCH ARTICLE



# Termitotrox venus sp. n. (Coleoptera, Scarabaeidae), a new blind, flightless termitophilous scarab from Cambodia

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## Abstract

*Termitotrox venus* **sp. n.** is described from Cambodia and represents the second discovery of *Termitotrox* Reichensperger, 1915 from the Indo-Chinese subregion of the Indomalayan region. Most of the type series was collected from refuse dumps in fungus garden cells of *Macrotermes* cf. *gilvus* (Hagen, 1858). *Macrotermes* Holmgren, 1910 was previously an unknown host of *Termitotrox* species. The new species is easily distinguished from all known congeners by having wing-shaped trichomes on the elytra and the larger body size, at 2.5 mm in length. We also describe the mouthparts to complement the description of the genus *Termitotrox*.

#### **Keywords**

Termitophily, Termitotroginae, Termitotrogini, Isoptera, Termitidae, Macrotermitinae, *Macrotermes*, new species, Indo-Chinese subregion, mouthparts

# Introduction

Members of the genus *Termitotrox* Reichensperger, 1915 are blind, flightless termitophilous scarabs associated with the fungus-growing termite genera *Protermes* Holmgren, 1910, *Odontotermes* Holmgren, 1912 or *Hypotermes* Holmgren, 1917 (Isoptera, Termitidae,

Macrotermitinae). The genus was previously known from the Ethiopian region (eight species) and the Indian subregion (two species) of the Indomalayan region (Krikken 2008), until the discovery of *Termitotrox cupido* Maruyama, 2012 from Cambodia, representing the first species of *Termitotrox* from the Indo-Chinese subregion of the Indomalayan region (Maruyama 2012a). Recently, we collected another undescribed species of *Termitotrox* in Cambodia from fungus garden cells of *Macrotermes* Holmgren, 1910 (also Macrotermitinae) – a previously unknown host of *Termitotrox*. This is the second discovery of the genus in the Indo-Chinese subregion of the Indomalayan region. In this paper the new species is described and biological information about it is provided.

## Materials and methods

In August 2014, we examined fungus gardens of the termite genera Macrotermes, Microtermes Wasmann, 1902, Odontotermes and Hypotermes in Siem Reap, Cambodia. After examining more than 300 fungus gardens, we found 8 Termitotrox beetles from fungus garden cells of Macrotermes cf. gilvus (Hagen, 1858) and Hypotermes makhamensis Ahmad, 1965, seven specimens in seven cells of three colonies of M. cf. gilvus and one specimen from one cell of *H. makhamensis*. The beetles were put in a killing tube (35 ml) with tissue paper and ethyl acetate; a day later they were removed from the tube and kept in 80% ethanol. All specimens were dried and mounted for morphological observation. Dissected genitalia and mouthparts were mounted in Euparal on a small glass plate (10×5 mm), and subsequently glued onto a paper card (6×5 mm) and pinned under the respective specimen (Maruyama 2004). A permanent mount of mouthparts was also made. Specimen photographs were taken using a Canon EOS 60D with a Canon MP-E 65 mm 1-5× macro lens and Kenko extension tubes and stacked using CombineZP software. Images of living beetle were taken using a Canon EOS 7D with a EF 100mm F2.8L Macro lens and Kenko extension tubes. Terminology of the species description follows Krikken (2008). All measurements in the paper are given in millimeters as follows: minimum length – maximum length (mean ± SD). The type series is deposited in Maruyama collection in the Kyushu University Museum, Fukuoka, Japan.

## Taxonomy

# Genus Termitotrox Reichensperger

Figs 5-8

- *Termitotrox* Reichensperger 1915: 16 (type species: *Termitotrox consobrinus* Reichensperger, 1915, by monotypy).
- *Aphodiocopris* Arrow 1920: 432 (type species: *Aphodiocopris minutus* Arrow, 1920, by monotypy).

Additional description. Maxillae (Fig. 5) small; mala toothed distally; basistipes and cardo with long setae on lateral side. Maxillary palpus 4-segmented and well developed; segment I small, bent outwards; segment II about 2 times as long as segment I; segment III small, only slightly longer and broader than segment I, slightly bent inwards; segment IV large, approximately twice as long as segment II; numerous digitiform sensillae present on ventrolateral side of proximal half of segment IV. Labial palpus strongly reduced. Mandibles (Figs 6, 7) asymmetrical, pointed apically, numerous serrate ridges on molar surface. Epipharynx (Fig. 8) with anterior margin feebly bisinuate, epitorma almost indistinct, pedia almost glabrous, chaetoparinae very strong and elongate.

**Comments.** See Krikken (2008) for generic review. No detailed mouthparts description has previously been provided for *Termitotrox*. Although this additional description is based on only two species, *T. cupido* and *T. venus*, the other members of *Termitotrox* are expected to share the same or similar character states based on their overall similarity of external morphology.

## Termitotrox venus Kakizoe & Maruyama, sp. n.

http://zoobank.org/8174A036-4FCC-4957-B788-2541A6CA13D1 Figs 1–13

**Type materials.** Holotype, male, deposited in Maruyama collection in the Kyushu University Museum: 1.0 km south of Angkor Wat, Angkor, Siem Reap, Cambodia, 22 VIII 2014, S. Kakizoe leg. Paratypes, deposited in Maruyama collection in the Kyushu University Museum: 1 female, 1.6 km southwest of Angkor Wat, Angkor, Siem Reap, Cambodia, 20 VIII 2014, S. Kakizoe leg.; 1 male, 1.0 km south of Angkor Wat, Angkor, Siem Reap, Cambodia, 24 VIII 2014, S. Kakizoe leg.; 2 males, 2 females, 0.77 km east of Angkor Wat, Angkor, Siem Reap, Cambodia, 24 VIII 2014, S. Kakizoe leg.; 1 female, 24 VIII 2014, S. Kakizoe leg.; 1 male, 1.0 km south of Angkor Wat, Angkor, Siem Reap, Cambodia, 24 VIII 2014, S. Kakizoe leg.; 2 males, 2 females, 0.77 km east of Angkor Wat, Angkor, Siem Reap, Cambodia, 24 VIII 2014, M. Maruyama & S. Kakizoe leg. (In fungus garden cells of *Macrotermes* cf. *gilvus*); 1 female, 1.7 km east of Neak Pean, Angkor, Siem Reap, Cambodia, 25 VIII 2014, S. Kakizoe leg. (In fungus garden cells of *Hypotermes makhamensis*).

Distribution. Northwestern Cambodia.

**Etymology.** *Venus* is the goddess of fertility, beauty and love in ancient Roman mythology and is often illustrated together with *Cupido*. The new species was found in the area where *Termitotrox cupido* was also found. Therefore, this species is named *Venus*. Noun in apposition.

**Diagnosis.** This species is similar to *Termitotrox cupido* in having the wing-shaped trichomes on the elytra but easily distinguished from it by the larger body and the development of the pronotal basomedian section and the elytral median projection.

**Description of holotype male.** General color (Figs 1–4, 11–13) uniformly dark brown, matt, body length 2.46 mm. *Head* (Figs 1–4). Surface generally evenly convex, only with a slight callosity at clypeofrontal transition. Lateral margin of head entirely, finely marginate. Clypeal outline evenly rounded. Clypeofrons brown, glabrous, dis-



Figures 1–4. Male habitus of *Termitotrox venus* sp. n. 1 dorsal view 2 lateral view 3 ventral view 4 anterolateral view

tinctly, moderately punctate, and 9 elongate deep punctures. Genal tip obtusely angular (in dorsal view); genal surface depressed with deep groove medially. Antennae (Fig. 3) yellowish brown with setate club. *Prothorax* (Figs 1–4). Prothorax dark brown, narrower than elytra, sides (in dorsal view) evenly rounded over anterior half. Anterolateral lobe rounded, edge slightly projecting downward (forming side of anterolateral propectoral ridge). Pronotal sides steeply declivous. Posterolateral section of pronotum rounded. Base of pronotum evenly rounded, immarginate; basolateral area with 1 fine ridge and numerous grooves around base. Pronotal surface glabrous. Costae densely punctate, intercostal sulci with distinctly wrinkled. Discal depression deep; surface, apart from some local micropunctation, smooth. Pronotal pattern of longitudinal costae as follows: median costa broad, becoming indistinct around apical 1/5; basomedian section triangular, surface deplanate, flattened except a longitudinal wrinkled furrow at middle. Central depression posterolaterally delimited by depressed area of paramedian costa. Paramedian costa broad, distinct, continuing to about 2/3 of pronotal length. Sublateral costa anteriorly broad, distinct, tapering posteriad to about 1/5 of pronotal length, reaching paramedian costa. Lateral costa anteriorly broad, distinct, extending from anterolateral lobe caudad, tapering to base of pronotum. Marginal costa posteriorly broad, ending at depressed basolateral area. Anterolateral part of propectus deeply excavate. Preprosternal apophysis distinct, with several setae. Propectus glabrous, brown. Posterolateral area of propectus with some ridges and grooves. Postprosternal surface with small, shallow, median impression. *Elytra* (Figs 1–2, 4). Semi-elliptical, strongly convex dorsally, as high as pronotum, dark brown, matt, with 7 interstrial costae and intervening striae, and with short adpressed trichomes at base of costae 2-5 forming wing-shaped patches. Humeral and apical elytral umbones absent; apicosutural edge nearly rectangular, strongly protruding above. Epipleuron wide. Elytral striae distinct, deeply impressed, with transverse weak costae from base to apex forming quadrate cells; striae 1 and 2 reaching basal half. Discal interstrial costae broadly trapezoidal (in cross-section), surface with dense, scattered punctures. Elytral pattern of interstrial costae as follows: costa 1 (next to suture) narrow, almost rectilinear; costa 2 tapering in front, stopping at basal half. Costa 3 complete, strongly developed, stoutly protruding in front to form median projection. Costae 4–8 complete, strongly developed. Costae 9 and 10 apparently fused together. *Mesosternum* (Fig. 3). Transverse mesometasternal groove between posterior edges of mesocoxae distinct, straight, not completely reaching mesocoxae. Mesosternum with fine peridiscal grooves arising from this transverse groove and two diagonal grooves, except in front; mesosternal surface dark brown, glabrous, flattened, moderately micropunctate. Metasternum (Fig. 3). Metasternum with very shallow median impression, glabrous, and with fine perimarginal groove all around; dark brown. Abdomen (Fig. 3). Venter with 5 visible fairly sclerotized sternites, all dark brown, matt, glabrous, without grooves, sparsely micropunctate. Pygidium dark brown, glabrous, base broadly margined; surface generally convex, densely micropunctate. Legs (Figs 1-4). Procoxa protuberant. Profemur brown, underside glabrous, densely micropunctate; outline broadly elliptical, emarginate distally. Protibia pale brown, broad, with sparse short setae, microsculpture poorly pronounced; shape strongly complanate with 2 external denticles, no basal serration; apex oblique-sinuate, transverse, with distinct apico-internal spine; internal side strongly dilated from slender base. Protarsus twice longer than width of tibial apex, slender, yellowish brown; segment 1 inserted in fine groove, as long as segments 2-4 combined. Mesocoxa dark brown, widely separated, slightly divergent anteriad. Mes-



Figures 5–10. Body parts of *Termitotrox venus* sp. n. 5 maxilla (without cardo) 6, 7 right mandible, in ventral and lateral view 8 epipharynx 9, 10 aedeagus in lateral and dorsal views.



Figures 11-13. Living Termitotrox venus sp. n. walking on a wall of the host termite nest inside.

ofemur dark brown, broadly elliptic in outline, distally emarginate, surface moderately micropunctate, glabrous. Mesotibia dark brown, with several setae, broad, dilated near base, nearly parallel-sided from apex, edges entire; tibial apex deeply emarginate, with pair of acuminate apico-internal spurs, external one long, slightly curved, internal one short, straight; upper side of mesotibia with fine longitudinal ridge near outer edge, weak costa at basal half, underside with fine sinuate ridge from base to apico-internal section; with long setae around apical quarter. Metatibia similar to mesotibia, but gently dilated apicad, with apex shallowly emarginate. Meso- and metatarsi dark brown, compacted-complanate, segments 1–4 short. Length of outer apical spur of metatibia 1/4 of metatibia, reaching base of tarsal segment 5. Aedeagus (Figs 9, 10).

Female. No significant sexual dimorphism is detected.

**Measurements.** Body length 2.26–2.70 (2.48±0.124); maximum width of head 0.84–0.93 (0.88±0.031); median dorsal length of pronotum 0.92–1.14 (1.01±0.064), maximum width 0.98–1.12 (1.04±0.053); sutural length of elytra 1.12–1.53 (1.35±0.115), maximum width 1.14–1.34 (1.24±0.067). N = 7.

Symbiotic host. *Macrotermes* cf. gilvus (see Discussion).

**Remarks.** Male aedeagus size ratio is the same rate as *Termitotrox cupido*, i.e. 44% of body length.

## Discussion

**Termite association.** Of the eight *Termitotrox venus* beetles recovered from fungus garden cells of *Macrotermes* cf. *gilvus* (Figs 11–13), seven were found on or inside the refuse dumps. The refuse dumps do not contain any fungal carpophores. The beetles appear camouflaged inside the refuse and move slowly, so they are difficult to collect. Only one specimen was found on the wall of fungus garden cell of *Hypotermes makhamensis* (Figs 11–13) despite more than 200 fungus garden cells of this termite were examined; this is probably accidental (it may be caused by the underground connection of the colonies of the two termite species ). Therefore, we think that the true host of *T. venus* is *M.* cf. *gilvus*.

All other known termitotrogines are associated with either Protermes, Odontotermes or Hypotermes (Krikken 2008; Maruyama 2012a). A phylogenetic analysis of fungusgrowing termites revealed that these three genera form a monophyletic group, with Odontotermes being paraphyletic with respect to Hypotermes; however, Macrotermes did not group with this clade, and is instead only distantly related (Aanen and Eggleton 2005). In contrast, *Termitotrox venus* and *T. cupido* have a clear synapomorphy in the wing-shaped trichomes on the elytra, so these species are apparently closely related to each other. Therefore, the host relationship between species of Termitotrox and genera of Macrotermitinae is unlikely to have arisen via co-cladogenesis. This type of relationship between termite hosts and termitotrogine scarabs is similar to that observed in Corythoderini. Corythoderines are also known to be associated with Odontotermes and Macrotermes (Tangelder and Krikken 1982; Bordat and Moretto 2010; Maruyama 2012b). This capacity to utilize phylogenetically unrelated hosts suggests that perhaps both Macrotermes and the group formed by Protermes+Odonto termes+Hypotermes produce similar nest odors, which are targeted by termitophilous scarabs in search of host colonies.

The pronotal basomedian section and the elytral median projection of *Termitotrox* venus form a structure (Figs 2, 4) similar to that seen in *Eocorythoderus incredibilis* Maruyama, 2012 (and, to a lesser extent in *Termitotrox cupido*), which was also found in a *Macrotermes* cf. gilvus nest in Siem Reap. Maruyama (2012b) revealed that this structure functions as a handle that allows the termite to grip the beetle and carry it. We did not observe *Termitotrox venus* being carried by worker termites during our survey, but this structure is probably used for the same behavior. In addition, the number of damaged specimens (broken legs, tibia or tarsi) of this species was lower than that of *E. incredibilis* (damaged/undamaged: *T. venus* 2/8, *E. incredibilis* 5/10, based on the type series from Maruyama (2012b)), hence, perhaps, *T. venus* could be mostly a synoekete (ignored by the hosts) except during certain periods, such as the movement of the host colony.

Using Wasmannian terminology (Wasmann 1894), Vårdal and Forshage (2010) suggested Termitotrogini may be synechthrans (treated with hostility by the hosts) because of the defensive morphology of the species known at that time. However, at least *T. venus* and *T. cupido* seem to be mainly synoeketes because both species appeared to be ignored by termites in the field. Based on both morphology and field observations, Corythoderini were proposed to be symphilic (Vårdal and Forshage 2010). Although symphilic behavior was recorded for *E. incredibilis* (Maruyama 2012b), the rate of specimen damage is nevertheless high, and this species has, in overall, a more defensive morphology compared to the other species of Corythoderini. Hence, the biology of *E. incredibilis* may vary from persecuted (synechthran) to integrated (symphile). On the other hand, *T. venus* may be a largely ignored (synoekete) but based on the morphology similar to that of *E. incredibilis* (trichomes, carrying "handle"), may at times exhibit symphilic behavior. The discoveries of these new combinations of life-styles in termitophilous beetles require a more flexible framework than that proposed by Wasmann (1894).

**Size difference.** *Termitotrox venus* is larger than *T. cupido*, and the beetle size seems to be correlated with the body size of the primary host of each of these species. Therefore, inquiline size may be affected by host size — a relationship paralleling that seen between termitophilous Staphylinidae and their hosts (Maruyama, personal observation).

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RESEARCH ARTICLE



# New records of Limoniidae and Pediciidae (Diptera) from Croatia

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#### Abstract

New records are provided for Limoniidae and Pediciidae from Croatia, with new distribution records for species in 12 different genera. Four genera and 18 species are newly recorded for Croatia. Until now, including this data, 87 Limoniidae and eight Pediciidae have been recorded from Croatia. In this paper we confirm presence of *Ormosia (Oreophila) bergrothi* (Strobl, 1895) and we give the first records of *Dicranota (Paradicranota) pavida* (Haliday, 1833) and *Molophilus (Molophilus) repentinus* Starý, 1971 from Balkan Peninsula.

#### **Keywords**

Lipsothrix, Paradelphomyia, Rhabdomastix and Tricyphona, Plitvice Lakes, Krka River, distribution

# Introduction

Pediciidae or hairy-eyed crane flies comprise slender tipuloid like flies. Species belonging to subfamily Pediciinae have carnivorous larvae and are connected with the wet environments (Kolcsár et al. 2012). The adults can be found near the larval habitats (brooks, springs, small rivers), and they are mostly found in riparian vegetation. Most of the species are mountainous. In some species males can produce massive swarms (e.g. *Pedicia* (*Amalopis*) occulta (Meigen, 1830), *Dicranota* (*Paradicranota*) pavida (Haliday, 1833)). Limoniidae or short-palped crane flies are a paraphyletic group (Petersen et al. 2010) of various sized tipuloid like flies, which larvae occupy different habitats (Ujvárosi 2005, Ujvárosi and Póti 2006, Kolcsár et al. 2013). A large part of genera bind to wet environments and some are truly aquatic (e.g. *Antocha*). From Western Palaearctic region 77 Pediciidae and 735 Limoniidae species are reported (Oosterbroek 2015).

The Croatian Pediciidae and Limoniidae fauna is less studied, despite the relatively large number of publications. The first study which contains records and description of *Dactylolabis dilatata* from Croatia was noted by Loew (1856). Since then Egger (1863), Strobl (1893, 1900, 1902), Langhoffer (1917), Lackschewitz (1928, 1940a, 1940b), Lackschewitz and Pagast (1941, 1942), Coe (1959), Nielsen (1959), Simova (1960), Mannheims (1967), Starý (1969, 1971a), Mendl (1984) and Savchenko et al. (1992) contributed additional distribution records from Croatia. Oosterbroek and Simova-Tosic (2004) gave a list of Pediciidae and Limoniidae species of Croatia. In the latest publication by Starý and Oosterbroek (2008) *Dicranomyia (Glochina) tristis* (Schummel, 1829) is reported for the first time in Croatia. At the present a total of four Pediciidae and 73 Limoniidae are reported from Croatia (Oosterbroek 2015). In this paper we offer new records of several genera of Pediciidae and Limoniidae collected from various sites in Croatia.

#### Material and methods

In the course of various ecological and taxonomic projects and surveys by Marija Ivković, many limonid and pedicid flies were collected by means of emergence traps set in springs, tufa barriers, streams and small rivers at nine sites in Plitvice Lakes National Park and at two sites at Krka National Park. Traps were emptied once a month, at the end of each month. Each trap had a surface area of 45 × 45 cm (and height 50 cm), was fixed in the sediment of the stream, and contained 2% formaldehyde; six traps were placed at each location on various types of microhabitats. At majority of sites traps were placed for one year (two sites at Krka River) or two years (majority of sites in Plitvice Lakes), but at three sites at Plitvice Lakes (spring of Bijela rijeka, tufa barrier Labudovac and tufa barrier Kozjak-Milanovac) they were placed for seven years, for additional details see Ivković et al. (2014). Each trap was recorded with the initial "P" and a number, e.g. "P4", is pyramid emergence trap no. 4. All flies were collected from March 2007 to October 2014.

Additional sampling using a sweep net took place between March 2011 and June 2014 at various sites in Croatia. All Limoniidae and Pediciidae specimens were placed in 80% ethanol. The literature used for identification is as follows: Alexander (1975), Blythe (2010), Dienske (1987), Podenas et al. (2006), Savchenko (1982), Starý (1971a, 1971b, 2004, 2009), Starý and Rozkošný (1970), Tjeder (1958, 1972),

**Table 1.** The list of sampling sites.

Site name	Longitude	Latitude	Altitude (m)
Spring Jankovac, Papuk Mountain	E 17°41'14"	N 45°31'06"	525
Dubočanka stream, Papuk Mountain	E 17°40'42"	N 45°29'11"	585
Brzaja, before N. Zvečeva, Papuk Mountain	E 17°30'53"	N 45°33'17"	502
Brzaja after N. Zvečeva, Papuk Mountain	E 17°31'53"	N 45°30'57"	368
Spring of Bijela rijeka, Plitvice Lakes	E 15°33'43"	N 44°50'05"	720
Upper reach of Bijela rijeka, Plitvice Lakes	E 15°33'33"	N 44°50'04"	715
Spring of Crna rijeka, Plitvice Lakes	E 15°36'28"	N 44°50'14"	680
Crna rijeka by the bridge, Plitvice Lakes	E 15°35'59"	N 44°50'22"	665
Tufa barrier Labudovac, Plitvice Lakes	E 15°35'59"	N 44°52'17"	630
Tufa barrier Kozjak-Milanovac, Plitvice Lakes	E 15°36'32"	N 44°53'39"	545
Tufa barrier Novakovića Brod, Plitvice Lakes	E 15°36'38"	N 44°54'07"	500
Stream Plitvica,Plitvice Lakes	E 15°36'27"	N 44°54'08"	555
Korana village, Plitvice Lakes	E 15°37'09"	N 44°55'33"	390
Spring of Krupa River	E 15°54'33"	N 44°11'49"	150
Spring Krčić	E 16°19'42"	N 44°01'48"	390
Spring of Krka River	E 16°14'07"	N 44°02'31"	265
Roški Slap, Krka River	E 15°58'22"	N 43°54'20"	55
Skradinski Buk, Krka River	E 15°57'55"	N 43°48'09"	45
Radmanove Mlinice, Cetina River	E 16°45'11"	N 43°26'16"	15

Ujvárosi and Bálint (2012) and Stary and Stubbs (2015). All the material listed here is deposited in the col. M. Ivković, University of Zagreb, Croatia (UZC) and Diptera Collection of the Faculty of Biology and Geology, Cluj-Napoca, Romania. GPS coordinates and altitudes for the localities where specimens were trapped and/or collected are given in Table 1.

## Results

## Faunistic records

The following format is used for the records given here: name of the site, followed by the sampling date (in the case of collections from the pyramid emergence traps, the trap number is also given, e.g. "P1" is pyramid emergence trap number 1), and the number of sampled specimens. New species for Croatia are listed with \* before the name of the species. All the sites are listed in Table 1. All mentioned distribution data are from Catalogue of the Craneflies of the World (Oosterbroek 2015).

## Pediciidae

## **Subfamily Pediciinae**

#### \*Dicranota (Dicranota) bimaculata (Schummel, 1829)

**New records. Spring of Bijela rijeka, Plitvice Lakes**, 5.x.2010, P4, 1 $\bigcirc$ ; same site, 29.v.2012, P3, 1 $\bigcirc$ 

**Comments.** Widespread in Europe except the Mediterranean region. In the Balkan Peninsula it has been recorded from Bulgaria.

#### \*Dicranota (Paradicranota) pavida (Haliday, 1833)

New records. Upper reach of Bijele rijeka, Plitvice Lakes, 5.x.2010, P4, 13, 19; spring of Crna rijeka, Plitvice Lakes, 29.v.2011, 13

**Comments.** Mostly Central and Western European distributed species. This is the first record for Balkan Peninsula.

#### \*Pedicia (Amalopis) occulta (Meigen, 1830)

New records. Spring of Bijela rijeka, Plitvice Lakes, 28.iv.2007, P2,  $1^{\circ}$ ; same site and date, P3, 13; same site and date, P6, 19; same site, 29.v.2007, P3, 13, 29; same site, 28.vi.2007, P2, 12; same site and date, P5, 13; same site, 25.vii.2007, P2, 13; same site, 30.viii.2007, P2, 13; same site and date, P3, 13; same site, 2.x.2007, P3, 13; same site, 29.x.2007, P2, 33; same site and date, P3, 23, 32; same site and date, P5, 1; same site, 29.xi.2007, P2, 1; same site and date, P3, 1; same site, 26.xii.2007, P3,  $2\delta$ ; same site, 2.iv.2008, P2,  $1\delta$ ,  $1\mathfrak{P}$ ; same site and date, P6,  $1\delta$ ; same site, 23.iv.2008, P2,  $1^{\circ}$ ,  $1^{\circ}$ ; same site and date, P3,  $1^{\circ}$ ; same site and date, P4,  $1^{\circ}$ ; same site and date, P6,  $1^{\circ}$ ; same site, 1.vi.2008, P2,  $1^{\circ}$ ; same site and date, P4,  $2^{\circ}$ ; same site, 30.vi.2008, P3, 1<sup>3</sup>; same site, 22.vii.2008, P4, 1<sup>2</sup>; same site, 27.viii.2008, P4,  $1^{\uparrow}$ ; same site and date, P6,  $1^{\circ}$ ; same site, 26.xi.2008, P2,  $1^{\uparrow}$ ; same site and date, P3, 19; same site, 3.iii.2009, P2, 13; same site, 1.iv.2009, P2, 23, 29; same site and date, P4, 1 $\bigcirc$ ; same site and date, P6, 1 $\bigcirc$ ; same site, 2.v.2009, P2, 2 $\bigcirc$ ; same site and date, P6,  $1^{\circ}$ ; same site, 30.x.2009, P2,  $1^{\circ}$ ,  $1^{\circ}$ ; same site, 8.iv.2010, P2,  $1^{\circ}$ ; same site and date, P3, 13; same site, 4.v.2010, P6, 19; same site, 4.vi.2010, P4, 13; same site, 25.vii.2010, P2,  $13^{\circ}$ ,  $19^{\circ}$ ; same site, 26.viii.2010, P4,  $23^{\circ}$ ; same site, 2.ix.2010, P3,  $19^{\circ}$ ; same site and date, P4,  $1^{\circ}$ ; same site and date, P6,  $1^{\circ}$ ; same site, 10.xii.2010, P3,  $1^{\circ}$ ,  $1^{\circ}$ ; same site, 25.ii.2011, P6,  $1^{\circ}$ ,  $1^{\circ}$ ; same site, 26.iv.2011, P4,  $2^{\circ}$ ; same site and date, P6,  $1\overline{\Diamond}$ ,  $1\overline{\heartsuit}$ ; same site, 29.v.2011, P4,  $1\overline{\Diamond}$ ,  $2\overline{\heartsuit}$ ; same site and date, P6,  $1\overline{\Diamond}$ ; same site, 27.vi.2011, P2, 13; same site, 2.viii.2011, P6, 13, 19; same site, 6.ix.2011, P3,

 $1^{\circ}$ ; same site and date, P4,  $2^{\circ}$ ; same site, 4.viii.2012, P3,  $1^{\circ}$ ; same site, 4.ix.2012, P3, 1 $\delta$ ; same site and date, P4, 2 $\delta$ , 1 $\mathfrak{P}$ ; same site, 6.xi.2012, P4, 1 $\delta$ ; same site and date, P5, 13, 19; same site and date, P6, 13; same site, 11.iii.2013, P2, 23; same site, 2.iv.2013, P3, 1 $\bigcirc$ ; same site and date, P4, 1 $\bigcirc$ ; same site, 3.v.2013, P4, 1 $\bigcirc$ ; same site, 7.viii.2013, P4, 1 $\bigcirc$ ; same site and date, P5, 1 $\stackrel{?}{\circ}$ ; same site, 29.x.2013, P3, 1 $\bigcirc$ ; upper **reach of Bijele rijeka, Plitvice Lakes**, 30.iii.2007, P2,  $1^{\circ}$ ; same site and date, P4,  $2^{\circ}$ ; same site, 28.iv.2007, P2, 1 $\bigcirc$ ; same site and date, P3, 2 $\bigcirc$ ; same site and date, P4, 2 $\bigcirc$ ; same site and date, P5,  $2^{\circ}$ ; same site, 29.v.2007, P2,  $2^{\circ}$ ; same site and date, P3,  $1^{\circ}$ ; same site and date, P4,  $1^{\circ}$ ; same site and date, P5,  $1^{\circ}$ ; same site and date, P6,  $1^{\circ}$ ; same site, 28.vi.2007, P2, 13, 29; same site and date, P3, 13; same site and date, P4, 13; same site, 25.vii.2007, P3,  $2^{\circ}$ ; same site and date, P4,  $6^{\circ}$ ,  $1^{\circ}$ ; same site, 30.viii.2007, P1, 1 $\bigcirc$ ; same site and date, P2, 3 $\bigcirc$ ; same site and date, P3, 1 $\bigcirc$ ; same site and date, P4,  $1^{\circ}_{\circ}$ ; same site, 2.x.2007, P1,  $2^{\circ}_{\circ}$ ,  $1^{\circ}_{\circ}$ ; same site and date, P2,  $2^{\circ}_{\circ}$ ; same site and date, P3,  $1^{\circ}$ ,  $1^{\circ}$ ; same site and date, P4,  $1^{\circ}$ ; same site and date, P6,  $1^{\circ}$ ; same site, 29.x.2007, P1, 29; same site and date, P5, 19; same site, 29.xi.2007, P1, 19; same site and date, P2,  $13^{\circ}$ ,  $39^{\circ}$ ; same site, 25.ii.2008, P1,  $13^{\circ}$ ; same site, 2.iv.2008, P2,  $13^{\circ}$ ; same site and date, P3,  $4^{\circ}_{\circ}$ ; same site and date, P4,  $2^{\circ}_{\circ}$ ,  $3^{\circ}_{\circ}$ ; same site, 23.iv.2008, P2, 13, 19; same site and date, P4, 33; same site and date, P5, 29; same site, 1.vi.2008, P5, 1 $\bigcirc$ ; same site, 30.vi.2008, P2, 2 $\bigcirc$ ; same site and date, P3, 1 $\bigcirc$ , 1 $\bigcirc$ ; same site and date, P4, 1<sup>3</sup>; same site, 22.vii.2008, P3, 1<sup>3</sup>; same site, 27.8.2008, P1, 1<sup>9</sup>; same site and date, P2,  $1^{\circ}$ ; same site and date, P3,  $1^{\circ}$ ; same site and date, P5,  $1^{\circ}$ ; same site, 29.xi.2008, P6, 1 $\bigcirc$ ; same site and date, 27.x.2008, P2, 2 $\bigcirc$ ; same site and date, P3, 1 $\bigcirc$ ; same site and date, P4, 13; same site and date, P6, 19; same site and date, 26.xi.2008, P2, 1 $\bigcirc$ ; same site and date, P4, 1 $\bigcirc$ ; same site, 3.iii.2009, P5, 1 $\bigcirc$ ; same site, 1.iv.2009, P3, 1 $\bigcirc$ ; same site and date, P5, 1 $\bigcirc$ ; same site, 2.v.2009, P4, 1 $\bigcirc$ , 1 $\bigcirc$ ; same site and date, P5,  $1^{\circ}$ ,  $1^{\circ}$ ; same site, 29.v.2009, P1,  $2^{\circ}$ ; same site and date, P2,  $1^{\circ}$ ; same site and date, P3, 13; same site, 26.vi.2009, P4, 19; same site and date, P5, 19; same site, 24.vii.2009, P4, 1 $\bigcirc$ ; same site and date, P5, 1 $\bigcirc$ ; same site, 27.viii.2009, P3, 1 $\bigcirc$ ; same site and date, P4, 1 $^{\circ}$ ; same site, 30.x.2009, P2, 3 $^{\circ}$ , 2 $^{\circ}$ ; same site, 8.iv.2010, P6, 1 $^{\circ}$ ; same site, 4.v.2010, P2,  $1^{\uparrow}$ ,  $2^{\bigcirc}$ ; same site and date, P4,  $1^{\bigcirc}$ ; same site, 1.vii.2010, P1, 1 $\Diamond$ ; same site and date, P4, 1 $\Diamond$ ; same site and date, P6, 1 $\Diamond$ ; same site, 25.vii.2010, P1,  $2\delta$ ; same site and date, P2,  $1\delta$ ; same site and date, P4,  $6\varphi$ ; same site and date, P6,  $1\delta$ ; same site, 26.viii.2010, P2, 23; same site and date, P4, 23, 32; same site and date, P6, 13; same site, 5.x.2010, P2, 23, 12; same site and date, P3, 13, 12; same site and date, P4, 13; same site, 2.xi.2010, P1, 42; same site and date, P3, 13; same site and date, P4,  $13^{\circ}$ ,  $32^{\circ}$ ; same site and date, P5,  $12^{\circ}$ ; same site, 10.xii.2010, P1,  $13^{\circ}$ ; spring of **Crna rijeka, Plitvice Lakes**, 29.v.2007, P3, 13; same site, 30.viii.2007, P5, 19; **Crna rijeka by the bridge**, 28.vi.2007, P3, 13;

**Comments.** Widespread in Europe, including the Balkan Peninsula. It is frequently collected in springs and headwater brooks (Ujvárosi and Bálint, 2012). In the Balkan Peninsula it is recorded from Albania, Bosnia and Herzegovina, Bulgaria, Greece, Macedonia, Montenegro, Serbia and Slovenia.

## \*Tricyphona (Tricyphona) immaculata (Meigen, 1804)

New records. Spring of Bijela rijeka, Plitvice Lakes, 28.x.2011, P6, 13; upper reach of Bijele rijeka, Plitvice Lakes, 29.v.2007, P4, 29; spring of Crna rijeka, Plitvice Lakes, 30.iv.2007, P1, 59; same site, 29.v.2007, P1, 59; tufa barrier Labudovac, Plitvice Lakes, 28.x.2011, P1, 13

**Comments.** Widespread species. In the Balkan Peninsula it is recorded from Bosnia and Herzegovina, Montenegro, Macedonia, Serbia and Slovenia.

#### Limoniidae

#### Subfamily Chioneinae

## \*Ellipteroides (Ellipteroides) lateralis (Macquart, 1835)

New record. Tufa barrier Kozjak-Milanovac, Plitvice Lakes, 1.vii.2012, P4, 13

**Comments.** The species is frequently collected in calcareous springs, fens and around the tufa sediment (Blythe 2010). It is widely distributed all around Europe.

#### Gonomyia (Gonomyia) tenella (Meigen, 1818)

New records. Spring of Bijela rijeka, Plitvice Lakes, 29.x.2007, P5, 1 $\bigcirc$ ; Korana Village, Plitvice Lakes, 29.vi.2007, P2, 1 $\bigcirc$ ; same site and date, P6, 1 $\bigcirc$ ; same site, 26.vii.2007, P1, 6 $\bigcirc$ ; same site and date, P2, 1 $\bigcirc$ ; same site and date, P3, 1 $\bigcirc$ ; same site and date, P6, 3 $\bigcirc$ , 12 $\bigcirc$ ; same site, 1.xi.2007, P1, 1 $\bigcirc$ ; same site and date, P6, 3 $\bigcirc$ 

**Comments.** A semi-aquatic species which can be found along flowing and standing waters (Kolcsár et al. 2013). In the Balkans it is recorded from Bulgaria, Croatia, Macedonia and Slovenia.

## \*Molophilus (Molophilus) brevihamatus Bangerter, 1947

## **New record. Spring of Krka River**, 7.vii.2011, 1

**Comments.** A characteristic species along mountainous waters in Carpathians, Alps and Balkan high mountains (Pirin, Rila). Probably the microclimate of the spring provides right condition for the species in lower altitude. In the Balkan Peninsula it is recorded from Bulgaria and Slovenia.

#### \*Molophilus (Molophilus) bifidus Goetghebuer, 1920

New record. Spring of Bijela rijeka, Plitvice Lakes, 4.9.2012, P1, 1

**Comments.** This species is associated with springs and head waters (Ujvárosi 2005). Previous records from the Balkan Peninsula are from Bosnia and Herzegovina, Bulgaria and Macedonia.

## Molophilus (Molophilus) obscurus (Meigen, 1818)

New records. Spring Krčić, 29.iv.2011, 2♂; Roški Slap, Krka River, 13.x.2011, 1♂

**Comments.** Common species, with wide distribution in Europe, including Balkan Peninsula.

#### \*Molophilus (Molophilus) repentinus Starý, 1971

## **New record. Korana Village, Plitvice Lakes**, 30.v.2007, P2, 1∂, 1♀

**Comments.** Mountains species with sporadic distribution in Alps and Carpathians and surrounding mountains area. Its presence in the low altitude (390 m) is explainable by the presence of mountainous microhabitats around fast flowing Korana River. First record for Balkan Peninsula.

#### \*Ormosia (Oreophila) bergrothi (Strobl, 1895)

**New record. Spring of Crna rijeka, Plitvice Lakes**, 25.vii.2007, P1,  $3^{\circ}$ ; same site, 30.viii.2007, P1,  $1^{\circ}$ 

**Comments.** This species has a sporadic distribution, with erroneous remark on type- locality. Oosterbroek and Simova-Tosic (2004) suggested that the type-locality is not in Croatia and removed the species from Croatian checklist. This is the first confirmation of this species present in Croatia and Balkan Peninsula.

## \*Rhabdomastix (Rhabdomastix) edwardsi Tjeder, 1967

**New records. Korana Village, Plitvice Lakes**, 29.vi.2007, P2, 13; same site, 29.ix.2008, P6, 19

**Comments.** Riverine species associated with sandy sediments mostly in mountainous areas (Boardman 2012). Reported from Albania, Austria, Bosnia and Herzegovina, Bulgaria, Czech Republic, France, Germany, Great Britain, Italy, Slovakia, Slovenia and Spain.

## \*Rhypholophus phryganopterus Kolenati, 1860

**New record. Spring of Crna rijeka, Plitvice Lakes**, 30.iv.2007, P1, 1

**Comments.** Montane species, distributed in Alps, Carpathians and Bulgarian mountains.

#### Subfamily Limnophilinae

- Eloeophila apicata (Loew, 1871)
- **New record. Upper reach of Bijele rijeka, Plitvice Lakes**, 25.vii.2007, P4, 1 **Comments.** Common species associated with sandy sediment (Ujvárosi 2005).

#### Eloeophila maculata (Meigen, 1804)

New records. Upper reach of Bijele rijeka, Plitvice Lakes, 2.x.2007, P5, 1 $\bigcirc$ ; Crna rijeka by the bridge, Plitvice Lakes, 31.viii.2007, P2, 1 $\bigcirc$ 

**Comments.** Common species with wide European distribution.

#### \**Eloeophila miliaria* (Egger, 1863)

New record. Brzaja, before N. Zvečeva, Papuk Mountain, 14.vi.2012, 1 $\bigcirc$ 

**Comments.** Less common species than *Eloeophila apicata* and *E. maculata*. Reported from Albania, Bulgaria and Serbia.

## Epiphragma (Epiphragma) ocellare (Linnaeus, 1760)

New records. Spring of Kupa River, 10.vi.2009, 1♀; spring of Crna rijeka, Plitvice Lakes, 29.v.2011, 1♂

**Comments.** Very common species with wide distribution.

### Hexatoma (Eriocera) chirothecata (Scopoli, 1763)

New records. Tufa barrier Novakovića Brod, Plitvice Lakes, 25.vii.2007, P4, 13; Korana Village, Plitvice Lakes, 26.vii.2007, P4, 12

**Comments.** Mostly distributed in southeastern Europe. Reported from majority of Balkan countries.

# \*Paradelphomyia (Oxyrhiza) senilis (Haliday, 1833)

**New records. Upper reach of Bijele rijeka, Plitvice Lakes**, 2.x.2007, P2, 1 $\Diamond$ ; same site and date, P4, 1 $\Diamond$ ; spring of Crna rijeka, Plitvice Lakes, 28.vi.2007, P1, 2 $\Diamond$ 

**Comments.** A wide distributed species in Palearctic region, frequently collected around different flowing waters (Ujvárosi 2005, Kolcsár et al. 2013). In the Balkan Peninsula it is recorded from Bulgaria, Greece and Macedonia.

#### Subfamily Limoniinae

## Antocha (Antocha) vitripennis (Meigen, 1830)

**New record. Tufa barrier Kozjak-Milanovac, Plitvice Lakes**, 27.vi.2013, P4, 13; **tufa barrier Novakovića Brod, Plitvice Lakes**, 1.vi.2008, P2, 13; **Korana Village, Plitvice Lakes**, 30.v.2007, P6, 13; same site and date, 29.vi.2007, P1, 13; same site and date, 30.vi.2008, P6, 13; **Radmanove mlinice, Cetina River**, 19.vii.2005, 13, 59.

Comments. A very common species.

#### Dicranomyia (Dicranomyia) chorea (Meigen, 1818)

New records. Spring of Bijela rijeka, Plitvice Lakes, 1.vii.2010, P6, 1♂; same site, 2.viii.2011, P6, 1♂; upper reach of Bijele rijeka, Plitvice Lakes, 27.viii.2009, P3, 2♀; 1.vii.2010, P3, 1♂; same site, 25.vii.2010, P3, 1♂; same site, 26.viii.2010, P1, 1♂, 1♀; same site and date, P3, 4♂, 2♀; same site and date, P4, 2♂; same site, 5.x.2010, P1, 1♀; same site and date, P2, 1♂; tufa barrier Kozjak-Milanovac, Plitvice Lakes, 29.x.2013, P1, 1♂; stream Plitvica, Plitvice Lakes, 30.viii.2007, P3, 1♂; Korana village, Plitvice Lakes, 29.vi.2007, P1, 1♂; Roški Slap, Krka River, 28.iv.2014, P4, 1♀; Comments. A very common species.

## Dicranomyia (Dicranomyia) didyma (Meigen, 1804)

**New records. Spring of Bijela rijeka, Plitvice Lakes**, 30.viii.2007, P5, 15 $\cancel{3}$ , 9 $\bigcirc$ ; same site and date, P6, 6 $\cancel{3}$ , 5 $\bigcirc$ ; same site, 2.x.2007, P2, 1 $\cancel{3}$ , 2 $\bigcirc$ ; same site and date, P5, 4 $\cancel{3}$ , 2 $\bigcirc$ ; same site, 29.x.2007, P5, 1 $\cancel{3}$ ; same site, 1.vi.2008, P5, 1 $\bigcirc$ ; same site, 22.vii.2008, P6, 4 $\cancel{3}$ ; same site, 27.viii.2008, P6, 1 $\bigcirc$ ; same site, 29.ix.2008, P5, 1 $\bigcirc$ ; same site, 27.viii.2008, P6, 1 $\bigcirc$ ; same site, 29.ix.2008, P5, 1 $\bigcirc$ ; same site, 30.x.2009, P5, 1 $\cancel{3}$ , 2 $\bigcirc$ ; same site, 8.xii.2009, P6, 1 $\cancel{3}$ , 1 $\bigcirc$ ; same site, 1.vii.2010, P6, 1 $\bigcirc$ ; same site, 26.viii.2010, P5, 4 $\cancel{3}$ , 8 $\heartsuit$ ; same site and date, P6, 1 $\cancel{3}$ ; same site, 5.x.2010, P2, 1 $\cancel{3}$ , 1 $\bigcirc$ ; same site and date, P5, 4 $\cancel{3}$ , 3 $\bigcirc$ ; same site and date, P6, 2 $\cancel{3}$ , 2 $\bigcirc$ ; same site, 29.v.2011, P6, 7 $\cancel{3}$ , 6 $\bigcirc$ ; same site, 2.viii.2011, P6, 2 $\cancel{3}$ , 1 $\bigcirc$ ; same site, 6.ix.2011, P6, 12 $\cancel{3}$ , 10 $\bigcirc$ ; same

site, 28.ix.2011, P6,  $2^{\bigcirc}$ ; same site, 3.ix.2013, P6,  $7^{\bigcirc}$ ,  $5^{\bigcirc}$ ; upper reach of Bijele rijeka, Plitvice Lakes, 24.vii.2009, P1, 13; same site and date, P3, 13; same site, 27.viii.2009, P1, 5 $^{\circ}$ , 1 $^{\circ}$ ; same site and date, P2, 1 $^{\circ}$ ; same site, 1.x.2009, P1, 8 $^{\circ}$ , 2 $^{\circ}$ ; same site and date, P2, 2 $\bigcirc$ ; same site and date, P3, 3 $\bigcirc$ , 4 $\bigcirc$ ; same site, 30.x.2009, P2, 1 $\bigcirc$ ; same site and date, P3,  $1^{\circ}$ ; same site, 1.vii.2010, P2,  $4^{\circ}$ ,  $4^{\circ}$ ; same site and date, P3,  $1^{\circ}$ ; same site, 25.vii.2010, P2, 43, 59; same site, 26.viii.2010, P1, 33, 39; same site and date, P3, 13, 32; same site and date, P4, 23; same site, 5.x.2010, P1, 13, 22; same site and date, P2,  $2\beta$ ; same site and date, P3,  $14\beta$ ; same site and date, P4,  $1\beta$ ,  $3\varphi$ ; same site, 2.xi.2010, P3, 32; same site and date, P4, 13, 82; same site, 10.xii.2010, P4, 12; tufa barrier Kozjak-Milanovac, Plitvice Lakes, 1.vi.2008, P4, 13; same site, 27.viii.2008, P5, 1 $\bigcirc$ ; same site, 1.vii.2010, P4, 1 $\bigcirc$ , 4 $\bigcirc$ ; stream Plitvica, Plitvice Lakes, 25.v.2007, 1 $\delta$ ; same site, 27.vii.2007, P2, 1 $\delta$ ; **Roški Slap, Krka River**, 28.iv.2011, 3 $\delta$ , 2 $\mathfrak{Q}$ ; same site, 25.v.2011,  $5^{\circ}$ ,  $3^{\circ}$ ; same site, 30.viii.2011,  $1^{\circ}$ ; same site, 13.x.2011,  $1^{\circ}$ ; same site, 6.xi.2013, P4, 1∂, 1♀; same site, 7.ii.2014, P4, 5∂, 1♀; same site, 5.iii.2014, P4, 1 $\bigcirc$ ; same site, 2.iv.2014, P3, 1 $\bigcirc$ ; same site and date, P4, 1 $\bigcirc$ , 4 $\bigcirc$ ; same site, 28.iv.2014, P4, 23, 39; same site, 2.vi.2014, P3, 13, 29; same site and date, P4, 33, 29; same site, 26.vi.2014, P3, 23, 19; same site and date, P4, 53, 19; same site, 26.vii.2014, P4, 13, 1 $\Im$ ; same site and date, P6, 2 $\Im$ ; **Skradinski Buk, Krka River**, 2.vi.2014, P3, 3 $\Im$ , 2 $\Im$ ; same site, 26.vi.2014, P4, 1♀

Comments. A very common and abundant species.

#### Dicranomyia (Dicranomyia) goritiensis (Mik, 1864)

**New records. Roški Slap, Krka River**, 28.iv.2011, 23; same site, 25.v.2011, 23; same site, 2.vi.2014, P4, 19; same site, 26.vii.2014, P2, 13; same site and date, P4, 13, 29; same site, 2.ix.2014, P4, 19; same site, 2.x.2014, P1, 29; same site, 27.x.2014, P2, 29; same site and date, P4, 23; **spring of Krka River**, 25.v.2011, 13; **spring of Krupa River**, 26.v.2011, 13

**Comments.** Sporadic distributed species, which is associated with moss and/or algae covered stones, mostly around waterfalls and rocky coast. In the Balkan Peninsula it is recorded from Croatia and Greece.

#### \*Dicranomyia (Dicranomyia) imbecilla Lackschewitz, 1941

**New records. Spring of Bijela rijeka, Plitvice Lakes**, 29.v.2011, P2, 1 $\bigcirc$ ; same site, 27.vi.2013, P1, 1 $\bigcirc$ ; **tufa barrier Kozjak-Milanovac, Plitvice Lakes**, 27.vi.2013, P5, 1 $\bigcirc$ ; **spring Krčić**, 29.iv.2011, 1 $\bigcirc$ ; same site, 26.v.2011, 3 $\bigcirc$ , 4 $\bigcirc$ 

**Comments.** The species is known from calcareous seepages depositing tufaceous substrate. The species known from Bulgaria, Czech Republic, Great Britain, Germany, Slovakia and Switzerland (Stary and Stubbs 2015).

#### \*Dicranomyia (Dicranomyia) lucida de Meijere, 1918

## New record. Roški Slap, Krka River, 2.x.2014, P1, 1♀

**Comments.** Relatively common species in woodlands around rivers. In the Balkan Peninsula it is reported from Bulgaria, Macedonia and Greece.

# Dicranomyia (Dicranomyia) mitis (Meigen, 1830) complex

**New records. Brzaja after N. Zvečevo, Papuk Mountain**, 13.vi.2012, 1 $\eth$ ; upper reach of Bijele rijeka, Plitvice Lakes, 29.v.2009, P1, 1 $\circlearrowright$ ; ; spring of Krka River, 7.vii.2011, 1 $\clubsuit$ 

**Comments.** A very common and widespread species.

#### Dicranomyia (Dicranomyia) modesta (Meigen, 1818)

## New record. Roški Slap, Krka River, 25.v.2011, 2

**Comments.** A common species which inhabits various habitats, but mostly found in forests.

#### \*Elliptera omissa Schiner, 1863

#### New record. Dubočanka stream, Papuk Mountain, 18.ix.2012, 1

**Comments.** Mountainous species. In the Balkan Peninsula it is reported from Bosnia and Herzegovina, Bulgaria, Montenegro, Serbia and Slovenia.

#### Limonia hercegovinae (Strobl, 1898)

New records. Upper reach of Bijele rijeka, Plitvice Lakes, 28.iv.2007, P4, 13; spring of Crna rijeka, Plitvice Lakes, 30.iv.2007, P1, 13, 19; spring Jankovac, Papuk Mountain, 13.vi.2012, 19

**Comments.** A widely distributed species, but mostly in Balkan Peninsula.

#### Limonia macrostigma (Schummel, 1829)

New record. Brzaja, after N. Zvečeva, Papuk Mountain, 13.vi.2012, 1∂ Comments. Widely distributed and common species.

## Limonia phragmitidis (Schrank, 1781)

New record. Brzaja, after N. Zvečeva, Papuk Mountain, 14.vi.2012, 2♀ Comments. Widely distributed and common species.

#### \*Lipsothrix nobilis Loew, 1873

New record. Crna rijeka by the bridge, Plitvice Lakes, 28.vi.2007, P3, 13; tufa barrier Labudovac, Plitvice Lakes, 31.v.2007, P2, 23

Comments. Less common species associated with woodland streams and springs.

#### \*Lipsothrix remota (Walker, 1848)

**New records. Roški Slap, Krka River**, 28.iv.2011, 1♂; same site, 25.v.2011, 1♂, 1♀ **Comments.** Less common species associated with woodland streams and springs.

#### Orimarga (Orimarga) virgo (Zetterstedt, 1851)

#### **New record. Spring of Krka River**, 7.vii.2011, 2

**Comments.** Species reported from the different types of calcareous rocky habitats, which are covered with moss and/or algae. In the Balkan Peninsula it is reported from Croatia, Greece and Slovenia.

#### Discussion

In this paper we report for the first time four genera (*Lipsothrix* Loew, *Paradelphomyia* Alexander, *Rhabdomastix* Skuse and *Tricyphona* Zetterstedt) and 18 species new for Croatia. Including our results, eight Pediciidae and 87 Limoniidae have now been recorded from Croatia.

Oosterbroek and Simova-Tosic (2004) summarized the literature data of Former Yugoslav states, in which they reported 72 limonid and four pedicid species from Croatia and additional limonid species is reported by Starý and Oosterbroek (2008). This number is only the fragment of the real species number, which is supported by our results. Tipuloidea fauna is better studied in some neighboring countries, Slovenia has 11 species of Pediciidae (P) and 106 of Limoniidae (L), while Hungary has 12 species of Pediciidae and 132 of Limoniidae, however a similar number of species was reported from Bosnia and Herzegovina (6 P and 69 L), Serbia (8 P and 68 L) and Montenegro (9 P and 59 L). It is evident from these numbers that much remains to be discovered about the Tipuloidea fauna of Croatia and entire Balkan Peninsula.

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RESEARCH ARTICLE



# Mosquito species (Diptera, Culicidae) in three ecosystems from the Colombian Andes: identification through DNA barcoding and adult morphology

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# Abstract

Colombia, one of the world's megadiverse countries, has a highly diverse mosquito fauna and a high prevalence of mosquito-borne diseases. In order to provide relevant information about the diversity and taxonomy of mosquito species in Colombia and to test the usefulness of DNA barcodes, mosquito species collected at different elevations in the departments of Antioquia and Caldas were identified combining adult morphology and barcode sequences. A total of 22 mosquito species from eight genera were identified using these combined techniques. We generated 77 barcode sequences with 16 species submitted as new country records for public databases. We examined the usefulness of DNA barcodes to discriminate mosquito species from the Neotropics by compiling 1,292 sequences from a total of 133 species and using the tree-based methods of neighbor-joining and maximum likelihood. Both methodologies provided similar results by resolving 105 species of mosquitoes separated into distinct clusters. This study shows the importance of combining classic morphological methodologies with molecular tools to accurately identify mosquitoes from Colombia.

### **Keywords**

Culicomorpha, nematocerous Diptera, Neotropical Region, species identification, combining methodologies

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# Introduction

Culicidae currently comprises 3,543 formally recognized species distributed throughout most types of habitats and ecosystems of the world (Harbach 2015). Mosquitoes, as a group, occur in a wide array of both aquatic and terrestrial habitats and have correspondingly variable morphological and behavioral adaptations to these (Becker et al. 2010). Due to their hematophagous behavior, mosquitoes are able to transmit many different disease agents such as viruses, bacteria, protozoans, and nematodes from one vertebrate host to another (Becker et al. 2010). The taxonomy of Culicidae has received special attention because of their vector capabilities and medical and veterinary importance (Harbach 2007, Becker et al. 2010). Thus, major mosquito vector species are generally the best studied and understood, both biologically and taxonomically (Harbach 2007).

Mosquito identification is traditionally based on dichotomous keys constructed from morphological features taken for a particular life stage or gender (Munstermann and Conn 1997). The morphological identification of mosquito species is hampered by intraspecific variation, the complexity of some features and the need for specimens in generally excellent condition (Zavortink 1974, Cywinska et al. 2006). The importance of gaining high resolution in mosquito species discrimination requires the implementation of additional tools that can be used for accurate identification (Munstermann and Conn 1997, Cook et al. 2005).

The DNA barcode method was postulated by Hebert et al. (2003a, 2003b) as a DNA sequence-based approach for the accurate identification of specimens and for species discovery. There are three different methods proposed to analyze the efficiency of DNA barcode data to discriminate species: similarity methods based on the match between the query sequence and the reference sequences, tree-based identifications, and the barcode gap (Hebert et al. 2003a, 2004, Ratnasingham and Herbert 2007, Austerlitz et al. 2009). Most published approaches to DNA barcoding use distance-based methods for species designation; however, alternative approaches using character-based phylogenetic analysis have been proposed (DeSalle et al. 2005). Although DNA barcode is now widely use, there are still many open questions about both the advantages and disadvantages of its use, as well as its definition as a methodology (Moritz and Cicero 2004, Brower 2006, Baker et al. 2009, Casiraghi et al. 2010, Jinbo et al. 2011, Wilson et al. 2011, Begsten et al. 2012, Collins and Cruickshank 2013, Stoeckle and Thaler 2014). Despite the known limitations of the method, DNA barcodes have been used to identify specimens and to flag potential new species (e.g. Scheffer et al. 2006, Nagoshi et al. 2011, Tavares et al. 2011, Baldwin and Weigt 2012, Huemer and Mutaten 2015, among others).

In mosquitoes, the effectiveness of the COI barcode marker for specimen identification has been tested in surveys of Canada (Cywinska et al. 2006), India (Kumar et al. 2007), the Iranian islands in the Persian Gulf (Azari-Hamidian et al. 2010), China (Wang et al. 2012), Amazonian Ecuador (Linton et al. 2013), Pakistan (Ashfaq et al. 2014), Singapore (Chan et al. 2014), and Belgium (Versteirt et al. 2015). These studies show correspondence between morphological species and DNA barcode clusters, but also point out the failure of the methodology to distinguish between very similar or cryptic species.

Colombia is located in the northwest of South America. It comprises a variety of biogeographic regions that have contrasting biophysical characteristics and high environmental variability (Etter et al. 2006). Related to this variety of ecosystems, Colombia has high levels of endemism and species richness and has been categorized as a mega-diverse country (Hernandez et al. 1992, Chaves and Arango 1998). Moreover, environmental variability may favor the development and persistence of a great diversity of mosquito species (Foley et al. 2007, Brochero and Quiñones 2008), as well as to favor both the immigration and biological invasion of non-endemic species including vector species (Barreto et al. 1996, Molina et al. 2000, Olano et al. 2001). Furthermore, Colombia is located in a region shown to be a potential hotspot for malaria endemicity and other mosquito borne disease outbreaks (Foley et al. 2008). In Colombia, malaria vector mosquitoes (genus Anopheles Meigen, 1818) have been extensively investigated, the morphological keys for their identification are up-to-date and available, and different genetic techniques have been developed to differentiate cryptic species complexes. Nevertheless, the overall diversity of mosquito fauna in Colombia is understudied and generally poorly known (Olano et al. 2001, Montoya-Lerma et al. 2011).

In order to improve the mosquito knowledge in Colombia, we tested the effectiveness of the barcoding methodology to support the reliable identification of Neotropical species of mosquitoes, previously identified with morphological characters, collected over three different altitudinal ecosystems of the Colombian Andes (Antioquia and Caldas departments).

### Materials and methods

#### Study area

The study area is located in the west central Andean region in Colombia. Fieldwork was conducted during September 2013 in rural areas of Antioquia and Caldas departments (Fig. 1). This study focused on three important biomes: paramo (Belmira, 3,200 masl), cloud forest (Rio Sucio, 1,960 masl), and tropical dry forest (La Pintada, 660 masl). Each sampling location was split into two habitat types: forest (habitat A) and anthropogenic disturbed area (habitat B). Forest sampling in the paramo plots consisted of dense shrubbery and scattered trees associated with sub-paramo zones. Additional collections were made in the rural area of Supia (Caldas) (mountain forest) at an elevation of 1,150 masl.

### Specimen collection

Mosquito adults were collected using one malaise trap and three Centers for Disease Control-CDC light traps in each habitat type (A or B) of the three biomes, totaling six



**Figure 1.** Map of Colombia indicating the sampling sites of mosquitoes collected in this study: Belmira, Antioquia, paramo, 3,200 masl (red circle); Rio Sucio, Caldas, cloud forest, 1,960 masl (green circle); La Pintada, Antioquia, tropical dry forest, 660 masl (blue circle); Supia, Caldas, rural area, 1,150 masl (pink circle). Modified from Instituo Geográfico Agustín Codazzi (www.igac.gov.co) and Wikimedia Commons (by Addicted04).

sites. CDC traps collected for 14 hours, between 5:00 pm and 7:00 am of the next day during two nights in each of our six locations plus the rural area of Supia. Malaise traps were placed and left for 48 hours in each location. Additional specimens were obtained by aspirating mosquitoes attracted to humans during the placement and operation of the traps. All data in the sampled localities were plotted following the protocols of Foley et al. (2010) using a GPS Garmin<sup>®</sup> Rino 530HCx. Weather data of average wind speed (km/h), temperature (°C), and relative humidity (%) were recorded with a Kestrel<sup>®</sup> 4000 Weather Tracker.

Wild-caught adults were killed using ethanol (90%) fumes in a lethal chamber to ensure DNA preservation. All mosquitoes were kept dry and individually transferred to labeled 1.5 ml tubes. Each tube was labeled, pierced with a mounted needle (to allow the escape of moisture) and placed in plastic bags containing silica gel. Specimen mounting techniques were conducted using the protocols of Walter Reed Biosystematics Unit-WRBU (Gaffigan and Pecor 1997).

### Adult identification using morphology

All the specimens collected were identified by female morphology and male genitalia. Since there is no single morphological key to facilitate the identification of mosquitoes of Colombia, genus level identifications were made with multiple approaches using the dichotomous keys of Lane (1953), Becker et al. (2010), Chaverri (2009), and the multi-entry web-based keys developed by the WRBU for South America (http:// wrbu.si.edu/southcom\_MQkeys.html). In addition, available dichotomous keys and original species descriptions were compiled through literature freely available on the WRBU website (www.wrbu.org). Important references used for species identifications by taxonomic groups included: Tribe Aedini (Arnell 1976, Reinert 2000), Anopheles (Komp 1942, Cova-Garcia 1961, González and Carrejo 2007), Culex Linnaeus, 1758 (Rozeboom and Komp 1950, Cova-Garcia et al. 1966, Bram 1967, Knight and Haeger 1971, Sirivanakarn 1982, Strickmann and Prat 1989, Pecor et al. 1992), Coquillettidia Dyar, 1905 (Shannon 1934, Cova-Garcia et al. 1966), Haemagogus Williston, 1896 (Kumm et al. 1946, Levi-Castillo 1951, Arnell 1973, Liria and Navarro 2009), Psorophora Robineau-Desvoidy, 1827 (Guedes and Souza 1964, Liria and Navarro 2003), Trichoprosopon Lane & Cerqueira, 1942 (Stone 1944, Zavortink 1979), Wyeomyia Theobald, 1901 (Judd 1998, Motta and de Oliveira 2000). Overall, Lane (1953) was used as a general reference.

#### Voucher specimens

Voucher specimens and associated genitalia preparations are stored in the entomological collections of the Zoologisches Forschungsmuseum Alexander Koenig (ZFMK), Bonn (Germany). Genomic DNA extracts are stored at -80 °C in the biobank collections of the ZFMK for future reference. Details of reference material are listed by genus in Suppl. material 1, as well as full collection site details including georeferences and environmental conditions. Fully digitized specimen data records are freely available in MosquitoMap (www.mosquitomap.org). COI barcode sequences can be accessed through GenBank (accession numbers KM592986 to KM593062; see Suppl. material 1).

### Laboratory protocols, DNA extraction, PCR, and sequencing

DNA was extracted from legs and occasionally abdomens (for specimens without legs or with deficient amplification) following standard protocols of the commercially available DNeasy Blood & Tissue Kit (QIAgen®). The COI barcode region was amplified using the forward primer LCO1490 (5'-GGTCAACAAATCATAAAGA-TATTGG-3') and the reverse primer C1N2191 (alias Nancy) (5'CCCGGTAAAAT-TAAAATATAAACTTC-3') (Simon et al. 1994). When those primers failed to amplify full-length sequences, the set LCO1490 and HCO2198 (5'-TAAACTTCA GGGT-GACCAAAAAATCA-3') (Folmer et al. 1994) were alternatively used. PCR amplification followed the protocol optimized by the Laboratories of Molecular Biology, Alexander Koenig Museum-ZFMK (Bonn, Germany). Each PCR reaction contained 2.5 µL of DNA, 2.3 µL of ddH20, 2 µL of Q-Solution, 10 µL of Qiagen Multiplex-Mix, and 1.6  $\mu$ M of each primer. The polymerase chain reaction (PCR) cycle conducted was: 95 °C for 15 min, 25 cycles of 94 °C for 35 s, 55 °C for 90 s and 72 °C for 90 s, 72 °C for 10 min and a 10 °C hold. The PCR product was visualized on 1.5% agarose gels, containing 0.5 mg/ml of GelRed®, followed by 1.75 microliters of the PCR product being removed and mixed with 1.75 µL of loading dye. Gels were run at 120 V for 30 min, prior to ultraviolet visualization. PCR products were cleaned using the commercially available QIAquick PCR Purification Kit (QIAgen®). Bi-directionally sequencing reactions were carried out by Macrogen© Inc. Chromatograms were edited in Geneious 7.0.6 (Biomatters<sup>®</sup> Ltd). Primer sequences were removed from edited contigs and only high-quality sequences of at least 600 bp were retained for data analysis. Sequences were aligned using Geneious 7.0.6.

### Additional sequences

To test variation in the barcoding region across a greater geographical area, barcoding sequences of mosquito species listed for the Neotropics were downloaded from BOLD and GenBank between December 2013 and February 2014 for all identified species with a minimum length of 480 bp of COI barcoding region, no stop codons and alignment without gaps (Suppl. material 2). In order to have a broader representation of mosquito genera, additional sequences of species from other biogeographic areas were downloaded for those Neotropical groups without available sequences. These taxonomic groups include: *Coquillettidia* (*Coquillettidia*) Dyar, 1905, *Culex* (*Neoculex*) Dyar, 1905, *Mansonia* (*Mansonioides*) Theobald, 1907, *Ochlerotatus* (*Protoculex*) Felt, 1904, *Orthopodomyia* Theobald, 1904, *Toxorhynchites* (*Lynchiella*) Lahille, 1904, and *Toxorhynchites* (*Toxorhynchites*) Theobald, 1901. Moreover, an unpublished data set of 45 sequences of mosquitoes collected in rural areas of Uraba (Antioquia, Colombia) during 2009 was also added to the analysis (Suppl. material 3).

Lutzomyia longipalpis (Lutz & Neiva, 1912) (Diptera: Psychodidae) was constrained as outgroup. We also included several other genera from three different families as outgroups, i.e. Chironomidae [Cricotopus bicinctus (Meigen, 1818), Chironomus decorus Goetghebuer, 1927, Chironomus kiiensis Tokunaga, 1936, Dicrotendipes tritomus (Thienemann & Kieffer, 1916), and Tanytarsus guerlus (Roback, 1957)], Simuliidae [Simulium ochraceum Walker, 1861, Simulium inaequale (Paterson & Shannon, 1927), Gigantodax abalosi Wygodzinsky, 1958, and Gigantodax basinflatus Wygodzinsky & Coscaron, 1989] and Dixidae [Dixella sp.]. A total of 11 outgroup taxa were included. All outgroup taxa sequences were downloaded from BOLD (Suppl. material 4).

### **Barcoding methodologies**

We used *similarity methods* based on the match between the query sequence and the reference database [e.g. BLAST (http://blast.ncbi.nlm.nih.gov/Blast.cgi) and BOLD Identification System (IDS) (http://www.boldsystems.org/index.php/IDS\_Ope-nIdEngine)] and clustering in *Tree-based identifications* (using Neighbour-joining and maximum likelihood approaches) in order to analyze the DNA barcode region of the mosquitoes collected in Colombia and assign individuals to a given species.

The Neighbour-Joining (NJ) tree (Saitou and Nei 1987) was based on the Tamura-Nei distance model (TN93) (Tamura and Nei 1993) as recommend by Srivathsan and Meier (2012). Bootstrap support values were calculated with 1,000 replicates. The NJ tree, distance matrices and bootstrap were generated using Geneious 7.0.6.

To perform the maximum likelihood analysis, the data set was divided into three partitions according to the codon positions of COI (first, second, and third positions). We determined the best choice of model for each partition using PartionFinder v1.1.0 (Lanfear 2012) under the Akaike Information Criterion (AIC) as recommended by Posada and Buckley (2004). The model chosen for position 1 was SYM+I+G, GTR+I+G for position 2 and GTR+I+G for position 3 of COI gene. Data were analyzed using Garli 2.0 (Zwickl 2006) based on best choice of the model predicted. Default settings (scorethresh-forterm = 0.05; significanttopochange = 0.0001; searchreps = 1) were used to perform 20 replicates. The tree with the highest likelihood was retained. Bootstrap support values were estimated from 1,000 replicates using the same independent models. Analytical runs of ML were performed at the Hydra cluster (Center for Astrophysics, Harvard University), a Linux based cluster running Grid Engine with more than 3,000 CPUs with AMD 64-bit processors.

# Results

A total of 77 mosquito specimens were collected from four sampling sites during our study (Table 1). Sampling success among traps varies greatly across location and habitat types. Belmira, the location with highest wind speed and precipitation, only reported one or no specimens for their two habitats. The specimens were identified to 22 species from eight genera. One species collected during this study in La Pintada forest habitat, *Wyeomyia luteoventralis* Theobald, 1901, is the first record for Colombia (Rozo-Lopez and Mengual, 2015).

### Adult identification using morphology

Twenty one species belonging to seven genera were successfully identified by morphological characteristics of adult females and male genitalia: Coquillettidia nigricans Coquillett, 1904 [n=1 female], Culex conspirator Dyar & Knab, 1906 [n=5 males], Cx. corniger Theobald, 1903 [n=8 females], Cx. declarator Dyar & Knab, 1906 [n=8; 5 females, 3 males], Cx. educator Dyar & Knab, 1906 [n=1 male], Cx. erraticus [n=2 males], Cx. erythrothorax Dyar, 1907 [n=1 female], Cx. lactator Dyar & Knab, 1906 [n=2 females], Cx. lucifugus Komp, 1936 [n=3 males], Cx. nigripalpus Theobald, 1901 [n=2; 1 female, 1 male], Cx. spinosus Lutz, 1905 [n=1 male], Cx. spissipes Theobald, 1903 [n=1 female], Cx. theobaldi Lutz, 1094 [n=1 male], Haemagogus janthinomys Dyar, 1921 [n=2 females], Hg. lucifer Howard, Dyar & Knab, 1913 [n=1 female], Ochlerotatus angustivittatus Dyar & Knab, 1907 [n=2; 1 female, 1 male], Oc. euiris Dyar, 1922 [n=1 female], Psorophora cingulata Fabricius, 1805 [n=1 female], Ps. ferox von Humboldt, 1819 [n=3; 2 females, 1 male], Trichoprosopon evansae Antunes, 1942 [n=1 female], and Wyeomyia luteoventralis [n=1 female]. One specimen of the genus Anopheles was identified by morphology to the group: neomaculipalpus / punctimacula. It was only possible to identify the species using its gene sequence.

### Mosquito barcoding

All the collected specimens were successfully amplified and sequenced, generating a total of 77 sequences with lengths ranging from 618 to 699 bp. Only high quality sequences were retained. Sequences from 14 species are submitted as new records for public databases: *Cq. nigricans, Cx. conspirator, Cx. educator, Cx. lactator, Cx. lucifugus, Cx. spissipes, Cx. theobaldi, Hg. janthinomys, Hg. lucifer, Oc. angustivittatus, Oc. euiris, Ps. cingulata, Tr. evansae, Wy. luteoventralis.* 

All the DNA sequences obtained were compared to those available, by 25 January 2015, in GenBank and BOLD Identification System. At the level of genus, BLAST accurately discriminated 92% of the genera previously identified, whereas BOLD accurately discriminated 70% of genera previously identified. Although BLAST repre-

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(habitat A) still

Macanita chariae				Sites			
most appende	Supia	Rio Sucio	1,960 masl	La Pintadá	a 660 masl	Belmira 3,	200 masl
Species	1,150 masl	Habitat A	Habitat B	Habitat A	Habitat B	Habitat A	Habitat B
Anopheles neomaculipalpus	1						
Coquillettidia nigricans					1		
Culex coniger		10					
Culex conspirator				5			
Culex spp. [coronator complex]		1	1	3			
Culex declarator	1			10			
Culex educator				1			
Culex erraticus				6	2		
Culex erythrothorax		1					
Culex lactator				3			
Culex lucifugus				6			
Culex nigripalpus	1			2			
Culex spinosus		1					
Culex spissipes					1		
Culex theobaldi				1			
Culex (Culex) sp.		1					
Culex (Melanoconion) sp.				4	1		
Haemagogus janthinomys					2		
Haemagogus lucifer					1		
Ochlerotatus angustivittatus	1			1			
Ochlerotatus euiris						1	
Psorophora cingulata					1		
Psorophora ferox				2	1		
Trichoprosopon evansae		1					
Wyeomyia luteoventralis				1			
Undetermined		1					
Total	4	16	1	45	10	1	0

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sented a higher percentage of accurate discrimination of genera in our queries, five specimens with a score of more than 89% were wrongly matched: *Trichoprosopon* was mismatched as *Anopheles*, *Wyeomyia* was mismatched as *Sabethes* Robineau-Desvoidy, 1827, *Haemagogus* was mismatched as *Ochlerotatus* (or *Aedes*), and for the sequence of *Haemagogus lucifer* mixed results of *Spilogona* Schnabl, 1911, *Haematobia* Lepeletier & Serville, 1828 (family Muscidae), and *Culex* were obtained.

By adding GenBank, BOLD, and unpublished sequences to our data set, it was possible to obtain taxon coverage of 68% of the mosquito genera and 34% of the mosquito species listed for Colombia. Moreover, our data set coverage for Neotropical mosquito species corresponds to 58% of mosquito genera and 12% of mosquito species. The data set of the COI sequences of the Neotropical mosquitoes comprises a total of 1,292 barcode sequences belonging to 133 species and 21 genera (with a minimum length of 640 bp). The alignment was unambiguous: no gaps and amino acid translations without stop codons, indicating that all sequences represented functional protein coding genes, not pseudogenes. The analyzed region starts at the position 45 and stops at position 693 of the COI gene of the mitochondrial genome of *Drosophila melanogaster* Meigen, 1830 (AJ400907) (Adams et al. 2000) used as the reference genome. The amino acid reading frame starts at the second base of the dataset. The data matrix shows 116 invariant sites and a mean A+T content of 67.4%.

The Neighbour-Joining and Maximum likelihood analyses showed 105 species, from the 133 Neotropical species with available barcodes, separated into distinct clusters. Genera represented by more than one taxon formed cohesive assemblages of five clusters [*Anopheles, Orthopodomyia, Psorophora, Toxorhynchites* Theobald, 1901, *Uranotaenia* Lynch Arribalzaga, 1891] in the NJ analysis and eight [*Anopheles, Coquillettidia, Culex, Orthopodomyia, Psorophora, Stegomyia* Theobald, 1901, *Toxorhynchites, Uranotaenia*] in the ML analysis.

The NJ tree based on Tamura-Nei genetic distances (Fig. 2) revealed most of the species clusters (100 species) with high bootstrap value (97-99). Conversely, 28 species were recovered as non-monophyletic groups during this analysis: An. punctimacula Dyar & Knab, 1906 and An. neomaculipalpus Curry, 1931 as paraphyletic groups, an overlapping of Ps. insularia (Dyar & Knab, 1906) and Ps. pygmaea (Theobald, 1903), four species of Culex [Cx. declarator, Cx. nigripalpus, Cx. conspirator, and Cx. spinosus], and 20 species of Anopheles (Nyssorynchus) Blanchard, 1902 [myzorynchella section = An. antunesi Galvao & Amarai, 1940, An. lutzii Cruz, 1901, An. pristinus Nagaki & Sallum, 2010; nuneztovari complex = An. goeldi Rozeboom & Gabaldon, 1941, An. dunhami, An. nuneztovari Gabaldon, 1940; oswaldoi complex = An. evansae (Brethes, 1926), An. galvaoi Causey, Deane & Deane, 1943, An. konderi Galvao & Damasceno, 1942, An. oswaldoi (Peryassu, 1922), An. rangeli Gabaldon, Cova-Garcia & Lopez, 1940; strodei complex = An. albertoi Unti, 1941, An. arthuri Unti, 1941, An. rondoni (Neiva & Pinto, 1922), An. strodei Root, 1926; albitarsis complex = An. albitarsis Lynch Arribalzaga, 1878, An. janconnae Wilkerson & Sallum, 2009, An. marajoara Galvao & Damasceno, 1942, An. oryzalimnetes Wilkerson & Motoki, 2009; and An. benarrochi Gabaldon, Cova-Garcia & Lopez, 1941].



**Figure 2.** Neighbour-Joining tree of the barcoding sequences of mosquito species listed for Neotropics, based on Tamura-Nei genetic distances. Terminal branches have been collapsed in order to save space (see original tree in Suppl. material 5 and 6). Branches in colors indicate non-monophyletic genera. Red clusters represent groups with problems to discriminate species. Names with green asterisk indicate non-monophyletic species. Bootstrap values above 60 (1,000 replicates) are given at the nodes.



**Figure 3.** Maximum likelihood tree of the barcoding sequences of mosquito species listed for the Neotropics. Terminal branches have been collapsed in order to save space (see original tree in Suppl. material 7 and 8). Branches in colors indicate non-monophyletic genera. Red clusters represent groups with problems to discriminate species. Names with green asterisk indicate non-monophyletic species. Bootstrap values above 60 (1,000 replicates) are given at the nodes.

The likelihood score for the best ML tree was -31,760.30891. The overall topologies of the ML (Fig. 3) and NJ trees compared favorably with exception of *Coquillettidia*, *Culex*, and *Stegomyia*, which resolved as monophyletic clusters. The ML tree revealed 92 species clusters with high bootstrap (95–99). The bootstrap on the remaining 13 species ranged from 65 to 93. ML analysis also recovered 28 species as non-monophyletic groups.

### Discussion

In this study 22 species belonging to eight genera and 11 subgenera were identified by combining morphological and molecular methodologies. Although our data and sampling size were limited, the combination of methodologies provided better success in species identification. Overall, congruence between morphology and barcode grouping, based on cluster monophyly with high (more than 95%) bootstrap support, was found in 18 of the 22 morphologically defined taxa (82%). Similarity methods based on the match between the query sequence and the reference database (more than 98% identity between BOLD/BLAST) only discriminated three mosquito species of this study. Similarity methods based on the match between the query sequence and the reference databases represent the least suitable method to discriminate mosquito species in this study.

Tree-based diagnostics are graphic criterion for species recognition, which describes genetic similarity in a visually satisfying style (Goldstein and DeSalle 2010). An important advantage of using a tree-based approach is that they present a direct sense of the statistical reliability and do not retrieve a positive result if no matching diagnostics are found (unlike the best match algorithm of BLAST and BOLD which retrieves the closest match but requires the user to interpret its reliability) (Goldstein and DeSalle 2010). Here, we compared two tree-based methods (neighbour-joining and maximum likelihood), which provided similar results. The NJ tree computed was in general agreement with previously published taxonomy based approaches (Hebert et al. 2003a, 2003b, Hebert et al. 2004, Hajibabaei et al. 2007, Cywinska et al. 2006, Kumar et al. 2007). Both tree-based analysis in this study showed 105 species of mosquitoes (from 133 total species) separated into distinct clusters. Since the NJ clustering performed considerably faster than ML approach and indeed has been used in the great majority of published barcoding studies, these results indicate it is an efficient choice for mosquito barcode analyses, similar to what is observed for other organisms (Little and Stevenson 2007, Elias et al. 2007).

The usefulness of DNA barcodes to discriminate mosquito genera of our dataset by using a tree-based approach was poorly supported. Approximately half of currently recognized genera represented by two or more species formed stable clusters in the NJ tree. Similarly, generic monophyly was weakly improved with the ML approach. In other insect genera, the monophyletic clusters based on DNA barcodes varies greatly depending on clustering method: e.g. Lepidoptera: Ithomiinae 50 to 61% recovered generic monophyly (Elias et al. 2007), Diptera: Chironomidae 40% (Ekrem et al. 2007, 2010), Simuliidae 62% (Rivera and Currie 2009) and Muscidae 40% (Renaud et al. 2012), Hymenoptera: Apoidea 100% success (Sheffield et al. 2009). It remains unclear whether this is due to lack of phylogenetic signal in COI at this depth, the type of tree-building method, or to the true lack of monophyly of genera as currently defined (Renaud et al. 2012). Conversely, a high level of correspondence at the species level is observed between morphology and molecular species limits in the tree-based approach in the present study. The performance of DNA-based specimen identification in Diptera using COI differs greatly in the literature, which varies from less than 50% to near 100% congruence levels (Cywinska et al. 2006, Smith et al. 2006, Whitworth et al. 2007, Rivera and Currie 2009, Meiklejohn et al. 2012, Renaud et al. 2012, Smit et al. 2013, Contrearas et al. 2014). In most of the studies, identification success rose upon relaxing the bootstrap requirement.

In tree-based methods, the non-monophyly at the species level represents the greatest challenge for taxon sampling and threshold approach (Meyer and Paulay 2005). Funk and Omland (2003) explained five possible reasons for non-monophyly at the species level, i.e. inadequate phylogenetic information, imperfect taxonomy, interspecific hybridization, incomplete lineage sorting, and unrecognized paralogy. There are cases in which tree-based analyses of DNA barcodes have failed to discriminate species of insects (Wiemers and Fiedler 2007, Whitworth et al. 2007, Foster et al. 2013). The non-monophyletic groups of our study includes thirty species of *Anopheles*, two species of *Psorophora*, and four or five, depending on the tree reconstruction method, species of *Culex*.

Culex was the most diverse and species rich genus in the sampled mosquitoes. *Culex* is a cosmopolitan genus and one of the largest groups of the family Culicidae (768 species divided among 26 subgenera). There are many areas of uncertainty regarding phylogenetic relationships within the genus, as well as some problems with the identification of some species. In our study, many difficulties arose attempting to identify specimens of Culex species. In many species, female identification is very problematic due to polymorphisms and ambiguities of the morphological characters (Forattini 2002, Laurito et al. 2013). Morphological identification of Culex species is based primarily on differences in male genitalia (Harbach 2011). Moreover, the presence of unknown species complexes within *Culex* makes species identification challenging (Harbach 2011, Laurito et al. 2013). As research previously suggested, the non-monophyly of given species of Culex approached during this study is not a surprise. Furthermore, in earlier studies only 42% of the Culex (Culex) previously identified morphologically to species were clustered with their conspecifics in the NJ tree (Laurito et al. 2013). Although our data are limited, present results point in the same direction and more taxonomic work is needed to assess the monophyly of Culex and the phylogenetic relationships within the genus.

Classification of the species within the genus *Anopheles* has always been challenging. The current system of subgeneric classification is based primarily on characteristics of the male genitalia (Harbach 2013). Many *Anopheles* species are morphologically indistinguishable and form cryptic species complexes, which require a molecular approach as the only effective tool for resolving their identification (Foster et al. 2013). The DNA barcode approach has been successfully used in corroborating lineages within the *Anopheles albitarsis* complex (Ruiz-Lopez et al. 2012). In the present study, difficulties in resolving species among the sequences of the subgenus *Anopheles (Nys-sorhynchus)* were expected. Most of the overlapping of species was obtained among one section (*myzorynchella* section) and four of the five complexes comprising this group (*nuneztovari, oswaldoi, strodei, albitarsis*). Even if species level could not be reached for this difficult subgenus, barcodes allowed us to clearly identify the different species complexes present. Single genes, including the COI barcode region, are poor at confirming morphologically defined species and to estimate phylogenetic relationships within the subgenus *Nyssorhynchus* (Foster et al. 2013). However, a multi-locus approach (COI barcode region, nuclear white and CAD genes) was able to discriminate *Nyssorhynchus* species with greater accuracy (Foster et al. 2013).

The most important factor affecting the accuracy of species identification through public databases is the coverage and reliability of available sequences (Ekrem et al. 2007, Wiemers and Fiedler 2007, Virgilio et al. 2010). Unfortunately, the GenBank and BOLD databases have many records believed to be from misidentified specimens (Harris 2003, Meier and Dikow 2004, Meier et al. 2008), which are only obvious by incorporating accurately identified specimens into databases (Meyer and Paulay 2005, Meier et al. 2006). The importance of voucher specimens which can be reexamined is paramount. Furthermore, erroneous sequences may lead to an underestimation of the potential for species discrimination by DNA barcodes (Pape et al. 2009, Collins and Cruickshank 2013). In total, sequences for 133 Neotropical species were included in the tree-based analysis, which displayed species discrimination among most of the taxa, but not for all. We expect that as more studies provide sampling data, the number of entries will increase in databases, which should greatly improve the accuracy of search queries. Nonetheless, results of best match retrieved from any of the databases should not be used indiscriminately without considering the reliability of the comparison. Although BLAST and BOLD Identification System retrieved incongruent identifications for some of our sequences, these search tools are useful to check for sequence contaminations, especially when a species is sequenced for first time.

It is clear that reference libraries with properly identified sequences will facilitate the association of conspecific specimens and the detection of identification errors. The DNA barcodes produced in this work allowed for the identification of females and damaged specimens, which could not be done using morphological characteristics alone. This study highlights the potential of barcoding methodology to resolve taxonomic problems associated with limitations in morphological identification, but also draws attention to its limitations to discriminate species in some Neotropical mosquito genera, especially in *Anopheles (Nyssorhynchus)* species complexes and some *Culex* species. A larger taxon barcode library with correctly identified vouchers will help future studies. Despite the limitations in our survey, the DNA barcodes produced in this work are an important contribution to increase the scope of reference libraries with properly identified sequences and to assist in the identification of mosquito species from Colombia.

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# Supplementary material I

### Mosquito specimens collected in the present study

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: occurrence

- Explanation note: Full collection site details, including geo-references and environmental conditions, of the mosquito taxa identified in this study.
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### Supplementary material 2

#### COI mosquito sequences downloaded from NCBI and BOLD

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: Text

- Explanation note: Sequences from NCBI and BOLD. Sequences downloaded between December 2013 and February 2014 for all identified species with a minimum length of 480 bp of COI barcoding region, no stop codons and possible alignment among the majority of the sequences. Total of 1,159 sequences. In blue: Additional species sequences from other geographic areas for those Neotropical groups without available sequences from the Neotropics.
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# Supplementary material 3

# Mosquito specimens collected in Uraba during 2009 (unpublish data)

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: occurrence

Explanation note: Additional sequences of mosquitoes collected in rural areas of Uraba (Antioquia, Colombia) during 2009, including sampling information.

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# Supplementary material 4

# Outgroup taxa used in the present study

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: Text

- Explanation note: Outgroup taxa used in the present study. Sequences downloaded from BOLD between December 2013 and February 2014.
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# Supplementary material 5

# NJ tree based on 1,292 sequences of Neotropical mosquitoes

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: picture

Explanation note: Original Neighbour-Joining tree of the barcoding sequences of mosquito species listed for the Neotropics, based on Tamura-Nei genetic distances.

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# Supplementary material 6

### NJ tree based on 1,292 sequences of Neotropical mosquitoes

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: multimedia

- Explanation note: Original Neighbour-Joining tree of the barcoding sequences of mosquito species listed for the Neotropics, based on Tamura-Nei genetic distances (.tre).
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# Supplementary material 7

### ML tree based on 1,292 sequences of Neotropical mosquitoes

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: picture

- Explanation note: Original Maximum likelihood tree of the barcoding sequences of mosquito species listed for the Neotropics.
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# Supplementary material 8

### ML tree based on 1,292 sequences of Neotropical mosquitoes

Authors: Paula Rozo-Lopez, Ximo Mengual

Data type: multimedia

- Explanation note: Original Maximum likelihood tree of the barcoding sequences of mosquito species listed for the Neotropics (.tre).
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RESEARCH ARTICLE



# Australian Allograpta Osten Sacken (Diptera, Syrphidae)

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#### Abstract

*Allograpta terraenovae* **sp. n.** and *Allograpta notiale* **sp. n.** are described from Australia. Notes on the Australian species of *Allograpta* and an identification key to them are also given. The lectotype of *Allograpta javana* Wiedemann is designated, and the species *Syrphus pallidus* Bigot is synonymized under *Allograpta australensis* (Schiner).

#### **Keywords**

Australia, flower fly, hoverfly, new species, description, identification key

# Introduction

*Allograpta* Osten Sacken, 1875 (Diptera, Syrphidae) is a world-wide genus with its greatest diversity in the Neotropics (Thompson et al. 2010). Adult flies are pollinators and flower visitors and larvae are mostly predators of soft-bodied Hemiptera (Rojo et al. 2003), although secondarily some species are phytophagous (Nishida et al. 2003, Zuijen and Nishida 2011, Weng and Rotheray 2009). The genus was recently reviewed (Mengual et al. 2009) and based on morphological characters and molecular evidence (Mengual et al. 2008a, 2008b, 2012), six different genera are currently recognized instead of the previous subgenera (Thompson 2012).

Only three *Allograpta* species were known for the Australian fauna, *Allograpta ala-macula* Carver, 2003, *A. australensis* (Schiner, 1868) and *A. pallida* (Bigot, 1884) (Mengual et al. 2009), but no key or review of these has been published. The aim of the present work is to review the Australian species of this flower fly genus and to describe two new species.

# Material and methods

New species are described in full, following the terminology by Thompson (1999). The holding collection of each specimen is indicated between square brackets after the label information. The abbreviations used for collections follow the standard of the *Systema Dipterorum* (Thompson 2013), and their equivalents are given below:

ANIC	Australian National Insect Collection, CSIRO, Canberra City, Australia.
AMS	Australian Museum, Sydney, Australia.
CNC	Canadian National Collections of Insects, Ottawa, Canada.
NMW	Naturhistorisches Museum Wien, Vienna, Austria.
QM	Queensland Museum, South Brisbane, Australia.
USNM	National Museum of Natural History, Washington D.C., United States of
	America.
UMO	University Museum of Natural History, Oxford, United Kingdom.
ZMUC	Zoological Museum, University of Copenhagen, Copenhagen, Denmark.
ZFMK	Zoologisches Forschungsmuseum Alexander Koenig, Bonn, Germany.

Italics in the description of type labels denote handwriting, the contents of each label is enclosed within double quotation (""), and the individual lines of data are separated by a double forward slash (//). All measurements are in mm and were taken using a reticule in a Leica M165 C microscope (Wetzlar, Hesse, Germany). Photographs were composed using the software Zerene Stacker 1.04 (Richland, Washington, USA) based on images of pinned specimens taken with a Canon EOS 7D mounted on a P–51 Cam-Lift (Dun Inc., Virginia, USA) and the help of Adobe Lightroom (version 5.6) (San Jose, California, USA). Distribution maps were created using SimpleMappr (Shorthouse 2010).

# Key to the Australian species of Allograpta

1	Metasternum bare; wing with apical brown macula. Male genitalia large, vis-
	ible from dorsal view (Fig. 1)alamacula Carver
_	Metasternum pilose; wing without apical macula. Male genitalia smaller,
	usually underneath tergum 5 <sup>th</sup> (Figs 2, 3, 5) <b>2</b>
2	Katepimeron, coxae, pro- and mesotarsus yellow (Fig. 3)
	australiensis (Schiner)

	Katepimeron and coxae black; pro- and mesotarsus with apical tarsomeres
	dark brown to black (Figs 5, 7, 15, 16)
3	Postalar callus yellow pilose (Figs 5, 7, 9, 11); occiput yellow pilose (Figs
	5-8); male frontal triangle yellow pilose (Figs 5, 6), with small medial brown
	macula dorsad to antenna (Fig. 6) terraenovae sp. n.
_	Postalar callus partially black pilose (Figs 10, 12, 16); occiput black pilose
	on dorsal 1/3 (Figs 10, 13-16); male frontal triangle black pilose (Fig. 13),
	with large brown macula dorsad to antennae and reaching laterally around
	antenna (Fig. 13)notiale sp. n.

### Australian species of Allograpta

### Allograpta alamacula Carver, 2003

Figure 1, 17

*Allograpta alamacula* Carver in Carver and Thompson 2003: 37; fig. 1 (habitus), fig. 2 (male genitalia). Type-locality: Australia, Queensland, Indooroopilly [HT male, ANIC].

**Diagnosis.** Face straight, with large tubercle; oral opening about 2 times as long as wide, with oral apex at level of antennal base; antennal pits confluent; plumula absent; subscutellar pile fringe absent; wing broadly bare basomedially, with apical dark macula; alula broad, as broad as cell bm; metasternum bare; abdomen elongate.

**Biology.** Carver reared her species from maggots preying on the whitefly species *Aleurocanthus t-signatus* (Maskell, 1896) (Hemiptera, Aleyrodidae) (Carver and Thompson 2003).

Distribution. Australia (New South Wales, Queensland); Fig. 17.

### Allograpta australensis (Schiner, 1868)

Figures 2-4, 17

- *Melithreptus australensis* Schiner, 1868: 347. Type-locality: Australia, Sydney [HT female, NMW]. Holotype presumably lost (Vockeroth 1971: 1628), corroborated by first author (XM).
- Syrphus pallidus Bigot, 1884: 93. Type-locality: "Australie" [HT male, UMO]. Kertész 1910: 124 (cat. cit.); Hull 1936: 194 (distr.). Syn. n.
- Sphaerophoria australensis: Kertész 1910: 135 (cat. cit.); Curran and Bryan 1926: 129 (descr. note, distr.); Hull 1936: 195 (distr.).
- Allograpta javana (Wiedemann, 1824) of Australian authors; Hardy, 1933: 13 (as Sphaerophoria javana; distr., syn. of australensis Schiner).
- *Allograpta australensis*: Vockeroth 1971: 1628 (rev. status, diff. *iavanus* Wiedemann, figures); Thompson and Vockeroth 1989: 441 (cat. cit.).
- Allograpta pallida: Thompson and Vockeroth 1989: 442 (cat. cit.).

**Diagnosis** (modified from Vockeroth 1971). Face produced forward below, ventral margin of head long and oblique; yellow maculae of  $2^{nd}$  tergum (sometimes fused in female) narrowed laterally but extending narrowly to margins of tergum; yellow fasciae of  $3^{rd}$  and  $4^{th}$  terga strongly narrowed laterally and not extending to margins of terga; apex of metafemur and broad base and apex of metatibia brown to yellow-brown or black, the yellow annulus (ring) at middle of metatibia very poorly defined (some males without annulus).

**Distribution.** Australia (Queensland, New South Wales, Northern Territory, Norfolk Island); Fig. 17.

**Biology.** In the ANIC collection, there is a series of males and females collected as larvae on flowers of *Eucalyptus*.

**Remarks.** The holotype of *Syrphus pallidus* Bigot was examined and was found to be the same species as *australensis* Schiner.

### Allograpta javana (Wiedemann, 1824)

*Syrphus iavanus* Wiedemann, 1824: 34. Type-locality: Indonesia, Java [LT here designated, NMW].

*Syrphus javanus*: Wiedemann 1830: 131 (emendation, redescription); Zimsen 1954: 19 (no type specimen in Copenhagen), here verified.

Sphaerophoria javana: Kertész 1910: 136 (cat. cit.).

*Xanthogramma javana*: Bezzi, 1928: 73 (descr. (A, P), distr.); Hull 1936: 195, 1937: 83 (distr.).

Miogramma javana: Frey 1946: 165 (comb.).

Helenomyia javana: Bańkowska 1962: 311 (comb.).

Allograpta javana: Vockeroth 1969: 130 (comb.).

Allograpta iavana: Thompson and Vockeroth 1989: 441 (cat. cit.).

**Diagnosis** (modified from Vockeroth 1971). Face nearly vertical, ventral margin of head shorter and more nearly horizontal; yellow maculae of 2<sup>nd</sup> tergum (usually fused in female) and yellow fasciae of 3<sup>rd</sup> and 4<sup>th</sup> terga not narrowed laterally, extending to margins of terga in their full width; apex of metafemur and broad base and apex of metatibia dark brown to black, the yellow annulus at middle of metatibia sharply defined.

**Distribution.** India, Sri Lanka, north to Mongolia, China, Korea, Primorye (Primorsky Krai, Russia) and Japan, east to New Guinea, Solomon Islands and Fiji.

**Remarks.** Allograpta javana (Wiedemann) was confused with A. australensis (Schiner) until Vockeroth (1971) separated the two. All material we have examined is of A. australensis and undescribed species, so the status of the A. javana in Australia is still dubious (Vockeroth also did not know A. javana from Australia). Allograpta javana is similar to A. australiensis in that the katepimeron is yellow, but differs from A. australiensis in the vertical face, not projecting anteriorly (Vockeroth 1971: 1629, figures 1 and 2); abdominal fasciae not narrowed laterally and extending to margins



**Figures 1–8.** Australian *Allograpta* species: **1** *Allograpta alamacula*, male, dorsal **2–4** *Allograpta australensis*: **2** male, dorsal **3** male, lateral **4** male, frontal **5–8** *Allograpta terraenovae* sp. n.: **5** male (holotype), lateral **6** male (holotype), frontal **7** female (paratype), lateral **8** female (paratype), frontal. Scale for lateral and dorsal views: 2 mm. Scale for frontal views: 1 mm.

in their fullest widest and the yellow medial annulus on metatibia distinct. Other morphological characters to distinguish these two species are: anepimeron black in *A. javana*, including dorsomedial portion of anepimeron (dorsomedial anepimeron yellow in *A. australensis*); male frons yellow pilose in *A. javana* (mainly black pilose with some yellow pili in *A. australensis*); male with occiput yellow pilose in *A. javana* (male with occiput black pilose on dorsal 1/3 and yellow pilose on basal 2/3 in *A. australensis*); and usually metabasitarsomere pale in *A. javana* (metabasitarsomere dark in *A. australensis*).

Allograpta javana is a species complex, whose components probably should be recognized as full species. In at least eastern New Guinea and throughout Oceania, the face is entirely yellow [Allograpta distincta (Kertész, 1899: 177)], whereas the western component always has a broad black medial vitta (typic form). In Oceania, A. amphotera Bezzi (1928: 74) and A. nigripilosa Hull (1944: 52) are related more closely to A. javana (typic form) than to A. distincta, as they retain the black facial vitta.

Zimsen (1954: 19) mentions that there was no type specimen in the Zoological Museum of Copenhagen (ZMUC) or in Vienna (NMW), but Groll (2013) indicates that some Diptera and Hymenoptera of the Wiedemann collection went to Vienna via W. von Winthem. At NMW, there is a pinned male labelled: "*Java //* Coll. Winthem" "*javannus* // det. Wiedem." "*javanus Wid // Java*" "*LECTOTYPE // Allograpta // iavana // WIEDE-MANN //* K. Ghorpade des. 1983" [red] "LECTOTYPE // Allograpta // javana Wied. // des. X. Mengual 2014" [red]. We do agree with Ghorpadé in considering this specimen as part of the type series studied by Wiedemann. This specimen is here designated as the lectotype to fix and ensure the universal and consistent interpretation of the name.

#### Allograpta terraenovae Thompson, sp. n.

http://zoobank.org/FAA81060-E319-4773-8F12-D8D9EA4D3BAB Figures 5–9, 11, 18

Allograpta 88–13 Thompson in litt.

**Type locality.** AUSTRALIA: Queensland, Jowalbinna, 6.7 km west of, 15°45'S, 144°12'E.

**Types.** Holotype male labelled: "6.7 km W 'Jowalbinna' // H.S., Qld 15°45'S, 144°12'E // 11 May 1989 // G. and A. Daniels" "Australian Museum // K402264" "Holotype // Allograpta // nsp. 88–13 // Thompson" [red] "HOLOTYPE // Allograpta // terraenovae // Thompson 2014" [red] [AMS].

*Paratypes*: AUSTRALIA, NEW SOUTH WALES: Marsfield, vii.1976, C.E. Chadwick [1 $\bigcirc$  AMS; AMK 404830]; Mooney Mooney Creek near Gosford, 3.xii.1976, D.K. McAlpine [1 $\bigcirc$  USNM; AMK 410638]. QUEENSLAND: 6.7 km West of 'Jowalbinna' H.S., 15°45'S, 144°12'E, 11.v.1989, G. & A. Daniels [1 $\checkmark$  AMS; AMK 402265]; Brisbane, 7.ix.1927, J. Mann [1 $\bigcirc$  QM; UQIC 220474]; Brisbane, C. Deane [1 $\bigcirc$  QM; UQIC 220475]; Brisbane, 10.x.1916, H. Hacker [1 $\bigcirc$ 



Figures 9–16. Australian *Allograpta* species: 9 *Allograpta terraenovae* sp. n., female (paratype), dorsal 10 *Allograpta notiale* sp. n., male (holotype), dorsal 11 *Allograpta terraenovae* sp. n., male (holotype), dorsal 12–16 *Allograpta notiale* sp. n.: 12 female (paratype), dorsal 13 male (holotype), frontal 14 female (paratype), frontal 15 female (paratype), lateral 16 male (holotype), lateral. Scale for lateral and dorsal views: 2 mm. Scale for frontal views: 1 mm.

QM; UQIC 222111]; Brisbane, C.F. Ashby [2<sup>3</sup>/<sub>2</sub> 2<sup>2</sup>/<sub>2</sub> ANIC; ANIC 33153]; Bundaberg, viii.1971, H. Frauca [1<sup>(2)</sup> ANIC; ANIC 33157]; 2 miles North of Bundaberg, 26.vi.1971, Tea-tree swamp, H. Frauca [1º ANIC; ANIC 33159]; 14 km West by North Hope Vale Mission, 4.v.1981, D.H. Colless [1<sup>Q</sup> ANIC; ANIC 33160]; 20 miles South of Ingham, 1.ix.1956, C. Deane [2<sup>Q</sup> QM; UQIC 220476, 220477]; 3km S Mt Spurgeon, 1100 m., 20–22.xi.1997, C. Burwell, open forest [1 Q QM, UQIC 221457]; 7-14 miles West of Herberton, via Watsonville, 1.v.1967, D.H. Colless [1° ANIC; ANIC 33154]; Brisbane, 25.x.1953, F.M. Hull [2° CNC]; Flinders Mem. Park, 23.v.1968, J.W. Boyes [1] CNC; USNM ENT00249235]; Brisbane, Toowong, 26.v.1968, J.W. Boyes [1° CNC]; Bluff Range, near Biggenden, ca. 2750 ft., v.1971, H. Frauca [1º ANIC]; Atherton, 3-18.xi.1972, A.M. Hemmingsen [13 ZMUC]; Carnarvon Stn, nr Piebald Spring (CN1M1), 821 m., 13.xii.2010-15.vi.2011, C. Zwick & C. Wilson, malaise trap, Eucalyptus/ Callistemon in rocky gully [1º QM; UQIC 222109]; Dunk Island, 25.viii.1927 [2<sup>Q</sup> QM; UQIC 222110, 220478]; Great Sandy National Park, Cooloola Section, 1–5.x.1996, Winterton, D.K. Yeates, C. Lambkin, malaise trap  $[1]^{\circ}$  QM; UOIC 220473]; Mandalay Point, Great Barrier Reef, 13.viii.1986, De Beer [1] QM; UQIC 220479]; Mount Glorious, 6.xi.1965, C.F. Ashby [1 d 1 2 ZFMK, 1 ANIC, 1 USNM; ANIC 33152, USNMENT 01028878]; Mount Glorious, 8.xi.1965, J.K. Guyomar [2& ANIC; ANIC 33156]; Mount Glorious Scrub Creek Road, Brisbane Forest Park, 17–24.x.1997, N. Power, malaise trap [1Å QM; UQIC 220471]; Mt. Abbott, upper slopes, 700–900 m., 10–12.iv.1997, C. Burwell [2] 5♀ QM; UQIC 221451, 221452, 221447, 221448, 221450, 221454, 221456; 2♀ ZFMK; UQIC 221449, 221453]; Mt. Robert, 5km SW, 300 m., 23.x.2000, S. Wright, brigalow [1° QM; UQIC 222112]; Petrie Park, Mayborough, 15.xi.1993, G. & A. Daniels [1º AMS; AMK 402262]; Samsonvale Cemetary, 9.vi.1996, S.G. Evans [1 QM; UQIC 221462]; Scrub Road, Brisbane Forest Park, 12–19.Ix.1997, S. Winterton, N. Power, D. White, malaise trap  $[1 \stackrel{\bigcirc}{_{-}} QM; UQIC 220472]$ ; The Blunder, 20.ix.1969, C.F. Ashby [1<sup>Q</sup> ANIC; ANIC 33158]; Toomba, Site 1, 390 m., 14–15.xii.2006, S. Wright, rainforest/paddock edge [1] QM; UQIC 222113]; Undara Volcanic National Park, The Bluff, 19.vii.1998, J.& R. Skevington [2] QM; UQIC 220459, 220461; 1 ZFMK, UQIC 220460]; Undara Volcanic National Park, The Bluff, 770 m., 11.vi.1997, J.& R. Skevington, hilltop [6] QM; UQIC 220463, 220464, 220465, 220466, 220467, 220468, 220470; 1 ZFMK, UQIC 220462]; Hilltop ~21 km South of Coen, 14.137538°S, 143.240945°E, 945 m., 2.xii.2014, J.H., A.M. & A.W. Skevington [26] CNC; CNC373686, CNC373687, CNC373688, CNC373689, CNC373690, CNC373691, CNC373692, CNC373693, CNC373694, CNC373695, CNC373696, CNC373697, CNC373698, CNC373699, CNC373700, CNC373701, CNC373703, CNC373704, CNC373705, CNC373706, CNC373702, CNC373707, CNC373708, CNC373709, CNC373710, CNC373711]; Kroombit Tops National Park, hilltop, 24.44818°S, 150.93520°E, 21.xii.2014, J.H., A.M. & A.W. Skevington [4<sup>2</sup>/<sub>7</sub> CNC; CNC384420, CNC384422, CNC384423,
CNC384424]; Kroombit Tops National Park, hilltop, 24.448183°S, 150.935200°E, 22.xii.2014, J.H., A.M. & A.W. Skevington [1] CNC; CNC384468]; Sheoak Ridge Nature Reserve near Julatten, 16.645258°S, 145.403366°E, 7.xii.2014, J.H., A.M. & A.W. Skevington [3ex CNC; CNC371573, CNC371574, CNC371575]; Sheoak Ridge Nature Reserve near Julatten, Summit of hilltop in dry sclerophyll forest, 16.645258°S, 145.403367°E, 8.xii.2014, J.H. Skevington [34] CNC, 4] ZFMK, 4 USNM; CNC371588, CNC371589, CNC371590, CNC371591, CNC371592, CNC371593, CNC371594, CNC371595, CNC371596, CNC371597, CNC371598, CNC371599, CNC371600, CNC371601, CNC371602, CNC371603, CNC371604, CNC371605, CNC371606, CNC371607. CNC371608, CNC371609, CNC371610, CNC371611, CNC371612, CNC371613, CNC371614, CNC371615, CNC371616, CNC371617, CNC371618, CNC371619, CNC371620, CNC371621, CNC371622, CNC371623, CNC371624, CNC371625, CNC371626, CNC371627, CNC371628, CNC371629]; University of Queensland, St. Lucia, 16.viii.1994, G. Gordh, pupated 17.viii.1994, emerged 22.viii.1994 [18] QM; UQIC 220480]; Wilston, Brisbane, 28.vi.1998, S.G. Evans, pupated 2-3.vii.1998, emerged 10.vii.1998 [29 QM; UQIC 221460, 221461]; Wilston, Brisbane, 22.vi.1998, C.J. Burwell, on *Eucalyptus* feeding on lerps [1° QM; UQIC 221463]; Eidsvold, xii.1922 [1<sup>3</sup> ANIC; ANIC 33155]; Kuranda, F. P. Dodd [1<sup>3</sup> USNM; USNMENT 01028922]; 52km SWbyS of Mt. Garnet, 700 m., 18.05°S, 144.52°E, 28.v.1977, I.F.B. Common & E.D. Edwards [19 USNM; USNMENT 01028948].

**Diagnosis.** Species with metasternum pilose and katepimeron and coxae black. Very similar to *Allograpta notiale* sp. n., but *A. terraenovae* sp. n. has yellow pilosity on postalar callus and occiput, and male frons is also yellow pilose, with a small medial brown macula dorsad to antennae.

**Description.** Male. *Head.* Face yellow, with medial black vitta, shiny, yellow pilose; gena yellow on anterior 1/2, black posteriorly, yellow pilose; lunule yellow laterally, black medially; frontal triangle yellow except narrowly black immediately dorsad to lunule, shiny, yellow pilose; vertical triangle black, shiny, black pilose; occiput mostly black, yellow on ventral 1/9, white pilose on basal 2/3, and yellow pilose on dorsal 1/3; antenna orange, except basoflagellomere brownish black on dorsal 1/3, black pilose; arista brownish orange.

Thorax. Postpronotum yellow, shiny; propleuron black, sparsely silvery-white pollinose, white pilose; scutum black except broad yellow laterally, sparsely black pollinose, yellow pilose; postalar callus yellow, yellow pilose; scutellum yellow, black pilose; plumula orange; calypter light brownish except medial 1/3 of margin and fringe yellow; pleuron black except anepisternum yellow on posterior 2/3, katepisternum yellow on dorsal 1/3, and katatergum yellow on dorsal 4/5, sparsely silvery-white pollinose, white pilose; metasternum pilose. *Legs*: coxae black, sparsely silvery-white pollinose, white pilose; trochanters brownish black; pro- and mesofemora yellow, white pilose except black pilose on apical 1/3 dorsoposteriorly; metafemur yellow on basal 2/3, black apically, pale pilose basally, black pilose apically; pro- and mesotibiae yellow, yellow pilose; metatibia black, black pilose; tarsi brownish black, black pilose. *Wing*: hyaline, bare on basal 2/3, microtrichose apically; microtrichose on apical 1/4 of cell  $r_1$ , apical 1/2 cell  $r_{2+3}$ , apical 2/3 of cell  $r_{4+5}$ , apical 3/4 of cell dm, apical 1/2 of cell cup, and broadly along posterior margins of alula and anal lobe.

*Abdomen.* 1<sup>st</sup> tergum yellow except narrowly black on apical margin, yellow pilose; 2<sup>nd</sup> tergum black except for large yellow medial fascia, which may be narrowly separated medially, shiny along basal and apical margins, black pollinose bordering yellow fascia, yellow pilose on basal 3/4, black pilose apically; 3<sup>rd</sup> and 4<sup>th</sup> terga black except for large arcuate yellow fascia, shiny along basal and apical margins, black pollinose bordering yellow fascia, black pilose; 5<sup>th</sup> tergum black, except for large triangular yellow maculae, black pilose; sterna yellow, white pilose except 4<sup>th</sup> sternum black pilose. *Male genitalia* black, shiny, black pilose.

*Female*. Similar to male except for normal sexual dimorphism and as follows: frons yellow laterally (about 1/4 of frons width) with a medial, broad, black vitta (about 1/2 of frons width); abdominal fascia narrower than in male, very narrow medially on terga 3 and 4 looking like two joined maculae.

Distribution. Australia (New South Wales, Queensland); Fig. 18.

**Etymology.** The specific epithet is derived from the combination of *terra* (land, earth) and *nova* (new), and it refers to Australia. Species epithet to be treated as an adjective.

**Biology.** There is a female with a puparium (AMK 404830) that was collected as a larva preying on *Eucalyptolyma maideni* Froggatt, 1901 (Hemiptera, Psyllidae). Another female (UQIC 221463) was reared from a larva found feeding on lerps (Hemiptera, Psyllidae) on *Eucalyptus*.

#### Allograpta notiale Thompson, sp. n.

http://zoobank.org/0AB09328-B716-4F48-8FDF-83CF3BB56F36 Figures 10, 12–16, 18

Allogratpa 88–14 Thompson in litt.

#### Type locality. AUSTRALIA: Queensland, Collinsville, 20°33'S, 147°50'E.

**Types.** *Holotype* male labelled: "Collinsville, Q. // 15–9–1950 // E.F.Riek." "Australian // National // Insect // Collection" [green] "*Allograpta // 88–14 //* Det. X. Mengual, 2012" "ANIC Database No // 29 033161" "HOLOTYPE // *Allograpta // notiale // Thompson 2014*" [red] [ANIC].

*Paratypes*: AUSTRALIA, QUEENSLAND: Brisbane, 25.x.1953, F.M. Hull [1 $\checkmark$  CNC]; 18 miles North of Cairns, 13.v.1970, R. & J. Matthews [1 $\checkmark$  ZFMK, 1 $\checkmark$  ANIC; ANIC 33162]; 2 miles West of Kuranda, 7.v.1967, D.H. Colless [1 $\bigcirc$  ANIC; ANIC 33163]; 7 km North North West of Coen, 17.iv.1989, G. & A. Daniels [1 $\bigcirc$  AMS; AMK 402266]; Brisbane, C.F. Ashby [1 $\bigcirc$  ZFMK]; Dunk Island, 25.viii.1927 [1 $\bigcirc$  QM; UQIC 222114]; hut near East Claudie River, Iron Range National Park, 28.xii.1995, G. & A. Daniels [1 $\bigcirc$  AMS; AMK 410379]; Eungella National Park, Chelmer'S, Road, 21.132822°S, 148.492683°E, 19.xii.2014, J.H., A.M. & A.W. Sk-



**Figures 17–18.** Distribution of the Australian *Allograpta* species: **17** *Allograpta alamacula* (pink circles) and *Allograpta australensis* (green inverted triangles) **18** *Allograpta terraenovae* sp. n. (orange triangles) and *Allograpta notiale* (blue circles).

evington [1 $^{\circ}$  CNC; CNC384271]; Kuranda, F.P. Dodd [1 $^{\circ}$  ANIC, 1 $^{\circ}$  2 $^{\circ}$  USNM; ANIC 33166, USNMENT 01028912, 01028876, 01028865]; Leo Creek Roads, McIlwraith Range, 30 km Northeast of Coen, 500 m., 29.vi–4.vii.1976, C.B. & S.R. Monteith [1 $^{\circ}$  QM; UQIC 220482]; Mount Glorious, 10.xi.1965, C.F. Ashby [1 $^{\circ}$ ANIC; ANIC 33165]; Mount Glorious Scrub Creek Road, Brisbane Forest Park, 17– 24.x.1997, N. Power, malaise trap [1 $^{\circ}$  QM; UQIC 220481]; Shiptons, Flat, Roberts' house, 250 m., 1.viii.2004, S. Wright, cleared paddocks [1 $^{\circ}$  QM; UQIC 222115]; Shiptons Flat, 15.47°S, 145.14°E, 18.v.1981, D.H. Colless [1 $^{\circ}$  USNM; USNMENT 01028856].

**Description.** Male. Similar to *Allograpta terraenovae* except differs as follows: gena all black; occiput black pilose on dorsal 1/3; scutum shiny except sparsely pollinose on anterior margin, black pilose except yellow pilose on lateral yellow vitta; postalar callus mainly black pilose, with a few intermixed yellow pili anteriorly; callypter brownish black; abdominal fasciae narrower, with a linear posterior margin (not emarginated medially).

*Female*. Similar to male except for normal sexual dimorphism and as follows: frons yellow laterally (about 1/6 of frons width) with a medial, broad, black vitta (about 2/3 of frons width).

Distribution. Australia (Queensland); Fig. 18.

**Etymology.** The specific epithet is derived from the Latin *notialis* meaning southern (Brown 1956: 731), and it refers to Australia. Species epithet to be treated as an adjective.

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RESEARCH ARTICLE



# Description of two new filtering carnivore Drusus species (Limnephilidae, Drusinae) from the Western Balkans

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Dedicated to Hans Malicky on the occasion of his 80th birthday

#### Abstract

Two new species of the genus *Drusus* (Trichoptera, Limnephilidae, Drusinae) from the Western Balkans are described. Additionally, observations on the biodiversity and threats to the region's endemic aquatic fauna are discussed. *Drusus krpachi* **sp. n.** is a micro-endemic of the Korab Mountains, Macedonia, and *D. malickyi* **sp. n.** is a micro-endemic of the Prokletije Mountains, Albania. Both new species are most similar to *D. macedonicus* but differ from the latter in the shape of segment IX, the shape of the tips of the intermediate appendages in lateral view, the shape of the inferior appendages, and the form and shape of the parameres. In addition, males of the European species of filtering carnivore Drusinae are diagnosed and illustrated, including *Cryptothrix nebulicola* McLachlan, *Drusus chrysotus* Rambur, *D. discolor* Rambur, *D. macedonicus* Schmid, *D. meridionalis* Kumanski, *D. muelleri* McLachlan, *D. romanicus* Murgoci and Botosaneanu, and *D. siveci* Malicky. These additions to the Western Balkan fauna demonstrate the significance of this region for European biodiversity and further highlight the importance of faunistic studies in Europea.

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#### **Keywords**

Caddisfly, aquatic diversity, Mediterranean, taxonomy, conservation, Southern Europe

# Introduction

The Western Balkans harbour high biodiversity including high numbers of endemic species. This has been attributed to historic climate conditions and the highly diverse geology of the region (e.g., Neubauer 2002, Reed et al. 2004, Chiari et al 2011) that permitted perseverance of taxa in glacial refugia (e.g., Tzedakis 2004, 2009; Médail and Diadema 2009), and the formation of diverse habitats. Thus, the Western Balkans are rich in endemic plant (e.g., Eastwood 2004, Petrova and Vladimirov 2010, Mereda et al. 2011, Redžić 2011), vertebrate and invertebrate species (e.g., Bianco 1998, Bănărescu 2004, Kryštufek 2004, Griffiths and Frogley 2004, Bohlen et al. 2006, Guéorguiev 2007, Deltshev 2008, Pešić and Glöer 2013). The Western Balkans also have been identified as a hotspot of aquatic biodiversity, with high endemism and cryptic diversity (Bănărescu 2004, Previšić et al. 2014a). Climate change and its detrimental effects on biodiversity (e.g., Hering et al. 2009, Bálint et al. 2011) have focused attention on freshwater biota throughout Europe, including the Western Balkans (e.g., Zakšek et al. 2009, Klobučar et al. 2013, Weiss et al. 2014).

Faunal studies on Western Balkan aquatic biodiversity recovered intriguing biogeographic patterns and several new species (e.g., Petkovski et al. 2009, Pešić and Glöer 2013, Vitecek et al. 2015). Research on the caddisfly fauna of the Western Balkans further suggests several factors, such as karstification, as drivers of speciation (Previšić et al. 2009, 2014b). The limnephilid subfamily Drusinae is particularly species-rich in the Western Balkans, including a high proportion of micro-endemics *sensu* Graf et al. (2008), i.e., taxa restricted to small geographic units within an ecoregion *sensu* Illies (1978) (Malicky 2004; Graf et al. 2008; Oláh 2010, 2011; Kučinić et al. 2011a,b; Oláh and Kovács 2013; Previšić et al. 2014a, b; Vitecek et al. 2015; Ibrahimi et al. pers. comm.).

The subfamily Drusinae Banks comprises roughly 110 species in 8 genera (Malicky 2004, 2005; Oláh 2010, 2011; Oláh and Kovács 2013; Previšić et al. 2014a; Vitecek et al. 2015; Ibrahimi et al. pers. comm.). Ecologically, most species are crenobiont (Graf et al. 2008), and as larvae fall into one of three different feeding groups: filtering carnivores, omnivorous shredders and scraping grazers (Pauls et al. 2008, Graf et al. 2009). The adults of each larval feeding group are also characterized by a set of synapomorphies (Vitecek et al. in press). Filtering carnivorous Drusinae males uniquely exhibit laterally positioned gland openings at the fifth abdominal sternite and parallel wing veins in the hind wing anal field (depicted in Vitecek et al. in press). The largest genus *Drusus* is paraphyletic (Pauls et al. 2008; Kučinić et al. 2011a; Oláh 2010, 2011; Oláh and Kovács 2013; Vitecek et al. 2015; Vitecek et al. in press; Ibrahimi et al. pers. comm.). The monotypic genus *Cryptothrix (C. nebulicola* McLachlan) is also a filtering carnivore (Bohle 1987, Graf et al. 2008), and thus represents another filtering carnivorous Drusinae *sensu* Pauls et al. 2008, the systematic position of which is discussed in Vitecek et al. (in press).

Here we describe two new filtering carnivore *Drusus* species. Additionally, we provide re-descriptions of filtering carnivorous Drusinae *sensu* Pauls et al. (2008) in order to facilitate identification of known filtering carnivorous Drusinae, and identification of new species.

#### Materials and methods

Adults were collected using sweep nets and by handpicking. Collected specimens were stored in 96% EthOH. Male and female genitalia were examined after being cleared in either KOH or lactic acid. Nomenclature of male genitalia of *Drusus* follows Nielsen (1957, for *Limnephilus flavicornis* Fabricius) using the simplifying terms "superior appendages" for the lateral processes of segment X (cerci *sensu* Snodgrass 1935), and "intermediate appendages" for the sclerite and the anterior process of segment X (paraproct *sensu* Snodgrass 1935). Nomenclature of larval morphological features follows Wiggins (1998) and Waringer and Graf (2011), nomenclature of primary setae and setal areas follows Wiggins (1998). Illustrations were prepared according to Thomson and Holzenthal (2010) in which pencil drawings made with a camera lucida are digitized, edited and digitally inked in Adobe Illustrator (v. 16.0.4, Adobe Systems Inc.).

Specimens are currently stored in the following collections: Collection Wolfram Graf (WG), Institute of Hydrobiology and Aquatic Ecology Management, University of Natural Resources, Max-Emanuelstrasse 17, A-1180 Vienna, Austria; Collection Ana Previšić (AP), Department of Biology, Faculty of Science, University of Zagreb, Rooseveltov trg 6, HR-10000 Zagreb, Croatia; Collection Mladen Kučinić (MK), Department of Biology, Faculty of Science, University of Zagreb, Rooseveltov trg 6, HR-10000 Zagreb, Croatia; Collection Mladen Kučinić (MK), Department of Biology, Faculty of Science, University of Zagreb, Rooseveltov trg 6, HR-10000 Zagreb, Croatia; Collection Mladen Kučinić (MK), Department of Biology, Faculty of Science, University of Zagreb, Rooseveltov trg 6, HR-10000 Zagreb, Croatia; Collection János Oláh [János Oláh Private Collection under national protection of the Hungarian Natural History Museum, Budapest, Hungary] (JO), Tarján u. 28, H-4032 Debrecen, Hungary.

Type specimens will be deposited in museum collections upon completion of the taxonomic work.

#### Taxonomy

#### Descriptions of the new species

*Drusus krpachi* Kučinić, Graf & Vitecek, sp. n. http://zoobank.org/74BBEB74-1232-4B4E-934B-D8BE6433DDCB Fig. 1

**Material examined. Holotype.** 1 male: Macedonia, Mavrovo National Park, Korab Mountains, česma Elem; N41.857, E 20.625; leg. Kučinić, Krpač, Mihoci; 15.VIII.2011. Currently deposited in coll. WG, will be deposited in the Croatian Natural History Museum, Zagreb, Croatia.



**Figure 1.** Male genitalia of *Drusus krpachi* sp. n. **A** left lateral view **B** paramere, dorsal view **C** caudal view **D** dorsal view **E** ventral view. Abbreviations: tVIII tergite VIII, IX segment IX, sup superior appendage, int intermediate appendage, inf inferior appendage; arrow 1 indicates spinose area of tergite VIII, arrow 2 indicates lateral protrusion of segment IX. Scale bar denotes 1 mm. Del. Vitecek.

**Paratypes.** 3 males: Macedonia, Mavrovo National Park, Korab Mountains, Reč; leg. Krpač, Mihoci, Kučinić; 01.VIII.2011. Currently deposited in coll. MK, two paratypes will be deposited in the Macedonian Museum of Natural History, Skopje, Republic of Macedonia, one paratype will be deposited in coll. WG.

Type locality. Macedonia, Korab Mountains.

**Diagnosis.** Males of the new species are most similar to *D. macedonicus*, but exhibit (1) a distally straight ventral half of segment IX; (2) a dorsally straight tip of the intermediate appendage distinctly separated by a proximal indentation and with small proximal and distal rough protrusions; (3) a conical inferior appendage with a proximal dorsal triangular protrusion; (4) parameres with three tines in the distal third in dorsal view. *Drusus macedonicus* males have a distally concave ventral half of segment IX, intermediate appendages with two rough rounded dorsad protrusions but lacking a distinct proximal indentation, distally tapering inferior appendages, and parameres with a single tine in the distal third in dorsal view.

**Description.** *Adults.* Habitus yellow; sternites and tergites fawn; cephalic and thoracic setal areas pale; cephalic and thoracic setation blond, abdominal setation scarce, blond; legs fawn; haustellum and intersegmental integument pale, whitish; wings yellow with blond setae on veins and the membrane. Male maxillary palp 3-segmented. Forewing length 11 mm, spur formula 1–3–3.

*Male genitalia* (Fig. 1). Tergite VIII (tVIII) fawn, setae absent; spinose area in lateral view approximately flat with slight dorsocaudal protrusion in cranial half, in dorsal view suboval; flanked by membranous, less sclerotized areas. Segment IX (IX) in lateral view ventrally straight distally; in caudal view dorsally approximately as wide as ventrally; with rounded lateral protrusion in the dorsal half (best seen in ventral view). Superior appendages (sup) in lateral view suboval, curved obtusely caudad in proximal third, proximally with slight dorsal protrusion, longest in anterioposterior axis: approximately 2.5 times longer than high; in dorsal view proximally slightly concave medially; medial transverse section oval. Intermediate appendages (int) in lateral view with subtriangular tip, rough areas concentrated on dorsal proximal and dorsal distal aspect; in dorsal view tips separated, oval, distally converging; in caudal view approximately triangular. Inferior appendages (inf; gonopods *sensu* Snodgrass 1935) in lateral view conical, proximally wide, distally slender, with proximal triangular protrusion dorsally; in ventral and dorsal view with small medial projection and slight notch. Parameres simple, in dorsal view with 3 tines in distal third: 2 mediolateral, 1 dorsal.

Female and pupa unknown. Larval description and indentification key provided by Vitecek et al. (in press).

**Etymology.** Named after V. Krpač, Macedonian entomologist and collector of the species.

**Distribution.** Micro-endemic of the Korab Mountains, Hellenic Western Balkans (ecoregion 6, Illies 1978) (Fig. 11).

#### Drusus malickyi Oláh & Vitecek, sp. n.

http://zoobank.org/4A7440AE-973D-4F33-858C-D53A48EEF743 Fig. 2

**Material examined. Holotype.** 1 male: Albania, Shkoder County, Shkoder District, Prokletije Mts, beech forest with brook above Okol; N42.42258, E19.76127; leg. Puskas 05.IX.2013. Currently deposited in coll. WG, will be deposited in János Oláh Private Collection under national protection of the Hungarian Natural History Museum, Budapest, Hungary (JO).

**Type locality.** Albania, Prokletije Mountains.

**Diagnosis.** The holotype of the new species is most similar to *D. macedonicus*, but exhibits (1) a sharp mediocaudal protrusion of segment IX; (2) a dorsally straight and rough tip of the intermediate appendage distinctly separated by a proximal indentation (3) a distinctly slender and constricted distal half of the inferior appendage in lateral view. *Drusus macedonicus* males have a mediocaudal and a ventrocaudal protrusion of segment IX, intermediate appendages with two rough rounded dorsal protrusions but lacking a distinct proximal indentation, and to a lesser degree constricted inferior appendages.



**Figure 2.** Male genitalia of *Drusus malickyi* sp. n. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

**Description.** *Adult, holotype.* Habitus yellow; sternites and tergites fawn; cephalic and thoracic setal areas pale; cephalic and thoracic setation blond, abdominal setation scarce, blond; legs fawn; haustellum and intersegmental integument pale, whitish; wings yellow with blond setae on veins and the membrane. Male maxillary palp 3-segmented, forewing length 10.9 mm, spur formula 1–3–3.

*Male genitalia* (Fig. 2). Tergite VIII fawn, setae scarce; spinate area in lateral view approximately flat with slight dorsad protrusion in anterior half, in dorsal view suboval; flanked by membraneous, less sclerotized areas. Segment IX in lateral view with sharp medial caudal protrusion, ventrally concave distally; in caudal view wider dorsally than ventrally; with irregular triangular, rounded lateral protrusion in dorsal half (best seen in dorsal and ventral

views). Superior appendages in lateral view suboval, curved obtusely caudad in proximal quarter, proximally with slight dorsal and distinct ventral protrusions, longest in anterioposterior axis: approximately 2.5 times longer than high; in dorsal view proximally concave medially; medial transverse section oval. Intermediate appendages in lateral view with subtriangular, dorsally rough tip; in dorsal view tips separated, wedge-shaped, approximately parallel; in caudal view approximately triangular. Inferior appendages in lateral view subtriangular, proximally somewhat bulbous, distally slender and distinctly constricted; in ventral and dorsal views with small medial protrusion and slight notch; in ventral view with longitudinal groove delimiting medial lobe. Parameres simple, in lateral view with 1 tine in distal third.

Female, pupa and fifth instar larva unknown.

Etymology. Named after Hans Malicky, trichopterologist.

**Distribution.** Micro-endemic of the Prokletije Mountains, Hellenic Western Balkans (ecoregion 6) (Fig. 11).

#### Re-descriptions of male filtering carnivore Drusinae sensu Pauls et al. (2008)

#### Cryptothrix nebulicola McLachlan, 1867

Fig. 3

**Material examined.** 1 male: Italy, Torino, Traversella, Fondo, Burdeivier brook; leg. Vincon; 12.VII.2012. 12 males: Italy, San Marco Pass; leg. Graf; 14.VIII.2000; in coll. WG.

Type locality. Switzerland, Canton of Valais, Maienwang (Grimselpass).

**Description.** *Adults.* Habitus dark; sternites and tergites brown; cephalic and thoracic setal areas pale; cephalic, thoracic and abdominal setation blond; legs light brown to fawn, proximally darker; haustellum and intersegmental integument pale, whitish; wings dark, with dark setae. Male maxillary palp 3-segmented; forewing length 8–10 mm; spur formula 1–2–2.

*Male genitalia* (Fig. 3). Tergite VIII brown, with lighter areas around alveoli; setation abundant; spinose area approximately rectangular in dorsal view; flanked by membraneous, less sclerotized areas. Segment IX in lateral view ventrally slightly concave distally; in caudal view wider dorsally than ventrally; with long, round, wedgeshaped protrusion in dorsal half (best seen in ventral view). Superior appendages in lateral view suboval, curved obtusely caudad in proximal fifth, proximally dorsal somewhat protuberant, tips slightly curved dorsad, longest in anterioposterior axis: approximately 2 times longer than high; in dorsal view medially concave, tips converging; medial transverse section lateroventrally curviconvex suboval. Intermediate appendages in lateral, dorsal and caudal views dorsally with 2 distinct tips, the proximal tip rounded, rough, the distal tip pointed, smooth; in caudal view approximately an isoceles trapezium. Inferior appendages in lateral view roughly triangular, proximally constricted, ventrocaudally slightly concave; in dorsal and ventral views tips converging; in ventral view with longitudinal groove delimiting medial lobe. Parameres simple, rodlike, medially and distally somewhat bulbous.



**Figure 3.** Male genitalia of *Cryptothrix nebulicola*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

Female depicted by Schmid (1956), Malicky (2004); larva in key presented by Waringer and Graf (2011), Vitecek et al. (in press); pupa unknown.

Distribution. Regionally in the Western Alps (ecoregion 4) (Fig. 11).

# Drusus chrysotus Rambur, 1842

Fig. 4

**Material examined.** 12 males: Austria, Krumbach, Soboth; N46.723, E15.0555; leg. Graf; 20.V.2004; in coll. WG.

Type locality. France, Rhône-Alpes, Haute-Savoie, Chamonix valley.



**Figure 4.** Male genitalia of *Drusus chrysotus*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

**Description.** *Adults.* Habitus: light brown to yellow; sternites and tergites light brown, abdominal tergite VII with distinct saddle; cephalic and thoracic setal areas pale; cephalic and thoracic setation blond, abdominal setation scarce, short, dark; legs fawn, proximally darker; haustellum and intersegmental integument pale, whitish; wings light brown to yellow with dark setae on veins and blond setae on membrane. Male maxillary palp 3-segmented; forewing length 14–16 mm; spur formula 1–3–3.

*Male genitalia* (Fig. 4). Tergite VIII light brown, with short, pale, translucent setae; spinose area in lateral view with distinct dorsal protrusion and dorsomedial caudal protrusion, in dorsal and caudal views tripartite; flanked by membraneous, less sclerotized areas. Segment IX in lateral view ventrally irregular concave distally; dorsally approximately as wide as ventrally in caudal view; with distinct approximately subtriangular, rounded protrusion in dorsal half (best seen in dorsal and ventral views). Superior appendages in lateral view curved obtusely ventrocaudad in proximal third, proximally with distinct dorsocranial protrusion, approximately as long as high, in dorsal view proximally concave medially; medial transverse section oval. Intermediate appendages in lateral view medially protruding caudad, dorsally with long, rough tip; in dorsal view fused into approximately heart-shaped, centrally indented structure; in caudal view ventrally broad with bulbous lateral protrusions, dorsally narrow, fused. Inferior appendages in lateral view conical, short; in ventral and dorsal views blunt, with blunt, short medial protrusion and slight notch; in ventral view with longitudinal groove delimiting medial lobe. Parameres simple with several tines on common base in distal third.

Female depicted by Schmid (1956), Malicky (2004); larva in key presented by Waringer and Graf (2011), Vitecek et al. (in press); pupa described in Bohle (1987).

**Distribution.** This species is widely distributed, occuring in and around the Alpine arc (ecoregion 4), the Western and Central Highlands (ecoregions 8 & 9) and was also found in the northern part of the Dinaric Alps (ecoregion 5) (Fig. 11).

#### Drusus discolor Rambur, 1842

Fig. 5

Material examined. 3 males: France, Mt. Canigou; N42.4864, E2.4139; leg. Graf; 12.VII.2012; in coll. WG. 2 males: France, St. Pierre de la Martin; N42.9597, E0.8290; leg. Graf; 22.VII.2012; in coll. WG. 7 males: Austria, Gurkursprung; leg. Wieser; 13.VII.1997; in coll. WG. 22 males: Switzerland, Val Munstair; N46.5852, E10.4544; leg. Graf; 20.VII.2006; in coll. WG. 1 male: Montenegro, Brodavac, right tributary of Peručica; N42.6859, E19.7364; leg. A. Previšić; 10.VII.2013; in coll. AP. Type locality. France. Phône Alper. Haute Savaie. Chamonix valley.

**Type locality.** France, Rhône-Alpes, Haute-Savoie, Chamonix valley.

**Description.** *Adults.* Habitus fawn to brown; sternites and tergites fawn to brown; cephalic and thoracic setal areas pale; cephalic, thoracic and abdominal setation blond; legs fawn, proximally darker; haustellum and intersegmental integument pale, whitish; wings blond-brown, with blond-brown setae on veins and blond setae on membrane. Male maxillary palp 3-segmented; forewing length 12–15 mm, spur formula 1–3–3.

*Male genitalia* (Fig. 5). Tergite VIII light brown, setation scarce, in lateral view with distinct cranial dorsal protuberance; spinose area in lateral view with distinct dorsal protrusion and dorsomedial caudal protrusion, in dorsal view suboval, caudally straight; flanked by membraneous, less sclerotized areas. Segment IX in lateral view ventrally distinctly concave distally; in caudal view dorsally approximately as wide as ventrally; with a distinct, caudally straight rounded protrusion indorsal half (best seen in ventral view). Superior appendages in lateral view suboval, curved obtusely caudad in proximal half, proximal



**Figure 5.** Male genitalia of *Drusus discolor*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

half with distinct dorsal protrusion, approximately as long as high; in dorsal view medially concave; medial transverse section suboval. Intermediate appendages in lateral view medially approximately straight, dorsally with rounded, rough tip; in dorsal view tips separate, oval, distally converging; in caudal view approximately triangular with dorsally diverging tips. Inferior appendages in lateral view conical; in ventral and dorsal views with distinct medial protrusion and distinct notch; in ventral view with longitudinal groove delimiting medial lobe. Parameres simple with single bulbously based tine in distal third.

Female depicted by Schmid (1956), Malicky (2004); larva in key presented by Waringer and Graf (2011), Vitecek et al. (in press); pupa unknown.

**Distribution.** This species is one of the most widespread Drusinae species, covering all major European mountain ranges from the Carpathians to the Pyrenees (ecoregions 1–10) (Fig. 11).

#### Drusus macedonicus Schmid, 1956

Fig. 6

**Material examined.** 1 male: Macedonia, Jablanica Mt., Labunište; N41.271841, E20.558136; leg. Kučinić and Krpač; 19.IX.2013; in coll. MK. 1 male: Macedonia, Pelister Mt., springs of Caparska reka; N41.003889, E21.167944; leg. Graf and Previšić; 07.VII.2010; in coll. WG.

Type locality. Macedonia, Pelister Mountains.

**Description.** *Adults.* Habitus yellow; sternites and tergites fawn; cephalic and thoracic setal areas pale; cephalic and thoracic setation blond, abdominal setation scarce, blond; legs fawn; haustellum and intersegmental integument pale, whitish; wings yellow with blond setae on veins and membrane. Male maxillary palp 3-segmented; forewing length 10–12 mm; spur formula 1–3–3.

*Male genitalia* (Fig. 6). Tergite VIII fawn, setation lateral, scarce; spinose area in lateral view approximately flat, in dorsal view suboval, tapering cranially; flanked by membraneous, less sclerotized areas. Segment IX in lateral view ventrally deeply concave distally, with distinct medial and ventral caudad protrusion; in caudal view slightly wider dorsally than ventrally; with sharp, caudally approximately straight protrusion in dorsal half (best seen in dorsal and ventral views). Superior appendages in lateral view irregularly suboval, curved obtusely caudad in proximal quarter, proximally with an irregular dorsal and irregular ventral protuberance, longest in anterioposterior axis: approximately 2.5 times longer than high; in dorsal view proximally slightly concave medially; medial transverse section suboval. Intermediate appendages in lateral view with two rough tips: 1 curved dorso-posteriorly, 1 central, rounded; in dorsal view posterior tips adjacent, parallel; in caudal view subtriangular with slender lateral projections. Inferior appendages in lateral view swith medial tip and notch, separated by slight notch; in ventral view with longitudinal groove delimiting medial lobe. Parameres simple, with single dorsal tine in distal third.

Female depicted by Schmid (1956), Malicky (2004); larva in key presented by Vitecek et al. (in press); pupa unknown.

**Distribution.** Micro-endemic of the Pelister and Jablanica Mountains, Hellenic Western Balkans (ecoregion 6) (Fig. 11).

#### Drusus meridionalis Kumanski, 1973

Fig. 7

**Material examined.** 10 males: Bulgaria, Vihren, Pirin Mountains, Okotovo-Banserishka, marshy spring; N41.7389, E23.4462; leg. Keresztes, Török, Kolcsár; 23.VIII.2013; in coll WG.

Type locality. Bulgaria, Rila and Pirin Mountains.

**Description.** *Adults.* Habitus yellow to brown; sternites and tergites yellow to brown; cephalic and thoracic setal areas pale; cephlic and thoracic setation blond, ab-



**Figure 6.** Male genitalia of *Drusus macedonicus*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

dominal setation scarce, short, dark; legs yellow to light brown, proximally darker; haustellum and intersegmental integument pale, whitish; wings yellow to fawn, with blond setae on veins and membrane. Male maxillary palp 3-segmented. Forewing length 12–14 mm; spur formula 1–3–3.

*Male genitalia* (Fig. 7). Tergite VIII yellow to brown, setae absent; spinose area in lateral view approximately flat, in dorsal view suboval, somewhat rectangular cranially; flanked by membraneous, less sclerotized area bearing single seta. Segment IX in lateral view ventrally slightly concave distally; in caudal view wider ventrally than dorsally; with distinct, approximately triangular, rounded protrusion in dorsal half (best seen in dorsal view). Superior appendages in lateral view suboval, curved obtusely caudad in proximal third, proximally with distinct dorsal protrusion, longest in anterioposterior axis: approximately 3 times longer



**Figure 7.** Male genitalia of *Drusus meridionalis*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

than high; in dorsal view proximally slightly concave medially; medial transverse section oval. Intermediate appendages in lateral view with rounded, rough tip; in dorsal view 2 separate parallel tips, each oval, rough; in caudal view subtriangular, dorsally with 2 separate tips. Inferior appendages in lateral view conical; in ventral and dorsal views slender with minute subtriangular medial protrusion and shallow notch; in ventral view with longitudinal groove delimiting medial lobe. Parameres simple, with single, bulbously based tine in distal third.

Female depicted by Kumanski (1973), Malicky (2004); larva in key presented by Vitecek et al. (in press); pupa unknown.

Distribution. Regionally in the Eastern Balkans (ecoregion 7) (Fig. 11).

**Comments.** This species was first described as a subspecies of *D. romanicus*, but was elevated to species rank (Vitecek et al. in press). It is morphologically distinct from *D. romanicus*, disjunct in distribution, and was recovered as a well-separated clade from *D. romanicus* in the phylogenetic analyses of Pauls et al. (2009) and Vitecek et al. (in press).

#### Drusus muelleri McLachlan, 1868

Fig. 8

**Material examined.** 1 male: Switzerland, Furkapass; N46.5888, E8.4327; leg. Graf; 21.VII.2006, in coll. WG.

Type locality. Switzerland, Canton of Uri, Hospental.

**Description.** *Adults.* Habitus dark; sternites and tergites brown; cephalic and thoracic setal areas pale, cephalic and thoracic setation blond, abdominal setation scarce, short, dark; coxa, trochanter, femur brown, tibia and tarsi fawn; haustellum and intersegmental integument pale, whitish; wings brown, smoky, with dark setae on veins and blond setae on membrane. Male maxillary palp 3-segmented; Forewing length 11–13 mm (Malicky 2004); spur formula 1–3–3.

*Male genitalia* (Fig. 8). Tergite VIII brown, setae absent; spinose area in lateral view convex with caudal ventral lobe, in dorsal view suboval with small medial protrusion; flanked by membraneous, less sclerotized areas. Segment IX in lateral view ventrally slightly concave distally; in caudal view wider dorsally than ventrally; with sharp caudally straight subtriangular protrusion in the dorsal half (best seen in dorsal and ventral views). Superior appendages in lateral view irregular, curved obtusely caudad in proximal quarter, proximally distinctly dilated, distally clavate, longest in anterioposterior axis: approximately 5 times longer than high; in dorsal view the proximal third concave medially; medial transverse section circular. Intermediate appendages in lateral view with long, rounded rough tip; in dorsal view tips separate, approximately parallel, proximally bulbous; in caudal view subtriangular. Inferior appendages in lateral view subtriangular, ventrally irregular, proximally sightly concave dorsally; in ventral and dorsal views broad, with small subtriangular medial protrusion and distinct notch; in ventral view with longitudinal groove delimiting medial lobe; in caudal view broad. Parameres simple, with single dorsal tine in distal third.

Female depicted by Schmid (1956), Malicky (2004), larva in key presented by Waringer and Graf (2011), Vitecek et al. (in press); pupa unknown.

Distribution. Regionally in the Western Alps (ecoregion 4) (Fig. 11).

# Drusus romanicus Murgoci and Botosaneanu, 1953

Fig. 9

Material examined. 1 male: Romania, Apuseni Mts., Garda de Sus, tributary of Ariesul Mare; N46.4508, E22.7982; leg. Oláh, Bajka, Balogh, Borics; 29.V.2013; in coll.



**Figure 8.** Male genitalia of *Drusus muelleri*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

WG. 1 male: Romania, Apuseni Mts., Muntii Giaului, Stiunea Muntele Baisorii, Lupinus stream; leg. Oláh, Balogh, Fekete; 18.VI.2013; in coll. WG. 1 male: Romania, Retezat Mts, Bucara Stream, 150 m below Bucara lake; N45.3570, E22.8753; leg. Bajka, Balogh, Borics, Borics; 10.VIII.2013; in coll. WG.

Type locality. Romania, Carpathian Mountains, spring areas of the Ialomita stream.

**Description.** *Adults.* Habitus brown to light brown; sternites and tergites brown to light brown; cephalic and thoracic setal areas pale; cephalic, thoracic and abdominal

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**Figure 9.** Male genitalia of *Drusus romanicus*. **A** left lateral view **B** paramere, lateral view **C** caudal view **D** dorsal view **E** ventral view. Scale bar denotes 1 mm. Del. Vitecek.

setation blond; legs light brown, proximally darker; haustellum and intersegmental integument pale, whitish; wings brown, proximally lighter, with blond setae on veins and membrane. Male maxillary palp 3-segmented; forewing length 12–14 mm; spur formula 1–3–3.

*Male genitalia* (Fig. 9). Tergite VIII brown, setae present; spinose area in lateral view approximately flat with slight dorsal protrusion, in dorsal view suboval, distally straight; flanked by membraneous, less sclerotized areas. Segment IX in lateral view dorsally with distinct notch distally, ventrally irregularly concave distally; in caudal

view ventrally wider than dorsally; with distinct subtriangular rounded protrusion in dorsal half (best seen in dorsal view). Superior appendages in lateral view elongate suboval, curved obtusely dorsocaudad in proximal quarter, proximally with round dorsal protrusion and irregular ventral protrusion, longest in anterioposterior axis: approximately 4.5 times longer than high; in dorsal view proximally distinctly concave medially; medial transverse section circular. Intermediate appendages in lateral view with rounded, rough tip; in dorsal view tips separate, laterally diverging; in caudal view subtriangular. Inferior appendages in lateral view conical, long, dorsally irregular, proximally slightly concave dorsally; in ventral and dorsal views proximal half robust, distal half slender with slight medial protrusion and shallow notch. Parameres simple, with medial hook-shaped tip bearing several smaller tines.

Female and pupa unknown; larva in key presented by Vitecek et al. (in press).

**Distribution.** Regionally in the Western and Southern Carpathians (ecoregion 10) (Fig. 11).

#### Drusus siveci Malicky, 1981

Fig. 10

Material examined. 5 males: Bosnia and Herzegovina, Sutjeska National Park, stream close to Čermerno; N43.2650, E18.5927; leg. Previšić, Miliša; 04.VII.2012; in coll. AP. Type locality. Montenegro, Andrijevica, Gnjili Potok.

**Description.** *Adults.* Habitus yellow to fawn; sternites and tergites fawn; cephalic and thoracic setal areas pale; cephalic, thoracic and abdominal setation blond; legs yellow to fawn; haustellum and intersegmental integument pale, whitish; wings fawn, with blond to brown setae on veins and blond setae on membrane. Male maxillary palp 3-segmented, forewing length 10–12 mm, spur formula 1–3–3.

*Male genitalia* (Fig. 10). Tergite VIII fawn, setation concentrated dorsally and posterolaterally, with slight dorsal protrusion; spinose area in lateral view approximately flat, in dorsal view oval; flanked by membraneous, less sclerotized areas. Segment IX in lateral view with medial caudad protrusion, ventrally irregularly concave distally; in caudal view wider dorsally than ventrally; with distinct rounded triangular protrusion in dorsal half (best seen in dorsal and ventral views). Superior appendages in lateral view suboval, curved obtusely caudad in proximal half, proximal half with distinct rounded protrusion, in dorsal view slightly concave medially; medial transverse section subcircular. Intermediate appendages in lateral view with pointed, hook-like tip arching dorsad; in dorsal and caudal views the tips fused; in caudal view subtriangular. Inferior appendages in lateral view conical, short, blunt, posteroventrally somewhat concave; in ventral view with medial protrusion and distinct notch. Parameres simple, with single bulbously based tine in distal third.

Female and pupa unknown; larva in key presented by Vitecek et al. (in press).

Distribution. Micro-endemic of the Dinaric Western Balkans (ecoregion 5) (Fig. 11).



Figure 10. Male genitalia of *Drusus siveci*. A left lateral view B paramere, lateral view C caudal view D dorsal view E ventral view. Scale bar denotes 1 mm. Del. Vitecek.

# Discussion

# Drusinae micro-endemics of the Western Balkans

Morphology of the new species as well as molecular phylogenetic analyses (Vitecek et al. in press) suggest that the new species belong to the monophyletic clade of filtering carnivorous Drusinae *sensu* Pauls et al. (2008), comprising *Drusus discolor*, *D. muelleri*, *D. chrysotus*, *D. siveci*, *D. romanicus*, *D. meridionalis* and *Cryptothrix nebulicola*. The *Drusus* species described here are similar to *D. macedonicus* Schmid. However, they differ distinctly in the morphology of the male genitalia, particularly the intermediate



**Figure 11.** Distribution of filtering carnivore Drusinae. Single records of endemic species are depicted as symbols, stroked or filled areas denote ranges of more widely distributed species with a higher number of occurrence records.

appendages, and are discretely distributed. Also, they are highly supported in phylogenetic analysis and form a highly supported clade comprising (D. malickyi + (D. krpachi + D. macedonicus) in the phylogenetic analysis of Vitecek et al. (in press). To our knowledge, the new species are small-scale endemics restricted to single mountain ranges.

Interestingly, the type localities of the new species are close to the known range of D. macedonicus (Fig. 11). Such small-scale distribution of distinct Drusinae species is well documented from the Western Balkans (Marinković-Gospodnetić 1976; Kučinić et al. 2011a; Oláh 2010, 2011; Oláh and Kovács 2013; Previšić et al. 2014a, b; Vitecek et al. 2015). Similarly, other taxa exhibit comparable distribution patterns, in which single mountain ranges represent the range of a species, or haplogroups (Ursenbacher et al. 2008, Stevanović et al. 2009, Zogaris et al. 2009, Karaman et al. 2011). The intriguing distribution patterns exhibited by some groups of species potentially result from the geological history of the region and historic and present-day climate conditions. Small-scale speciation of Drusinae presumably is facilitated by intrinsic traits of the subfamily, such as their occurrence at higher elevations (Pauls et al. 2006, 2009), a putatively low long-distance dispersal potential (Müller-Peddinghaus 2011, Geismar et al. 2015), and might be further enhanced by habitat fragmentation, e.g., by regional karstification. Occurrence of Drusinae could therefore serve as proxy to occurrence of other aquatic invertebrate taxa, particularly to crenobiont taxa exhibiting the same or similar traits.

### Western Balkan aquatic diversity

The Western Balkans represent a hot-spot of species richness and endemicity in Europe (Griffiths et al. 2004, Guéorguiev 2007, Kenyeres et al. 2009, Jaskuła 2011). In particular, the faunas of isolated habitats such as coldwater springs and streams, caves or the profundal zone of large lakes contribute to high species richness in the region (Petkovski et al. 2009, Wilke et al. 2010, Pešić and Glöer 2013). Such taxa probably are more susceptible to factors promoting speciation, such as climatic and geological processes (e.g., karstification, see Previšić et al. 2009, 2014b), especially if their dispersal potential is low.

The description of the two new micro-endemic *Drusus* species increases the number of Western Balkan Drusinae species. Drusinae richness in the Western Balkans currently comprises 40 species including 13 species (30 %) that were discovered since 2010, of which 32 are endemic to the Western Balkans (Graf et al. 2008; Oláh 2010, 2011; Schmidt-Kloiber and Hering 2012; Oláh and Kovács 2013; Previšić et al. 2014b; Vitecek et al. 2015; Ibrahimi et al. pers. comm.; this study).

Thus, endemism rates of Western Balkan Drusinae are high, and are further augmented by the description of the two new micro-endemic *Drusus* species. Global and anthropogenic habitat changes are among the greatest threats to micro-endemic and endemic freshwater species (Hering et al. 2009, Tierno de Figueroa et al. 2010, Bálint et al. 2011, Conti et al. 2014). Water extraction for human consumption intensified by tourism, agriculture, and hydroelectricity are the primary modes of global anthropogenic habitat modification of freshwaters (Foster 1991, Polhemus 1993, Dudgeon 2006). Hydropower plants were identified as the greatest threat to European freshwater biodiversity (Freyhof 2012, Schwarz 2012, Zarfl et al. 2014, http://riverwatch.eu).

Recent published taxonomic works treating the Western Balkans, including the present one describing two new micro-endemic *Drusus* species, have demonstrated the significance of the region for European biodiversity. However, progressing socioeconomic change and anthropogenic habitat modification threaten the freshwater biodiversity of the Western Balkans, and potentially will result in the loss of yet-to-be discovered species.

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RESEARCH ARTICLE



# Four new species of Cymatodera Gray from central and southern Mexico (Coleoptera, Cleridae, Tillinae)

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# Abstract

Four new species of *Cymatodera* are described from Mexico: *C. tortuosa* Burke & Rifkind, **sp. n.** from Hidalgo and Tamaulipas; *C. ortegae* Burke, **sp. n.** from Colima, Jalisco and Michoacan; *C. gerstmeieri* Burke & Rifkind, **sp. n.** from Chiapas; and *C. mixteca* Burke & Rifkind, **sp. n.** from Puebla and Guerrero. Male genitalia and other characters of taxonomic value are illustrated.

# Keywords

Cleridae, Cymatodera, Mexico, brachyptery, Chiapas, Nevado de Jalisco, Sierra de Manantlan, Tehuacan

# Introduction

As part of an ongoing effort to catalog the diversity of Mexican Cleridae, the present work describes four new species of *Cymatodera* Gray from the central and southern states of Mexico. As previously discussed (Rifkind 2014, 2015), the diversity of the clerid genus *Cymatodera* in Mexico is extensive, but our knowledge of the group remains rudimentary. Many dozens of species await description and many more are likely to be discovered, particularly in areas distant from paved roads. Recent descriptions of species belonging to this genus include many endemics (Burke 2013; Burke and Zolnerowich 2014; Rifkind et al. 2010; Rifkind 2014, 2015), and here again, it is quite likely that the tally will increase as collecting efforts reach further into habitats

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such as relictual cloud forest and isolated mountain ranges that are known centers of endemism. Much attention has been focused on the rapid destruction or degradation of natural habitat in Mexico and other parts of Latin America, and the insufficiency of current resources dedicated to cataloguing disappearing biodiversity (Blackman et al. 2014; Armesto et al. 2007; Santibañez and Santibañez 2007; Trejo and Dirzo 2000). One positive trend is a recent increase in the number of trained taxonomists from these countries. Several workers in Mexico, for example, have undertaken faunistic and systematic studies of Cleridae with the result that the pace of description there is now higher than it has been in nearly a century (Burke 2013; Burke and Zolnerowich 2014; Rifkind et al. 2010, Toledo-Hernández et al. 2015). Should this trend continue, there may be more hope than previously thought for the assessment and conservation of critically threatened habitats in Mexico.

#### Material and methods

Genitalia extraction and dissection procedures are similar to those outlined by Ekis (1977). Most of the morphological terminology used follows the works of Ekis (1977), Rifkind (1996) and Opitz (2010). Morphology of the male genitalia and pygidia are considered of primary importance as characters for the determination of new species in this descriptive work. Specimens were observed using a Leica MZ 7.5 stereomicroscope. Images were taken and measured using a Leica DFC 500 digital camera, and stacked using the software Zerene Stacker V. 1.04.

The following abbreviations are used in the description of the holotypes: TL = Total body length, HW = Maximum head width, HL = Head length, PW = Maximum pronotal width, PL = Pronotal length, EW = Maximum elytral width, EL = Elytral length.

Type material is deposited in the following collections:

CASC	California Academy of Sciences Collection, San Francisco, California, USA
CIUM	Colección de Insectos de la Universidad Autónoma del Estado de Morelos,
	Centro de Investigación en Biodiversidad y Conservación, UAEM, Mexico.
CNIN	Colección Nacional de Insectos, Instituto de Biología, UNAM, DF, Mexico
EMEC	Essig Museum of Entomology, University of California, Berkeley, USA
FMNH	Field Museum of Natural History, Chicago, Illinois, USA
JEWC	James E. Wappes Collection, San Antonio, TX, USA
KSUC	Kansas State University Museum of Entomological and Prairie Arthropod
	Research Collection, Kansas State University, Manhattan, KS, USA
JNRC	Jacques Rifkind Collection, Valley Village, CA, USA
RHTC	Robert H. Turnbow Jr. Collection, Enterprise, AL, USA
SEMC	University of Kansas, Snow Entomological Museum, Lawrence, KS, USA
TAMU	Texas A&M Insect Collection, Texas A&M University, College Station,

- TX, USA
- WFBM William F. Barr Entomological Museum, University of Idaho, Moscow, ID, USA

# Taxonomy

# Cymatodera tortuosa Burke & Rifkind, sp. n.

http://zoobank.org/00F91AE7-8F5D-4A03-B70B-DD1B2BF851A0 Figs 1, 6, 11, 16, 20, 23

**Type material** (n = 2). Holotype, red labeled, male: Mexico, Hidalgo, La Florida, municipio de Cardonal, Sitio 1A, 4-V-2014, S. Quiñonez; holotype deposited in CNIN. Paratype: 1 female: Mexico, Tamaulipas Mpio. Tula, La Presita, Canon de Coyote, 1,900 m, 16-III-1987, P. Kovarik, R. Jones, R. Trevino; paratype deposited in TAMU.

**Differential diagnosis.** The new species can be separated from congeners by its unique combination of body form, antennal shape, size, color, and elytral fascia pattern. *Cymatodera tortuosa* superficially resembles a number of species that share a similar pattern of fuscous and testaceous elytral banding, such as *C. balteata* LeConte, *C. sirpata* Horn, *C. undulata* (Say), and *C. wolcotti* Barr. *Cymatodera tortuosa*, however, can be readily differentiated from those species based on clear differences in the male and female pygidium as well as discontinuity in geographic distribution. Specifically, the new species has the male pygidium distinctly modified (Fig. 11) compared to similar species; the female pygidium is moderately, V-shaped emarginate (Fig. 16), rather than broadly rounded posteriorly, as observed in similar species is restricted to that region, whereas those listed previously are distributed for the most part in the south and southwestern United States, with only *C. balteata* ranging into the Mexican border states of Nuevo Leon and Tamaulipas.

**Description.** Holotype male. Form elongate, slender; metathoracic wings present and fully developed. TL = 11.9 mm. Color: Head, pronotum and thorax piceous; elytra slightly lighter; antennae and mouthparts brunneous; legs testaceous; abdomen brunneous mesally, becoming pale testaceous laterally. Elytra with a pattern of testaceous markings and maculae as follows: the anterior 1/2 with a roughly X-shaped marking; each elytron at posterior 1/4 with a large, irregularly bordered, ovate macula narrowly separated at the elytral suture (Fig. 1).

Head: HL = 2.15 mm, HW = 1.95 mm. Measured across eyes wider than pronotum; surface smooth, moderately shiny, moderately, finely punctate; frons bi-impressed; sparsely clothed with short, very fine, recumbent setae, more profusely vested behind the eyes with longer setae; eyes moderately small, subsinuate, taller than wide, moderately emarginate in front, moderately protuberant laterally, separated by approximately 2.5 eye-widths. Antennae slender; loosely composed; extending beyond posterior margin of elytra; third antennomere about two times the length of second antennomere, fourth antennomere slightly longer than third antennomere; antennomeres 5–10 subequal in length; antennomere 4–10 weakly serrate, serration very gradually increasing distally; last antennomere flattened apically, as long as tenth antennomere (Fig. 6).

Thorax: PL = 2.75 mm, PW = 1.9 mm. Pronotum elongate; widest at middle; middle slightly broader than anterior margin; sides constricted laterally; more strongly



Figures 1–5. Habitus of: I *Cymatodera tortuosa* sp. n. (holotype male) 2 *Cymatodera ortegae* sp. n. (paratype male) 3 *Cymatodera gerstmeieri* sp. n. (holotype male) 4 *Cymatodera mixteca* sp. n. (holotype male)
5 *Cymatodera pallida* (male).

constricted behind middle; disc flat; moderately impressed in front of middle; subbasal tumescence feebly pronounced; surface rugulose; moderately, finely punctate; vested with short, fine, pale recumbent setae intermingled with less numerous, long erect setae, the latter more numerous laterally. Prosternum wider than long, rugulose,
moderately puncticulate, scarcely vested with fine, pale, semirecumbent setae. Mesosternum smooth, shiny, feebly, coarsely, deeply punctate. Metasternum convex; rugose; moderately, shallowly punctate; moderately clothed with pale, semirecumbent setae, vestiture more abundant medially.

Legs: Femora clothed with short, recumbent setae interspersed with a few erect and semierect setae; tibiae moderately vested with some short and long erect and semierect setae; femora and tibiae transversely rugose.

Elytra: EL = 8.8 mm, EW = 2.6 mm. Anterior margin arcuately emarginate; wider than widest portion of pronotum; humeri pronounced; sides subparallel; widest on posterior fourth; disc convex; apex moderately dehiscent; elytral sculpturing as follows: anterior third set with regular, rather coarse and deep striae that abruptly diminish after anterior third and disappear entirely on posterior half; punctations at elytral base coarse and deep; surface moderately clothed with short, very fine, pale, recumbent setae intermixed with very few fine, long, erect setae.

Abdomen: Ventrites 1-5 rugulose, strongly convex; shallowly, moderately punctate; subequal in length; each ventrite with a pair of large, pale, shallow impressions near sides; surface clothed with fine, pale, moderately long, recumbent setae. Fifth ventrite (Fig. 11) moderately smooth; sides moderately oblique and arcuate; posterior margin broadly, deeply emarginate, emargination reaches medial portion of segment, posterolateral angles acuminate; sixth ventrite (Fig. 11) longer than wide, surface rugulose, with a pair of oblique, V-shaped, longitudinal carinae that initiate on anterior 1/4 and end slightly beyond segmental mid-length; highly modified distally, posterior margin deeply emarginate, emargination U-shaped, with the posterolateral angles each produced as a dorsally recurved and conspicuously elongate extension, the last third of each posterolateral extension bearing an internal spine or acuminate protuberance. Fifth tergite (Fig. 20) moderately convex; rugulose; lateral margins subparallel; posterior margin bisinuate, broadly, moderately deeply, triangularly emarginate at middle. Sixth tergite (Fig. 20) conspicuously longer than wide; subtriangular; surface strongly convex; rugulose; moderately, shallowly punctate; lateral margin oblique; posterior margin narrowly, moderately rounded, much surpassed by posterolateral extensions of the sixth ventrite.

Aedeagus 2.8 mm long; moderately robust; ratio of length of paramere to whole tegmen 0.45:1; tegmen fully covering phallus; parameres robust throughout their length; lateral margins feebly obtuse, subparallel, pointed distally; phallobase wide; phallic plate armed with a row of denticles along dorsal margin; phallobasic apodeme robust distally, moderately short; phallobasic struts rather slender throughout their length (Fig. 23).

Female: The female paratype is somewhat longer than the male, having a total body length of 13.8 mm. The female also differs from the male holotype by having the sixth ventrite strongly, deeply, V-shaped emarginate, lacking the pair of elongate, posterolateral extensions observed in the male (Figs 11, 20). Additionally, the female has a slightly paler integument than the male, with a less distinct elytral pattern anteriorly by comparison.



Figures 6–10. Antennae of: 6 Cymatodera tortuosa 7 Cymatodera ortegae 8 Cymatodera gerstmeieri 9 Cymatodera mixteca 10 Cymatodera pallida.

**Distribution.** Presently known only from the northern portion of Hidalgo and the southeastern part of Tamaulipas, Mexico. The vegetation at the collecting localities is predominantly pine-oak forest (Fig. 29).

**Etymology.** The specific epithet *tortuosa* (from the Latin *tortilis* or *tortuosus*, meaning twisted or winding), is a reference to the intricate and elaborate structure of the male pygidium of this species.

*Cymatodera ortegae* Burke, sp. n. http://zoobank.org/27646149-9E6B-4397-ACD2-DB7E55F37A7C Figs 2, 7, 12, 17, 21, 22, 24

**Type material** (n = 30). Holotype red labeled, male: Jalisco, road to microondas Los Mazos, Sierra Manantlán, 1425-1610 m, 19°42'N, 104°24'W, 12 km SSD Autlan, mixed hardwood forest 15-VII-1993, R. L. Westcott; holotype deposited in CASC. Paratypes yellow labeled: 1 male: same data as holotype (WFBM); 1 female: Mexico:

Jalisco, 81 km E of El Grullo, 6-X-1992, R. Turnbow (RHTC); 2 males, 1 female: Mex: Jalisco, N slope Nevado de Colima, 8000', 17-VII-1990, J. Wappes (JEWC); 1 male, Jalisco, Autlán, Res. de la Biosfera Manantlán, Est. Cientifica Las Jovas, 19°35'443"N, 104°16'468"W, 30-VIII-2001, Col. V. H. Toledo (CIUM); 1 male, 2 females: Mexico, Jalisco, 2 km S La Manzanilla, 12-X-2001, F. Hovore (JNRC); 1 female: Mexico, Jalisco, Nevado de Colima, 8200', Parque Nacional, 10.7 mi N Hwy 54, 17-IX-1986, [no collector data] (KSUC); 1 male: Mexico, Jalisco, 24.8 km SW Ciudad Guzman, 2286 m, 2-VII-1988, R. S. Anderson, pine-oak forest (JNRC); 1 male: Mexico: Sierra de Manantlan, Jalisco, Las Joyas, 1870 m, 18-VII-1985, J. Doyen, black and white light (EMEC); 1 male, 1 female: Mexico, Sierra de Manantlan, Jalisco, 1800-1900 m, 17-VII-1985, J. Doven (EMEC); 4 females: Mexico, Jalisco, 5.4 km NE de Apango, 19 48 N, 103 41 W, 20-X-1996, beating dead leaf clumps of Quercus sp., R. L. Westcott (WFBM); 1 female: Mexico, Jalisco, Manantlan, Lab. Nat. Las Joyas, 8-VII-1988, F. A. Noguera and Y. A. Rodriguez (CNIC); 1 female: Mexico, Jalisco, 19 km E El Jazmin, (SW Ciudad Guzman), 2005 m, 19-VII-1993, pine-oak forest, R. L. Westcott, collected on Quercus sp. (WFBM); 2 males, 2 females: Mexico, Jalisco, km 3.5-4 Nevado de Colima, 24-VII-2011, R. Turnbow (RHTC); 1 female: Mex: Colima, nr El Terrero, 7800', 18-VII-199, J. E. Wappes (JEWC); 1 male: Mexico, on elderberry stems, lot 72-11927, 27-VII-1972, Racine and Turk (WFBM); 1 male: Mexico, Colima, NW slope Nevado de Colima, 17-VII-1990, E. Giesbert (JNRC); 1 female: Colima, W rd. to El Terrero, 5000', 3-5-X-1992, J. E. Wappes (JEWC); 1 male: Mexico, Michoacan, 2 km N Tancitaro, 2700-800 m, 26-I-1947, 53, T. H. Hubbell (JNRC).

**Differential diagnosis.** The undulate fascia pattern on the elytral ground, the testaceous to slightly greenish integumental color, general body shape, and geographic distribution of the new species will, in combination, serve to separate it from other species of *Cymatodera. Cymatodera ortegae* appears to be allied to several Mexican congeners that share similar body shape, integumental color, brachypterous condition, and a reduced anterior elytral margin. Of these, *C. barri* Rifkind, *C. maculifera* Barr, and *C. monticola* Rifkind are most similar. Unlike *C. ortegae*, however, the males of *C. barri* and *C. maculifera* possess a distinct pair of feebly to moderately developed tubercles on the median posterior portion of the metasternum. Both sexes of these species lack the irregular, infuscate elytral pattern of the new species. *Cymatodera monticola* possesses distinctly different terminalia from *C. ortegae*, as well as sinuate elytral apices.

**Description.** Holotype male. Medium sized, moderately slender anteriorly, rather robust posteriorly; brachypterous, TL = 13.1 mm. Color: Head, pronotum, thorax, femora and anterior portion of tibiae brunneous; posterior portion of femora and tarsomeres testaceous; antennae and mouthparts fuscous; abdomen fuscous, slightly darker than thorax, distal portion of abdominal segments with a depressed testaceous mark; elytral ground light testaceous, with a slight greenish tinge. Each elytron bearing a pair of irregular, sinuate, darkened fasciae: the first fascia located on anterior third, extending from the elytral suture to the epipleural fold, conspicuously slender proximal to elytral suture then abruptly widening before epipleural fold; the second fascia







Figures 11–22. Pygidia of: 11 *Cymatodera tortuosa* (male, ventral view) 12 *C. ortegae* (male, ventral view) 13 *C. gerstmeieri* (male, ventral view) 14 *C. mixteca* (male, ventral view) 15 *C. pallida* (male, ventral view) 16 *C. tortuosa* (female, ventral view) 17 *C. ortegae* (female, ventral view) 18 *C. gerstmeieri* (female, ventral view) 19 *C. mixteca* (female, ventral view) 20 *C. tortuosa* (male, dorsal view) 21 *C. ortegae* (male, dorsal view) 22 *C. ortegae* (female dorsal view).

located at the elytral mid-length, moderately wide, extending from the elytral suture to before the epipleural fold. Punctation on elytral ground infuscate (Fig. 2).

Head: HL = 2.9 mm, HW = 2.5 mm. Large, measured across eyes wider than pronotum; surface rugose; frons bi-impressed; surface moderately punctate; clothed with short, fine, recumbent setae intermixed with long, semierect and erect setae; eyes moderately small, form subsinuate, longer than wide, moderately emarginate in front, very feebly bulging laterally, separated by approximately 3 eye-widths. Antennae slender; loosely composed; extending beyond posterior margin of elytra; antennomeres 2–3 subequal in length; fourth antennomere slightly longer than third; fifth antennomere very slightly longer than fourth, antennomeres 5–10 subequal in length, antennomeres 4–10 weakly serrate; last antennomere flattened apically, somewhat acuminate distally, approximately the same length as ninth antennomere (Fig. 7).

Thorax: PL = 4.05 mm, PW = 2.1 mm. Pronotum elongate; widest at middle; middle slightly broader than anterior margin; sides constricted subapically; more constricted behind middle; moderately impressed in front of middle; subbasal tumescence pronounced; surface rugose, moderately punctate, punctation rather coarse; clothed with short, pale, semirecumbent setae, intermingled with long, stiff, semierect pale setae. Prosternum convex; wider than long; surface rugose, shining, very feebly punctate. Mesosternum feebly convex; surface rugulose, moderately, deeply punctate, vested with fine, pale, recumbent setae. Metasternum convex; shortened longitudinally; surface rugulose, devoid of tubercles or carinae, moderately, shallowly punctate. Scutel-lum conspicuously wider than long; moderately setose.

Legs: Femora clothed with short, recumbent setae intermingled with less numerous erect and semierect setae; tibiae vested with short and long erect and semierect setae; femora and tibiae transversely rugose; metathoracic legs with tarsomeres longer than those of pro- and mesothoracic legs.

Elytra: EL = 7.7 mm, EW = 3.2 mm. Form: subovate (brachypterous type). Anterior margin arcuately emarginate; narrower than widest portion of pronotum; humeri very feebly indicated; sides widest on posterior fourth; disc convex; apex rounded, broadly dehiscent, not covering sixth ventrite; surface smooth, moderately clothed with short, fine, pale, recumbent setae intermixed with long, pale, fine, erect setae; sculpturing consisting of small, coarse punctures and larger punctation irregularly arranged from base to apex, punctures becoming less numerous behind anterior third, interstices about  $3 \times$  the diameter of punctures at elytral base.

Abdomen: Ventrites 1–5 rugulose; shallowly, moderately punctate; each segment with a pair of large, shallow impressions near sides; surface clothed with short, recumbent setae intermixed with less numerous, long, semi-erect setae. Fifth ventrite (Fig. 12) moderately convex; sides oblique, moderately arcuate; posterior margin broadly, very deeply emarginate. Sixth ventrite (Fig. 12) subquadrate; protruding laterally (visible in dorsal view); rugose; surface moderately concave; somewhat punctate; lateral margins oblique, feebly arcuate; posterior margin broadly, very deeply emarginate; posterior angles somewhat blunt, slightly procurved inwardly. Fifth tergite (Fig. 21) convex, rugulose; lateral margins slightly oblique; posterior margin narrowly, shallowly emarginate. Sixth tergite (Fig. 21) subquadrate; moderately convex; surface rugulose; lateral margins moderately oblique; posterolateral angles rounded; posterior margin broadly, shallowly, triangularly emarginate.

Aedeagus 2.3 mm long; ratio of length of parameres to whole tegmen 0.4:1; tegmen fully covering phallus; parameres subparallel, pointed at apex, lateral margins fee-



Figures 23–26. Male genitalia of: 23 *Cymatodera tortuosa* 24 *Cymatodera ortegae* 25 *Cymatodera gerstmeieri* 26 *Cymatodera mixteca*.

bly oblique; phallobase moderately broad; phallus with copulatory piece feebly tapered distally; phallic plate armed with a row of moderately long denticles along the dorsal margin, these denticles increasing in size toward distal end; phallobasic apodeme short,

robust, dilated distally; phallobasic struts slender throughout their length, each as long as phallobasic apodeme (Fig. 24).

Females of the type series differ from males by having the sixth ventrite (Fig. 17) with lateral margins moderately oblique, feebly arcuate, and posterior margin very feebly, shallowly, narrowly emarginate, rather than broadly, deeply, semicircularly emarginate, as observed in males (Fig. 12); additionally, females have the sixth tergite (Fig. 22) subquadrate, with the lateral margins oblique and the posterior margin very feebly, shallowly, narrowly emarginate. Females closely resemble males in other respects.

**Variation.** Length of males range from 7.9–13.1 mm, females from 7.6–13.3 mm. Specimens examined have considerable variation in body size, and also in the shape of the fasciate pattern on the elytral ground, which ranges from almost incomplete and very narrow, to conspicuously wide, covering most part of the elytral ground. The color of these fasciae is also somewhat variable, ranging from dark testaceous to dark greenish. The elytral ground color ranges from light testaceous to light greenish.

**Distribution.** Available specimens were collected in the central-occidental part of Mexico, in the states of Colima, Jalisco and Michoacan. Distribution appears limited to the southern tip of the Sierra Madre Occidental (Fig. 29).

**Note.** This new species appears to be confined to mid-to-high altitude mountainous environments in the central-west portion of Mexico: specimens were collected on the slopes of Volcan Nevado de Colima in the states of Colima and Jalisco, and Cerro Tancitaro, in the northwestern region of the Mexican state of Michoacan. These mid and high altitude areas are dominated by pine and pine-oak forest stands.

**Etymology.** The specific epithet is a patronymic honoring Cristina Ortega, a friend of the first author.

### Cymatodera gerstmeieri Burke & Rifkind, sp. n.

http://zoobank.org/76E3C983-C185-4C7A-9BE2-1CF65B2858C7 Figs 3, 8, 13, 18, 25, 27

**Type material** (n = 8): Holotype, red labeled, male: Mexico, Chiapas, El Aguacero, 680 m, 17-VI-1990, at light, R. A. Cunningham; holotype deposited in CSCA. Paratypes, yellow labeled: 4 males, 2 females: same data as holotype (JNRC), except 2 males and 1 female collected on 16-VI-1990, and 1 male collected on 01-IX-1990; 1 female: Mexico Chiapas, Aguacero, 16 km W Ocoz[ocuautla], 1-7-VII-1986, 2500', J. E. Wappes (JEWC).

**Differential diagnosis.** *Cymatodera gerstmeieri* is similar to a number of New World tilline species that share a testaceous to ferrugineous integument and a median, dark fascia on the elytral ground; those closest include *Cymatodera mitae* Burke, *Bogcia disjuncta* Barr, and *Cymatodera insignis* Schenkling. The new species can be separated from the former as follows: male specimens of *C. gerstmeieri* have the eleventh antennomere medially depressed, acuminate posteriorly, and approximately 2× longer than tenth antennomere (Fig. 8), while males of *C. mitae* have the eleventh antennomere medially depressed.



Figures 27-28. First and second ventrites of: 27 Cymatodera gerstmeieri (male) 28 Cymatodera mitae (male).

cylindrical in shape, rounded posteriorly, and approximately  $2.5-3\times$  the length of tenth antennomere. Furthermore, males of *C. gerstmeieri* have a feebly developed longitudinal carina on the first ventrite, but lack a carina on the second ventrite (Fig. 27), whereas males of *C. mitae* have a well-developed longitudinal carina on the first ventrite, but also a somewhat less developed carina on the second (Fig. 28). The females of both species lack these carinae, but can be distinguished by the presence of a fuscous macula located on each elytral humeral angle in *C. mitae*, absent in *C. gerstmeieri*. The feebly to moderately serrate antennomeres 4-10 of *C. gerstmeieri* (Fig. 8) will easily separate it from *Bogcia disjuncta*, which has strongly serrate antennae. *Cymatodera gerstmeieri* somewhat recalls the Central American species *C. insignis*, with which it shares similar integumental color, antennal structure, and a median, slightly oblique, dark fascia. However, *C. insignis* bears a dark macula on the humeral angles, a longitudinal black macula at the posterolateral margin of the pronotum on either side, and has the posterolateral margins of the elytral ground narrowly darkened. These markings are absent in *C. gerstmeieri*.

**Description.** Holotype male. Moderately small, rather robust, metathoracic wings complete, TL = 10.2 mm. Color: head, pronotum, prosternum, mesosternum ferrug-ineous-brown, the rest of the body uniformly testaceous brown. Each elytron with a fuscous, irregular fascia near the mid-length, extending from the elytral suture to the eighth row of striae, but not reaching the epipleural fold (Fig. 3).

Head: HL = 1.2 mm, HW = 1.85 mm. Measured across eyes wider than pronotum; surface feebly rugose, shiny; frons bi-impressed; moderately, coarsely punctate; vested with short, recumbent setae interspersed with some erect, stiff setae on and behind eyes; eyes rather rounded, moderately large, slightly longer than wide, feebly emarginate in front, bulging laterally. Antennae reaching posterior margin of pronotum; second antennomere  $0.75 \times$  longer than third antennomere, antennomeres 3-10subequal in length; antennomeres 2-3 subcylindrical; fourth antennomere feebly serrate; antennomeres 5-10 moderately serrate; last antennomere flattened apically, posterior margin acuminate, about  $2 \times$  the length of tenth antennomere (Fig. 8).

Thorax: PL = 2.9 mm, PW = 1.5 mm. Pronotum rugose; moderately punctate; anterior margin as wide as middle; sides feebly constricted subapically; slightly more

constricted behind middle; disc flat, very feebly impressed in front of middle; subbasal tumescence absent; surface clothed with moderately long, semirecumbent setae interspersed with some long, erect setae. Prosternum sparsely vested, feebly punctate. Mesosternum convex; surface shiny, moderately, shallowly punctate. Metasternum with surface rugulose, shiny, moderately, shallowly puncticulate.

Legs: Moderately vested with semierect and some recumbent setae; femora puncticulate, rugulose; tibia moderately punctate, rugulose.

Elytra: EL = 6.9 mm, EW = 4.4 mm. Broader than pronotum; humeri pronounced, rounded; sides subparallel, widest portion behind posterior third; disc flattened above; surface moderately rugulose; apices rounded, feebly dehiscent; elytral declivity gradual; integument clothed with short, semierect setae intermixed with fewer long, erect setae; surface bearing coarse punctation arranged in regular striae that gradually become smaller and shallower on posterior half, punctation not reaching elytral apex; interstices at elytral base about  $1.2 \times$  the width of punctation.

Abdomen: Ventrites 1–5 rugulose, moderately, finely punctate, clothed with short, pale, fine, recumbent setae. First ventrite rather convex, subquadrate, conspicuously elevated at posterior 1/4 with a transverse, arcuate carina which does not attain posterolateral angles (Fig. 27). Fifth ventrite (Fig. 13) feebly convex; subquadrate in shape; surface shiny, moderately, shallowly, finely punctate; lateral margins oblique, somewhat arcuate; posterolateral angles rounded; posterior margin feebly, broadly emarginate. Sixth ventrite (Fig. 13) reduced; broader than long; subtriangular; lateral margins oblique, arcuate; hind margin moderately acuminate; surface feebly convex, shiny, moderately, finely, shallowly puncticulate. Fifth tergite subtriangular in shape, rugulose; lateral margin moderately oblique, posterior margin truncate. Sixth tergite subtriangular, broader than long; lateral margins strongly oblique, feebly arcuate; converging posteriorly; extending beyond apical margin of sixth ventrite.

Aedeagus 1.2 mm long; feebly sclerotized; wide; ratio of length of paramere to whole tegmen 0.45:1; tegmen partially covering phallus; parameres subtriangular; lateral margins obtuse, strongly oblique, pointed distally; phallobase wide; phallic plate armed with two long rows of moderately large denticles along dorsal and distal margins; phallobasic apodeme slender, somewhat shorter than endophallic struts; phallobasic struts slender throughout their length,  $1.5 \times$  the length of phallobasic apodeme (Fig. 25).

Females can be distinguished from males based on the structure of the pygidium. The sixth ventrite and the sixth tergite are broadly rounded posteriorly in females (Fig. 18). Other characters are constant in both sexes.

**Variation.** Length of males ranges from 8.7–10.2 mm; length of females from 9.5–13.1 mm. The elytral fascia is slightly variable in width, extending from the elytral suture to the epipleural fold in one male and one female, but incomplete in remaining individuals. Two males and one female in the type series have slightly paler elytra than the male holotype.

**Distribution.** All specimens in the type series were collected in the locality of El Aguacero, approximately 10 miles northwest of Ocozocuautla, Chiapas, Mexico. The



Figure 29. Map of central and south Mexico showing geographic position of collecting localities for: *Cymatodera tortuosa* (red circles); *C. gerstmeieri* (green circle); *C. mixteca* (yellow circles); and *C. ortegae* (blue circles).

elevation at the type locality is approximately 650 m and the vegetation is predominantly tropical deciduous forest (Fig. 29).

**Etymology.** We name this beetle for Prof. Dr. Roland Gerstmeier (Technische Universität München, Germany), in recognition of his many contributions to the study of Cleridae.

# Cymatodera mixteca Burke & Rifkind, sp. n.

http://zoobank.org/580DD2DD-2760-4140-9FCC-5D1E2B495D65 Figs 4, 9, 14, 19, 26

**Type material** (n = 16): Holotype, red labeled, male: Cacaloapan, Puebla, Mexico, 26-IV-1962, L. A. Stange. Holotype deposited in CASC. Paratypes, yellow labeled: 1 male, 1 female: same data as holotype (FMNH); 2 males: Mexico, Puebla, 2 mi SW Tehuacan, 5300', 4-X-1975, blacklight trap, 2300-0600, Powell (EMEC); 1 male: Tehuacan, Puebla, Mexico, 23-VI-1953, P. D. Hurd (JNRC); 1 female: 82 km NE Tehuacan, Puebla, Mexico, 5480 ft, rt. 2A, km 242, 7-VI-1948, desert, at light, F. Werner and W. Nutting (KSUC); 2 males, 1 female: Mexico, Puebla, 10 km N Tehuacan, 1650 m, 20-VII-1987, J. T. Doyen (EMEC); 1 female: Mexico, Puebla, 5 mi SW Zapotitlan, 8-VII- 1973, Mastro and Schaffner (TAMU); 1 female: Mexico, Puebla, 8 mi SE Tehuitzingo, 29-VI-1961, 4100', University of Kansas, Mexico expedition (SEMC); 1 male: Mexico, Puebla, 6 mi SW Tehuacan, 7-VII-1973, taken at light, Mastro and Schaffner (TAMU); 3 females: Mexico, Guerrero, Mexcala, 29-VI-1959 P. D. Hurd (EMEC);

**Differential diagnosis.** Cymatodera mixteca is most similar to the allopatric Cymatodera pallida Schaeffer, but the two species can be readily differentiated based on the structure of the antennae. Antennomeres 2–3 of C. mixteca are about the same length and width but shorter and narrower than the fourth antennomere (Fig. 9), while antennomeres 2–4 are about the same length and width but shorter and narrower than the fifth antennomere in C. pallida (Fig. 10). The elytral integument in C. mixteca is uniformly pale-testaceous to testaceous (Fig. 4) while C. pallida has a faint, wide, transversal, dark-testaceous band on the last third of the elytral ground (Fig. 5). In addition, C. mixteca is restricted to central Mexico while C. pallida is found in the southwest portion of the United States and the northern state of Chihuahua, Mexico. Male pygidia of C. mixteca and C. pallida (Figs 14–15) closely resemble one another and will not serve to separate these species. The similar and possibly sympatric species C. cylindricollis Chevrolat is darker and moderately larger than C. mixteca.

**Description.** Holotype male. Small, moderately slender, metathoracic wings complete. TL = 9.3 mm. Color: head, pronotum, prosternum, mesosternum and mouthparts testaceous; remainder of body pale testaceous (Fig. 4).

Head. HL = 1.1 mm, HW = 1.6 mm. Measured across eyes wider than pronotum; surface feebly rugose, shiny; frons not bi-impressed; moderately, finely punctate; vested with pale, short, recumbent, fine setae interspersed with some erect, fine, long and less numerous setae; eyes moderately rounded, large, slightly longer than wide, feebly emarginate in front, conspicuously bulging laterally. Antennae long, extending to posterior half of elytral length; second and third antennomere small, slender, about the same length; fourth antennomere about  $3 \times$  longer than third antennomere, antennomeres 4–10 robust, moderately elongate, subequal in length; antennomeres 2–3 subcylindrical; antennomeres 4–10 moderately serrate; last antennomere acuminate posteriorly, flattened apically, about the same length of tenth antennomere (Fig. 9).

Thorax: PL = 1.7 mm, PW = 0.9 mm. Pronotum moderately rugose, feebly, finely punctate; anterior margin as wide as middle and posterior margin; sides feebly constricted subapically; more constricted behind middle; disc flat, feebly impressed in front of middle; anterior pronotal impression present, subbasal tumescence obvious; surface moderately clothed with pale, stiff, short and long, semirecumbent setae. Prosternum sparsely vested, feebly, finely punctate. Mesosternum convex; surface shiny, smooth, moderately, shallowly punctate. Metasternum with surface feebly rugose, moderately, shallowly puncticulate. Scutellum ovoid, wider than long, posteriorly emarginate.

Legs: Moderately vested with pale, fine, recumbent setae intermixed with some scattered, very long, pale, stiff setae; femora transversally rugulose; tibia feebly punctate, longitudinally, finely rugulose.

Elytra: EL = 4.9 mm, EW = 2.1 mm. Broader than pronotum; humeri pronounced, rounded; sides slightly ovoid; widest portion at posterior fourth; disc moderately flat-

tened above, slightly depressed medially, smooth; apices subtriangular, feebly dehiscent; elytral declivity steep; integument clothed with short, pale, fine, recumbent setae intermixed with long, erect, pale, stiff setae; sculpture consisting of moderately coarse punctation arranged in regular striae that gradually become smaller and shallower on toward apex, punctation disappear before elytral apex; interstices at elytral base smooth, about 2.0× the width of punctation.

Abdomen: Ventrites 1–4 shiny, smooth; feebly, finely punctate; clothed with few short, pale, fine, recumbent setae; posterior margins truncate; lateral margins not depressed. Fifth ventrite (Fig. 14) conspicuously wider than long; surface smooth, shiny, moderately concave; lateral margins oblique, finely arcuate; posterior margin broadly, shallowly emarginate. Sixth ventrite (Fig. 14) small; broader than long; subtriangular; surface shiny, smooth, very finely rugulose, medially convex; lateral margins strongly oblique, arcuate, hind margin broadly, very shallowly emarginate; posterolateral angles rounded. Fifth tergite subquadrate; rugulose; lateral margin moderately oblique, posterior margin truncate. Sixth tergite subquadrate, broader than long; surface concave; lateral margins oblique, feebly arcuate; posterior margin truncate; posterolateral angles broadly rounded. Sixth tergite extending slightly beyond the apical margin of sixth ventrite.

Aedeagus 0.9 mm long; feebly sclerotized; moderately wide; ratio of length of paramere to whole tegmen 0.3:1; tegmen partially covering phallus; parameres ovoid; lateral margins obtuse, oblique, pointed distally; phallobase wide; phallic plate devoid of denticles, distal portion of phallic plate spinous, spines reduced; phallus rounded at apex, conspicuously wide at middle; phallobasic apodeme robust, swollen distally, longer than phallobasic struts; phallobasic struts moderately robust, swollen distally, approximately  $1.2 \times$  the length of phallobasic apodeme (Fig. 26).

Females of the type series can be differentiated from males by the shape of the sixth ventrite. This segment is broadly rounded posteriorly (Fig. 19), rather than shallowly emarginate, as in males (Fig. 14). Remaining characters are constant in both sexes.

**Variation.** Length of males ranges from 6.3–8.4 mm; length of females from 7.1–7.9 mm. Individuals in the type series vary somewhat in integument color, ranging from pale testaceous to brown. Such color variation is observable in male and female members in the type series. Remaining characters in the type series remain consistent.

**Distribution.** The type series was collected from various localities in the Sierra Mixteca of Mexico, specifically in the south and southwestern portion of the state of Puebla, and in central Guerrero state (Fig. 29). This region is characterized by tropical deciduous to thorny forest habitats.

**Etymology.** The specific epithet makes allusion to the regional home of the Mixteca people, and of this new species.

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RESEARCH ARTICLE



# A new, mesophotic Coryphopterus goby (Teleostei, Gobiidae) from the southern Caribbean, with comments on relationships and depth distributions within the genus

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# Abstract

A new species of western Atlantic *Coryphopterus* is described from mesophotic depths off Curaçao, southern Caribbean. *Coryphopterus curasub* **sp. n.**, is similar to *C. dicrus* in, among other features, having two prominent pigment spots of roughly equal intensity on the pectoral-fin base, the pelvic fins fused to form a disk, and no pelvic frenum. The two species can be differentiated by body depth (shallower in *C. curasub* at origin of dorsal fin and caudal peduncle); differences in the pigmentation on the head, trunk, and basicaudal region; and usually by total number of rays (spinous plus soft) in the second dorsal fin (10-11,usually 11, in *C. curasub*, 10 in *C. dicrus*). *Coryphopterus curasub* differs from other *Coryphopterus* species that have a prominent pigment spot on the lower portion of the pectoral-fin base (*C. punctipectophorus* and *C. venezuelae*) in, among other features, lacking a pelvic frenum. *Coryphopterus curasub* was collected between 70 and 80 m, the deepest depth range known for the genus. Collections of *C. venezuelae* at depths of 65–69 m extend the depth range of that species by approximately 50 m. Mitochondrial cytochrome c oxidase subunit I (COI) data corroborate the recognition of *C. curasub* as a distinct species but do not rigorously resolve its relationships within the genus. A revised key to the western Atlantic species of *Coryphopterus* is presented.

#### Keywords

*Coryphopterus curasub, Coryphopterus dicrus*, submersible, Substation Curaçao, Deep Reef Observation Project (DROP), DNA barcoding, phylogeny

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# Introduction

Coryphopterus gobies live in mostly shallow warm waters of the western Atlantic and eastern Pacific Oceans, dwelling on sand around coral and rocky reefs or hovering above or perching on reef structures. Twelve species are known from the western Atlantic and one from the eastern Pacific (Böhlke and Robins 1960, 1962; Thacker and Cole 2002; Victor 2007; Baldwin et al. 2009). Most Coryphopterus species inhabit depths < 40 m, but C. hyalinus Böhlke & Robins has been recorded to 52 m; C. eidolon Böhlke & Robins and C. thrix Böhlke & Robins to 54 m; C. dicrus Böhlke & Robins to 56 m; C. lipernes Böhlke and Robins to 60 m; C. glaucofraenum Gill to 61 m; and C. personatus (Jordan & Thompson) to 70 m (Böhlke and Robins 1960, 1962; Thacker and Cole 2002; Feitoza et al. 2005, Robertson and Van Tassell 2015). Published depth data for C. glaucofraenum, however, as well as that for C. tortugae (Jordan) (5-32 m) and C. venezuelae Cervigón (1-20 m), must be interpreted cautiously because of historical confusion about the taxonomy of this group, as should those of C. hyalinus and C. personatus for the same reason (Baldwin et al. 2009). Recent submersible diving to 300 m off Curaçao in the southern Caribbean as part of the Smithsonian Institution's Deep Reef Observation Project (DROP) resulted in the collection of three specimens of C. venezuelae at 65-69 m and four specimens of an unidentified Coryphopterus at 70-80 m. We describe the specimens from 70-80 m as a new species and comment on its relationships within the genus.

Thacker and Cole (2002) investigated species relationships within *Coryphopterus* based on morphology and one mitochondrial gene (ND2). Their phylogeny suggests that *Coryphopterus* is restricted to the western Atlantic and eastern Pacific and that the Indo-Pacific genus *Fusigobius*, which Randall (1995) synonymized with *Coryphopterus*, is distinct. Based on study of the western Atlantic *Lophogobius cyprinoides*, Thacker and Cole (2002) and Thacker and Roje (2011) hypothesized that *Coryphopterus* is more closely related to *Lophogobius* than it is to *Fusigobius*. The eastern Pacific *C. nicholsii* (Bean), which is sister to *Lophogobius* + *Coryphopterus* in the phylogenies of Thacker and Cole (2002) and Thacker and Roje (2011), has been re-relegated to the monotypic genus *Rhinogobiops* Hubbs (Thacker 2011, Van Tassell 2011).

A new western Atlantic species, *Coryphopterus kuna* Victor, was described in 2007 but not included in the molecular phylogeny of Thacker and Roje (2011). Further, neither *C. tortugae* nor *C. venezuelae* was included in previous phylogenetic work, even though both appear to be valid *Coryphopterus* species. Although Longley and Hildebrand (1941) and Böhlke and Robins (1960) considered *C. tortugae* (Jordan) to be a synonym of *C. glaucofraenum*, Garzón-Ferreira and Acero (1990) redescribed it as valid. Victor (2008) concurred and also described a variant of *C. tortugae* as new species, *C. bol.* Based on an integrative molecular and morphological analysis, Baldwin et al. (2009) also recognized *C. tortugae* as valid but relegated *C. bol* to the synonymy of *C. venezuelae*. In addition to the new species described here, tissue samples of all known *Coryphopterus* species except the Gulf of Mexico species *C. punctipectophorus* Springer are now available and were incorporated into the genetic analyses conducted in this study. As noted by Baldwin et al. (2009), Thacker and Cole's (2002) DNA sequence from Belize previously thought to be from *C. punctipectophorus* (GenBank Accession No. AF391396) is actually from *C. dicrus*.

#### Materials and methods

Four specimens of the new species and three of *C. venezuelae* were collected using Substation Curaçao's (http://www.substation-Curacao.com) manned submersible *Curasub*. The sub has two flexible, hydraulic arms, one of which is equipped with a quinaldine-ejection system and the other with a suction hose. Anesthetized fish specimens were captured with the suction hose, which empties into a vented plexiglass cylinder attached to the outside of the sub. At the surface, the specimens were photographed, tissue sampled, and preserved. Preserved specimens were later photographed to document preserved pigment pattern and X-rayed with a digital radiography system. Counts and measurements follow Randall (2001). Format for dorsal-fin formula follows Birdsong et al. (1988). Head pore terminology follows Akihito et al. (1988). Measurements were made weeks to months after preservation and were taken to the nearest 0.1 mm with digital calipers or an ocular micrometer fitted into a Zeiss stereomicroscope.

Tissue samples for DNA Barcoding were stored in saturated salt-DMSO (dimethyl sulfoxide) buffer (Seutin et al. 1991). DNA extraction, PCR, sequencing cytochrome c oxidase subunit I (COI), and editing COI sequences were performed as outlined by Weigt et al. (2012a). A neighbor-joining tree (Saitou and Nei 1987) was generated using PAUP\*4.1 (Swofford 2002) on an analysis of Kimura two-parameter distances (Kimura 1980). The neighbor-joining analysis reveals genetic distances in COI among individuals and clusters them into genetically distinct lineages, which, in teleost fishes, correspond well with species (e.g. Baldwin and Weigt 2012, Weigt et al. 2012b). Interspecific phylogenetic relationships were hypothesized for Coryphopterus based on maximum parsimony analysis of the COI sequences using heuristic searches in PAUP\*4.1 (Swofford 2002). Characters were equally weighted and left unordered. The resulting equally parsimonious trees were summarized using the strict consensus method, and nodal support was estimated from 1,000 replicates of the bootstrap, utilizing random addition sequence and TBR branch swapping (Swofford 2002). The outgroup for the neighbor-joining analysis was a species of Fusigobius, a basal genus in the crested goby group that includes Coryphopterus, Lophogobius, and Rhinogobiops (Thacker and Roje 2011). Outgroups for the parsimony analysis were Fusigobius and Rhinogobiops.

GenSeq nomenclature (Chakrabarty et al. 2013) and GenBank accession numbers for DNA sequences derived in this study are presented along with museum catalog numbers for voucher specimens in the Appendix. GenBank accession numbers for *Coryphopterus* sequences included in the analyses that were published by Baldwin et al. (2009) are GQ367306–GQ367475, and those for *Lophogobius cyprinoides* sequences published by Weigt et al. (2012b) are JQ840574.1 and JQ842196.1. GenBank accession numbers for *Fusigobius duospilus* and *Rhinogobiops nicholsii* are JX462852 and HQ909488, respectively.

# Results

#### Coryphopterus curasub sp. n.

http://zoobank.org/5C60A7B0-58D9-4896-81EF-A0D1DD28873D Yellow-spotted sand goby Figs 1–2

#### Type locality. Curaçao, southern Caribbean

Holotype. USNM 406373, Smithsonian DNA number CUR 11373, 33.3 mm SL, female, *Curasub* submersible, sta. 11-05, southern Caribbean, Curaçao, east of downline off Substation Curaçao dock, near 12°05.069'N, 68°53.886'W, 80 m, quinaldine, 30 May 2011, D. R. Robertson, B. Brandt, A Schrier, K. Stewart.

**Paratypes.** USNM 430037, CUR 13302, 30.0 mm SL, male, *Curasub* submersible, sta. 13-29, southern Caribbean, Curaçao, east of downline off Substation Curaçao dock, near 12°05.069'N, 68°53.886'W, 70–72 m, quinaldine, 30 October 2013, C. C. Baldwin, D. R. Robertson, B. Brandt, C. Castillo, L. Ybarrondo. USNM 431328, CUR 14003, 31.0 mm SL, male, *Curasub* submersible, sta. 14-01, southern Caribbean, Curaçao, east of downline off Substation Curaçao dock, near 12°05.069'N, 68°53.886'W, 73 m, quinaldine, 17 March 2014, C. C. Baldwin, D. R. Robertson, B. Brandt, C. Castillo, H. Reichardt. USNM 430019, CUR 13303, 17.5 mm SL, immature (same collection locality as USNM 430037), cleared and stained.

**Generic assignment.** The combination of six spines in the first dorsal fin, fewer than 20 rays in the second dorsal fin, pelvic fin with one spine and five soft rays, head pores present, no free pectoral-fin rays, no scales on top of head, and no prominent crest on top of head anteriorly from first dorsal fin support the placement of *C. curasub* in the genus *Coryphopterus* (Murdy 2002).

**Diagnosis.** A species of *Coryphopterus* distinguishable from its congeners by the following combination of characters: total second dorsal-fin rays (spinous plus soft) 10-11, usually 11; total anal-fin rays (spinous plus soft) 10; pectoral-fin rays 19-20; pelvic fins united; no pelvic frenum; pectoral-fin base with two prominent dark spots (yellow with dark spotting in life) of roughly equal intensity, one on dorsal portion of fin base and one on ventral part; no distinct black blotch behind orbit above opercle; no dark triangular blotch immediately behind middle of orbit; blotches of pigment on trunk mostly yellow; few melanophores and yellow dots interspersed among yellow blotches of pigment on trunk; no black ring of pigment surrounding anus; dark triangular blotch variously developed beneath anteroventral corner of orbit; basicaudal blotch cross-shaped, with prominent anterior projection; and two yellow/orange blotches on base of caudal fin situated immediately behind basicaudal blotch.

**Description.** Dorsal-fin rays VI + I, 9-10 (9 in one paratype, 10 in other specimens), total second dorsal-fin rays 10 or 11 (10 in one paratype, 11 in other specimens); anal-fin rays I, 9; all soft dorsal- and anal-fin rays branched. Pectoral-fin rays 19, 19 (paratypes) or 20, 20 (holotype); all pectoral rays branched except splint-like uppermost and lowermost rays. Pelvic-fin rays I, 5; all soft rays branched; fins unit-



**Figure 1.** *Coryphopterus curasub* sp. n., type specimens: **A**, **B** USNM 406373, holotype, Smithsonian DNA number CUR 11373, 33.3 mm SL, female – after preservation (**A**) and before preservation (**B**) **C** USNM 431328, Paratype, CUR 14003, 31.0 mm SL, male, before preservation **D** USNM 430019, Paratype, CUR 13303, 17.5 mm SL, immature, before preservation and clearing and staining. Note that the dark color on the posterior portion of the caudal fin is an artifact of flash photography and does not reflect the existence of dark pigment. Photos by Ian Silver-Gorges (**A**) and D. R. Robertson and C. C. Baldwin (**B–D**).

ed, no frenum. Total caudal-fin rays (including procurrent rays) 30 (holotype) or 31 (cleared and stained paratype; can't assess number from radiographs of other paratypes); segmented caudal rays17; branched caudal rays12 (6+6); unbranched caudal rays 18 in holotype (9+9), 19 (10+9) in cleared and stained paratype. Dorsal-fin formula 3-22110. Vertebrae 10 precaudal +16 caudal. Epineurals 10 pairs. Ribs on vertebrae 3-10. Anal-fin pterygiophores anterior to first haemal spine 2. Gill rakers on first arch, including rudiments, 2 + 8 (holotype and adult paratypes), 0 + 8 in cleared and stained juvenile paratype. Branchiostegal rays 5. Numerous scales abraded and missing on all specimens, one paratype with approximately 22 scales in longitudinal series, 6 scales between origin of second dorsal fin obliquely downward to anal fin, and approximately 17 circum-peduncular scales.

Measurements of holotype in parentheses following extremes for holotype plus two adult paratypes. Juvenile paratype bent and not measured prior to clearing and staining. Body elongate, maximum depth from base of dorsal-fin spines 17–19% SL (17); body compressed, greatest width just posterior to gill opening 14–15% SL (14); head length 30–32% SL (32); snout length (to fleshy edge of orbit) 6.0–7.1% SL (6.0); greatest fleshy orbit diameter 9.3–10% SL (9.3); least fleshy interorbital 1.3–3.3% SL (3.3); caudal-peduncle length 23–24% SL (23); least caudal-peduncle depth 11–12% SL (11); length of dorsal-fin base 39–46% SL (46); first dorsal spine 14–16% SL (16); second dorsal spine 15–18% SL (17); third and longest dorsal spine 16–20% SL (18); sixth and shortest dorsal spine 7.0–8.4% SL (8.4); seventh dorsal spine (first element of second dorsal fin) 14–16% SL (16); last dorsal soft ray 16–17% SL (16); anal-fin spine 8.7–11% SL (11); last anal soft ray 20–22% SL (20); pectoral fin 32–36% SL (36), fin reaching vertical through second anal-fin soft ray, longest pectoral rays the 11<sup>th</sup>-14<sup>th</sup> rays from top of fin; pelvic fin 24–32% SL (27), fin reaching origin of anal fin or terminating slightly before origin, longest pelvic ray the fourth.

Trunk, belly, and pre-pelvic region scaled, head and predorsal region naked; scales ctenoid except on pre-pelvic region, where they are cycloid. Upper jaw with several rows of small conical teeth, outermost teeth largest but smaller than outermost dentary teeth; dentary with outer row of fairly large conical teeth and several inner rows of smaller teeth; innermost teeth intermediate in size between teeth of outermost row and those adjacent to it. No teeth on vomer, palatines, or pterygoids. Anterior nare opening on short tube; posterior nare a simple opening. Head pores prominent: nasal pore, anterior interorbital pore, posterior interorbital pore, infraorbital pore, postorbital pore, pore at each end of lateral sensory canal, pore at each end of posterior lateral canal, and three preopercular pores (pores B', C and D [both single], E, F, G, H', K', L', M', N, O'). A very low, thin ridge of tissue extending from just posterior to interobital region to base of first dorsal fin.

When photographed against a light background (Fig. 2A), the following color pattern visible in holotype. TRUNK: ground color white, several irregular horizontal rows of yellow/orange irregular-shaped blotches, most blotches bordered by and peppered with small black melanophores; uppermost row comprising approximately 11 blotches distributed along dorsal body margin from head (with two-three blotches) to caudal



**Figure 2.** Comparison of **A** *Coryphopterus curasub* sp. n., holotype, USNM 406373, 33.3 mm SL, and its most similar congener **B** *Coryphopterus dicrus*, USNM 413296, 30 mm SL. Note the differences in the shape of the basicaudal pigment marking (with distinct anterior projection in *C. curasub*), body depth (shallower in *C. curasub*), head pigment (absence of a distinct blotch of black pigment immediately posterior to the orbit and presence of a black triangle of pigment beneath the anteroventral portion of orbit in *C. curasub* (present and absent, respectively, in *C. dicrus*), and trunk pigment (blotches predominantly yellow with few melanophores interspersed among them in *C. curasub* vs. blotches predominantly orange/ rust with numerous melanophores interspersed among them in *C. dicrus*).

peduncle; second row shortest, comprising approximately eight blotches and extending from just posterior to posterodorsal margin of orbit to vertical through third or fourth dorsal-fin spine; third row comprising approximately 15 blotches and extending from middle of posterior margin of orbit to caudal peduncle-anterior blotches of this row united to form irregular stripe; lowermost row comprising five prominent blotches that extend from vertical through anterior origin of second dorsal fin to posterior portion of caudal peduncle and several less conspicuous blotches anterior of and within this series; this row continuing anteriorly onto pectoral-fin base and head as an irregular yellow stripe that passes along ventral margin of orbit and terminates on posterior end of upper jaw; yellow blotches on anterior portion of trunk and head better defined by peripheral melanophores than those on posterior portion of trunk; scattered small spots of yellow/black pigment interspersed among blotches in most rows. HEAD: head pigment also including short yellow/black stripe on snout; yellow stripe on ventral portion of head from posterior end of lower jaw to preopercle; triangle of black pigment beneath anteroventral margin of orbit; scattered black dots on upper lip, on snout, and beneath eye; two small black blotches of pigment on operculum; some whitish pigment extending posterodorsally from dark triangle beneath orbit; pupil black, iris brown; ventral portions of head and trunk mostly white except for streak of yellow pigment beneath opercular opening. CAUDAL PEDUNCLE: dark crossshaped basicaudal blotch present on central portion of caudal peduncle and caudal-fin base, the anterior projection of cross prominent; two yellow spots bordering dorsal and ventral ends of blotch posteriorly and extending onto bases of several caudal-fin rays. FINS: first dorsal fin translucent, with three irregular yellow stripes; second dorsal fin with three or four irregular yellow stripes; anal fin with broad stripe of pale yellow pigment on middle of fin; caudal fin with blotches of yellow pigment forming an arc on basal portion of fin, some of this pigment extending distally along caudal rays as barely noticeable yellow streaks; pectoral and pelvic fins mostly clear; yellow/black blotch on dorsal portion of pectoral-fin base extending onto basal portion of dorsalmost pectoral rays; lower portion of pectoral-fin base with well-defined, round, yellow/orange blotch with dark dots. When photographed against a black background (Fig. 1B), numerous small, white, round to oblong spots visible on membranes of all fins; distal margin of anal fin with pale blue stripe.

Male paratypes (Fig. 1C) with similar coloration except black triangle of pigment beneath anteroventral corner of eye less conspicuous; melanophores present on lower jaw; iris mottled whitish/bronze; pectoral fin with pale yellow pigment; and pelvic fin with black-spotted yellow patches. IMMATURE SPECIMEN (Fig. 1D) paler but with most pigment described above developing; diagnostic cross-shaped marking on caudal peduncle not formed, and only lower black-spotted yellow blotch on pectoralfin base well formed; most prominent pigment comprising yellow stripes on head, five black-spotted yellow blotches on trunk in lowermost row, and black-spotted yellow blotch in line with this row on base of caudal fin.

Color of holotype in alcohol (Fig. 1A). Ground color of head and trunk light tan, overlain by assorted dark circles, stripes, and irregular markings. Scattered melanophores and blotches present along base of spinous dorsal fin and on dorsal portion of trunk. Most prominent trunk pigment located just ventral to lateral midline as a row of six mostly circular blotches of roughly equal size except the second from anterior and last, which are small relative to the others; this row of pigment markings originating at a vertical through second element of second dorsal fin and terminating on caudal peduncle. Head with several circular blotches in row posterior to posterodorsal portion of orbit; irregular stripe of pigment extending posteriorly from middle of orbit; irregular stripe-like mark extending posteriorly from posteroventral portion of orbit; scattered melanophores on snout, lacrimal, and upper jaw; dark triangle of pigment beneath anteroventral corner of orbit; and two irregular streaks of pigment on ventral portion of operculum. Dark portion of basicaudal blotch as described above, but no melanophores present on remainder of caudal fin or on anal and pelvic fins. First dorsal fin with small bits of dark pigment on membranes of second, third and fifth spines. Pectoral-fin base with one dorsal and one ventral circular blotches, the former extending as short series of melanophores posteriorly onto bases of dorsal rays of fin.

Distribution. Known from 70-80 m off Curaçao, southern Caribbean.

**Habitat.** Notes recorded during the submersible dive on which the 33.0 mm SL paratype (USNM 431328) was collected indicate that it occurred on sand with rubble patches on a 45°slope.

**Etymology.** Named for the manned submersible *Curasub*, which is owned and operated by Substation Curaçao, in recognition of the contributions of this vehicle to increasing our knowledge of the Caribbean deep-reef fish fauna.

**Common name.** "Yellow-spotted sand goby" refers to the yellow spots on the trunk and the collection habitat.

Morphological comparisons. Coryphopterus curasub is most similar to C. dicrus (Fig. 2) and keys to that species in the most recent dichotomous key to western Atlantic Coryphopterus (Baldwin et al. 2009). They share the presence of two dark circular markings on the pectoral-fin base that are of roughly equal intensity (except in the juvenile C. curasub, in which only the lower spot is prominent), the absence of a distinct black blotch or triangle of pigment behind the eye above the opercle in adults, the presence of a united pelvic fin in which the fourth rays are longer than the fifth, and the absence of a pelvic frenum. They usually differ in total number of rays (spinous plus soft) in the second dorsal fin (10–11, usually 11, in *C. curasub*, 10 in *C. dicrus*); absence of a dark triangular blotch immediately posterior to the orbit in C. curasub (present in *C. dicrus*); presence of a dark triangular blotch beneath the anteroventral portion of the orbit in the largest specimen (33.3 mm SL holotype) of C. curasub (absent in similarly large specimens of C. dicrus); blotches of pigment on the trunk typically yellow in *C. curasub*, orange to rusty brown in *C. dicrus*; few yellow spots with tiny melanophores among the yellow blotches of pigment on the trunk in C. curasub vs. many rusty spots with tiny melanophores between the rusty brown blotches in C. dicrus; configuration of the basicaudal blotch (a cross-shaped blotch with a distinct anterior projection in C. curasub vs. a dumbbell-shaped bar in C. dicrus); the two yellow/ orange blotches on the base of the caudal fin situated immediately behind the basicaudal blotch in C. curasub vs. superimposed on and contributing to the upper and lower heads of the basicaudal bar of C. dicrus; maximum body depth from base of spinous dorsal fin (17-19% SL in C. curasub, 20-26% SL in C. dicrus - Böhlke and Robins 1960); least depth of caudal peduncle (11-12% SL in C. curasub, 13-15% SL in C. dicrus - Böhlke and Robins 1960). Coryphopterus curasub differs from all other western Atlantic Coryphopterus (C. alloides Böhlke & Robins, C. eidolon, C. glaucofraenum, C. hyalinus, C. kuna, C. lipernes, C. personatus, C. punctipectophorus, C. thrix, C. tortugae, and C. venezuelae) in having two round dark marks on the pectoral-fin base that are of roughly equal intensity in adults. It further differs from C. hyalinus, C. lipernes, and C. personatus in lacking a black ring around the anus; from those species and C. alloides in having the pelvic fin united; from C. glaucofraenum, C. tortugae, and C. venezuelae in lacking both a pelvic frenum and a distinct black blotch or triangle behind the eye above the opercle; and from C. kuna in having 10 or 11 total second dorsal-fin rays, 10 total anal-fin rays, and 19–20 pectoral-fin rays (vs. 9, 9, and 15, respectively).

Of the 14 apomorphic morphological characters of Coryphopterus species tabulated by Thacker and Cole (2002) for inclusion in their phylogenetic analysis of the genus, C. curasub has (character 1) no pelvic frenum, (3) the fifth (innermost) pelvic-fin ray shortened relative to the fourth, (4) a low ridge of tissue on top of the head, (8) orange or gold coloration on the body, and (10) three stripes of pigment on the head. The presence of a low ridge of tissue on the head characterizes all Coryphopterus species and is thus uninformative. Likewise, although Thacker and Cole (2002) scored most species as lacking orange or gold coloration, in a more thorough analysis of fresh color patterns in western Atlantic Coryphopterus, Baldwin et al. (2009) noted the presence of yellow/orange/gold pigment in all species. Coryphopterus curasub shares with C. dicrus, C. alloides, C. personatus, C. hyalinus, and C. *lipernes* the absence of a pelvic frenum, with those taxa and *C. eidolon* a shortened fifth pelvic-fin ray (relative to the fourth), and with C. eidolon, C. thrix, C. dicrus, C. glaucofraenum, C. tortugae, C. venezuelae, C. urospilus, and C. punctipectophorus the presence of three stripes of pigment on the head. In C. curasub, the lowermost stripe (on the cheek) is yellow and lacks melanophores, which are present in the other species. The homology of the pigment stripes is thus questionable. Thacker and Cole (2002) list several apomorphic characters (11-13) related to basicaudal pigment, but the configuration of the basicaudal blotch in C. curasub is unique among Coryphopterus species. Thacker and Cole's (2002) 14th character, the presence or absence of a pigment spot on the pectoral-fin base, insufficiently describes the variation in this character in Coryphopterus. Of the various configurations-no spots, one spot dorsally, one spot ventrally, two spots with upper spot more intense, two spots of roughly equal intensity-only C. curasub and C. dicrus have two spots of equal intensity among Coryphopterus species and outgroup taxa. In summary, of the potentially informative, putative apomorphic characters exhibited by *C. curasub*, only C. dicrus shares all of them.

**Genetic comparisons.** COI sequences derived from tissue samples from the four type specimens of *C. curasub* and three specimens of *C. venezuelae* collected by submersible as part of this study (Appendix) were combined with 173 previously published COI sequences for western Atlantic *Coryphopterus* (Baldwin et al. 2009, Weigt et al. 2012b) in a neighbor-joining analysis (Fig. 3). Intraspecific divergence in COI for *C. curasub* was 0.1% as compared to 17–23% interspecific divergence between *C. curasub* and other western Atlantic *Coryphopterus* species, including *C. dicrus* (18%, Table 1). Intraspecific divergences for all western Atlantic *Coryphopterus* species were < 1% except for *C. alloides* (3.7%), likely reflecting, as suggested by Baldwin et al. (2009), a cryptic species that awaits investigation. Phylogenetic relationships within *Coryphopterus* (43 ingroup sequences selected from the entire COI data set to maximize geographical coverage of each species), the eastern Pacific *C. urospilus*, and the western Atlantic *Lophogobius cyprinoides*. A strict consensus of 24 trees resulting from a maximum parsimony analysis (Fig. 4) does not resolve the



**Figure 3.** Neighbor-joining tree derived from COI sequences for western Atlantic species of *Coryphopterus*. The tree was rooted on *Fusigobius duospilus*. Divergence represented by scale bar = 3%. Note: *C. punctipectophorus* from the Gulf of Mexico was not available for inclusion in this analysis.

relationships of *C. curasub* with confidence. A clade comprising *C. venezuelae*, *C. glaucofraenum*, and *C. tortugae* is strongly supported (99%), as are clades comprising *C. venezuelae* and *C. glaucofraenum* (80%), the hovering species *C. hyalinus* and *C. personatus* (100%), and the planktivores *C. lipernes* + *C. hyalinus* + *C. personatus* (63%). As noted by Thacker and Cole (2002) based on ND2 mitochondrial and morphological data, *Lophogobius* is closely related to *Coryphopterus*, here appearing in a poorly supported clade that also comprises *C. dicrus* and the eastern Pacific *C. urospilus*. Adding more loci to the genetic analysis as well as *C. punctipectophorus* and the eastern- and Indo-Pacific species of *Lophogobius* may help resolve interspecific

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	curasub	dicrus	lipernes	hyalinus	personatus	tortugae	glaucofraenum	venezuelae	tbrix	eidolon	alloides	kuna
C. curasub (n=4)	0.10											
C. dicrus (n=24)	18.00	0.60										
C. lipernes (n=7)	19.40	21.70	0.10									
C. hyalinus (n=11)	18.00	19.60	14.90	0.10								
C. personatus (n=11)	19.00	19.00	15.60	7.20	0.10							
C. tortugae (n=21)	17.40	17.50	19.60	21.10	20.10	0.20						
C. glaucofraenum (n=26)	17.80	20.60	20.70	21.70	21.50	12.10	0.20					
C. venezuelae (n=39)	17.40	18.30	21.40	20.90	20.10	9.90	9.50	0.40				
C. thrix $(n=7)$	22.90	21.30	21.90	21.10	19.70	19.00	21.20	19.60	0.20			
C. eidolon (n=21)	22.90	19.40	25.20	19.20	18.00	19.70	12.10	18.90	19.60	0.10		
C. alloides (n=4)	17.90	17.90	21.90	17.90	18.40	20.00	21.30	18.90	20.20	18.50	3.70	
<i>C. kuna</i> (n=5)	22.70	24.90	26.40	23.20	25.70	25.50	27.90	26.00	24.90	25.80	23.60	0.50



**Figure 4.** The strict consensus of a maximum parsimony analysis of the COI region of 42 individuals of *Coryphopterus* and *Lophogobius cyprinoides*. *Fusigobius duospilus* and *Rhinogobiops nicholsii* were outgroups in the analysis. Numbers above branches represent bootstrap support values > 50. Note: *C. punctipecto-phorus* from the Gulf of Mexico was not available for inclusion in this analysis.

and generic relationships. The placement of *C. kuna* outside of the western Atlantic *Coryphopterus* + *Lophogobius cyprinoides* clade warrants further morphological and molecular investigation.

**Depth distributions.** Depth ranges of *Coryphopterus* species are shown in Figure 5. *Coryphopterus curasub*, which is known from 70–80 m, is the only member of the genus that has a narrow depth range completely confined to mesophotic depths. *Coryphopterus venezuelae*, *C. glaucofraenum*, *C. dicrus*, *C. eidolon*, *C. thrix*, *C. hyalinus*, *C. lipernes*, and *C. personatus* inhabit depths as deep as 52–70 m, but they have broad depth ranges that extend as shallow as 1–6 m. The only *Coryphopterus* species in addition to *C. curasub* that we have collected using the *Curasub* submersible are *C. hyalinus* – one specimen from 33 m, and *C. venezuelae* – three specimens from 65–69 m. Prior to this study, *C. venezuelae* was known from 1–20 m (Robertson and Van Tassell 2015), and thus our new collections of the species off Curacao extend its known range by nearly 50 m.



**Figure 5.** Depth ranges for *Coryphopterus* species. Data are from Böhlke and Robins (1960, 1962); Thacker and Cole (2002); Feitoza et al. (2005); Robertson and Van Tassell (2015); the Florida Museum of Natural History online fish catalog – http://specifyportal.flmnh.ufl.edu/fishes/; the Florida Fish and Wildlife Conservation Commission online catalog http://myfwc.com/research/saltwater/specimen-collections/sis/ichthyology/; the Smithsonian National Museum of Natural History online catalog – www. vertebrates.si.edu//search/fishes); and this study.

# Revised key to the Western Atlantic species of Coryphopterus

(Modified from Baldwin et al. 2009)

1	Black ring surrounding anus; pelvic fins separate, frenum absent2
_	No black ring around anus; pelvic fins separate or fused, frenum present or
	absent4
2	One anterior interorbital pore
_	Two anterior interorbital pores Coryphopterus hyalinus
3	Total second dorsal-fin rays (spinous plus soft) typically 11; total anal-fin
	rays (spinous plus soft) typically 11; in life, head with orange pigment, body
	translucent with several square- or rectangular-shaped orange blotches inter-
	nally; preserved specimens lacking conspicuous postorbital stripes of melano-
	phores but with dark "mask" around eye Coryphopterus personatus
-	Total second dorsal-fin rays (spinous plus soft) typically 10; total anal-fin
	rays (spinous plus soft) typically 10; in life, head and body predominantly
	yellow, with blue-white stripes extending posteriorly from dorsal and ventral
	portions of orbit; a dusky internal stripe along posterior section of vertebral
	column; preserved specimens with postorbital stripes of melanophores and
	scattered spots over entire body Coryphopterus lipernes
4	No distinct black blotch behind eye above opercle in adults; pigment mark
	above opercle, if present, no larger or darker than other marks behind eye;
	pelvic fins separate or fused, frenum present or absent (see Baldwin et al.
	2009 for additional comments)
-	Distinct black blotch or triangle behind eye above opercle in adults, blotch
	usually larger and darker than other pigment in stripe behind eye; pelvic fins
	fused to form disc, frenum present (see Baldwin et al. 2009 for additional
5	Tatal and for more (minore plus of) 8.0 (mullion), more plus of 15
)	10 tai anai-nn rays (spinous pius son) 8–9 (usually 9), pectorai-nn rays 1)–
	Total anal fin rays (spinous plus soft) 10, 11, pectoral fin rays 17, 20, pelvic
_	for fused frequences present or absent $7$
6	Total second dorsal-fin rays (spinous plus soft) 9: total anal-fin rays (spinous plus
0	soft) 9: pectoral-fin rays (spinous pius soft) 9; total and initialys (spinous pius soft) 9:
	bead and body with orange spots and blotches sometimes a flag of dark pigment.
	on 1st–3rd dorsal spines: pelvic fins fused to form a disc. <i>Corryphopterus kuna</i>
_	Total second dorsal-fin rays (spinous plus soft) 10: total anal-fin rays (spinous
	plus soft) 9 (rarely 8); pectoral-fin rays 16–17; black blotch or bar between
	2nd and 3rd dorsal spines; in life, head and anterior portion of body mottled
	orange, posterior portion of body mottled vellow; pelvic fins separate
	Coryphopterus alloides
7	Pectoral-fin base with two prominent dark spots of equal intensity, one
	dorsally and one ventrally; upper spot usually with swath of melanophores

extending posteriorly onto pectoral-fin rays; sides of body freckled with scattered large and small blotches of melanophores (blotches associated with Pectoral-fin base with or without two prominent dark spots; if two spots present, upper spot more intense; sides of body with few dark markings (with few to many yellow spots in life) or with three rows of light markings (coral pink/orange in life); pelvic frenum present......9 Total second dorsal-fin rays (spinous plus soft) 10; a dark triangle immediately 8 behind orbit; no dark triangle under front of orbit; basicaudal mark a vertical dumbbell that in life incorporates two large orange spots on the base of the caudal fin; maximum body depth beneath spinous dorsal fin 20-26% SL, least depth of caudal peduncle 13–15% SL, depth range 0–56 m...... Coryphopterus dicrus Total second dorsal-fin rays (spinous plus soft) 10 or 11, usually 11; no dark triangle behind orbit; dark triangle variously developed beneath front part of orbit; basicaudal blotch cross-shaped, the anterior horizontal projection prominent; in life, two large yellow spots on base of caudal fin just posterior to basicaudal blotch; maximum body depth beneath spinous dorsal fin 17-19% SL; least depth of caudal peduncle 9-12% SL; depth range 70-80 m......Coryphopterus curasub sp. n. 9 Pectoral-fin base without prominent dark markings, but may have scattered melanophores; sides of body with few if any dark markings (with yellow to orange spots and stripes in life) except for several dark streaks internally along spinal cord and a thin dark basicaudal bar..... Coryphopterus eidolon Pectoral-fin base with prominent markings; sides of body with or without numerous dark markings .....10 10 Pectoral-fin base with distinct, often large, pigment spot dorsally, spot usually dark above, diffuse below, often with dots trailing ventrally; ventral dots coalescing into a separate spot in some specimens (ventral spot, if present, less intense than dorsal spot); total second dorsal-fin rays (spinous plus soft) 9-10; second dorsal spine filamentous; in life, first dorsal fin without orange stripes ...... Coryphopterus thrix Pectoral-fin base with prominent dark spot or blotch *ventrally*; total second dorsal-fin rays (spinous plus soft) 11; second dorsal spine not filamentous; in life, first dorsal fin with two broad orange stripes ..... .....Coryphopterus punctipectophorus Body usually pale, pigment primarily comprising three rows of markings on 11 side of body; lower row comprising small, mostly vertically elongate markings, some of which may be crescent shaped or some part of an X-shape but rarely well-defined X's; height of any X-shaped markings considerably less than eye diameter; pigment mark above opercle usually a triangle, and basicaudal pigment usually a central bar ...... Coryphopterus tortugae Body heavily pigmented or pale but no vertically elongate or crescent-shaped markings in ventral row of pigment on side of body; height of any X-shaped

# **Discussion and conclusions**

Exploratory submersible diving to 300 m off Curaçao is resulting in the discovery of numerous new fish species, only a few of which have been described to date (Baldwin and Robertson 2013, 2014; Baldwin and Johnson 2014). In addition to the new Coryphopterus described here, numerous seven-spined gobies that represent undescribed species in the Chriolepis/Psilotris/Varicus group have been collected, as have several new species of Lythrypnus-like gobies and a putative new species of Palatagobius. A new genus and species of deep-reef goby, Antilligobius nikkiae Van Tassell, Tornabene, and Colin, was recently described from deep reefs at several localities in the Caribbean (including Curaçao) and Bahamas (Van Tassell et al. 2012). Deep-reef fish faunas in general have been poorly studied globally, and the recent new-species discoveries suggest that our knowledge of the deep-reef gobiid fauna in the southern Caribbean, and likely circumglobally, is far from complete. One question of interest is whether deepreef species generally represent single offshoots of largely shallow-reef clades or form natural evolutionary groups. Antilligobius nikkiae, which inhabits depths of 73-150 m, appears to have its closest relative in shallower water than it inhabits. Rüber et al. (2003) hypothesized that Antilligobius belongs within the monophyletic Microgobius group of the tribe Gobiosomatini, specifically as the sister group of the monophyletic Microgobius. Tornabene et al. (2012) tabulated depth distributions for Microgobius species, which occur from < 1 m to at least 75 m, but most occur at depths < 20 m. The COI data analyzed in this study do not rigorously resolve relationships among Cor*yphopterus* species, but morphological data suggest that *C. curasub* may be most closely related to C. dicrus. Depth ranges of those species (1-56 m for C. dicrus, 70-80 m for C. curasub) do not overlap, suggesting that if they are sister species, depth-mediated speciation may have been involved in their evolution. Additional morphological and genetic analyses of Coryphopterus and other goby genera are in progress in efforts to

investigate patterns of speciation and historical invasions of deep tropical reefs. Filling gaps in our knowledge of deep-reef species diversity is critical to meaningful hypotheses about the evolution of the deep-reef fauna, and we therefore continue to seek funding for exploratory diving aboard the *Curasub*.

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# Appendix

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Links between DNA voucher specimens, GenBank accession numbers, and cytochrome c oxidase subunit I (COI) sequences of *Coryphopterus curasub* sp. n. and *C. venezuelae*.

Catalog Number/DNA Number	GenBank No.	GenSeq Designation
Coryphopterus curasub sp. n.		
USNM 406373, CUR 11373, Holotype	KT020955	Geneseq-1 COI
USNM 430037, CUR 13302, Paratype	KT020957	Genseq-2 COI
USNM 431328, CUR 14003, Paratype	KT020961	Genseq-2 COI
USNM 430019, CUR 13303, Paratype	KT020958	Genseq-2 COI
Coryphopterus venezuelae		
USNM 413804, CUR 12273	KT020959	Genseq-4 COI
USNM 413992, CUR 13008	KT020956	Genseq-4 COI
USNM 430016, CUR 13328	KT020960	Genseq-4 COI

**REVIEW ARTICLE** 



# Taxonomic etymology – in search of inspiration

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#### Abstract

We present a review of the etymology of zoological taxonomic names with emphasis on the most unusual examples. The names were divided into several categories, starting from the most common – given after morphological features – through inspiration from mythology, legends, and classic literature but also from fictional and nonfictional pop-culture characters (e.g., music, movies or cartoons), science, and politics. A separate category includes zoological names created using word-play and figures of speech such as tautonyms, acronyms, anagrams, and palindromes. Our intention was to give an overview of possibilities of how and where taxonomists can find the inspirations that will be consistent with the ICZN rules and generate more detail afterthought about the naming process itself, the meaningful character of naming, as well as the recognition and understanding of names.

#### **Keywords**

ICZN, taxonomy, mythology, pop-culture, word-play, politics

# Introduction

The irresistible desire to classify things has always been an important element of culture and science. In the pre-modern era this resulted in, for example, the creation of a list of angels in Dante's *Divine Comedy* (14th century), or a catalogue of various curiosities in Caspar Schott's *Physica curiosa* (17th century). In *The Infinity of Lists* Eco (2009) provides an exhaustive discussion about various catalogues and registers. In the zoological context, it also led to classifications of life forms that resulted in first taxonomical systems. At the

same time, the creation of lists is inextricably linked with the process of 'naming'. Of the two, Foucault (1994) suggested that "*finding words that will at last name accurately that which has never been named before*" is probably the most difficult task.

According to Foucault (1994) "natural history does not have to establish a system of names based upon representations that are difficult to analyse, but only to derive it from a language that has already been unfolded in the process of description", i.e. names are, in a sense, already accessible or derived through the tradition of zoological nomenclature. The examples described in our paper show the difficulties and problems, but also solutions and reasons for decisions hidden behind the process of choosing names for new taxa. The name may also reveal the circumstances under which a given taxon was named and at the same time shows how a name may lead to completely erroneous assumptions about the characteristic of species.

Zoological nomenclature has evolved over the centuries according to any prevailing official system (poly– or binominal), language used, derivation and inspiration hidden behind the names. The first attempts to unify rules of nomenclature and to make it more regular date back to the end of 19<sup>th</sup> century with, for example, '*Rules for Zoological Nomenclature*' proposed by Strickland (1878) or '*The Merton Rules*' by Allen (1897). However, more decades were needed to establish a consistent and official set of principles. Those rules are now widely accepted by the international scientific community and described in the International Code of Zoological Nomenclature (ICZN 1961). The idea of the Code was best expressed by J. C. Bradley in the preface to the first ICZN edition:

Like all language, zoological nomenclature reflects the history of those who have produced it, and is the result of varying and conflicting practices. Some of our nomenclatural usage has been the result of ignorance, of vanity, obstinate insistence on following individual predilections, much, like that of language in general, of national customs, prides, and prejudices. Ordinary languages grow spontaneously in innumerable directions; but biological nomenclature has to be an exact tool that will convey a precise meaning for persons in all generations.

The regulations of the Code (based on ICZN 1999) concern among others the system of zoological nomenclature, including the alphabet used in naming taxa and derivation of names. According to the ICZN the name should be compact, euphonious, and memorable. This recommendation was given to avoid situations like those of Dybowski (1926), who proposed a series of names for new amphipod species that were over 30 letters long, with the record-holder having 52 letters – *Gammaracanthuskyto-dermogammarus loricatobaicalensis*; these names were later invalidated. Except for these technical and linguistic recommendations, it seems that the Code gives the taxonomist *carte blanche* in creating new names. The main limitation for authors is that the process of naming new taxa should not cause offence. This clause is fully justified as the history of zoological taxonomy includes cases when etymology was used for inappropriate purposes, e.g., the case of Wilhelm Blandowski, whose descriptions of new species of fish
were insulting members of the Philosophical Institute of Victoria who were in conflict with Blandowski (Kean 2005).

Free rein in choosing a name is of significance when describing a large series of taxa. There have been cases when an author simply used ordinal numbers as specific names of new species (Spencer 1969). Many other inspirations are hidden behind the taxonomic names, starting from the most obvious such as general morphology or type locality, through various word-plays (e.g., anagrams or acronyms) and names dedicated to politicians, musicians or comic book characters. Here we make the review of the variety of inspirations for zoological names and discuss some of the most important queries associated with the process of naming and recognition of names.

# Form and function

In the history of taxonomy, the most common animal names are probably those reflecting species morphology, habitat and sometimes even behaviour. Names referring to morphological features can be further divided into several subcategories such as size – one of the largest beetles ever described is *Titanus giganteus* (Linnaeus 1771). A name may also reflect the colour of the animal (the black garden ant *Lasius niger*) or a specific pattern present on the body of the described taxon, like zigzags, dots or stripes (the cowry *Cypraea ziczac* (Linnaeus 1758)).

An inspiration for the name can be a shape of a whole animal e.g., hammer-like bivalves from the genus *Malleus* (Lat. Hammer) (Lamarck 1799); or its appendages, e.g., the tanaidacean *Apseudes batillus* that was named after its wide, shovel-like rostrum (Lat. *batillus* – shovel) (Bamber 2007).

Some of the names describe a species habitat as for the synanthropic spider *Tegenaria domestica* (Lat. *domesticus* – belonging to the house) (Clerck 1757) or in the case of parasites the name of the host e.g., louse *Pediculus humanus* (Linnaeus 1758). Names may reflect life strategies and behaviour, for example the shell-inhabiting tanaidacean *Pagurapseudes inquilinus* from the Latin *inquilinus*, meaning a tenant or lodger (Bamber 2007) and *Macrolabrum mansoris* – from *mansoris* – guest or sojourner (Bamber 2009).

In many cases names provide an appropriate impression of a particular species but there are examples when the name can be misleading or reveals some unusual circumstances that led to the name. One of those unusual cases is that of the New Guinean Greater Bird–of–paradise. Its scientific name given by Linnaeus is *Paradisea apoda* (Linnaeus 1758) and direct translation of its specific Latin epithet means 'legless'. The specimens available for description indeed did not have legs as members of the tribes who collected the specimens had removed the legs during preparation. Lack of this essential information led to further theories that this bird was a visitor from paradise, spending its entire life in the air.

As it was mentioned above many names can literally describe some characters of the species. Many others are a type of metaphor that shows the intuitive leap between the species and the name. Some of those names are clearly anthropomorphic. This attribu-

tion of human form fits into our need to generate assumptions that animals share some of their physical or mental capacities with humans. Inspirations described in this category are often the taxonomist's personal response to the animal. For example the name of the beetle with an unusual head shape *Agathidium akallebregma* (Miller and Wheeler 2005) can be translated as 'ugly face', while the name of the tanaidacean *Pseudoleptochelia anorexia* is associated with its slender morphology (Bird and Bamber 2000). The physical characters might also generate some sort of assumption about mental features. For example the tanaidacean species *Pseudoleptochelia ebriosus* characterized by a presence of red eyes has a name derived from the latin word for drunkard (Bamber and Bird 1997).

### You name the place and I will be there

Knowledge about the distribution of species is very important for the understanding of their biology and ecology. No wonder that there is a series of species named after their type locality or range. The name may point directly the place where the animal was found e.g., the amphipod *Gammarus varsoviensis* collected in the oxbow lake near Polish capital city – Warsaw (Jażdżewski 1975) or a wider geographic area e.g., *Colossendeis fijigrypos* – pycnogonid from Fiji (Bamber 2004). Sometimes the inspiration may also reveal the sense of humor e.g., the name of pycnogonid *Pallenopsis* (*Pallenopsis*) desperado (Bamber 2005) is a pun on the French 'd'Esperance' (of Esperance – the type locality).

# Gods and demons

Since the beginning of the binominal system, zoological nomenclature has been influenced by mythologies and religions. Inspirations from Greek mythology were and probably still are in the lead mostly because of numerous names (mostly butterflies) created by Linnaeus and other pioneers of taxonomy (Tolman and Levington 2004). This situation was most probably associated with the classical education in those days. Linnaeus studied Greek and literature, and mythology of the ancient Greece was the basis of education in many European countries (Huxley 2007). One of the innumerable examples from this category is that of the Little Owl that owes its Latin name - Athene noctua, to the Greek goddess Athena (Scopoli 1769). In this case it is easy to track the association between the species and its mythic archetype as both Athena and owls are symbols of wisdom; moreover the Little Owl was sacred to Athena. A further example is the name of a brittlestar, Gorgonocephalus caputmedusae. The name can be translated as 'head of Gorgon' and 'head of Medusa' (Medusa was one of the gorgons with serpent-like hair) and reflects multifurcate and curled arms present in this species (Linnaeus 1758, Leach 1815). Furthermore, the name of the small nepticulid moth Pectinivalva (Casanovula) minotaurus, a species characterized by expanded antennae, refers to the Minotaur's horns (Hoare and Nieukerken 2013).

Hellenic culture of the ancient Greece has also a great influence on forming of the Judaeo-Christian tradition, which is also present in zoological nomenclature. The Mediterranean bivalve Arca noae (Linnaeus 1758), owes its name to its similarity to the Biblical Ark. The spider name Aptostichus asmodaeus was dedicated to Asmodeus (King of Demons), a fallen angel mentioned in the Book of Tobias, in reference to the type locality, Mount Diablo State Park (Bond 2012). There are also some taxonomic names derived from various versions of the Devils name, starting from the original Hebrew term Satan, through Lucifer, which refers to planet Venus and ending with Mephistopheles, from a German Faust legend. A blind catfish associated with aquifers was named Satan (Hubbs and Bailey 1947), while a nematode that was described as the deepest ever recorded land animal (found in terrestrial deep subsurface of South Africa, 2.2 miles underground) was named Halicephalobus mephisto (Borgonie et al. 2011). The name of Lucifer (Lat. *lux* – light and *ferre* – to carry) is often used as a name for animals that have abilities to produce light e.g., the dragonfish Lucifer (Döderlein 1882) but also in more direct sense, like in case of tanaidacean Pakistanapseudes lucifer (Błażewicz-Paszkowycz and Bamber 2012) which was named after the Devil, owing to its bifurcated claws on pereopods.

Other beliefs are represented in names mainly from Scandinavian mythology, e.g., some common Eurasian butterflies are named after Thor and Frigga – Thor's fritillary (*Boloria thore*) and Frigga's fritillary (*Boloria frigga*) respectively (Hübner 1803). One of the most recognizable examples of names inspired by religion from the Indian subcontinent is *Stegodon ganesa*, a Pliocene mammal closely related to extant elephants. The species was found in India and its specific name regards to mythical Ganesa, one of the most important gods from Hindu pantheon, usually shown with the head of elephant (Saegusa 1987).

#### **Monsters and cryptids**

Legendary or mythical animals can be found in civilizations from various regions of the world and taxonomists find inspirations from them and the related field of cryptozoology. One of the best known is the myth of the Yeti – a mysterious anthropoid cryptid inhabiting the Himalayan Mountains; a carabid beetle was named *Agra yeti* (Erwin 1982) because of its big feet.

Monsters that suck blood and the vital power from people are known in many legends and myths. Some of these demons sucked vital powers from men, and one of the oldest, Empusa from Ancient Greece, lent its name to a praying mantis *Empusa* (Illiger 1798). This myth 'evolved' to more modern versions, including the current cultural obsession with zombies and vampires. Three genera of South-American bats were named *Vampyrodes, Vampyrops, Vampyressa* (Thomas 1889, 1900). A deep-sea cephalopod, *Vampyroteuthis infernalis*, is even more terrifying, because its name means 'vampire squid from hell' (Chun 1910).

# The first muse

Acede was the first original Boeotian muse, a muse of songs and voice, and we can say that music has inspired humans since the ancient times. Scientists have often declared their enthusiasm for music, both as listeners and performers. Albert Einstein said that "life without playing music is inconceivable" and he often declared that music was an undeniable help in his work. He is not alone, with a paper concerning amphipod reproductive traits (Grabowski et al. 2014) being dedicated to the Norwegian duet Ylvis. The authors wrote in the acknowledgements that Ylvis "songs and videos kept us in a continuous good mood when analysing the data and writing the manuscript". It is unsurprising that many taxa have been named after famous musicians, bands or even music styles and genres, but it is remarkable that these names represent so diverse range of inspirations, from well known pop classics (The Beatles – polychaete worm Bushiella beatlesi, Michael Jackson – fossil paguroid Mesoparapylocheles michaeljacksoni) (Rzhavsky 1993, Fraaije et al. 2012) through major progressive rock bands (Pink Floyd - spider genus *Pinkfloydia*) (Dimitrov and Hormiga 2011), jazz icons (Miles Davis - trilobite *Milesdavis*) (Liberman 1994) and local folk ensembles (Argentinean band Los Chalchaleros - caviomorph rodent Salinoctomys loschalchalerosorum) (Mares et al. 2000). Not excluded are classical music (Ludwig van Beethoven - isopod crustacean Gnathia beethoveni) (Paul and Menzies 1971) and obscure avant-garde genres known to only small groups of enthusiasts (e.g., Dutch post-punk group The Ex – gastropod Depressizona exorum) (Geiger 2003).

Most of these names explicitly refer to the person or group; however some of the linkages between music and the morphology or biology of a new taxon result in intriguing names. The generic name of the ichneumonid wasp *Metallichneumon neurospastarchus* refers to the heavy metal band Metallica. This wasp parasitizes sphingid moth caterpillars and *neurospastarchus* derives from the Greek words *neurospasta* (puppet) and *archos* (ruler), linking the famous Metallica album 'Master of Puppets' with "*mindless nature of lepidopterous larvae*" (Sime and Wahl 2002).

# **Reading and writing**

Writing is an indispensable element of scientific work. If done well, it provides the most effective method of communication with other scientists. Yet, according to a well-known reporter Ryszard Kapuściński (2008) to write one single page one probably needs to read at least 100 pages first. In consequence, all one's previous reading (including favourite popular fiction) has a great influence on writing style, both in life and in scientific texts. Taxonomic inspirations associated with book titles, famous authors and fictional characters from literature are numerous amongst taxonomic eponyms. It is worth mentioning that some writers were at the same time professional scientists. Vladimir Nabokov is famous for his novel "Lolita", however the name of the lycaenid butterfly *Nabokovia* (Hemming 1960) was dedicated to him not because of

his writing, but due to his passion for taxonomic studies on butterflies. Nabokov was a curator of the lepidopteran collection at Harvard University's Museum of Comparative Zoology in the 1940s and he even published articles concerning this subject (e.g., Nabokov 1944). Many names of butterflies are derived from the main protagonists in his novels, such as *Madeleinea lolita* (Balint 1993).

In the early twentieth century the American entomologist Alexandre Arsène Girault dedicated some of the names of the wasps he studied to influential writers such as William Shakespeare (*Shakespearia*) (Girault 1928), Johann Wolfgang von Goethe (*Goetheana*) (Girault 1920) and Henry Wadsworth Longfellow (*Tineobius longfellowi*) (Girault 1923). In the years since then, the names of writers and books used in taxonomy have changed notably.

Many names have been created in honour of the British author of fantasy novels, Terry Pratchett, especially after the characters from his Discworld series. Roger Bamber was especially fond of these books and named 30 species of 'Discworld' tanaidaceans, three of which are *Apseudes atuini* – derived from Great A'Tuin, the giant space turtle on whose back four elephants hold up the Discworld, *Tanaella dongo* – Crocodile Dongo, a creature that runs a pub in the town of Dijabringabeeralong, on the Last Continent, and *Bathytanais greebo* – Nanny Ogg's cat Greebo (Bamber 2005). There are also many species named after characters created by J. R. Tolkien in his trilogy *Lord of The Rings*, including the tanaidacean, *Gollumudes*, from Gollum who dwelt in damp muddy caves (Bamber 2000), and a beetle, *Pericompsus bilbo*, whose hairy feet refer to the hobbit Bilbo Baggins (Erwin 1974). New bestselling classics, like J. K. Rowling's *Harry Potter* series of novels, inspired the names such as the dinosaur *Dracorex hogwartsia* (Bakker et al. 2006). We can expect that, with the appearance of new 'bestsellers', the number of new taxa with names derived from literature will continue to increase.

### The big screen

Actors, directors and movie characters have been an inspiration for some taxonomists. Former governor of California and actor, Arnold Schwarzenegger, was commemorated in the name *Agra schwarzeneggeri* since males of this species have thickened thighs, which are similar to his well-muscled limbs (Erwin 2002). The name *Agra liv* was dedicated to the Hollywood actresses Liv Tyler. As described by the author, she survived the impending apocalypse in the Armageddon movie, but devastation of tropical rainforests may also lead to an 'Armageddon' for this small beetle (Erwin 2002). People from other side of the camera also have been commemorated in taxonomy. Director Steven Spielberg was honoured for his three Jurassic Park movies with a pterosaur species named *Coloborhynchus spielbergi* (Veldmeijer 2003).

Movie characters, rather than the actors themselves, sooner or later feature in zoological names. Terminator was a deadly robot from the future and authors who described the African spider *Hortipes terminator* found similarities in the appearance of its pedipalps to Terminator's weapon (Bosselaers and Jocqué 2000). Batman, masked defender of Gotham City, is a pop culture icon. The name of the North-American fish *Otocinclus batmani* (Lehmann 2006) is associated with a black spot on its caudal fin, the shape being similar to the image generated by the searchlight used by Commissar Gordon to summon Batman. Perhaps inevitably, the popular movie series *Star Wars* by George Lucas was also an inspiration for many taxonomists. Master Yoda's ears were so similar to appendages on the sides of the head of the small parasitic isopod *Albunione yoda*, that authors gave its name after this Jedi (Markham and Boyko 2003).

Some characters from cartoons have also inspired taxonomists. According to the authors, the male aedeagus of a beetle *Adelopsis dumbo* look like the large and floppy ears of the elephant Dumbo (Gnaspini and Peck 2001).

#### From Aristotle to Darwin

Many species and genera have been named in honour of scientists. These have appeared since the time of the 'Father of modern taxonomy' Carolus Linnaeus. He named the Common Shag *Phalacrocorax aristotelis* (Linnaeus 1958) to honour Aristotle. The list of scientists appreciated by their colleagues and followers is very long, and some of them were honoured many times. We can even say that the number of taxa named after a researcher can be considered a signifier of their importance in the history of science. Pioneers of taxonomy Linnaeus and Lamarck were commemorated in many taxonomic names, e.g., the snake *Calamaria linnaei* and polychaete worm *Pomatoceros lamarckii* (Boie 1827, Quatrefages 1866).

Nevertheless, judging from the zoological nomenclature Charles Darwin has been probably the most inspiring and influential scientist. There are over 300 taxa from nine phyla named after him (Miličić et al. 2011). These include over 250 species, many genera (e.g., ostracod *Darwinula* or hemipteran *Darwinysius*) but also some higher ranks like the ostracod Infraorder Darwinulocopina (Sohn 1988). His work "On the Origin of Species" became the fundamental text for biologists but it was also highly contested and criticized, mostly because of the lack of the true understanding of his ideas. So, as we might expect, a person who has contributed much to improve the understanding of evolutionary biology would also be memorialized with a taxonomic name. A cyprinid fish (*Dawkinsia*) was named after Richard Dawkins, author of the popular science books "The Selfish Gene" and "The Blind Watchmaker", for "*his contribution to the public understanding of science and, in particular evolutionary science*" (Pethiyagoda et al. 2012).

# Politicians and the powerful

Politicians always have great influence on peoples' lives for good or bad. A large group of scientific names commemorates former presidents of the United States, including

Abraham Lincoln (the parasitic wasp *Lincolna* (Girault 1940)) and Franklin Delano Roosevelt (the amphipod *Neomegamphopus roosevelti* (Shoemaker 1942)). From more recent times, George W. Bush, Vice President Dick Cheney, and Secretary of Defense Donald Rumsfeld were honoured in the names of Leiodidae beetles: *Agathidium bushi*, *A. cheneyi*, and *A. rumsfeldi* (Miller and Wheeler 2005).

The emperors of Japan were treated with great dignity and respect, and were even considered deities. Hirohito, the longest-reigning emperor (1926–1989) was also a marine biologist and inspired the name of a sea gastropod *Rotaovula hirohitoi* (Cate 1973). A beetle, *Aegomorphus wojtylai*, was recently dedicated to a former head of the Catholic Church, Karol Wojtyła (Pope John Paul II) (Hilszczański and Bystrowski 2005).

There is clearly a negative aspect to "political taxonomy". The German entomologist Oscar Scheibel (1937) honoured the leader of the National Socialist German Workers' Party, Adolf Hitler, by describing *Anophthalmus hitleri* – a species of carabid beetle from the Balkans. This endemic insect occurs only in five caves in Slovenia, and is now a victim of Hitler's notoriety as specimens are being illegally collected as 'memorabilia' of the Third Reich. The restricted range and very specific habitat of this endemic beetle has pushed it to the brink of extinction.

#### **Brands and sponsors**

Some of the most widely known international brands, or companies, are reflected in taxonomic names. Probably the world's best known drink manufacturer, Coca–Cola©, was included in the name of the wasp *Oxybelus cocacolae* (Verhoeff 1968), although more subtle links between companies' and species' names have been made. A species of ant from Madagascar, *Proceratium google*, named in recognition of Google©, has a great "*ability to hunt down obscure prey* [Sic!]" (Fischer 2005).

Financial and logistic support to scientific expeditions are also rewarded in the names given to new species: a team of palaeontologists found and described a new Palaeocene mammal, *Roberthoffstetteria nationalgeographica*, (Goin et al. 2003) during an expedition supported by the publisher of the National Geographic magazine. Conservation efforts can be an outcome of cooperation between business and nature-protection agencies: a model example happened in Bolivia where the online casino GoldenPalace.com paid hundreds of thousands of dollars for the rights to name the new species of primate, *Callicebus aureipalatii* (Lat. *aureus* – golden and *palatium* – palace), in the Madidi National Park. Moreover, the company decided to pay an annual subsidy for this national park (Wallace et al. 2006).

# **Playing with words**

In the introduction we said that taxonomists have (almost) free rein in choosing the name for new taxa and this no-restriction policy is most evident in this category. It includes various types of word plays, figures of speech and rhetorical devices. In most cases it is hard to say that there is any significant 'inspiration' hidden behind these names. Some of the examples from this category might be called controversial and may even bend the rules of the ICZN.

Tautonyms are names where the specific epithet is repeated after the genus. They were widely used for, and often characterize, common European animals such as the Roe Deer – *Capreolus capreolus*, or Fox – *Vulpes vulpes* (Linnaeus 1758). This tautonymy has a certain euphonious effect, in contrast to some true 'tongue twisters' such as the nematode *Xyzzors* (Inglis 1966) or snail, *Zyzzyxdonta* (Solem 1976).

An early example of wordplay is the taxonomic work of English marine biologist Elford Leach. In his 1818 monograph he described a series of isopod genera that were anagrams of 'Caroline' or 'Carolina': *Anilocra, Cirolana, Conilera, Lironeca, Nerocila, Olencira*, and *Rocinela* (Leach 1818). Later this idea was picked up by Hansen (1890) who described *Alcirona* and *Lanocira*, and Nierstrasz (1931) who described *Orcilana*.

Palindromes, i.e. words that can be read the same forward and backward, are not so common in zoological nomenclature and are usually applied to genus names e.g., the bethylid hymenopteran *Afgoiogfa* (Argaman 1988), and more rarely in species names, as for the syrphid fly *Xela alex* (Thompson 1999).

Acronyms are often used to honour an institution or project that was involved in collecting the material or financially supported the taxonomic studies e.g., *Pseudotanais soja* (Błażewicz et al. 2013) a tanaidacean species collected during expedition of SoJaBio (Sea of Japan Biodiversity Studies). Błażewicz-Paszkowycz and Bamber (2013) named tanaidacean *Acinoproskelos vermes* after Latin plural for "worms", being both a reference to the parasitic nematodes within the type specimen, and also the acronym of the World Register for Marine Species (WoRMS). There is, however, an example of an acronym–name that probably appeals to those taxonomists spending a lot of time in the laboratory – *Afropolonia tgifi* (Thank God it's Friday) (Goff 1983).

Finding good names when a large series of taxa has to be described can be time consuming and various 'tricks' are sometimes used to facilitate this. Taxonomists might use the same word stem and supplement it with a different prefix or suffix. This was used by the British, American entomologist Kirkaldy (1904), who proposed numerous hemipteran names with the Greek stem '-chisme' including: *Marichisme, Peggichisme,* and *Ochisme.* At first glance this series is unremarkable but phonetically in English it results in the creation of short phrases: 'Mary kiss me!', 'Peggy kiss me' and 'O kiss me!'. By a decision of the Zoological Society of London, Kirkaldy was criticized in 1912 for this frivolity but it seems that it has not discouraged other taxonomists from similar usage, as Evenhuis (2002) described a fossil fly in the genus *Carmenelectra* (named after the singer, actress and model) with the specific name 'shechisme'.

Another method for naming dozens of species was used by Kearfott (1907) who described a series of new moths from the genus *Eucosma* with names created in the same way distinguished only in their consonants: e.g., *E. bobana, E. cocana, E. dodana* 

or *E. fofana*. Spencer (1969) described several agromyzid flies from the genus *Ophiomyia* using ordinal numbers for the specific names: *O. prima*, *O. secunda*, *O. tertia* and so on. Riedel et al. (2013) also did not have easy task as he needed to find names for 101 species of curculionid beetles. In this case, the Papua New Guinea Telephone Directory, appeared to be helpful and Riedel use the names of people found there as the stem with the Latin suffix '-orum' e.g., *Trigonopterus hitoloorum*, *T. kanawiorum*, *T. koveorum* or *T. lekiorum*.

There are many names that are created from Latin words that are sometimes meaningful taking into account the taxonomic context e.g., *Sphenoptera incerta* (unsure) (Jakovlev 1887) or *Leptura dubia* (Scopoli 1763) (doubtful). Names may also present an unusual combination of words as in the African cicada, *Imbecilla cretinica* (Dworakowska 1974).

# **Concluding remarks**

While names are often derived from the attributes of a species (e.g., its morphology or behaviour) it may also be a result of subjective state of taxonomist's mind at the time of naming, including those who were assisted by the music of a certain band or composer. Sometimes a curious name is used simply to get other people interested in a given taxon, but more often than not it has some stronger basis. Nevertheless, it is generally very difficult to analyse all types of taxonomic inspiration since we have to follow only the information provided in the etymology section of cited papers, and we do not wish speculate about the taxonomist inspirations even if the name generates some questions about the author's decision.

Continuous findings of new taxa results in a obvious necessity of naming them. The name is very important because it allows to communicate about a given species with other scientists. Therefore, it seems that it is also important to ask what we can learn from a name? This function of the name may show a need for accuracy in scientific naming. Some people could say that the name itself should be as informative as possible, and give us also some basic information about the species. Taxonomist may use a very simple words to describe the taxon or try to be more subtle in their inspirations like for example link the species morphology with description of a mythological creature or god (Winston 1999).

On the other hand naming process reflects also fads and trends present in the modern world. Those fads are present in all languages and it is not surprising that are also visible in taxonomy. There are numerous names that are very attractive but cannot give us any meaningful message, that is significant from the biological point of view. At the same time the name may educate us about culture, history or politics, however it is important to ask if this is really the function of taxonomic eponym? It is probably not, nevertheless, it is worth mentioning that naming of large series of morphologically similar new species that we know almost nothing about often do not allow to maintain the informative function of the names.

Naming process may also result in problems of ethical nature. For example the name of the sponsor, company or a certain politician commemorated in a taxonomic name may caused (often not intended) questions associated with the policy of those companies or accusations of showing allegiance. However, one could say also that if the sponsoring allows for a development of knowledge it is still consistent with the ethical standards. In some cases the name can be also a political statement because according to the etymology sections taxonomist approve the politics of presidents and ministers which are mentioned in the taxonomic names. On the other hand it is worth to remember that it may result in completely opposite reactions of other researchers. Moreover, we cannot be sure that we will approve also all future decisions of the active politician. Finally we may also consider the fact that taxonomic names may also completely lose their meaning, like in case of *A. wojtylai* which was synonymised just three years after description (Hilszczański 2008).

Analysis of species names raises questions about our understanding of names. Some names may lose impact or 'recognition' over the years. We do not know if we will be able to comprehend the inspiration associated with the Star Wars saga 200 years from now, and knowledge of music bands like The Beatles or authors like Longfellow might not be present among many biology students today. Nevertheless, taxonomic names persist and will still be used by other taxonomists even if their etymology will not be clear for everyone or becomes outdated. Taking these cultural changes into account it is interesting to ask if it is appropriate that scientists should have such a wide choice of names. We did not intend to find the final answer to this question here, but propose that each taxonomist should consider it during their own work. We can only repeat after Brown (1956) that invention of names is an art and simplicity is often the most appropriate choice.

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This paper was planned with Roger Bamber as co–author as we unanimously agreed that his great experience in field of zoology, taxonomical nomenclature, ICZN rules and a subtle sense of humour were the right attributes for this issue. Shortly after preliminary arrangements had been made for the structure of the manuscript, our plans had to be changed as we were informed about Roger's illness. Our intention was to finish the paper with a dedication to our friend Roger, who undoubtedly enjoyed unusual etymology for taxonomic names and could as no other connect modern taxonomy with Prachett's *Discworld*, Tolkien's trilogy or McCulley's *Zorro*. Closer look in this paper reveals some of Roger's taxonomical inspirations.

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