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Cyphastrea kausti sp. n. (Cnidaria, Anthozoa, Scleractinia), a new species of reef coral from the Red Sea

Jessica Bouwmeester[1], Francesca Benzoni[2], Andrew H. Baird[3], Michael L. Berumen[1]

1 *Red Sea Research Center, King Abdullah University of Science and Technology (KAUST), Thuwal, 23955-6900, Kingdom of Saudi Arabia* **2** *Department of Biotechnology and Biosciences, University of Milano-Bicocca, Piazza della Scienza 2, Milan, Italy* **3** *ARC Centre of Excellence for Coral Reef Studies, James Cook University, Townsville, QLD 4811, Australia*

Corresponding author: *Jessica Bouwmeester* (jessica.bouwmeester@kaust.edu.sa)

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Abstract

A new scleractinian coral species, *Cyphastrea kausti* **sp. n.**, is described from 13 specimens from the Red Sea. It is characterised by the presence of eight primary septa, unlike the other species of the genus, which have six, ten or 12 primary septa. The new species has morphological affinities with *Cyphastrea microphthalma*, from which it can be distinguished by the lower number of septa (on average eight instead of ten), and smaller calices and corallites. This species was observed in the northern and central Red Sea and appears to be absent from the southern Red Sea.

Keywords

Merulinidae, Saudi Arabia, biodiversity, coral reef, taxonomy, KAUST

Introduction

The genus *Cyphastrea* Milne Edwards & Haime, 1848 has an Indo-West Pacific distribution range, from the Western Indian Ocean to the Central Pacific, and contains 19 nominal species, nine of which are considered valid (Hoeksema 2014). The genus was previously included in the family Faviidae Gregory, 1900, but recent molecular work has heavily re-organised the classification of the major scleractinian families and

Cyphastrea is now placed in the Merulinidae Verrill, 1865 (Fukami et al. 2008, Budd et al. 2012, Huang et al. 2014).

In his revision of the genus, based on macro-morphological characters of the skeleton and the tissue of the coral polyps, Matthai (1914) recognised five species, i.e., *C. microphthalma* (Lamarck, 1816), *C. serailia* (Forskål, 1775), *C. chalcidicum* (Forskål, 1775), *C. gardineri* (Matthai, 1914), and *C. suvadivae* (Gardiner, 1904). However, the last three were later declared junior synonyms of *C. serailia* by Wells (1954) and Chevalier (1975). Veron et al. (1977) then reinstated *C. chalcidicum* and declared *C. gardineri* as junior synonym of *C. microphthalma* rather than *C. serailia*.

More recent regional work on the taxonomy, biodiversity, and distribution of scleractinian corals in the Red Sea originally recognised two *Cyphastrea* species in the region, *C. microphthalma* and *C. serailia*, synonymising again *C. chalcidicum* with *C. serailia* (Scheer and Pillai 1983, Sheppard and Sheppard 1991). However, DeVantier et al. (2000) and Veron (2000, 2002) subsequently reinstated *C. chalcidicum* and described a new species from the northern Red Sea, *C. hexasepta* Veron et al. 2000, leading to four valid species of *Cyphastrea* in the Red Sea: *C. microphthalma*, *C. serailia*, *C. chalcidicum*, and *C. hexasepta*.

The genus *Cyphastrea* has recently been formally revised within the Merulinidae, incorporating molecular phylogeny, macromorphology, micromorphology, and microstructure (Huang et al. 2014) but it has yet to be revised at the species level. Indeed, the species boundaries within the genus have not yet been investigated using an integrated systematic approach, such as for example in the genera *Plesiastrea* (Benzoni et al. 2011), *Blastomussa* (Benzoni et al. 2014), *Australomussa*, and *Parascolymia* (Arrigoni et al. 2014).

The genus *Cyphastrea* is characterised by its compact coenosteum (Milne Edwards and Haime 1848). The genus is composed of colonial species, with only extracalicular budding. The corallites are monomorphic, monticules are absent, and the coenosteum is generally spinose. Calice width is smaller than 4 mm, septa are in three cycles or less, costosepta are not confluent, and are unequal in relative thickness. The columella is trabecular and compact, and paliform lobes are weak or moderate (Huang et al. 2014). Species in this genus are commonly described based on the septal arrangement (Wijsman-Best 1980, Sheppard and Sheppard 1991, Veron 2000). Among the species currently recognised in the Red Sea, *C. chalcidicum* and *C. serailia* both have two cycles of six septa that are identical in *C. serailia* and alternating in *C. chalcidicum*, *C. hexasepta* has six primary septa, and *C. microphthalma* has ten primary septa (Veron 2000). Here we describe a new species that resembles *C. microphthalma* in the field but has eight septa: *C. kausti*, sp. n. first observed in 2011 in Al Fahal Reef, offshore from the King Abdullah University of Science and Technology, Thuwal, Saudi Arabia.

Methods

Colonies of *Cyphastrea kausti* sp. n. (13 in total) were sampled on SCUBA in 2013 during several expeditions along the coast of the Saudi Arabian Red Sea, from Magna

Figure 1. Map of the Red Sea showing the sampling sites of *Cyphastrea kausti* sp. n. The red dot indicates the locality of the holotype, the dark red dot indicates the locality of the paratype, and the green dots indicate the localities of the remaining sampled material.

in the Gulf of Aqaba in the north to the Farasan Islands in the south (Fig. 1). Digital images of living corals were taken in the field with a Sony DSC-W80 camera and Sony MPK-WB underwater housing and the depth recorded with a dive computer, when possible. Coral specimens were collected with hammer and chisel and tagged. From each colony, a small fragment was subsampled and preserved in absolute ethanol for molecular analysis. The remaining corallum was placed for 24 hours in sodium hypochlorite to remove all coral tissue, rinsed in fresh water, and dried for microscopic

Table 1. Micromorphologic characters of *Cyphastrea kausti* sp. n. compared to *C. microphthalma*. Average number of septa for seven randomly selected corallites, average calice and corallite width for five randomly selected corallites, and number of corallite centres visible in 1 cm^2. Standard deviation is indicated for averages.

	***Cyphastrea kausti* sp. n.**		***Cyphastrea microphthalma***	
aveage number septa	MNHN-IK-2012-14236*	8.0 ± 0.3	MNHN-IK-2012-14002*	9.9 ± 0.4
	SA607	8.1 ± 0.4	SA159	9.7 ± 0.8
	SA1121	8.1 ± 0.7	SA552	9.6 ± 0.5
	SA1103	7.7 ± 0.8	SA734	9.9 ± 0.4
			SA100	9.9 ± 0.4
	Average:	**8.0 ± 0.4**	**Average:**	**9.8 ± 0.5**
average calice diameter [mm]	MNHN-IK-2012-14236*	1.11 ± 0.06	MNHN-IK-2012-14002*	1.19 ± 0.09
	SA607	1.04 ± 0.03	SA159	1.37 ± 0.05
	SA1121	0.83 ± 0.12	SA552	1.34 ± 0.10
	SA1103	1.05 ± 0.03	SA734	1.28 ± 0.09
			SA100	1.19 ± 0.09
	Average:	**1.01 ± 0.13**	**Average:**	**1.27 ± 0.11**
average corallite diameter [mm]	MNHN-IK-2012-14236*	1.77 ± 0.14	MNHN-IK-2012-14002*	1.82 ± 0.12
	SA607	1.61 ± 0.09	SA159	2.14 ± 0.11
	SA1121	1.36 ± 0.11	SA552	2.12 ± 0.17
	SA1103	1.73 ± 0.07	SA734	2.09 ± 0.05
			SA100	1.81 ± 0.08
	Average:	**1.62 ± 0.19**	**Average:**	**2.01 ± 0.18**
number corallites per cm^2	MNHN-IK-2012-14236*	19	MNHN-IK-2012-14002*	33
	SA607	11	SA159	19
	SA1121	31	SA552	11
	SA1103	19	SA734	11
			SA100	19
	Average:	**20 ± 8.2**	**Average:**	**15 ± 4.6**

* holotype

observation. The cleaned skeletons were then photographed with a Canon G9 digital camera. Macro and micromorphological characters were examined using light microscopy (Zeiss Stemi 2000 dissecting microscope) and scanning electron microscopy (SEM), respectively. For SEM, a small fragment of clean skeleton was ground at the base with sandpaper, mounted on a stub using double-sided carbon tape, sputter-coated with a 3 nm layer of conductive gold-palladium (AuPd) film, and examined using a Quanta 200 FEG SEM at the KAUST Imaging & Characterization Core Lab. Samples of *C. kausti* sp. n. were compared to samples of the other species present in the Red Sea, which were located at the KAUST Biodiversity collection. Specimens of *C. kausti* sp. n. were morphologically compared with those of *C. microphthalma*, morphologically the most closely resembling species based on *in situ* observations and skeletal examination. The morphometric characters (Table 1) were determined post-imaging with a digital measurement analysis tool calibrated on the image scale bar in Adobe Photoshop CS3. The holotype and a paratype of *Cyphastrea kausti* sp. n. were deposited at the National Museum of Natural History (MNHN), Paris, France. All other material is located at the King Abdullah University of Science and Technology (KAUST), as part of its Red Sea Biodiversity collection. The holotype of *C. microphthalma* was examined from images taken by A.F. Budd, available online at http://www.corallosphere.org.

Abbreviations

KAUST	King Abdullah University of Science and Technology, Thuwal, Saudi Arabia
MNHN	National Museum of Natural History, Paris, France

Taxonomic account

Family Merulinidae Verrill, 1865

Genus *Cyphastrea* Milne Edwards & Haime, 1848

Astrea (pars) Lamarck, 1816, not *Astrea* Lamarck, 1801
Cyphastrea Milne Edwards & Haime, 1848
Solenastrea (pars) Milne Edwards & Haime, 1850, not *Solenastrea* Milne Edwards & Haime, 1848

Type species. *Astrea microphthalma* Lamarck, 1816; original designation, Milne Edwards & Haime, 1848

Cyphastrea kausti Bouwmeester & Benzoni, sp. n.

http://zoobank.org/39E6E02E-B176-4ADB-9175-0E8C29C8D74C
Figures 2, 3a, c, e, 4

Material examined. Type material. *Holotype:* MNHN-IK-2012-14236 (KAUST SA1307). Type locality: Fsar (Thuwal), N 22°13.78', E 39°01.73', depth 13.6 m, coll. J. Bouwmeester 20/10/2013.

Paratype: MNHN-IK-2012-14237 (KAUST SA522). Sodfa (Thuwal), N 22°12.07', E 38°57.52', depth 2.0 m, coll. D. Huang, 24/04/2013.

Other material (Red Sea, Saudi Arabia). SA443 Qita al Kirsh (Thuwal), N 22°25.60', E 38°59.77', coll. F. Benzoni, 18/03/2013; SA446 Qita al Kirsh (Thuwal), N 22°25.60', E 38°59.77', coll. F. Benzoni , 18/03/2013; SA498 Sodfa (Thuwal), N 22°12.07', E 38°57.52', depth 10.4 m, coll. D. Huang, 24/04/2013; SA607 Abu Madafi (Thuwal), N 22° 3.73', E 38°45.82', depth 6.1 m, coll. J. Bouwmeester, 28/04/2013; SA643 Tahla (Thuwal), N 22°17.04', E 39° 3.10', depth 6-12 m, coll.

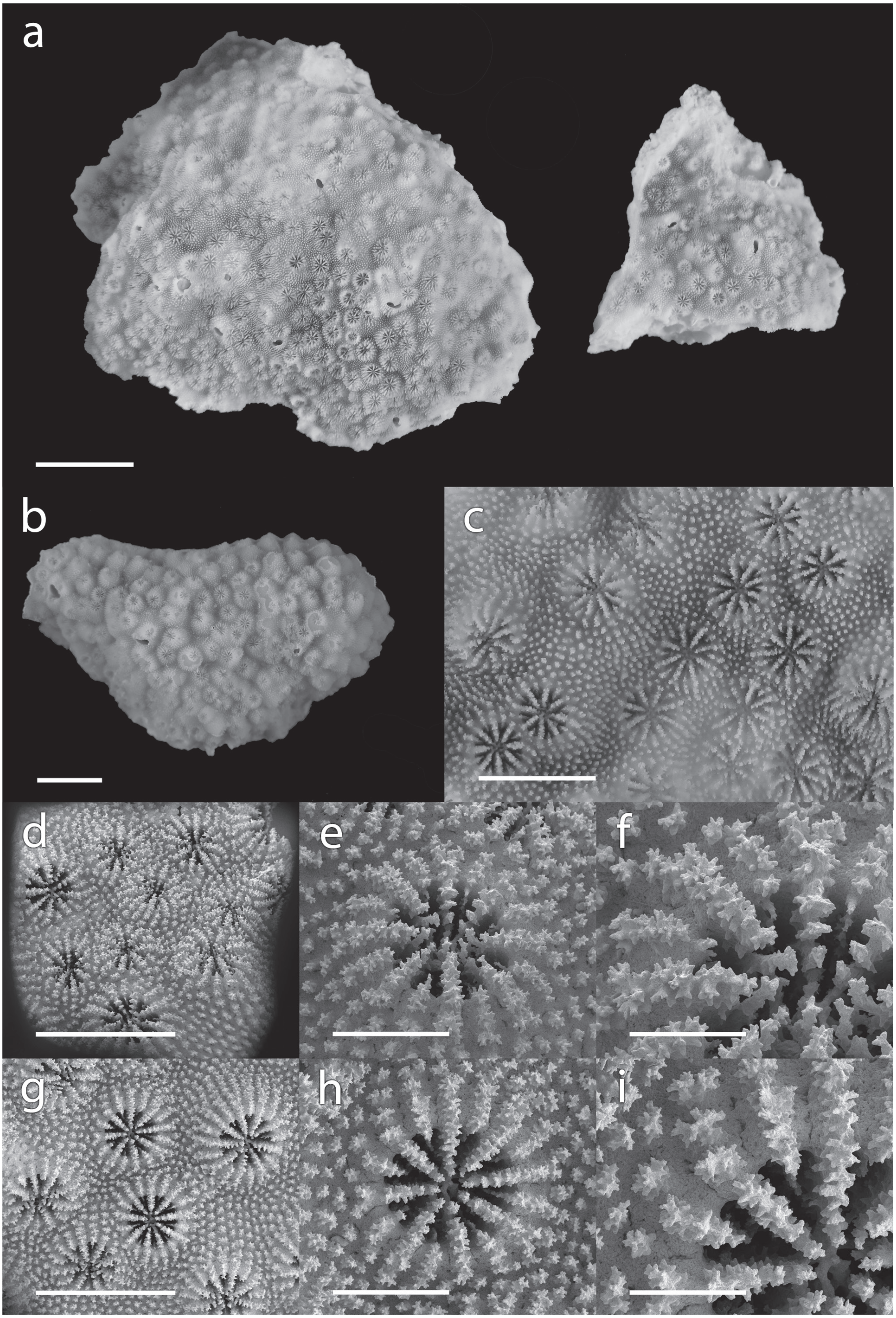

Figure 2. *Cyphastrea kausti* sp. n. **a** holotype, two fragments (MNHN-IK-2012-14236) **b** paratype (MNHN-IK-2012-14237) **c** skeletal detail of holotype (MNHN-IK-2012-14236) **d–f** SEM images of SA1121 **g–i** SEM images of SA1103. Scale bars: 10 mm (**a, b**); 3 mm (**c, d, g**); 1 mm (**e, h**); 500 µm (**f, i**).

J. Bouwmeester, 08/07/2013; SA644 Tahla (Thuwal), N 22°17.04', E 39° 3.10', depth 6-12 m, coll. J. Bouwmeester , 08/07/2013; SA973 Magna (Gulf of Aqaba), N 28°24.23', E 34°44.44', coll. F. Benzoni, 29/09/2013; SA1103 Shaybarah (Al Wajh), N 25°21.69', E 36°54.75', coll. F. Benzoni, 03/10/2013; SA1121 Marker 9 (Yanbu), N 24°26.56', E 37°14.86', coll. F. Benzoni, 04/10/2013; SA1165 Marker 9 (Yanbu), N 24°26.56', E 37°14.86', depth 12.5 m, coll. J. Bouwmeester, 04/10/2013; SAE015 Fsar (Thuwal), N 22°13.78', E 39°01.73', depth 12m, coll. J. Bouwmeester, 21/09/2014.

Description of holotype. The holotype is part of a 12 cm high and 17 cm wide encrusting colony living on an inclined surface (Figure 4a–b), and is constituted of two fragments sampled from a single colony (Figure 2a). The first fragment is the bigger of the two, bell-shaped, 4.5 cm high and 3.8 cm wide (Figure 2a, left). The second fragment is smaller, triangular-shaped, 2.5 cm high with a 2.5 cm base (Figure 2a, right). The number of septa is eight but in a small number of corallites (4/40), the number of septa is seven or nine. The septa are exsert and carry densely ornamented spines. The costae are composed of a line of ornamented spines, which continue on the coenosteum, adding to its already dense and elaborate arrangement of ornamented spines. The columella is trabecular and surrounded by a crown of paliform lobes (Figure 2c). Extra-calicular budding can be observed on both fragments (Figure 2a). The calice diameter of the corallite is 1.11 ± 0.06 mm and the corallite diameter is 1.77 ± 0.14 mm. The corallite density varies from 13 to 22 corallites per cm^2 (Figure 2c).

Diagnosis. The number of primary septa in *Cyphastrea kausti* sp. n. is typically eight (Figures 2e, h, 3e) but in some cases, seven, nine, or even ten septa can be observed in some corallites of the same colony. However, the majority of corallites have eight primary septa and the average number of septa throughout examined samples is 8.0 ± 0.4 (61 corallites examined from four specimens). This character distinguishes it from *C. microphthalma*, which on average has 9.8 ± 0.5 primary septa per corallite, although in one case a corallite with eight septa was observed (Table 1).

In *C. kausti* sp. n. the calice diameter of the corallite is 1.01 ± 0.13 mm and the corallite diameter is 1.62 ± 0.19 mm. This is smaller than in *C. microphthalma*, which has a calice diameter of 1.27 ± 0.11 mm and a corallite diameter of 2.01 ± 0.18 mm (Table 1).

The corallite density is highly variable between colonies of *Cyphastrea kausti* sp. n. but also within a colony, and varies from 11 to 31 corallites per cm^2 (e.g. Figures 2c, e, 3c). The corallite density is generally lower on convex surfaces, and higher on concave surfaces, but remains highly variable between colonies, and is not dependant on depth. Indeed, SA607, which had the lowest number of corallites per cm^2, was sampled at 6 m depth, shallower than the other colonies examined here. In a similar way, corallite density is also highly variable in *C. microphthalma* and varies from 11 to 33 corallites per cm^2.

The first order septa are clearly exsert, the second order septa are weak and never reach the columella, and the third cycle of septa is absent (Figures 2, 3a, c, e). A distinct crown of eight ornamented paliform lobes, corresponding to the number of first

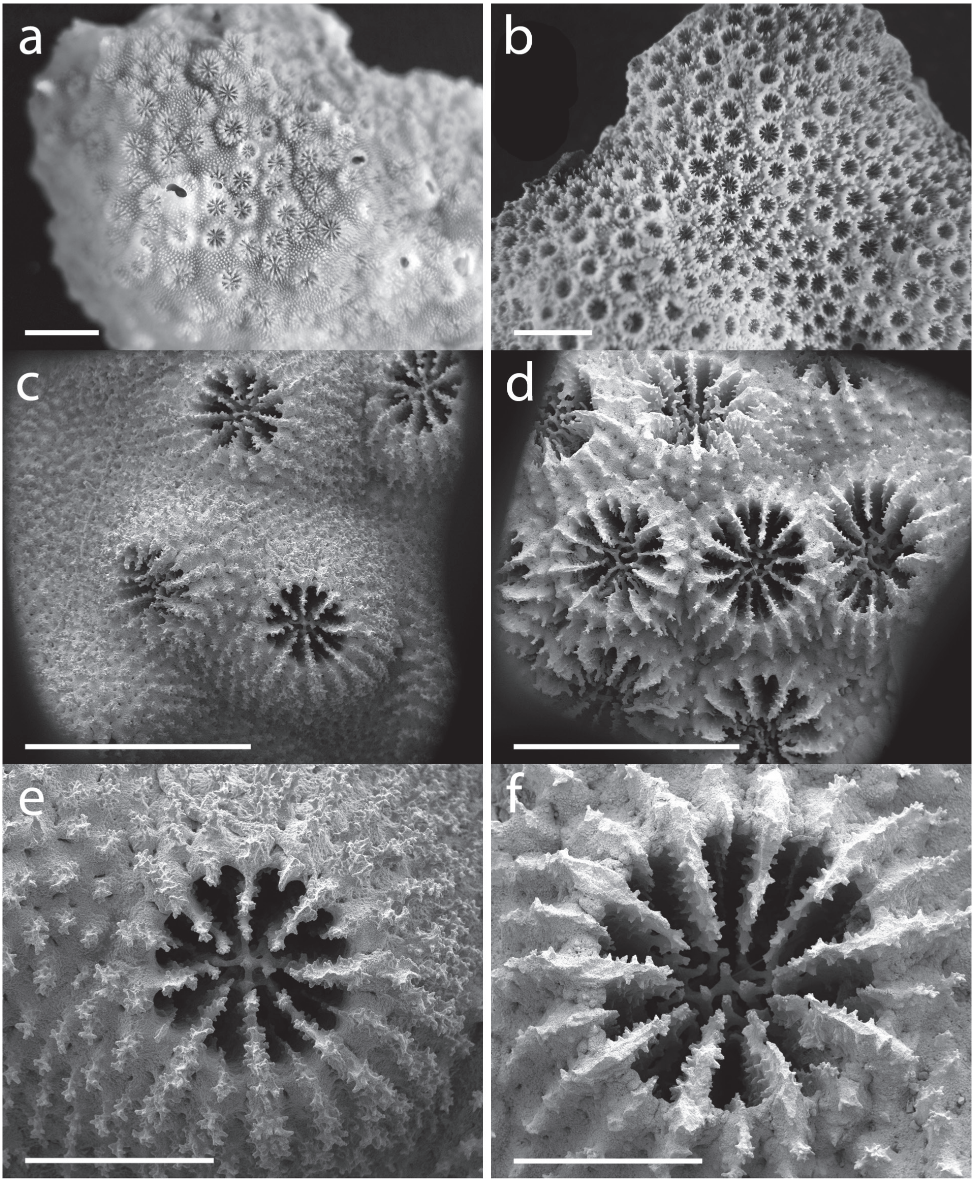

Figure 3. **a, c, e** *Cyphastrea kausti* sp. n. **a** holotype (MNHN-IK-2012-14236) **c, e** SEM images of SA607 **b, d, f** *Cyphastrea microphthalma* **b** holotype (MNHN-IK-2012-14002, photo by AF Budd, with permission from MNHN-Paris) **d, f** SEM images of SA159. Scale bars: 5 mm (**a, b**); 3 mm (**c, d**); 1 mm (**e, f**).

order septa, surrounds the columella (Figure 2c). Columella is trabecular and compact (Figures 2h–i, 3c, e). Septal teeth are prominent and ornamented, and granules are scattered on the septal face (Figure 2e–f). Costae are composed of a continuous or dotted line of ornamented spines, which become more and more spaced out while

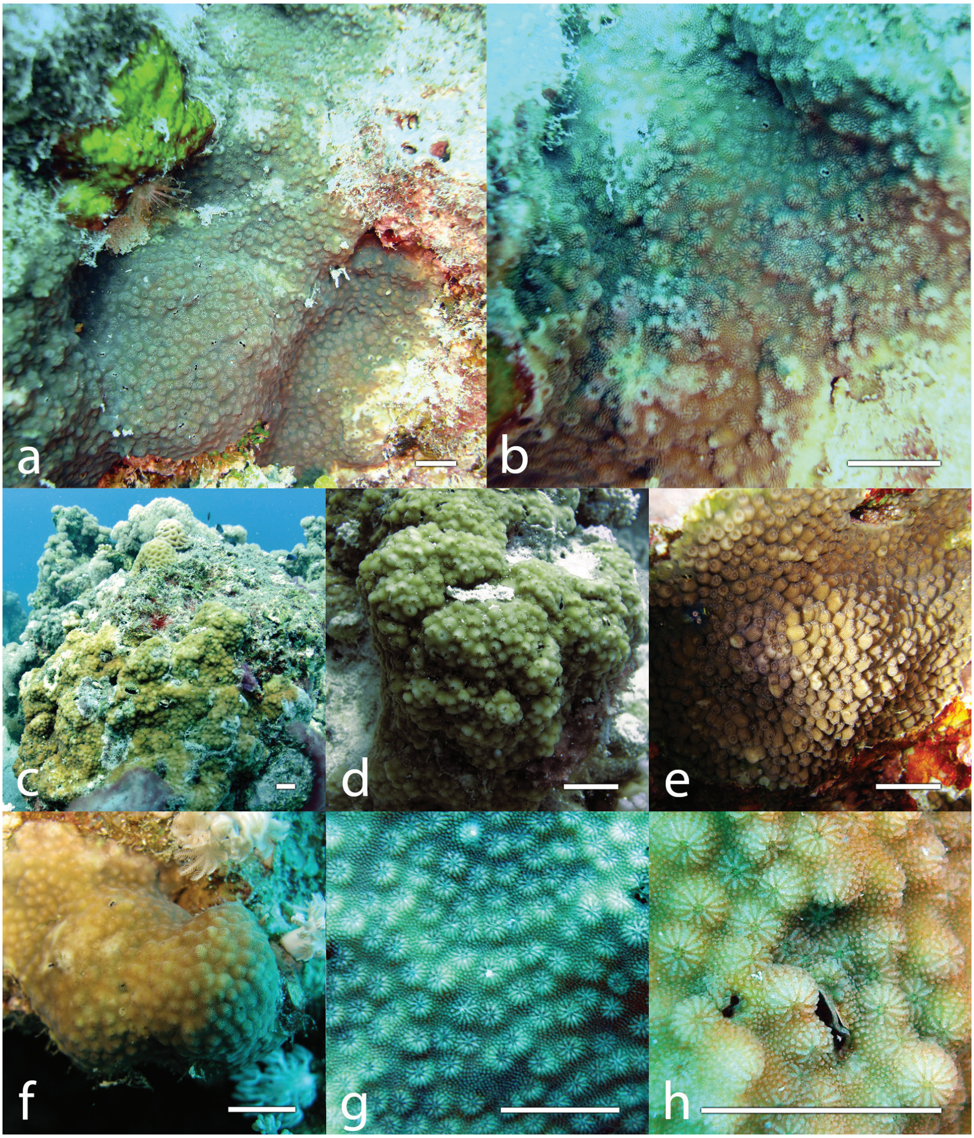

Figure 4. *Cyphastrea kausti* sp. n. **a–b** holotype (MNHN-IK-2012-14236) living colony (Fsar, 13.6 m depth) **c** SAE015 **d** SA498 **e** SA607 **f** SA446 **g** SA644 **h** 7887. Scale bars: 1 cm.

extending on the coenosteum, blending in with the already dense arrangement of ornamented spines, which covers the remaining of the coenosteum (Figure 2c–i). The height of each corallite is also variable, leading to colonies with corallites appearing more exsert than in others (e.g. Figure 4e-f).

Field characteristics and identification. Colonies of *Cyphastrea kausti* sp. n. are encrusting (Figures 4a–c) to submassive (Figure 4d), often growing on inclined substrate, and are found mostly at 6–12 m depth although they have been observed at 2.0–13.6 m depth. They appear similar to *C. microphthalma* but close observation of the corallites will reveal the typical eight-arm snowflake septal arrangement (Fig. 2, 3a, c, e). The size of the colonies is variable but generally is 10–60 cm. Colour in the field is cream, yellow, or brown.

Etymology. This species is named after the King Abdullah University of Science and Technology (KAUST), which has facilitated a considerable increase in marine biodiversity research in the Red Sea since its opening in 2009. Moreover, *Cyphastrea kausti* sp. n. was first observed by the authors on Al Fahal, a reef in front of KAUST, in Thuwal, Saudi Arabia.

Distribution. *Cyphastrea kausti* sp. n. has been recorded in the northern and central Red Sea, from Magna in the Gulf of Aqaba, to Thuwal (Figure 1). It was not found in the southern Red Sea, where a similar sampling effort was made.

Key to the species of *Cyphastrea* from the Red Sea

The primary septa cycle contains:

1	Six primary septa	**2**
–	Eight primary septa	***C. kausti* sp. n.**
–	Ten primary septa	***C. microphthalma***
2	Secondary septa cycle absent	***C. hexasepta***
–	Secondary septa cycle present	**3**
3	Primary and secondary septa alternating	***C. chalcidicum***
–	Primary and secondary septa subequal	***C. serailia***

Discussion

Cyphastrea kausti sp. n. is morphologically closest to *C. microphthalma* based on the presence of a primary and a secondary septa cycle, a missing third septa cycle, a crown of paliform lobes surrounding the columella (although generally more distinct in *C. kausti* sp. n.), the growth form, the densely ornamented coenosteum, and the density of corallites. The two species can, however, be distinguished by the average number of septa (8.0 ± 0.4 for *C. kausti* sp. n. and 9.8 ± 0.5 for *C. microphthalma*) and by the average calice and corallite size (Figure 3a–f). The calice and corallite diameter are on average smaller in *C. kausti* sp. n. (1.01 ± 0.13 mm and 1.62 ± 0.19 mm) than in *C. microphthalma* (1.27 ± 0.11 mm and 2.01 ± 0.18). In their description of *C. microphthalma* Sheppard and Sheppard (1991) report that Red Sea specimens have a tendency to contain mostly eight primary septa and that this form could be established as a new species in further work, which is done here. Field observations and sampling performed throughout the entire Saudi Arabian coast of the Red Sea between 2013 and 2014 provide evidence that both *C. kausti* sp. n. and *C. microphthalma* are present in the Red Sea and co-occurring in some regions, often in similar habitats. However, *C. kausti* sp. n. appears to be restricted to the central and northern Red Sea and is not found in the southern half of the Red Sea, while *C. microphthalma* extends out of the Red Sea and is distributed throughout the Indo-Pacific Ocean with an eastern range limit in Tahiti, French Polynesia, in the Central-Pacific Ocean (Sheppard and Sheppard 1991).

The simultaneous presence of both male and female gametes in colonies of *C. kausti* sp. n. during reproductive surveys shows that it is hermaphroditic and likely to spawn in June in the central Red Sea, along with numerous other species including the congeneric *C. serailia*, and *C. chalcidium*, while *C. microphthalma* was observed to spawn in May (Bouwmeester et al. 2015). Further reproductive surveys are, however, necessary to establish if indeed a reproductive barrier is present between *C. kausti* sp. n. and *C. microphthalma* with each species spawning in a different month, or whether one or both of the two species spawn over two consecutive months, as observed in the region for *Acropora humilis*, *Goniastrea edwardsi*, and *Echinopora hirsutissima*, which released gametes during consecutive months in a given year (Bouwmeester et al. 2015).

A molecular phylogeny of all *Cyphastrea* species remains necessary to test and establish species boundaries within the genus. The phylogenetic position of *C. kausti* sp. n. will be investigated in further work, integrating a molecular as well as a macromorphological and a micromorphological approach.

Conclusion

Cyphastrea kausti sp. n. is described from the Saudi Arabian Red Sea based on morphological analyses. The eight-septa arrangement in the first septa cycle distinguishes it from other described species in the genus. *Cyphastrea kausti* sp. n. is further recognized by a crown of paliform lobes around the columella and corallite and calice sizes smaller than in *C. microphthalma*, to which it is morphologically closely related.

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RESEARCH ARTICLE

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Two new species of the *Clubiona corticalis*-group from Yunnan Province, China (Araneae, Clubionidae)

Pan-Long Wu[1], Guo Zheng[2], Feng Zhang[1]

1 *The Key Laboratory of Invertebrate Systematics and Application, College of Life Sciences, Hebei University, Baoding, Hebei 071002, P. R. China* **2** *College of Life Science and Chemistry, Shenyang Normal University, Shenyang, Liaoning 110034, P. R. China*

Corresponding author: *Feng Zhang* (dudu06042001@163.com)

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Abstract

The present paper describes two new *Clubiona corticalis*-group species collected from Xishuangbanna, Yunnan Province of China: *Clubiona submoralis* **sp. n.** (♀♂) and *C. pollicaris* **sp. n.** (♀♂).

Keywords

Sac spiders, *Paraclubiona*, *Atalia*, taxonomy, South-East Asia

Introduction

Clubiona Latreille, 1804, the largest genus of the Clubionidae, currently includes 468 species (Platnick 2014) widely distributed around the world (except South America). Because of its high species diversity, several revisions have been published by Simon (1932) for the French species, Lohmander (1944) for Swedish species, Wiehle (1965) for German species, Wunderlich (2011) for European species, Edwards (1958) for the North American species, Dondale and Redner (1982) for Canadian and Alaskan species, Mikhailov (1990, 1991, 1995, 2002, 2012) for Palaearctic species, and Deeleman-Reinhold (2001) for southeast Asian species.

Table 1. A list of *Clubiona corticalis*-group species in China.

	Species name	Known sex	Distribution
1	*C. altissimoides* Liu et al., 2007	♂♀	Yunnan
2	*C. applanata* Liu et al., 2007	♂♀	Yunnan
3	*C. brachyptera* Zhu, Ren & Chen, 2012	♂♀	Hainan
4	*C. cordata* Zhang & Zhu, 2009	♂♀	Sichuan, Xizang
5	*C. cylindrata* Liu et al., 2007	♂♀	Yunnan
6	*C. didentata* Zhang & Yin, 1998	♂	Yunnan
7	*C. kurosawai* Ono, 1986	♂♀	Taiwan
8	*C. lamina* Zhang, Zhu & Song, 2007	♂	Yunnan
9	*C. lyriformis* Song & Zhu, 1991	♀	Hubei
10	*C. moralis* Song & Zhu, 1991	♂♀	Hubei
11	*C. parallela* Hu & Li, 1987	♂♀	Xizang
12	*C. pyrifera* Schenkel, 1936	♂♀	Gansu
13	*C. qiyunensis* Xu, Yang & Song, 2003	♂♀	Fujian, Anhui
14	*C. taiwanica* Ono, 1994	♂♀	Yunnan, Taiwan
15	*C. tengchong* Zhang, Zhu & Song, 2007	♂	Yunnan
16	*C. yaginumai* Hayashi, 1989	♂♀	Taiwan
17	*C. submoralis* sp. n.	♂♀	Yunnan
18	*C. pollicaris* sp. n.	♂♀	Yunnan

Clubiona corticalis-group was first recognized by Simon (1932). *Atalia* Thorell, 1887 (type species *A. concinna* Thorell, 1887, belongs to the *corticalis*-group) and subgenus *Paraclubiona* Lohmander, 1944 (with type species *C. corticalis*) are currently considered as junior synonyms of *Clubiona*. Species of this group can be recognized by the following characters: inflated tegulum; long filiform, spiniform, or short embolus; simple and weakly developed retrolateral tibial apophysis; the anterior position of copulatory openings (Mikhailov 1995).

Currently, the *corticalis*-group includes 48 species mainly distributed in Eurasia and Australia (Mikhailov 1995, Deeleman-Reinhold 2001, Liu et al. 2007). Among these, at least 18 species have been recorded from China (see Table 1).

While examining *Clubiona* specimens collected from Xishuangbanna Prefecture, Yunnan Province of China, we found two new species belonging to the *corticalis*-group, which are described in this paper.

Material and methods

All specimens studied are stored in 75% ethanol and deposited in the Institute of Zoology, Chinese Academy of Sciences in Beijing (IZCAS). All specimens were examined under a Tech XTL-II stereomicroscope. The photos, drawings and measurements were prepared using a Leica M205A stereomicroscope equipped with a DFC450 CCD camera and a drawing tube. Carapace length was measured from the anterior margin to the posterior margin of the carapace medially. The eyes were measured as the maximum diameter of the lens in dorsal or frontal view. The measurements of legs are shown as total length (femur, patella, tibia, metatarsus, tarsus). The epigyne was cleared in a solution of potassium hydroxide (KOH) and transferred to 75% ethanol for taking photos, drawing and measuring. All measurements are in millimeters.

The following abbreviations are used: ALE, anterior lateral eyes; AME, anterior median eyes; B, bursae; C, conductor; CO, copulatory openings; E, embolus; FD, fertilization ducts; MOA, median ocular area; PLE, posterior lateral eyes; PME, posterior median eyes; PPA, prolateral patellar apophysis; RFA, retrolateral femoral apophysis; RPA, retrolateral patellar apophysis; RTA, retrolateral tibial apophysis; S, spermathecae.

Taxonomy

Clubiona submoralis sp. n.

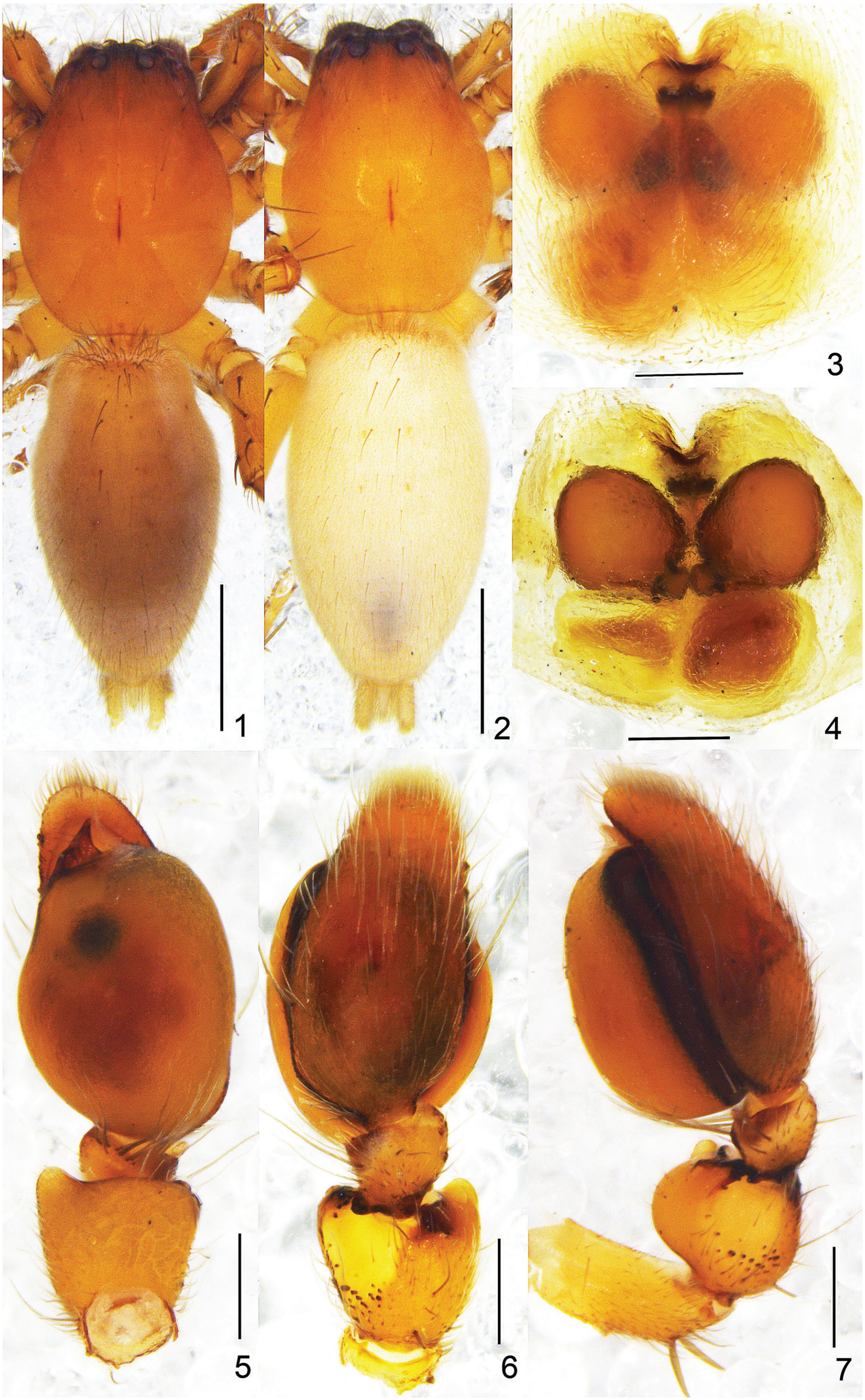
http://zoobank.org/3299A436-1170-4959-9BE3-95190B84F461
Figs 1–12

Type material. Holotype ♂, CHINA, Yunnan Province, Xishuangbanna Prefecture, Mengla County, Menglun Town, XTBG (Xishuangbanna Tropical Botanical Garden), Yunnan Rubber Plantation (575 m; 21°54.46'N, 101°15.98'E), 21 July 2007, Guo Zheng leg. Paratypes: 9 ♂, 10 ♀, same data as holotype.

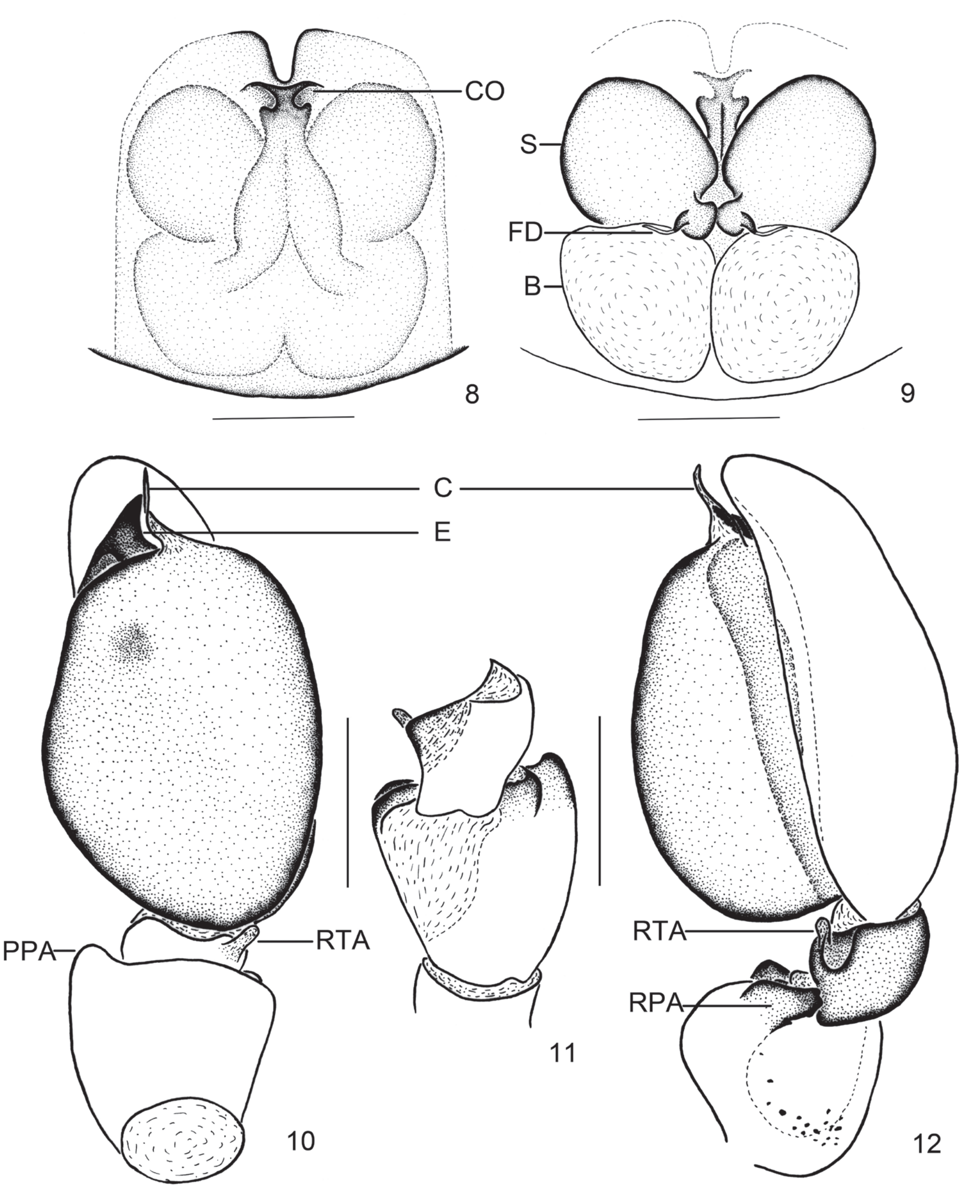
Diagnosis. Among the Chinese species of the *corticalis*-group, this new species resembles *C. moralis*, but differs by: a wider and triangular embolus (filiform and coiled in *C. moralis*), presence of only one round, black marking on tegulum (several fan-shaped markings in *C. moralis*); fertilization ducts opening in the medio-ventral side of the spermathecae (anteriorly in *C. moralis*); and the spermathecae almost as large as the bursae (the bursae twice longer than the spermathecae in *C. moralis*) (Figs 3–12).

Etymology. The species name refers to a close resemblance between the new species and *C. moralis*.

Description. Male. Total length 4.36–4.86. Holotype (Fig. 1): body 4.73 long; carapace 2.14 long, 1.96 wide; abdomen 2.49 long, 1.30 wide. Carapace brown. Median furrow longitudinal. In dorsal view, anterior eye row recurved, posterior eye row recurved. Eye sizes and interdistances: AME 0.10, ALE 0.11, PME 0.11, PLE 0.10; AME–AME 0.07, AME–ALE 0.06, PME–PME 0.27, PME–PLE 0.15. MOA 0.31 long, front 0.30 wide, back 0.48 wide. Chelicerae dark brown, promargin with six teeth, retromargin with three teeth. Endites brown, longer than wide. Labium dark brown, longer than wide. Sternum 1.19 long, 0.80 wide. Abdomen oval, brown, with conspicuous anterior tufts of hairs, dorsum with fine brown hairs. Legs yellow, both tibia I and II with two pairs of ventral spines, both metatarsi I and II with one pair of ventral spines. Measurements of legs: I 5.50 (1.55, 0.75, 1.57, 1.07, 0.56), II 6.02 (1.65, 0.80, 1.80, 1.21, 0.56), III 4.73 (1.26, 0.61, 1.13, 1.30, 0.43), IV 6.73 (1.78, 0.71, 1.65, 2.01, 0.58).



Figures 1–7. *Clubiona submoralis* sp. n. **1** male habitus, dorsal view **2** female habitus, dorsal view **3** epigyne, ventral view **4** vulva **5** left male palp, ventral view **6** same, dorsal view, showing tibial apophysis **7** same, retrolateral view. Scale bars: 1 mm (**1–2**); 0.2 mm (**3–7**).



Figures 8–12. *Clubiona submoralis* sp. n. **8** epigyne, ventral view **9** vulva **10** left male palp, ventral view **11** tibial apophysis, dorsal view **12** left male palp, retrolateral view. Scale bars: 0.25 mm (**8–12**).

Palp (Figs 5–7, 10–12). Femur unmodified; patella swollen, almost globular, twice wider than tibia, and 1.5 wider than femur, with short and rounded pro- and retrolateral apophyses, retrolateral side with short modified spines near the base; tibia with small retro-ventral membranous apophysis. Cymbium longer than tegulum. Tegulum inflated, with a round, black marking medially; embolus short, wide, almost triangular in ventral view; conductor membranous, folded in the middle position, and almost threefold longer than wide.

Female. Total length 4.45–4.92. One paratype (Fig. 2) measured, body 4.47 long; carapace 1.87 long, 1.43 wide; abdomen 2.49 long, 1.39 wide. Eye sizes and interdistances: AME 0.10, ALE 0.11, PME 0.09, PLE 0.07; AME–AME 0.08, AME–ALE 0.04, PME–PME 0.24, PME–PLE 0.14. MOA 0.22 long, front 0.23 wide, back 0.41 wide. Sternum 0.96 long, 0.70 wide. Measurements of legs: I 4.53 (1.31, 0.66, 1.21, 0.84, 0.51), II 5.14 (1.48, 0.74, 1.41, 0.97, 0.54), III 4.22 (1.25, 0.57, 0.90, 1.13, 0.37), IV 6.14 (1.66, 0.70, 1.40, 1.89, 0.49). Coloration lighter than in male. Other characters as in male.

Epigyne (Figs 3–4, 8–9). Copulatory openings located anteriorly; in ventral view, the anterior part of copulatory ducts well visible and extending posteriorly, then connecting to bursae; spermathecae located anterior to bursae, both of them almost spherical, and with the same size as bursae.

Distribution. China (Yunnan).

Clubiona pollicaris sp. n.

http://zoobank.org/A815DA83-1925-4B07-BB84-D2DC0D5DA3C8
Figs 13–19, 23–27

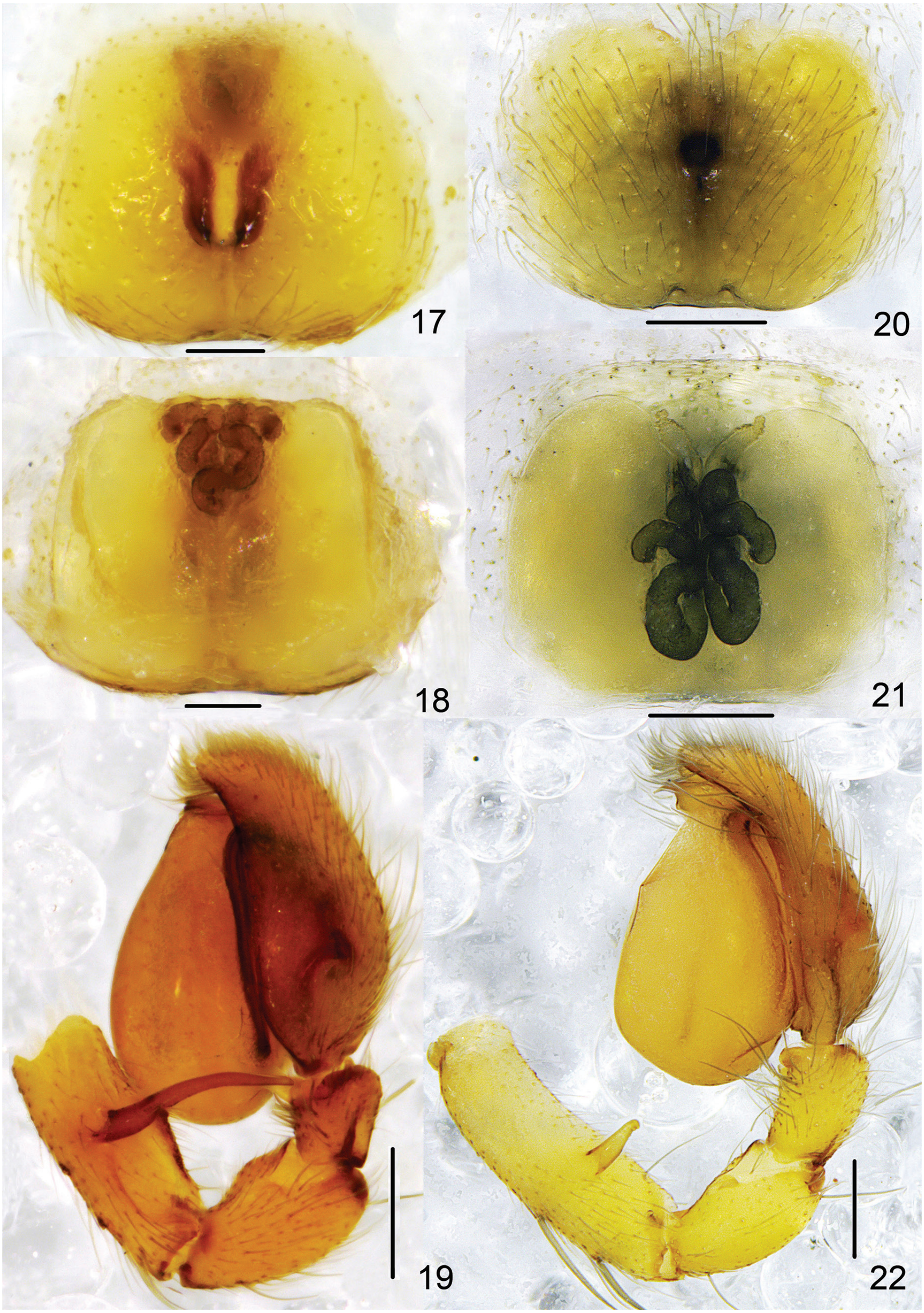
Type material. Holotype ♂, CHINA, Yunnan Province, Xishuangbanna Prefecture, Mengla County, Menglun Nature Reserve (710 m; 21°57.70'N, 101°11.89'E), 7 August 2007, Guo Zheng leg. Paratypes: 6 ♂, 6 ♀, same data as holotype.

Diagnosis. The new species differs from all other *Clubiona* species by a very long retrolateral femoral apophysis (almost as long as femur) and resembles *C. qiyunensis* (Figs 20–22; also see Wu and Zhang (2014): 211, f. 13–23), but differs by: a smaller and inconspicuous embolus; the much longer femoral apophysis (in *C. qiyunensis*, femoral apophysis is short, less than diameter of femur); a thumb-shaped prolateral patellar apophysis; anteriorly situated spermathecae; and rectangular bursae (Figs 15–19, 23–27).

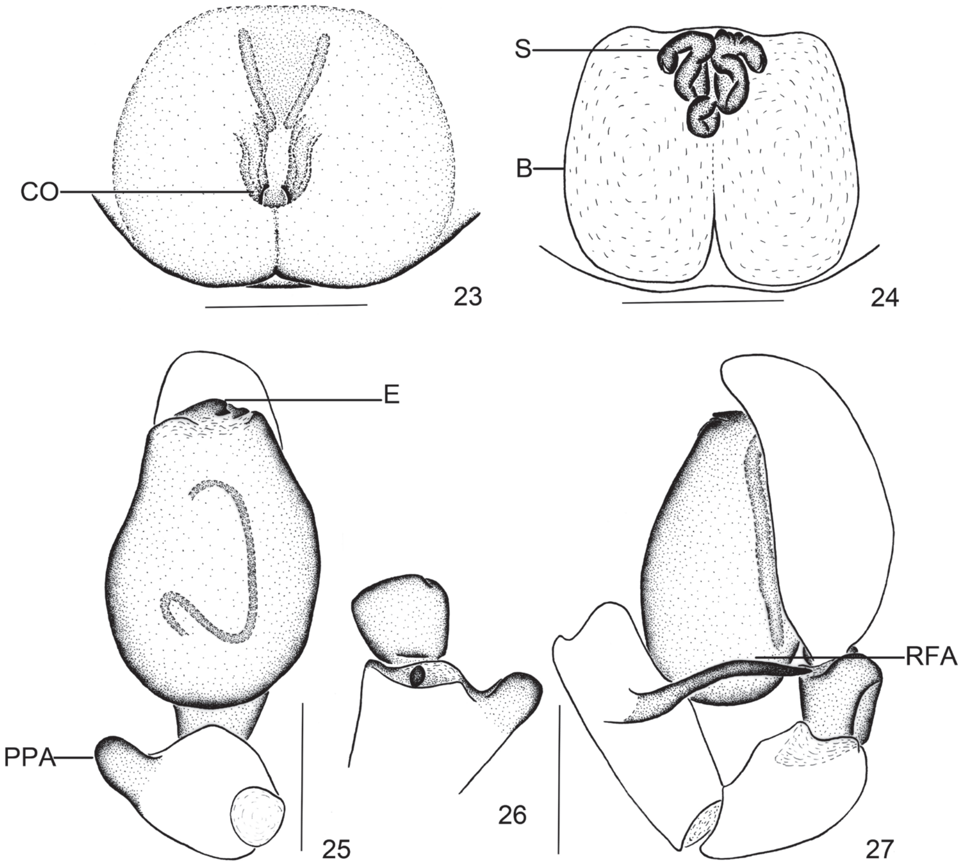
Etymology. The species name is derived from the Latin word “*pollicaris*”, and refers to the prolateral patellar apophysis, which in ventral view is thumb-shaped.

Description. Male. Total length 3.20–3.49. Holotype (Fig. 13) body 3.27 long; carapace 1.51 long, 1.45 wide; abdomen 1.61 long, 0.90 wide. Carapace brown. Median furrow longitudinal. Both anterior and posterior eye row recurved in dorsal view. Eye sizes and interdistances: AME 0.06, ALE 0.10, PME 0.09, PLE 0.08; AME–AME 0.06, AME–ALE 0.03, PME–PME 0.19, PME–PLE 0.08. MOA 0.21 long, front 0.19 wide, back 0.37 wide. Chelicerae brown, promargin with five teeth, retromargin with four teeth. Both endites and labium brown, longer than wide. Sternum 0.83 long, 0.56 wide. Abdomen oval, brown, with conspicuous anterior tufts of hairs, dorsum with fine yellow hairs. Legs brown, both tibia I and II with two pairs of ventral spines, both metatarsi I and II with one pair of ventral spines. Measurements of legs: I 3.09 (0.84, 0.47, 0.83, 0.62, 0.33), II 3.46 (0.90,

Figures 13–16. *Clubiona pollicaris* sp. n. **13** male habitus, dorsal view **14** female habitus, dorsal view **15** left male palp, ventral view **16** same, dorsal view, showing patellar apophysis. Scale bars: 1 mm (**13–14**); 0.2 mm (**15, 16**).



Figures 17–22. 17–19 *Clubiona pollicaris* sp. n. **17** epigyne, ventral view **18** vulva **19** left male palp, retrolateral view; **20–22** *Clubiona qiyunensis* Xu, Yang & Song, 2003. **20** epigyne, ventral view **21** vulva **22** left male palp, retrolateral view. Scale bars: 0.1 mm (**17, 18**) 0.2 mm (**19–22**).



Figures 23–27. *Clubiona pollicaris* sp. n. **23** epigyne, ventral view **24** vulva **25** left male palp, ventral view **26** patellar apophysis, dorsal view **27** left male palp, retrolateral view. Scale bars: 0.25 mm (**23–27**).

0.50, 1.02, 0.69, 0.35), III 2.92 (0.84, 0.43, 0.63, 0.76, 0.26), IV 4.11 (1.18, 0.50, 0.96, 1.15, 0.32).

Palp (Figs 15–16, 19, 25–27). Femur modified, with long and thin retrolateral apophysis originating from median part, apophysis longer than tibia and subequal in length to femur; patella twice longer and 1.5 wider than tibia, with a round apophysis and thumb-shaped prolateral apophysis; tibia without apophyses; cymbium shorter than tegulum; tegulum inflated; sperm duct obvious, almost straight in retrolateral view; embolus short, originating from the apical tegulum prolaterally, directed almost horizontally.

Female. Total length 3.12–3.92. One paratype (Fig. 14) measured: body 3.90 long, carapace 1.52 long, 1.13 wide; abdomen 2.09 long, 1.18 wide. Eye sizes and interdistances: AME 0.07, ALE 0.09, PME 0.08, PLE 0.07; AME–AME 0.08, AME–ALE 0.05, PME–PME 0.20, PME–PLE 0.12. MOA 0.21 long, front 0.20 wide, back

0.35 wide. Sternum 0.85 long, 0.57 wide. Measurements of legs: I 2.69 (0.84, 0.38, 0.64, 0.51, 0.32), II 3.09 (0.90, 0.46, 0.79, 0.60, 0.34), III 2.63 (0.80, 0.35, 0.55, 0.67, 0.26), IV 3.92 (1.02, 0.50, 0.93, 1.10, 0.37). Coloration slightly lighter than in male; other characters as in male.

Epigyne (Figs 17–18, 23–24). Copulatory openings small, located almost in the centre of the epigynal plate; in ventral view copulatory ducts inconspicuous; spermathecae long, tubular and sinuous; bursae large, membranous, almost rectangular.

Distribution. China (Yunnan).

Comments. Retrolateral femoral apophysis is known only in three species of Clubionidae: *Clubiona pollicaris*, *C. qiyunensis* and *C. brachyptera*.

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RESEARCH ARTICLE

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A faunistic study on the leafhoppers of northwestern Iran (Hemiptera, Cicadellidae)

Tandis Abdollahi[1], Ali Reza Jalalizand[1], Fariba Mozaffarian[2], Michael Wilson[3]

1 *Department of Plant protection, Faculty of Agriculture, Islamic Azad University, Isfahan (Khorasgan) Branch, Isfahan, Iran* **2** *Insect Taxonomy Research Department, Iranian Research Institute of Plant Protection, Tehran, 19395, P.O. Box 1454, Iran* **3** *Department of Natural Sciences, National Museum of Wales, Cardiff, U.K.*

Corresponding author: *Tandis Abdollahi* (tandis_ab@yahoo.com)

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Abstract

The leafhopper fauna of northwestern Iran: Azarbaijan-e-Sharghi, Azarbaijan-e-Gharbi and Ardabil provinces is listed from previously published records and from our current work. Sixty-nine species are included with four species (*Mogangella straminea* Dlabola, 1957, *Doratura stylata* (Boheman, 1847), *Macrosteles sordidipennis* (Stål, 1858) and *Psammotettix seriphidii* Emeljanov, 1962) listed as new for Iran and *Balclutha punctata* (Fabricius, 1775), as a new record for the region. A distribution map of the species in northwestern Iran is given.

Keywords

Cicadellidae, leafhoppers, fauna, Azarbaijan-e-Sharghi, Azarbaijan-e-Gharbi, Ardabil

Introduction

The Auchenorrhyncha consists of approximately 42000 described worldwide species which have adopted varied life habits (Larivière et al. 2010). Moreover, they play an important role in the food chains due to their high biomass in the herb layer and provide a food source for other insects and animals (Nickel 2003).

The family Cicadellidae (leafhoppers), is the largest family within the Hemiptera, with approximately 19,500 described species in more than 40 subfamilies (Oman et al. 1990). Metcalf (1962–1968) considered the leafhoppers to represent a superfamily (the Cicadelloidea) and divided them into a number of families, currently subfamilies or tribes. Following Dietrich (2005), Cicadellidae are included in the superfamily Membracoidea with the Membracidae (treehoppers). Most Cicadellidae species tend to feed from phloem fluid (except some Cicadellinae and most Typhlocybinae) (Biedermann and Niedringhaus 2009). Moreover, some species may cause both direct and indirect damage during their feeding activity, which is sometimes economically important. The most important form of indirect damage is caused by phytoplasmas and viruses, vectored mostly by Cicadellidae (Weintraub and Beanland 2006).

The earliest available record of Auchenorrhyncha from Iran is Gardenhire (1959) who recorded some species as agricultural pests. Jiri Dlabola, from the Czech Republic, studied considerable numbers of Auchenorrhyncha species from Iran in the 1970s, which led to the discovery of more than 100 new Cicadellidae species in a long series of papers (Dlabola 1974, 1977, 1980, 1981, 1982, 1983, 1985). More recently other authors have published on the fauna: Karimzadeh et al. (1998); Haghshenas and Khajeali (2000); Lashkari et al. (2009); Taghizadeh et al. (2010); Mozaffarian and Taghizadeh (2010); Mozaffarian and Emeljanov (2010); Mozaffarian et al. (2010); Mozaffarian and Sanborn (2010, 2012, 2013); Mozaffarian and Gnezdilov (2011); Gnezdilov and Mozaffarian (2011); Moazaffarian and Wilson (2011); Moosavi and Sadeghi Namaghi (2012); Mozaffarian (2012a, b); Taghizadeh (2012); Zohdi et al. (2012); Aghagoli-Marzijarani et al. (2013), Mozaffarian (2013) and Abdollahi et al. (2013, 2014). There have been a wide range of researchers who mainly focused on the Auchenorrhyncha as pests in both agricultural and forest ecosystems in Iran such as: Gharib (1966); Kheyri and Alimoradi (1969); Kheyri (1989, 1992); Rajabi and Mirzayans (1989); Behdad (1992, 1993); Khajehali et al. (2001); Nematollahi and Khajehali (2000); Yarmand et al. (2006); Aghagoli-Marzjarani et al. (2010); Taghizadeh et al. (2010) amongst others.

Northwestern Iran (the study area) covers nearly 100,503 square kilometers and consists of three provinces: Azarbaijan-e-Sharghi, Azarbaijan-e-Gharbi and Ardabil. It is located in Irano-Turanian zoogeographical region (Firouz 2005) and in the northwest plateau of Iran. It is limited between the Caspian Sea and Caspian district in the east, Caucasus mountains in the north, Anatolian Plateau and Mesopotamian region in the west and a part of Zagros, called Humid Zagros, in the south. Hence, it is expected that the fauna of this area will be influenced by the faunal elements of all mentioned regions rather than just the Iranian Plateau. The area is considered to be the crossroads of the two main mountains of Iran (Alborz and Zagros), a part of Alpine Himalayan orogenic belt (Dewey et al. 1986) with deep valleys and has a variety of altitudes from 256 m to 2896 m. It is differentiated from other parts of Iran by the highest latitude (39°40'N) and the coldest recorded temperature (-35 °C) (Hedge and Wendelbo 1978). Zarudny (1911) considered this part of Iran as a zoogeographic zone, with a fauna similar to the Caucasus. This area was also considered as a different

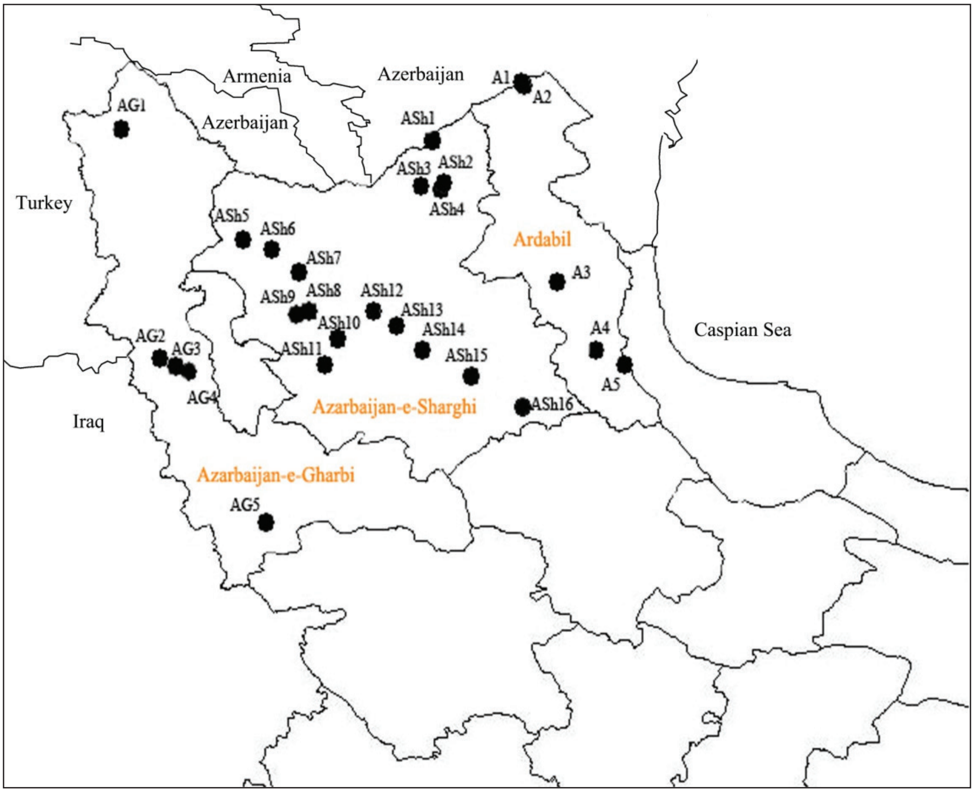

Figure 1. Distribution map of leafhoppers in northwestern Iran (For codes see Table 1).

area from other parts of Zagros by Emeljanov (1974). Hedge and Wendelbo (1978) recognized part of Iran as an endemic zone named Armeno-Kurdic due to the distribution patterns of endemic phanerogamic plants.

The aim of this research was to collect and identify the leafhoppers in northwestern Iran and to prepare a checklist as a starting point for gathering the sporadic publications and studying the fauna for the whole of Iran. A total number of 69 species belonging to 11 subfamilies are recorded.

Material and methods

The present study was carried out in three northwestern provinces including, Azarbaijan-e-Sharghi, Azarbaijan-e-Gharbi and Ardabil provinces (Figure 1). During 2007 (August, September and January) and 2008 (January) field trips were made and leafhopper specimens were collected using a sweep net. A total of 2340 specimens consists of newly collected specimens along with other specimens located in the Hayk Mirzayans Insect Museum (Tehran, Iran, which had been collected since 1968) were studied and identi-

Table 1. List of the localities, their coordinates and the codes

Locality names	Coordinates	Locality names	Coordinates
Ajabshir	37° 35'N, 46°11'E (ASh11)	Moghan	39°37'N, 47°47'E (A1)
Bonab	37°26'N, 45°57'E (ASh9)	Eskanlu	39°12'N, 47°04'E (ASh1)
Bostanabad-Siah chaman	37°41'N, 46°59'E (ASh14)	Parsabad	39°36'N, 47°49'E (A2)
Heiran	37°41'N, 48°24'E (A4)	Sarein	38°11'N, 48°05'E (A3)
Kaleibar, Arabshahi	38°51'N, 47°08'E (ASh4)	Siah chaman-Basmenj	37°52'N, 46°46'E (ASh13)
Kaleibar, 1863m	38°52'N, 46°58'E (ASh3)	Sufian	38°16'N, 45°58'E (ASh7)
Kaleibar, 1732m	38°54'N, 47°09'E (ASh2)	Tabriz	37°58'N, 46°03'E (ASh8)
Kandovan	37°46'N, 46°17'E (ASh10)	Tabriz-Shabestar	38°15'N, 45°58'E (ASh7)
Khalkhal	37°35'N, 48°38'E (A5)	Tabriz-Bostanabad	37°58'N, 46°35'E (ASh12)
Mahabad	36°27'N, 45°42'E (AG5)	Uromieh	37°25'N, 47°42'E (AG3)
Maku	39°17'N, 44°31'E (AG1)	Uromieh, Mirzabad	37°32'N, 45°04'E (AG4)
Marand	38°25'N, 45°45'E (ASh6)	Uromieh-Sarv	37°38'N, 44°50'E (AG2)
Miyaneh-Siah chaman	37°30'N, 47°23'E (ASh15)	Zonuschay	38°29'N, 45°31'E (ASh5)
Miyaneh-Zanjan	37°17'N, 47°48'E (ASh16)		

fied. The identifications were made using the works of Le Quesne (1965, 1969), Ribaut (1952), Biedermann and Niedringhaus (2009) and Emeljanov (1997). Vouchers of all species are deposited in the Hayk Mirzayans Insect Museum. In addition to the identification, literature records were also taken into consideration and a distribution map for the leafhoppers of northwestern Iran was prepared by ARCMAP version 9.3.0.1770.

List of taxa

The genera and species from northwestern Iran recorded through the present study and other publications are as follows (* indicates species not found in the present study). For those species with specimens examined, a reference to the authority used for the identification is included in parenthesis following the taxon heading. The classification used follows mainly Oman et al. (1990) with changes based on more recent literature e.g., Zahniser and Dietrich (2013) for Deltocephalinae.

Subfamily: Agalliinae
Tribe: Agalliini

Agallia firdausica Dlabola, 1981*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. Iran; Saudi Arabia (Dlabola 1980).

Anaceratagallia laevis (Ribaut, 1935)

Anaceratagallia laevis: Le Quesne 1965: 52, figs 267–268.

Material examined. Azarbaijan-e-Sharghi: 1♂, 2♀, Ajabshir, Yaichi village, 1922 m, 37°35'27.2"N, 46°11'03.7"E, 15.January.2008, leg. Mozaffarian (Fig. 1, ASh11).

Dlabola (1981) reported this species from Sufian (Fig. 1, ASh7).

Worldwide distribution. East Palaearctic, Europe (Albania, Britain I., Bulgaria, Canary Is., Channel Is., Cyprus, French mainland, Greek mainland, Hungary, Italian mainland, Moldova, Portuguese mainland, Romania, South Russia, Sardinia, Sicily, Spanish mainland, Ukraine, Yugoslavia), Near East, North Africa (De Jong 2013).

Austroagallia sinuata (Mulsant & Rey, 1835)

Austroagallia sinuata: Le Quesne 1965: 50, figs 253–255, 257.

Material examined. Azarbaijan-e-Sharghi: 1♂, 1♀, Kaleibar, Arabshahi, 1391 m, 38°51'42.7"N, 47°08'01.1"E, 3.September.2007, leg. Mozaffarian (Fig. 1, ASh4).

Dlabola (1971, 1981) reported this species from Sufian, Maku and Miyaneh-Siah chaman (Fig. 1, Ash7, AG1, ASh15)

Worldwide distribution. Afro-tropical region, East Palaearctic, Europe (Austria, Balearic Is., Belgium, Britain I., Bulgaria, Canary Is., Crete, Cyprus, Greek mainland, Hungary, Italian mainland, Moldova, Portuguese mainland, Romania, South Russia, Sardinia, Sicily, Slovakia, Spanish mainland, Switzerland, Yugoslavia), Near East, North Africa (De Jong 2013).

Subfamily: Aphrodinae
Tribe: Aphrodini

Aphrodes bicinctus (Schrank, 1776)*

Localities. Sufian, Marand (Dlabola 1981) (Fig. 1, ASh7, ASh6).

Worldwide distribution. Europe (Albania, Austria, Belgium, Britain I., Bulgaria, Corsica, Crete, Croatia, Cyprus, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Lithuania, Madeira, Republic of Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Russia Central, Russia North, South Russia, Sardinia, Sicily, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia) (De Jong 2013).

Subfamily: Cicadellinae
Tribe: Cicadellini

Cicadella viridis (Linnaeus, 1758)

Cicadella viridis: Le Quesne 1965: 24, fig. 115.

Material examined. Ardabil: 8 ♂♀, Heiran, 1527 m, 37°41'07.4"N, 48°23'57.4"E, 18.January.2007, leg. Mozaffarian, Light trap (Fig. 1, A4).

Ardabil: 1♂, 7♀, 10 km to Parsabad, 39°36'8.3"N, 47°48'45.5"E, 18.January. 2007, leg. Mozaffarian (Fig. 1, A2).

Azarbaijan-e-Sharghi: 25 ♂♀, Eskanlu, Aras river, 290 m, 39° 12'13.4"N, 47° 04'23.2"E, 3.Sebtember.2007, leg. Mozaffarian & Nematian (Fig. 1, ASh1).

Azarbaijan-e-Gharbi: 1♂, Maku, Cheshme Soraya, 900 m, 22.August.1994, leg. Ebrahimi & Sarafrazi (Fig. 1, AG1).

Dlabola (1981) reported this species from Zonuschay (Fig. 1, ASh5).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Belgium, Britain I., Bulgaria, Croatia, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Romania, Russia Central, Russia North, South Russia, Sardinia, Sicily, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region, Oriental region (De Jong 2013).

Comment. Behdad (1993) reported this species as a rice pest.

Subfamily: Deltocephalinae
Tribe: Athysanini

Conosanus obsoletus (Kirschbaum, 1858)

Conosanus obsoletus: Ribaut 1952: 95, 99, figs 137–138; Le Quesne 1969: 109, figs 593, 596.

Material examined. Azarbaijan-e-Gharbi: 15♂♀, Mahabad, KoushkDareh, 1499 m, 36°27'08.6"N, 045°42'32.9"E, 28.August.2007, leg. Mozaffarian & Nematian (Fig. 1, AG5).

Dlabola, 1981 reported this species from Sufian and Marand. (Fig. 1, ASh7, ASh6).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Azores, Belgium, Britain I., Bulgaria, Cyprus, Czech Republic, Danish mainland, Estonia, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Sicily, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region, North Africa (De Jong 2013).

Eohardya miyaneha Dlabola, 1971*

Localities. Miyaneh- Siah chaman (Dlabola 1971) (Fig. 1, ASh15).

Worldwide distribution. Iran (Dlabola 1971).

Euscelis alsius Ribaut, 1952

Euscelis alsius: Ribaut 1952: 95, fig. 130.

Material examined. Ardabil: 2♂♀, Moghan, 65 m, 39°37'30.7"N, 47°46'57.5"E, 19.January.2007, leg. Mozaffarian (Fig. 1, A1).

Ardabil: 21♂♀, Parsabad, 80 m, 39°36'8.3"N, 47°48'45.5"E, 18.January.2007, leg. Mozaffarian (Fig. 1, A2).

Ardabil: 1♂, 1♀, 12 km to Khalkhal, 1998 m, 37°35'41.8"N, 48°37'54.3"E, 17.Janaury.2007, leg. Mozaffarian, Light trap (Fig. 1, A5).

Azarbaijan-e-Sharghi: 74♂♀, Tabriz, Khosroshahr, 1346 m, 37°58'28"N, 46°02'55"E, 21-30.August.2007, leg. Lotfalizadeh, Malaise trap (Fig. 1, ASh8).

Azarbaijan-e-Sharghi: 1♂, Sahand mountain, Kandovan, 2661 m, 37°45'47.7"N, 46°17'39.8"E, 1.September.2007, leg. Mozaffarian (Fig. 1, ASh10).

Dlabola (1981) reported this species from Zonuschay and Sufian (Fig. 1, ASh5, ASh 7).

Worldwide distribution. East Palaearctic, Europe (Bulgaria, French mainland, Greek mainland, Italian mainland, Portuguese mainland, Sicily, Spanish mainland, Yugoslavia), Near East, North Africa (De Jong 2013).

Handianus bejbienkoi Dlabola, 1959

Handianus bejbienkoi: Emeljanov 1964: 523, fig. 188: 26–27.

Material examined. Ardabil: 1♂, 10 km to Parsabad, 39°36'8.3"N, 47°48'45.5"E, 18.January.2007, leg. Mozaffarian (Fig. 1, A2).

Dlabola 1981 reported this species from Zonuschay and Maku (Fig. 1, ASh5, AG1).

Worldwide distribution. East Palaearctic, Europe (Ukraine), Near East (De Jong 2013).

Hardya anatolica Zachvatkin, 1946*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. Europe (Bulgaria, Greek mainland, Italian mainland, Romania, Yugoslavia), Near East (De Jong 2013).

Hardya iranicola Zachvatkin, 1946*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. Iran (Nast 1972).

Platymetopius chloroticus Puton, 1877 *

Localities. Sufian, Zonuschay (Dlabola 1981) (Fig. 1, ASh7, ASh5).

Worldwide distribution. East Palaearctic, Europe (South Russia, Ukraine), Near East (De Jong 2013).

Platymetopius safavii Dlabola, 1971*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. Iran (Dlabola 1981).

Platymetopius shirazicus Dlabola, 1974*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. Iran (Dlabola 1981).

Tribe: Chiasmini

Aconura jakowlefi Lethierry, 1876 *

Localities. Sufian, Zonuschay (Dlabola 1981) (Fig. 1, ASh7, ASh5).

Worldwide distribution. East Palaearctic, Europe (South Russia), Near East (De Jong 2013).

Chiasmus conspurcatus (Perris, 1857)*

Localities. Miyaneh-Siah chaman (Dlabola 1971) (Fig. 1, ASh15).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria (doubtful), Bulgaria, Canary Is., French mainland, Greek mainland, Italian mainland, Portuguese mainland, Romania, Sardinia, Sicily, Slovenia, Spanish mainland, Switzerland, Yugoslavia), Near East (De Jong 2013).

Doratura marandica Dlabola, 1981*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. Iran (Dlabola 1981).

Doratura stylata (Boheman, 1847)

Doratura stylata: Le Quesne 1969: 67, figs 329–331; Biedermann and Niedringhaus 2009: 298.

Material examined. Aradebil: 11♂♀, Sarein. Ardestan, 1700 m, 2.July.1997, leg. Barari & Mofidi (Fig. 1, A3).

Azarbaijan-e-Sharghi: 1♂, Bonab, 13.6 m, 37° 26'14.4"N, 045° 57'56.7"E, 27.August, 2007, leg. Mozaffarian & Nematian (Fig. 1, ASh9).

Azarbaijan-e-Sharghi: 9♂♀, Kandovan, 2645 m, 37° 45'45.8"N, 46° 17'40.5"E, 18.January.2008, leg. Mozaffarian. (Fig. 1, ASh10).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Belgium, Britain I., Bulgaria, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Russia Central, Russia North, South Russia, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region, North Africa (De Jong 2013).

This species is newly recorded from Iran.

Doraturopsis heros (Melichar, 1902)*

Localities. Zonuschay, Marand (Dlabola 1981) (Fig. 1, ASh5, ASh6).

Worldwide distribution. Europe (South Russia, Ukraine) (De Jong 2013).

Tribe: Cicadulini

Stenometopiellus iranicus Zachvatkin, 1946*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. Iran (Dlabola 1981); Uzbekistan (Nast 1972).

Tribe: Goniagnathini

Goniagnathus brevis (Herrich-Schäffer, 1835)

Goniagnathus brevis: Emeljanov 1964: 501, fig. 180: 7, 8; Biedermann and Niedringhaus 2009: 283.

Material examined. Azarbaijan-e-Sharghi: 1♂, Kaleibar, 1863 m, 38°52'13.5"N, 46°58'14.5"E, 2.September.2007, leg. Mozaffarian (Fig. 1, ASh3).

Abdollahi et al. (2013) reported this species from the above locality.

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Belgium, Bulgaria, Cyprus, Czech Republic, European Turkey, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Moldova, Poland, Portuguese mainland, Romania, South Russia, Sicily, Slovakia, Spanish mainland, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, North Africa (De Jong 2013).

Goniagnathus guttulinervis (Kirschbaum, 1868)*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. Afro-tropical region, East Palaearctic, Europe (Balearic Is., Canary Is., French mainland, Greek mainland, Hungary, Italian mainland, Portuguese mainland, South Russia, Sardinia, Sicily, Spanish mainland, The Netherlands (doubtful), Ukraine), Near East, North Africa (De Jong 2013).

Goniagnathus minor Kusnezov, 1928*

Localities. Miyaneh- Siah chaman (Dlabola 1971) (Fig. 1, ASh15).

Worldwide distribution. Ukraine (Nast 1972).

Tribe: Hecalini

Hecalus glaucescens (Fieber, 1866)*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. East Palaearctic, Europe (Bulgaria, Cyprus, Greek mainland, Italian mainland, South Russia, Sicily, Slovakia, Spanish mainland, Ukraine, Yugoslavia), Near East, North Africa (De Jong 2013).

Tribe: Limotettigini

Limotettix striola (Fallén, 1806)

Limotettix striola: Emeljanov 1964: 529, fig. 190: 2–5; Biedermann and Niedringhaus 2009: 322.

Material examined. Ardabil: 1♀, Moghan, Parsabad, 9.May.1969, leg. Abaii. (Fig. 1, A1)

Azarbaijan-e-Sharghi: 1♀, Tabriz, Gharachaman, 1600 m, 16.January.1976, leg. Boroumand & Pazouki (Fig. 1, ASh8).

Dlabola (1981) reported this species from Sufian (Fig. 1, ASh7).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Azores, Belgium, Britain I., Bulgaria, Cyprus, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Russia Central, Russia North, South Russia, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region, North Africa (De Jong 2013).

Tribe: Macrostelini

Balclutha flavella Linnavuori, 1962*

Localities. Zonuschay, Marand (Dlabola 1981) (Fig. 1, ASh5, ASh6).

Worldwide distribution. Israel (Nast 1972).

Balclutha punctata (Fabricius, 1775)

Balclutha punctata: Emeljanov 1964: 507, fig. 182: 3; Biedermann and Niedringhaus 2009: 286.

Material examined. Ardabil: 32♂♀, Heiran, 1527 m, 37°41'07.4"N, 48°23'57.4"E, 18.January.2007, Light trap, leg. Mozaffarian (Fig. 1, A4).

Worldwide distribution. Australian region, East Palaearctic, Europe (Albania, Austria, Britain I., Bulgaria, Cyprus, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Russia Central, Russia North, South Russia, Sardinia, Sicily, Slovakia, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region, North Africa, Oriental region (De Jong 2013).

This species is newly recorded from northwestern Iran.

Balclutha rhenana Wagner, 1939*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. East Palaearctic, Europe (Austria, Bulgaria, Czech Republic, Finland, Germany, Greek mainland (doubtful), Slovakia, Switzerland, The Netherlands, Yugoslavia) (De Jong 2013).

Macrosteles chobauti Ribaut, 1952

Macrosteles chobauti: Ribaut 1952: 48, figs 26–28.

Material examined. Azarbaijan-e-Sharghi: 51♂♀, Kandovan, 2645 m, 37°45'45.8"N, 46°17'40.5"E, 18.January.2008, leg. Mozaffarian (Fig. 1, ASh10).

Abdollahi et al. (2014) also reported this species from the above locality.

Worldwide distribution. Europe (Bulgaria, French mainland, Greek mainland) (De Jong 2013), France, Israel (Nast 1972).

Macrosteles fieberi (Edwards, 1889)*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. East Palaearctic, Europe (Austria, Britain I., Bulgaria, Czech Republic, Finland, French mainland, Germany, Greek mainland (doubtful), Ireland, Moldova, Norwegian mainland, Poland, Romania, South Russia, Slovakia, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region (De Jong 2013).

Macrosteles laevis (Ribaut, 1927)*

Localities. Zonuschay, Maku (Dlabola 1981); Miynaeh-Zanjan, Miyaneh-Siah chaman, Tabriz-Shabestar (Dlabola 1971) (Fig. 1, ASh5, AG1, ASh16, ASh15, ASh7).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Belgium, Britain I., Bulgaria, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Iceland, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Romania, Russia Central, Russia North, South Russia, Slovakia, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region (De Jong 2013).

Comment. Kheyri (1989) reported this species as a sugar beet pest in most sugar beet growing areas in Iran.

Macrosteles sexnotatus (Fallén, 1806)

Macrosteles sexnotatus: Emeljanov 1964: 507, fig. 182: 26, 27; Biedermann and Niedringhaus 2009: 288.

Material examined. Azarbaijan-e-Sharghi: 30♂♀, Sahand mountain, Kandovan, 2661 m, 37°45'47.7"N, 46°17'39.8"E, 1.September.2007, leg. Mozaffarian (Fig. 1, ASh10).

Abdollahi et al. (2013) reported this species from the above locality.

Worldwide distribution. East Palaearctic, Europe (Austria, Azores, Belgium, Britain I., Bulgaria, Canary Is., Cyprus, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Iceland, Ireland, Italian mainland, Latvia, Lithuania, Madeira, Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Russia Central, South Russia, Sardinia, Sicily, Slovakia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, North Africa (De Jong 2013).

Macrosteles sordidipennis (Stål, 1858)

Macrosteles sordidipennis: Emeljanov 1964: 507, fig. 182: 36, 37; Biedermann and Niedringhaus 2009: 290.

Material examined. Azarbaijan-e-Sharghi: 1♂, Sahand Mountain, Kandovan, 2661 m, 37°45'47.7"N, 46°17'39.8"E, 1.September.2007, leg. Mozaffarian (Fig. 1, ASh10).

Worldwide distribution. East Palaearctic, Europe (Austria, Britain I., Czech Republic, Danish mainland, Finland, Germany, Hungary, Norwegian mainland, Poland, Russia North, Sweden, The Netherlands) (De Jong 2013).

This species is newly recorded from Iran.

Tribe: Opsiini

Concavifer marmoratus Dlabola, 1960*

Localities. Zonuschay (Dlabola 1981) (Fig. 1, ASh5).

Worldwide distribution. Iran, Israel, Kazakhstan, Tadzhikistan (Nast 1972).

Neoaliturus fenestratus (Herrich-Schäffer, 1834)*

Localities. Tabriz (Dlabola 1981) (Fig. 1, ASh8).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Balearic Is., Belgium, Bulgaria, Canary Is., Cyprus, Czech Republic, Danish mainland, European Turkey, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Latvia, Lithuania, Moldova, Poland (doubtful), Portuguese mainland, Romania, Russia Central, Russia North, South Russia, Sardinia, Sicily, Slovakia, Spanish mainland, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, North Africa (De Jong 2013).

Neoaliturus haematoceps (Mulsant Rey, 1855)*

Localities. Marand, Zonuschay, Sufian, Maku (Dlabola 1981) (Fig. 1, ASh6, ASh5, ASh7, AG1).

Worldwide distribution. Afghanistan, Algeria, Austria, Canary Is., Cyprus, Czechoslovakia (Bohemia, Moravia, Slovakia), Egypt, France, German FR., Greece, Hungary, Iran, Italy (also Sardinia and Sicily), Jordan, Lebanon, Libya, Madeira Archipelago, Mongolia, Morocco, Poland, Romania, Spain, Syria, Tunisia, Turkey (Anatolia), Armenia, Azerbaijan, Georgia, Kazakhstan, Kirghizia, Moldavia, s.Russia, Turkmenia, Ukraine, Uzbekistan, Yugoslavia (Nast 1972).

Comment. Kheyri (1989) reported this species as an economic pest on sugar beet from Isfahan, Kerman, Fars, Khorasan, Azarbaijan and Karaj.

Neoaliturus opacipennis (Lethierry, 1876)*

Localities. Miyaneh-Zanjan, Bostanabad-Siah chaman, Siah chaman-Basmenj (Dlabola 1971) (Fig. 1, ASh16, ASh14, ASh13).

Worldwide distribution. Europe (Cyprus, French mainland, Germany, Greek mainland, Italian mainland, South Russia, Sardinia, Sicily, Spanish mainland, Switzerland, Ukraine), Near East, North Africa (De Jong 2013).

Comment. Kheyri and Alimoradi (1969) reported this species as a vector of curly top virus in Khorasan, Fars, Isfehan, Kerman, Ahvaz and Karaj.

Neoaliturus pulcher (Haupt, 1927)*

Localities. Zonuschay (Dlabola 1981) (Fig. 1, ASh5).

Worldwide distribution. Iran, Israel, Georgia, Kazakhstan, Tadzhikistan (Nast 1972).

Opsius cypriacus Lindberg, 1958*

Localities. Zonuschay (Dlabola 1981) (Fig. 1, ASh5).

Worldwide distribution. Europe (Cyprus, Greek mainland, Ukraine), Near East (De Jong 2013).

Opsius discessus (Horváth, 1911)*

Localities. Zonuschay, Marand (Dlabola 1981) (Fig. 1, ASh5, ASh6).

Worldwide distribution. East Palaearctic, Europe (South Russia), Near East (De Jong 2013).

Opsius pallasi (Lethierry, 1874)*

Localities. Zonuschay, Marand (Dlabola 1981) (Fig. 1, ASh5, ASh6).

Worldwide distribution. Europe (South Russia) (De Jong 2013).

Opsius scutellaris (Lethierry, 1874)*

Localities. Zonuschay (Dlabola 1981) (Fig. 1, ASh5).

Worldwide distribution. Afro-tropical region, East Palaearctic, Europe (Canary Is.), Near East, North Africa (De Jong 2013).

Pseudophlepsius binotatus (Signoret, 1880)*

Localities. Zonuschay, Sufian (Dlabola 1981) (Fig. 1, ASh5, ASh7).

Worldwide distribution. Europe (South Russia) (De Jong 2013).

Tribe: Paralimnini

Mogangella straminea Dlabola, 1957

Mogangella straminea: Emeljanov 1964: 541, fig. 194: 13, 14.

Material examined. Azarbaijan-e-Sharghi: 1♂ 1♀, Marand, 12.July.2007, Light trap, leg. Lotfalizadeh (Fig. 1, ASh6).

Worldwide distribution. East Palaearctic, Europe (Moldova, Ukraine), Near East (De Jong 2013).

This species is newly recorded from Iran.

Paramesus major Haupt, 1927*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. East Palaearctic, Europe (Austria, Bulgaria, Czech Republic, Germany (doubtful), Greek mainland, Hungary, Poland, South Russia, Yugoslavia), Near East (De Jong 2013).

Paramesus paludosus Ribaut, 1952*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. France, Italy, Kazakhstan, Moldavia, Ukraine (Nast 1972).

Psammotettix alienus (Dahlbom, 1850)

Psammotettix alienus: Ribaut 1952: 243, figs 579–580; Emeljanov 1964: 541, fig. 194: 8, 9; Biedermann and Niedringhaus 2009: 337.

Material examined. Ardabil: 4♂♀, Moghan, 18.May.1978, leg. Abaii (Fig. 1, A1).

Azarbaijan-e-Sharghi: 3♂♀, Marand, 1610 m, 28.July.1976, leg. Broumand & Pazouki (Fig. 1, ASh6).

Azarbaijan-e-Sharghi: 1♀, Tabriz, Gharachaman, 1600 m, 16.July.1976, leg. Broumand & Pazouki (Fig. 1, ASh8).

Dlabola (1981) reported this species from Marand, Sufian and Zonuschay, Maku and in 1971 from Tabriz-Bostanabad, Miyaneh-Zanjan, Miyaneh-Siah chaman, Siah chaman-Basmenj (Fig. 1, ASh6, ASh7, ASh5, AG1, ASh12, ASh16, ASh15, ASh13).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Belgium, Bulgaria, Canary Is., Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Latvia, Lithuania, Madeira, Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Russia Central, Russia North, South Russia, Sicily, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, Ukraine, Yugoslavia), Near East, Nearctic region, North Africa (De Jong 2013).

Comment. Nematollahi and Khajehali (2000) reported this species as a vector for wheat dwarf virus on *Zea* (maize) in Isfahan.

Psammotettix pictipennis (Kirschbaum, 1868)*

Localities. Miyaneh-Zanjan (Dlabola 1971); Marand, Sufian (Dlabola 1981) (Fig. 1, ASh16, ASh6, ASh7).

Worldwide distribution. East Palaearctic, Europe (Austria, Bulgaria, Greek mainland, Hungary, Moldova, Romania, South Russia, Slovenia, Spanish mainland, Ukraine, Yugoslavia), Near East (De Jong 2013).

Psammotettix seriphidii Emeljanov, 1962

Psammotettix seriphidii: Emeljanov 1964: 539, fig. 193: 1, 2.

Material examined. Ardabil: 87♂♀, 12 km to Khalkhal, 1998 m, 37°35'41.8"N, 48°37'54.3"E, 17.Janaury.2007, leg. Mozaffarian, light trap (Fig. 1, A5).

Ardabil: 1♂, 10 km to Parsabad, 80 m, 39°36'8.3"N, 47°48'45.5"E, 18.January.2007, leg. Mozaffarian (Fig. 1, A2).

Ardabil: 3♂♀, Moghan, 65 m, 39°37'30.7"N, 47°46'57.5"E, 19.January.2007, leg. Mozaffarian (Fig. 1, A1).

Azarbaijan-e-Sharghi: 6♂♀, AjabShir, Yaichi village, 1922 m, 37°35'27.2"N, 46°11'03.7"E, 15.January.2008, leg. Mozaffarian (Fig. 1, ASh11).

Azarbaijan-e-Sharghi: 2♂♀, Kaleibar, 1732 m, 38°54'25.2"N, 47°09'11.5"E, 3.September.2007, leg. Mozaffarian (Fig. 1, ASh2).

Azarbaijan-e-Sharghi: 1♂, Kandovan, 2645 m, 37°45'45.8"N, 46°17'40.5"E, 18.January.2008, leg. Mozaffarian (Fig. 1, ASh10).

Azarbaijan-e-Sharghi: 20♂♀, Kandovan, 1978 m, 37°44'15.8"N, 46°19'55.1"E, 18.January.2008, leg. Mozaffarian (Fig. 1, ASh10).

Azarbaijan-e-Sharghi: 4♂♀, Eskanlu, Aras river, 290 m, 39°12'13.4"N, 47°04'23.2"E, 3.September.2007, leg. Mozaffarian & Nematian (Fig. 1, ASh1).

Azarbaijan-e-Gharbi: 11♂♀, Uromieh, MirzaAbad, 1450 m, 21.July.1976, leg. Boroumand & Pazouki (Fig. 1, AG4).

Worldwide distribution. Kazakhstan (Nast 1972).

This species is newly recorded from Iran.

Sorhoanus medius (Mulsant Rey, 1855)*

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7)

Worldwide distribution. Bulgaria, France, Italy, Switzerland, Altai Mts., Kazakhstan, Kirghizia, Russia, Siberia, Ukraine, Yugoslavia (Nast 1972).

Tribe: Phlepsiini

Phlepsius intricatus (Herrich-Schäffer, 1838)

Phlepsius intricatus: Emeljanov 1964: 516, fig. 185: 5, 6; Biedermann and Niedringhaus 2009: 305.

Material examined. Azarbaijan-e-Sharghi: 2♂, Tabriz, Khosroshahr, 1346 m, 37°58'28"N, 46°02'55"E, 21-30.August.2007, leg. Lotfalizadeh, Malaise trap (Fig. 1, ASh8).

Dlabola (1981) reported this species from Zonuschay and in 1974 from Uromieh (Fig. 1, ASh5, AG3).

Worldwide distribution. Europe (Albania, Austria, Balearic Is., Bulgaria, Czech Republic, European Turkey, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Moldova, Portuguese mainland, Romania, South Russia, Sardinia, Sicily, Slovakia, Slovenia, Spanish mainland), Near East, North Africa (De Jong 2013).

Tribe: Scaphoideini

Anoplotettix magnificus Emeljanov, 1962 *

Localities. Sufian (Dlabola 1981) (Fig. 1, ASh7).

Worldwide distribution. Azarbaijan, Georgia (Nast 1972).

Subfamily: Dorycephalinae

Tribe: Eupelicini

Eupelix cuspidata (Fabricius, 1775)

Eupelix cuspidata: Ribaut 1952: 325, figs 868–871.

Material examined. Azaibaijan-e-Sharghi: 4♂♀, Ajabshir, Yaichi village, 1922 m, 37°35'27.2"N, 46°11'03.7"E, 15.January.2008, leg. Mozaffarian (Fig. 1, ASh11).

Dlabola (1981) reported this species from Zonuschay, Marand and Sufian (Fig. 1, ASh5, ASh6, ASh7).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Balearic Is., Belgium, Britain I., Bulgaria, Canary Is., Corsica, Croatia, Cyprus, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Lithuania, Moldova, Norwegian mainland, Poland, Portuguese mainland, Romania, Russia Central, Russia North, South Russia, Sardinia, Sicily, Slovakia, Slovenia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, North Africa (De Jong 2013).

Paradorydium aristidae (Zachvatkin, 1953)*

Localities. Zonuschay, Maku (Dlabola 1981) (Fig. 1, ASh5, AG1).

Worldwide distribution. East Palaearctic, Europe (South Russia, Ukraine), Near East (De Jong 2013).

Subfamily: Iassinae

Tribe: Iassini

Batracomorphus irroratus Lewis, 1834*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. East Palaearctic, Europe (Albania, Austria, Belgium, Britain I., Bulgaria, Czech Republic, Danish mainland, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Lithuania, Moldova, Poland, South Russia, Slovakia, Switzerland, Ukraine, Yugoslavia), Near East (De Jong 2013).

Subfamily: Idiocerinae

Tribe: Idiocerini

Rhytidodus resaicus Dlabola, 1974*

Localities. Uromieh (Dlabola 1974) (Fig. 1, AG3)

Subfamily: Macropsinae

Tribe: Macropsini

Hephathus unicolor (Lindberg, 1926)*

Localities. Zonuschay (Dlabola 1981) (Fig. 1, ASh5).

Worldwide distribution. East Palaearctic, Europe (Romania (doubtful), South Russia (doubtful), Ukraine (doubtful), Yugoslavia (doubtful)), Near East (De Jong 2013).

Hephathus freyi (Fieber, 1868)*

Localities. Siah chaman-Miyaneh (Dlabola 1971) (Fig. 1, ASh15).

Worldwide distribution. Europe (Balearic Is., Bulgaria, French mainland, Greek mainland (doubtful), Italian mainland, Portuguese mainland, Sicily, Slovakia, Spanish mainland, Yugoslavia), Near East, North Africa (De Jong 2013).

Subfamily: Typhlocybinae

Tribe: Empoascini

Empoasca punjabensis Singh-Pruthi, 1940*

Localities. Zonuschay, Maku (Dlabola 1981); Siah chaman-Miyaneh, Miyaneh-Zanjan, Tabriz-Shabestar, Uromieh-Sarv (Dlabola 1971) (Fig. 1, ASh5, AG1, ASh15, ASh16, ASh7, AG2).

Worldwide distribution. Europe (Bulgaria, French mainland, Greek mainland, South Russia, Ukraine, Yugoslavia), Near East, Oriental region (De Jong 2013).

Comment. Kheyri (1989) reported this species as an economic pest on sugar beet from Isfahan, Kerman, Fars and Karaj.

Kyboasca bipunctata (Oshanim, 1871)*

Localities. Miyaneh- Siah chaman, Tabriz-Shabestar, Miyaneh-Zanjan (Dlabola 1971); Sufian (Dlabola 1981) (Fig. 1, ASh15, ASh7, ASh16, ASh7).

Worldwide distribution. East Palaearctic, Europe (Austria, Bulgaria, Czech Republic, Danish mainland, Finland, Germany, Hungary, Italian mainland, Moldova, Poland, Romania, South Russia, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region (De Jong 2013).

Tribe: Erythroneurini

Tamaricella ribauti (Zachvatkin, 1947)*

Localities. Zonuschay (Dlabola 1981) (Fig. 1, ASh5).

Worldwide distribution. Europe (Crete, South Russia, Ukraine) (De Jong 2013).

Tamaricella tamaricis (Puton, 1872)*

Localities. Miyaneh-Zanjan, Miyaneh-Siah chaman (Dlabola 1971) (Fig. 1, ASh16, ASh15).

Worldwide distribution. Europe (Bulgaria, Crete, Cyclades Is., Cyprus, French mainland, Greek mainland, Italian mainland, Romania, South Russia, Sardinia, Sicily, Spanish mainland, Ukraine) (De Jong 2013).

Zyginidia pullula (Boheman, 1845)*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. Europe (Albania, Austria, Bulgaria, Corsica (doubtful), Czech Republic, Danish mainland, Finland, French mainland, Germany, Greek mainland, Hungary, Italian mainland, Romania, Slovakia, Spanish mainland, Sweden, Switzerland, Ukraine, Yugoslavia), Near East (De Jong 2013).

Zyginidia sohrab Zachvatkin, 1947*

Localities. Miyaneh-Zanjan, Siah chaman-Miyaneh, Uromieh-Sarv (Dlabola 1971) (Fig. 1, ASh16, ASh15, AG2).

Worldwide distribution. Europe (Cyprus, Greek mainland, South Russia, Ukraine), Near East (De Jong 2013).

Tribe: Typhlocybini

Edwardsiana rosae (Linné, 1758)*

Localities. Siah chaman-Miyaneh (Dlabola 1971) (Fig. 1, ASh15).

Worldwide distribution. East Palaearctic, Europe (Austria, Belgium, Britain I., Bulgaria, Cyprus, Czech Republic, Danish mainland, Estonia, Finland, French mainland, Germany, Greek mainland, Hungary, Ireland, Italian mainland, Latvia, Moldova, Norwegian mainland, Poland, Romania, Russia Central, Russia North, South Russia, Sicily, Slovakia, Spanish mainland, Sweden, Switzerland, The Netherlands, Ukraine, Yugoslavia), Near East, Nearctic region, Oriental region (De Jong 2013).

Subfamily: Ulopinae

Tribe: Ulopini

Ulopa trivia Germar, 1821*

Localities. Marand (Dlabola 1981) (Fig. 1, ASh6).

Worldwide distribution. Albania, Austria, Belgium, Bulgaria, Cyprus, Czechoslovakia (Bohemia, Moravia, Slovakia), Denmark, France, German DR, German FR, Great Britain (England), Greece, Hungary, Italy, Palestine, Poland, Portugal, Romania, Spain, Turkey (Anatolia), Azerbaijan, Armenia, Georgia, Moldavia, Russia, Ukraine, Yugoslavia (Nast 1972).

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First record of Jacobsoniidae (Coleoptera) from China with description of a new species of *Sarothrias* Grouvelle

Wen-Xuan Bi[1,2], Chang-Chin Chen[3], Mei-Ying Lin[1]

1 *Key Laboratory of Zoological Systematics and Evolution, Institute of Zoology, Chinese Academy of Sciences, Beichen West Road, Chaoyang, Beijing, 100101, China* **2** *Room 401, No. 2, Lane 155, Lianhua South Road, Shanghai, 201100, China* **3** *NPS office, Tianjin New Wei San Industrial Company, Limited, China*

Corresponding authors: *Wen-Xuan Bi* (insectb@163.com); *Mei-Ying Lin* (linmeiying@ioz.ac.cn)

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Abstract

The family Jacobsoniidae Heller (短跗甲科) is newly recorded from China upon the discovery of *Sarothrias sinicus* Bi & Chen, **sp. n.** (中华短跗甲) from Motuo, Southeast Xizang. Description and illustrations of the habitus and major diagnostic features of the new taxon are provided. A key to the species of *Sarothrias* and some ecological notes on the new species are presented.

Keywords

Jacobsoniidae, *Sarothrias*, new record, new species, taxonomy, Oriental region

Introduction

The family Jacobsoniidae Heller, 1926 was considered *incertae sedis* within the series Bostrichiformia by Löbl and Burckhardt (1988), Lawrence and Newton (1995) and Lawrence et al. (1999a, b). Lawrence et al. (2010) provisionally placed Jacobsoniidae within the series Derodontiformia, along with families Derodontidae and Nosodendridae. In the current study, the results of a cladistic analysis based on morphological characters of adults and larvae indicated that Jacobsoniidae is sister to part of the Staphylinoidea (Lawrence et al. 2011).

Jacobsoniidae includes 21 known species in three genera: *Sarothrias* Grouvelle, 1918, *Saphophagus* Sharp, 1886 and *Derolathrus* Sharp in Sharp & Scott, 1908 (Háva and Löbl 2005). They can be recognized by the minute (0.65–2.5 mm), narrowly elongate (about 2.1–3 times as long as wide), yellowish brown body, an elongate prothorax, lack of a visible scutellum, and markedly elongate metaventrite (at least 2.5 times as long as mesoventrite). All species are poorly represented in collections and little is known about their biology (Lawrence and Leschen 2010; Philips et al. 2002).

Currently, 13 species of *Sarothrias* have been described (Háva and Löbl 2005; Pal 1998). They are restricted to humid tropical areas which close to the equator with the exception of *S. hygrophilus* Pal, 1998 from northeast India. They can be primarily defined and separated from *Saphophagus* by tarsal formula 3-3-3 instead of 5-5-5, and from *Derolathrus* by antennal club 3-segmented instead of 1- or 2-segmented (Burckhardt and Löbl 1990; Lawrence and Leschen 2010).

During July to August 2014, the first author participated in an expedition to Motuo (=Mêdog), Xizang, the third time that he visited that area. A small but remarkable beetle collected during this expedition represented an interesting, unexpected result in that it is an undescribed species of *Sarothrias* which belongs to the Jacobsoniidae – a family so far not recorded from China. In this paper, we describe a new species, *Sarothrias sinicus* Bi & Chen, sp. n., based on this specimen. An identification key to the species of *Sarothrias* is also included. Based on Lin and Yang (2012), Jacobsoniidae is the 147th family of Coleoptera recorded from China.

Results

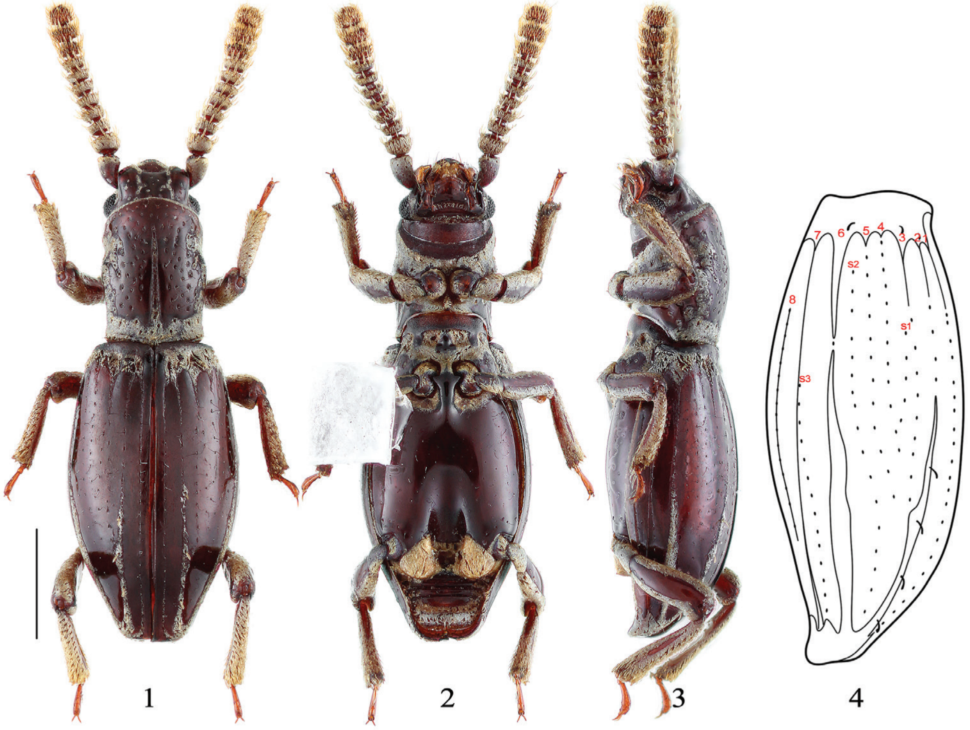
Sarothrias sinicus Bi & Chen, sp. n.

http://zoobank.org/69D14BBA-16B8-49AC-8E45-AA254FB6C862
Figs 1–5

Type material. Holotype: male, "China: Xizang, Motuo / Baricun / 2014.VII.27 1850 m / leg. Wen-Xuan Bi" [white label printed]. The holotype was deposited in the Insect Collection of Shanghai Normal University, Shanghai, China (SNUC).

Diagnosis. This new species can be separated from most congeners by the elytra, which are predominantly shiny, and elytral row 5 unimpressed; the pronotum is devoid of squamiform setae. It differs from *S. hygrophilus* Pal by the elytra, with row 1 largely represented by fine punctures, row 2 interrupted by punctures after basal quarter, and with supplementary series (s1) between rows 3 and 4; the pronotum has secretions on the lateral margins instead of on the side below the protrusions. It also resembles *S. papuanus* Ślipiński but can be easily distinguished by the elytra, bearing four squamiform setae in the distal half of row 2, row 3 merging with row 2 and with s1 between rows 3 and 4; the pronotum is devoid of secretions.

Description. Male (Figs 1–3). Body length 2.20 mm, elongate, convex, dark-brown, dorsal surface largely shiny except whitish secretions in parts of pronotum and



Figures 1–4. Habitus of *Sarothrias sinicus* Bi & Chen, sp. n., Holotype, male. **1** dorsal view **2** ventral view **3** lateral view **4** left elytron in three-quarter view (s1 to s3: supplementary series, s2 = s in Poggi 1991). Scale bar: 0.5 mm (**4** not to scale).

elytra; legs, excluded tarsi, pubescent and covered with yellowish secretions except two narrow longitudinal shiny band at both sides of femora and tibiae, tarsi brown; wings fully developed.

Head broader than long, width across eyes 0.45 mm, clypeus smooth, rounded anteriorly, fronto-clypeal suture clearly visible; frons slightly broad anteriorly with sides moderately raised, sparsely and distinctly setigerous punctured; eyes large, rounded, nearly as long as half of head length, coarsely facetted. Antenna length 0.88 mm, scape slightly elongate, antennomeres 2–11 transverse, with secretions on antennomeres 1–8, squamiform setae at apex of scape and on antennomeres 2–10 and normal setae on antennomeres 9–11.

Pronotum length 0.67 mm, width 0.48 mm, subparallel-sided, widest near middle; anterior margin rounded; disc with a shallow median groove, extending from the anterior one-fifth to little above base, slightly broad posteriorly; punctures on disc of similar size, shape and distribution to those on head; median groove, lateral margins and one-sixth of pronotal base with secretions. Scutellum invisible.

Elytra length 1.38 mm, width 0.79 mm, fusiform, widest slightly after middle; basal transverse bulge well developed, with subbasal band of secretions on which three

Figure 5. Habitat of *Sarothrias sinicus* Bi & Chen, sp. n., taken on the way from Baricun to Renqinbeng, Motuo, Xizang, alt. 1850 m.

subbasal depressions on each elytron, each depression with one squamiform setae posteriorly. Each elytron with striae or fine puncture forming 8 rows and 3 supplementary series (Fig. 4) of which 6 rows and 2 supplementary series are visible in dorsal view; rows 1 to 3 impressed at basal quarter and continued as a row of fine punctures, row 2 once again impressed after middle and connected with row 1 subapically, row 3 joined row 2 at apical one-third, rows 4 and 5 represented by fine punctures and disappearing anterior to apical one-third, row 6 largely impressed but intercepted by single puncture at basal one-third, rows 7 and 8 impressed, of which the former extending subapically and the latter starting at basal quarter and extending half of elytra length; three supplementary series (s1, s2, s3) represented by fine punctures, of which s1 present between rows 3 and 4 which starting at basal quarter and joined row 4 anterior to basal half, s2 (=s in Poggi 1991) present between rows 5 and 6 which starting at basal one-sixth and ending subapically, s3 separate from row 7 at basal two-fifths and ending subapically between row 6 and 7; apical half of row 2 and entire length of row 6 with secretions, of which the former secretionary row with four squamiform setae asymmetrically arranged.

Prosternum and mesoventrite largely covered with secretions. Metaventrite covered with the same secretions after mesocoxal insertions (cavities) and between metacoxal insertions, other parts shiny with very sparser and fine punctures; with deep median impression in apical half which with rounded sides and not delimited by lateral ridges. Legs moderately long, tarsi 3-segmented.

Ecological notes. Adult *Sarothrias* have been collected in leaf litter and rotten wood, but the larvae are as yet unknown (Lawrence and Leschen 2010). The only specimen of the new species was collected by beating a branch with dead leaves on the way from Baricun (= Bari village) to Renqinbeng (= Renqinbeng Temple), Motuo, Xizang at altitude of 1850 m (Fig. 5). The vegetation types are subtropical evergreen broadleaf forest which has been well protected, although slightly disturbed by locals by grazing. A variety of staphylinids, endomychids and cerambycids were collected at the same time. Another mysterious termitophilous lucanid, *Penichrolucanus cryptonychus* (Zhang, 1988), which is located in the same area and known only from its original description shares a similar generic distribution with *Sarothrias*.

Distribution. China: Xizang (Tibet) Autonomous Region, Motuo County.

Etymology. The new species is named after the country of the type locality.

Key to the species of *Sarothrias*

(modified from Pal 1998)

1 Elytra entirely mat and covered by secretions, except a narrow shiny stripe along the suture and which is devoid of secretions **2**

– Elytra partly shiny, not completely covered by secretions **6**

2 Terminal antennomere with a whorl of squamiform setae as on antennomeres 2 to 10 .... **3**

– Terminal antennomere devoid of a whorl of squamiform setae **4**

3 Sides of pronotum with 2–3 squamiform setae; antennomere 2 twice as long as antennomere 3. Sabah .... ***S. crowsoni*** **Löbl & Burckhardt, 1988**

– Sides of pronotum devoid of setae; antennomere 2 about 1.4× as long as antennomere 3. Moluccas .... ***S. audax*** **Ślipiński & Löbl, 1995**

4 Epipleural keel of elytra extending more towards base than lateral keel **5**

– Epipleural and lateral keels of elytra ending at about the same level. New Britain .... ***S. boumei*** **Ślipiński, 1986**

5 Median depression of metaventrite indistinctly delimited, narrowing towards apex beyond middle; mat, covered by secretions. Seychelles .... ***S. eximius*** **Grouvelle, 1918**

– Median depression on metaventrite well delimited laterally , gradually narrowing towards apex; apical portion shiny, devoid of secretions. Fiji .... ***S. fijianus*** **Löbl & Burckhardt, 1988**

6 Secretions on head and pronotum strongly expanded, those on elytra forming longitudinal stripes which are separated by shiny stripes. South India .... ***S. indicus*** **Dajoz, 1978**

– Dorsal surface of body predominantly shiny; pattern formed by secretions on elytra different 7

7 Pronotum with 2–3 squamiform setae on sides **8**

– Pronotum devoid of squamiform setae **9**

8 Elytral secretions forming apical drop-shaped loop; antennomere 11 with squamiform setae. Sumatra .... ***S. dimerus*** **(Heller, 1926)**

– Elytral secretions strongly reduced; antennomere 11 without squamiform setae. New Caledonia............................. ***S. pacificus*** **Ślipiński & Löbl, 1995**

9 Elytron with row 5 entirely deeply impressed, merged. with row 4 apically. Queensland .... ***S. lawrencei*** **Löbl & Burckhardt, 1988**

– Elytron with row 5 at most impressed near base and then continued as separate punctures, not joined with row 4 .... **10**

10 Elytron with rows 1 and 3 impressed only at base and then continued as a row of separate punctures **11**

– Elytron with rows 1 and 3 well impressed, row 3 with impression at least surpassing middle of elytron **12**

11 Elytron with row 2 presented two squamiform setae apically, disconnected with row 3; devoid of s1. Papua New Guinea .... ***S. papuanus*** **Ślipiński, 1986**

– Elytron with row 2 presented four squamiform setae after middle, connected with row 3; with s1 between rows 3 and 4 (Fig. 4). Southwest China .... ***S. sinicus*** **sp. n.**

12 Rows 2 and 3 of elytron entirely impressed, join far before level of metacoxae and at level of last puncture of row 5. Malaysia... .... ***S. amabilis*** **Ślipiński & Löbl, 1995**

– Rows 2 and 3 of elytron join near the level of metacoxae or behind it, further below level of last puncture of row 5, row 3 completely or incompletely impressed.. **13**

13 Row 3 of elytron entirely impressed before joining with row 2. New Guinea..... .... ***S. morokanus*** **Poggi, 1991**

– Row 3 of elytron impressed but interrupted just before joining with row 2, where it is represented by separate punctures. Northeast India..................... ....***S. hygrophilus*** **Pal, 1998**

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Re-establishment of *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 as a separate valid species (Coleoptera, Carabidae)

Claudio Ghittino[1], Enrico Busato[2], Achille Casale[3]

1 *Istituto Zooprofilattico Sperimentale dell'Umbria e delle Marche (IZSUM), Via Dalla Chiesa 78, 05100 Terni, Italy* **2** *Dipartimento di Scienze Agrarie, Forestali e Alimentari (DISAFA), Università di Torino, Largo Braccini 2, 10095 Grugliasco (TO), Italy* **3** *c/o Dipartimento di Scienze della Natura e del Territorio (DIPNET), sezione Zoologia, Università di Sassari, Via Muroni 25, 07100 Sassari, Italy*

Corresponding author: *Claudio Ghittino* (c.ghittino@izsum.it)

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Abstract

Carabus (*Cathoplius*) *aliai* was described as a separate species by Escalera in 1944 but since the 1950–60s it has been considered as a subspecies of *C.* (*Cathoplius*) *stenocephalus* Lucas, 1866. This downgrading was adopted after examining only a few specimens, due to their rarity in collections. In recent years, an important population of this taxon was rediscovered in the Tan-Tan area in southern Morocco. By combining field observations with laboratory breeding experiments including hybridization trials, and through the morphological examination of a representative number of individuals, it is confirmed that *C. aliai* is indeed a valid species. Despite close geographic distribution, the morphological and biological characteristics of *C. aliai* and *C. stenocephalus ifniensis* Zarco, 1941, its northern substitutive taxon, are very different. *Carabus aliai* adults are characterized by a smaller size, a slender silhouette, a more brilliant aspect, a narrower pronotum, a coarser elytral sculpture, longer legs, and a wider and a little more curved apex of the median lobe of the aedeagus. *Carabus aliai* larvae are also characterized by a much smaller size and the *C. aliai* pupa has a narrower thoracic area and a different chaetotaxy compared to that of *C. stenocephalus ifniensis*. Contrary to this, *C. aliai* has a life cycle belonging to the annual univoltine winter semelparous type. Moreover, the duration of its development cycle is shorter. *Carabus aliai* is a sabulicolous steppe-wandering species with an intensive running activity, while *C. stenocephalus ifniensis* is a more sedentary taxon. Crossbreeding experiments showed a marked reproductive isolation between *C. aliai* and *C. stenocephalus ifniensis*. When F1 hybrids were crossed with one another, a very high mortality rate during embryonic, larval and pupal development was evident and no vital F2 neo-adults were obtained.

Morphological and biological differences, together with the reproductive failure in *C. aliai* × *C. stenocephalus ifniensis* hybrids, clearly indicate that *C. aliai* is a separate *Cathoplius* species that is distributed in an area south of the Anti-Atlas chain, from Plage Blanche (Guelmim) to Lemsid and Bou Kra (south of Laâyoune). *Carabus aliai* is therefore both a Saharan desert endemic and an Atlantic resident. Moreover, it is the southernmost *Carabus* species of the western Palaearctic region.

Keywords

Carabus ground beetles, Saharan desert endemism, Atlantic element, life cycle, hybridization

Introduction

The subgenus *Cathoplius* C.G. Thomson, 1875 within the Genus *Carabus* Linnaeus, 1758 forms a very homogenous and peculiar lineage of strictly snail-predating ground beetles adapted to live in arid habitats with scarce xerophilous vegetation along the Moroccan Atlantic coast (Busato et al. 2014). This subgenus has been postulated as the only representative of Cathopliogenici, a possible sister group of the "Neocarabi" of Bengtsson (Deuve et al. 2012).

Carabus (*Cathoplius*) *aliai* is a desert-dwelling taxon that was first described by Manuel Martinez de la Escalera in August 1944. This description was based on the examination of only one specimen, a female found by the Spanish geologist Manuel Alía in April 1943 in El Khaloua which is 15 km north of present Abteh, in former Cape Juby strip, southern territories of the Spanish protectorate in Morocco (Escalera 1944). At approximately the same time (September 1944), it was also described by Francisco Español and named as *C.* (*Cathoplius*) *mateui*. This description was based on three specimens, a female found by Joaquin Mateu in February 1944 in Agti Baba Ali in the Sabkhat Tislatine area, 50 km southwest of present Laâyoune, in former Saguia el Hamra, Spanish Sahara, and a male and a female found in May 1944 in Tan-Tan, in former Cape Juby strip, in the southern territories of the Spanish protectorate in Morocco (Español 1944).

Dr Joaquin Mateu, to whom this contribution is dedicated, really discovered and first studied *C. aliai*. In November 1944, he collected a further seven specimens (five males and two females) south of Laâyoune, in an area comprised between Izik plateau, Lemsid (Ougnit) and Metmarfag (Asreifa) to the west and Bou Kra to the east. This area is characterized by the presence of "graras", land depressions where water cumulates during the raining season, allowing a certain vegetation to grow and consequently snails to dwell in. It was here that *C. aliai* specimens were observed during early morning or late afternoon while running, mating or eating snails of the family Helicidae. In addition to the biogeographic and biological notes, in his paper on the carabid beetles of Spanish Sahara, Mateu (1947) established *C. mateui* as a junior synonym of *C. aliai*.

Some years later, French entomologists dealing with the Moroccan carabid fauna reconsidered *C. aliai* as a subspecies of *C.* (*Cathoplius*) *stenocephalus* Lucas, 1866 (Antoine 1955; Kocher 1963). This downgrading was adopted after examining one of the specimens collected by Mateu in 1944, plus another doubtful specimen found by

André Reymond in 1954 at Aoreora, Plage Blanche (75 km west of Guelmim / 30 km north of the Oued Drâa outlet, in the southern part of the former French protectorate in Morocco). As far as we know, few other specimens of *C. aliai* have been found until December 2003, when several individuals of this taxon were sampled by Jaroslav Kaláb in the vicinity of the city of Tan-Tan, Tan-Tan province, southern Morocco.

Modern catalogues and checklists of the Genus *Carabus* either consider *C. aliai* as a "strong" subspecies of *C.* (*Cathoplius*) *stenocephalus* (Deuve 1994; 2004; 2012; 2014), together with the "weak" subspecies *escalerae* Csiki, 1927, *susicus* Antoine, 1941 and *ifniensis* Zarco, 1941, or as a subspecies of *C.* (*Cathoplius*) *asperatus* (Dejean, 1826) in the broad sense (Březina 1994; 1999; Bousquet et al. 2003; Lorenz 2005). In this study, we collected data from field observations and laboratory breeding experiments, including hybridization trials with the northern substitutive taxon *C. stenocephalus ifniensis*. By combining this information with morphological examinations of a representative number of individuals, both pre–imaginal stages and adults, it was concluded that *C. aliai* is indeed a separate species, as considered in the original descriptions (Escalera 1944; Español 1944) and as hypothesized in our previous contribution (Busato et al. 2014).

Methods

Field observations on *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 are the results of nine surveys (March–April 1992; December 1999; January 2000; January, February–March & April 2006; November 2008; January & December 2010) carried out in the Guelmim, Tan-Tan and Laâyoune areas, southern Morocco. From December 14–19, 2010, one of the authors (C. G.) sampled 11 adult specimens (7 males and 4 females) and 10 larvae (1 first instar and 9 second instars) of this taxon on the Tan-Tan northern plateau (Hameidia Tellia: 2 km west of Tan-Tan city, 90–110 m a.s.l., 28°27'13"N; 11°07'53"W, and 7 km west of Tan-Tan city, 190 m a.s.l., 28°26'21"N; 11°10'30"W, in Ben Khlil rural commune, Tan-Tan province). In addition, observations on the behavior of specimens in their natural environment were recorded.

Life cycle investigations included the maintenance and breeding of *C. aliai* adults and larvae in the laboratories of the State Veterinary Institute (IZSUM) in Terni, Italy. Breeding activities started on 21 December 2010 and ended on 30 April 2013. The breeding methodology used was basically that reported by Malausa (1977). Insects were exposed to varying temperatures (from 15/19 °C in December to 17/21 °C in April, night/day) and to a gradually increasing photoperiod (from 9 to 14 hours of daylight).

Carabus aliai mating pairs were maintained in a 50 × 30 × 30 cm (length × width × height) transparent terrarium with a perforated top for aeration. A 15 cm substrate, composed of a mixture of argillaceous soil (40%) and sand (60%) was placed on the bottom of each terrarium. One third of the substrate surface was covered with a layer of moss. The substrate was kept moist by periodic spraying with water. Adults were *ad libitum* fed with live *Theba pisana pisana* (O.F. Müller, 1774) snails. In order to compare their life cycles, *C.* (*Cathoplius*) *stenocephalus ifniensis* adults (previously collected on the beach of Sidi Ifni city, 10 m a.s.l., 29°23'52"N; 10°10'40"W, Sidi Ifni province, central Morocco) were bred in parallel with *C. aliai*. Two mating pairs of both taxa were introduced in the terrariums on 21 December 2010 and two others on 1 December 2011 as a control group.

Larvae were reared individually in cylindrical opaque plastic containers (10 cm in diameter, 13 cm height) filled with 9 cm of the same substrate and kept at the same temperature as adults. Larvae were fed with live *T. pisana pisana* snails of the appropriate size (shell width 10–20 mm). In each container, a piece of bark [5 × 3 × 1(h) cm] was placed on top of the substrate as a shelter for the larva. Neo-adults were kept in mini-terrariums [20 × 12 × 15(h) cm] with a 5 cm substrate and a layer of moss, and *ad libitum* fed with live *T. pisana pisana* snails until their complete sclerification.

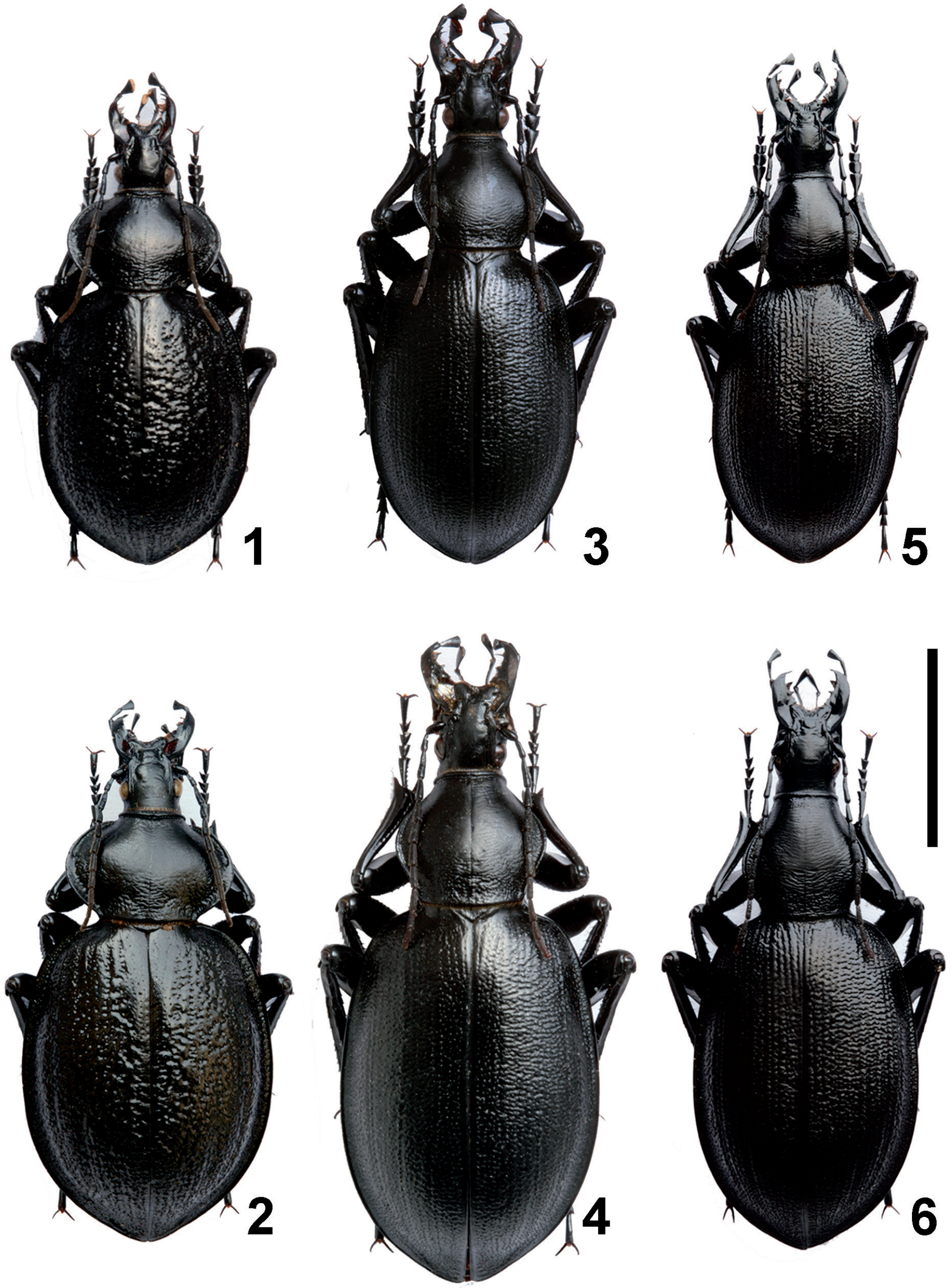
Following pure breeding, hybridization experiments between virgin *C. aliai* and *C. stenocephalus ifniensis* were carried out. Four mating pairs of the combination *C. aliai* ♂ × *C. stenocephalus ifniensis* ♀ (AIF1) and four of the combination *C. stenocephalus ifniensis* ♂ × *C. aliai* ♀ (AIF2) were bred at the same temperatures and at the same photoperiod previously reported, starting from December 1, 2011. Due to the different size between *C. aliai* (generally smaller) and *C. stenocephalus ifniensis* (generally larger), small females of *C. stenocephalus ifniensis* and large males of *C. aliai* were used to obtain AIF1 hybrid, while large females of *C. aliai* and small males of *C. stenocephalus ifniensis* were used to obtain AIF2 hybrid. Once obtained, AIF1 and AIF2 hybrids were then crossed with one another to investigate their fecundity. The F1 crossbreeding experiments were done in quadruplicate starting on December 3, 2012.

Representative specimens of *C. aliai* and *C. stenocephalus ifniensis* used for comparative morphological studies are conserved dry or under alcohol in authors' collections.

Results

Morphology of adults. Morphological differences observed among adults of the various *Cathoplius* taxa were in agreement with those reported by Antoine (1955) (Figures 1–6). The size of *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 and *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941 adults was found to be a peculiar feature: *C. aliai* specimens were in general smaller (shorter and narrower), while specimens of its northern substitutive *C. stenocephalus ifniensis* were larger (longer and wider). The total length from apex of mandibles to apex of elytra (L) was 26.5–33.0 mm in *C. aliai* (males 26.5–28.5 mm, females 27.0–33.0 mm) and 28.0–36.0 mm in *C. stenocephalus ifniensis* (males 28.0–30.0, females 30.0–36.0 mm). The general aspect was also found to be very peculiar: *C. aliai* specimens were bright black in color on their dorsal side, while specimens of *C. stenocephalus ifniensis* were dull black.

The head was slender in *C. aliai* and broad in *C. stenocephalus ifniensis*. The pronotum was narrow and elongated, longer than wide, with a very reduced lateral furrow in *C. aliai*, while it was transverse, wider than long, with a developed lateral furrow in *C. stenocephalus ifniensis*. On average, the ratio between length of pronotum and



Figures 1–6. *Cathoplius* species adults. Habitus of *Carabus asperatus* (Dejean, 1826) from Oualidia (**1** male **2** female) *C. stenocephalus ifniensis* Zarco, 1941 from Sidi Ifni (**3** male **4** female) and *C. aliai* Escalera, 1944 from Tan-Tan (**5** male **6** female). Scale bar: 1 cm.

maximum width of pronotum (PL/PW) was 6.0/5.5 mm = 1.09 in *C. aliai* and 5.5/7.0 mm = 0.79 in *C. stenocephalus ifniensis*. In both taxa the prosternal apophysis was elongated, but while its apex was subquadrate in *C. stenocephalus ifniensis* it was rounded in *C. aliai*. The elytra were parallel-sided and flattened in *C. aliai*, ovoid and convex in *C. stenocephalus ifniensis*. On average, the ratio between length of elytra and maximum width of elytra (EL/EW) was 17.5/11.5 mm = 1.52 in *C. aliai* and 19.5/13.0 mm = 1.50 in *C. stenocephalus ifniensis*. In females of both taxa, the apex of the elytra was more acuminate than the one of males. The elytral sculpture was composed by deep striae with coarse punctuation in *C. aliai* and by slightly marked striae with fine punctuation in *C. stenocephalus ifniensis*. Legs were proportionally longer in *C. aliai* than in *C. stenocephalus ifniensis*.

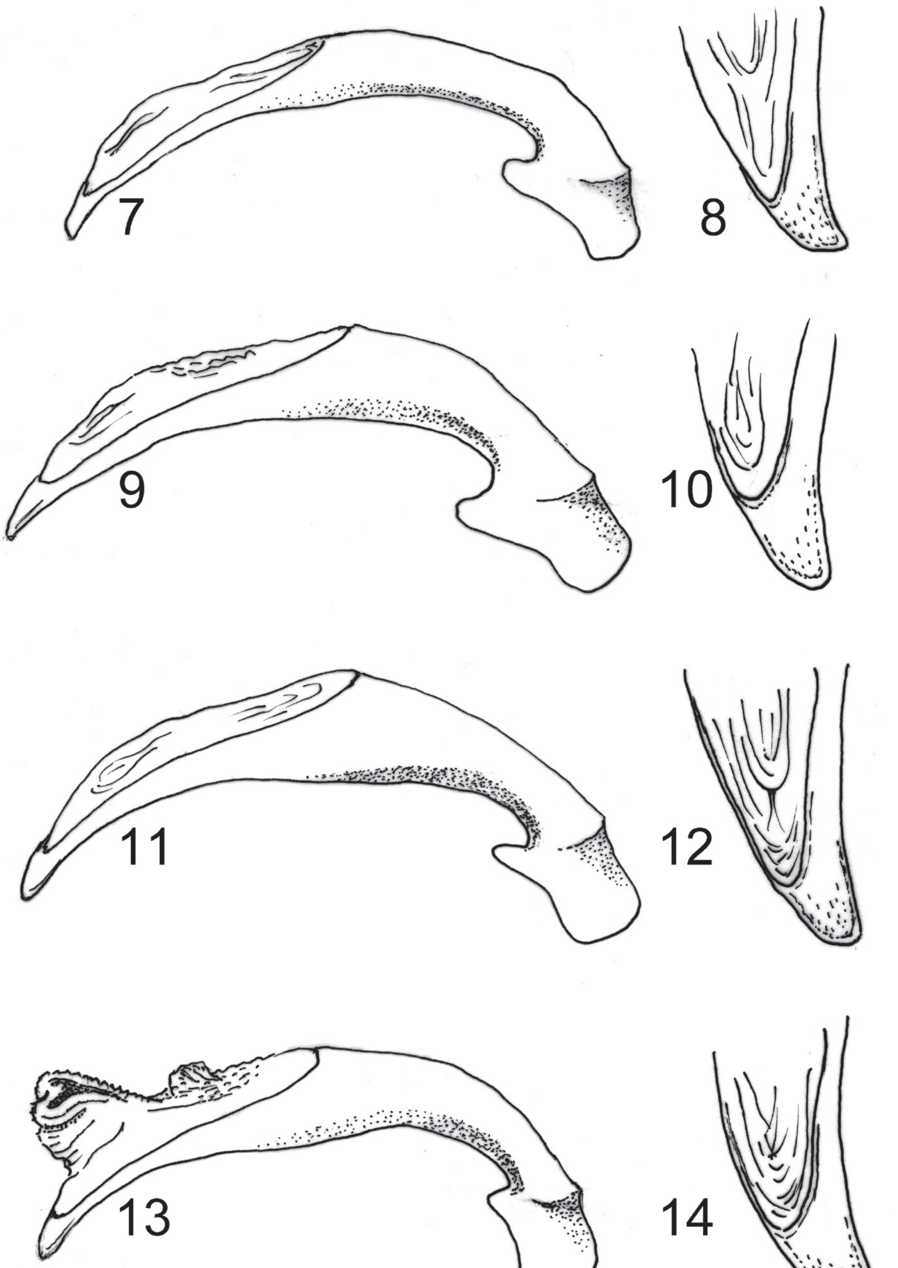
Both male and female genitalia were found to be very homogeneous within representatives of subgenus *Cathoplius*. The median lobe of the aedeagus in all taxa was similar either in size or in shape (Figures 7–14). In *Carabus asperatus* (Dejean, 1826) the median lobe was slightly smaller, more elongate and its apex was more acutely shortened, while in *C. aliai* Escalera, 1944 the apex was wider and a little more curved than in *C. stenocephalus susicus* Antoine, 1941 and *C. stenocephalus ifniensis* Zarco, 1941. Parameres were similar.

The endophallus in *Cathoplius* species showed a peculiar morphological feature (Figures 15–17). In the distal aggonoporial area, two small sclerites, slightly different in *C. aliai* and in *C. stenocephalus* from those of *C. asperatus*, were present. In the saccellar area, a tumid, v-shaped body covered with dense, prominent scales and spines, was evident. The ostial lobe was well developed. The female inner reproductive tract of the various *Cathoplius* taxa was found to be identical, being fully membranous with no sclerified structures. External genitalia, in the specific gonocoxite 2 of the ovipositor, were also very similar in all species (Figures 18–19).

C. aliai × *C. stenocephalus ifniensis* hybrids showed intermediate features when compared to parental species. Hybrids were morphologically more similar to the parent female than to the male: hybrid AIF1 (♂ *C. aliai* × ♀ *C. stenocephalus ifniensis*) (L 28.0–36.0 mm) was a little more similar to *C. stenocephalus ifniensis* than hybrid AIF2 (♂ *C. stenocephalus ifniensis* × ♀ *C. aliai*) (L 27.0–34.0 mm) which in turn was more similar to *C. aliai* (Figures 20–23).

Morphology of pre-imaginal stages. Morphological differences observed among the pre–imaginal stages of the various *Cathoplius* taxa were corresponding to those reported by Busato et al. (2014). *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 larvae were found to be very small, being much shorter and more slender than the ones of *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941. Their total length varied according to the development period and to the quantity of food ingested. Minimum and maximum length and width of the three larval instars of *C. aliai* and *C. stenocephalus ifniensis* are reported in Table 1.

The *C. aliai* newborn larvae (Figure 24) measured 9.5 × 2.0 mm (length × width), versus 11.0 × 2.5 mm in *C. stenocephalus ifniensis*. Before burying, the *C. aliai* third instar larvae measured 26.0–27.0 × 6.5 mm (males) or 29.0–30.0 × 6.5 mm (females),



Figures 7–14. *Cathoplius* species aedeagi. Right lateral aspect of the median lobe (left) and details of its apex (right) in *Carabus asperatus* from Oualidia (**7–8**) *C. stenocephalus susicus* from Aglou (**9–10**) *C. stenocephalus ifniensis* from Sidi Ifni (**11–12**) and *C. aliai* from Tan-Tan (**13–14**). Scale bars: 1 mm.

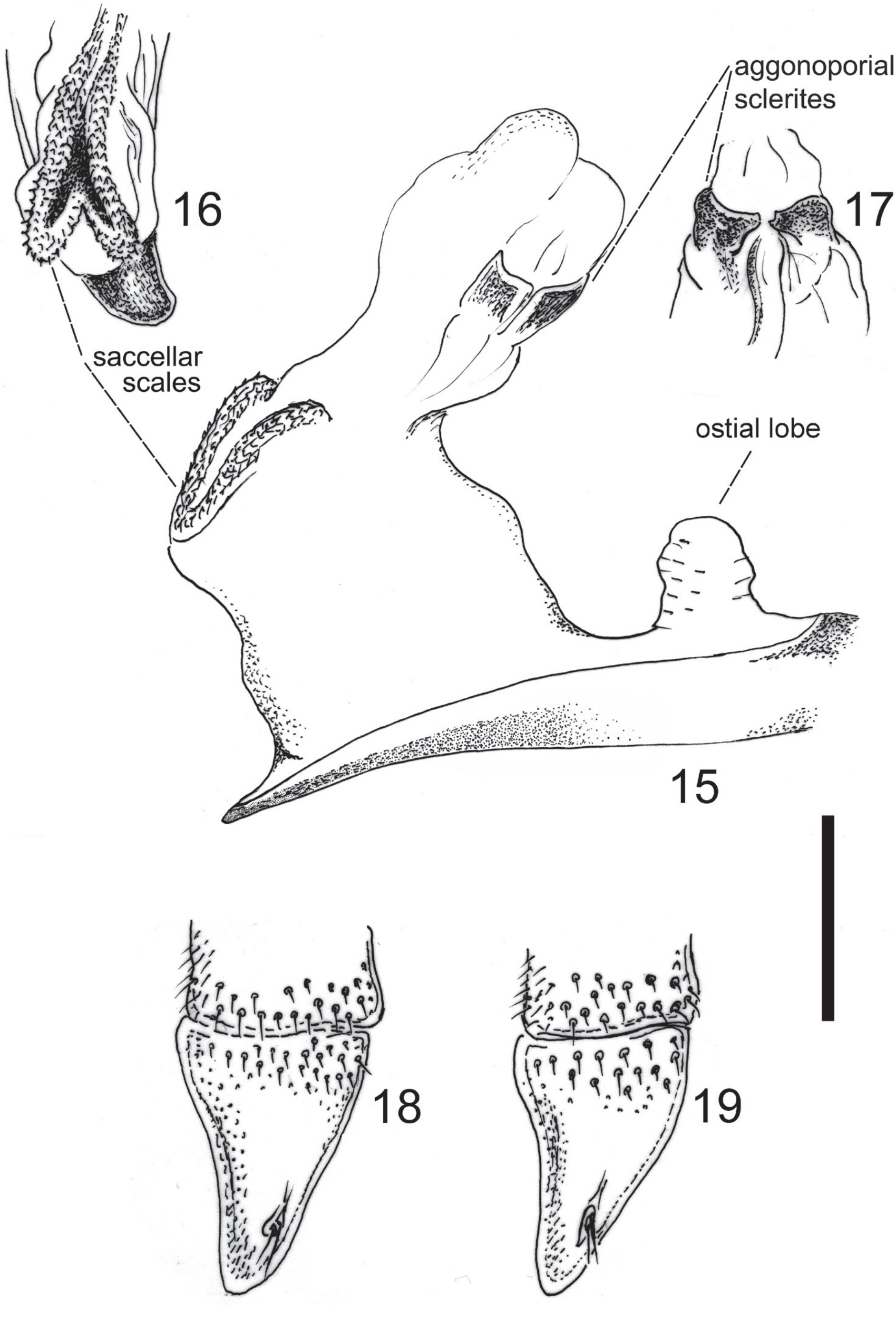
Table 1. Size of *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 and *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941 pre–imaginal stages.

Pre–imaginal stages	*C. aliai* (length × width, mm)	*C. s. ifniensis* (length × width, mm)
1st instar larva:		
- newborn	9.5–10.0 × 2.0	11.0–11.5 × 2.5
- before ecdysis	15.0–16.5 × 2.5	16.5–18.0 × 3.0
2nd instar larva:		
- after ecdysis	16.0–16.5 × 3.5	17.5–18.0 × 4.0
- before ecdysis	20.0–21.5 × 4.5	23.0–24.5 × 5.0
3rd instar larva:		
- after ecdysis	21.0–23.0 × 6.0	24.0–26.0 × 6.0
- before burying	26.0–30.0 × 6.5	28.0–34.0 × 7.0
- pre–pupa	23.0–26.0 × 6.5	25.0–29.0 × 7.0
Pupa	20.0–22.0 × 9.0	22.0–25.0 × 9.5

versus 27.0–29.0 × 7.0 mm (males) or 32.0–34.0 × 7.0 mm (females) in *C. stenocephalus ifniensis*. In both taxa, the length/width ratios of the *frontoclypeolabrum* of the three larval instars were similar. For the size difference, appendages (antennae, mouthparts and legs) looked shorter in *C. aliai* than in *C. stenocephalus ifniensis* but length ratios between segments were similar.

The aspect of the pupa reflected that of the adult. The *C. aliai* pupae (Figures 25–27) were smaller (20.0–22.0 × 9.0 mm, length × width) and with a narrower and more elongated thoracic area compared to those of *C. stenocephalus ifniensis*, that were larger (22.0–25.0 × 9.5 mm) and with a wider thoracic area. Pupal chaetotaxy was found to be an important character capable of discriminating the two taxa: the *C. aliai* pupa showed tufts of scarce and thin setae on the metanotum, tufts of thick, strong and long setae from urotergite I to V, isolated setae on urotergite VI and VII, and two small groups of setae on urotergite VIII. Contrary to this, in the *C. stenocephalus ifniensis* pupa, setae on metanotum were extremely scarce, thin and barely visible, while thicker, stronger and longer setae were present from urotergite I to VI and on urotergite VIII but not on urotergite VII (see Busato et al. 2014).

Field observations. The biotope where *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 was found is a sandy and rocky desert area located 15–20 km far from the Atlantic coast, at an elevation comprised between 50 and 200 m a.s.l. (Figure 28). The Tan-Tan northern plateau, in Tan-Tan province, southern Morocco, is a north–south oriented 60 km long and 6–10 km wide mountain (from the Oued Drâa outlet to the Oued Boukhchibia, a tributary of the Oued Chbika). Its xerophilous vegetation is composed mainly by shrubs of *Launaea arborescens* (Battandier, 1888), *Lycium intricatum* Boissier, 1838 and *Tetraena gaetula* (Emberger & Maire, 1928), as well as by the arboreal spurge *Euphorbia regis-jubae* J. Gay, 1847 and the cactus–like spurge *Euphorbia officinarum echinus* (J.D. Hooker & Cosson, 1874). Snails of the family Helicidae inhabiting this vegetation are represented by three species of *Theba* [*T. subdentata me-*



Figures 15–19. *Cathoplius* species male and female genitalia. Endophallus of *Carabus asperatus* from Oualidia, with emphasis on the aggonoporius, saccellus and ostial lobe (**15**). Details of the saccellar (**16**) and aggonoporial (**17**) areas in *C. aliai* from Tan-Tan. Gonocoxite 2 of the ovipositor in *C. asperatus* from Oualidia (**18**) and in *C. aliai* from Tan-Tan (**19**). Scale bar: 1 mm.

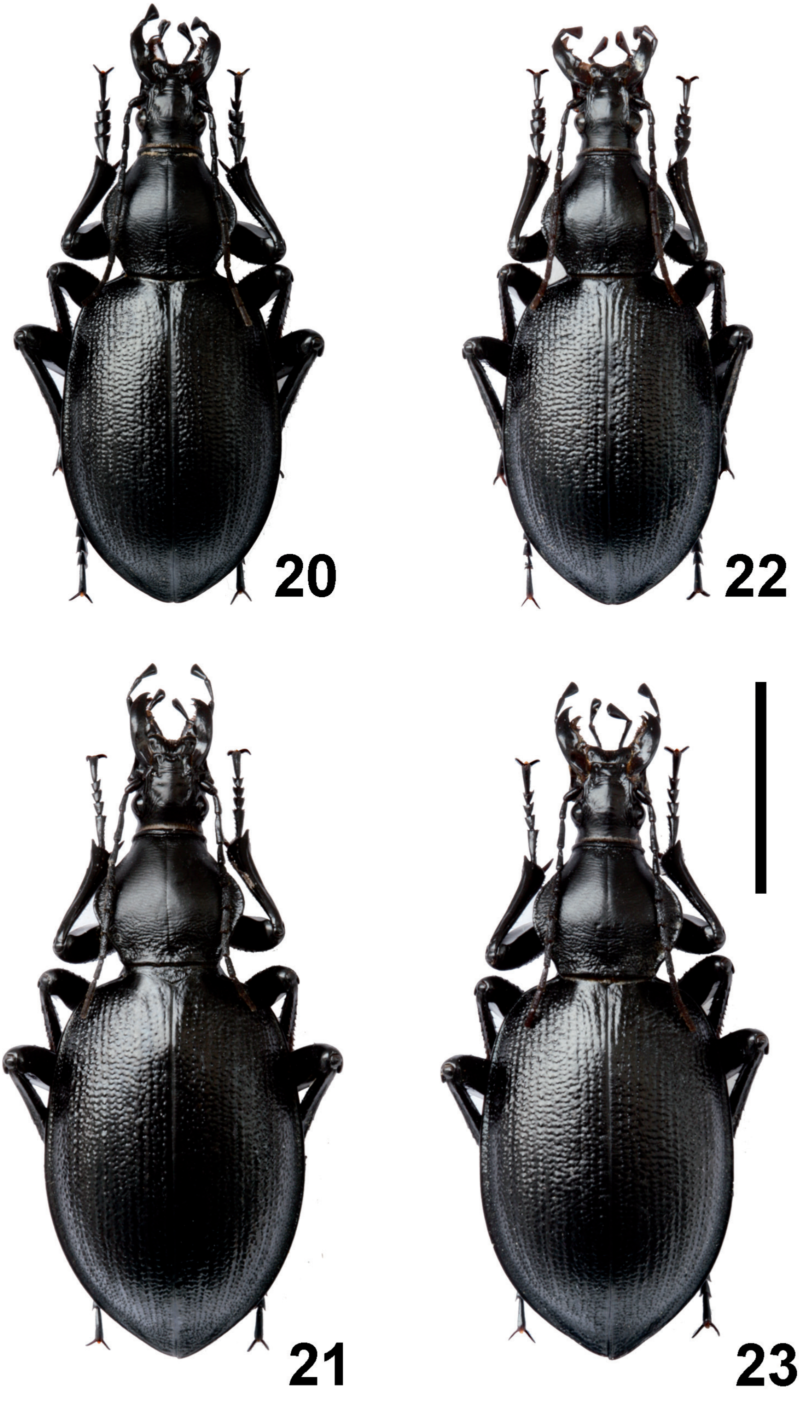
ridionalis (Sacchi, 1955), *T. chudeaui* (Germain, 1908) and *T. sacchii* Gittenberger & Ripken, 1987] and by *Eremina dillwyniana* (Pfeiffer, 1851). *T. subdentata meridionalis* is the smallest and most common snail, particularly abundant on *L. arborescens* and *E. regis-jubae* shrubs, while *Eremina dillwyniana* is the largest and generally related to *E. echinus*. Major carabids cohabiting with *C. aliai* are *Calosoma* (*Campalita*) *maderae* (Fabricius, 1775), *Calosoma* (*Caminara*) *olivieri* Dejean, 1831, *Scarites* (*Scallophorites*) *buparius* (Forster, 1771), *Sphodrus leucophthalmus* (Linnaeus, 1758) and *Anthia* (*Termophilum*) *sexmaculata* (Fabricius, 1787), the last three species being more common. The climate in the Tan-Tan area is typical of a steppe–desert, with low rainfall (102 mm/year) exclusively distributed in late autumn and winter (Meregalli 2005). Despite these extreme environmental conditions, the area hosts highly interesting endemic taxa such as *Eurycleonus talamellii* Meregalli, 2005, the largest cleonine weevil of the western Palaearctic region.

At the time of the visit to the Tan-Tan northern plateau in 2010, temperatures ranged from 15–17 °C (night) to 19–25 °C (day). *Carabus aliai* adults and larvae were found from December 17–19 in coincidence with light rains. Adults were seen especially in the late afternoon, coming out from their burrows from underneath the shrubs. Females were preying upon *Theba* snails while staying hidden into the shrubs. Males were seen running quickly from one shrub to another in search of both snails to eat and females to mate. Burrows consisted in oblique holes about 20 cm deep. *Carabus aliai* adults were observed while eating mainly *T. chudeaui* and secondarily *T. subdentata meridionalis*, but not *T. sacchii* and *E. dillwyniana*. Larvae were only seen inside *T. subdentata meridionalis* shells adhering to *L. arborescens* branches, at a height from the ground up to 1.5 m. Ecdysis was taking place inside snail shells, where exuviae were normally found. One second instar larva was observed while running on *L. arborescens* branches, searching for a new snail to prey upon. Predation did not result in snail falling to the ground, instead the shell remained adherent to the shrub branch during the larval meal.

Laboratory observations. After an adaptation period of one–two hours, during which *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 adults were running frenetically in the terrariums, both males and females started digging their own burrows underneath the moss. Burrows were represented by oblique holes with a diameter of 1.5–2 cm reaching the bottom of the terrarium. Adults were hiding inside the burrows during the day and normally came out for feeding and mating in the late afternoon to early morning. Only when barometric pressure was decreasing, did we detect a prolonged running activity throughout the day, especially in males.

Adults of *C. aliai*, fed with *Theba pisana pisana* (O.F. Müller, 1774) snails, were very voracious. Due to their elongate head and narrow pronotum they were able to penetrate deep inside the snail's shell. Each meal lasted for approximately an hour with males and 2–3 hours with females. Males were killing and partially eating an average of 5–6 snails per night. Female meal totals were 1–3 snails per night. Meals were preferably consumed close to the moss side.

Mating occurred during the night and throughout the whole oviposition period. Males were frequently observed on top of females who were feeding. Each mating



Figures 20–23. *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 × *C. stenocephalus ifniensis* Zarco, 1941 hybrids. Habitus of hybrid AIF1 (**20** male **21** female) and hybrid AIF2 (**22** male **23** female) from our laboratories. Scale bar: 1 cm.

lasted for approximately an hour. One day after mating, oviposition took place. Females, after digging an oblique gallery in the soil, started laying eggs by inserting their abdomen at a depth and distance between eggs of 1–1.5 cm. Females layed from 1–6 but up to 12–20 eggs per night. When laying was complete, females sealed the gallery. When laid, eggs were diaphanous white in color and measured 4.5 × 1.5 mm. With time, they gradually became light yellow in color. Their size increased during embryonic development, reaching 5.0 × 2.0 mm. The embryonic development was completed in 15 days. Under laboratory conditions, the duration of the oviposition period in *C. aliai* was 100–110 days, from December to March. During this time, the four females laid 277, 282, 298 and 307 eggs, respectively. Two oviposition periods, separated by a short pause (10 days) in January, were noticed. On average, the total productivity of *C. aliai* in 95 laying days was 291 eggs/female, with a laying frequency of 3 eggs/night. Following reproduction, all *C. aliai* breeders died without undergoing summer diapause.

In contrast to *C. aliai*, the reproduction cycle of *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941 consisted in a first winter and a second spring oviposition cycle, separated from each other by a short burial period (30 days). When the spring cycle ended, breeders buried again and fell into a long summer diapause that lasted until the following winter time, when a scarce number of eggs and viable larvae was produced before breeders died. The average total productivity of *C. stenocephalus ifniensis* in 120 laying days was 338 eggs/female (laying frequency of 2.8 eggs/night), subdivided in 242 eggs laid in 75 days during the first oviposition cycle (3.2 eggs/night) and 96 eggs laid in 45 days during the second cycle (2.1 eggs/night).

After hatching, *C. aliai* larvae remained in the egg cell for a day to complete sclerification. Once on the surface, newborn larvae begun running around the terrarium searching for snails to feed on. When snails were found, larvae penetrated the shell by keeping their ventral side of the body adhering to the shell walls. Despite their small size [10.0 × 2.0 mm (length × width)], *C. aliai* first instar larvae were found to be very aggressive, being able to kill also large *T. pisana pisana* snails. For the completion of the first instar period, two meals (each lasting 1.5 days) with snails of small size (shell width 10 mm) were necessary. The pre-ecdysis period and ecdysis lasted 2 days. Ecdysis always took place inside snail shells. Exuviae were then found inside the shells.

The *C. aliai* second instar larvae (16.0 × 3.5 mm in size) began feeding after integument sclerification, which stood one day. For the completion of the second instar period, three meals (each lasting 1.5 days) with snails of intermediate size (shell width 15 mm) were necessary. The pre-ecdysis period and ecdysis lasted 2.5 days. Ecdysis generally took place inside snail shells or in a pit dug under the shelter or into the soil.

After integument sclerification of one day, the third instar larvae (22.0 × 6.0 mm in size) began feeding again. For the completion of the third instar period, four meals (each lasting 2 days) with snails of large size (shell width 20 mm) were necessary. Then, the mature larvae (28.0 × 6.5 mm in size) stopped feeding and started digging a large cell in the soil for pupating. Inside the pupal cell, pre-pupas (24.0 × 6.5 mm in size) and pupas (21.0 × 9.0 mm in size) occupied 2/3 of the cell length. After emergence

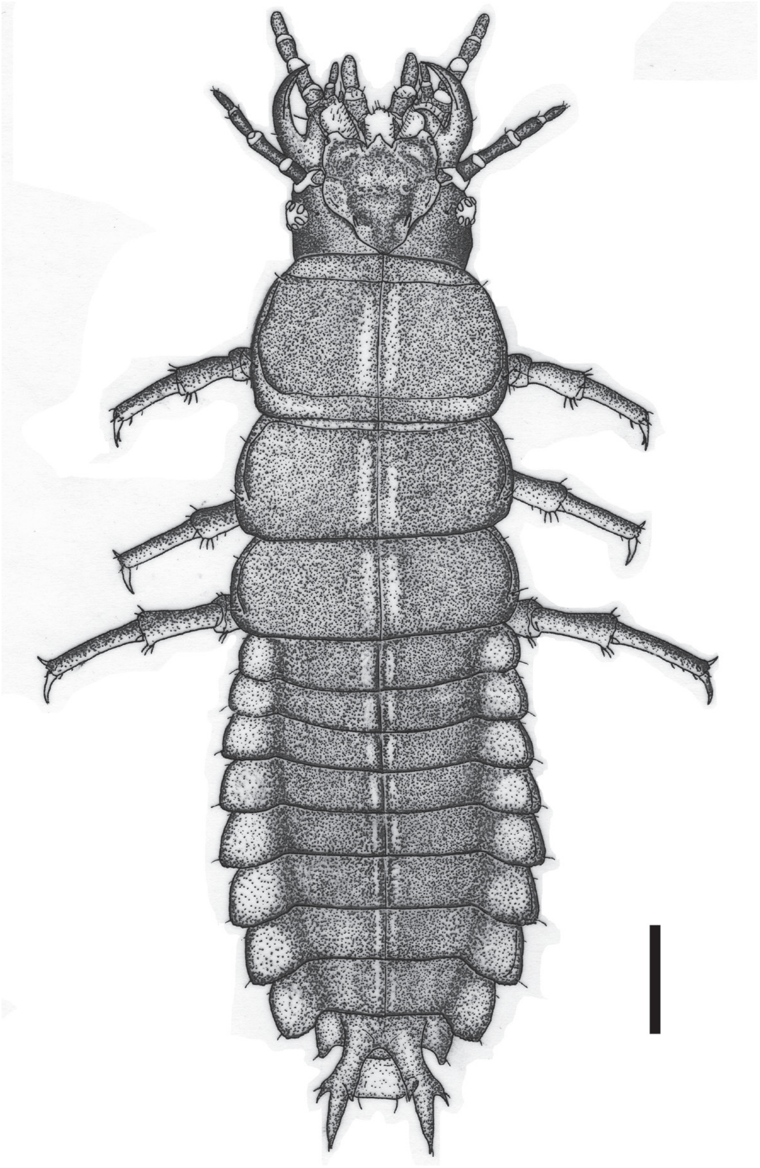


Figure 24. *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 first instar larva: dorsal aspect. Scale bar: 1 mm.

from the pupa, *C. aliai* neo-adults remained in the pupal cell for about 3 days before rising to the surface.

Under laboratory conditions, the duration of the development cycle from egg fertilization to the rising of neo-adult averaged 2 days shorter in *C. aliai* compared to *C. stenocephalus ifniensis* (70 vs. 72 days). While embryonic development lasted 15 days in both species, larval development on the surface (from egg hatching to digging of the

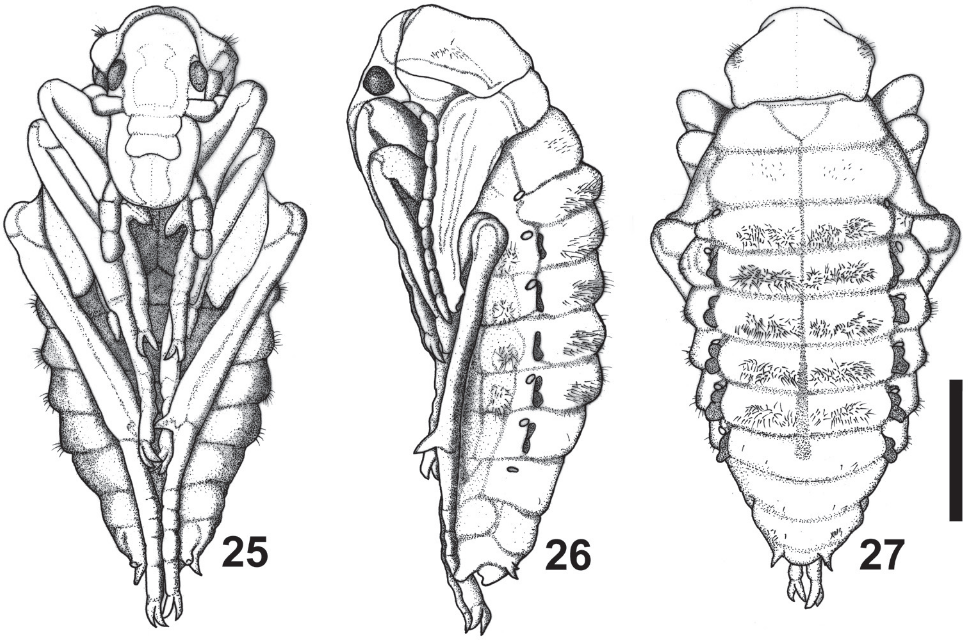
Table 2. Differences between *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 and *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941 life cycles at the same laboratory conditions [temperature 15/19–17/21 °C (night/day); photoperiod 9–13 hours of daylight].

Life cycle phases	***C. aliai* (mean days)**	***C. s. ifniensis* (mean days)**
Embryonic development:	15	15
- mating – oviposition	1	1
- oviposition – hatching of larvae	14	14
Larval development on the surface:	23	24
- 1st larval instar	6	7
- 2nd larval instar	8	8
- 3rd larval instar	9	9
Burial phase:	32	33
- pre–pupa	8	9
- pupa	21	19
- adult emergence – rising to the surface	3	5
Duration of development cycle	**70**	**72**
Completion of adult sclerification	21	21
Time from hardening to burying	2	15
Burial phase (gonad maturation)	–	30
Spring oviposition cycle	–	50 or 0 *
Burial phase (summer diapause)	250	180 or 220 *
Winter oviposition cycle	95	75
End of oviposition – death	15	15
Life span (larva + pupa + adult)	**453**	**458**

* Different duration depending on the number of oviposition cycles

pupal cell), as well as the burial phase, were one day shorter in *C. aliai* than in *C. stenocephalus ifniensis* (23 vs. 24 and 32 vs. 33 days, respectively). More specifically, the duration of the first instar period (6 vs. 7 days), the duration of the prepupal period (8 vs. 9 days) and the time between emergence from the pupa and the rising to the surface of neo-adults (3 vs. 5 days) were a little shorter in *C. aliai* than in *C. stenocephalus ifniensis*, while the duration of the pupal instar was a little longer (21 vs. 19 days) (Table 2).

Under laboratory conditions, the rising to the surface of *C. aliai* neo-adults took place from the beginning of February through mid-April. The mean survival rate from egg to adult was 53% and the final ratio between males and females was of 1:1. Males rose to the surface a little earlier than females. Once on the surface, neo-adults fed on *T. pisana pisana* snails until their total hardening, which was completed in approximately 3 weeks. During this period, neo-adults were very aggressive, killing a large number of snails throughout the day. Both males and females were partially eating an average of 9–10 snails per day. Even with optimal temperature and humidity conditions, a few days after completing sclerification neo-adults buried themselves and fell into a long summer diapause (8–9 months, starting from April–May) without reproducing. Adults rose to the surface again at the end of November, commencing their oviposition cycle that lasted for 3.5 months until mid-March. Egg production averaged 250 eggs/female.



Figures 25–27. *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 pupa: ventral, lateral and dorsal aspect. Scale bar: 5 mm.

Following reproduction, adults died. Females died a few days after discontinuing oviposition. Males survived approximately 2 weeks longer than females. No feeding attempts were observed during this period. The average life span of *C. aliai* under laboratory conditions was 15 months (including 2 months as pre-imaginal stages) (Table 2).

In *C. stenocephalus ifniensis* the pattern was found to be similar (the mean survival rate from egg to adult was 52% and the final ratio between males and females was of 1:1), but the reproduction cycle was different. Neo-adults of this taxon, after rising to the surface and completing sclerification in approximately 3 weeks, remained on the surface for about two weeks and then underwent a burial phase that lasted for a month. During this phase, gonad maturation was taking place. By mid-March, precocious neo-adults rose to the surface and started reproducing, but their cycle was short (1.5 months) and egg production was scarce (average of 100 eggs/female). Only one third of neo-adults took part in the spring oviposition cycle, while the majority of individuals remained buried without reproducing. At the end of April–beginning of May, adults that reproduced buried themselves and fell into diapause for 6 months. At mid-November, all individuals rose to the surface and started reproducing. The winter oviposition cycle lasted for 2.5 months and egg production was high (average of 250 eggs/female). Following reproduction, adults died. When considering the two oviposition cycles, the average egg production in *C. stenocephalus ifniensis* was 350 eggs/female (100 eggs/female more than in *C. aliai*). The average life span of *C. stenocephalus ifniensis* under laboratory conditions was 15 months (Table 2).

Figure 28. The *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 biotope in Tan-Tan, southern Morocco. Photo by Dr M. Meregalli.

Hybrids behavior. Hybridization between *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 and *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941 led to the following results:

1) *C. aliai* ♂ × *C. stenocephalus ifniensis* ♀ *crossbreeding* (AIF1). Mating occurred normally, but in a less intensive manner than in pure *C. aliai* and *C. stenocephalus ifniensis* breeding. Oviposition took place a day after copulation. Females layed from 1 to 4 eggs per night. The average total productivity was 87 eggs/female over a period of 65 days. Following reproduction, adults buried themselves and fell into diapause. Besides a reduced oviposition rate compared to the normal one of the parental taxa, hybridization resulted in a reduced hatching rate and an increased pre-imaginal mortality rate that led to a F1 mean survival rate from egg to adult of 33%, versus 53% in *C. aliai* and 52% in *C. stenocephalus ifniensis*. The duration of the development cycle in hybrid AIF1 was 74 days, subdivided into 15 days for embryonic development, 25 days for larval development on the surface and 34 days for the burial phase. Sclerification of neo-adults was completed in about 3 weeks. Afterwards, neo-adults remained on the surface for 3–4 weeks before falling into diapause. Hybrids had a sturdiness similar to that of pure *C. aliai* or *C. stenocephalus ifniensis* specimens, with a life span of 12–15 months.

The F1 crossbreeding (AIF1 × AIF1) led to a reduced fecundity rate (41 eggs/female over a period of 45 days), to neonatal mortality with scarcely viable F2 pre-imaginal stages (70% mortality rate for first instar larvae, 48% for second instar larvae, 32% for third instar larvae, 80% for pre-pupas and pupas) and to stillbirth with non-viable F2 imagoes (100% mortality rate inside the pupal cell, without any rising to the surface of neo-adults).

2) *C. stenocephalus ifniensis* ♂ × *C. aliai* ♀ *crossbreeding* (AIF2). The behavior was found to be similar to that seen in the previous combination. The average total productivity was 69 eggs/female over a period of 75 days. Two oviposition periods, separated by a short pause (15 days), were noticed. Also in this case, hybridization was responsible for a reduced hatching rate and an increased pre-imaginal mortality rate that led to a F1 mean survival rate from egg to adult of 31%. The duration of the development cycle in hybrid AIF2 was 72 days, subdivided into 15 days for embryonic development, 24 days for larval development on the surface and 33 days for the burial phase. After sclerification, neo-adults remained on the surface for about 2 weeks before falling into diapause.

The F1 crossbreeding (AIF2 × AIF2) led to a reduced fecundity rate (33 eggs/female over a period of 53 days), to neonatal mortality with scarcely viable F2 pre-imaginal stages (82% mortality rate for first instar larvae, 50% for second instar larvae, 34% for third instar larvae, 84% for pre-pupas and pupas) and to stillbirth with non-viable F2 imagoes (100% mortality rate inside the pupal cell).

Discussion

Morphology. The subgenus *Cathoplius* within the Genus *Carabus* includes ground beetles characterized by strongly sclerified integument, black in color, brachypterous, with elytra that are joined along the suture, an achetous, ellipsoidal, silphoid or cychrized pronotum, and a narrow and very elongate head (Busato et al. 2014). According to their close geographic vicariance and their clinal morphological variation along the Atlantic Moroccan coast (from stocky, tenebrionid-like northern forms to more elongate, cychrized southern forms), *Cathoplius* are considered as belonging to a single species [*Carabus asperatus* (Dejean, 1826)] in some catalogues (Březina 1994; 1999; Bousquet et al. 2003; Lorenz 2005). However, in most taxonomic treatments, *Cathoplius* taxa are ascribed to two species: *C. asperatus*, monotypic, with a northern distribution, and *C. stenocephalus* Lucas, 1866, polytypic, with a southern distribution, *C. aliai* Escalera, 1944 being considered as the southernmost subspecies of the latter (Antoine 1955; Kocher 1963; Deuve 1994; 2004; 2012; 2014). The *C. stenocephalus* taxonomic subdivision into subspecies was adopted after examining a scarce number of individuals coming from the southern parts of the distributional area (Zarco 1941; Escalera 1944; Español 1944; Mateu 1947). This fact has led to the statement of considering *C. aliai* as a simple subspecies of *C. stenocephalus* and not as a separate species.

The morphological differences between *C. aliai* and *C. stenocephalus* (*sensu lato*) are remarkable. The pronotum profile and the elytral sculpture are very peculiar. Despite their close range of distribution (only 50 km are dividing Sidi Ifni Beach from Plage Blanche), differences are striking when *C. aliai* adults are compared to those of *C. steno-*

cephalus ifniensis Zarco, 1941. These differences include a smaller size, a much slender silhouette, a more brilliant aspect, longer legs, and a wider and a little more curved apex of the median lobe of the aedeagus. Differences are also present referring to pre-imaginal stages: *C. aliai* larvae are much smaller than those of *C. stenocephalus ifniensis* (9.5 × 2.0 mm vs. 11.0 × 2.5 mm in newborn larvae) and the *C. aliai* pupa is also smaller, with a narrower thoracic area and a different chaetotaxy. Both imaginal and pre-imaginal features are an index of the specific differentiation between the two taxa.

Life cycle. The 2010 raining season in the Tan-Tan area began on November 30, with a storm (17 mm of rain at a temperature of 16–23 °C) that induced the rising of *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 adults from their summer diapause. Oviposition occurred some days later, with eggs hatching on December 13–14 and first ecdysis on December 18–19, when second instar larvae were found. The second ecdysis occurred at the laboratory on December 28–31 and the burying of third instar larvae on January 7–9, with rising to the surface of neo-adults on February 8–15. Out of the ten wild larvae found, two died as second instar and two as third instar, and out of the six surviving adults, four were males and two were females. The 60% survival rate noticed for wild larvae matches the mean survival rate of 53% obtained at the laboratory from egg to adult, but the ratio between males and females does not (2:1 in nature vs. 1:1 at the laboratory). This is probably due to the fact that, after emergence, precocious males are rising to the surface earlier than first females.

In mid-March *C. aliai* neo-adults buried themselves and fell into a summer diapause that lasted until late autumn, when terrariums were abundantly watered. When watering was carried out during late spring or in summer, adults were rising to the surface, feeding for some days and burying again without mating. The beginning of reproduction in representatives of subgenus *Cathoplius* depends exclusively on the start of the natural raining season (Busato et al. 2014). In *C. stenocephalus ifniensis* Zarco, 1941 the raining season normally starts at the beginning of November, while in *C. aliai* it starts at the beginning of December. The reason for this difference is that on the western slopes of the Anti-Atlas chain, where Sidi Ifni is located and where *C. stenocephalus ifniensis* dwells, the first rain usually falls a month earlier than in the desert area around Tan-Tan.

While under laboratory conditions the oviposition period in *C. aliai* lasted for over three months, in nature this is not the normal case. Aridity and cold weather at night are responsible for a lack of active *Theba* snails, and correspondingly no activity of *C. aliai* adults and larvae is present in January in the Tan-Tan area. In nature, the oviposition period probably lasts for a month, from the beginning to the end of December, and after reproduction all adults die. The *C. aliai* life cycle can therefore be placed into the winter breeders with short larval development type *sensu* Paarmann (1979) and into the annual univoltine winter semelparous type *sensu* Matalin (2007). The last breeding type is well adapted to desert conditions with short winter rains. Contrary to this, the *C. stenocephalus ifniensis* life cycle belongs to the late autumn breeders type and is iteroparous, where adults who bred in late autumn become active again and breed a second time in early spring before summer diapause.

One of the most striking features in representatives of the subgenus *Cathoplius* is their high fecundity rate. Species belonging to this subgenus are one of the most prolific among the known ground beetles (Busato et al. 2014). In our experiments, *C. aliai* females laid 250–300 eggs in 100 days that resulted in 125–150 neo-adults. In nature, the number of laid eggs and the consequent number of neo–adults is certainly lower, due to the shortened oviposition period and to predation. Ordinarily, each female should lay an average of 80 eggs in a month resulting in 40 neo-adults.

The mean duration of the development cycle, from egg fertilization to the rising of neo-adults, was found to be shorter in *C. aliai* than in *C. stenocephalus ifniensis*. Results of the present study are in agreement with those obtained by Busato et al. (2014) where, at a constant temperature of 21±1 °C, the development cycle was a day shorter in *C. aliai* than in *C. stenocephalus ifniensis* (69 and 70 days, respectively). At temperatures varying from 15/19 to 17/21 °C (night/day) the development cycle was a little longer in both taxa, but indeed shorter in *C. aliai* than in *C. stenocephalus ifniensis* (70 vs. 72 days). In nature, this fact is likely due to the desert habitat of the Tan-Tan area, where snails activity is reduced to shorter periods compared to that on the Sidi Ifni beach.

From the behavioral point of view, *C. aliai* is a typical sabulicolous steppe wandering species (Mateu 1947). A remarkable and intensive running activity, more evident in males, was noticed both in the field and in terrariums. Moreover, adults dig burrows underneath shrubs where they hide during the day for protection from dehydration and predation. This way of life is common to other cohabiting steppe wandering species, such as *Calosoma* (*Campalita*) *maderae* (Fabricius, 1775) and *Calosoma* (*Caminara*) *olivieri* Dejean, 1831 (Ghittino, personal observations). Contrary to *C. aliai*, running activity in *C. stenocephalus ifniensis* was found infrequently. *Carabus stenocephalus ifniensis* is a more sedentary taxon, carrying out the major part of its life inside the *Lycium intricatum* and *Tetraena gaetula* shrubs that are present on the Sidi Ifni coastal area, where snails belonging to the species *Theba subdentata meridionalis* (Sacchi, 1955) and *T. solimae* (Sacchi, 1955) are dwelling. This sedentary behavior was confirmed at a laboratory level, where running activity in terrariums was rarely observed even when barometric pressure was decreasing.

In the field, *C. aliai* adults were observed mainly feeding upon *T. chudeaui* (Germain, 1908) and secondarily upon *T. subdentata meridionalis* snails. This is likely due to the major shell width, the larger aperture and the lack of a parietal denticle in *T. chudeaui*. The fact that *Eremina dillwyniana* (Pfeiffer, 1851) snails, which are abundant in the Tan-Tan area, were not eaten by *C. aliai* is an additional proof that representatives of subgenus *Cathoplius* feed exclusively on *Theba* snails. First and second instar *C. aliai* larvae were only found inside *T. subdentata meridionalis* shells adhering to *Launaea arborescens* branches. The completion of the first pre-imaginal stage on vegetation is probably essential for *C. aliai* larval survival, in order to avoid predation from associated carabids. At the Tan-Tan biotope several very aggressive species are present, such as *Anthia* (*Termophilum*) *sexmaculata* (Fabricius, 1787), *Scarites* (*Scallophorites*) *buparius* (Forster, 1771) and *Sphodrus leucophthalmus* (Linnaeus, 1758) (Ghittino, personal observations). *T. subdentata meridionalis* snails are probably the prey of choice for *C. aliai* larvae for their minor shell width, the smaller aperture and the presence of a well developed parietal denticle that better protects larvae from aggressions.

As predators of live *Theba* snails, *Cathoplius* should play an important role in the ecosystem by reducing *Theba* proliferation which can be detrimental to both conservation of the scarce vegetation in arid areas and to animal husbandry, by preventing important livestock parasitic diseases among grazing land animals (Busato et al. 2014). *Theba* snails are involved in the life cycle of sheep and goat lungworms by harboring the infective stage of some protostrongylid nematodes (Cabaret 1987; Grewal et al. 2003). Sheep and goat breeding is a common activity in the Moroccan Saharan provinces. Parasitic diseases affecting sheep and goat lungs are frequent in this area and can be diagnosed both clinically and at slaughtering (Ghittino, personal observations).

We could not find any reports of *Theba subdentata meridionalis* or *T. chudeaui* acting as intermediate hosts for sheep and goat lungworms but the presence of the pathology in animals and the observation of worm stages in snails are evidence of their involvement in the disease life cycle. Differently from other local snails, *T. subdentata meridionalis* and *T. chudeaui* live on shrubs that are particularly appreciated by sheep and goats (e.g. *Launaea arborescens* and *Lycium intricatum*) which then can be easily infected through accidental ingestion of parasitized snails.

Under laboratory conditions, *C. aliai* adults and larvae were found to be very aggressive against *Theba* snails. Adults killed up to 5–6 snails per night over a period of three months. For completing the development cycle, each larva fed on an average of nine snails and to reach complete sclerification neo-adults killed up to 9–10 snails per day over a period of three weeks. These data indicate that *C. aliai* is an efficacious natural enemy of *Theba* snails that can also reduce the impact of some economically important ruminant diseases. Conservation of *C. aliai* in southern Morocco is therefore recommended.

Hybridization. The results of our hybridization experiments between *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 and *C.* (*Cathoplius*) *stenocephalus ifniensis* Zarco, 1941 are in agreement with those obtained by Busato et al. (2014). The few discrepancies in the current study are likely due to the different breeding temperatures applied. Despite a normal mating attitude of parents, a shorter oviposition period and a reduced number of laid eggs was noticed in both crossbreeding combinations compared to what observed in pure *C. aliai* and *C. stenocephalus ifniensis* breeding. This fact was more evident for the AIF2 (*C. stenocephalus ifniensis* ♂ × *C. aliai* ♀) than for the AIF1 (*C. aliai* ♂ × *C. stenocephalus ifniensis* ♀) combination. Besides this, F1 hybrids underwent a high mortality rate during embryonic, larval and pupal development that led to a 40% reduction of the final survival rate compared to that of *C. aliai*. F1 hybrids showed also a longer duration of their development cycle (72 days in AIF2 hybrid and 74 days in AIF1 hybrid vs. 70 days in *C. aliai*).

The F1 hybrids crossbreeding (AIF1♂ × AIF1♀ and AIF2♂ × AIF2♀) led to a further reduction of laid eggs and a very high mortality rate during embryonic, larval and pupal development. This was also more evident in the AIF2 than in the AIF1 combination. Only a few F2 immature imagoes were obtained (3 out of 120 larvae for AIF1 and 1 out of 100 larvae for AIF2), but these specimens died in the pupal cell within 10 days from emergence. Since no vital neo-adults were available, it was impossible to carry out subsequent F2 hybrids crossbreeding.

Through our crossbreeding experiments, that employed a high number of F1 hybrids (4 mating pairs for each combination) maintained at natural conditions with regards to temperature and photoperiod, we demonstrated that a marked reproductive isolation between *C. aliai* and *C. stenocephalus* is present. This isolation corroborates the distinction at a species level of the two taxa and is similar to the isolation observed between *C. stenocephalus* and *C. asperatus* (Busato et al. 2014).

Conclusions

Data generated in this study, together with those obtained by Busato et al. (2014), clearly indicate that *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 is an independent species and not a mere subspecies of *C.* (*Cathoplius*) *stenocephalus* Lucas, 1866. Despite their close geographic distributional area, and the fact that they are geographical substitutes, the morphology and biology are very different between *C. aliai* and *C. stenocephalus ifniensis* Zarco, 1941. In *C. aliai*, adults are characterized by a shorter size and a more slender silhouette, larvae are smaller and pupae have a different chaetotaxy. The life cycle in *C. aliai* belongs to the annual univoltine winter semelparous type, where adults of the parental generation die after the breeding period ends. The duration of the development cycle, from eggs fertilization to the rising of neo-adults, is shorter in *C. aliai* then in *C. stenocephalus ifniensis* [70 vs. 72 days at temperatures varying from 15/19 to 17/21 °C (night/day)]. As a way of life, *C. aliai* is a typical sabulicolous steppe wandering species with an intensive running activity, while *C. stenocephalus ifniensis* is a more sedentary taxon carrying out the major part of its life inside shrubs. Feeding on *Theba* snails is common to both species and centers mainly on the predation of *T. subdentata meridionalis* (Sacchi, 1955) and *T. solimae* (Sacchi, 1955) (in the case of *C. stenocephalus ifniensis*) and on *T. subdentata meridionalis* and *T. chudeaui* (Germain, 1908) (in the case of *C. aliai*). Besides the different morphology and biology, results of crossbreeding experiments are indicative of a marked reproductive isolation between *C. aliai* and *C. stenocephalus*. When F1 hybrids are crossed with one another, progenies undergo a very high mortality during embryonic, larval and pupal development. Only a few F2 hybrids can be obtained, but these specimens usually die within some days from emergence.

According to these results, three *Cathoplius* species are spread along the Moroccan Atlantic coast: *C.* (*Cathoplius*) *asperatus* (Dejean, 1826) in the north, *C. stenocephalus* Lucas, 1866 in the center and *C. aliai* Escalera, 1944 in the south (Figure 29). *Carabus aliai* is distributed in an area south of the Anti-Atlas chain, from Plage Blanche (Guelmim province) to Lemsid and Bou Kra (south of Laâyoune/north of Boujdour). This area is approximately 450 km in length. *Carabus aliai* is therefore both a Saharan desert endemic and an Atlantic species. Moreover, it is the southernmost *Carabus* species of the western Palaearctic region.

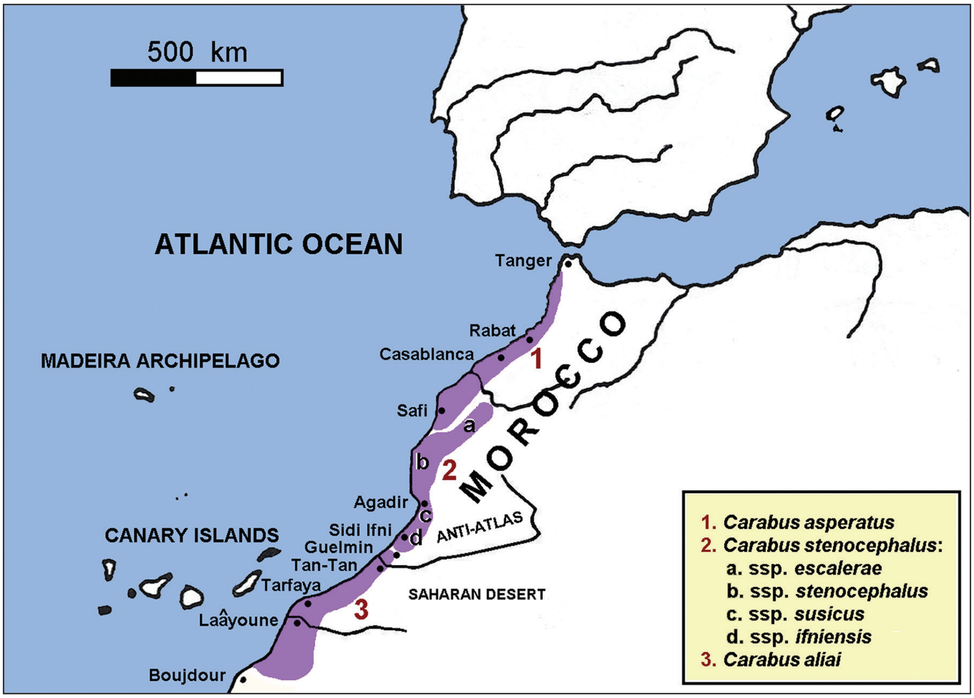


Figure 29. Distribution map of *Carabus* (*Cathoplius*) *aliai* Escalera, 1944 and its sister species.

A distributional gap, probably due to the lack of *ad hoc* investigations, is actually present between northern and southern *C. aliai* populations. Northern populations dwell in Guelmim (Plage Blanche) and Tan-Tan (Ben Khlil, Tan-Tan, Abteh) provinces, while southern populations are spread from present Laâyoune (Izik plateau, Sabkhat Tislatine, Bou Kra) to Boujdour (Lemsid, Metmarfag) provinces. The 200 km area in between, corresponding to present Tarfaya province, is represented by a very sandy desert with a few spots suitable for *C. aliai* such those around Akhfennir, Daoura and El Hagounia. Searches of this area during the favorable season will probably allow the identification of new *C. aliai* populations.

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RESEARCH ARTICLE

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Description of a new species of *Wormaldia* from Sardinia and a new *Drusus* species from the Western Balkans (Trichoptera, Philopotamidae, Limnephilidae)

Simon Vitecek[1], Ana Previšić[2], Mladen Kučinić[2], Miklós Bálint[3], Lujza Keresztes[4], Johann Waringer[1], Steffen U. Pauls[3], Hans Malicky[5], Wolfram Graf[6]

1 *Department of Limnology and Oceanography, University of Vienna, Althanstrasse 14, A-1090 Vienna, Austria* **2** *Department of Biology, Faculty of Science, University of Zagreb, Rooseveltov trg 6, HR-10000 Zagreb, Croatia* **3** *Senckenberg Biodiversity and Climate Research Centre (BiK-F), Senckenberganlage 25, D-60325 Frankfurt a.M., Germany* **4** *Hungarian Department of Biology and Ecology, Babeş-Bolyai University, Clinicilor 5–7, 400006 Cluj-Napoca, Romania* **5** *Sonnengasse 13, Lunz am See, Austria* **6** *Institute of Hydrobiology and Aquatic Ecology Management, University of Natural Resources and Life Sciences, Max Emanuel-Strasse 17, A-1180 Vienna, Austria*

Corresponding author: *Simon Vitecek* (simon.vitecek@univie.ac.at)

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Abstract

New species are described in the genera *Wormaldia* (Trichoptera, Philopotamidae) and *Drusus* (Trichoptera, Limnephilidae, Drusinae). Additionally, the larva of the new species *Drusus crenophylax* **sp. n.** is described, and a key provided to larval *Drusus* species of the *bosnicus*-group, in which the new species belongs. Observations on the threats to regional freshwater biodiversity and caddisfly endemism are discussed.

The new species *Wormaldia sarda* **sp. n.** is an endemic of the Tyrrhenian island of Sardinia and differs most conspicuously from its congeners in the shape of segment X, which is trilobate in lateral view. The new species *Drusus crenophylax* **sp. n.** is a micro-endemic of the Western Balkans, and increases the endemism rate of Balkan Drusinae to 79% of 39 species. Compared to other Western Balkan *Drusus*, males of the new species are morphologically most similar to *D. discophorus* Radovanovic and *D. vernonensis* Malicky, but differ in the shape of superior and intermediate appendages. The females of *D. crenophylax* **sp. n.** are most similar to those of *D. vernonensis*, but differ distinctly in the outline of segment X. Larvae of *D. crenophylax* sp. n. exhibit toothless mandibles, indicating a scraping-grazing feeding ecology.

Keywords

Caddisfly, Europe, larval key, taxonomy, conservation, Mediterranean, hydropower

Introduction

The Mediterranean area is a flora and fauna biodiversity hot-spot. The Tyrrhenian islands and the Balkans, in particular, are noteworthy for their high number of plant endemics (Médail and Quezél 1997, 1999; Nikolić et al. 2008; Fenu et al. 2010; Bacchetta et al. 2012), and mammal and invertebrate endemics (Holdhaus 1924, Vigne 1992, Muccedda et al. 2002, Griffiths et al. 2004, Grill et al. 2007). Freshwater biodiversity has recently become a focus of attention throughout Europe, including the Mediterranean region with the Western Balkans and Sardinia (e.g., di Sabatino 2003, Zakšek et al. 2009, Tierno de Figueroa et al. 2013, Klobučar et al. 2013, Weiss et al. 2014).

The genus *Wormaldia* currently comprises 204 species (Morse 2014) of which 36 species occur in Europe (Malicky 2005, Graf et al. 2008). Most species are widely distributed, but also several apparently highly endemic species have been described (Graf et al. 2008, Martínez-Menéndez and González 2011). Aquatic stages of the genus, with few exceptions, prefer crenal and rhithral sections of alpine to lowland streams, are caseless and behave as passive filter feeders using characteristic nets (Graf et al. 2008). Species in the genus exhibit characteristic male genitalia, but also comparatively high variability, particularly of the phallic structures (Malicky 2004, Martínez-Menéndez and González 2011, Neu pers. comm.), resulting in the description of several subspecies.

The genus *Drusus* is in the subfamily Drusinae Banks, and comprises 84 species (Malicky 2004, 2005; Kučinić et al. 2011a; Oláh 2010, 2011; Oláh and Kovács 2013). Larvae of the group prefer eucrenal to epirhithral sections of cold alpine or montane streams and brooks. Feeding ecology of *Drusus* larvae is complex, and three different feeding guilds can be distinguished based on the shape of larval mandibles and leg setation: filtering carnivores, omnivorous shredders, and scraping grazers (Pauls et al. 2008, Graf et al. 2009). Taxonomic richness of Drusinae is particularly high in the Western Balkans, including a high number of micro-endemics (Malicky 2004; Graf et al. 2008; Oláh 2010, 2011; Kučinić et al. 2011a, b; Oláh and Kovács 2013, Previšić et al. 2014a, b).

In this paper we describe a new species of *Wormaldia* and a new grazer *Drusus* species, including a key to the hitherto known larval stages of the *bosnicus*-group, in which *Drusus crenophylax* sp. n. belongs.

Materials and methods

Adults were collected using sweep nets and immature stages by handpicking. Collected specimens were stored in 70% and 96% EthOH, for morphological and molecular analyses, respectively.

Male and female genitalia were examined after being cleared in either KOH or lactic acid. Nomenclature of male genitalia of *Wormaldia* McLachlan follows Nielsen (1957, for *Wormaldia occipitalis* Pictet), 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

Table 1. Detailed list of *Drusus* specimens used for mtCOI analysis. Abbreviations: M adult male, F female; L larva; U unknown.

Species	Locality	Specimen ID/Stage	Accession #	Collectors
D. crenophylax	44°32.932'N, 17°23.562'E	fDsp4501M/M	KP793082	Dmitrović, Šukalo
D. crenophylax	44°33.003'N, 17°23.580'E	fDsp4502L/L	KP793083	Dmitrović, Šukalo
D. crenophylax	44°33.003'N, 17°23.580'E	fDsp4503L/L	KP793081	Dmitrović, Šukalo
D. crenophylax	44°33.003'N, 17°23.580'E	fDsp3401F/F	KP793084	Dmitrović, Šukalo
D. crenophylax	44°33.003'N, 17°23.580'E	fDsp3402F/F	KP793085	Dmitrović, Šukalo
D. vernonensis	41°0.887'N, 21°10.448'E	DdphPEIM1/M	KC881524	Kučinić, Graf
D. vernonensis	41°0.887'N, 21°10.448'E	DdphPEIM2/M	KP793087	Kučinić, Graf
D. vernonensis	41°0.887'N, 21°10.448'E	DdphPEIM3/M	KP793086	Kučinić, Graf
D. discophorus	Macedonia, Jablanica Mts.	fDds0110M/M	KP793089	Kučinić
D. discophorus	Macedonia, Jablanica Mts.	fDds0112F/F	KP793088	Kučinić

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 inked in Adobe Illustrator (v. 16.0.4, Adobe Systems Inc.).

Molecular genetic sequence data were used to support larval association and assess relationships to previously described *Drusus* species. DNA extraction and amplification of a 541-bp-long fragment of the mtCOI gene using standard primers (forward primer: Jerry, Simon et al. 1994, reverse primer: S20, Pauls et al. 2006) was performed as outlined by Pauls et al. (2008) and Previšić et al. (2009b). Sequences were edited manually using Geneious version R7 (http://www.geneious.com, Kearse et al. 2012) and aligned using MAFFT (Katoh and Standley 2013). Sequences were deposited in GenBank under Accession nos: KC881524, KP793081–KP793089 (Table 1). Inter- and intraspecific genetic distances (uncorrected *p*-distances) were calculated in Mega 4.0.1 (Tamura et al. 2007).

Taxonomy

Wormaldia sarda Graf & Malicky, sp. n.

http://zoobank.org/F02C5CF5-9043-463F-809B-FCD5D2B8FBD2

Material examined. Holotype. 1 male pupa, holotype: Sardinia, Gola di Gorruppo; 40°11.122'N, 9°30.104'E; 350 m a.s.l.; 28.03.2001; leg. Monika Hess, Ulrich Heckes; currently in coll. W. Graf, will deposited in the Biologiezentrum des Oberösterreichischen Landesmuseums, Linz, Austria.

Type locality. Italy, Sardinia.

Diagnosis. Morphology of the male terminalia suggests placement of the new species in *Wormaldia*. The species is unique in the European Trichoptera fauna, and easily differentiated from all other *Wormaldia* species by the combination of the following characters: (1) presence of median subtriangular protrusion in the distal half of the harpago, (2) membraneous dorsoproximal portion and trilobate lateral portions of segment X, and (3) distinct sclerotized structures visible on the invaginated phallus.

Description. *Adults* (in pupa). Habitus dark, sclerites and tergites brown; cephalic and thoracic setal areas pale; cephalic, thoracic and abdominal setation dark brown; legs light brown, proximally darker; haustellum and intersegmental integument pale cream. Wings brown mottled with golden patches. Male maxillary palp 5-segmented. Spurformula 2–4–4 in males.

Male genitalia (Fig. 1A–D). Segment IX in lateral view subrectangular, bulging anteriad; dorsal quarter reduced to a narrow transverse bridge, ventral 3/4ers broad (Fig. 1A). Segment X in lateral view trilobate: unpaired dorsal lobe strongly convex with a bicuspid apex, dorsoproximally membraneous; 1 lateromedian lobe, subovate, pointed on either side; 1 ventral lobe, posteriad, pointed on either side (Fig. 1A, B). Superior appendages suboval, curved dorsad in lateral view, flat with a rounded apex in dorsal and ventral view (Fig. 1A, C, D). Invaginated phallus terminally with a dorsal pair of sclerotized, laterad divergent tines and a ventral sclerotized plate; internally with 4 distinct tines (Fig. 1A, D). Coxopodite subovate in lateral view, ventrally with a sharp mediolaterad ridge (Fig. 1A, C). Harpago subovate in lateral view, in ventral view distally with a median subtriangular serrated protrusion flattened dorsoventrally (Fig. 1A, D).

Mature pupa (Fig. 2D–F). Mandibles tubular, dilated at the apex (Fig. 2E,F). Abdominal dorsal sclerites as in Fig. 2D.

Female and larva unknown.

Etymology. The species epithet refers to the island of Sardinia, the type locality.

Drusus crenophylax Graf & Vitecek, sp. n.

http://zoobank.org/4FBB2D55-59BD-46AB-8E39-B34F2D892C79

Material. Holotype. 1 male: Bosnia and Herzegovina, Cvrcka river; 44°32.932'N 17°23.562'E; 393 m a.s.l.; 01.10.2014; leg. Dejan Dmitrović, Goran Šukalo; specimen identifier: fDsp4501M. Paratypes: 2 females: Bosnia and Herzegovina, Spring of Cvrcka river, Vilenjska vrela; 44°33.003'N, 17°23.580'E; 456 m a.s.l.; 12.09.2012; leg. Dejan Dmitrović; specimen identifiers: fDsp3401F, fDsp3402F. 4 males, 3 females, 19 larvae: Bosnia and Herzegovina, Spring of Cvrcka river, Vilenjska vrela; 44°33.003'N, 17°23.580'E 456 m a.s.l.; 12.09.2012; leg. Dejan Dmitrović, Goran Šukalo; specimen identifiers for 3 larvae: fDsp4502L, fDsp4503L, fDsp4504L. Holotype and paratypes currently in coll. W. Graf, will deposited in the Biologiezentrum des Oberösterreichischen Landesmuseums, Linz, Austria.

Type locality. Bosnia and Herzegovina, Republika Srpska, Cvrcka River.

Diagnosis. Males of the new species are most similar to *Drusus discophorus* Radovanovic and *D. vernonensis* Malicky, but exhibit (1) subtriangular superior appendages in lateral view, (2) subtriangular, low tip of the intermediate appendage in lateral view, and (3) simple, rounded tips of intermediate appendages in caudal view. *Drusus disco-*

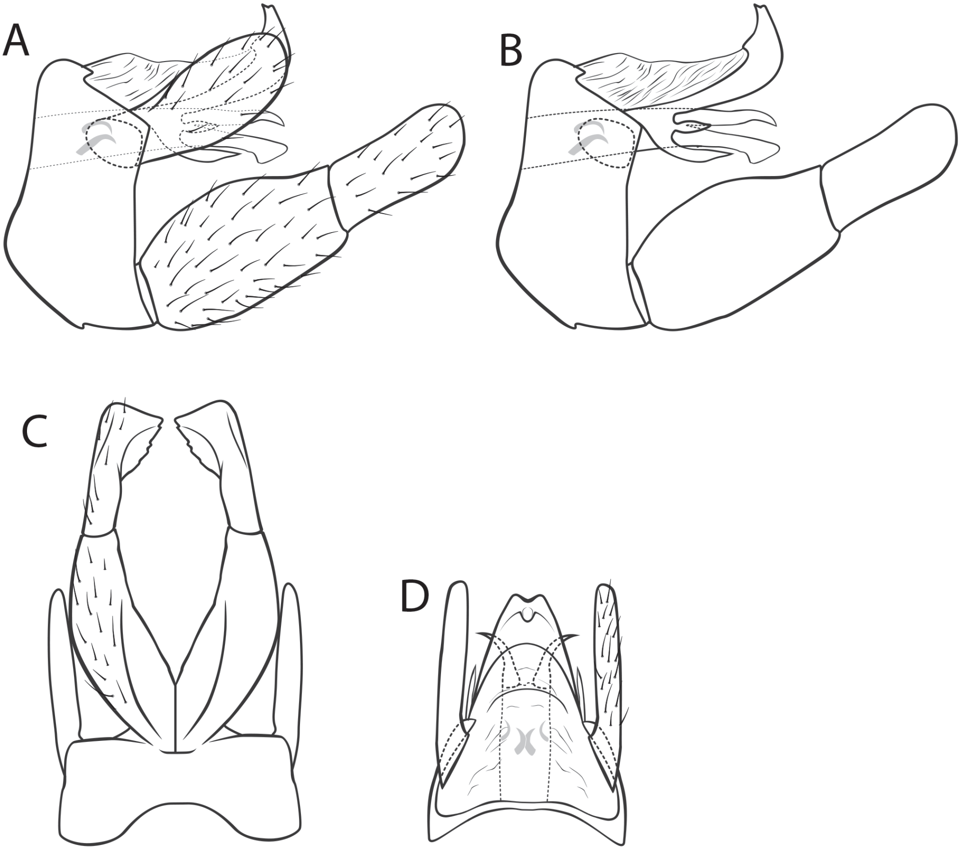


Figure 1. Male genitalia of *Wormaldia sarda* sp. n. **A** right lateral view, intact **B** right lateral view, superior appendage removed **C** ventral view **D** dorsal view.

phorus males have suboval superior appendages and a high round tip of the intermediate appendage in lateral view; *D. vernonensis* males have round superior appendages in lateral view and trilobate tips of intermediate appendages in caudal view.

Females of the new species show the reduced median lobe of the vaginal sclerite and high base of the lateral lobe of segment IX as typical for Balkan Drusinae, and are most similar to *Drusus vernonensis*, but exhibit (1) a sharp dorsal notch of segment X in lateral view, and (2) segment X with 2 round median lobes in dorsal view. *Drusus vernonensis* females have a rounded dorsal outline of segment X and lack the median lobes of segment X.

Larvae of the new species are most similar to *Drusus klapaleki* Marinković-Gospodnetić and *D. serbicus* Marinković-Gospodnetić, but exhibit (1) a semicircular area dorsomedially on the pronotum anterior the pronotal ridge void of white recumbent setae, (2) lateral gills, and (3) a subtriangular pronotal ridge in lateral view. Larvae of *D. klapaleki* have white recumbent setae covering the whole pronotum, and larvae of *D. serbicus* lack lateral gills and have an annular pronotal ridge.

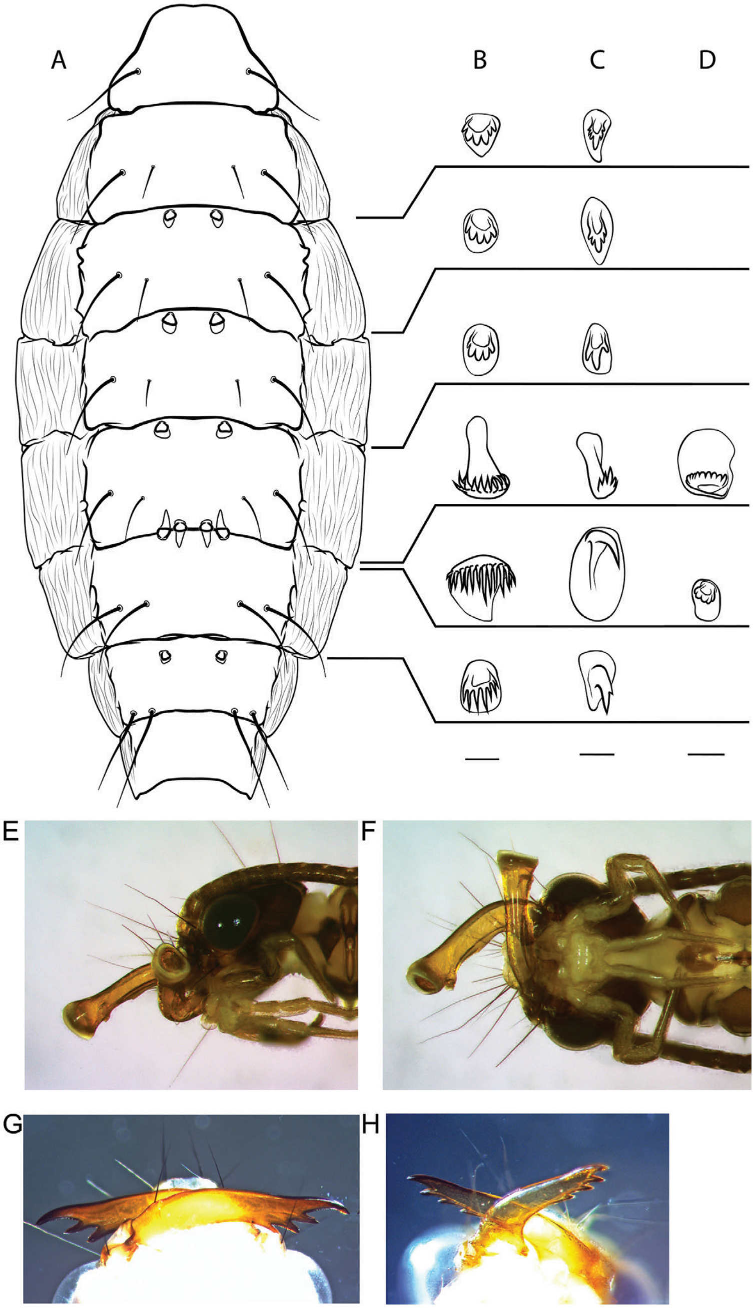


Figure 2. Pupal characteristics of *Wormaldia sarda* sp. n., *Philopotamus montanus*, and *Wormaldia* spp. **A** generalized pupal abdomen in dorsal view, depicting the position of the dorsal sclerites **B** dorsal sclerites of *Philopotamus montanus* **C** dorsal sclerites of *Wormaldia occipitalis* **D** dorsal sclerites of *W. sarda* sp. n. **E** head of *W. sarda* pupa in left lateral view **F** head of *W. sarda* pupa in ventral view **G** pupal mandibles of *Philopotamus montanus* in ventral view **H** pupal mandibles of *Wormaldia copiosa* in ventral view. Scale bars: 100 µm (**B**); 50 µm (**C, D**).

Description. *Adults*. Habitus dark; sclerites 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 smoky, with dark setae. Male maxillary palp 3-segmented. Forewing length 11–13.2 mm, spur formula 1–3–3 in males; forewing length 13–14.5 mm, spur formula 1–3–3 in females.

Male genitalia (Fig. 3A–E). Tergite VIII dark brown, in dorsal view cranially distinctly incised, with lighter areas around fused alveoli; setation concentrated at laterocranial borders of spinate areas; spinate area as two ± triangular laterocaudal lobes medially connected by a band of spines, embracing a medial, indent less sclerotized area (translucent in cleared specimens) with scarce spines. Ninth abdominal segment (IX) ventrally wider than dorsally in caudal view; in lateral view medially with a sharp caudad protrusion and a ventral protrusion, embracing the base of the inferior appendices. Superior appendages in lateral view subtriangular, somewhat Y-shaped with a shorter dorsal and a longer ventral protrusion separated by a slight indentation. Intermediate appendages in lateral view blocky with 2 tips, the proximal sharp, the distal high, rounded, rough; in dorsal view the tips parallel, extending laterally: a bar-shaped, laterally rounded distal tip and a sharp proximal tip, separated by a rounded excision with round edges; in caudal view approximately triangular, tips rounded. Inferior appendages (gonopods *sensu* Snodgrass 1935) in lateral view proximally wide, medially slightly constricted with a slight dorsal triangular protrusion, curved dorsadly in the slender posterior third; in dorsal, ventral and caudal view proximal part laterad, distal part approximately straight in dorsoventral plane, curved dorsad; in caudal view tips distinctly slender; setal alveoli fused, creating a rugged, less sclerotized ventral area. Parameres simple, with a distinct medial thorn-like spine and 2 proximal spines in the proximal half.

Female genitalia (Fig. 3F–I). Segment IX setation abundant, concentrated in the caudal half; lateral lobe of segment IX membraneous, in lateral view oblique triangular, the ventral edge about twice as long as the dorsal edge, with a dorsal sclerotized setose part protruding caudally; in dorsal and ventral view slender, projecting lateradly; in caudal view dorsal sclerotized setose part somewhat triangular. Segment X in lateral view with a proximal and a distal part, defined by a sharp dorsal notch; in dorsal view trapezoidal, with rounded shoulders, 2 small dorsal median lobes, and distally with 2 triangular, sharp-tipped lateral lobes, each with a lateral rounded setose and a small median rounded protrusion; ventrally unsclerotized, open. Supragenital plate in lateral view sinuously-edged quadrangular with a small, rounded dorsal protrusion, caudal line slightly indent; in ventral view quadrangular, in caudal view quadrangular, dorsally slightly wider than ventrally. Vulvar scale in lateral view triangular, rather straight, longer than the supragenital plate; in ventral view slender with 3 lobes: 2 lateral lobes, digitiform, roundly oval, straight; 1 median, short (reduced), of greater width than length: length approximately 1/6th of that of lateral lobes.

Fifth instar larva (Fig. 4A–I). Head capsule hypognathous, finely granulated with a field of microspinules dorsal to each eye, dark brown dorsally, fading to yellow ven-

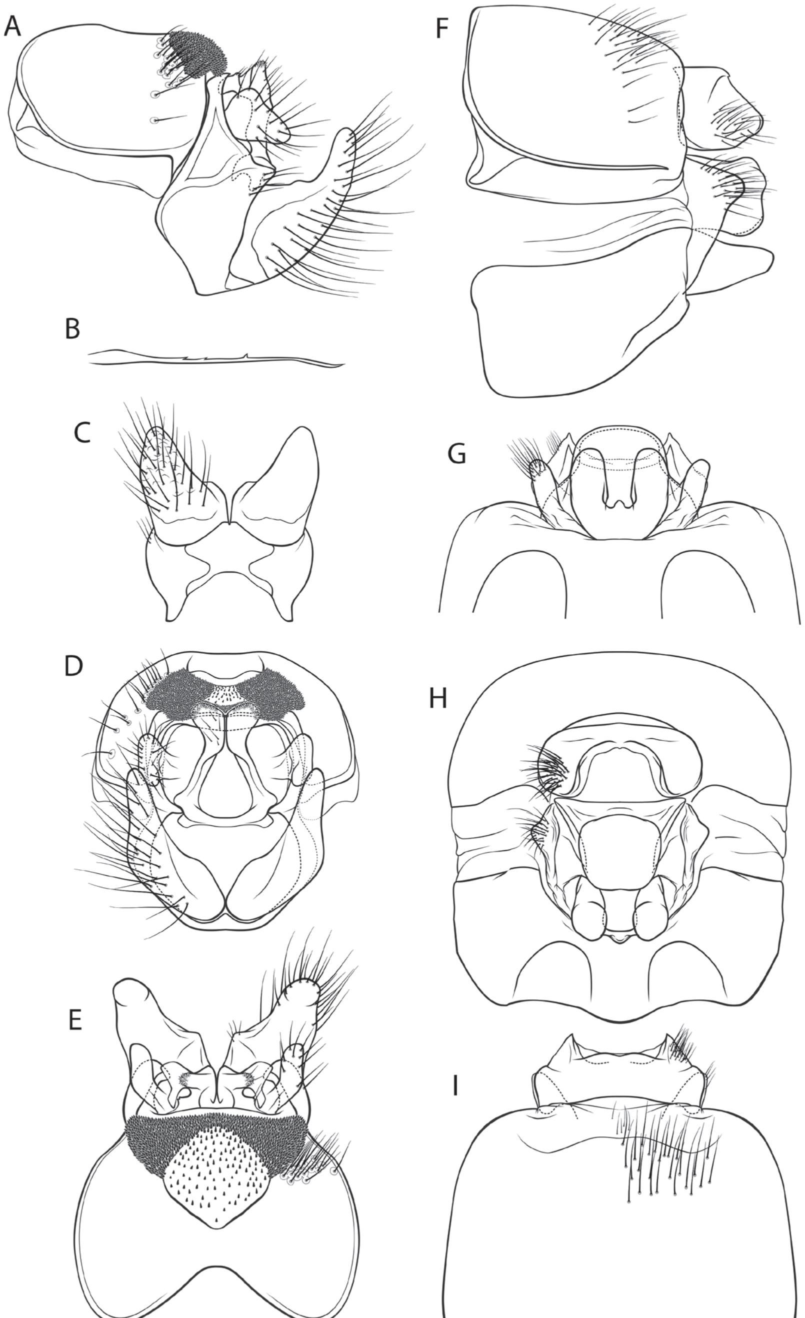


Figure 3. Genitalia of *Drusus crenophylax* sp. n. **A–E** male genitalia: **A** right lateral view **B** paramere in right lateral view **C** ventral view **D** caudal view **E** dorsal view **F–I** female genitalia: **F** right lateral view **G** ventral view **H** caudal view **I** dorsal view. Scale bar: 1 mm.

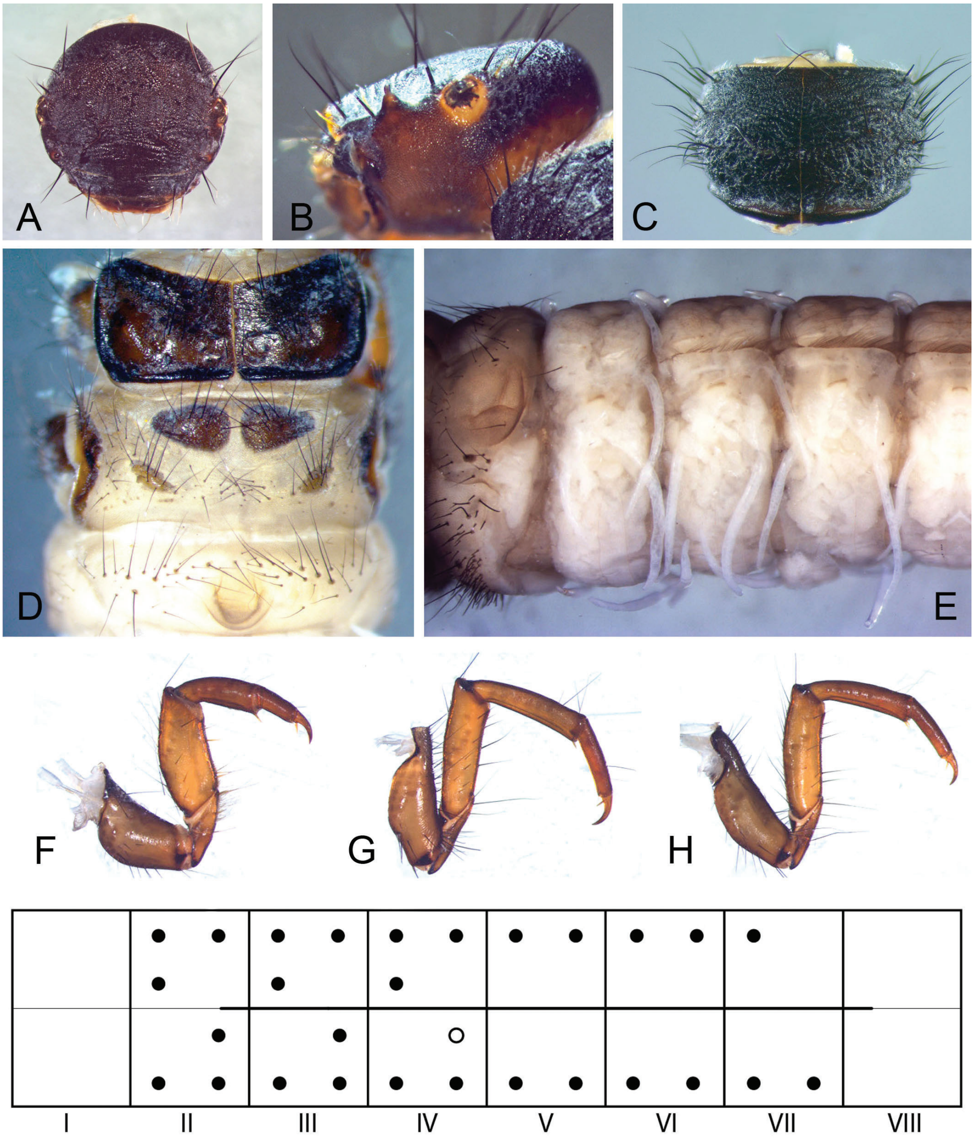
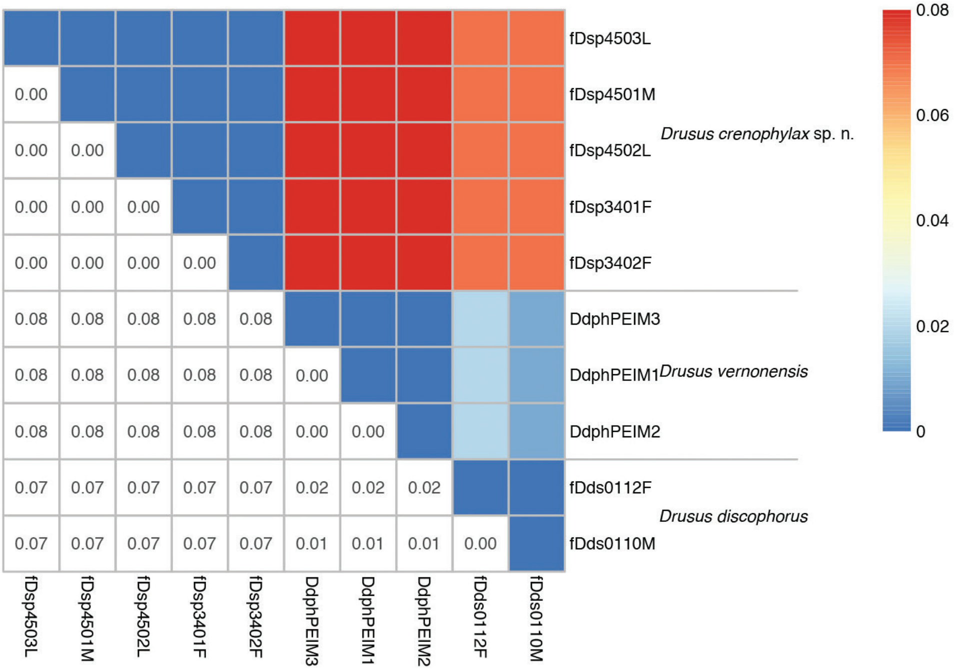


Figure 4. Larval characteristics of *Drusus crenophylax* sp. n. **A** head, frontal view **B** head, left lateral view **C** pronotum dorsal view **D** meso- and metathorax with abdominal segment I, dorsal view **E** abdominal segments I-V, left lateral view **F** left thoracic leg I, frontal view **G** left thoracic leg II; frontal view **H** left thoracic leg III, frontal view; bottom, gill and lateral line diagram, positions of gills are depicted as black circles, position of lateral line bold.

trally; 18 pairs of primary setae present: #1, 4, 6, 10, 12, 13 yellow and #6, 13 short, inconspicuous, the rest dark brown, long (Fig. 4A); antennae located on high carinae, each carina about as high as long, both strongly curved mediad (Fig. 4B); mandibles toothless. Pronotum dark brown, coarsely granulated; distinct medial ridge present,



	fDsp4503L	fDsp4501M	fDsp4502L	fDsp3401F	fDsp3402F	DdphPEIM3	DdphPEIM1	DdphPEIM2	fDds0112F	fDds0110M
fDsp4503L										
fDsp4501M	0.00									
fDsp4502L	0.00	0.00								
fDsp3401F	0.00	0.00	0.00							
fDsp3402F	0.00	0.00	0.00	0.00						
DdphPEIM3	0.08	0.08	0.08	0.08	0.08					
DdphPEIM1	0.08	0.08	0.08	0.08	0.08	0.00				
DdphPEIM2	0.08	0.08	0.08	0.08	0.08	0.00	0.00			
fDds0112F	0.07	0.07	0.07	0.07	0.07	0.02	0.02	0.02		
fDds0110M	0.07	0.07	0.07	0.07	0.07	0.01	0.01	0.01	0.00	

Figure 5. Distance matrix (lower left) and colour heat map (upper right) showing uncorrected inter- and intraspecific *p*-distances of the partial mtCOI sequence (541 bp) between *Drusus crenophylax* sp. n., *D. vernonensis* and *D. discophorus*. For detailed information on the haplotypes, see Table 1.

rounded, steeper anteriorly in lateral view; recumbent white setae present, but lacking in a semicircular area anterior the pronotal ridge (Fig. 4C); pronotal horn present. Mesonotum completely covered by 2 sclerites, dark brown, with darker apodemes; edges black; *sa*1 comprising 4–6 setae, *sa*2 and *sa*3 connected, comprising 28–34 setae in total on each sclerite (Fig. 4D). Metanotum with 3 pairs of sclerites: anteriomedian sclerites subtriangularly ovoid, dark brown with 11–19 setae; posteromedian sclerites rhomboid, pale brown, with 13–15 setae; lateral sclerites long, curved dorsally in lateral view, pale brown fading to yellow ventrally with a dark median spot and 21–25 setae (Fig. 4E). Legs yellow-light brown, dorsally and distally darker (Fig. 4F–H). Abdomen white (Fig. 4G), dorsal gills from II praesegmental position to VI praesegmental position, lateral gills from II praesegmental position to IV praesegmental position, ventral gills from II prasegmental position to VII postsegmental position; lateral line from last quarter of II to first quarter of VIII (Fig. 4I); abdomen I with 1 dorsal and 2 lateral protuberances, posterior sclerites absent on lateral protuberances, setal areas *sa*1–3 fused dorsally and ventrally (Fig. 4D, E), sternum bearing 2 setae with distinct basal plates; abdomen VIII with 2 long and 2–4 short posterodorsal setae on either side; abdomen IX with 1 posterodorsal seta on either side, dorsal sclerite IX semicircular, pale brown with 7 long and several shorter setae. Case simple, constructed of mineral particles.

Molecular species delimitation and larval affiliation. Analysis of the genetic distance of mtCOI between *Drusus crenophylax* sp. n. and the in the adult stage morpho-

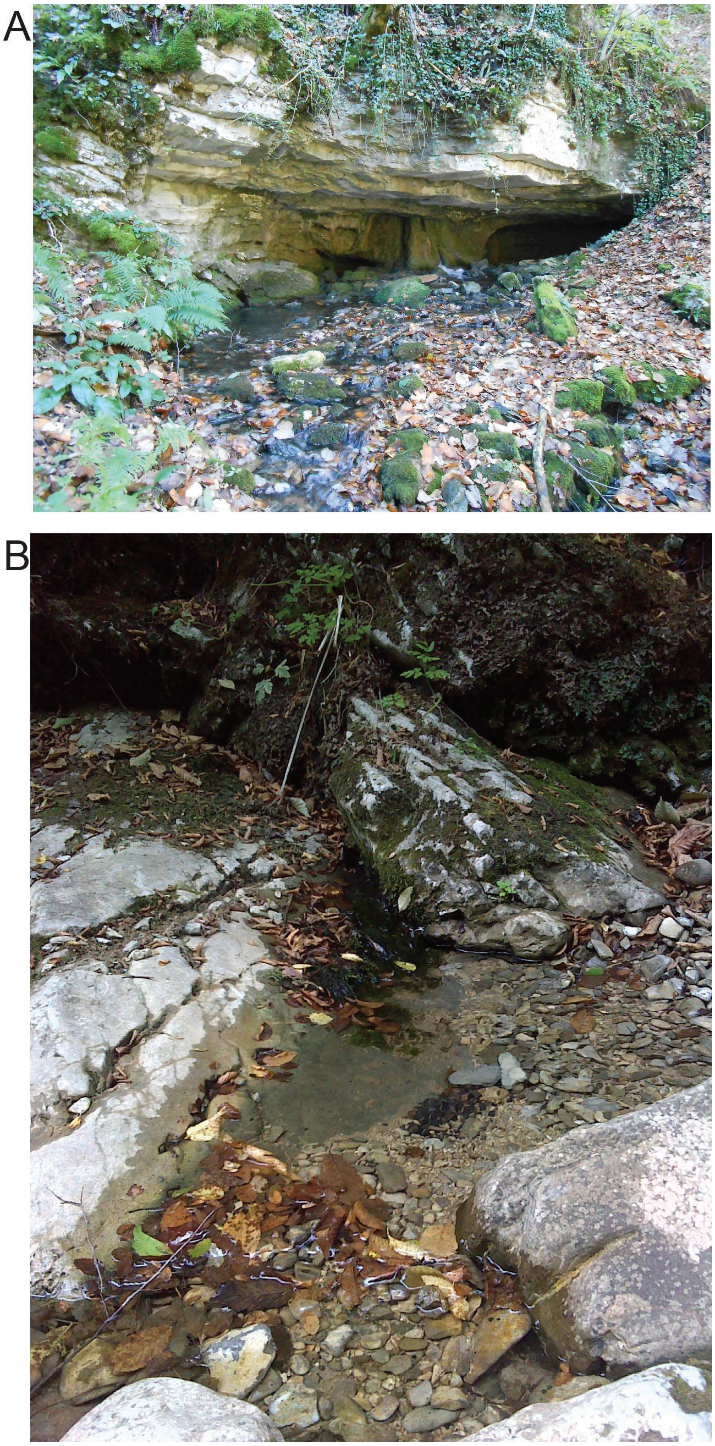


Figure 6. Habitat of *Drusus crenophylax* sp. n. at the type locality. **A** collection site of the larval paratypes **B** collection site of the male holotype.

logically most similar species, *D. discophorus* and *D. vernonensis*, clearly supports the recognition of the new species. Uncorrected *p*-distances recorded in a fragment of the mtCOI gene (ranging from 2–8%; Fig. 5), agree with the interspecific distances commonly recorded in Limnephilidae (e.g., Graf et al. 2005; Kučinić et al. 2011a; Previšić et al. 2014a, b) and other caddisfly families (e.g., Hydropsychidae; Pauls et al. 2010). Also, all haplotypes of *Drusus crenophylax* sp. n. adults were completely identical to another and those of undescribed *Drusus*-larvae collected at the locus typicus, enabling confident affiliation of larvae and adults of *D. crenophylax* sp. n.

Ecology and distribution. Drusinae species typically are members of crenal species communities, and mainly inhabit crenal sections of cold streams. Larval *D. crenophylax* were collected at eucrenal sections of the Cvrcka River (Fig. 6A, B) and behave as epilithic grazers, as indicated by mandible morphology (Pauls et al. 2008, Graf et al. 2009). Based on regional collection data, we assume that the species is a micro-endemic restricted to the watershed of the Cvrcka river.

Etymology. The species epithet is a compound name, combining *ϰϱηνον* ('well, spring, fountain' in Ancient Greek) and φυλαξ ('guard, keeper, protector' in Ancient Greek), terms that reflect the high degree of niche specificity of *Drusus* species, the majority of which inhabit crenal sections of streams (Graf et al. 2008).

Key to Drusinae larvae of the *bosnicus*-group

Drusinae have evolved into three distinct subclades reflecting feeding ecology of larvae (Pauls et al. 2008, Graf et al. 2009). The grazer clade *sensu* Pauls et al. 2008 represents the largest clade, comprising over 70 species in several subclades (Malicky 2004, 2005; Kučinić et al. 2011a; Oláh 2010, 2011; Oláh and Kovács 2013). Larvae of scraping grazers species characteristically develop toothless mandibles (Pauls et al. 2008, Graf et al. 2009, Waringer and Graf 2011). In the Western Balkans, the grazing *bosnicus*-group represents a group of morphologically similar endemics and comprises according to Marinković-Gospodnetić (1976) *Drusus bosnicus* Klapálek, *D. klapaleki* Marinković-Gospodnetić, *D. medianus* Marinković-Gospodnetić, *D. plicatus* Radovanović, *D. radovanovici* (Marinković-Gospodnetić), *D. ramae* Marinković-Gospodnetić, *D. septentrionis* (Marinković-Gospodnetić) and *D. vespertinus* Marinković-Gospodnetić (Kučinić et al. 2011a).

Larvae of the *bosnicus*-group also develop, with the exception of *D. ramae* (Kučinić et al. 2010), a field of microspinules close to each eye (Kučinić et al. 2011a, b; Waringer et al. 2015). Further, carinae of *D. bosnicus*, *D. radovanovici*, *D. septentrionis* and *D. medianus* are high and curved mediad. Larvae of *D. crenophylax* sp. n. share those characters and can be integrated in the following dichotomous key (Waringer et al. 2015):

1 Head with flat vertex .... ***Drusus bosnicus*** (Kučinić et al. in press)
– Vertex evenly rounded **2**
2 Pronotum with thin long, yellow setation ....
.... ***Drusus radovanovici*** (fig. 17 in Kučinić et al. 2011a)

– Pronotum without thin long, yellow setation..**3**

3 Pronotum with numerous short, white, recumbent setae **4**

– Pronotum without numerous short, white, recumbent setae........................ .... ***Drusus septentrionis*** (figs 4, 5 in Kučinić et al. 2008)

4 Dorsal pronotal hump smoothly rounded ....***Drusus medianus*** (fig. 43 in Kučinić et al. 2010, figs 20–22 in Kučinić et al. 2011b)

– Dorsal pronotal hump with distinct ridge...**5**

5 Anterior metanotal sclerites narrowly subtriangular (width / length ratio ≥ 2.0)...***Drusus vespertinus*** (Previšić et al. 2009a)

– Anterior metanotal sclerites broadly subtriangular (width / length ratio < 2.0)..**6**

6 In lateral view, dorsal pronotal ridge annular, posterior section sharply descending...***Drusus serbicus*** (Waringer et al. 2015)

– In lateral view, posterior section of dorsal pronotal ridge gently descending 7

7 White recumbent setae cover the entire pronotum...................................... ....***Drusus klapaleki*** (Kučinić et al. 2011b)

– White recumbent setae lacking in a semicircular area anterior to the pronotal ridge ....***Drusus crenophylax* sp. n.**

Discussion

Systematic significance of *Wormaldia sarda* sp. n.

The Tyrrhenian islands and Sardinia in particular have been renowned for their relictual fauna and flora for a long time (Holdhaus 1924) and represent one of the Mediterranean biodiversity hotspots (Grill et al. 2007). *Wormaldia sarda* sp. n. represents an addition to the distinct Sardinian biodiversity. As no species similar to *W. sarda* sp. n. are recorded from neither northern Africa nor mainland Europe, it is likely that this species is restricted to Sardinia, as are several other species such as *Crunoecia irrorata sarda* Curtis, *Stactobia ericae* Malicky or *Hydropsyche sattleri* Tobias (Graf et al. 2008). However, the geological history and geographic proximity of the Tyrrhenian islands – Sardinia and Corsica in particular (Vigliotti et al. 1990) – suggest that some species may occur on both islands. For instance, *Leptodrusus budtzi* Ulmer or *Micrasema togatum* Hagen occur also on Corsica, or other Mediterranean islands (Graf et al. 2008).

The distinct apomorphic characters, particularly the modified segment X and the very different pupal characters (mandibles, dorsal abdominal sclerites; Fig. 2D–F), might warrant establishing a new genus for this species. The pupal characteristics alone are strikingly different from those of either *Wormaldia* or *Philopotamus* (Lepneva 1964; Fig. 2). However, since pupae of only three species of *Wormaldia* are described (Nielsen 1942, Lepneva 1964) the range of genus-level pupal characters remains unknown. Further, modifications of segment X are common in southeast Asian species of *Wormaldia* (Malicky 2010). Tooth-like structures on segment X similar to the ones observed in *W. sarda* sp. n. are present in *Wormaldia* species from Thailand (e.g., *W. acheloos* Malicky & Chantaramongkol, *W. congina* Malicky & Chantaramongkol, *W. lot* Malicky & Chantaramongkol), or Sulawesi (*W. otaros* Neboiss). Nevertheless, *Wormaldia* species with a phallus shaped as in *W. sarda* sp. n. have not yet been described. Since the whole genus is in need of revision (Malicky 2005, Malicky unpubl. data), we refrain, in the interest of taxonomic stability, from creating a new genus.

Aquatic diversity of the Western Balkans under threat

Endemic freshwater species are particularly vulnerable to global change and (anthropogenic) habitat degradation (Hering et al. 2009, Tierno de Figueroa et al. 2010, Bálint et al. 2011, Conti et al. 2014). The Balkans is rich in apparently endemic freshwater species (Griffiths et al. 2004). Recent taxonomic efforts in the Western Balkans increased the number of endemic Drusinae taxa to 31 of 39 described Drusinae species (Previšić et al. 2014b, Vitecek et al. unpubl. data). Further, several endemic species of *Chaetopteryx* were recently described from the Western Balkans (Oláh et al. 2012, Kučinić et al. 2013) indicating the need for further systematic investigations on an underestimated diversity of southeastern Europe.

The construction of hydropower dams in emerging economies is currently one of the greatest threats to freshwater biodiversity (Zarfl et al. 2014). Small hydropower plants fed by small cold-water mountain rivers such as the Cvrcka River are currently under construction throughout the Western Balkans (Freyhof 2012, Schwarz 2012), and gravely threaten the habitats that harbour endemic highland caddisflies such as Drusinae (Previšić et al. 2014a, Vitecek et al. unpubl. data, this study), or *Chaetopteryx* species (Kučinić et al. 2013). The description of *Drusus crenophylax* sp. n. highlights the importance of biodiversity research in southern Europe, and demonstrates that the currently prevailing energy policy will likely result in the loss of known and unknown biodiversity.

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RESEARCH ARTICLE

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Two new *Neuratelia* Rondani (Diptera, Mycetophilidae) species from Western Palaearctic: a case of limited congruence between morphology and DNA sequence data

Olavi Kurina[1], Erki Õunap[1,2], Kadri Põldmaa[3]

1 *Institute of Agricultural and Environmental Sciences, Estonian University of Life Sciences, Kreutzwaldi st 5D, 51014 Tartu, ESTONIA* **2** *Department of Zoology, Institute of Ecology and Earth Sciences, University of Tartu, Vanemuise 46, 51014 Tartu, ESTONIA* **3** *Department of Botany, Institute of Ecology and Earth Sciences, University of Tartu, Ravila 14a, 50411 Tartu, ESTONIA*

Corresponding author: *Olavi Kurina* (olavi.kurina@emu.ee)

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Abstract

Two new Mycetophilidae species, *Neuratelia jabalmoussae* **sp. n.** and *Neuratelia salmelai* **sp. n.** are described on the basis of material collected from Lebanon, Estonia and Finland. Detailed figures of male terminalia and photographs of general facies are provided along with discussions of their morphological distinction from sibling species. For the first time molecular characters are used to distinguish new fungus gnat species. Molecular analysis relies on cytochrome oxidase subunit one (COI) but has additionally been corroborated by information from the 28S and ITS2 regions of nuclear ribosomal DNA. Situations where morphological and molecular data provide conflicting evidence for species delimitation are discussed. A new country record from Georgia is provided for *Neuratelia caucasica*.

Keywords

Mycetophilidae, *Neuratelia*, new species, Western Palaearctic, systematics, molecular analysis, COI, ITS2, 28S

Introduction

The genus *Neuratelia* Rondani, 1856 forms a well delimited clade in the subfamily Sciophilinae (Mycetophilidae), as sister group to the remaining Sciophilinae (Borkent & Wheeler, 2013). According to Søli et al. (2000) it is characterised by the following combination of characters: laterotergite setose, M and CuA clearly branched but base of M_1 obsolete, R_5 strongly sinuate, C produced about one fifth of the distance between apex of R_5 and apex of M_1, and tibia with distinct setae. Very little is known about their biology; according to Laffoon (1965) the larvae of one species were found in moss. This is, however, challenged by Hutson et al. (1980). Altogether 31 extant species are known from across the world including 16 species from the Palaearctic region (Matile 1974, Zaitzev 1994, Sasakawa 2004), 13 species from the Nearctic region (Borkent and Wheeler 2013) and one from both the Neotropical and Oriental regions (Bechev 2000). Additionally, three species have been described from fossils (Evenhuis 2014). Among the Palaearctic species seven are so far known to occur in the Western Palaearctic. There are no keys to cover all described species of the world, of only the Palaearctic region or even just in Europe. For Western Palaearctic species, the most exhaustive one is the key by Zaitzev (1994) that excludes, however, several European species.

So far, alpha-taxonomy of fungus gnats has been carried out using traditional taxonomic methods, primarily morphological examination. Though in recent years nucleotide data have been implemented to address the phylogeny of this group (e.g. Rindal et al. 2009a, 2009b, Ševčík et al. 2013, 2014), to associate sexes of one species (Kurina et al. 2011) and in population genetic studies (Dörge et al. 2014). Hippa and Ševčík (2014) provided mitochondrial 12S and 16S sequences in the description of *Nepaletricha sigma*. Despite that, no molecular information has so far been utilised for delimitation of a new fungus gnat species. This is surprising, as using a 658-bp fragment from the 5' end of the mitochondrial cytochrome oxidase gene subunit 1 (COI) – the so-called 'DNA barcode' (see Hebert et al. 2003) – has become an increasingly common practice in discriminating insect species during recent years (e. g. Yassin 2008, Huemer and Hebert 2011, Riedel et al. 2013). Sometimes, acquiring additional genetic data from other loci has also been used to corroborate findings discovered by studying DNA barcodes (e.g. Õunap and Viidalepp 2009, Raupach et al. 2010, van Nieukerken et al. 2012).

The aim of this article is to publish taxonomic and faunistic information about Western Palaearctic *Neuratelia* specimens that the senior author has accumulated over recent years. Both morphological and molecular data were used for species delimitation. This resulted in describing two new species – one from Estonia and Finland and another from Lebanon.

Material and methods

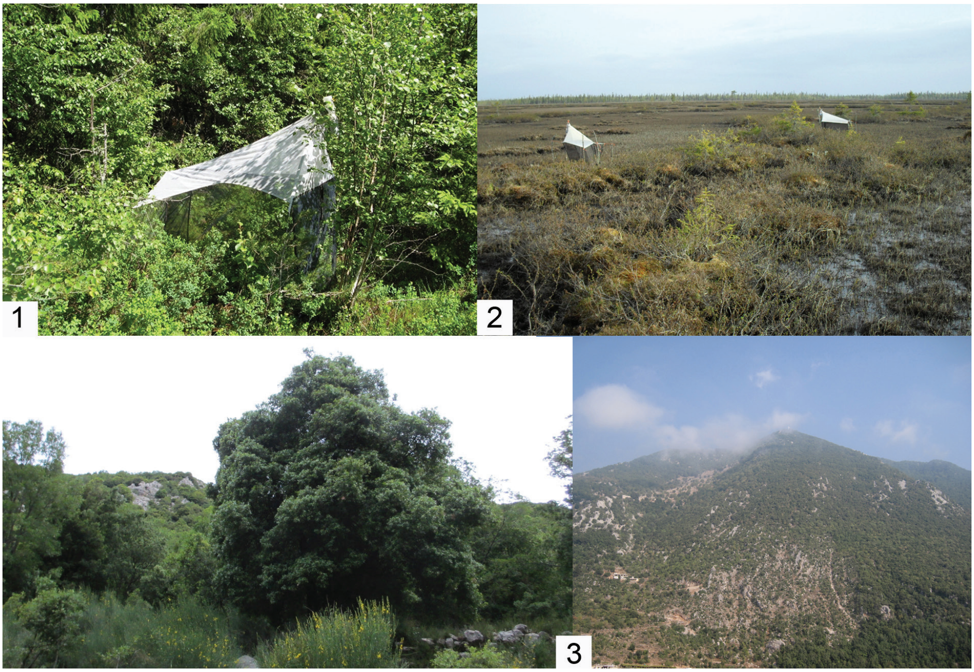
Collection, preparation, illustration and morphological study

The examined material of two new species was collected from Estonia and Finland using Malaise traps, and from Lebanon by light trapping, respectively. The Estonian locality lies at the herb rich edge of a mixed forest (Fig. 1) while the Finnish localities are predominantly wet fen habitats (Fig. 2) with variable vegetation irrigated by occasional springs. All Finnish localities are from the northern part of the country. In Lebanon, the material was collected from Jabal Moussa Biosphere Reserve, north-east of Beirut, characterised by karstic mountains with evergreen sclerophyllous vegetation (Fig. 3). The additional studied *Neuratelia* material was collected from Georgia, Greece, Slovakia, Finland and Estonia by sweep netting and Malaise trapping.

All specimens were stored initially in ethyl alcohol within which parts of them – after studying under a stereomicroscope Leica S8APO – are still preserved. For more detailed study of male terminalia, they were detached and macerated in a 10% solution of KOH, followed by neutralization and washing in distilled water. The remaining chitinous parts were thereafter inserted into glycerine for study, including black and white illustrations, and preserved as glycerine preparations in polyethylene microvials (see also Kurina 2003). A few specimens including their terminalia were slide mounted in Euparal following the method described by Hippa and Kurina (2012). The current preservation method of each specimen is indicated in the material section. The measurements are given as the range of measured specimens followed by the mean value, while measurements from the holotypes are given in square brackets. The ratios of the three apical palpal segments are given as 3rd:4th:5th. All measurements are taken from specimens in alcohol. Morphological terminology follows Søli et al. (2000).

The habitus photos have been made in an alcohol medium using a Canon 7D camera with a Canon MP-E65 (F2.8 1–5×) lens (see Kurina et al. 2011). The photos of thorax and terminalia were combined using the software LAS V.4.1.0. from multiple gradually focused images taken by a Leica DFC 450 camera attached to a Leica 205C stereomicroscope or Leica DM 6000 B compound microscope, respectively. Adobe Photoshop CS5 was used for editing the figures and compiling the plates. Black and white illustrations of the terminalia were prepared using a U-DA drawing tube attached to an Olympus CX31 compound microscope.

The material is deposited in the Institute of Agricultural and Environmental Sciences, Estonian University of Life Sciences [former Institute of Zoology and Botany], Tartu, Estonia (IZBE), in the Zoological Museum, University of Turku, Finland (ZMUT) and in the personal collection of J. Salmela, Rovaniemi, Finland (JSPC).



Figures 1–3. Collecting localities of *N. salmelai* sp. n. (**1, 2**) and *N. jabalmoussae* sp. n. (**3**). **1** Palupõhja in Estonia (holotype) **2** Kaita-aapa (Sodankylä) in Finland (a paratype) **3** Mar Elias in Jabal Moussa Biosphere Reserve, Lebanon (holotype).

Molecular techniques

The genomic DNA was extracted using High Pure PCR Template Preparation Kit (Roche Diagnostics GmbH, Mannheim, Germany). Anterior segments of the abdomen that had been stored after dissection of genitalia were crushed and used for the extraction. This process was carried out following the manufacturer's instructions for extraction of genetic material from mammalian tissue.

In total, one mitochondrial and two nuclear markers were sequenced. A 658-bp 'barcoding' fragment from close to the 5' terminus of the mitochondrial gene cytochrome C oxidase subunit 1 (*COI*), was amplified and sequenced using primers LCO1490 (5'-GGT CAA CAA ATC ATA AAG ATA TTG G-3') and HCO2198 (5'-TAA ACT TCA GGG TGA CCA AAA AAT CA-3') (Folmer et al. 1994). A 695-701-bp fragment covering expansion segments D1 and D2 of the nuclear 28S rRNA gene was sequenced using primers D1F (5'-GGG GAG GAA AAG AAA CTA AC-3') (Abraham et al. 2001) and D2R (5'-TTG GTC CGT GTT TCA AGA CGG G-3') (Belshaw and Quicke 1997). In the case that this preferable treatment was not successful, the desired part of the 28S was sequenced in two fragments, combining D1F with D1R (5'-CAA CTT TCC CTT ACG GTA CT-3') (Abraham et al. 2001) and D2R with D2F (5'-AGA GAG AGT TCA AGA GTA CGT G-3') (Belshaw and Quicke 1997). In addition, a fragment of the internal transcribed spacer 2 region (ITS) located between the 5.8S rRNA and 28S rRNA genes was sequenced using primers ITS2A (5'-TGT GAA CTG CAG GAC ACA T-3') and ITS2B (5'-TAT GCT TAA ATT CAG GGG GT-3') (Beebe and Saul 1995). PCR was performed in a total volume of 25 µl, with the reaction mixture containing 1X HOT FIREPol® Blend Master Mix Ready to Load (Solis BioDyne, Tartu, Estonia), 10 pmol of primers and 20-80 ng of purified genomic DNA. PCR was carried out in an Eppendorf Mastercycler epigradient thermocycler (Eppendorf AG, Hamburg, Germany). Its conditions involved an initial denaturation at 95 °C for 15 min, 35 cycles of 30 s at 95 °C, 30 s at 45–60 °C (depending on primers) and 1 min at 72 °C, followed by a final extension at 72°C for 10 min. PCR products were visualised on a 1.2% agarose gel, and 20 µl of the PCR solution was treated with fast alkaline phosphatase and exonuclease I (Thermo Scientific, Pittsburgh, USA). In some cases, direct sequencing from PCR solution was not possible due to multiple products. To sequence these samples, desired products were cut from agarose gel and extracted using a High Pure PCR Product Purification Kit (Roche). DNA cycle sequencing was performed either by Macrogen Europe (Amsterdam, Netherlands) or by the Estonian Biocentre (Tartu, Estonia). Both DNA strands were sequenced for all studied markers.

Phylogenetic analysis

Consensus sequences were created with Geneious R7 (Biomatters Ltd., Auckland, New Zealand) or Sequencher 5.1 (Gene Codes, Ann Arbor, MI, USA). Sequences were double-checked by eye and aligned using ClustalW (Thompson et al. 1994) in BioEdit 7.2.5 (Hall 1999). Two phylogenetic analyses were performed using either only COI or all three regions (COI, 28S, ITS2). *Neuratelia minor* was used as an outgroup in all phylogenetic analyses.

For COI, a neighbour-joining tree implementing Kimura 2-parameter model (a standard model analysing DNA barcode data, see e.g. Waugh 2007, Õunap and Viidalepp 2009, Hausmann et al. 2013) was constructed in MEGA6 (Tamura et al. 2013). Clade credibilities were assessed by bootstrapping (1000 replications). The tree was visualised using MEGA6.

For the concatenated dataset, data were first divided into three subsets according to the markers used (COI, 28S and ITS). Thereafter, PartitionFinder 1.1.1 (Lanfear et al. 2012) was used to select the most effective partitioning scheme and best substitution model for each partition. Accroding to PartitionFinder results, COI and ITS were treated together as one partition keeping 28S separately for ML analysis with RAxML 7.7.1 (Stamatakis et al. 2008). A GTR+I substitution model was implemented on both partitions. Analysis was run using the default settings of the RAxML online platform (http://embnet.vital-it.ch/raxml-bb/index.php). Ten slow ML searches, one thorough ML search and 100 rapid boostrap replications were performed. The results of bootstrapping were drawn on a single best-scoring ML tree. Phylograms were visualised with FigTree v1.4.0 (http://tree.bio.ed.ac.uk/software/figtree/).

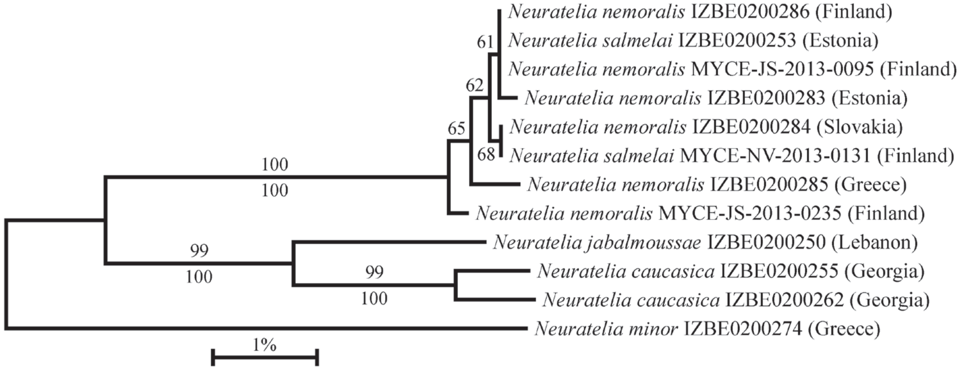


Figure 4. Neighbour-joining tree of the COI 'barcode' region of *Neuratelia* spp. Scale bar: Kimura 2-parameter genetic distance. Bootstrap supports are presented above the branches. Maximum likelihood analysis of the concatenated (COI, 28S, ITS2) dataset yielded a tree with similar topology, bootstrap supports for the divergencies obtained in this analysis are given below the branches. Support values inferior to 60 are not shown.

Table 1. Voucher numbers, depositories and GenBank accession codes of studied *Neuratelia* specimens. *COI*: cytochrome oxidase subunit I; *28S*: 28S rRNA; *ITS2*: internal transcribed spacer 2.

Species	Voucher No	Depository	COI	28S	ITS2
Neuratelia minor	IZBE0200274	IZBE	KP715935	KP715924	KP715947
Neuratelia nemoralis	IZBE0200283	IZBE	KP715936	KP715925	KP715948
Neuratelia nemoralis	IZBE0200284	IZBE	KP715937	KP715926	KP715949
Neuratelia nemoralis	MYCE-JS-2013-0095	ZMUT	KP715938	KP715927	KP715950
Neuratelia nemoralis	IZBE0200286	IZBE	KP715939	×	KP715951
Neuratelia nemoralis	IZBE0200285	IZBE	KP715940	KP715928	KP715952
Neuratelia nemoralis	MYCE-JS-2013-0235	ZMUT	KP715941	KP715929	KP715953
Neuratelia salmelai	IZBE0200253	IZBE	KP715942	KP715930	KP715954
Neuratelia salmelai	MYCE-NV-2013-0131	ZMUT	KP715943	KP715931	KP715955
Neuratelia jabalmoussae	IZBE0200250	IZBE	KP715944	KP715932	KP715956
Neuratelia caucasica	IZBE0200255	IZBE	KP715945	KP715933	KP715957
Neuratelia caucasica	IZBE0200262	IZBE	KP715946	KP715934	KP715958

Results

The morphology of studied material distinguished three previously known species of *Neuratelia* and a group of specimens, clearly delimited by characters of male terminalia. This group, represented by specimens from Estonia and Finland, resembles the widespread *N. nemoralis* (Meigen, 1818) and hereafter described and referred to as a new species – *N. salmelai* sp. n. In addition, another group of three specimens from different localities in Jabal Moussa Biosphere Reserve (Lebanon) had slight differences from *N. caucasica* Zaitzev, 1994 – a species only known from Caucasus. In the latter case, the species is described as *N. jabalmoussae* sp. n. but the morphological differences are diminutive underpinning the necessity of including DNA sequence data for species discrimination.

Sequencing the 'barcode region' of COI was successful for all specimens included in the current study. The success rate was lower for ITS2 and 28S rDNA, as all attempts to sequence 28S failed for one individual of *N. nemoralis*, and for a few specimens, only half of 28S or ITS was obtained (Table 1). GenBank accession numbers for all sequences are presented in Table 1. The NJ tree constructed on the basis of barcodes divided the studied specimens into three well-supported clusters differing from each other by at least 4%. One group comprised only *N. caucasica* and another only *N. jabalmoussae*, whereas *N. nemoralis* and *N. salmelai* were intermingled in the third clade (Fig. 4). Studying the concatenated dataset resulted with almost identical results, as *N. jabalmoussae* and *N. caucasica* remained clearly separate sister taxa with *N. nemoralis* and *N. salmelai* remaining inseparable on the ML tree (Fig. 4).

The species

Neuratelia jabalmoussae sp. n.

http://zoobank.org/95794D4E-8664-4BB5-80F2-762C125858BF
Figs 6, 7, 8, 9, 10, 15

Type material. *Holotype.* 1♂, LEBANON, Kesrouane Mar Elias, 34°03'06,9"N, 35°46'00,5"E, 1138 m a.s.l., at light, 27.v.–4.vi.2012, J. Kullberg leg. (IZBE0200250, slide mounted in Euparal with terminalia in glycerine). *Paratypes.* 1♂, LEBANON, Kesrouane Mar Geryes, 34°03'20,9"N, 35°44'28,9"E, 749 m a.s.l., at light, 26.v.–2.vi.2012, J. Kullberg leg. (IZBE0200251, in alcohol with terminalia in glycerine);1♂, LEBANON, Kesrouane Ghbele, 34°03'25,5"N, 35°43'02,5"E, 884 m a.s.l., at light, 26.v.–30.v.2012, J. Kullberg leg. (IZBE0200252, in alcohol).

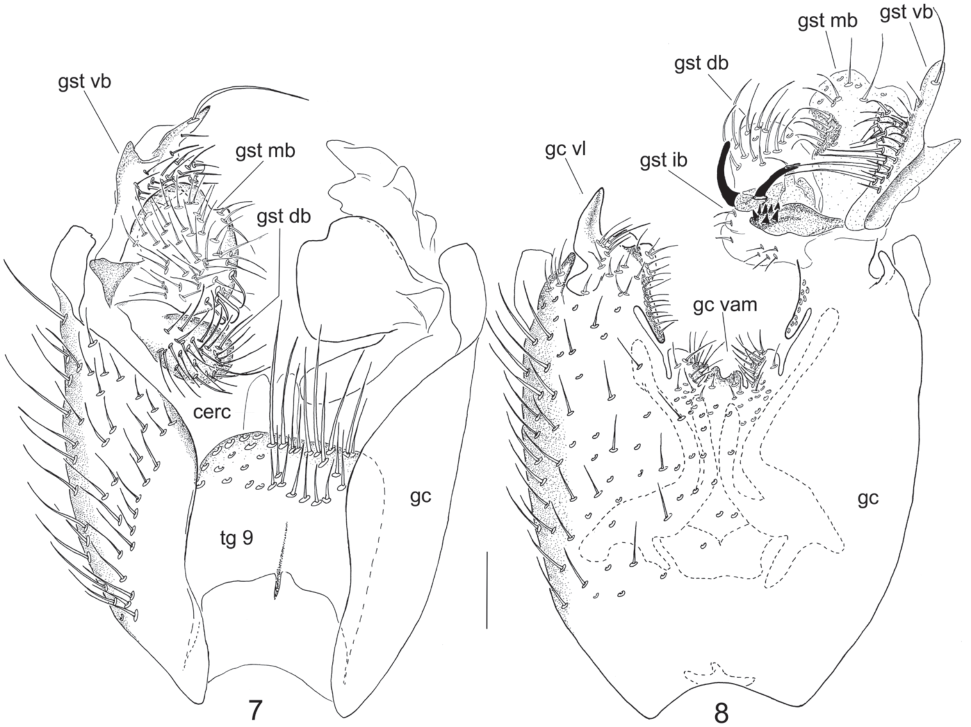
Description. Male (Fig. 6). Body length 5.4–5.8, 5.6 [5.6] mm (n=3).

Head dark brown, with numerous pale to yellowish setae. Three ocelli in a shallow and wide triangular arrangement, with laterals separated from eye margins by a distance slightly more than their own diameter. Face conical, about 0.8 times as wide as maximum height; clypeus rectangular, about 0.6 times as wide as high; both brown, setose, with setae on clypeus stronger than those on face. Mouthparts yellow. Palpus five segmented, yellowish with apex of fifth segment brownish. Ratios of three apical palpal segments 1.0 : 1.68–1.85, 1.77 [1.85] : 1.97–2.00, 1.98 [2.00]. Scape and pedicel light brown to brown, flagellomeres light brown, with short yellowish setae. First flagellomere yellowish at basal third. Flagellum evenly tapering; first flagellomere clavate, 2.9–3.1 times as long as broad apically, 2–13 flagellomeres cylindrical, fourth flagellomere about 1.7–2.5 times as long as broad, apical flagellomere slightly conical, 4.8–5.4 times as long as broad at base.

Thorax. All parts brown to dark brown, all setae yellow to light brownish. Mesonotum with evenly arranged numerous setae. Scutellum wholly setose with about 10

Figures 5–6. Habitus of *Neuratelia* species. **5** *N. salmelai* sp. n., terminalia detached **6** *N. jabalmoussae* sp. n. Scale bar = 1 mm.

stronger setae along the margin, not arranged to distinct pairs. Antepronotum with 8–9 and proepisternum with 4–7 setae of unequal size, laterotergite with 22–26 setae and mediotergite with ca 14–16 setae medially on lower part. Other pleural parts bare. Halteres pale yellow, setose.

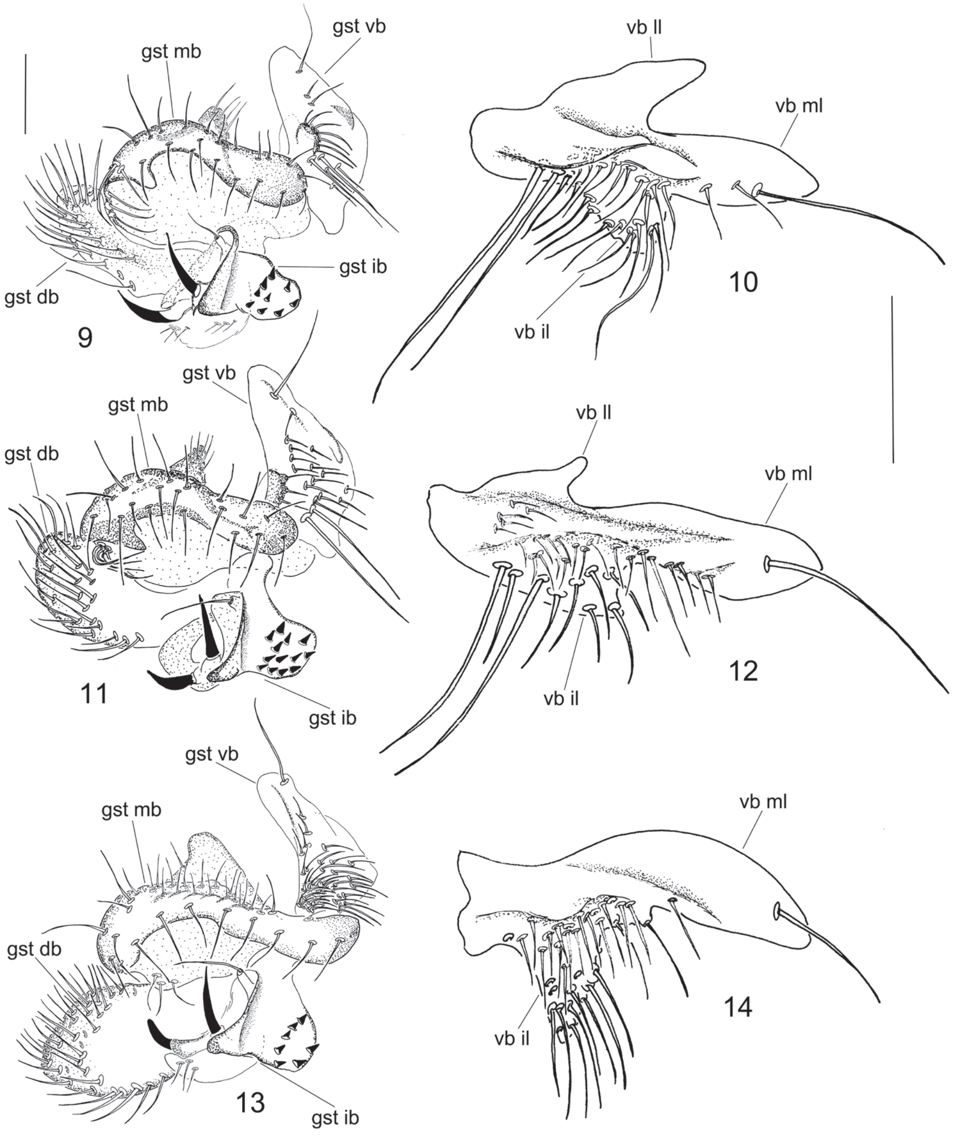


Figures 7–8. Male terminalia of *N. jabalmoussae* sp. n., dorsal view (**7**) and ventral view (**8**). Scale bar = 0.1 mm. Abbreviations: cerc = cerci; gc = gonocoxite; gc vam = ventroapical margin of gonocoxite; gc vl = ventroapical lobe of gonocoxite; gst db = dorsal branch of gonostylus; gst ib = internal branch of gonostylus; gst mb = medial branch of gonostylus; gst vb = ventral branch of gonostylus; tg 9 = IX tergite.

Legs. All coxae yellow, basally infuscated. All trochanters brown. All femora and tibiae yellow, tarsi seem considerably darker because of dense setae. Foretibia with 1–2 ad, 0–2 d and 2–3 pd. Midtibia with 4–7 a, 2–3 d, 1–2 av and 2–4 pd. Hind tibia with 7–8 a, 1–2 ad (1 at apex), 4–6 d, 0–1 pd, 5 p and with a posterior apical comb of setae. Ratio of femur to tibia for fore-, mid- and hind legs: 0.86–0.94, 0.9 [0.9]; 0.77–0.86, 0.82 [0.77]; 0.71–0.75, 0.73 [0.75]. Ratio of tibia to basitarsus for fore-, mid- and hind legs: 0.91–0.94, 0.93 [0.94]; 1.26–1.3, 1.27 [1.3]; 1.51–1.54, 1,53 [1.51].

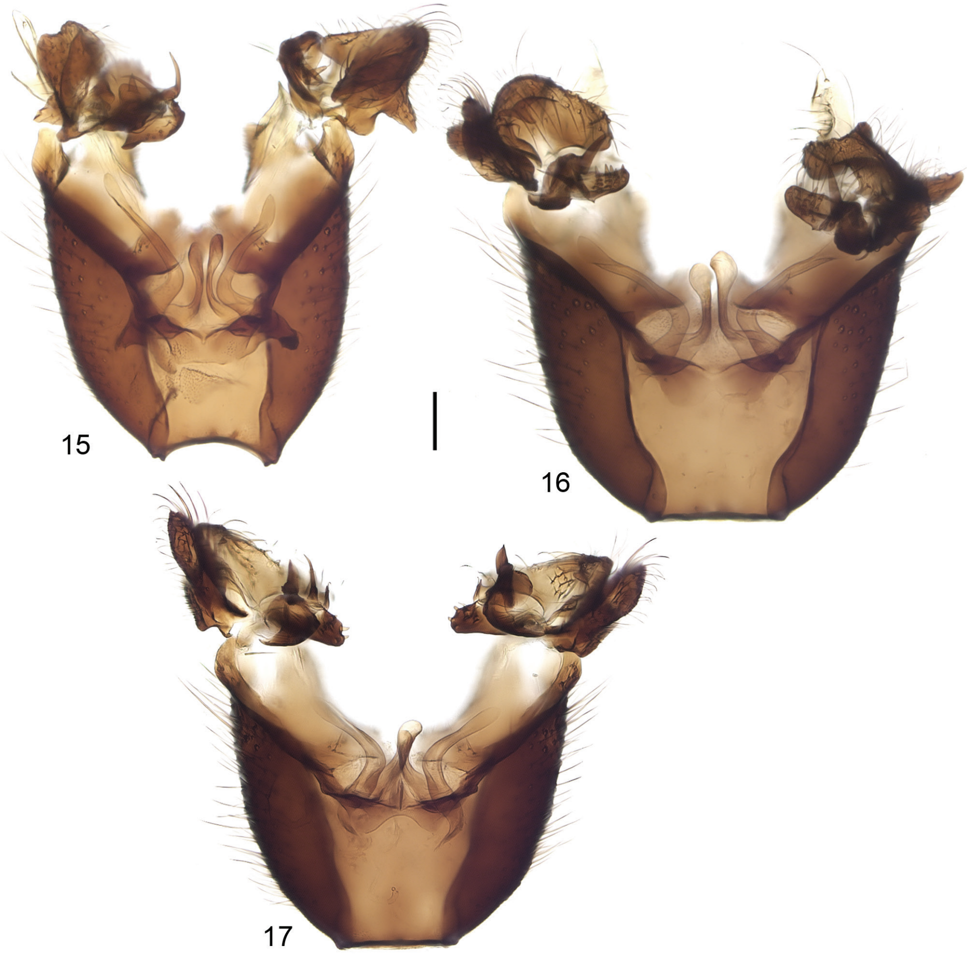
Wing hyaline, length 5.0–5.03, 5.02 [5.03] mm (n=3). All veins brown, costal and radial veins somewhat darker. Both surfaces of veins setose, except bare bM-Cu and r-m. Wing membrane with micro- and macrotrichia on both surfaces. Costa reaches very little from R_5 to M_1. Sc reaches costa at about one sixth between R_s and tip of R_1. R_5 sinuate. R_s about as long as crossvein r-m. M_1 basally obsolete: observable vein begins distally from middle of R_1. Cubital fork begins proximally from apex of Sc.

Abdomen with tergites brown and sternites yellowish. Tergites 6–7 somewhat darker. Terminalia (Figs 7, 8, 9, 10, 15) dark brown. Tergite 9 apically almost straight, with wide basal incision about one third of height of tergite. Basolateral



Figures 9–14. Male terminalia of *N. jabalmoussae* sp. n. (**9, 10**), *N. caucasica* Zaitzev, 1994 (**11, 12**) and *N. minor* (Lundström, 1912) (**13, 14**). **9**, **11**, **13** internal view of gonostylus **10**, **12**, **14** ventral view of ventral branch of gonostylus. Scale bars = 0.1 mm. Abbreviations: gst db = dorsal branch of gonostylus; gst ib = internal branch of gonostylus; gst mb = medial branch of gonostylus;gst vb = ventral branch of gonostylus; vb il = internal lobe of ventral branch of gonostylus; vb ll = lateral lobe of ventral branch of gonostylus; vb ml = medial lobe of ventral branch of gonostylus.

portions of tergite 9 narrow and proximally pointed. Setae on tergite 9 similar to these on the gonocoxite, the posteriormost ones slightly stronger than the others. Cerci separated, protruding over tergite 9, with strong apical setae deviating from



Figures 15–17. Male terminalia, dorsal view, tergite 9 removed. *N. jabalmoussae* sp. n. (**15**), *N. caucasica* Zaitzev, 1994 (**16**) and *N. minor* (Lundström, 1912) (**17**). Scale bar = 0.1 mm.

other setosity. The gonocoxite with a complex ventroapical lobe laterally; ventrobasally with wide shelving incision; ventroapical margin medially with lateral well delimited sub-circular and a medial apically concave setose structures. Dorsomedial margin of the gonocoxite slightly sinuous. The gonostylus with four branches. The dorsal branch simply oval, setose. The ventral branch trifid with 1) internal lobe setose including two stronger internally directed setae, 2) middle lobe elongated with a strong subapical seta, and 3) lateral lobe similar to internal lobe except being bare. Medial branch setose with a well delimited medial hump. Internal branch complex with two strong pointed spines and a lobe bearing 9 short spines ventrally on its apical part. The medial branch of the gonostylus connected with apical part of ventroapical lateral lobe of the gonocoxite. Parameres not protruding over ventroapical margin of gonocoxite.

Female. Unknown.

Biology. Unknown.

Etymology. The species is named after the type locality in Jabal Moussa Biosphere Reserve, Lebanon; the specific epithet is a noun in genitive case.

Specific discussion. *Neuratelia jabalmoussae* sp. n. is very similar to *N. caucasica*, into which it also runs to in the key by Zaitzev (1994) because of having the foretibia slightly shorter than the fore basitarsus. Also the male terminalia of these two species are extremely similar, differ in details as follows: 1) paramers not expanded apically (Fig. 15), while they are well expanded in *N. caucasica* (Fig. 16), 2) the lateral lobe of the trifid ventral branch of the gonostylus prominent, about half of the size of medial lobe (Fig. 10), while it is minute in *N. caucasica*, about one fifth of the size of medial lobe (Fig. 12), and 3) internal branch of gonostylus has 8 short spines on a separate lobe (Fig. 9), while there are 13 spines in *N. caucasica* (Fig. 11). Both species share the general outline of male terminalia also with Western Palaearctic species *N. minor* (Lundström, 1912) and with *N. microdigitata* Sasakawa, 2004, known from Japan. However, *N. minor* has the foretibia slightly longer than fore basitarsus and the ventral branch of gonostylus bifid instead of being trifid. *N. microdigitata* has the internal branch of gonostylus with finger-like processes apically on a separate lobe (cf. Sasakawa 2004: fig. 4) instead of short spines as in other three species. All four species have the similar branching of the gonostylus and two strong pointed spines on internal branch of the gonostylus.

Neuratelia caucasica Zaitzev, 1994

Figs 11, 12, 16

Studied material. GEORGIA. 2♂♂ 2♀♀, Surami, 42°01'34,2"N, 043°29'52,5"E, 941 m a.s.l., sweeping, O. Kurina leg. 18.v.2012 (IZBE0200255– IZBE0200258, in alcohol); 2♂♂ 1♀, Borjomi, 41°50'9,2"N, 043°19'56,7"E, 936 m a.s.l., sweeping, O. Kurina leg. 21.v.2012 (IZBE0200259, ♂, on pin with terminalia in glycerine; IZBE0200260, IZBE0200261, in alcohol); 3♂♂ 4♀♀, Mtirala NP, near visitor centre, 41°40'20,7"N, 041°52'31,8"E, 465 m a.s.l., sweeping, O. Kurina leg. 20.v.2013 (IZBE0200262, ♂, slidemounted in Euparal with terminalia in glycerine; IZBE0200263–IZBE0200268, in alcohol); 1♂ 2♀♀, Kintrishi NP, 41°45'11,7"N, 041°58'38,4"E, 453 m a.s.l., sweeping, O. Kurina leg. 22.v.2013 (IZBE0200269–IZBE0200271, in alcohol).

Specific discussion. Having been described from North Caucasus (Krasnodarsk region), the species has not been recorded since and the studied material represents the first records from Georgia. According to male terminalia the species is close to *N. minor* and *N. jabalmoussae* sp. n.

Neuratelia minor (Lundström, 1912)

Figs 13, 14, 17

Studied material. SLOVAKIA. 1♂, NP Muránska planina, Murán 3.5 km NE, sweeping, 48°45'46,5"N, 020°04'55,9"E, 483 m a.s.l. 30.v.2009, O. Kurina leg. (IZBE0200272, on pin with terminalia in glycerine; earlier published in Ševčík and Kurina 2011: 101); 1♂, NP Muránska planina, Šiance, sweeping, 48°46'14,7"N, 020°05'33,0"E, 656 m a.s.l. 30.v.2009, O. Kurina leg. (IZBE0200273, on pin with terminalia in glycerine; earlier published in Ševčík and Kurina 2011: 101). GREECE. 1♂, Central Macedonia, Kerkini lakes area, Vironia village, Beabies site, 41°19'15,4"N, 023°13'39,6"E, 1150 m a.s.l., Malaise trap, 19.–25.v.2008, G. Ramel leg. (IZBE0200274, slide mounted in Euparal).

Specific discussion. *Neuratelia minor* was described and figured by Lundström (1912: figs 8, 9) from Romania. Because the type material was subsequently destroyed, Matile designated neotype from Hungary and provided also a new figure of male terminalia (Matile 1974: fig. 6). Both figures are sufficiently detailed, presenting a bifid ventral branch of the gonostylus that clearly discriminates the species morphologically from *N. caucasica* and *N. jabalmoussae* sp. n. *Neuratelia minor* has a more eastern distribution in the Western Palaearctic but is also found in France and the Eastern Palaearctic (Chandler 2013).

Neuratelia salmelai sp. n.

http://zoobank.org/1554A8EF-A6FF-484D-9555-4855836A4263

Figs 5, 18, 20, 22, 24, 26, 28

Type material. *Holotype.* 1♂, ESTONIA. Palupõhja, Kaha (ME 57), Malaise trap, 58°25'54,68"N, 026°14'28,90"E, 29.vi.–8.vii.2009, V. Soon leg. (IZBE0200253, slide mounted in Euparal with terminalia in glycerine). *Paratypes.* 1♂, FINLAND. Lkor: Sodankylä, Kaita-aapa, Malaise trap, 67°50'45,5"N, 026°33'17,6"E, 5.vi.–3.vii.2012, J. Salmela leg. (IZBE0200254, in alcohol with terminalia in glycerine); 1♂, FINLAND. Lkoc: Kittilä, Kielisenpalo, Malaise trap, 68°01'16,6"N, 025°03'46,9"E, 26.vi–24.vii.2007, J. Salmela leg. (MYCE-NV-2013-0093 in ZMUT, in alcohol with terminalia in glycerine); 1♂, FINLAND. Lkoc: Kittilä, Vuotsonperän-jänkä, Malaise trap, 67°37'15,9"N, 025°26'43,6"E, 25.vi.–24.vii.2009, J. Salmela leg. (MYCE-NV-2013-0131 in ZMUT, in alcohol with terminalia in glycerine); 1♂, FINLAND, Lkor: Sodankylä, Pomokaira 67°52'19,2"N, 026°12'46,8"E, 11.6.-10.7.2013, J. Salmela leg. Malaise trap Salix swamp with seepages (DIPT-JS-2014-0199 in JSPC, in alcohol).

Description. Male (Figs 5, 18). Body length 5.8–6.5, 6.2 [5.8] mm (n=4).

Head (Fig. 18) brown to dark brown, with numerous setae. Three ocelli in a shallow and wide triangular arrangement, with laterals separated from eye margins by a distance about twice of their own diameter. Face conical, about 0.9 times as wide as

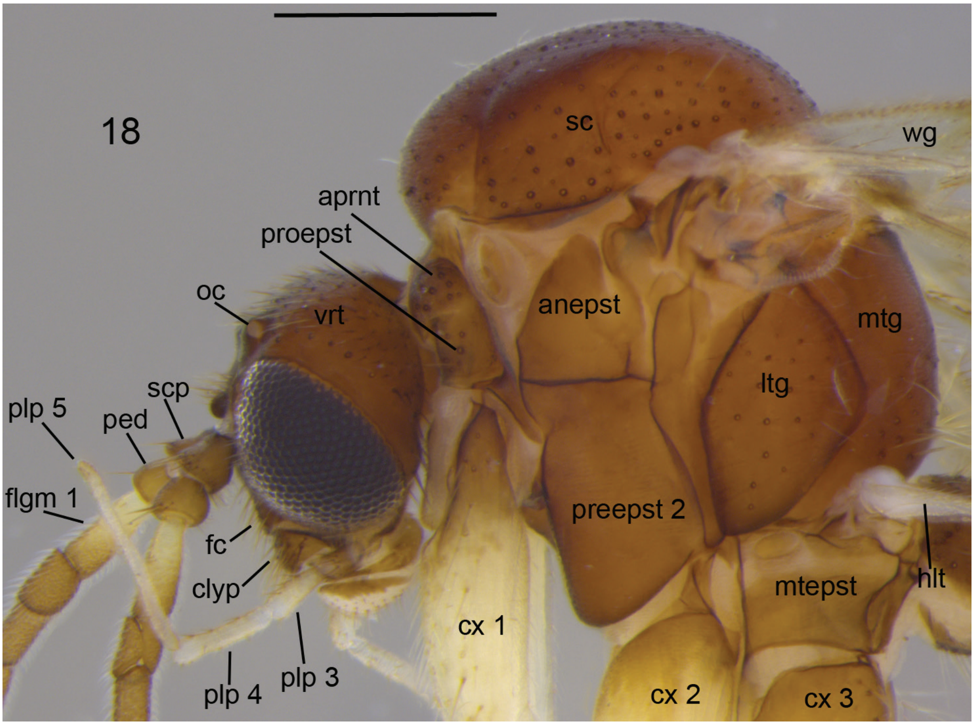


Figure 18. Head and thorax of *N. salmelai* sp. n. Scale bar = 0.5 mm. Abbreviations: anepst = anepisternum; aprnt = antepronotum; clyp = clypeus; cx = coxa; fc = face; flgm = flagellar segment; hlt = halter; ltg = laterotergite; mtepst = metepisternum; mtg = mediotergite; oc = ocellus; ped = pedicell; plp = palpal segment; preepst = preepisternum; proepst = proepisternum; sc = scutum; scp = scape; vrt = vertex; wg = wing.

maximum height; clypeus rectangular, about 0.6 times as wide as high; both brown, setose, with setae on clypeus stronger than those on face. Mouthparts light brown. Palpus five segmented, yellowish brown with second segment and apex of fifth segment darker. Ratios of three apical palpal segments 1.0 : 1.37–1.65, 1.52 [1.56] : 1.62–1.82, 1.72 [1.71]. Scape and pedicel brown, pedicel somewhat lighter; flagellomeres light brown, with short pale setae. First flagellomere yellowish at basal half. Flagellum evenly tapering; first flagellomere clavate, 2.5–3.3 times as long as broad apically, 2–13 flagellomeres cylindrical, fourth flagellomere about 2.3–2.6 times as long as broad, apical flagellomere slightly conical, 5.2–6 times as long as broad at base.

Thorax (Fig. 18). All parts brown to dark brown, all setae yellow to light brownish. Mesonotum with evenly arranged numerous setae. Scutellum with about 10 setae along the margin, not arranged to distinct pairs. Antepronotum with 10–13 and proepisternum with 5–8 setae of unequal size, laterotergite with 17–26 setae and mediotergite with ca 12–20 setae medially on lower part. Other pleural parts bare. Halteres yellow, setose.

Legs. All coxae yellow with basal fourths brown. In case of two paratypes, cx_3 entirely light brownish with darker basal half. All trochanters brown. All femora and tibiae yellow, tarsi seem darker because of dense setae. Foretibia with 2–3 ad, 1–3 d and 2–3 pd. Midtibia with 6–10 a, 0–4 d, 4–5 av and 2–3 pd. Hind tibia with 8–10 a, 1–2 ad (1 at apex), 7–8 d, 5–7 p and with a posterior apical comb of setae. Ratio of femur to tibia for fore-, mid- and hind legs: 0.86–0.91, 0.88 [0.91]; 0.72–0.87, 0.80 [0.87]; 0.72–0.77, 0.75 [0.72]. Ratio of tibia to basitarsus for fore-, mid- and hind legs: 0.81–1.00, 0.9 [0.9]; 1.22–1.33, 1.27 [1.22]; [1.66].

Wing hyaline, length 4.1–5.0, 4.52 [4.49] mm (n=4). All veins brown, costal and radial veins somewhat darker. Both surfaces of all veins setose. Wing membrane with micro- and macrotrichia on both surfaces. Costa reaches very little from R_5 to M_1. Sc reaches costa at about quarter between R_s and tip of R_1. R_5 sinuate. R_s about as long as crossvein r-m. M_1 basally obsolete: observable vein begins distally from middle of R_1. Cubital fork begins proximally from apex of Sc.

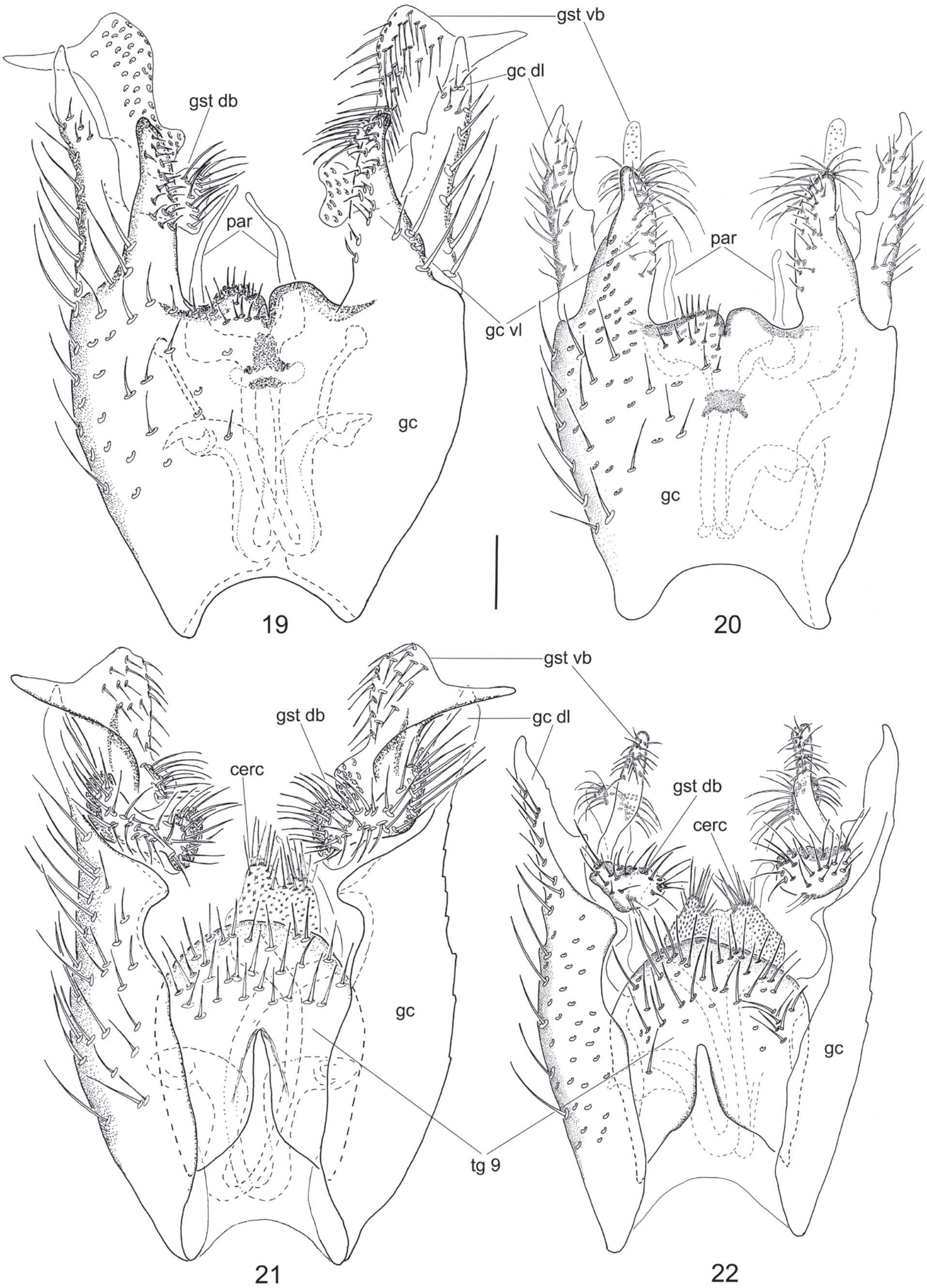
Abdomen with tergites brown to dark brown and with sternites yellow to brownish yellow. Terminalia (Figs 20, 22, 24, 26, 28) dark brown. Tergite 9 apically rounded, with deep and narrow basal incision about half of height of tergite. Basolateral portions of tergite 9 tapering. Setae on tergite 9 similar to these on the gonocoxite. Cerci fused, protruding over tergite 9, with strong apical setae deviating from other setosity. The gonocoxite with elongated dorsoapical and ventroapical lobes. Dorsoapical lobe of the gonocoxite dorsobasally right-angled and apically tapering, both well exposed in lateral view and with subapical medially directed hump. Dorsomedial margin of the gonocoxite slightly undulating. Ventroapical lobe of the gonocoxa apically rounded and subapically somewhat deformed. The gonocoxite ventrobasally with wide shelving incision and ventroapically well sclerotised, with a medial cleft. The gonostylus with two branches. The dorsal branch kidney-shaped, slightly widening towards medial line. The ventral branch elongated, apically evenly rounded, with a clear medial widening which bears strong setae well deviating from other setosity of the branch. The medial widening of the ventral branch of gonostylus connected with apical part of the ventroapical lobe of gonocoxite. Parameres long, sinuous, protruding over ventroapical margin of gonocoxite.

Female. Unknown.

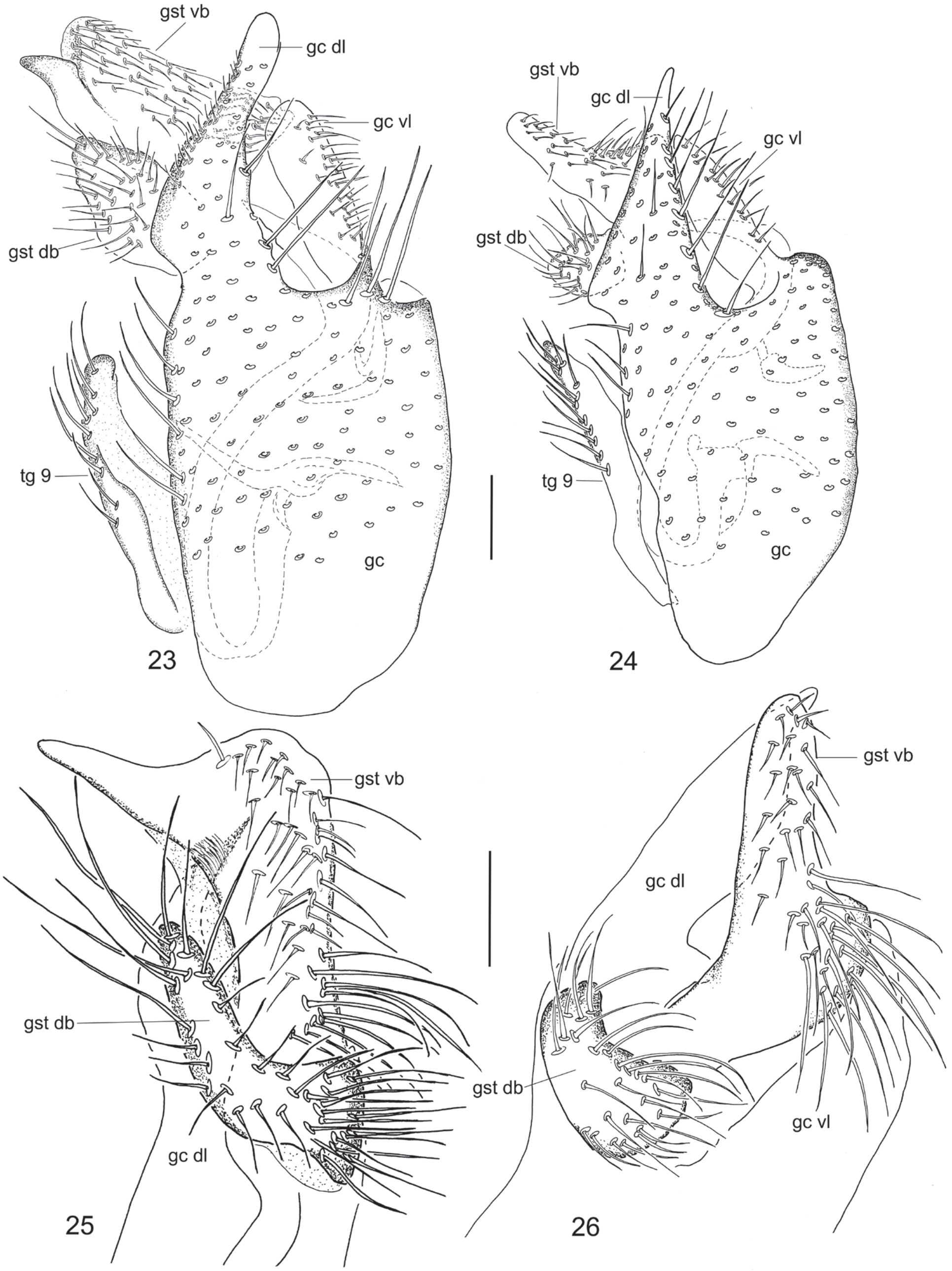
Biology. Unknown.

Etymology. The species is named in honour of Dr. Jukka Salmela, who kindly provided us the material collected from Finland.

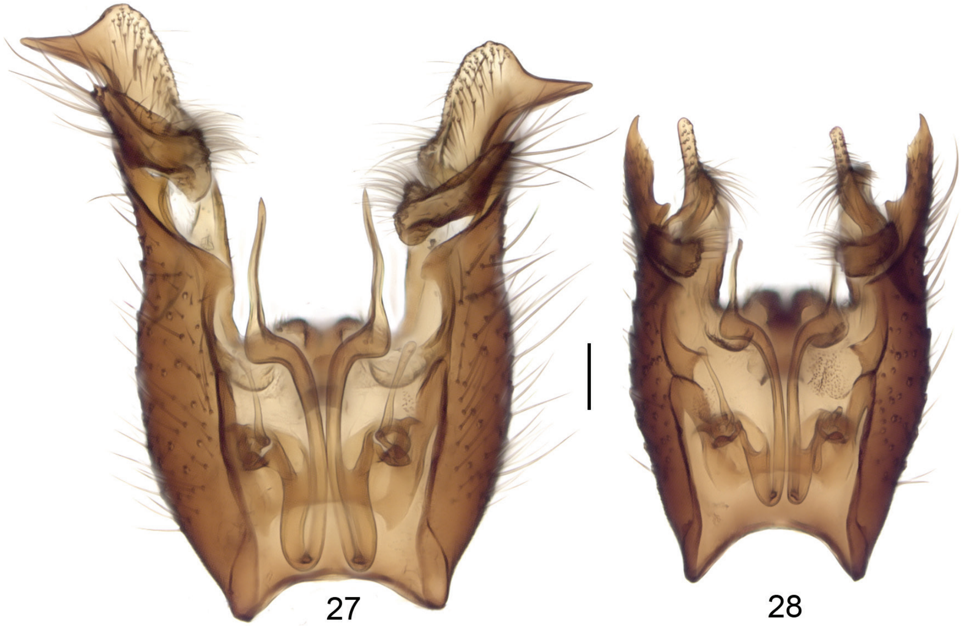
Specific discussion. Following the key by Zaitzev (1994) the new species runs to *N. sintenisi* Lackschewitz, 1937, as its foretibia is usually shorter than fore-basitarsus. However, this character seems to be variable, as these are of equal length in one paratype, guiding to *N. nemoralis*, a species with so far reported Holarctic distribution (Laffoon 1965, Zaitzev 1994, but see specific discussion under the latter). In sharing the general outline of male terminalia, *N. salmelai* resembles in addition to *N. nemoralis* also to *N. kamijoi* Sasakawa, 2004 from Japan. All three species have gonocoxite with protruding lobes dorsoapically and ventroapically, and two-branched gonostylus. *N. kamijoi* has the dorsoapical lobe of gonocoxite with clear subapical tooth (cf. Sasakawa 2004: fig. 5) while in the other two species it is more simple. *Neuratelia salmelai* differs markedly



Figures 19–22. Male terminalia of *N. nemoralis* (Meigen, 1818) (**19**, **21**) and *N. salmelai* sp. n. (**20**, **22**). Ventral view (**19**, **20**) and dorsal view (**21**, **22**). Scale bar = 0.1 mm. For abbreviations: see Figs 7–8, except: gc dl = dorsoapical lobe of gonocoxite; par = parameres.



Figures 23–26. Male terminalia of *N. nemoralis* (Meigen, 1818) (**23**, **25**) and *N. salmelai* sp. n. (**24**, **26**). Lateral view (**23**, **24**) and internal view of the gonostylus (**25**, **26**). Scale bar = 0.1 mm. For abbreviations: see Figs 7–8 and 19–22.



Figures 27–28. Male terminalia, dorsal view, tergite 9 removed. *N. nemoralis* (**27**) and *N. salmelai* sp. n. (**28**). Scale bar = 0.1 mm.

from *N. nemoralis* as follows: 1) dorsoapical lobe of gonocoxite apically tapering (apically evenly rounded in *N. nemoralis*), 2) dorsal branch of gonostylus kidney-shaped and slightly widening towards medial line (elongated, curved and sharply widening towards medial line in *N. nemoralis*), and 3) ventral branch of gonostylus apically evenly rounded with medial widening that bears strong setae deviating well from other setosity of the lobe (ventral branch of gonostylus apically pointed with subapical widening that bears normal setae not deviating from other setosity of the branch in *N. nemoralis*).

Neuratelia nemoralis **(Meigen, 1818)**

Figs 19, 22, 23, 25, 27

Studied material. FINLAND. 1♂, Lkor: Savukoski, Törmäoja, Malaise trap, 67°50'48,5"N, 029°28'20,8"E, 14.vi–10.vii.2012, J. Salmela leg. (MYCE-JS-2013-0095 in ZMUT, in alcohol with terminalia in glycerine); 1♂, Lkor: Sodankylä, Tarmpomapää, Malaise trap, 67°59'14,0"N, 025°55'09,4"E, 1.–29. ii.2009, J. Salmela leg. (MYCE-JS-2013-0235 in ZMUT, in alcohol with terminalia in glycerine); 1♂, Ab: Turku, Pomponrahka, Malaise trap, 2011, J. Salmela leg., (IZBE0200286, in alcohol with terminalia in glycerine). ESTONIA. 2♂♂, Matsalu NP, Matsalu village, window trap, 58°44'04"N, 023°42'42"E, 29.v–17.vi.2009, I.

Süda leg. (IZBE0200275, IZBE0200276, in alcohol); 7♂♂, Palupõhja, Kaha (ME 57), Malaise trap, 58°25'54,68"N, 026°14'28,90"E, 31.v–15.vi.2009, V. Soon leg. (IZBE0200277– IZBE0200282, in alcohol; IZBE0200283, slide mounted in Euparal). SLOVAKIA. 1♂, NP Slovenský raj, Javorina Mt., sweeping, 48°53'23,1"N, 020°15'20,8"E, 1112 m a.s.l., 4.vi.2009, O. Kurina leg (IZBE0200284, in alcohol with terminalia in glycerine; earlier published in Ševčík and Kurina 2011: 101). GREECE. 1♂, Central Macedonia, Kerkini lakes area, Vironia village, Beabies site, 41°19'15,4"N, 023°13'39,6"E, 1150 m a.s.l., Malaise trap, 19.–25.v.2008, G. Ramel leg. (IZBE0200285, in alcohol with terminalia in glycerine).

Specific discussion. This is a widely distributed species in the Palaearctic region (Zaitzev 1994, Sasakawa 2004, Chandler 2013), and as far as we know supposed to extend also to North America (Laffoon 1965, Chandler 2013). The species was first reported from the Nearctic region by Coquillett (1900: 391) and thereafter by Johannsen (1911: 264, Fig. 145) and Fisher (1937: 171, Plate 12: Fig. 12), while all subsequently published information is of secondary nature. However, Fisher (1937: 171) already questioned conspecificity of the Nearctic material and as far as we can judge from the figures of both authors, these represent a different species. Thus, the occurrence of *N. nemoralis* in the Nearctic region remains open with need for the future study.

Discussion

This study combines for the first time the results of morphological and molecular analyses for delineating species of fungus gnats. As a common practice in insect taxonomy, we relied on characters of male genitalia and the mitochondrial COI barcoding, respectively. In one case, however, these two types of data provided conflicting evidence for species delimitation in the fungus gnat genus *Neuratelia*. Therefore, additional characters were sought by sequencing also the 28S and ITS2 regions of the nuclear ribosomal DNA. The latter is becoming increasingly applicable in delimitation of insect taxa (e.g. Rokas et al. 2002, Wilkerson et al. 2004, Haarto and Ståhls 2014). While COI has successfully been used in studies on fungus gnat taxonomy (Martinsson et al. 2011, Rindal et al. 2009b, Ševčík et al. 2013, 2014) and ecology (Põldmaa et al. 2015), ITS2 has been incorporated only in a few studies (Ševčík et al. 2013, 2014).

Taxonomic work on insects has mostly been carried out on the basis of morphological examination. In many cases where studying external characters fail to yield unequivocal results, genital morphology has been proven to be valuable source of additional information (Hosken and Stockley 2004). In more complicated cases, however, even the most detailed morphological study can remain inconclusive. One of the ‘classic’ scenarios where morphological examination may produce questionable results is allopatry. Thus, solving the taxonomic status of morphologically similar allopatric populations has for long time been one of the key questions for systematists. There has been no clear practice how to handle such cases, as acquisition of diagnostic characters does not always happen in the same order or at the same rate for different groups of organisms (e. g. Mutanen et al. 2012). Morphological study of *N. caucasica* and *N. jabalmoussae* presents one more case following the 'classic' scenario: these species have clearly separate geographic ranges located no less than a thousand kilometres away from each other but their morphological differences are minute. However, the genetic distance (quantified according to the Kimura 2-parameter model) calculated from the COI barcoding region is only 1.5% between the two specimens of *N. caucasica*, but ranges from 4.0% to 4.3% between that species and *N. jabalmoussae* (Fig. 4). Many studies have shown that intraspecific genetic distance in the barcode region is several times smaller than the interspecific genetic distance (e. g. Hausmann et al. 2011, Huemer et al. 2014, Pentinsaari et al. 2014). Average intraspecific genetic distance remains under 1% in different insect orders, with only few known exceptions (e. g. Hebert et al. 2010, Park et al. 2011, Pentinsaari et al. 2014). The 4% difference between *N. caucasica* and *N. jabalmoussae* exceeds usual intraspecific genetic variation in insects for more than 4 times, suggesting these taxa truly are different species. This conclusion is further corroborated by few substitutions and small length variation, both in the ITS2 and 28S regions of rDNA distinguishing the two species. The three gene regions thus provide evidence for considering *N. caucasica* and *N. jabalmoussae* to represent distinct species.

The situation with *N. salmelai* and *N. nemoralis* is, however, much more intriguing. Regarding these species, there are five COI barcode haplotypes in our data matrix that have a maximum 1% genetic distance. Specimens of *N. salmelai* and *N. nemoralis* are impossible to distinguish from each other on the basis of barcode data, as the holotype of *N. salmelai* from Estonia carries COI sequence that is identical to that of two specimens of *N. nemoralis* from two different regions of Finland. A Finnish paratype of *N. salmelai*, on the other hand, has COI sequence identical to that of a specimen of *N. nemoralis* from Slovakia. Such a situation has been called 'barcode sharing' in literature (e.g. Hausmann et al. 2011, 2013). Additionally, the relatively rapidly evolving sequences of nuclear rDNA 28S and ITS2, did not allow us to distinguish *N. salmelai* and *N. nemoralis*. 28S was identical in all studied specimens of both species, whereas one individual of *N. nemoralis* from Greece had ITS2 haplotype identical to that of both specimens of *N. salmelai*, which differ from the remaining specimens of *N. nemoralis* by one substitution in ITS2. Thus, delineating *N. salmelai* from *N. nemoralis* on the basis of current molecular data is not possible regardless of whether a distance-based or character-based (DeSalle et al. 2005) approach is selected.

In contrast to the failure of genetic markers to distinguish *N. salmelai* and *N. nemoralis*, their male terminalia were remarkably different. The differences are more pronounced than among the other three species included in this study. Most likely these taxa represent recently diverged species that still share the genetic diversity of their common ancestor. The evolution of insect genitalia can be more rapid than diversification of commonly studied markers (Raupach et al. 2010, Hausmann et al. 2013). Another possibility, hybridisation between females of *N. nemoralis* and males of some other fungus gnat species, deserves less credit for at least two reasons. First, though interspecific hybridisation sometimes occurs in closely related insects, hybrids usually are confined to clear hybrid zones or exist in particular sympatric populations (Mallet et al. 2011, Sánchez-Guillén et al. 2014). Therefore they constitute only a small proportion of the total population. In the current case the five males of *N. salmelai* are 26% of the 19-individual sample of *N. nemoralis* group in our study. This is an unrealistically high proportion for hybrids, as material was randomly collected from different parts of these species' ranges, not concentrating on a particular region where hybridisation could occur. Second, if there really had been hybridisation, it would be natural to assume that putative hybrids (i. e. *N. salmelai*) share genetic material with specimens collected from geographically close localities. This is not the case, as no geographic pattern was detected when collecting localities of *N. salmelai* and *N. nemoralis* were taken into account. Apparently, the *N. salmelai*/*N. nemoralis* species pair is one of the rare occasions where nucleotide data from common markers and morphological characters do not corroborate each other. Large-scale barcoding projects have shown that such cases usually constitute no more than one or two per cents of the total diversity of insects (e. g. Mutanen et al. 2012, Huemer et al. 2014, Pentinsaari et al. 2014).

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New records of Muscidae (Diptera) from Mediterranean countries

Marija Ivković[1], Adrian C. Pont[2]

1 *Department of Zoology, Division of Biology, Faculty of Science, University of Zagreb, Rooseveltov trg 6, 10000 Zagreb, Croatia* **2** *Oxford University Museum of Natural History, Parks Road, Oxford OX1 3PW, UK*

Corresponding author: *Adrian C. Pont* (pont.muscidae@btinternet.com)

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Abstract

New records are provided for Muscidae from four different Mediterranean countries, with new distribution records for species in ten different genera. Seven species are newly recorded for Croatia, four species for Montenegro and one species for Bosnia & Herzegovina. In this paper we give the first confirmation of an aquatic larval stage for *Lispocephala brachialis* (Rondani, 1877), *Lispocephala spuria* (Zetterstedt, 1838) and *Lispocephala mikii* (Strobl, 1893). A first record of the species *Coenosia lyneborgi* Pont, 1972 since its original description is also provided.

Keywords

Limnophora, Croatia, Sierra Nevada, *Coenosia lyneborgi*, Montenegro, Bosnia & Herzegovina

Introduction

Muscid flies are one of the largest groups of Diptera in Europe, with approximately 600 species (Oosterbroek 2006). The family Muscidae comprises seven subfamilies among which the subfamily Coenosiinae, commonly known as "hunter flies", is entirely predaceous in both the larval and adult stages. Larvae of the other subfamilies may be predaceous or saprophagous in decaying organic matter, while most adults feed on nectar. Some adults feed on blood or on the tissues of wounded animals (flies of the subfamilies Muscinae and Azeliinae) and they are facultative vectors of diseases (Oosterbroek 2006, Gregor et al. 2002).

In this paper new records of various genera of Muscidae collected from a number of different localities in the eastern Mediterranean and from localities in the Sierra Nevada, Spain, are offered. The fauna of Muscidae from the Mediterranean countries and especially from the eastern Mediterranean is poorly known and has not been sufficiently explored. Apart from the papers by Coe (1960, 1962a,b, 1968a,b) on the countries of the former Yugoslavia, the only recent paper involving the Balkan Peninsula is that by Pont and Ivković (2013) that deals with the genus *Limnophora* Robineau-Desvoidy from some sites in Croatia. “Rearing” records from emergence traps are exclusively from Croatia, while the net-collected records are from different countries, including Croatia. The majority of records are of “hunter flies” as many of them have aquatic larvae and, as all the collecting took place around streams and river banks, the preponderance of these flies was to be expected.

Material and methods

In the course of various ecological and taxonomic projects and surveys by M.I., many muscid flies were collected by means of emergence traps set in streams and small rivers at five sites in Plitvice Lakes National Park and at two sites at Krka National Park, both in Croatia. 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 (Fig. 1) and for additional details see Ivković et al. (2014). Each trap was recorded with the initial “P” and a number, e.g. “P5" is pyramid emergence trap no. 5. All flies were collected from March 2007 to October 2014. The occasional presence in the emergence traps of a species that does not have aquatic or semi-aquatic larvae is an anomaly that we cannot explain at present. It may be the result of fluctuations in the water level, allowing an adult fly or a drifting puparium to enter the trap. Such anomalies have been discussed by Malicky (2002).

Additional sampling using an aspirator and a sweep net took place between March 2011 and June 2014 at various localities in the eastern Mediterranean part of Europe and from April to June 2013 in the Sierra Nevada, Spain (western Mediterranean). All Muscidae specimens were placed in 80% ethanol and sent to A.C.P. for identification. They were passed through 2-ethoxyethanol (24 hours) and ethyl acetate (24 hours), and then dried, mounted and labelled. Inevitably, many were freshly emerged and not fully hardened, but nevertheless almost every specimen could be identified to species. The monographs by Hennig (1955–1964), Gregor et al. (2002) and Pont and Ivković (2013) were used for identification. All the material listed here is deposited in the Natural History Museum, London, UK (BMNH) and the Oxford University Museum of Natural History, Oxford, UK (OUMNH). GPS coordinates and altitudes for the localities where specimens were trapped and/or collected are given in Table 1.

Figure 1. Spring of Bijela Rijeka, Plitvice Lakes, Croatia, emergence trap.

Results

Faunistic records

The following format is used for the records given here: country, 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), and the number of sampled specimens. All the sites are listed in Table 1.

Subfamily Azeliinae

Genus *Thricops* Rondani, 1856

Remarks. A Holarctic genus. The 26 European species are largely confined to higher altitudes. Twelve species are known from the Balkan Peninsula. Adults are well-known to visit flowers, where they feed on both nectar and pollen (Pont 1993). Larvae are terrestrial and are facultative to obligate carnivores (Skidmore 1985).

Thricops nigrifrons (Robineau-Desvoidy, 1830)

New record. CROATIA: upper reach of Bijela rijeka, Plitvice Lakes, vii.2010, emergence trap P6, 1♀.

Comments. Widespread and common in the West Palaearctic. New for Croatia.

Table 1. The list of sampling sites.

Site name	Longitude	Latitude	Altitude (m)
Río Sucio, Las Barreras (Órgiva), Sierra Nevada, Spain	W 03°26'03"	N 36°54'22"	500
Barranco Frío, Hoya Carlos, Sierra Nevada, Spain	W 03°23'44"	N 36°56'47"	1560
Río Chico, Soportújar, Sierra Nevada, Spain	W 03°24'47"	N 36°55'42"	746
Río Aguas Blancas, Cenes de la Vega, Sierra Nevada, Spain	W 03°30'53"	N 37°09'55"	760
Río Genil, Barranco San Juan, Sierra Nevada, Spain	W 03°23'24"	N 37°08'08"	1200
Río Maitena, Desembocadura, Sierra Nevada, Spain	W 03°24'54"	N 37°09'01"	1018
Djedovica by Rupnica, Papuk Mountain, Croatia	E 17°31'54"	N 45°36'17"	366
Dubočanka stream, Papuk Mountain, Croatia	E 17°40'42"	N 45°29'11"	585
Channel Sava River-Odra River, village Kuče, Croatia	E 16°08'11"	N 45°40'20"	99
*Spring of Bijela rijeka, Plitvice Lakes, Croatia	E 15°33'43"	N 44°50'05"	720
*Upper reach of Bijela rijeka, Plitvice Lakes, Croatia	E 15°33'33"	N 44°50'04"	715
*Upper reach of Crna rijeka, Plitvice Lakes, Croatia	E 15°36'30"	N 44°50'10"	670
*Tufa barrier Labudovac, Plitvice Lakes, Croatia	E 15°35'59"	N 44°52'17"	630
*Tufa barrier Kozjak-Milanovac, Plitvice Lakes, Croatia	E 15°36'32"	N 44°53'39"	545
*Korana village, Plitvice Lakes, Croatia	E 15°37'09"	N 44°55'33"	390
Spring Krčić, Croatia	E 16°19'42"	N 44°01'48"	390
Stream Strmica by Golubić, Croatia	E 16°13'42"	N 44°05'15"	245
Spring of Krka River, Croatia	E 16°14'07"	N 44°02'31"	265
*Roški Slap, Krka River, Croatia	E 15°58'22"	N 43°54'20"	55
*Skradinski Buk, Krka River, Croatia	E 15°57'55"	N 43°48'09"	45
Spring Glavaš, Cetina River, Croatia	E 16°25'48"	N 43°58'36"	385
Jabučica stream, National Park Sutjeska, Bosnia and Herzegovina	E 18°37'02"	N 43°17'24"	767
Spring Bukovica, Durmitor Mountain, Montenegro	E 19°06'42"	N 43°03'30"	1346
Bukovica Stream, Durmitor Mountain, Montenegro	E 19°09'38"	N 43°01'17"	1240
Spring Jeremija, Kolašin, Montenegro	E 19°34'07"	N 42°50'10"	1070
River Murinska rijeka, Montenegro	E 19°53'01"	N 42°39'09"	1000
Alipaša's springs, Montenegro	E 19°49'33"	N 42°33'00"	930

*use of emergence pyramid traps

Subfamily Phaoniinae

Genus *Helina* Robineau-Desvoidy, 1830

Remarks. This a speciose genus, well represented in all biogeographic regions. There are 83 European species, of which18 have been found in the Balkan Peninsula. Adult *Helina* are found in many diverse environments. Larvae are carnivorous and develop mainly in moss or humus soil.

Helina moedlingensis (Schnabl, 1911)

New record. MONTENEGRO: spring Bukovica, Durmitor Mountain, 5.vii.2012, 1♂.

Comments. Widespread in the Palaearctic, but nowhere common. New for Montenegro.

Helina reversio (Harris, 1780)

New record. CROATIA: spring Glavaš, Cetina River, 4.vi.2014, 1♀.

Comments. A very common, widespread and eurytopic species in the Palaearctic. Larvae are terrestrial, found in a wide variety of microhabitats (Skidmore 1985).

Genus *Phaonia* Robineau-Desvoidy, 1830

Remarks. Another speciose genus, present in all biogeographic regions. There are 81 European species of which 14 are known from the Balkan Peninsula. Adults are mostly found on flowers or resting on tree trunks, wooden posts, etc. Larvae are carnivorous and live in soil, in fungi and in decaying wood. Some live in the tunnels of wood boring beetles (Scolytidae) and feed on their larvae.

Phaonia cincta (Zetterstedt, 1846)

New record. CROATIA: tufa barrier Labudovac, Plitvice Lakes, v.2012, emergence trap P6, 1♂.

Comments. Widespread in Europe, but nowhere common. Larvae develop in sap runs in broad-leaved trees where they prey on the larvae of other Diptera. New for Croatia.

Phaonia rufiventris (Scopoli, 1763)

New record. CROATIA: tufa barrier Kozjak-Milanovac, Plitvice Lakes, ix.2008, emergence trap P5, 1♀.

Comments. A common West Palaearctic species. Larvae have been found in decaying wood and fungi. New for Croatia.

Subfamily Mydaeinae

Genus *Hebecnema* Schnabl, 1889

Remarks. This is a small genus of some 35 species. There are six species in Europe, five of which are known from the Balkan Peninsula. Larvae are obligate carnivores and live mostly in dung.

Hebecnema vespertina (Fallén, 1823)

New record. CROATIA: upper reach of Bijela rijeka, Plitvice Lakes, 2.x.2007, emergence trap P6, 1♀.

Comments. A Holarctic species, and common throughout the Palaearctic region. New for Croatia.

Subfamily Coenosiinae

Tribe Limnophorini

Genus *Limnophora* Robineau-Desvoidy, 1830

Remarks. A large genus, found in all biogeographic regions. There are 27 species in Europe of which 13 are known from the Balkan Peninsula. Species of the genus *Limnophora* are usually associated with clean water courses (Rozkošný and Gregor 2004) although Skidmore (1985) writes that *Limnophora riparia* (Fallén) can tolerate high levels of pollution. Both adults and larvae are predaceous (Werner and Pont 2006). The larvae of many species are found among aquatic mosses in streams and rivers (Rozkošný and Gregor 2004). A key to Croatian species, with four new records and one new species, was given by Pont and Ivković (2013).

Limnophora caesia (Villeneuve, 1936)

New record. SPAIN: Río Chico, Soportújar, Sierra Nevada, 17.iv.2013, 1♀.

Comments. Southern Europe, but an uncommon species.

Limnophora croatica Pont & Ivković, 2013

New records. CROATIA: stream Strmica by Golubić, 12.iv.2012, 1♀; spring of Krka River, 7.vii.2011, 1♂ 2♀; Roški Slap, Krka River, 6.vii.2011, 1♀; same site, 30.viii.2011, 2♀; same site, 13.x.2011, 3♂ 2♀; same site, 6.xi.2013, emergence trap P1, 3♂ 1♀; same site and date, emergence trap P3, 1♀; same site and date, emergence trap P4, 1♂ 3♀; same site, 5.iii.2014, emergence trap P3, 1♀; same site and date, emergence trap P4, 1♂ 2♀; same site, 2.iv.2014, emergence trap P1, 1♂ 2♀; same site and date, emergence trap P3, 1♂ 2♀; same site and date, emergence trap P4, 6♂ 6♀; same site, 28.iv.2014, emergence trap P1, 1♀; same site and date, emergence trap P3, 1♂; same site and date, emergence trap P4, 4♂ 3♀; same site, 2.vi.2014, emergence trap P1, 1♂ 5♀; same site and date, emergence trap P3, 1♂; same site and date, emergence trap P4, 2♂ 4♀; same site, 26.vi.2014, emergence trap P1, 1♂ 4♀; same site and date, emergence trap P2, 1♂; same site and date, emergence trap P4, 11♂ 18♀; same site, 26.vii.2014, emergence trap P1, 8♂ 5♀;

same site and date, emergence trap P2, 1♂ 1♀; same site and date, emergence trap P3, 1♂; same site and date, emergence trap P4, 6♂, 10♀; same site, 2.ix.2014, emergence trap P1, 6♂ 4♀; same site, 2.x.2014, emergence trap P1, 1♀; same site, 27.x.2014, emergence trap P1, 1♀; same site and date, emergence trap P3, 1♂; Skradinski Buk, Krka River, 2.vi.2014, emergence trap P4, 1♂ 1♀; same site and date, emergence trap P6, 1♀; same site, 26.vi.2014, emergence trap P1, 3♀; same site and date, emergence trap P4, 4♂ 7♀; same site and date, emergence trap P5, 1♀; same site, 26.vii.2014, emergence trap P3, 1♀; same site and date, emergence trap P4, 3♀; tufa barrier Labudovac, Plitvice Lakes, viii.2011, emergence trap P5, 1♀; spring of Bijela rijeka, Plitvice Lakes, v.2011, emergence trap P2, 1♂; same site, vii.2013, emergence trap P5, 1♂.

Comments. So far this species is known only from Croatia.

Limnophora olympiae Lyneborg, 1965

New records. CROATIA: tufa barrier Kozjak-Milanovac, Plitvice Lakes, ix.2009, emergence trap P4, 1♀. **MONTENEGRO:** Alipaša's springs, 8.vii.2012, 1♂.

Comments. Widespread in the West Palaearctic. New for Montenegro.

Limnophora pandellei Séguy, 1923

New records. SPAIN: Barranco Frío, Hoya Carlos, Sierra Nevada, 1♀; Río Aguas Blancas, Cenes de la Vega, Sierra Nevada, 13.v.2013, 1♀; Río Maitena, Desembocadura, Sierra Nevada, 13.v.2013, 1♂; Río Genil, Barranco San Juan, Sierra Nevada, 29.v.2013, 1♂ 1♀.

Comments. Widespread in the West Palaearctic.

Limnophora pulchriceps (Loew, 1860)

New records. CROATIA: Roški Slap, Krka River, 6.vii.2011, 2♂; same site, 13.x.2011, 1♀; same site, 28.iv.2014, emergence trap P1, 1♀; same site and date, emergence trap P4, 2♂ 1♀; same site, 2.vi.2014, emergence trap P2, 1♀; same site and date, emergence trap P4, 1♂ 4♀; same site, 26.vi.2014, emergence trap P1, 1♂ 1♀; same site and date, emergence trap P4, 3♂ 2♀; same site, 26.vii.2014, emergence trap P1, 3♂ 3♀; same site and date, emergence trap P4, 5♂ 2♀; same site, 2.ix.2014, emergence trap P1, 1♂ 2♀; same site, 2.x.2014, emergence trap P4, 1♀; tufa barrier Labudovac, Plitvice Lakes, v.2012, emergence trap P3, 1♂ 1♀; same site, vii.2012, emergence trap P3, 5♂ 16♀; same site, viii.2012, emergence trap P3, 1♂; same site, vii.2013, emergence trap P7, 1♂ 1♀; same site, viii.2013, emergence trap P7, 1♂ 1♀; tufa barrier Kozjak-Milanovac, Plitvice Lakes, ix.2010, emergence trap P5, 1♀; same site, v.2011, emergence trap P4, 1♂; same site, vii.2011, emergence trap P5, 2♂ 4♀; same site, viii.2011, emergence trap P5, 1♀; same site, ix.2011, emergence trap P3, 1♀; Dubočanka stream, Papuk Mountain, 18.ix.2012, 1♂ 1♀.

Comments. Described from Croatia (Dalmatia) and found in southern Europe and the Middle East.

Limnophora riparia (Fallén, 1824)

New records. CROATIA: Skradinski Buk, Krka River, 6.xi.2013, emergence trap P1, 5♂ 6♀; same site and date, emergence trap P2, 2♂ 1♀; same site and date, emergence trap P3, 3♂; same site and date, emergence trap P5, 1♀; same site and date, emergence trap P6, 1♂ 1♀; same site, 5.iii.2014, emergence trap P1, 2♂ 2♀; same site, 2.iv.2014, emergence trap P1, 2♂ 1♀; same site and date, emergence trap P2, 1♂; same site, 28.iv.2014, emergence trap P1, 1♂ 1♀; same site and date, emergence trap P5, 1♂; same site, 2.vi.2014, emergence trap P1, 1♂ 2♀; same site and date, emergence trap P2, 2♂ 3♀; same site and date, emergence trap P3, 1♀; same site and date, emergence trap P4, 2♀; same site and date, emergence trap P6, 1♀; same site, 26.vi.2014, emergence trap P1, 13♂ 17♀; same site and date, emergence trap P2, 10♂ 4♀; same site and date, emergence trap P4, 9♂ 11♀; same site and date, emergence trap P5, 1♂ 4♀; same site and date, emergence trap P6, 1♂; same site, 26.vii.2014, emergence trap P1, 116♂ 137♀; same site and date, emergence trap P2, 41♂ 95♀; same site and date, emergence trap P3, 12♂ 21♀; same site and date, emergence trap P4, 31♂ 37♀; same site and date, emergence trap P5, 4♂; same site and date, emergence trap P6, 1♀; same site, 2.ix.2014, emergence trap P1, 4♂ 5♀; same site and date, emergence trap P2, 1♂; same site and date, emergence trap P4, 6♂ 10♀; same site and date, emergence trap P5, 1♂ 1♀; same site and date, emergence trap P6, 4♂ 2♀; same site, 2.x.2014, emergence trap P1, 1♀; same site and date, emergence trap P2, 1♀; same site, 27.x.2014, emergence trap P1, 1♀; same site and date, emergence trap P2, 1♂ 1♀; same site and date, emergence trap P4, 1♂, 4♀; Roški Slap, Krka River, 6.vii.2011, 2♂; same site, 6.xi.2013, emergence trap P4, 2♀; same site, 2.iv.2014, emergence trap P4, 1♀; same site, 28.iv.2014, emergence trap P4, 1♀; same site, 26.vi.2014, emergence trap P1, 1♂; tufa barrier Labudovac, Plitvice Lakes, v.2009, emergence trap P2, 1♂; same site, vi.2009, emergence trap P2, 1♂; same site and date, emergence trap P3, 1♂ 1♀; same site, vii.2009, emergence trap P2, 1♂ 2♀; same site and date, emergence trap P3, 1♂ 10♀; same site, viii.2011, emergence trap P2, 1♀; same site and date, emergence trap P5, 2♂ 1♀; same site, ix.2011, emergence trap P1, 1♂; same site, x.2011, emergence trap P2, 1♀; same site, viii.2012, emergence trap P3, 1♀; same site, v.2013, emergence trap P6, 1♂; same site, vi.2013, emergence trap P3, 1♀; same site, vii.2013, emergence trap P2, 1♂; same site, viii.2013, emergence trap P5, 3♀; tufa barrier Kozjak-Milanovac, Plitvice Lakes, vii.2009, emergence trap P5, 1♂; same site, viii.2011, emergence trap P5, 1♀; same site, vi.2012, emergence trap P5, 2♀; same site, vii.2012, emergence trap P5, 1♂ 3♀; same site, ix.2012, emergence trap P5, 1♀; same site, vi.2013, emergence trap P3, 1♂; same site, vi.2013, emergence trap P5, 1♀. **SPAIN:** Río Sucio, Las Barreras (Órgiva), Sierra Nevada, 17.iv.2013, 1♂.

Comments. Widespread and common throughout the Palaearctic region, and closely associated with fast-flowing rivers and streams.

Limnophora setinerva Schnabl, 1911

New records. CROATIA: spring Glavaš, Cetina River, 3.vi.2014, 1♀; Roški Slap, Krka River, 28.iv.2014, emergence trap P2, 1♂; same site, 2.vi.2014, emergence trap P4, 1♂ 1♀; same site, 26.vi.2014, emergence trap P6, 1♂; upper reach of Bijela rijeka, Plitvice Lakes, viii.2010, emergence trap P3, 1♂; tufa barrier Labudovac, Plitvice Lakes, vii.2013, emergence trap P7, 1♀; tufa barrier Kozjak-Milanovac, Plitvice Lakes, ix.2010, emergence trap P5, 2♂; **MONTENEGRO:** River Murinska rijeka, 11.vii.2013, 1♂; Alipaša's springs, 11.vii.2013, 2♀; same site, 8.vii.2012, 2♂ 4♀; spring Jeremija, Kolašin, 6.vii.2012, 1♀; Bukovica stream, Durmitor Mountain, 6.vii.2012, 1♂ 1♀. **BOSNIA & HERZEGOVINA:** Jabučica stream, National Park Sutjeska, 4.vii.2012, 1♂. **SPAIN:** Barranco Frío, Hoya Carlos, Sierra Nevada, 17.iv.2013, 1♂ 1♀; Río Chico, Soportújar, Sierra Nevada, 17.iv.2013, 3♂ 1♀; Río Sucio, Las Barreras (Órgiva), Sierra Nevada, 17.iv.2013, 1♂;Rio Genil, Barranco San Juan, Sierra Nevada, 13.v.2013, 1♂; Río Aguas Blancas, Cenes de la Vega, Sierra Nevada, 2♂; same site, 29.v.2013, 2♂; Río Genil, Barranco San Juan, Sierra Nevada, 29.v.2013, 3♂.

Comments. Widespread in the southern Palaearctic and in the Oriental region. New for Bosnia & Herzegovina and Montenegro.

Limnophora triangula (Fallén, 1825)

New records. CROATIA: tufa barrier Kozjak-Milanovac, Plitvice Lakes, viii.2012, emergence trap P5, 1♀; same site, viii.2012, emergence trap P5, 1♀; Djedovica by Rupnica, Papuk Mountain, 14.vi.2012, 1♀.

Comments. Common throughout the Palaearctic region.

Genus *Lispe* Latreille, 1797

Remarks. *Lispe* is also a large genus, found in all biogeographic regions, with 31 species known from Europe and 14 from the Balkan Peninsula. Adults are predaceous and can be found around standing and running water, where they actively hunt other small invertebrates even in hot, sunny, open habitats (Werner and Pont 2006). Larvae are semi-aquatic and also predaceous, living in organic sand and mud (Skidmore 1985).

Lispe tentaculata (De Geer, 1776)

New records. CROATIA: Korana village, Plitvice Lakes, 29.vi.2007, emergence trap P1, 1♂ 1♀; same site and trap, 26.vii.2007, 6♂ 1♀; same site and trap, viii. 2008, 2♂ 2♀; same site, 26.vii.2007, emergence trap P2, 1♂; same site, 1.ix.2007, emergence trap P5, 1♀; same site, 29.vi.2007, emergence trap P6, 3♂ 3♀; same site and trap, 26.vii.2007, 7♂ 4♀; same site and trap, viii.2008, 1♂.

Comments. The most widespread species of the genus and common throughout the Palaearctic and Nearctic regions. Adults are aggressive predators of Culicidae and Chironomidae.

Genus *Spilogona* Schnabl, 1911

Remarks. *Spilogona* is a genus primarily of high altitudes and high latitudes. Of the 85 European species, only three are known from the Balkan Peninsula. Adults and larvae are predaceous (Werner and Pont 2006). Adults are mostly found in the vicinity of water, whilst the few known larvae are terrestrial and subaquatic.

Spilogona dispar (Fallén, 1823)

New record. MONTENEGRO: spring Bukovica, Durmitor Mountain, 5.vii.2012, 1♂.

Comments. Widespread in the western Palaearctic. New for Montenegro.

Tribe Coenosiini

Genus *Coenosia* Meigen, 1826

Remarks. A speciose genus, found in all regions. Some 80 species are known from Europe, of which 24 are found in the Balkan Peninsula. Species are found in meadows, forests and damp habitats. Both adults and larvae are predaceous. Larvae are terrestrial, living in a wide range of habitats (Skidmore 1985).

Coenosia albicornis Meigen, 1826

New record. CROATIA: tufa barrier Labudovac, Plitvice Lakes, v.2009, emergence trap P5, 1♂.

Comments. Widespread in the western Palaearctic. New for Croatia.

Coenosia lyneborgi Pont, 1972

New record. SPAIN: Río Aguas Blancas, Cenes de la Vega, Sierra Nevada, 13.v.2013, 1♂ 1♀

Comments. This is the first record of the species since its description in 1972, and it is still known only from the Sierra Nevada, Spain. This is a unique species of *Coenosia* as it has only one pair of frontal setae, set high on the frons (see Pont 1972: fig. 1).

Coenosia nigridigita Rondani, 1866

New record. CROATIA: spring Glavaš, Cetina River, 3.vi.2014, 1♀; channel Sava River-Odra River, village Kuče, 17.iv.2011, 1♀.

Comments. A southern European species. New for Croatia.

Coenosia testacea (Robineau-Desvoidy, 1830)

New record. CROATIA: upper reach of Crna rijeka, Plitvice Lakes, viii.2008, emergence trap P4, 1♀.

Comments. Throughout the Palaearctic region.

Coenosia tigrina (Fabricius, 1775)

New records. CROATIA: Korana village, Plitvice Lakes, 29.vi.2007, emergence trap P4, 1♀; same site, 26.vii.2007, emergence trap P2, 1♀; Channel Sava River-Odra River, village Kuče, 17.iv.2011, 2♂.

Comments. A Holarctic species. The larvae live in the soil and are predators of earthworms (Morris and Cloutier 1987).

Genus *Lispocephala* Pokorny, 1893

Remarks. A small genus in Europe with only 12 species, six of which are known from the Balkan Peninsula. Adults are predaceous on other small insects. No larvae of the European species have been described, but it was suspected that they would be aquatic as the adults are usually found in the vicinity of running water. This is confirmed by the records of the three species given here, all of which were caught in emergence traps set in the water.

Lispocephala brachialis (Rondani, 1877)

New records. CROATIA: tufa barrier Kozjak-Milanovac, Plitvice Lakes, vi.2012, emergence trap P5, 1♀; spring Krčić, 23.iv.2011, 1♂.

Comments. Central and southern Europe and North Africa. This is the first confirmation of an aquatic life-cycle for this species.

Lispocephala mikii (Strobl, 1893)

New record. CROATIA: Roški Slap, Krka River, 2.ix.2014, emergence trap P4, 1♂.

Comments. This species was described from Croatia and is a Mediterranean and Afrotropical species. This is the first confirmation of an aquatic life-cycle for this species.

Lispocephala spuria (Zetterstedt, 1838)

New record. CROATIA: spring of Bijela rijeka, Plitvice Lakes, vii.2012, emergence trap P6, 1♀.

Comments. Throughout the Palaearctic region, but an uncommon species. This is the first confirmation of an aquatic life-cycle for this species. New for Croatia.

Genus *Spanochaeta* Stein, 1919

Remarks. Only two species of *Spanochaeta* are known, *S. dorsalis* and an Afrotropical species doubtfully referred to this genus.

Spanochaeta dorsalis (von Roser, 1840)

New record. CROATIA: Roški Slap, Krka River, 6.xi.2013, emergence trap P4, 1♀.

Comments. Nothing is known of the biology of this species but the present rearing indicates that the larvae are aquatic. Throughout Europe, and also in East Africa. New for Croatia.

Discussion

The *Fauna Europaea* site for the family Muscidae (Pont 2005) has not been updated since it first went online, and over the past decade a number of new records have been published, new material has been identified by A.C.P., and records in some of the older publications have been re-assessed. For this reason some of the records presented in this paper are not actually new even though they do not appear on the *Fauna Europaea* site. Moreover, the *Fauna Europaea* website did not separate Serbia and Montenegro (Pape et al. 2015).

Including the new records given here, current totals for the countries of the former Yugoslavia are as follows:

Bosnia-Herzegovina:	45	(11 in *Fauna Europaea*)
Croatia:	91	(79 in *Fauna Europaea*)
Slovenia:	93	(85 in *Fauna Europaea*)
Macedonia:	39	(39 in *Fauna Europaea*)
Serbia:	45	(89 in *Fauna Europaea* for
Montenegro:	17	Serbia and Montenegro combined)

For comparison, 138 species are known from the Greek Mainland and 258 from Spain. It is evident from these figures that much remains to be discovered about the muscid fauna of the Balkan Peninsula, and areas of mountainous and/or temperate broad-leaf forest should prove to be particularly rich in biodiversity.

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