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**Highly disjunct and highly infected millipedes – a new cave-dwelling species of *Chiraziulus* (Diplopoda: Spirostreptida: Cambalidae) from Iran and notes on Laboulbeniales ectoparasites**

Ana Sofia P.S. REBOLEIRA<sup>1</sup>, Mohamad Javad MALEK HOSSEINI<sup>2</sup>, Saber SADEGHI<sup>3</sup> & Henrik ENGHOFF<sup>4</sup>

<sup>1,4</sup>Zoological Museum, Natural History Museum of Denmark, University of Copenhagen, Universitetsparken 15, DK-2100 Copenhagen Ø, Denmark.

<sup>2</sup>Department of Biology, Payame Noor University (PNU), Yasouj, Iran.

<sup>2,3</sup>Department of Biology, College of Sciences, Shiraz University, Iran.

<sup>1</sup> Email: [sreboleira@snm.ku.dk](mailto:sreboleira@snm.ku.dk) (corresponding author)

<sup>1</sup> [urn:lsid:zoobank.org:author:338DE845-4839-4EF5-B684-587C021F076C](http://urn:lsid:zoobank.org:author:338DE845-4839-4EF5-B684-587C021F076C)

<sup>2</sup> [urn:lsid:zoobank.org:author:54DE1791-C6F7-4C81-99DD-736BFC2F7A9F](http://urn:lsid:zoobank.org:author:54DE1791-C6F7-4C81-99DD-736BFC2F7A9F)

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**Abstract.** *Chiraziulus* is a highly disjunct, hitherto monotypic genus of cambalid millipedes, geographically isolated in Iran by more than 7000 km from its presumed closest relatives in East Asia and North America. Recent fieldwork in caves of Iran has provided several specimens of this genus, allowing the description of *Chiraziulus troglopersicus* sp. nov. The intraspecific variability of the type species, *C. kaiseri* Mauriès, 1983, is illustrated with scanning electron micrographs. *Chiraziulus* is characterized by exceedingly long microtrichose gonopod flagella which from their insertion points on the posterior face of the anterior gonopod coxites first point distad instead of basad or basad-posteriad as in most other flagelliferous Cambalidea (and Julida), then traverse a groove on the mesal surface of the anterior gonopod coxites, making a full (360°) loop. The same feature is also illustrated for the first time in the genus *Cambala*. The patterns and prevalence of the infection with a species of ectoparasitic fungus of the genus *Rickia* (order Laboulbeniales) in the type material of *C. kaiseri* is described. An updated review of the cave-adapted fauna of Iran is given.

**Key words.** Cave-adapted, troglobiont, karst, Zagros, Iran, ectoparasitic fungus.

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## Introduction

The millipede order Spirostreptida is mainly distributed in the tropical and temperate zones of the Southern Hemisphere, and in warm-temperate North America. In the Palearctic subregion there are

some marginal occurrences in East Asia, and very few, disjunct occurrences in the western Palaearctic: *Odontostreptus* spp. in Morocco, *Archispirostreptus* spp. in the Middle East, and the so far monotypic genus *Chiraziulus* Mauriès, 1983, in Iran (Shelley & Golovatch 2011: fig. 29). Whereas *Odontostreptus* Attems, 1914 and *Archispirostreptus* Silvestri, 1895 belong to the family Spirostreptidae, suborder Spirostreptidea, *Chiraziulus* is the only W Palaearctic representative of the suborder Cambalidea (Shelley & Golovatch 2011: fig. 30).

*Chiraziulus* and its type species, *C. kaiseri*, were described by Mauriès (1983) who erected it as a subgenus of *Nannolene* Bollman, 1887. Golovatch (1983) tentatively suggested that full generic status might be warranted for *Chiraziulus*, and Mauriès (1987) listed it as a full genus. *Nannolene*, in its current conception (Hoffman 1999), includes numerous nominal species from the western USA and the Hawaiian Islands. Although relationships within Cambalidae are not at all clear, and the delimitation of the family vis-à-vis other cambalidean families is equally uncertain (Enghoff *et al.* in press), the fact remains that *Chiraziulus* is a highly disjunct genus, separated from all other Cambalidea by distances of over 2000 km; if its closest relative is really *Nannolene*, the distance will surpass 7000 km.

Iran is located in a biogeographically interesting transition area, but the cave-adapted fauna of the country is still poorly known (Malek Hosseini *et al.* 2015). The Zagros Mountains are rich in caves, and many remain unexplored. This mountain range extends from Turkey and Iraq to Iran, and is oriented from northwest to southeast, comprising nearly one-fifth of the surface area of Iran. Only eight cave-adapted species (i.e., troglobionts for terrestrials and stygobionts for aquatics) have been recorded from Iran so far: four amphipods, *Niphargus alisadri* Esmaeili-Rineh & Sari, 2013; *N. daniali* Esmaeili-Rineh & Sari, 2013; *N. khayyami* Hekmatara, Zakšek, Heidari Baladehi & Fišer, 2013 and *N. valachicus* Doboreanu & Manolache, 1933; one terrestrial isopod, *Protracheoniscus gakalicus* Kashani, Malek Hosseini & Sadeghi, 2013; one species of goblin spider, Oonopidae, *Trilacuna qarzi* Malek Hosseini & Grismado, 2015; and two cave fishes, the barb *Gara typhlops* Bruun & Kaiser, 1944 and the loach *Paracobitis smithi* Smith, 1976 (Esmaeili-Rineh & Sari 2013; Hekmatara *et al.* 2013; Kashani *et al.* 2013; Malek Hosseini *et al.* 2015).

We revise the genus *Chiraziulus*, describe a new species from a cave in Iran and document intraspecific variation of *C. kaiseri*. New observations on the flagellum in a species of the North American genus *Cambala* Gray, 1832, are also included. Re-examination of the type material of *C. kaiseri* led to the discovery of the presence of ectoparasitic fungi of the order Laboulbeniales; we also include information about this infection.

## Material and methods

New specimens of *Chiraziulus* (Fig. 1) were collected in two caves in Iran: Ghar Sarab Cave (*Ghar* is Persian for “cave”), located at 87 km northwest of Hamadan city, near Sarab village, altitude: 2200 m asl, coordinates 35°14'41.7" N, 48°15'40.6" E, length: 2959.9 m and Neyneh Cave, located at 45 km southwest of Dehdasht city, Kohgiluyeh and Boyerahmad Province, coordinates 30°40'18" N, 50°21'71" E, 770 m asl (Fig. 2).

Specimens were studied using a Leica Wild M10 microscope equipped with an ocular micrometer. Parts of specimens were mounted in glycerine on temporary slides for microscope study. Microphotographs were taken using a Leica digital camera M205A mounted on a stereo microscope Leica DFC 420. Images were processed with a Leica Application Suite program, and final stacking was made using ZereneStacker software. For scanning electron microscopy, parts of specimens were critical point dried in a Tousimis Autosamdi 815, series A. Other specimens were transferred to 96% ethanol, then to acetone, air-dried, mounted on aluminium stubs, coated with platinum/palladium and studied in a JEOL JSM-6335F scanning electron microscope. Images were processed with Adobe Photoshop CS6.

Material is deposited in the Zoological Museum, Natural History Museum of Denmark, University of Copenhagen (ZMUC), the Collection of the Biology Department, Zoological Museum of Shiraz University, Shiraz, Iran (ZM CBSU) and the Natural History Museum of Vienna (NHMW).

## Results

### *Systematics*

Class Diplopoda Blainville-Gervais, 1844

Order Spirostreptida Brandt, 1833

Suborder Cambalidea Cook, 1895

Family Cambalidae Bollman, 1893

Genus *Chiraziulus* Mauriès, 1983

*Nannolene* (*Chiraziulus*) Mauriès, 1983: 250

*Chiraziulus* (as full genus) – Mauriès 1987: 197.

### Type species

*Nannolene* (*Chiraziulus*) *kaiseri* Mauriès, 1983.



**Fig. 1.** Habitus of *Chiraziulus troglopersicus* sp. nov., paratype.



## Diagnosis

A genus of medium-sized blind cambalid millipedes resembling the Nearctic genus *Nannolene*, with longitudinal crests on the body rings and ozopores placed on prominent tubercles (Fig. 1). Sharing with *Nannolene* remarkably long gonopodal flagella, which make a complete loop (360°) from their insertion points on the anterior gonopods towards the interior part of the posterior gonopods, differing from *Nannolene* by having the coxites of the anterior gonopods larger than the telopodites. First pair of male legs not modified.

## Distribution

Iran (Fig. 2).



**Fig. 2.** Distribution of species of *Chiraziulus* Mauriès, 1983 in Iran (Mauriès 1987). *C. troglopersicus* sp. nov.: red dot (1); *C. kaiseri*: yellow dots (2–6). 1. Neyneh Cave. 2. “Chiraz” “montagne greuse au nord de la ville”. 3. 19 km W of Shiraz. 4. 5 km N of Persepolis. 5. Oasis 95 km N of Bandarabass. 6. Sarab Cave.



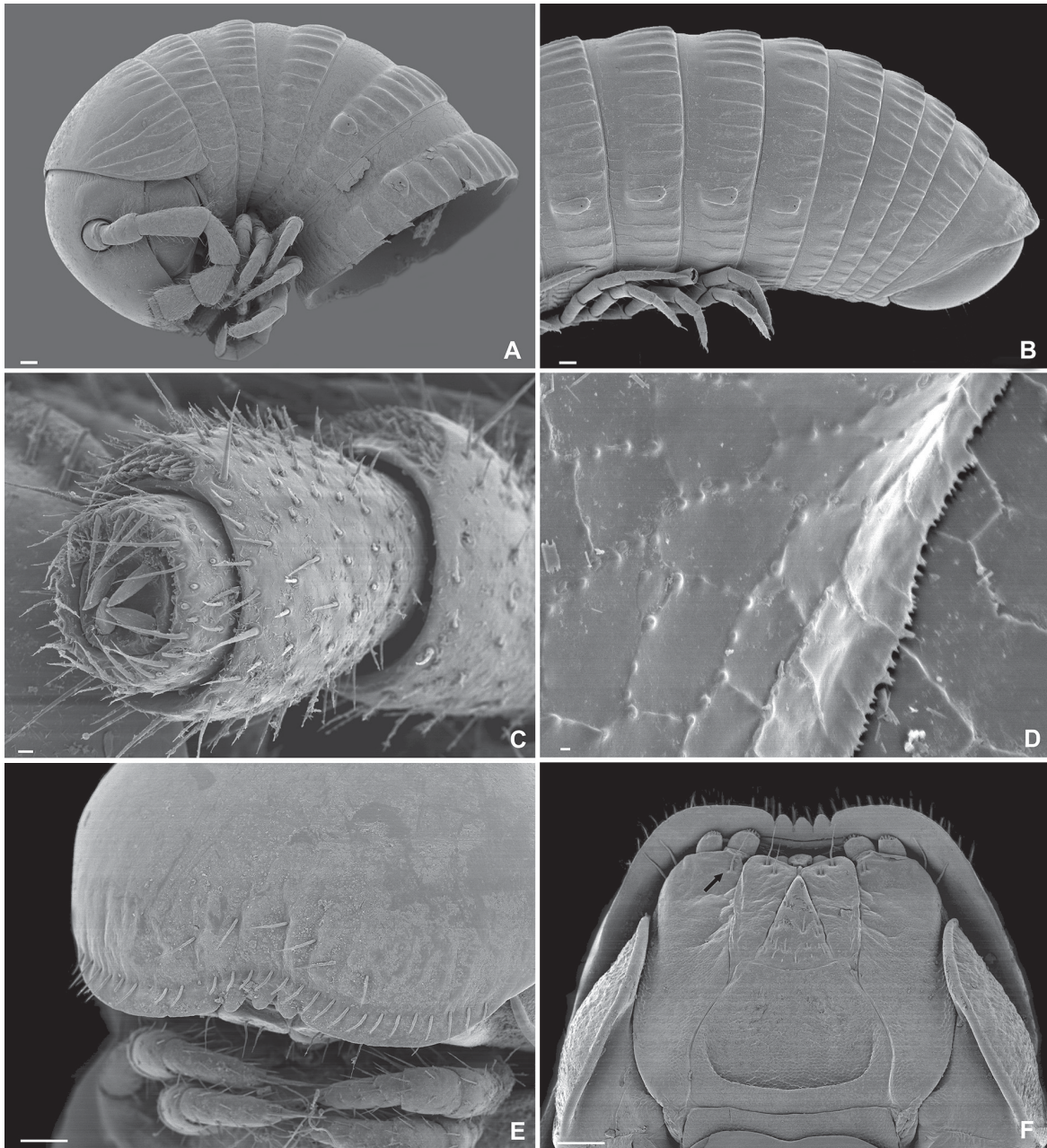
*Chiraziulus kaiseri* (Mauriès, 1983)

Figs 3–8, 11–12

*Nannolene* (*Chiraziulus*) *kaiseri* Mauriès, 1983: 252, figs 4–10.

*Nannolene* (*Chiraziulus*) *kaiseri* – Golovatch 1983: 166.

*Chiraziulus kaiseri* – Enghoff & Moravvej 2005: 63.



**Fig. 3.** *Chiraziulus kaiseri* (Mauriès, 1983), paratypes. Scanning electron micrographs. **A.** Head and first body rings in lateral view. **B.** Last body rings ("tail") in lateral view. **C.** Tip of antenna. **D.** Detail of the limbus; notice lines of beadlike structures between cuticular scutes. **E.** Detail of labrum in frontal view. **F.** Gnathochilarium in ventral view; the arrow shows the distomesal setae on stipes. Scale bars: A–B, E–F = 100 µm; C = 10 µm; D = 1 µm.

## Diagnosis

A species of *Chiraziulus*, differing from *C. troglopersicus* sp. nov. by having triangular anterior gonopods in anterior view, the posterior branch of the telepodite smaller than the anterior branch, and the posterior gonopod with a simple, sharp, spinelike process.

## Type material examined

IRAN: Holotype (♂) and numerous paratypes, Shiraz, sandstone mountains N of the town, 1600 m asl. Very dry, scattered vegetation including the first spring flowers, under stones and clods, 16 Feb. 1937, E.W. Kaiser leg. (ZMUC).

## Additional material examined

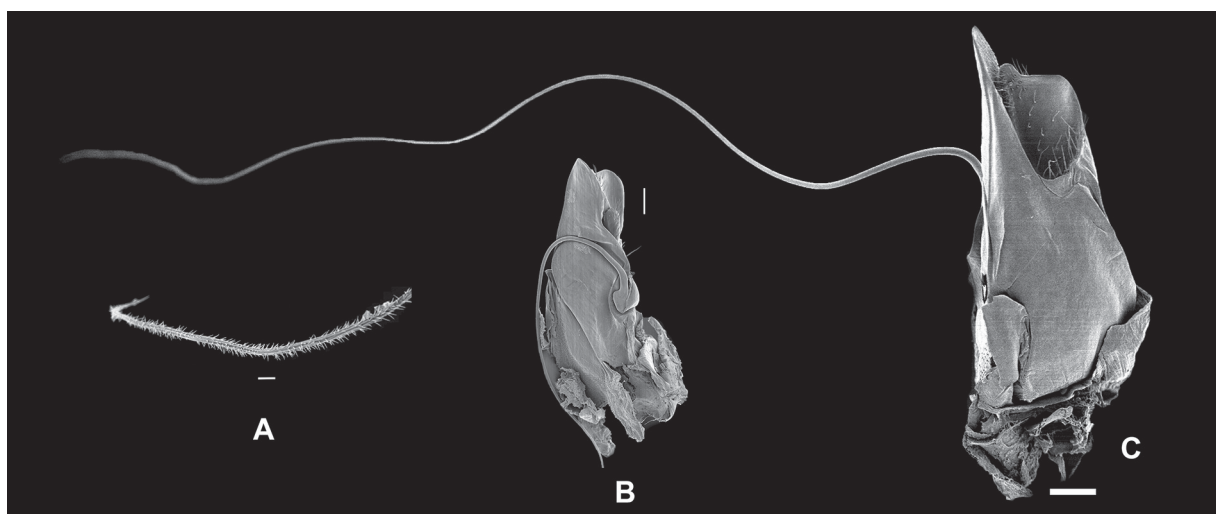
IRAN: 1 ♂, Sarab Cave, 22 Jul. 2002, Alexandri leg. (ZMUC); 2 ♂♂, 2 ♀♀, 19 Str. km W of Shiraz, G. Pretzmann leg. (NHMW).

## Description

Eyes absent, body depigmented, with prominent tergal crests. Ozopores on protruding tubercles, starting on 5<sup>th</sup> body ring (Fig. 3A, B). Antenna with four long apical cones and bacilliform sensilla seated in small depressions on external part of the 5<sup>th</sup> and 6<sup>th</sup> antennomeres (Fig. 3C). Labrum and supralabral chaetotaxy as in Fig. 3E. Gnathochilarial stipes with three setae, two distolateral and one distomesal seta, the latter close to promentum. Lingual lamella with two setae. Promentum moderately setose (Fig. 3F). Limbus with numerous tiny denticles (Fig. 3D). Borders between cuticular scutes on part of body with lines of beadlike structures (Fig. 3D), similar to those observed by Akkari & Enghoff (2011) and Enghoff (2014) in other millipedes.

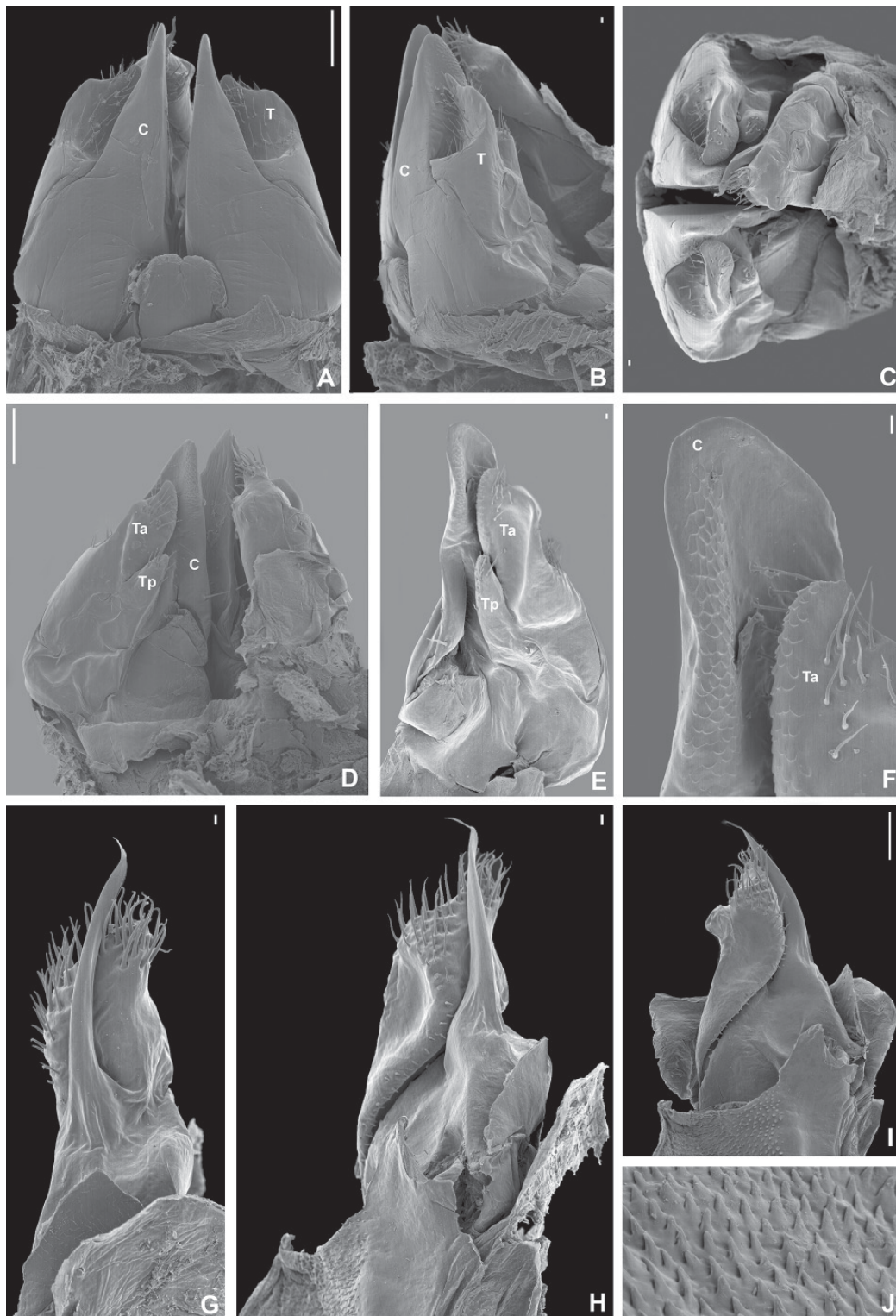
## Male

Anterior gonopods (Figs 4, 5A–F) with tongue-shaped, parallel-sided mid-sternal lobe. Each coxite approximately like an equilateral triangle folded at right angles along vertical axis; resulting surfaces facing anterior and mesad; coxite hence narrow triangular in anterior view, ending in rather sharp point (Fig. 5A); coxite in mesal view with a plane surface traversed by oblique groove for accommodation of flagellum (Fig. 4); posterior margin sinuous, in apical view *c.* 30% of it with rugose-scaly microsculpture



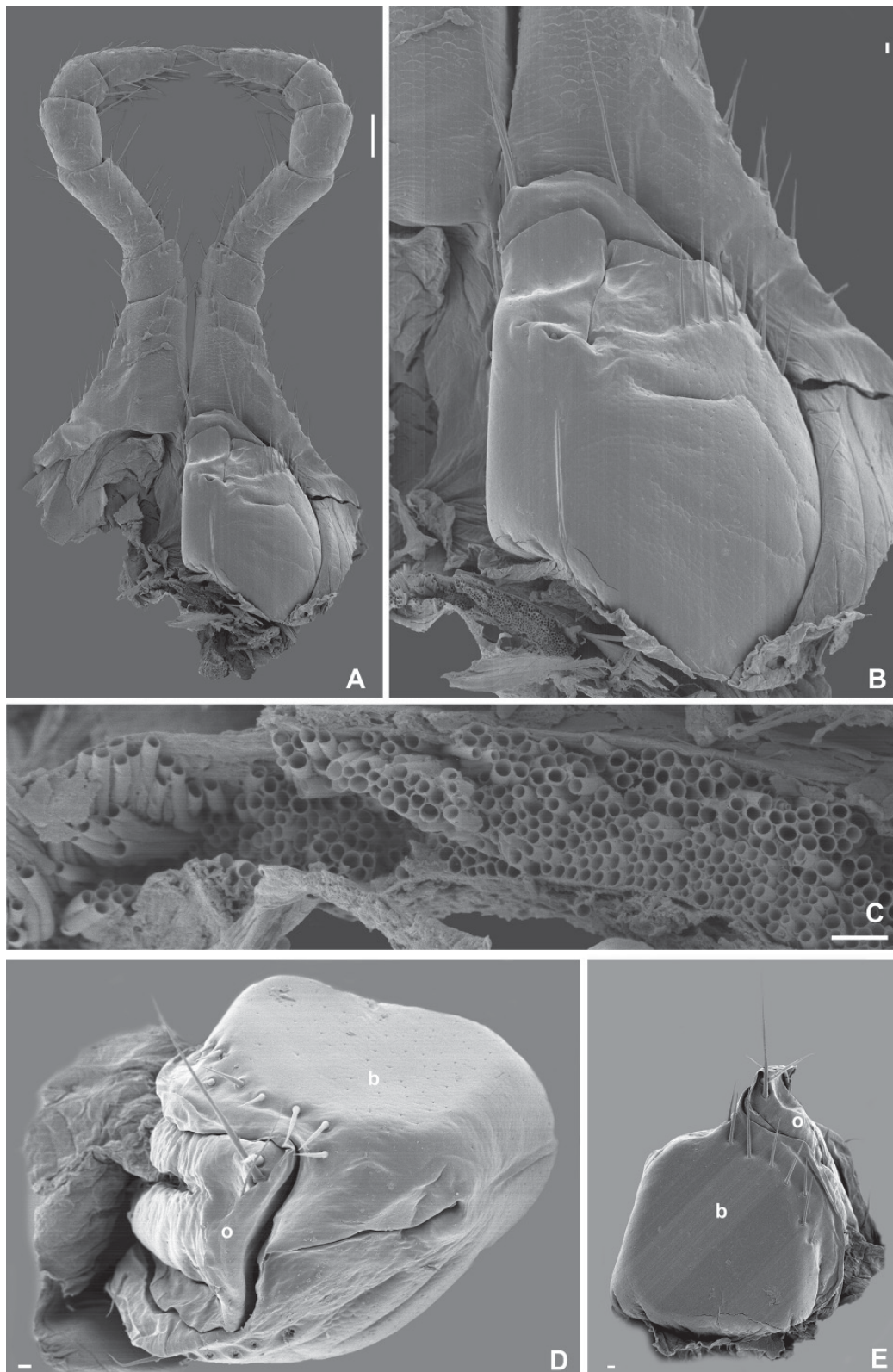
**Fig. 4.** *Chiraziulus kaiseri* (Mauriès, 1983), ♂, paratype, anterior gonopod. Scanning electron micrographs. **A.** Tip of flagellum. **B.** Flagellum insertion and loop across anterior gonopod coxite. **C.** Anterior gonopod showing the entire extension of the flagellum. Scale bars: A = 10 µm; B–C = 100 µm.





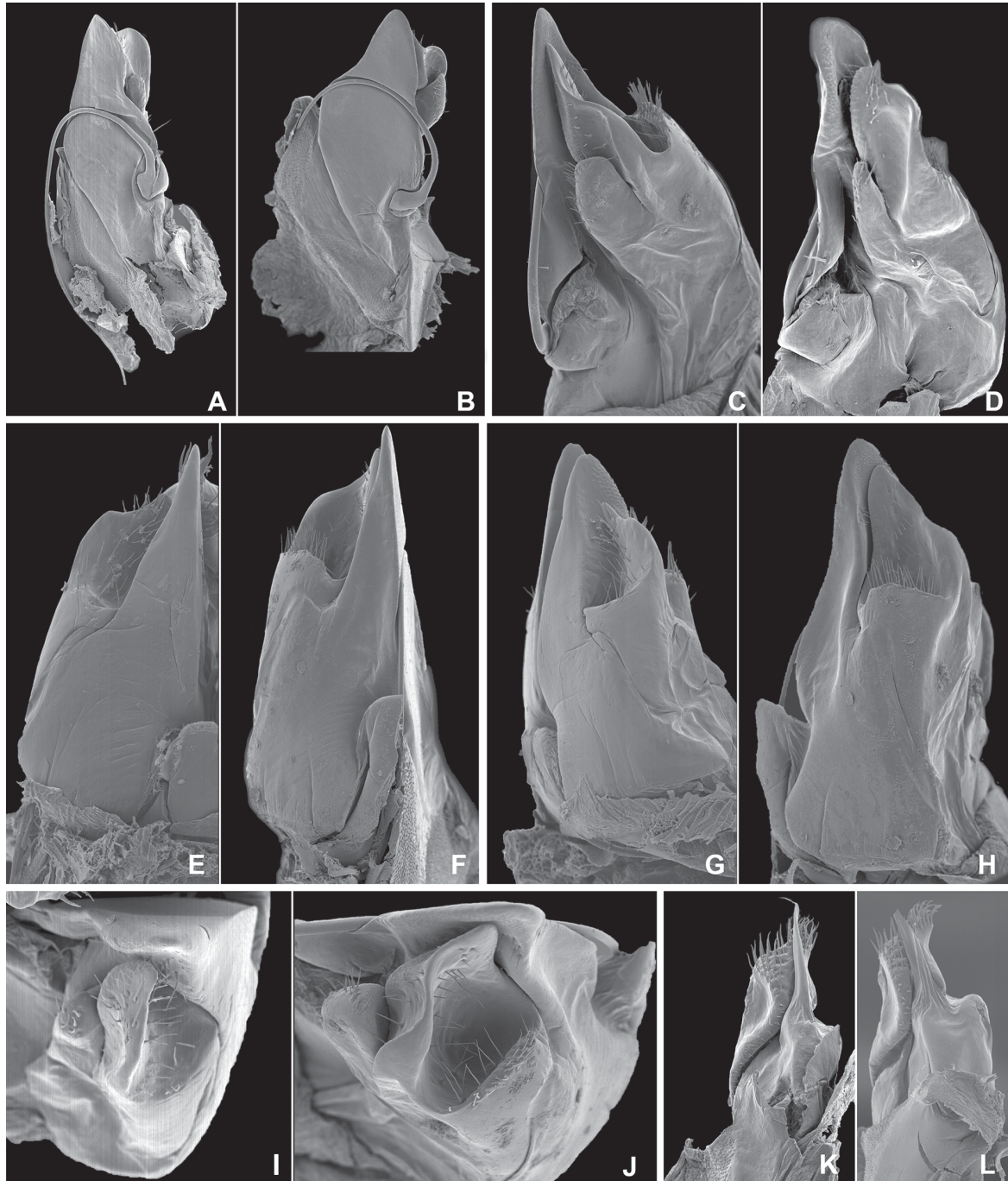
**Fig. 5.** *Chiraziulus kaiseri* (Mauriès, 1983), ♂, paratypes, gonopods. Scanning electron micrographs. **A.** Anterior gonopods in anterior view. **B.** Anterior gonopods with one posterior gonopod in lateral view. **C.** As B, in apical view. **D.** As B, in posterior view. **E–F.** Anterior gonopod with detail of the processes on the tip of anterior gonopods. **G–I.** Posterior gonopod. **J.** Detail of the mesal sternal part. Abbreviations: C = coxal process; T = telopodite; Ta = anterior lobe of telopodite; Tb = posterior lobe of telopodite. Scale bars: A, D, I = 100 µm; B–C, E–H = 10 µm.





**Fig. 6.** *Chiraziulus kaiseri* (Mauriès, 1983), female vulva. Scanning electron micrographs. **A.** Vulva *in situ* behind second pair of legs, posterior view. **B.** Details of right vulva behind second pair of legs. **C.** Detail of microtubular structure under the vulva. **D.** Vulva in ventral view. **E.** Vulva in anterior view. Abbreviations: o = operculum; b = bursa. Scale bars: A = 100  $\mu\text{m}$ ; B–E = 10  $\mu\text{m}$ .

on lateral surface. Telopodite large, two-lobed; anterior lobe (regarded as part of the coxite (e) by Mauriès) larger (*Ta*), but not reaching tip of coxite (Fig. 5D–E), apical margin concave (Fig. 5F); in anterior view *Ta* broadly visible lateral to coxite, with transverse “step”, distal to ‘step’ with several setae; in posterior view *Ta* longitudinally concave, apically with several setae and with mesal margin (facing rugosity of coxite) rugose; posterior lobe of telopodite (*Tp*) (T = telopodite of Mauriès) much shorter than *Ta*, rod-



**Fig. 7.** *Chiraziulus kaiseri* (Mauriès, 1983). Comparison of the paratype (A, C, E, G, I and K) and specimen from Ghar Sarab Cave (B, D, F, H, J and L). **A–B.** Left anterior gonopods, mesal view. **C–D.** Anterior gonopod, posterior view. **E–F.** Anterior gonopod, anterior view. **G–H.** Anterior gonopod, lateral view. **I–J.** Anterior gonopod, apical view. **K–L.** Posterior gonopods.



shaped, apically broadly rounded, with several setae (Fig. 5D). Rugose tips of coxite and telepodite (*Ta*) of anterior gonopod forming a pair of “forceps” (Fig. 5F). Flagellum exceedingly long, inserted below mid-height of posterior margin of coxite, initially directed distad, then turning anteriad and traversing mesal surface of coxite in slightly curved groove, then turning basad-posteriad, probably accommodated in basal oblique groove (Fig. 4B–C); apical part of flagellum densely covered with *c.* 5  $\mu\text{m}$  long hair-like, retrorse processes (Fig. 4A). Posterior gonopods (Fig. 5G–I) apically covered with setae and with a long slender, spine-like, apically twisted process. Mesal sternal part of gonopods forming trough densely covered with sharktooth-like denticles (Fig. 5J).

### Female

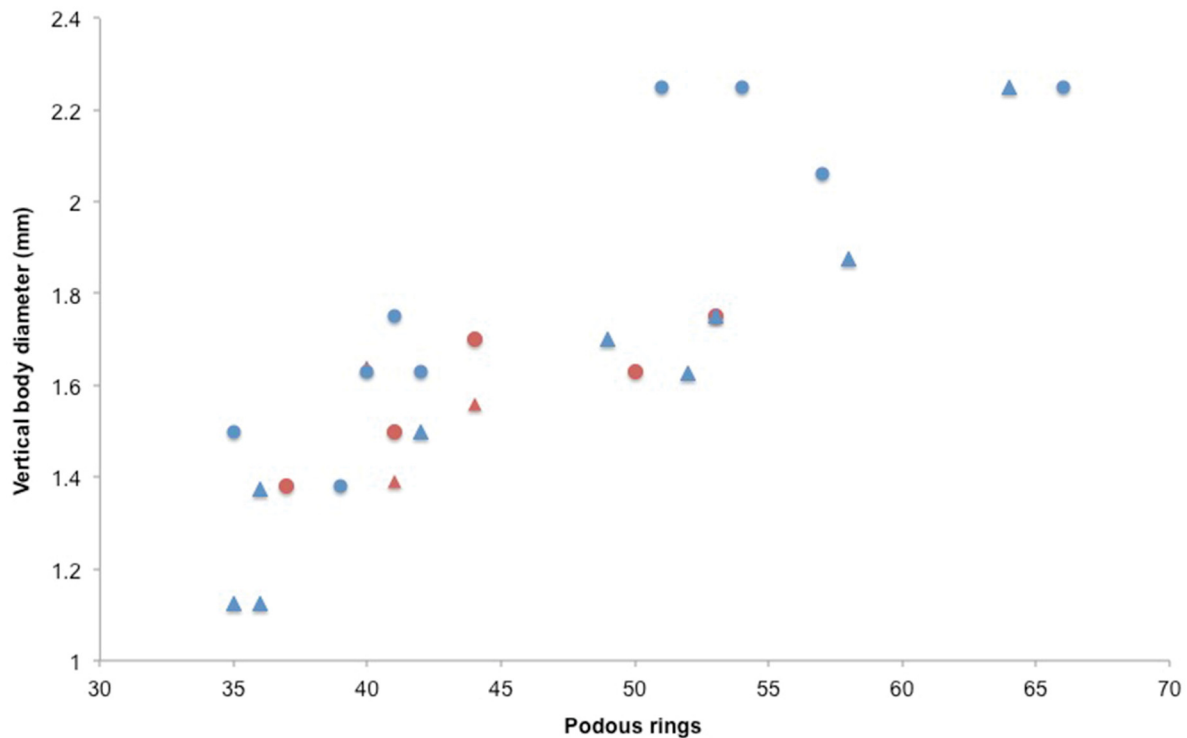
Vulva *in situ* as in Fig. 6A–B. Peculiar, microtubular tissue present below insertion level of bursa (Fig. 6C). Operculum (o) smaller than bursa, with 2 large apical setae; bursa massive, enveloping operculum, with simple slit-shaped crest and 8 + 8 setae along distal margins (Fig. 6D, E).

### Distribution

Known from several localities in Iran (Fig. 2).

### Intraspecific variability of *Chiraziulus kaiseri*

In addition to the type locality and a few sites in its vicinity, *Chiraziulus kaiseri* has been recorded from the south of Iran (Golovatch 1983). Examination of an adult male from an isolated cave (Sarab Cave) in northwestern Iran, far from the type locality, leads us to include it in *C. kaiseri* as well and to regard the differences in the male gonopods as intraspecific variability (Fig. 7).



**Fig. 8.** Number of podous rings and midbody vertical diameter of the studied specimens. *Chiraziulus kaiseri* (Mauriès, 1983) in blue; *Chiraziulus troglopersicus* sp. nov. in red. ▲ = adult ♂♂; ● = juveniles and ♀♀.



The anterior gonopods of the specimen from Sarab Cave shows some differences compared to the type material, especially in the shape of the posterior branch of the telepodite (Fig. 7E–J). No remarkable difference was observed in the posterior gonopods (Fig. 7K–L). Some variability was also observed in the chaetotaxy of the gnathochilarial stipes (Fig. 3F). In the specimens from the type locality, Sarab Cave and 19 km W of Shiraz, there are two distolateral and one distomesal setae, whereas in the specimen illustrated by Golovatch (1983: 168, fig. 12), all three setae are distolateral.

*Chiraziulus troglopersicus* sp. nov.

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Figs 1, 9–10

### Diagnosis

A species of *Chiraziulus*, differing from *C. kaiseri* by anterior gonopods not presenting a triangular shape in anterior view, by having equally sized anterior and posterior lobes of anterior gonopod telepodites, and by posterior gonopod having a spine-like process ending in a halberd-like structure.

### Etymology

From Greek *trōglo*: cave, common prefix for terrestrial cave-dwelling animals, and Latin *persicus*: Iranian.

### Type material

#### Holotype

IRAN: ♂, Kohgiluyeh and Boyerahmad Province, Khaeiz protected area, Mt. Khaeiz, 45 km southwest of Dehdasht city, Neyneh Cave, 30°40'18" N, 50°21'71" E, altitude 770 m (gonopods coated for SEM), 17 May 2012 (ZMUC).

#### Paratypes

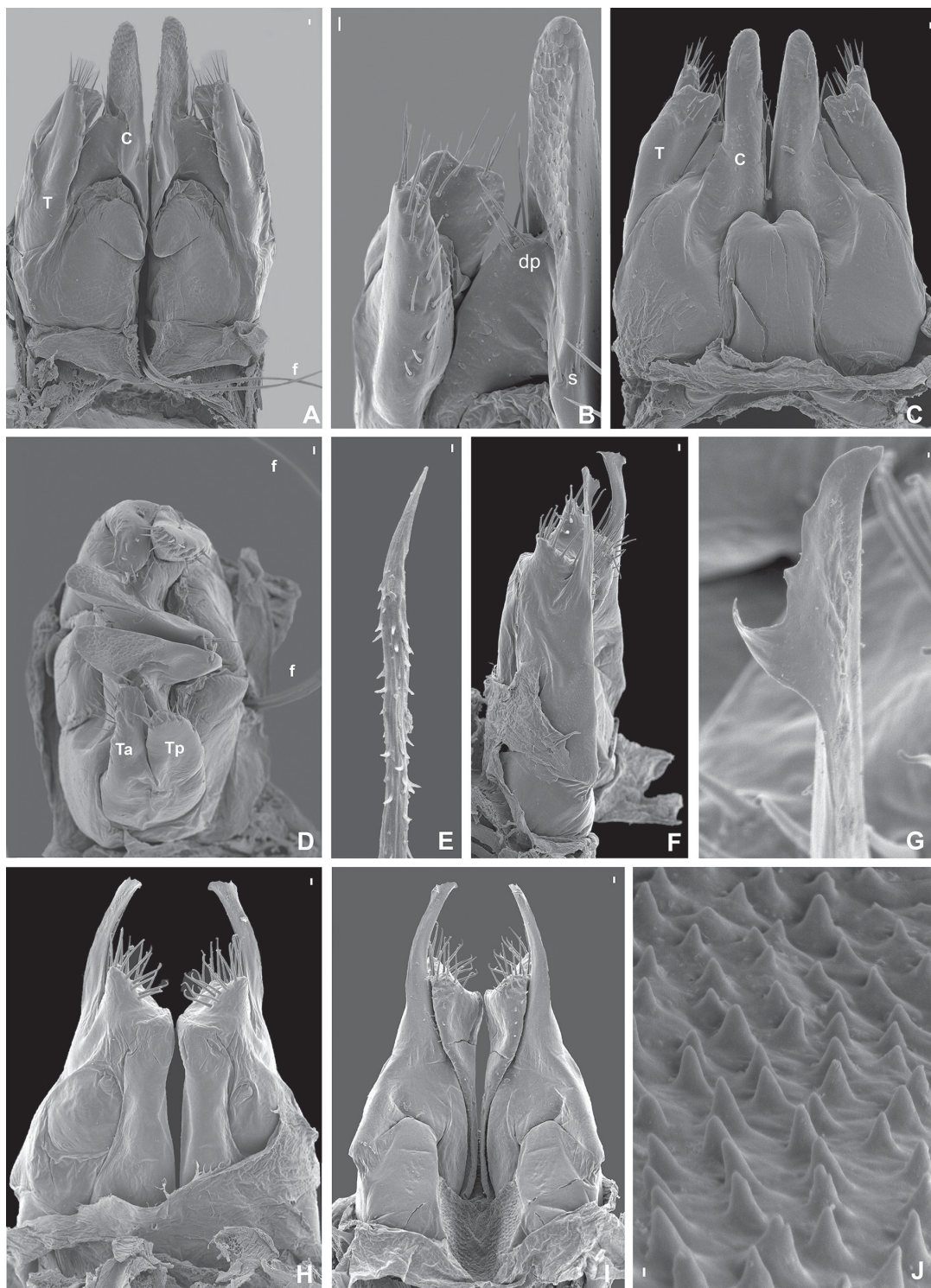
IRAN: same data as the holotype: 4 ♀♀ (ZMUC); 1 ♀ and 3 ♂♂ (ZM CBSU).

### Description

Males: 41–44 podous + 2 apodous rings, diameter 1.39–1.64 mm. Females: 39–51 podous + 2 apodous rings, and 53 + 0 podous rings, diameter 1.38–1.75 mm. Habitus typical of the genus, eyes absent. Six supralabral setae. Ozopore starting on 5<sup>th</sup> body ring (except for one female paratype that presents teratological traits and in which ozopores start on 6<sup>th</sup> body ring).

#### Male (Fig. 9)

Anterior gonopods (Fig. 9A–D) with remarkably long flagellum (Fig. 9A, D–E), making a full loop from its insertion point on posterior face of anterior gonopod coxites pointing distad. Anterior gonopods with blade-shaped and apically rounded coxite (not of a triangular shape in anterior view as in *C. kaiseri*) (Fig. 9A–D); each coxite with 2 setae in mesal anterior view (*s*) (Fig. 9B) and a lateral ridge with 4 setae at mid-height (*dp*); external lateral margin slightly concave, with rugose-scaly microsculpture (Fig. 9B). Telepodite shorter than coxite, bi-lobed (Figs. 9A–D). Anterior and posterior lobes of telepodite (*Ta* and *Tp*) about the same size; anterior lobe with 8 setae, slightly concave on side facing posterior lobe; posterior lobe with 8 parallel setae. Flagellum (*f*) extremely long, covered with *c.* 5 μm long hair-like, retrorse processes and a pointed tip (Fig. 9E). Posterior gonopods (Fig. 9F–J) apically covered with setae and with a long slender spine-like process ending in a halberd-like expansion. Mesal sternal part of gonopods forming a trough covered with sharktooth-like denticles (Fig. 9I–J).



**Fig. 9.** *Chiraziulus troglopersicus* sp. nov., ♂, paratype, gonopods. Scanning electron micrographs. **A.** Anterior gonopods in posterior view. **B.** Detail of the process on the tip of an anterior gonopod in posterior view. **C.** Anterior gonopods in anterior view. **D.** As C, in apical view. **E.** Tip of the flagellum. **F.** Posterior gonopods in lateral view. **G.** Tip of the long spine-like process of the posterior gonopod. **H.** Posterior gonopods in posterior view. **I.** As H, in anterior view. **J.** Detail of the mesal sternal part of the gonopods. Abbreviations: C = coxal process; T = telepodite; f = flagellum; s = setae; dp = distal process. Scale bars: A–D, F, H–I = 10 µm; E, G, J = 1 µm.



### Habitat

The new species was discovered in the Neyneh Cave (Fig. 10), located in the mountainous Khaeiz protected area, in Zagros Mountains, southwestern Iran. Neyneh Cave is situated in the warmer part of the province where the temperature at the surface reaches to 50°C during the summer. The temperature inside the cave is 25°C; the relative humidity is very high, 95–99.9%, and the CO<sub>2</sub> level is 613–700 ppm. There was no water flow in the cave. This cave is located in cliffs and has more than 700 m of horizontal maze development. It is located in an area with difficult access, thus protected from human pressure; however, there is some destruction due to the activities of treasure hunters. *Chiraziulus troglopersicus* sp. nov. specimens were found in complete darkness, crawling on guano and cave walls. Some colonies of bats produce enough guano to cover the cave soil, providing an important source of organic matter for this subterranean trophic chain. Species richness in Neyneh Cave is relatively high and includes several arthropod taxa (Acari, Araneae, Oniscidea, Collembola, Orthoptera, Coleoptera), as well as some vertebrate taxa, such as gekkos, bats and hystricid rodents.

### Notes on the gonopods of *Cambala annulata* (Say, 1821)

SEM study of one male of *Cambala annulata* (USA, NC, Surry Co., Elkin, 12 October 1975, R.M. Shelley leg.) revealed that the anterior gonopod in this species, like that of *Chiraziulus* (and *Nannolene*), has a remarkably long flagellum (Fig. 11A) with a similar insertion, and with a similar full-loop course in part involving a groove on the mesal side of the anterior gonopod coxite. The sharktooth-like structure of the mesal gonopodal sternal part is also present (Fig. 11B).

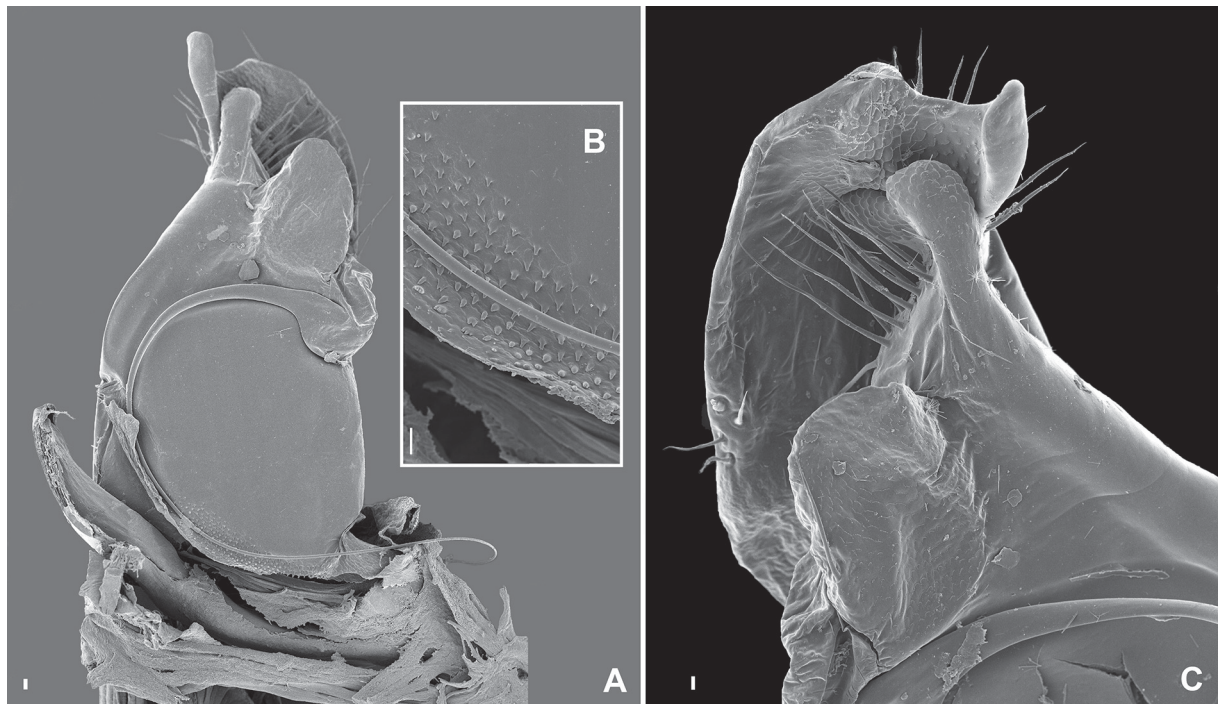
### Occurrence of ectoparasitic fungi on *Chiraziulus kaiseri*

Studies of the obligate ectoparasitic fungi of the order Laboulbeniales associated with millipedes are scarce (e.g., De Kesel *et al.* 2013; Santamaria *et al.* 2014; Enghoff & Santamaria 2015; Reboleira &

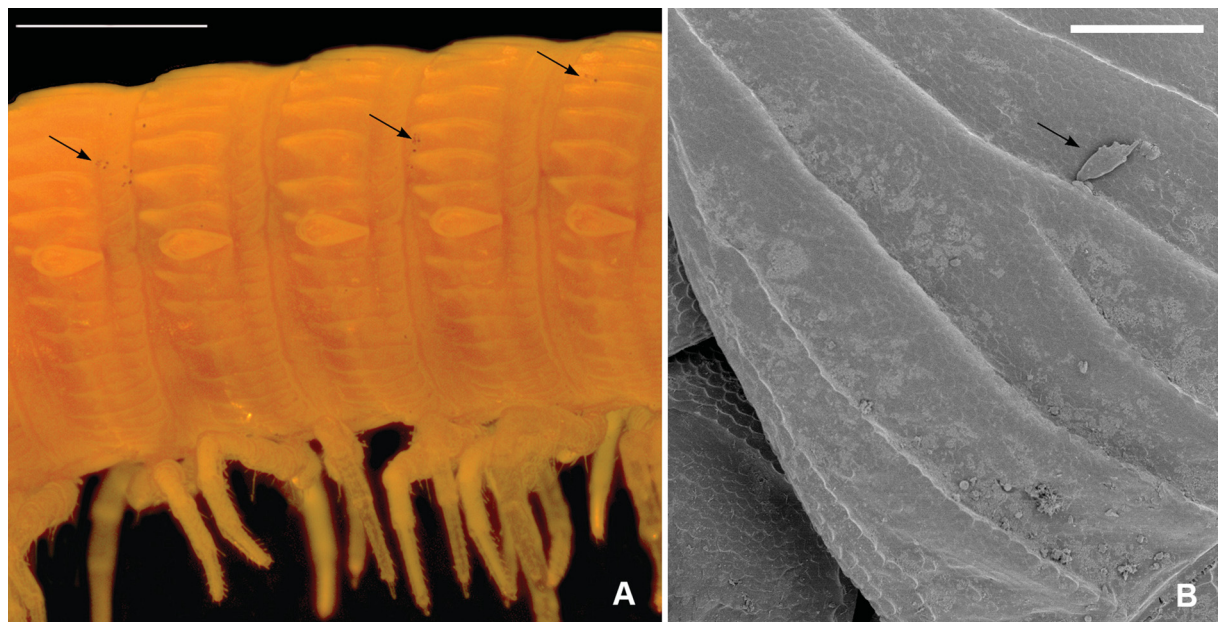


**Fig. 10.** *Chiraziulus troglopersicus* sp. nov., habitat; Neyneh Cave in Zagros Mountains, Iran.





**Fig. 11.** *Cambala annulata* (Say, 1821), anterior gonopod. **A.** Mesal view. **B.** Detail of the mesal sternal part. **C.** Apical part of coxal processes and telepodite. Scale bars: A = 100  $\mu\text{m}$ ; B–C = 10  $\mu\text{m}$ .



**Fig. 12.** *Chiraziulus kaiseri* (Mauriès, 1983), paratypes infected with ectoparasitic fungi of the genus *Rickia* Cavara, order Laboulbeniales. **A.** Arrows indicate black dots on the body rings corresponding to the insertion of the fungi. **B.** Scanning electron micrograph of the collum with one fungus (arrow). Scale bars: A = 1 mm; B = 100  $\mu\text{m}$ .

**Table 1.** Numbers of *Chiraziulus kaiseri* (Mauriès, 1983) specimens infected with the ectoparasitic fungus *Rickia* sp.

	Infected		Non-infected	
	Male	Female	Male	Female
Type locality	30	31	2	2
19 km W of Shiraz	2	2	–	–
Sarab Cave	1	–	–	–

Enghoff 2015). These fungi live only on the surface of living animals, and during the revision of the type material of *Chiraziulus kaiseri*, deposited in ZMUC (Fig. 12), almost all type specimens were found to be infected with an undescribed species of the genus *Rickia*, which in spite of its small size can be detected due to the dark coloration of the foot and appendages (Fig. 12A).

Some Laboulbeniales have a preference for growing on specific parts of the millipede body, and the transmission mechanisms play an important role for the position of the fungus on the host (Enghoff & Santamaria 2015; Reboleira & Enghoff 2015). In the case of *Rickia* sp. on *C. kaiseri*, the fungus is randomly distributed along the millipede (Fig. 12): it was found on body rings, legs, head, antennae and surprisingly also on the paraprocts (anal valves), both in adults and juveniles, suggesting that transmission does not exclusively take place during copulation as occurs in other species (Santamaria *et al.* 2014).

Of the 70 specimens studied, 66 were infected, which means that the infection rate may be around 94% (Table 1). This is significantly higher than the 64% infection rate recorded for populations in caves of the callipodidan *Lusitanipus alternans* (Verhoeff, 1893), infected with *Diplopodomycetes lusitanipodos* Santam., Enghoff & Reboleira in central Portugal, and than the 63% recorded for the blaniulid *Archiboreoiulus pallidus* (Brade-Birks, 1920) infected with *Troglomyces triandrus* Santam. & Enghoff, but still lower than the 100% infections reported for *Troglomyces pusillus* Santam. & Enghoff on the blaniulid *Iberoiulus cavernicola* Ceuca, 1967 (Enghoff & Santamaria 2015; Reboleira & Enghoff 2015). The four specimens from 19 km W of Shiraz were also all infected.

More remarkably, the specimen of *Chiraziulus kaiseri* from Sarab Cave, despite being part of a disjunct population in the north of Iran, was also infected with the same species of *Rickia*.

## Discussion

*Chiraziulus kaiseri* presents a considerable degree of intraspecific variation even within the type material. The new population found isolated in a cave in the northwest of Iran is also parasitized by the same *Rickia* (morpho)species as the type material. These fungi are normally highly host specific (De Kesel 1996), and the presence of the same ectoparasitic species in disjunct populations is herewith recorded for the second time for millipedes, as it was recently reported for *Diplopodomycetes lusitanipodos* Santam., Enghoff & Reboleira and *Troglomyces manfrediae* Colla (Santamaria *et al.* 2014).

The highly relict character of the genus *Chiraziulus* had already been pointed out by Golovatch (1983), due to its remarkable disjunct distribution relative to other Cambalidae, which are distributed mainly in North and Central America, Hawaii and Australia. The closest relative of *Chiraziulus* may be *Nannolene* (including *Hawaicambala* Mauriès, 1983), as suggested by the original classification of *C. kaiseri* in *Nannolene* (Mauriès 1983). A synapomorphy could be the exceedingly long gonopodal flagellum which from its insertion point on the posterior face of the anterior gonopod coxites points distad instead of basad or basad-posteriad as in most other flagelliferous Cambalidea (and Julida), and then makes a full

(360°) loop. Illustrations of the flagellar loop in some Hawaiian and Californian species of *Nannolene* are provided by Silvestri (1904: figs 28–29) and Chamberlin (1922: pl. 1, fig. 6), respectively. However, not all *Nannolene* species have such extraordinary flagella (see, e.g., Silvestri 1904: fig. 19); also a specimen of *Nannolene keiferi* Chamberlin, 1943, has flagella of normal length which do not make the 360° loop. On the other hand, we could show that this peculiar type of flagellum is also present in the genus *Cambala*. Clearly, the genus-level taxonomy and phylogeny of Cambalidae needs a closer study.

Mauriès (1983) mentioned the following characters in support of his new subgenus *Chiraziulus*:

1. Anterior gonopod coxites with much longer parasagittal lobes than in *Nannolene* s.s.
2. First pair of male legs unmodified (although there is some uncertainty about the first pair of legs in *Nannolene* s.s.)
3. Lack of eyes
4. Presence of longitudinal crests and grooves on the metaterga
5. The geographical isolation

We can support the first and fourth of these characters, although we have re-interpreted the “parties coxales parasagittales” as belonging to the telopodites. The second, third and fifth characters are less convincing: the male of *N. keiferi* that we have examined has unmodified first legs, several (sub)genera of, e.g., Julidae and Blaniulidae include eyed as well as blind species, and finally, geographical isolation *per se* should not be used as an argument for creating a new taxon, as this could easily lead to circular argumentation. For the time being, we maintain *Chiraziulus* as a full genus, following Mauriès (1987).

The microtrichose flagellum in both species of *Chiraziulus* is probably used for sperm competition, cf. Barnett & Telford (1996), Enghoff (1985, 1992, 1995) and Frederiksen & Enghoff (2015).

The anterior gonopod coxite and telopodite in *Chiraziulus* seem to form a “forceps” with the opposing scaly-rugose edges. This is probably an analogue of the “forceps” formed by the promerite (= anterior gonopod) and mesomerite (part of posterior gonopod) in several Julidae, which has been shown to grip the operculum of the vulva during copulation (Haacker & Fuchs 1970; Tadler 1996; see also Read 1990). Mauriès interpreted both parts of the forceps as belonging to the coxite, but the SEM images clearly show that it is composed of apical parts of the coxite and the anterior branch of the telopodite.

We here add a cave-adapted millipede to the short list of Iranian cave fauna by describing a new species of *Chiraziulus*, raising to ten the number of cave-adapted species known for the country.

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## References

Akkari N. & Enghoff H. 2011. On some surface structures of potential taxonomic importance in families of the suborders Polydesmidea and Dalodesmidea (Polydesmida, Diplopoda). *Zookeys* 156: 1–24. <http://dx.doi.org/10.3897/zookeys.156.2134>



- Barnett M. & Telford S.R. 1996. Sperm competition and the evolution of millipede genitalia. *In*: Geoffroy J.-J., Mauries J.-P. & Nguyen Duy-Jacquemin M. (eds) *Acta Myriapodologica*: 331–339. Mémoires du Muséum national d'Histoire naturelle 169, Muséum national d'Histoire naturelle, Paris.
- Chamberlin R.V. 1922. Notes on West Indian millipedes. *Proceeding of the United States National Museum* 61 (10): 1–19.
- De Kesel A. 1996. Host specificity and habitat preference of *Laboulbenia slackensis*. *Mycologia* 88 (4): 565–573. <http://dx.doi.org/10.2307/3761150>
- De Kesel A., Haelewaters D. & Gerstmans C. 2013. Two interesting species of *Rickia* (Laboulbeniales) from coastal habitats in Belgium and the Netherlands. *Sterbeeckia* 32: 6–10.
- Enghoff H. 1985. The millipede family Nemasomatidae. With the description of a new genus, and a revision of *Orinisobates* (Diplopoda: Julida). *Entomologica Scandinavica* 16 (1): 27–67. <http://dx.doi.org/10.1163/187631285X00045>
- Enghoff H. 1992. *Dolichoiulus* – a mostly Macaronesian multitude of millipedes. With the description of a related new genus from Tenerife, Canary Islands (Diplopoda, Julida, Julidae). *Entomologica Scandinavica, Supplement* 40: 1–158.
- Enghoff H. 1995. A revision of the Paectophyllini and Calyptophyllini: millipedes of the Middle East (Diplopoda, Julida, Julidae). *Journal of Natural History* 29: 685–786. <http://dx.doi.org/10.1080/00222939500770241>
- Enghoff H. 2014. A mountain of millipedes I: An endemic species-group of the genus *Chaleponcus* Attems, 1914, from the Udzungwa Mountains, Tanzania (Diplopoda, Spirostreptida, Odontopygidae). *European Journal of Taxonomy* 100: 1–75. <http://dx.doi.org/10.5852/ejt.2014.100>
- Enghoff H. & Moravvej S.A. 2005. A review of the millipede fauna of Iran (Diplopoda). *Zoology in the Middle East* 35: 61–72. <http://dx.doi.org/10.1080/09397140.2005.10638104>
- Enghoff H. & Santamaria S. 2015. Infectious intimacy and contaminated caves – three new species of ectoparasitic fungi (Ascomycota: Laboulbeniales) from blaniulid millipedes (Diplopoda: Julida) and inferences about their transmittal mechanisms. *Organisms, Diversity and Evolution* 15 (2): 249–263. <http://dx.doi.org/10.1007/s13127-015-0208-8>
- Enghoff H., Golovatch S., Short M., Stoev P. & Wesener T. *In press*. Diplopoda – Taxonomic overview. *In*: Minelli A. (ed) *The Myriapoda 2. Treatise on Zoology – Anatomy, Taxonomy, Biology*. Brill, Leiden.
- Esmaeili-Rineh S. & Sari A. 2013. Two new species of *Niphargus* Schiödte, 1849 (Crustacea: Amphipoda: Niphargidae) from two caves in Iran. *Journal of Natural History* 47 (41–42): 2649–2669. <http://dx.doi.org/10.1080/00222933.2013.802041>
- Frederiksen S.B. & Enghoff H. 2015. East African odontopygid millipedes 4: A restricted redefinition of the genus *Rhamphidarpoidea* Kraus, 1960, a related new genus, five new species, and notes on solenomere function (Diplopoda; Spirostreptida; Odontopygidae). *Zootaxa* 3926 (1): 541–60. <http://dx.doi.org/10.11646/zootaxa.3926.4.5>
- Golovatch S.I. 1983. A contribution to the millipede fauna of Iran (Diplopoda). *Annalen des naturhistorischen Museums Wien* 85/B: 157–169.
- Haacker U. & Fuchs S. 1970. Das Paarungsverhalten von *Cylindroiulus punctatus* Leach. *Zeitschrift für Tierpsychologie* 27 (6): 641–648. <http://dx.doi.org/10.1111/j.1439-0310.1970.tb01892.x>
- Hekmatara M., Zakžek V., Heidari B.M. & Fišer C. 2013. Two new species of *Niphargus* (Crustacea: Amphipoda) from Iran. *Journal of Natural History* 47 (21–22): 1421–1449. <http://dx.doi.org/10.1080/00222933.2012.743616>

- Hoffman R.L. 1999. *Checklist of Millipeds of North and Middle America*. Special Publication 9, Virginia Museum of Natural History, Martinsville.
- Kashani G.M., Malek Hosseini M.J. & Sadeghi S. 2013. First recorded cave-dwelling terrestrial isopods (Isopoda: Oniscidea) in Iran with a description of a new species. *Zootaxa* 3734 (5): 591–596. <http://dx.doi.org/10.11646/zootaxa.3734.5.8>
- Malek Hosseini M.J., Grismado C., Sadeghi S. & Bakhshi Y. 2015. Description of the first cave dwelling species of the spider genus *Trilacuna* Tong & Li from Iran (Araneae: Oonopidae). *Zootaxa* 3972 (4): 549–561. <http://dx.doi.org/10.11646/zootaxa.3972.4.6>
- Mauriès J.-P. 1983. Cambalides nouveaux et peut connus d'Asie, d'Amérique et d'Océanie. I. Cambalidae et Cambalopsidae (Myriapoda, Diplopoda). *Bulletin du Muséum national d'Histoire naturelle Paris, section A, 4<sup>e</sup> série* 5 (1): 247–276.
- Mauriès J.-P. 1987. Cambalides nouveaux et peut connus d'Asie, d'Amérique et d'Océanie. II. Pseudonannolenidae, Choctellidae (Myriapoda, Diplopoda). *Bulletin du Muséum national d'Histoire naturelle Paris, section A, 4<sup>e</sup> série* 9 (1): 169–199.
- Read H.J. 1990. The generic composition and relationships of the Cyndroiulini – a cladistic analysis (Diplopoda, Julida: Julidae). *Entomologica Scandinavica* 21: 97–112. <http://dx.doi.org/10.1163/187631290X00085>
- Reboleira A.S.P.S. & Enghoff H. 2015. Redescription of *Lusitanipus alternans* (Verhoeff, 1893) (Diplopoda, Callipoda, Dorypetalidae) and ecological data on its Laboulbeniales ectoparasites in caves. *Zootaxa* 3957 (5): 567–576.
- Santamaria S., Enghoff H. & Reboleira A.S.P.S. 2014. Laboulbeniales on millipedes: the genera *Diplopodomycetes* and *Troglomyces*. *Mycologia* 106 (5): 1027–1038. <http://dx.doi.org/10.3852/13-381>
- Shelley R.M. & Golovatch S.I. 2011. Atlas of myriapod biogeography. I. Indigenous ordinal and supra-ordinal distributions in the Diplopoda: perspective on taxon origins and ages, and a hypothesis on the origin and early evolution of the class. *Insecta Mundi* 158: 1–134.
- Silvestri F. 1904. Myriopoda. In: Sharp D. (ed.) *Fauna Hawaiiensis*. Vol. 3: 323–338. The University Press, Cambridge. <http://dx.doi.org/10.5962/bhl.title.4628>
- Tadler A. 1996. Functional morphology of genitalia of four species of julidan millipedes (Diplopoda: Nemasomatidae; Julidae). *Zoological Journal of the Linnean Society* 118: 83–97. <http://dx.doi.org/10.1111/j.1096-3642.1996.tb01263.x>

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