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A new troglobiotic species of *Hyalella* (Crustacea, Amphipoda, Hyalellidae) with a taxonomic key for the Brazilian species

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Abstract

The freshwater crustaceans from the order Amphipoda occur mainly in cold and temperate climates. However, in the tropics, these animals can be more abundant in subterranean environments, where the temperatures are milder than in surface. Despite being accepted that the number of species of freshwater amphipods in South America is lower when compared to other regions, recent descriptions have shown that its diversity is certainly underestimated. In this study, a new species of the genus *Hyalella* is described for Brazil, the fourth troglobiotic species of *Hyalella* for the country and the sixth in the world. The new species was found on the epikarst of a cave in São Paulo state, Southeastern Brazil. Besides, the new species shows typical characteristics from organisms adapted to the subterranean environments, a pattern also observed in the other troglobiotic species of the genus.

Key words: epikarst, freshwater crustacean, morphological adaptations, troglomorphism

Resumo

Os crustáceos de água doce da ordem Amphipoda ocorrem principalmente em climas frios e temperados. Contudo, nos trópicos, estes animais podem ser mais abundantes em ambientes subterrâneos, onde as temperaturas são mais amenas do que na superfície. Apesar de ser aceito que o número de espécies de anfípodas de água doce na América do Sul seja baixo quando comparado com outras regiões, descrições recentes mostram que sua diversidade é certamente subestimada. Neste estudo, uma nova espécie do gênero *Hyalella* é descrita para o Brasil, a quarta espécie troglóbia de *Hyalella* no país e a sexta no mundo. A nova espécie foi encontrada no epicarste de uma caverna do estado de São Paulo, sudeste do Brasil. Além disso, a nova espécie apresenta características típicas de organismos adaptados aos ambientes subterrâneos, um padrão também observado nas demais espécies troglóbias do gênero.

Palavras-chave: epicarste, crustáceos de água doce, adaptações morfológicas, troglomorfismo

Introduction

The freshwater species from the order Amphipoda inhabit a wide variety of habitats. They are mainly abundant in cold and temperate climates while exhibiting a low diversity in the tropics. In warmer climates, most species occur in subterranean environments where the temperatures are milder than on surface (Barnard & Barnard 1982; Väinölä *et al.* 2008). Less than 10% of the freshwater species described for the order occurs in the Neotropical region, and the diversity in South America comprises only 10 families, 22 genera and 74 species, although this diversity is certainly underestimated (Fišer *et al.* 2013).

Among the freshwater families of the Neotropics, Hyalellidae have the highest number of species and they are the dominant epigean organisms, and are also endemic to the New World (Bousfield 1996). This family is only composed of the genus *Hyalella* Smith, 1874, and currently there are 61 species described and Brazil has the greatest diversity, with 18 taxa (Bueno *et al.* 2013; Lowry & Myers 2013).

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The diversity of the genus has been increased in recent years by the descriptions of new epigean and hypogean species (Cardoso *et al.* 2011; Bastos–Pereira & Bueno 2012; Rodrigues *et al.* 2012; Bastos–Pereira & Bueno 2013; Bueno *et al.* 2013). This figure could be higher due to a high number of difficult to identify cryptic species. Molecular studies have already shown that the diversity of *Hyalella* is underestimated, so that the regional diversity is very high, like in desert springs in California and Nevada, USA. For some of these unexplored areas, the number of species could be 500 only in North America (Witt *et al.* 2006; Väinölä *et al.* 2008).

Although many freshwater amphipods occur in caves, there are few *Hyalella* species described for these habitats. Currently, there are only five species of the genus inhabiting exclusively subterranean environments: *H. anophtalma* Ruffo, 1957, *H. caeca* Pereira, 1989, *H. muerta* Baldinger, Shepard & Threloff, 2000 and *H. spelaea* Bueno & Cardoso, 2011 occur in caves, and *H. imbya* Rodrigues & Bueno, 2012 occurs in hypothelminorheic habitat (Rodrigues *et al.* 2012). Three of these occur in Brazil: *H. caeca* and *H. spelaea* in São Paulo state, southeastern of the country and *H. imbya* in Rio Grande do Sul state, in the southernmost Brazil.

Brazil may turn out as a hotspot for subterranean environments, because it has large regions of limestone rocks and a high number of caves, which could support a high diversity of subterranean organisms (Fišer *et al.* 2013). Furthermore, Ferreira (2005) stressed that inventories of cave animals in Brazil are fragmentary, which might lead us to the assumption that the diversity in caves of poorly studied taxa, such as *Hyalella*, should be much higher than previously known.

The aim of this study is to describe a new cave species of *Hyalella* from Brazil, which is the first epikarstic species of the genus, as well as to provide an identification key for the Brazilian species of *Hyalella*.

Material and methods

Specimens were collected in the "Areias de Cima" cave which is part of the "Areias system", formed by three caves interconnected by a stream (for details, see "Habitat" and "Conservation").

The description is based on the main taxonomic characteristics of the genus, such as the shape and arrangement of the setae from gnathopods, uropods and telson, according mainly to González *et al.* (2006), Rodrigues *et al.* (2012) and Bueno *et al.* (2013). The terminology for setae follows Zimmer *et al.* (2009) and the classification of the mandibles is in sensu of Watling (1993).

The head and total body sizes of all the specimens collected were measured under a stereoscopic microscope with a millimetric scale. The body measurement was made from the tip of the head to the base of telson. A male paratype and a female allotype were dissected and theirs appendages were mounted on permanent slides. The description and all the drawings of the new species were based on the slides of the male paratype and the female allotype.

The type material is deposited in the Museu Nacional do Rio de Janeiro (MNRJ) and Coleção de Crustáceos da Universidade Federal de Lavras (CCUFLA).

Taxonomy

Order Amphipoda Latreille, 1816

Suborder Senticaudata Lowry & Myers, 2013

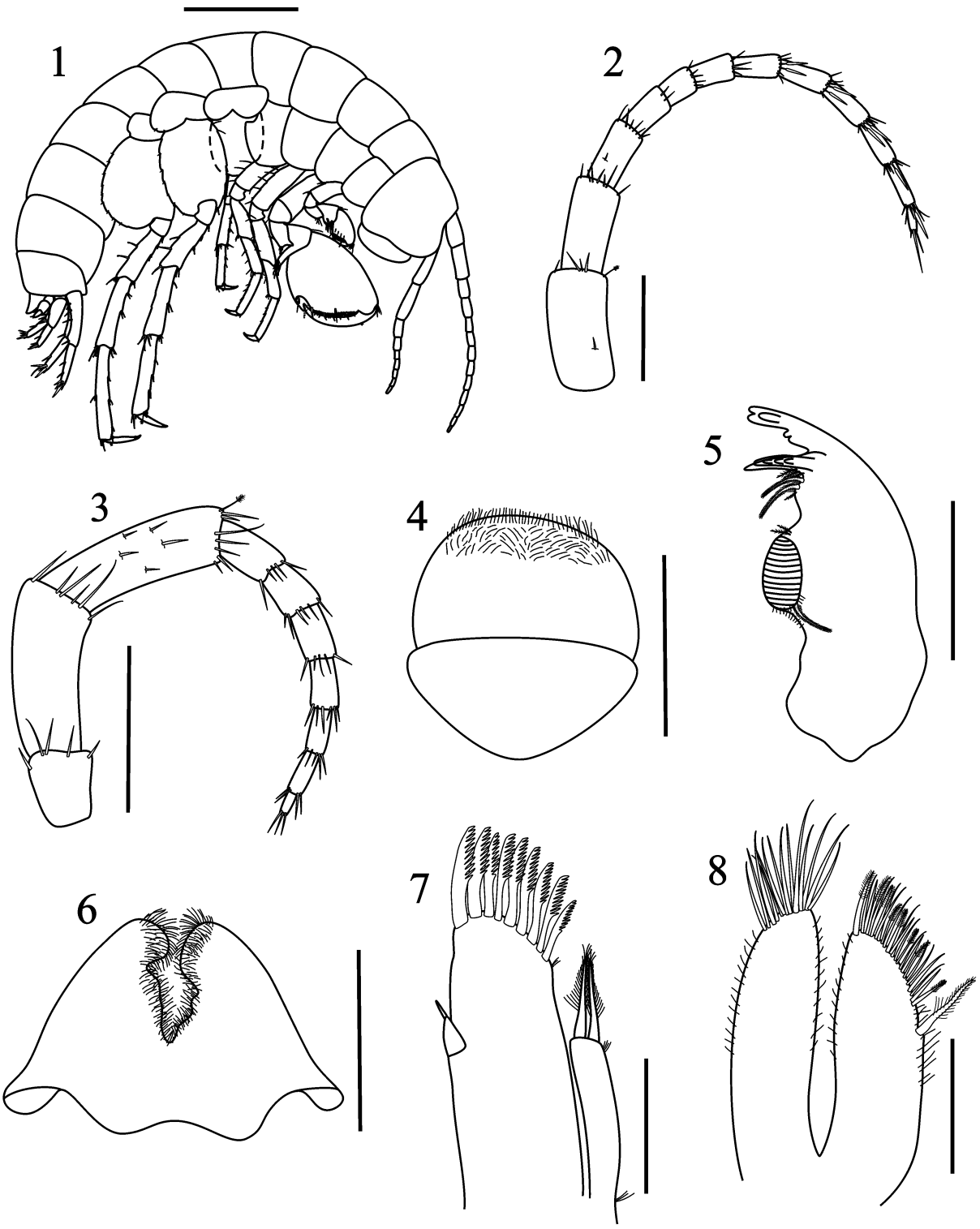
Family Hyalellidae Bulycheva, 1957

Genus *Hyalella* S. I. Smith, 1874

Hyalella epikarstica n. sp.

Material examined. Holotype male, body length = 3.95 mm, head length = 0.36 mm, MNRJ 24771.

Paratypes: CCUFLA 0344 with the same data as the holotype (one slide of a male and two entire individuals).

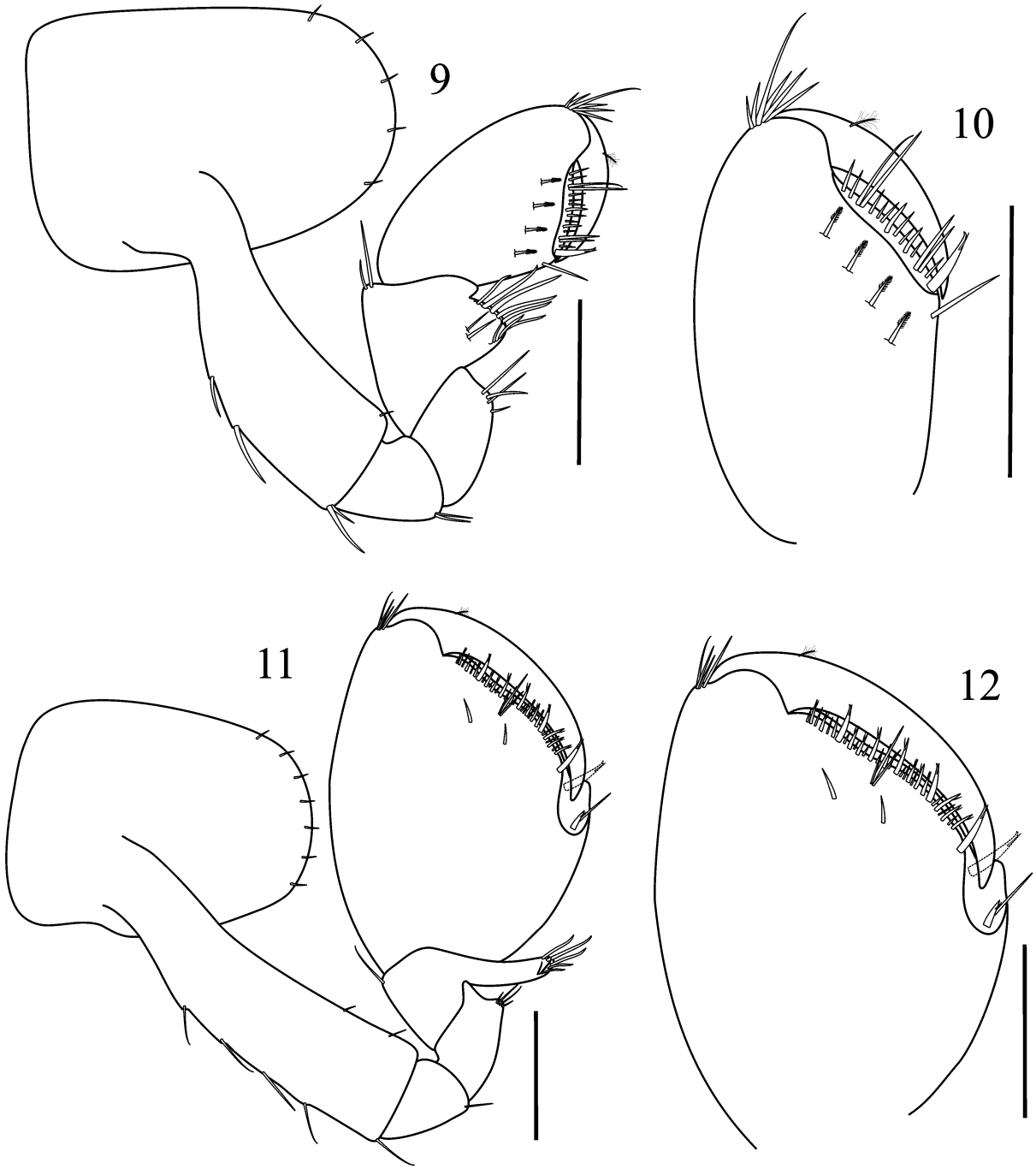


FIGURES 1–8. *Hyalella epikarstica* **n. sp.** paratype male (CCUFLA 0344), Mean body lenght: 3.1 ± 0.85 mm, mean head length: 0.26 mm ± 0.1 mm. (1) Habitus from holotype (MNRJ 24771), body length = 3.95 mm, head length = 0.36 mm; (2) Antenna 1; (3) Antenna 2; (4) Upper lip; (5); Mandible (6) Lower lip; (7) Maxilla 1; (8) Maxilla 2. Scales: (1) represents 1 mm; (2–4) represents 0.2 mm; (5–6) represents 0.05 mm; (7–8) represents 0.1 mm.

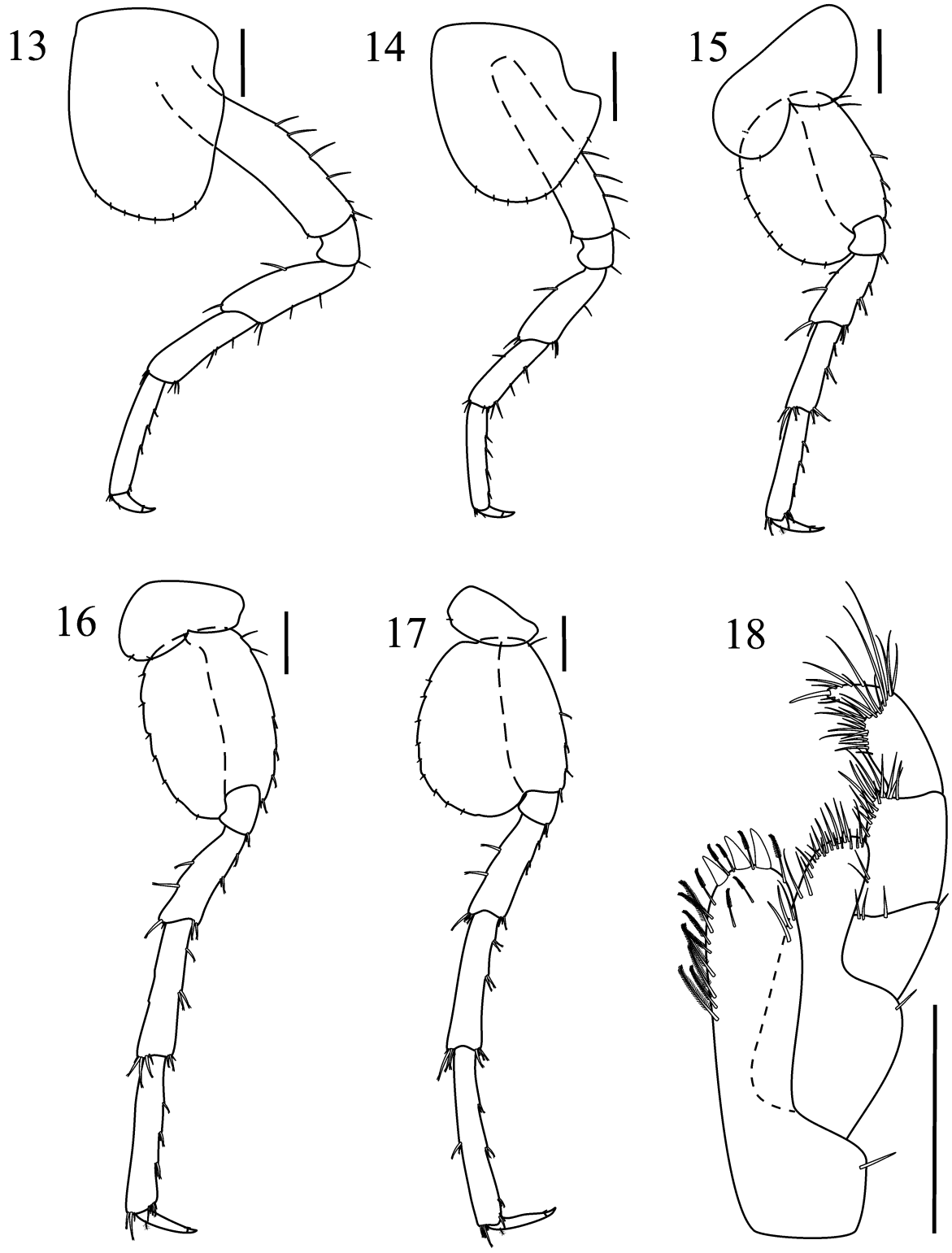
Type locality. Brazil, São Paulo state: Areias de Cima cave (24º35′28.5″S 48º42′08.3″W), São Paulo state, Iporanga municipality, Areias stream (Betary river basin), 225 meters of altitude, July, 7, 2012, Ferreira, R.L. coll.

Diagnosis. Body surface smooth. Eyes absent. Antenna 1 longer than antenna 2, flagellum with 9 articles. Antenna 2 less than half body length, flagellum with 7 articles. Maxilla 2 inner plate with only one long and strong

papposerrate apical seta. Maxilliped inner plate with three strong and very long cuspidate setae apically; palp smaller than inner plate. Gnathopod 1 propodus length less than twice maximum width, hammer shaped, inner face with 4 pappose setae; carpus border not pectinate, only with simple setae. Gnathopod 2 carpus wider than long, posterior lobe slim without pectinate border, with few short simple setae; propodus ovate; palm sub-equal to posterior margin, slope oblique. Peraeopod 6 and 7 much more longer than others. Uropod 1 inner ramus of male with a long curved seta apically and four simple setae. Uropod 3 shorter than telson, peduncle longer than ramus and with only one long cuspidate seta with an accessory seta. Telson as long as wide, with two short simple apical setae. Sternal gills tubular on segments 3 to 7.



FIGURES 9–12. *Hyalella epikarstica* **n. sp.** paratype male (CCUFLA 0344), Mean body lenght: 3.1 ± 0.85 mm, mean head length: 0.26 mm ± 0.1 mm. (9) Gnathopod 1; (10) Gnathopod 1 propodus and dactylus; (11) Gnathopod 2; (12) Gnathopod 2 propodus and dactylus. Scales: (9–12) represents 0.2 mm.

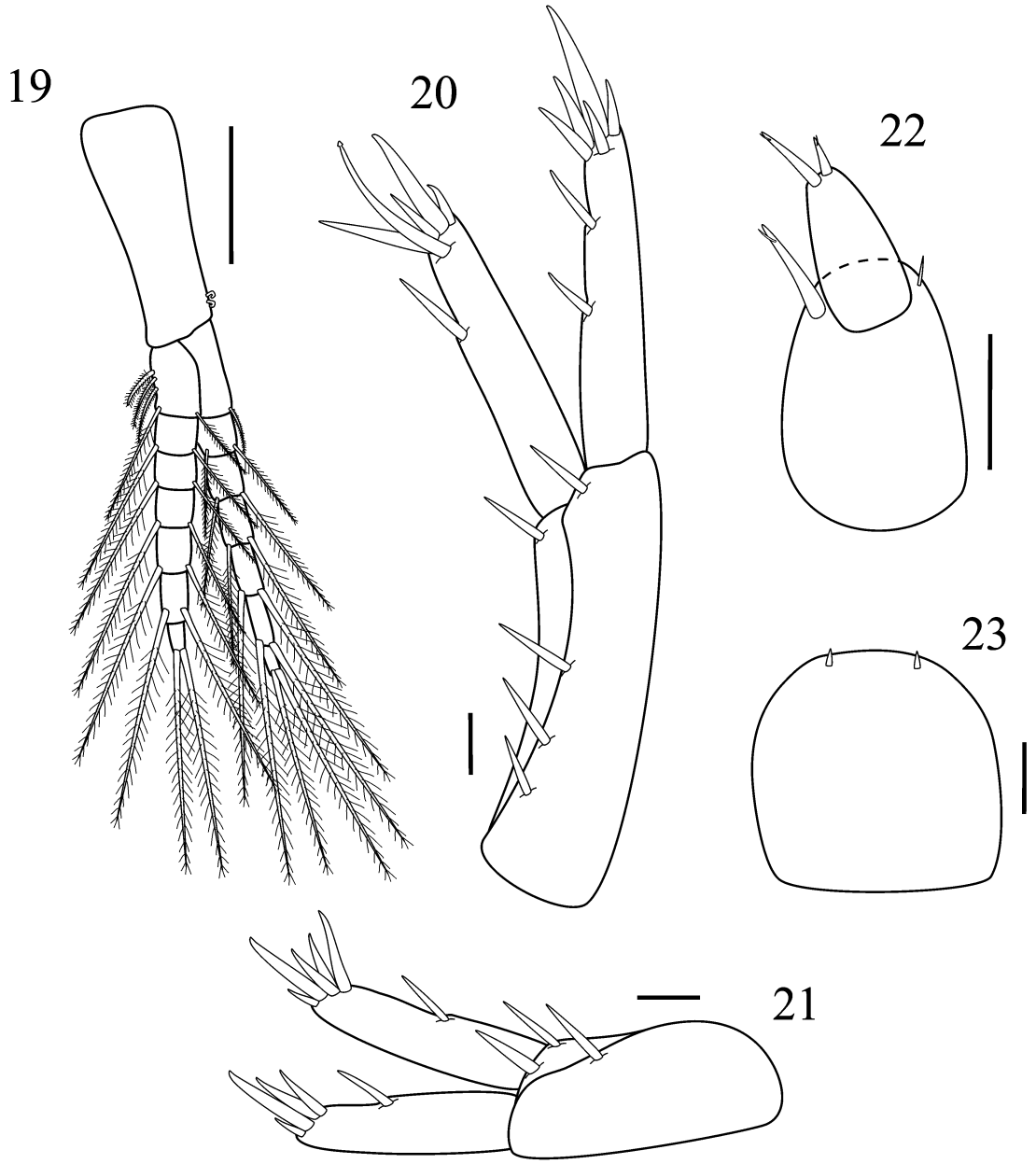


FIGURES 13–18. *Hyalella epikarstica* **n. sp.** paratype male (CCUFLA 0344), Mean body lenght: 3.1 ± 0.85 mm, mean head length: 0.26 mm ± 0.1 mm. (13) Peraeopod 3; (14) Peraeopod 4; (15) Peraeopod 5; (16) Peraeopod 6; (17) Peraeopod 7; (18) Maxilliped. Scales: (13–18) represents 0.2 mm.

Description of male. (Fig. 1). Mean body length: 3.1 ± 0.85 mm, mean head length: 0.26 mm ± 0.1 mm (n=4). Body surface smooth; epimeral plates not acuminate.

Head as the same size as the first pereon segment, rostrum absent. Eyes absent (Fig. 1).

Antenna 1 (Fig. 2) longer than antenna 2, less than half body length; peduncle surpassing head length; flagellum with 9 articles, longer than peduncle; aesthetascs occurring distally on flagellum after article 4.



FIGURES 19–23. *Hyalella epikarstica* **n. sp.** paratype male (CCUFLA 0344), Mean body lenght: 3.1 ± 0.85 mm, mean head length: 0.26 mm ± 0.1 mm. (19) Pleopod; (20) Uropod 1; (21) Uropod 2; (22) Uropod 3; (23) Telson. Scales: (19) represents 0.2 mm; (20–23) represents 0.05 mm.

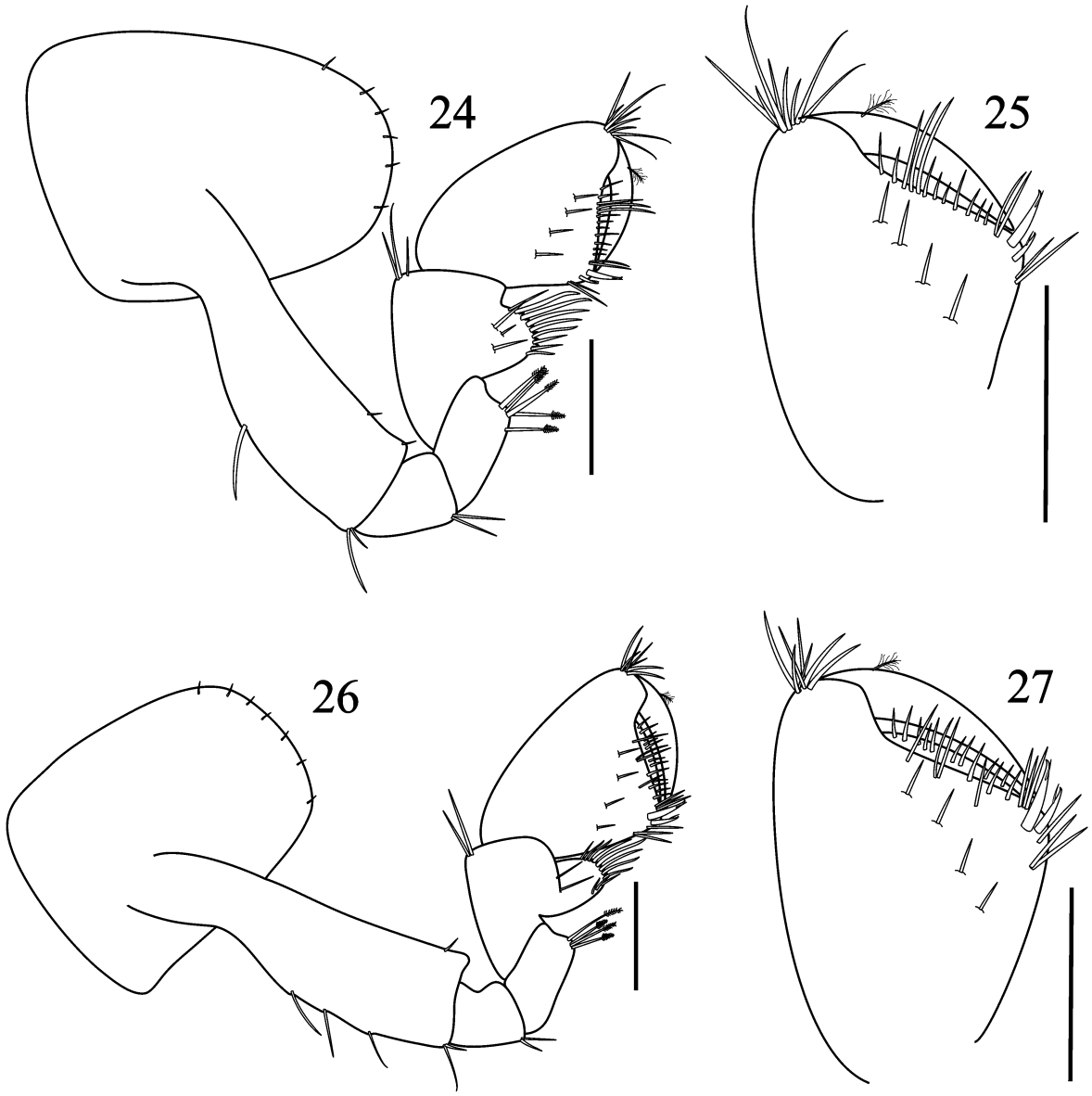
Antenna 2 (Fig. 3) peduncle not surpassing the second peraeonite, less than half body length, peduncle slender, longer than head; flagellum with 7 articles, shorter than peduncle.

Upper lip (Fig. 4) margin rounded; distal margin covered by setules on ventral and dorsal faces.

Basic amphipodan mandible, without palp; incisor toothed; molar large, cylindrical and triturative, with setules around its circumference; left lacinia mobilis with five teeth (Fig. 5), setal row on left mandible with two main pappose setae plus accessory setae; right lacinia mobilis with two teeth; setal row on right mandible with two main pappose setae plus accessory setae.

Lower lip (Fig. 6) outer lobes rounded and distally notched, with setules on ventral and dorsal faces; strongly irregular surface between the outer lobes.

Maxilla 1 (Fig. 7) palp uniarticulate, short, longer than wide, reaching less than half length the distance between the base of the palp and and the apex of the outer plate, with a long and strong seta; inner plate slender, shorter than outer plate, with two long papposerrate apical setae presenting long setules, with few minute setae on the inner margin; outer plate with nine serrate setae.



FIGURES 24–27. *Hyalella epikarstica* **n. sp.** allotype female (CCUFLA 0344), body length = 4.79 mm, head length = 0.46 mm. (24) Gnathopod 1; (25) Gnathopod 1 propodus and dactylus; (26) Gnathopod 2; (27) Gnathopod 2 propodus and dactylus. Scales: (24–27) represents 0.2 mm.

Maxilla 2 (Fig. 8) inner plate slightly shorter than outer plate, with only one long and strong papposerrate apical seta, eight serrulate and several simple apical setae; outer plate with abundant long simple setae; outer and inner plates with several setules.

Maxilliped (Fig. 18) inner plate longer than wide, with three strong and very long cuspidate setae apically, several pappose setae on apical and inner margins; outer plate shorter than inner plate, with several simple setae mediomarginally and apically; palp sub-equal in length as outer plate and shorter than inner plate, 4-articulate; article 1 longer than wide, outer and inner faces with few simple setae; article 2 longer than wide, inner margin with several long simple setae; article 3 longer than wide, outer and inner margins with several extremely long simple setae; dactylus unguiform with few simple setae, shorter than other articles, inner border with several simple setae; distal nail sub-equal to dactylus.

Gnathopod 1 (Fig. 9) subchelate; coxal plate wider than long, with simple setae on the anteromarginally; basis, ischium and merus with simple setae posteromarginally; carpus longer than wide, shorter than propodus, with posterior lobe produced and forming a scoop-like structure, without pectinate margin, with few simple setae; propodus width about ¾ of maximum length, hammer-shaped (Fig. 10), without setae on anterior margin, without comb-scales, inner face with four serrate setae, with a simple seta on the disto-posterior margin; palm slope

oblique, margin slightly concave, palm with many simple setae, posterior distal corner with one long and strong cuspidate seta with an accessory seta; dactylus claw-like surpassing the palm, without comb-scales, with one plumose seta anteriorly and few setae on the inner curvature.

Gnathopod 2 (Fig. 11) subchelate; basis hind margin with five simple setae; merus with few simple setae posteromarginally; carpus wider than long, posterior lobe nawrolly produced between merus and propodus, without pectinate border, with few short simple setae; propodus ovate (Fig. 12), length 1.1 maximum width, without comb-scales; palm sub-equal to posterior margin of propodus, slope oblique, with one row of several cuspidate setae with an accessory setae and simple setae, posterior distal corner with two long and strong cuspidate setae and with a deep cup for dactylus; dactylus claw-like, congruent with palm, plumose seta anteriorly, without comb-scales.

Peraeopods 3 to 7 simple. Peraeopod 3 (Fig. 13) coxal plate longer than wide, width about half its length, with small simple setae on the border; merus and carpus posterior margin with cluster of simple setae; propodus posterior margin with cuspidate setae; dactylus less than half-length of propodus. Peraeopod 4 (Fig. 14) coxal plate excavated posteriorly, wider than long, with small simple setae on the border; merus and carpus posterior margin with clusters of simples setae; propodus posterior margin with simple setae; dactylus less than half-length of propodus. Peraeopod 5 (Fig. 15) coxal plate wider than long, with two lobes and small simple setae on the border; merus, carpus and propodus margin with 10 marginal clusters of 1-5 cuspidate setae with an accessory setae; dactylus less than half-length of propodus. Peraeopod 6 (Fig. 16) coxal plate wider than long, with two lobes and small simple setae on the border; merus, carpus and propodus margin with 10 marginal clusters of 1–5 cuspidate setae with an accessory setae; dactylus less than half-length of propodus. Peraeopod 7 (Fig. 17) coxal plate wider than long with small simple setae on the border; merus, carpus and propodus margin with 10 marginal clusters of 1–5 cuspidate setae with an accessory setae; dactylus less than half-length of propodus. Peraeopod 3 sub-equal to peraeopod 4; peraeopod 5 shorter than others; peraeopods 6 sub-equal to 7, both longer than others.

Pleopods (Fig. 19) peduncle shorter than rami, with two coupling spines; both rami with several plumose setae.

Uropod 1 (Fig. 20) peduncle longer 1.3 times than rami; outer ramus longer than inner ramus; outer ramus with two dorsal simple setae and five simple setae apically, one much more longer than the others; inner ramus with one dorsal simple seta, male with a long curved seta apically on the ramus, four simple setae apically; peduncle with five simple setae dorsally.

Uropod 2 (Fig. 21) shorter than uropod 1; inner ramus with only one dorsal simple seta and five distal simple setae; outer ramus with only one dorsal simple seta and four distal setae; peduncle longer and wider than rami, with three simple setae.

Uropod 3 (Fig. 22) shorter than telson, than peduncle of uropod 1 and peduncle of uropod 2; inner ramus absent; outer ramus uniarticulate; peduncle longer than wide, with only one long cuspidate seta with an accessory seta; ramus shorter than peduncle; basal width 3 times the width of ramus apex, with two cuspidate setae with an accessory seta.

Telson (Fig. 23) entire, apically rounded, as long as wide, with two short simple apical setae; plumose setae may be present laterally.

Coxal gills sac-like, present on peraeonites 2 to 6. Sternal gills tubular present on peraeonites 3 to 7. Both coxal and sternal gills are extremely reduced in size.

Female. Mean body length: 4 ± 0.8 mm, mean head length: 0.3 ± 0.1 mm (n=2). Antenna 1 similar in shape to male, flagellum with 10 articles; antenna 2 similar in shape to male, flagellum with 7 articles. Gnathopod 1 (Fig. 24) different to male gnathopod 1; carpus longer than wide, without comb-scales; with posterior lobe produced and forming a scoop-like structure, without pectinate margin, with few simple setae; propodus (Fig. 25) as long as wide, "hatchet-shaped", palm longer than posterior margin of propodus, without comb-scales, inner face with four simple setae, palm slope transverse, posterior distal corner with two long and strong cuspidate seta with an accessory seta; dactylus claw-like. Gnathopod 2 (Fig. 26) similar in size and shape to gnathopod 2; different in shape to male gnathopod 2 and smaller; propodus (Fig. 27) as long as wide, subchelate, inner face with four simple setae, palm transverse, without comb-scales. Telson similar in shape to male.

Habitat. The epikarst is defined as the heterogeneous interface between unconsolidated material (soil, sediments and modified carbonate rock) that is partially saturated with water and capable of delaying or storing water and locally rerouting vertical infiltration to the deeper regional phreatic zone of the karst aquifer (Jones *et al*. 2004). The main traits of the epikarst habitats usually prevent direct sampling, and most species known from this habitat are indirectly collected especially in dripping pools (Pipan & Culver 2005).

This is the case of *H. epikarstica*. Although there are three distinct streams in the Areias de Cima cave, no specimen was recorded in these lotic habitats. The specimens were only collected in a single place within the cave, located in the eastern branch of the system (Figs. 28, 29 and 31). Specimens were found swimming in small travertine pools, which were being filled by percolating water coming from a small crack in the cave wall (Fig. 32). During five visits to the cave, specimens were only recorded two times, when the area was under heavy rain. We believe that the amplified flow of water from the epikarst has washed out few specimens that were carried out to the travertine pools. In the other episodes in which we have visited the cave, the travertive pools were almost empty, clearly indicating that they are not persistent habitats, thus, incapable to maintain populations.

There are other amphipod species that are considered epikarstic, as *Niphargobates orophobata* Sket, 1981 and *Niphargus fongi* Fišer & Zagmajster, 2009. Specimens from the former were collected from a jet of percolating water in Panina cave, Slovenia, while specimens from the latter were found in pools of percolating water, some of which temporarily dry up. According to Fišer & Zagmajster (2009) the narrow distribution range of *N. fongi* suggests that the species lives in limestone fissures, possibly in the epikarst ecotone. This is the same situation observed for *H. epikarstica*, what strongly suggests that the species is associated to these "above cave" compartments.

Conservation. The karst of Iporanga is associated with carbonate rocks of the Açungui Group, which was formed between 1.45 billion and 540 million years ago (Campanha *et al.* 2008; 2010). The caves and their special features developed during the Quaternary Period (between 1.8 million years and the present) and are still active (Karmann 1994). The external vegetation comprises the Brazilian Atlantic Forest, which is well preserved in the area (Fig. 33).

The Areias de Cima cave is part of the "Areias system", which is divided into three caves connected by the Areias stream. The Areias de Cima cave is the first (upstream) cave in the system, possessing around 5.5 km of linear projection, being divided in two conduits, interconnected near the entrance (Fig. 29). The specimens were found in the left branch of the cave (Fig. 31). To date, 17 troglobitic species (16 invertebrates and one vertebrate) have been recorded for the whole system (Trajano 2007; Ázara & Ferreira 2013), which is considered the richest system for troglobitic fauna in Brazil. This cave system has been studied for over 100 years and only in recent samplings few specimens of this new species were found, which strongly suggests its epikarstic status.

The cave system is protected, since it is located within a Conservation Unit (Parque Estadual Turístico do Alto Ribeira – PETAR). Tourist visits are prohibited, so that the cave is extremely preserved.

Etymology. The species epithet "epikarstica" refers to the species habitat. The word is feminine in gender.

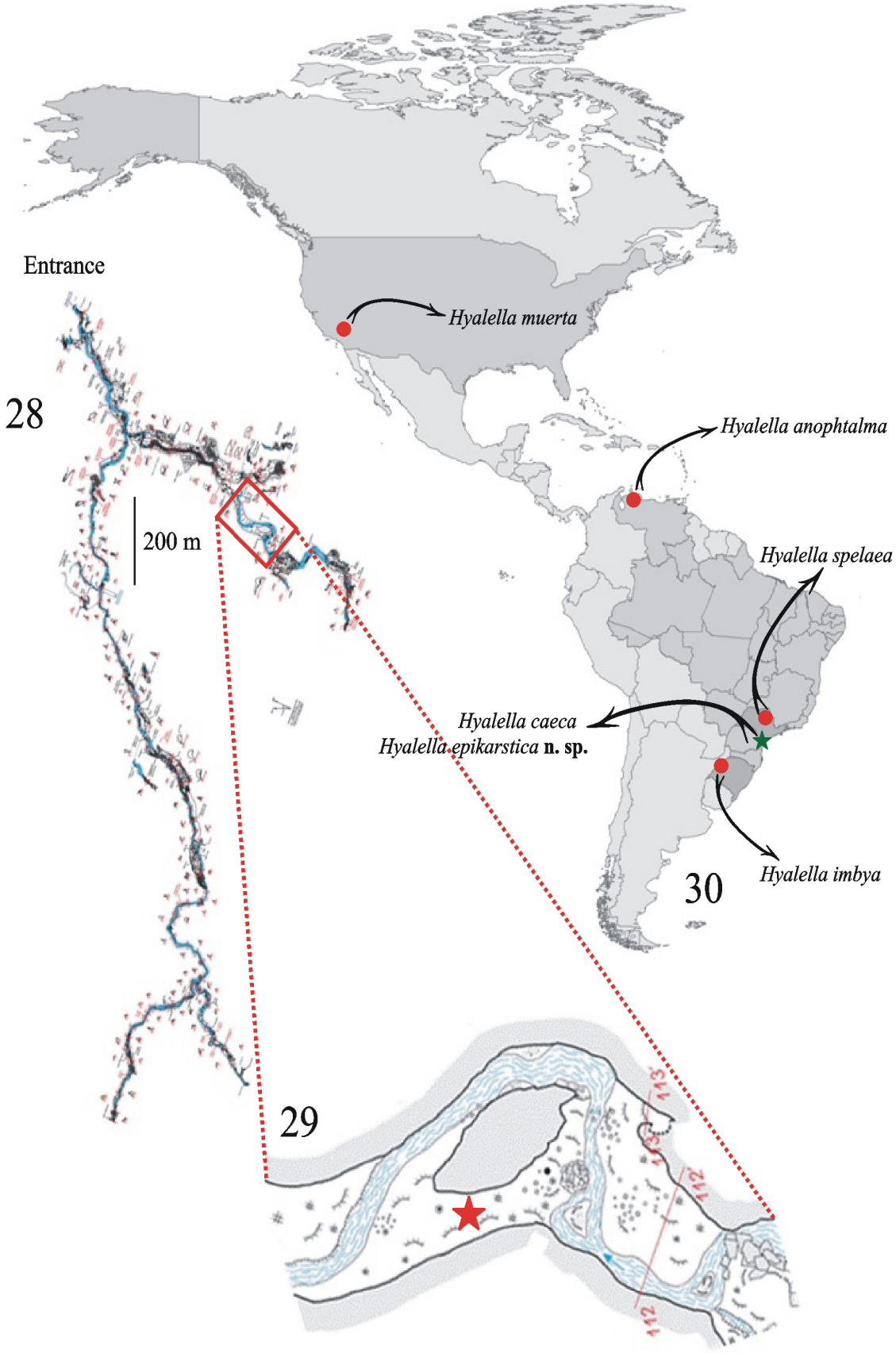
Remarks. *Hyalella epikarstica* has similar characteristics and also adaptations as other troglobiotic species of the genus. The new species shares the following characters with *H. imbya* and *H. muerta*: presents the antenna 1 longer than antenna 2; absence of comb-scales on both gnathopods; and sternal gills present on peraeonites 3–7. However, *H. epikarstica* differs from *H. muerta* by the presence of a curved seta on the inner ramus of uropod 1 and differs from *H. imbya* on the palm slope of gnathopod 1. The palm slope on gnathopod 1 on *H. imbya* is transversal and on *H. epikarstica* is oblique, a characteristic very unusual in the genus for this appendage.

Hyalella epikarstica is different in almost all the morphological characteristics when compared to *H. anophtalma*, like size proportions of antennae; presence of comb-scales in gnathopods; arrangement of sternal gills; presence of an apical seta on telson; and presence of a curved setae on uropod 1.

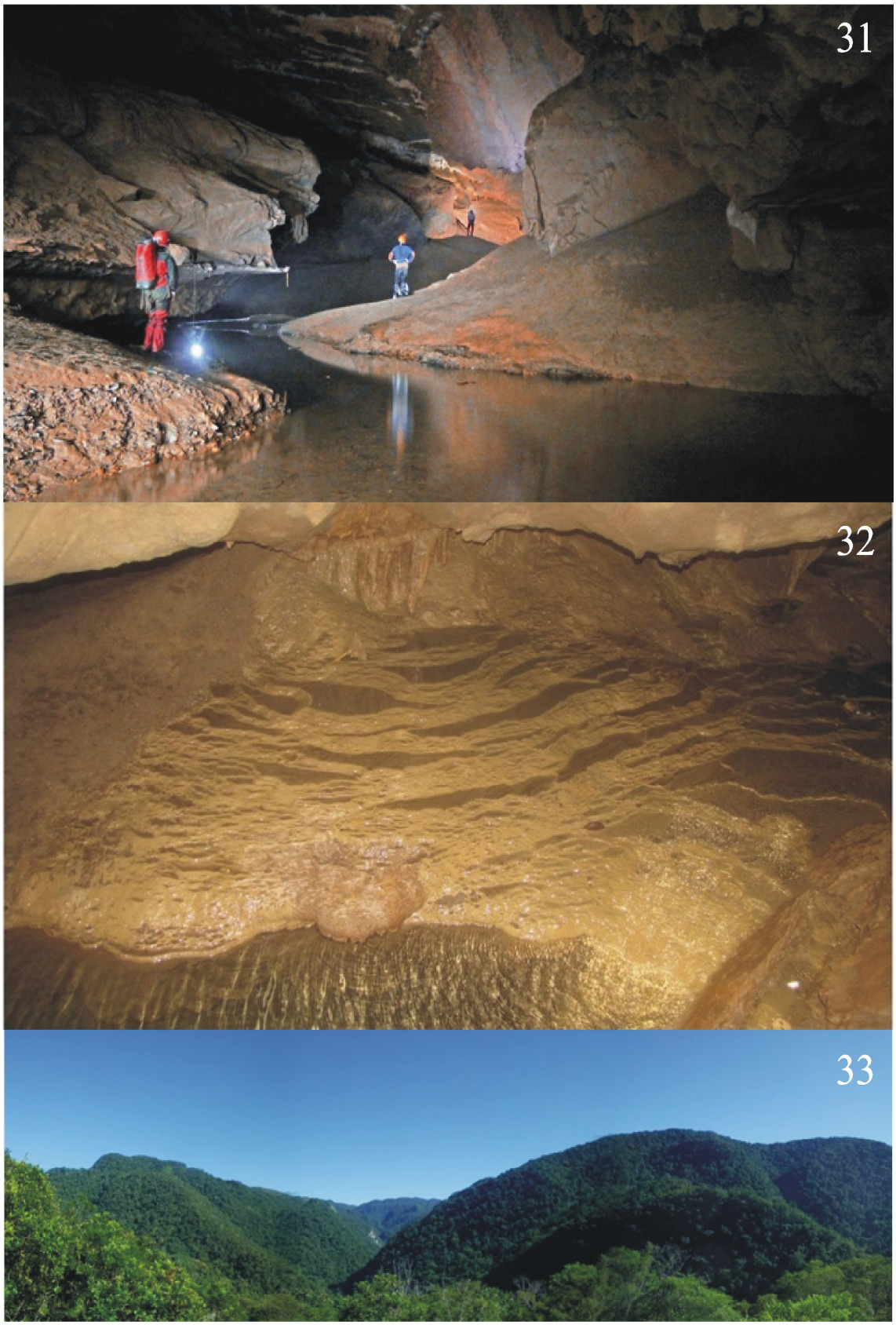
Possibly because both species live close to each other, *H. epikarstica* share many morphological characteristics with *H. caeca*, such as: absence of comb-scales on gnathopods; presence of sternal gills on peraeonites 3–7; number and arrangement of setae on telson; and the presence of only one papposerrate apical seta on the inner plate of maxilla 2. Although, unlike the new species, *H. caeca* does not have a curved seta on the inner ramus of uropod 1; antenna 1 is shorter than 2; carpus of gnathopod 1 is longer than propodus and bears serrate setae and denticles on the carpus distal lobe, forming a pectinate margin, absent on *H. epikarstica.*

Finally, *H. spelaea* differs from *H. epikarstica* in the following: reduced but present eyes; absence of a curved seta on inner ramus of uropod 1; sternal gills on peraeonites 2–7; antenna 1 smaller than antenna 2; and presence of comb-scales on gnathopod 1.

Moreover, *H. epikarstica* has unique morphological characteristics: extreme reduction in the size of sternal and coxal gills; lower lip with strongly irregular surface between the outer lobes; maxilliped inner plate with three very long cuspidate setae apically and palp extremely reduced with long simple setae; distal lobe basis of carpus on gnathopod 1 and 2 without pectinate margin, serrate setae, denticles or comb-scales, only with few simple setae; and palm slope oblique on both male gnathopods.



FIGURES 28–30. (28) Schematic drawing of Areias de Cima cave, showing its division in two conduits, interconnected near the entrance; (29) Detail of the eastern branch of the system in Areias de Cima cave; the red star indicates the place where the specimens of *Hyalella epikarstica* **n. sp.** were collected; (30) Distribution map showing the places of occurrence of the six troglobiotic species of *Hyalella*.



FIGURES 31–33. (31) Left branch of the Areias de Cima cave; (32) Small travertine pools, the only place within the cave where the specimens of *Hyalella epikarstica* **n. sp.** were found; (33) Brazilian Atlantic Forest, the well preserved external vegetation of the Areias de Cima cave.

Discussion

Hyalella epikarstica is the fourth troglobiotic species of the genus described for Brazil and the sixth in the world (Fig. 30). The troglobiotic species of *Hyalella* exhibit many morphological characteristics compared to epigean species, that could be adaptations to the subterranean environment: reduction of body size; reduction or absence of eyes; reduction of size and number of gills; elongation of antenna 1 and increase of the length of aesthetascs; modifications in some mouthparts, as the maxilliped, with the increase in size and number of the setae; pronounced reduction of size of carpus on gnathopod 2; decrease in quantity or absence of serrate setae on carpal lobe; pronounced elongation of propodus; palm with acute oblique slope and deep cup for dactylus; absence of comb-scales in both gnathopods; elongation of peraeopods 6 and 7; reduction of the size of uropod 3; and decrease in number or absence of setae on telson.

Jones *et al.* (1992) have demonstrated that elongation of the antennae in cave populations of *Gammarus minus* results in a more accurate sensorial ability compared to epigean populations. The antennae of male amphipods have specialized structures – aesthetascs – capable of detecting the pheromones from females, facilitating their gathering for reproduction. Furthermore, apparently the elongation of antennae and the reduction of the eyes are inversely related on these crustaceans. The reduction or absence of eyes could lead to an improvement on the functioning of the non-visual sensorial system, since both compete for the information processing in the central nervous system (Jones & Culver 1989). Thus, the absence of visual information, through the reduction or absence of the eyes, could improve the efficiency of the processing of non-visual signals, of the antennae.

The reduction in body size and the elongation of the locomotory appendages, as the peraeopods, can also be considered as an adaptation to the cave habitat of some troglobiotic species of the genus *Niphargus* (Trontelj *et al.* 2012). Besides, geographically and genetic independent communities of many species from the genus exhibit the same variations in morphological characters, indicating the existence of a convergent evolution for adaptation to the subterranean life. However, minor morphological variations between species within the genus can be a result of the competition with other organisms in the community, as well as adaptations to specific microhabitats (Trontelj *et al.* 2012).

Further studies are necessary to check if the morphological structures of the mouthparts, gnathopods and the other appendages from the troglobiotic species of *Hyalella* are specific adaptations to the cave environment. As these species inhabit sites completely isolated from each other, it may be that the morphological traits observed are a result of convergent evolution, caused by multiple invasions to the subterranean environment and probably are not synapomorphic. However, a cladistic analysis and the reconstruction of the phylogeny need to be conducted to support this hypothesis.

In order to better understand the real functions of the morphological traits shown by the troglobiotic species of *Hyalella*, studies on the taxonomy, phylogeny, geometric morphology, behavior and physiology must be performed in future. Thus, we might eventually be able to infer how the selection pressure has acted in such cave organisms.

Key to the species of *Hyalella* from Brazil

1	Body surface smooth	2
–	Body with flanges on some peraeonites and some pleonites	14
2	Eyes absent or reduced	16
–	Eyes present and pigmented	3
3	Uropod 1 of male without curved seta on inner ramus	4
–	Uropod 1 of male with curved seta on inner ramus	9
4	Dorsal margin of uropod 3 without setae	5
–	Dorsal margin of uropod 3 with setae	*H. dielaii*
5	Gnathopod 2 with propodus much less than twice its width	6
–	Gnathopod 2 with propodus almost twice its width	*H. longistila*
6	Slender setae on apical margin of telson	7
–	Strong setae on apical margin of telson	*H. meinerti*
7	Inner face of gnathopod 1 with a row of five or less serrate setae	8
–	Inner face of gnathopod 1 with a row of 10 or more serrate setae	*H. warmingi*
8	Flagellum of antenna 2 with 14–17 articles	*H. minensis*
–	Flagellum of antenna 2 with 19–25 articles	*H. gracilicornis*

9	Sternal gills present on segments 2–7	10
–	Sternal gills present on segments 3–7	*H. montenegrinae*
10	Telson with more than two apical setae	11
–	Telson with only two strong apical setae	*H. carstica*
11	Gnathopod 2 dactylus shorter than propodus palm	12
–	Gnathopod 2 dactylus as long as propodus palm	13
12	Rami of uropod 3 with only cuspidate setae distally	*H. xakriaba*
–	Rami of uropod 3 with cuspidate and simple setae distally	*H. brasiliensis*
13	Peduncle of uropod 3 with four to six cuspidate apical setae	*H. curvispina*
–	Peduncle of uropod 3 with more than seven cuspidate apical setae	*H. castroi*
14	Uropod 1 of male with curved seta on inner ramus	15
–	Uropod 1 of male without curved seta on inner ramus	*H. pseudoazteca*
15	Flanges present only on pleonite 1–2	*H. kaingang*
–	Flanges present on peraeonite 7 and pleonite 1–3	*H. pleoacuta*
16	Sternal gills present on segments 3–7	17
–	Sternal gills present on segments 2–7	*H. spelaea*
17	Antenna 1 longer than antenna 2	18
–	Antenna 1 shorter than antenna 2	*H. caeca*
18	Posterior lobe of carpus with a row of serrate setae	*H. imbya*
–	Posterior lobe of carpus with a row of simple setae	*H. epikarstica* **n. sp.**

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