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HYALELLA MAYA, A NEW HYALELLIDAE SPECIES (CRUSTACEA: AMPHIPODA) FROM A CENOTE IN THE YUCATAN PENINSULA, MEXICO

Aurora Marrón-Becerra¹, Margarita Hermoso-Salazar² and Vivianne Solís-Weiss^{3,C}

Abstract

Hyalella maya n. sp. from a sinkhole (cenote) in Quintana Roo, Mexico, is described herein, based on morphological characters. It is the third species recorded in Mexico and the second species of *Hyalella* found in the Aktun-Ha sinkhole. It can be distinguished from its closest species, *Hyalella azteca* from Veracruz, by the shape of the palp of maxilla 1, the number of setae in the posterior margin of the basis, the relative palm length and the truncate process of gnathopod 2 in males, the number of setae in the lobe of the basis of pereopod 7, and the shape and setation of the telson. Those characters have been found to be useful for species distinction in the complex by González and Watling (2002). The main differences between *Hyalella cenotensis* and *Hyalella maya* n. sp. are: the presence of eyes, the number of articles in the flagellum in antennae 1 and 2, the relative length of antennae to the total body length, the relative length of the rami in uropod 3 to the peduncle length, and the number of setae in the telson.

Introduction

Up to now, about 70 species of the genus *Hyalella* have been described, with approximately 60 species distributed in South America (Horton and Lowry, 2013) and 12 species in North America, Central America and the Caribbean (Fig.1, Table 1). These last are: *Hyalella azteca* (De Saussure, 1858); *H. faxoni* (Stebbing, 1903); *H. texana* (Stevenson and Peden, 1973); *H. montezuma* (Cole and Watkins, 1977); *H. caribbeana* (Bousfield, 1996); *H. longicornis* (Bousfield, 1996); *H. muerta* (Baldinger, Shepard and Threlhoff, 2000); *H. sandra* (Baldinger, Shepard and Threlhoff, 2000); *H. meraspinosa* (Baldinger, 2004); *H. cenotensis* (Marrón-Becerra, Hermoso-Salazar and Solís-Weiss, 2014); *H. spinicauda* (Soucek and Lazo-Wasem, et al., 2015) and *H. wellborni* (Soucek and Lazo-Wasem, et al., 2015). The *Hyalella azteca* complex is considered to be present only in North America, Central America and the Caribbean, with no records from South America (González and Watling, 2001; 2002; González, 2003; González and Watling, 2003a-d). There are now around 40 “provisional” species included in the *H. azteca* complex: 33 in the U.S. (Witt et al., 2006) and seven in Canada (Hogg et al., 1998; Witt and Hebert, 2000). The reason to call them “provisional species” is that all of them have been differentiated through molecular studies, but they have not yet been formally described.

In the past, *Hyalella azteca* (De Saussure, 1858) was considered as one species, widely distributed in the American continent but not as a complex of species. This was due to the absence of evident morphological variability, as well as to the lack of carefully detailed morphological studies (González and Watling 2002). In Mexico, *H. azteca* was the only epicontinental, epigean species of amphipod recorded, with a wide distribution in the whole country. However, we now know that this species is, in fact, a complex of several species, taxonomically close, but that their ecological and morphological characteristics have not been sufficiently studied to separate them accurately (González and Watling, 2002; Brito et al., 2014; Soucek and Lazo-Wasem, et al., 2015).

De Saussure (1858) described *Hyalella azteca* as *Amphitoe aztecus* from material collected in Veracruz state and Lago de Chapultepec, Mexico City. However, the description, as well as the illustrations, are not detailed enough and the type locality was not specified. That is why González and Watling (2002) redescribed it, making a morphologically-detailed diagnosis based on the syntypes, to try and clarify its taxonomic status.

Currently, two species of *Hyalella* (Fig. 1) are known in Mexico: *H. cenotensis*, with no eyes, from a cenote in Tulum, Quintana Roo, and *H. azteca*, with well-developed eyes and with a distribution probably limited to the state of Veracruz, Mexico (González and Watling, 2002; Graening et al., 2012).

The objective of this study is to describe a new species of *Hyalella*, collected in a sinkhole in the Yucatan Peninsula, to compare it to the other species of the genus in the area (*H. cenotensis* and *H. azteca*) and to other species from North America and the Caribbean. In addition to contributing to the knowledge of this complex of species, we wish to demonstrate that more studies, focused on this genus of amphipods, are needed in these fragile, endangered environments.

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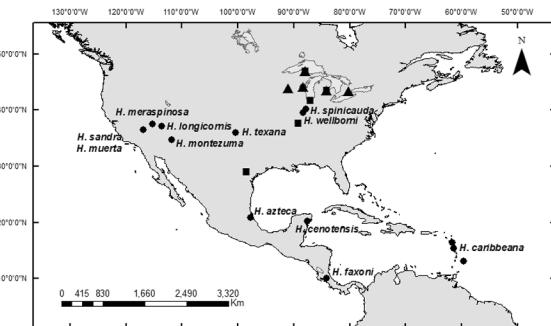


Figure 1. Map with the location of the species of *Hyalella* in North America, Central America and the Caribbean region. Circles indicate the type locality of each species. Triangles indicate the localities of *H. spinicauda*. Squares indicate the localities of *H. wellborni*.

Method

Samples were collected manually at a depth of 1-2 m, while snorkeling at Cenote Aktun-Ha, Quintana Roo, Mexico ($20^{\circ}16.48'N$, $87^{\circ}29.20'W$), in April 2008 on dense algal mats. The upper level taxon was identified following Lowry and Myers (2013). The specimens collected are deposited in the National Collection of Crustaceans, Instituto de Biología of the Universidad Nacional Autónoma de México (UNAM). Specimens were dissected and body parts were mounted on permanent slides. The total length was measured from the tip of the head to the base of the telson, using an optical microscope with a micrometric scale on the objective lens. The terminology for the setae follows Zimmer et al. (2009). Characters for the key were taken from Soucek et al. (2015).

Scanning electron photographs were taken from paratypes (one female and one male) with a Hitachi SU1510 scanning electron mi-

Table 1. *Hyalella* species recorded in North America, Central America and the Caribbean in chronological order.

Species	Distribution	Country	Habitat	References
<i>H. azteca</i> (De Saussure, 1858)	Veracruz	Mexico	freshwater, epigean, benthic	De Saussure (1858); González and Watling (2002)
<i>H. faxonii</i> Stebbing, 1903	Reventado Volcano and Barva Volcano	Costa Rica	freshwater, epigean, littoral	Stebbing (1903)
<i>H. texana</i> Stevenson and Peden, 1973	Clear Creek Spring, Texas	USA	freshwater, epigean, benthic	Stevenson and Peden (1973)
<i>H. montezuma</i> Cole and Watkins, 1977	Montezuma Well, Yavapai, Arizona	USA	freshwater, epigean, planktonic	Cole and Watkins, (1977)
<i>H. caribbeana</i> Bousfield, 1996	Pond and lakes in Grand Terre islands, Guadeloupe, Dominica, Barbados and probably in other lesser Antilles windward islands.	Guadeloupe, Dominica and Barbados	freshwater, epigean, benthic	Bousfield (1996)
<i>H. longicornis</i> Bousfield, 1996	Utah	USA	freshwater, epigean, benthic	Bousfield (1996)
<i>H. muerta</i> Baldinger, Shepard and Threlloff, 2000	Texas Springs and Travertine Springs, Death Valley National Park, Inyo, California	USA	freshwater, hypogean, benthic	Baldinger et al. (2000)
<i>H. sandra</i> Baldinger, Shepard and Threlloff, 2000	Texas Springs and Travertine Springs, Death Valley National Park, Inyo, California	USA	freshwater, epigean, benthic	Baldinger et al. (2000)
<i>H. meraspinosa</i> Baldinger, 2004	Ash Springs, Lincoln, Nevada	USA	freshwater, epigean, benthic	Baldinger (2004)
<i>H. cenotensis</i> Marrón-Becerra, Hermoso-Salazar and Solis-Weiss, 2014	Cenote Aktún-Ha, Tulum, Quintana Roo	Mexico	freshwater, hypogean, benthic	Marrón-Becerra et al. (2014)
<i>H. spinicauda</i> Soucek and Lazo-Wasem, 2015	Ponds, lakes and rivers in Illinois, Indiana, Michigan, Wisconsin and Texas, USA and Ontario, Canada	USA and Canada	freshwater, epigean, benthic	Soucek et al. (2015)
<i>H. wellborni</i> Soucek and Lazo-Wasem, 2015	Ponds, lakes and rivers in Illinois, Indiana, Michigan, Wisconsin and Texas, USA and Ontario, Canada	USA and Canada	freshwater, epigean, benthic	Soucek et al. (2015)
<i>H. maya</i> n. sp.	Cenote Aktún-Ha, Tulum, Quintana Roo	Mexico	freshwater, epigean, benthic	In this study

croscope (SEM) from the Laboratory of Microscopy and Photography of Biodiversity I, at the Instituto de Biología, UNAM.

The number of articles in the flagellum of antennae 1 and 2 were compared between males' and females' paratypes using a linear regression of the total body length and the number of articles of at least 10 organisms of each sex.

Results

Order AMPHIPODA Latreille, 1816
Suborder Senticaudata Lowry and Myers, 2013
Infraorder Talitrida Rafinesque, 1815

Parvorder Talitridira Rafinesque, 1815
 Superfamily Talitroidea Rafinesque, 1815
 Family Hyalellidae Bulycheva, 1957
 Genus *Hyalella* S. I. Smith, 1874

***Hyalella maya* n. sp.**

Material examined: Holotype male, size 3.85 mm (Cat. No. CNCR 31502), from algae in the outer area of Cenote Aktun-Ha, Quintana Roo, Mexico (20°16.48'N, 87°29.20'W) in April 2008. Paratypes: male, size 4.10 mm, 1 ovigerous female, size 4.15 mm (Cat. No. CNCR 31503), locality was the same as holotype. Collectors: Vivianne Solis-Weiss and Sarita Frontana Uribe.

Type locality: Cenote Aktun-Ha, Quintana Roo, México (20°16.48'N, 87°29.20'W).

Etymology: The name is derived from the Mayan civilization that ruled the area in the past.

Habitat: In algae, freshwater (pH 7.2, water temperature 24.85°C, dissolved oxygen 1.93 mg/l).

Diagnosis: Eyes present. Tergites of pleon 1 and 2 with dorsoposterior carina. Head length: slightly shorter than the length of pereonites 1 and 2 combined, reaching more than half the length of pereonite 2. Antenna 1 is shorter than antenna 2; flagellum with nine articles. Antenna 2 is almost 1.3 times longer than antenna 1, longer than half body length; flagellum with 10 articles. Maxilla 1, vestigial palp, uni-articulated, short, rounded with an apical seta; inner plate with three strong, pappose apical setae. Lower lip, distal lobes rounded. Gnathopod 2 (males), carpus posterior lobe is approximately 1.5 times the width of merus, palm oblique without an evident, truncated process or distinct notch; length of palm is similar to the posterior margin of gnathopod 2. Pereiopod 7 basis with up to 12 short, fine setae. Telson is wider than long; distal margin is rounded with two long, separated, simple setae, with three short, sub-marginal setae on both left and right sides.

Male description (Figs. 2-3): Size 3.85 mm (holotype). Coxae 1-4 are sub-rectangular, longer than wide, inferior margin is rounded with small setules, coxa 4 with a posterior excavation. Coxae 5-7 are shorter than coxae 1-4; coxa 5 with two subequal lobes, coxa 6 with two unequal lobes, and coxa 7 with a single lobe.

Pleon (Fig. 2A): pleonites 1 and 2 with dorsoposterior carina, ventral margin of epimeral plates 2 and 3 slightly pointed.

Head (Figs. 2A, 5C): typically, gammaridean, smooth surface; length is less than pereionites 1 and 2 combined, reaching half of pereonite 2; eyes are present and rostrum is absent.

Antenna 1 (Figs. 2A, 3H, 5C): shorter than antenna 2, length reaching beyond two-thirds of the fourth pereionite; peduncle is longer than head, reaching more than half the length of first pereionite. Peduncle articles become gradually smaller in length and width toward their distal portion; close to the middle length of the ventral margin for the first peduncle article, a short cuspidate seta and one at the distal end; flagellum with nine articles, longer than peduncle, basal article of flagellum not elongate; no accessory flagellum.

Antenna 2 (Figs. 2A, 3I, 5C): almost 1.3 times longer than antenna 1, length is slightly longer than half the total body length, reaching half the length of the sixth pereionite. Peduncle articles increase gradually in length and decrease in width toward their distal end; flagellum with 10 articles, length 1.16 times the peduncle length.

Buccal parts (Fig. 3): upper lip (Fig. 3C), distal margin is rounded with numerous setules, longer and more distant toward the lateral ends; distal surface of the outer surface has two rows of short setules, very close toward the middle of the row. Lower lip (Fig. 3B): distal lobes are rounded distally; apices are relatively separated from one another with numerous setules; no inner lobes; mandibular lobes are rounded with distal ends slightly directed outward.

Mandibles (Figs. 3E-F): incisor with six teeth, distal tooth stronger than the proximal five; strong and triturative molar (Figs. 9E-F); left lacinia mobilis (Fig. 3E) is more developed than right, with five teeth; length of the external tooth is subequal to the distal tooth of the incisive process; right lacinia mobilis has two teeth, each with inner margin denticulate (Figs. 3F, 9G); left mandible ranker row has three longer and two shorter pappose setae, and a large accessory pappose seta at the end of the molar process in both mandibles; palp is lacking.

Maxilla 1 (Fig. 3A): outer plate of maxilla 1 is slightly longer than 2.5 times the width of the inner plate, distal margin with nine serrate setae; vestigial palp is uni-articulated, longer than wide, with rounded apex and a simple seta; palp length is slightly shorter than a third of the distance of the palp base, to the end of the longest seta of the outer plate; inner plate is slender and shorter than outer one, almost reaching the palp base, distal margin with two to three pappose setae.

Maxilla 2 (Fig. 3D): plates are subequal in length and shape; inner plate is almost half the length of the inner margin with two plumose setae; distal margin of both plates has simple setae.

Maxilliped (Fig. 3G): inner plate is shorter than outer; distal margin has three cuspidate setae of equal size with plumose setae; inner margin has plumose setae; outer plate is elongate; distal and inner margins have numerous simple setae; palp, longer than inner plate, is composed by four articles. The first article has three simple setae at the far-

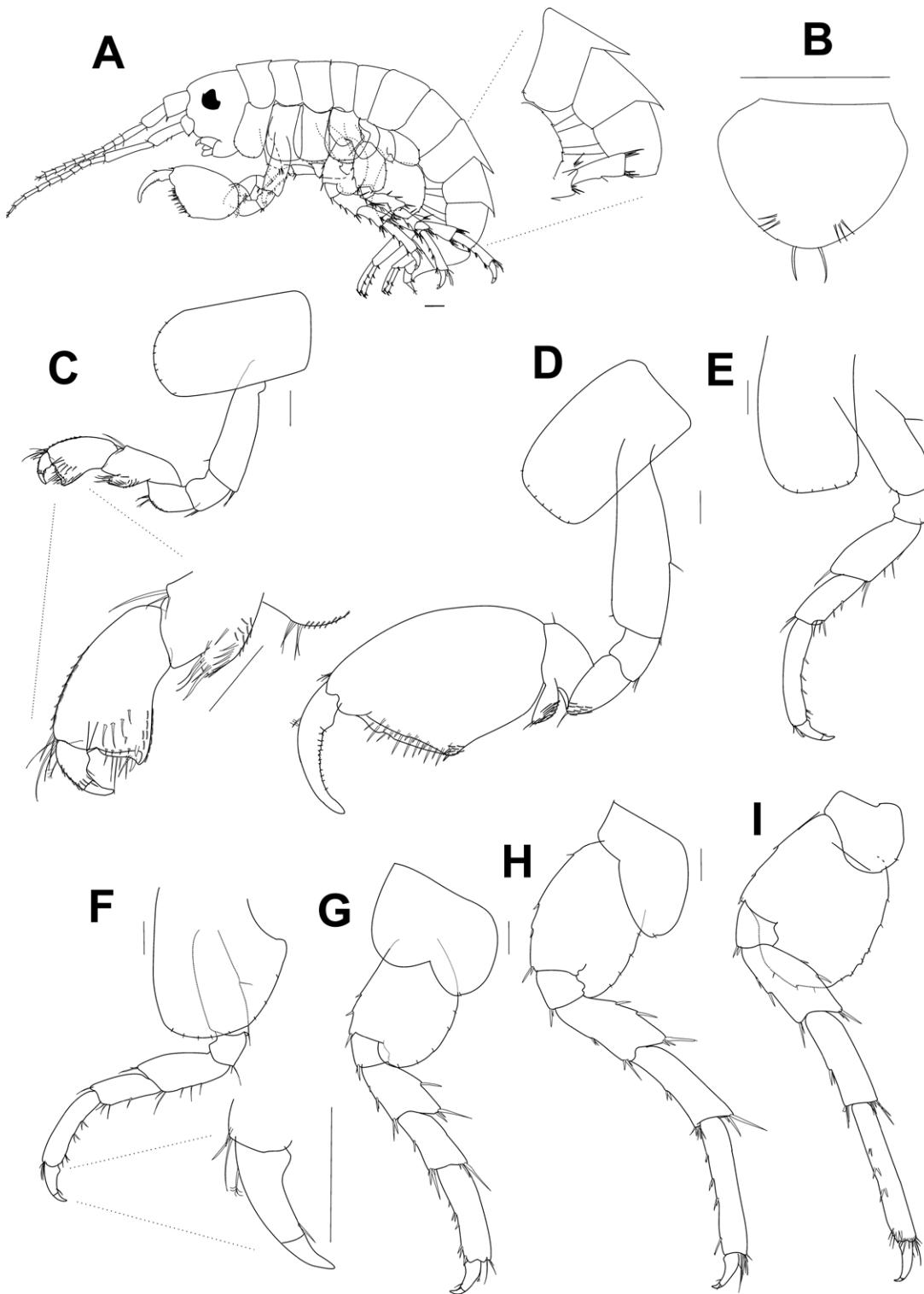


Figure 2. *Hyalella maya* n. sp., paratype, male (3.85 mm). A) complete body and mucronations on pereionites 1-2; B) telson; C) gnathopod 1; D) gnathopod 2; E-I) Pereiopods 3-7. Scale bars = 100 microns.

ther, anterior end of the inner margin; the second article has numerous simple setae in the inner margin and two in the outer margin; the third article has five simple setae at the distal, inner margin and eight simple setae at the apicolateral margin; the fourth article unguiform, distal end with three simple setae, shorter than the nail; nail reaches approximately half as long as the fourth article.

Gnathopod 1 (Figs. 2C, 6B): sub-chelate, hammer-shaped, and shorter than gnathopod 2; basis is elongate, approximately 3.5 times longer than wide; posterior margin is without setae; ischium is short, with maximum width and length subequal to the length of the merus, distal posterior end, and with two simple setae; merus is longer than wide, shorter

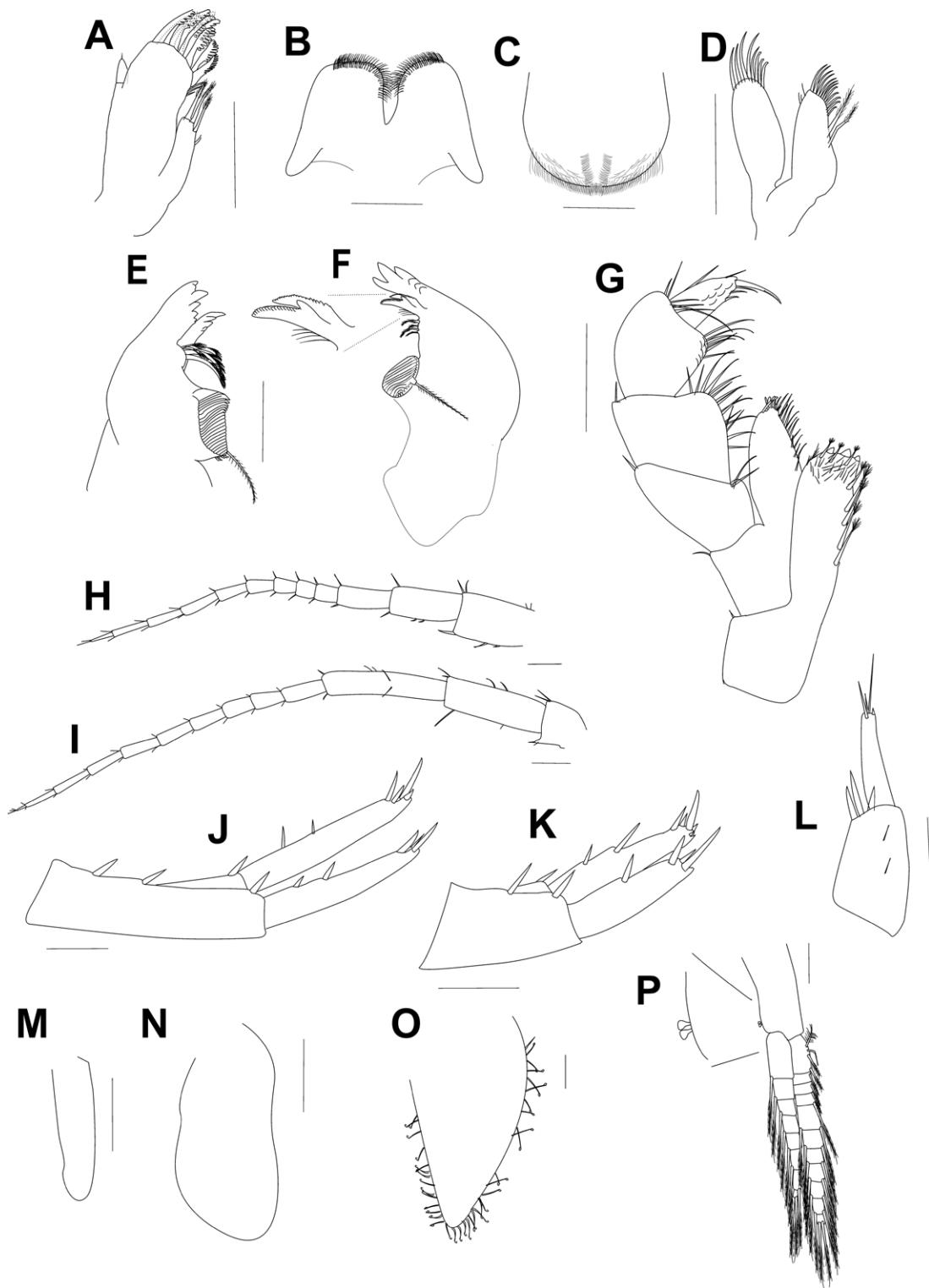


Figure 3. *Hyalella maya* n. sp., paratype, male (3.85 mm). A) maxilla 1; B) lower lip; C) upper lip; D) maxilla 2; E) left mandible; F) right mandible; G) maxilliped; H) antenna 1; I) antenna 2; J-L) uropods 1-3; M) sternal gills; N) coxal gills; O) oosteguities; P) pleopods showing the retinacula. Scale bars = 100 microns.

than the carpus, distal posterior, margin end with simple setae, and comb scales on mid-posterior surface (Figs. 2C, 7B-C); carpus is longer than wide, almost as long as the propodus, posterior margin of lobe, with several simple setae, and two simple setae on the medial surface; lobe, posterior surface of both sides has comb scales near the margin (Figs. 7B-C), distal anterior margin has two simple setae; propodus is 1.76 times longer than wide, distal end of anterior margin, with four simple setae, anterodistal and posterodistal margin, with comb scales; inner surface has four stout, simple setae in a row, and below it, a simple, short seta; palm transverse has long setae, posterior end with a robust seta and cup for dactyl, and with a robust seta in the inner face, near the cup of dactyl; dactyl, claw-like, nail is present, anterior end has a plumose seta, posterior margin has simple seta, and apex has comb scales.

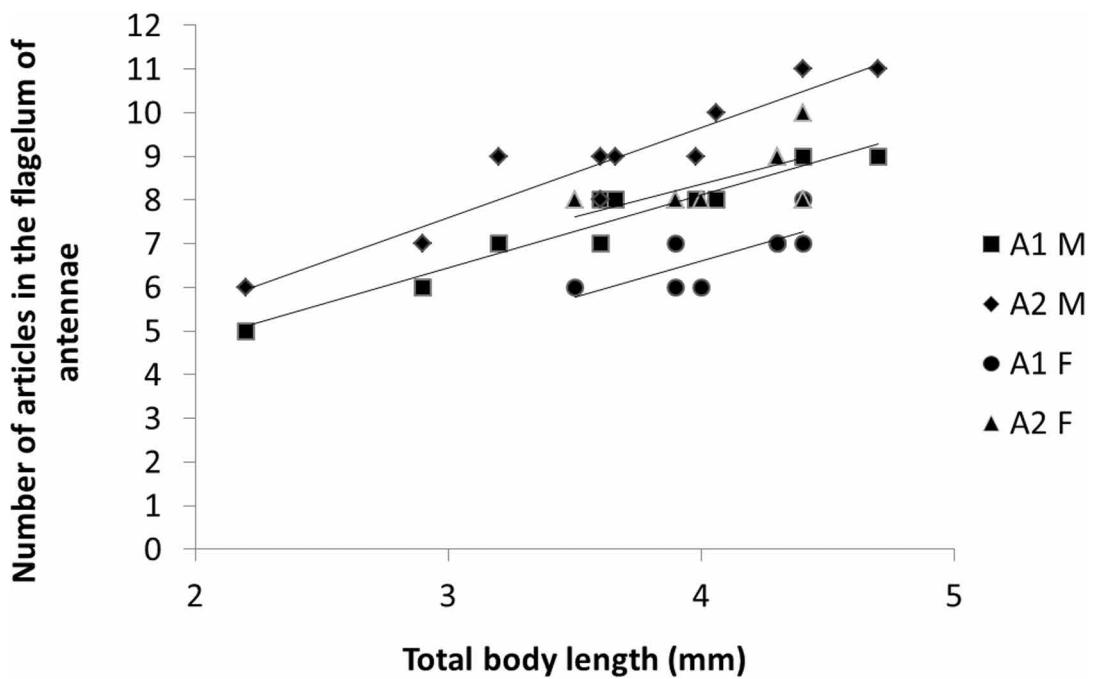


Figure 4. Linear regression between body length and number of articles in the flagellum of the antennae. The males (A1 M and A2 M) have more articles than the females (A1 F and A2 F). The number of articles increases with size of body.

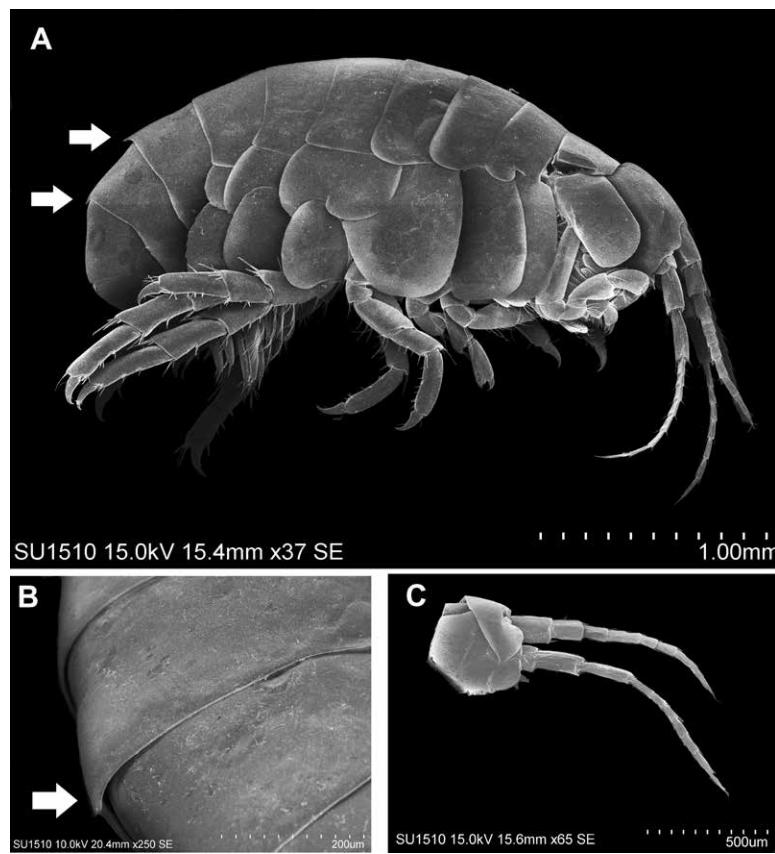


Figure 5. Paratype, ovigerous female (4.15 mm). A) complete body, arrows show the mucronations; B) pleon mucronations, arrow shows the mucronation. Paratype, male (4.10 mm). C) head.

posterior margin has two simple setae; merus is longer than ischium, posterior margin has three simple setae, antero-distal edge has two simple setae, and posterodistal edge has three simple setae; carpus is shorter than merus and longer than ischium; posterior margin of the carpus has two simple setae, antero-distal edge has three simple setae, and posterodistal edge has four simple setae; propodus, posterior margin has two cuspidate setae with an accessory

Gnathopod 2 (Figs. 2D, 6E-F, 7A): sub-chelate, palm is slightly oblique; basis is elongate; posterior margin is almost half its length with a simple seta, anterodistal end with one short simple seta, and distal end of posterior margin with two simple setae; ischium is short, sub-quadrangular, shorter than merus, posterodistal end with three simple setae; merus is short, distal end of the posterior margin with three simple setae, distal half of the posterior surface of the inner and outer surfaces with comb scales; carpus is shorter than propodus, distal end of the anterior margin with two simple setae; posterior lobe is elongate, almost 1.5 times the width of merus, with several sub-marginal, pappose setae; both the inner and outer surfaces of lobe have comb scales; propodus is rectangular and palm is subequal to the posterior margin; slope is slightly oblique, irregular, with several long, simple setae; anterior edge is without any evident, truncated process and without any evident notch at the base (Fig. 7A); posterior distal corner has strong setae and cup for dactyl; dactyl are claw-like, congruent with palm, and without comb scales.

Pereopods (Figs. 2E-I): simple, gradually longer posteriorly, and pereopod 5 is shorter than fourth and sixth. Pereopod 3, basis is elongate with a simple seta at half the length of the posterior margin, anterior distal end with a short simple seta, and distal posterior end has two simple setae; ischium is sub-quadrangular, and distal end of

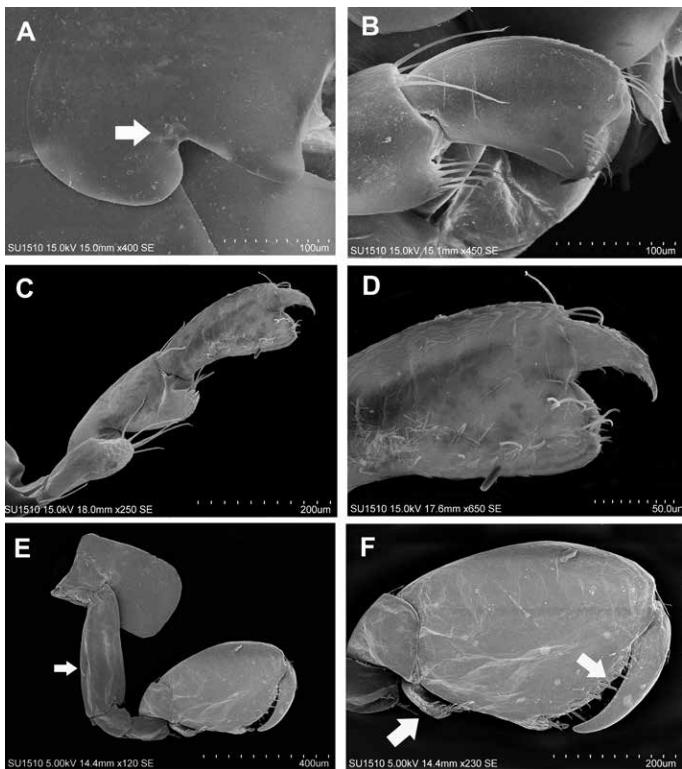


Figure 6. Paratype, ovigerous female (4.15 mm). A) arrow shows the copulatory notch on pereonite 2; B) gnathopod 1 (female); C) gnathopod 2 (female); D) propodus and dactyl, gnathopod 2 (female). Paratype, male (4.10 mm). E) gnathopod 2 (male), arrow shows the posterior setae on the basis; F) propodus and dactyl, gnathopod 2 (male), arrow shows carpus posterior lobe with comb setae and the lack of an evident notch in the palm.

seta, and anterodistal and posterodistal end with three simple setae; dactylus has a claw-like, nail present, at the first, proximal third of the anterior margin with a plumose seta; posterior margin has a simple seta close to the nail (Figs. 2F, 8F). Pereopod 4 is similar in shape to pereopod 3, but longer; coxa is wider than coxa 3 with a posterior excavation; basis is almost at half-length of the posterior margin, with one seta. Pereopods 5-7 are similar in shape; coxa of pereopod 5 is wider than long with two unequal lobes and posterior lobe is slightly longer; coxa of pereopod 6 has no anterior lobe and posterior lobe reaches half as long as the basis; coxa of pereopod 7 is short, with no anterior lobe. Basis of pereopods 5-7 have a rounded and denticulate posterior lobe; lobe of pereopod 7 basis is widely expanded and posterior margin has nine setae.

Pleopods (Fig. 3P): 1-3 are not modified, biramous and elongate, with numerous long plumose setae; peduncle's inner margin has two short retinacula (coupling hooks).

Uropods (Figs. 3J-L): uropod 1 is longer than uropod 2; length of peduncle is longer than rami, proximal half of dorsal margin has two stout, simple setae; ramus has two dorsal setae, three simple ones and one connate distal seta; inner ramus is scarcely longer than outer ramus, without curved seta. Uropod 2 has a sub-rectangular peduncle; length is longer than rami, dorsal margin is almost at distal half with a stout, simple seta and distal end with an apical, simple seta; inner ramus is slightly longer than outer one; dorsal margin has two stout setae and four distal, stout setae; outer ramus has dorsal margin with a simple, stout seta and distal end with three simple, stout setae. Uropod 3's total length is subequal to the peduncle length of uropod 2; peduncle is slightly longer than ramus with distal end having three robust setae; ramus styliform its apex truncate with three apical, simple setae and one connate seta.

Telson (Fig. 2B): slightly wider than long, entire, fleshy, and smooth dorsal surface with three short, sub-marginal plumose setae on both sides (left and right), reaching the distal margin; distal margin is rounded with two separated, apical setae.

Gills. Coxal gills are small, simple, and saclike, on segments 2-7 (Fig. 3N).

Sternal gills are tubular, on pereonites 3-7 (Fig. 3M).

Female (Figs. 5-9) differences: Size is 4.15 mm; Antennae 1 and 2 have flagellum with fewer articles (Figs. 4, 5A). Gnathopod 1 has similar size and shape to gnathopod 2; gnathopod 2, propodus is smaller and more slender than the

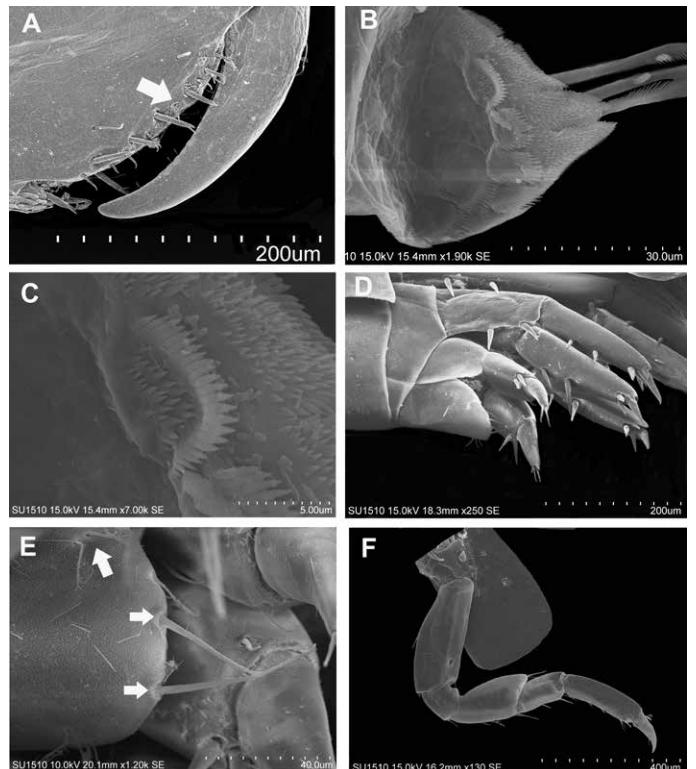


Figure 7. Paratype, male (4.10 mm). A) palm, arrow shows the absence of an evident notch; B) ventral face, carpus posterior lobe with comb scales; C) comb scales on the carpus posterior lobe; D) uropods 2-3. Paratype, ovigerous female (4.15 mm). E) telson, arrows show the two distal separated setae and the submarginal setae; F) pleopod 3.

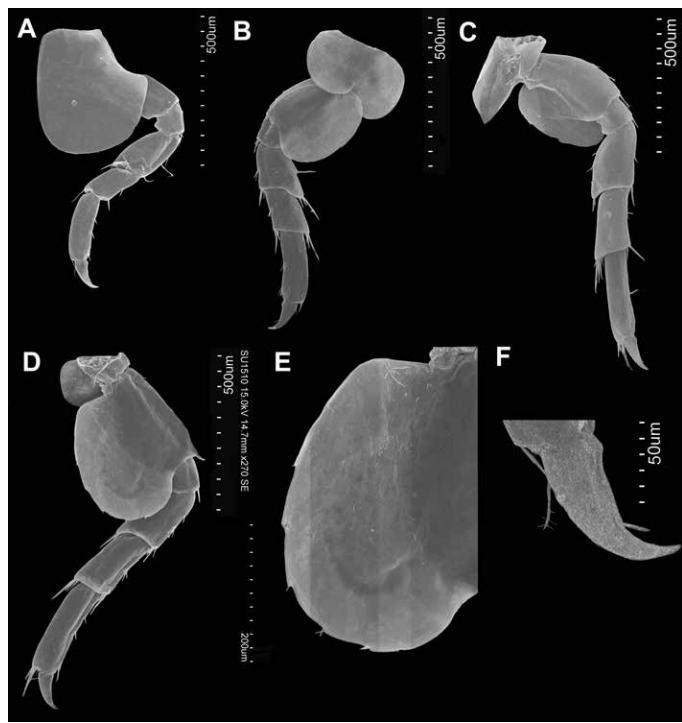


Figure 8. Paratype, ovigerous female (4.15 mm). A) pereopod 4; B) pereopod 5; C) pereopod 6; D) pereopod 7; E) posterior lobe, pereopod 7; F) dactylus and nail close up, pereopod 4.

García-Schroeder and Araujo (2009), said that the number of articles in the flagellum of the antennae increases with the size of the animal. In the redescription of González and Watling (2002), *H. azteca* is larger than the species described herein, but *H. maya* n. sp. had a larger number of articles in the flagellum; likewise, in the two pairs of antennae, the number of segments increases with the size of the specimen and differs between males and females (Fig. 4). Antenna 2 in *H. azteca* is less than half the length of the total length of body, while in *H. maya* n. sp. it is longer than half the total length of body (Fig. 2A). Other important differences are: the shape of the palp of maxilla 1, in *H. azteca* it is rounded and in *H. maya* n. sp. it is shorter and rounded with a distal seta (Figs. 3A; 9D); *H. maya* n. sp. has one seta on the posterior margin of the basis of gnathopod 2 (Fig. 6E), while *H. azteca* has two. The relationship between the length of the palm and the length of the posterior margin is another character that distinguishes *H. maya* n. sp.: the first length is similar to the second, while in *H. azteca*, the length of the palm is shorter than the posterior margin; in *H. maya* n. sp. the telson is described as being wider than long, with a rounded apex and a pair of apical setae widely separated (Fig. 7E), while in *H. azteca*, the width is equal to the length, the apical setae are apposed, and the margin is pointed (Table 2); finally, in *H. maya* n. sp. a smaller number of setae are present in the basis of the lobe of pereopods 5-7 (Figs. 8B-E), compared to *H. azteca*.

male, length to the dactyl is almost twice the maximum width, parachelated, and palm reverse oblique (Figs. 6C-D). Pereonite 2 has an anterior excavation or notch for the amplexus (Fig. 6A).

Oostegites foliaceous, setae end in a curl on the margin of pereonites 2-7 (Fig. 3O).

Remarks: The lack of clear morphological characters that could help distinguish the different species in the *H. azteca* complex make it difficult to identify them accurately. That is why it is considered a complex with cryptic phenotypes by González and Watling (2002), who declared that the characters that help distinguish the species of this complex are: the relative length of the antenna, the number of setae in the internal plate of maxilla 1, the setation in the palp of the maxilliped, the number and organization of the setae of the propodus of gnathopod 1, the setation of the posterior margin of the basis, the propodus shape and the irregular shape of the palm in gnathopod 2 of the males, the shape of the epimeral plates, the setation and proportions of the ramus and the peduncle of uropod 3, and, finally, the shape and setation of the telson.

Hyalella maya n.sp. is morphologically close to *H. azteca*. However, important differences are present. Geisler (1944), Stevenson and Peden (1973) and

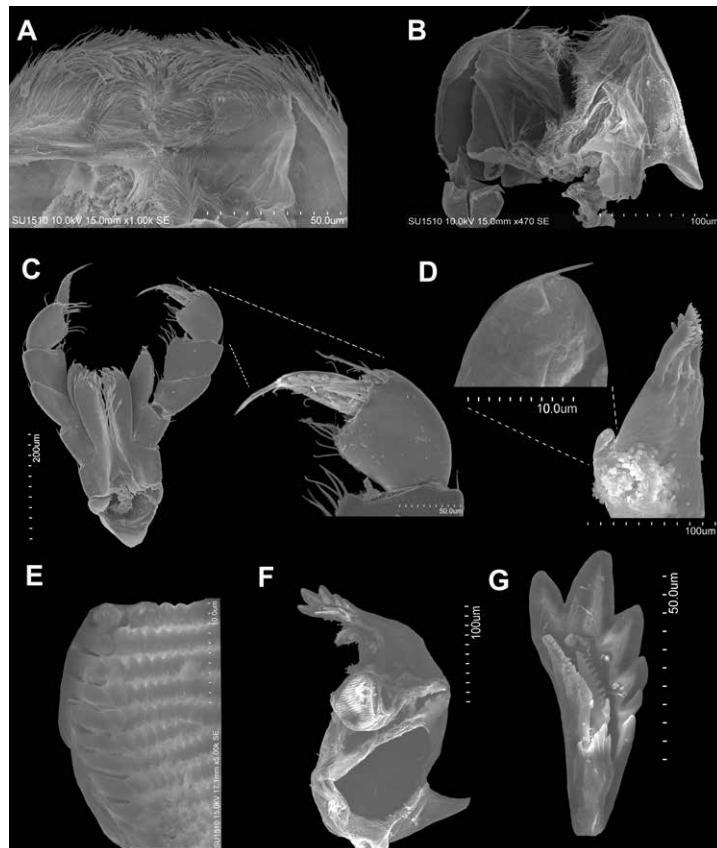


Figure 9. Paratype, ovigerous female (4.15 mm). A) upper lip; B) lower lip; C) maxilliped with a close-up of the third and fourth article and the nail of the peduncle; D) maxilla 1, close-up to the palp with an apical seta; E) molar process; F) right mandible; G) close-up to the incisor process and lacinia mobilis.

Table 2. Morphological comparison between *Hyalella maya* n. sp., *H. spinicauda*, *H. wellborni* and *H. azteca* redescription (HBL = half body length, FTTS = First two thoracic segments, PM = Posterior margin, W = width, L = length).

Morphological Characteristics	<i>H. azteca</i> (redescription of González and Watling, 2002)	<i>H. spinicauda</i> Soucek and Lazo-Wasem, 2015	<i>H. wellborni</i> Soucek and Lazo-Wasem, 2015	<i>Hyalella maya</i> n. sp.
Size (mm)	7.8	5.76	4.28	3.85
Length of antenna 2	A2 < HBL	A2 < HBL (30%)	A2 < HBL (40%)	A2 ≥ HBL (>40%)
No. articles flagellum antenna 1	7	<9
No. articles flagellum antenna 2	8	<10
Relation between length of head and two first thoracic segments	H < FTTS	H < FTTS	H < FTTS	H < FTTS
Maxilla 1 outer plate, number of pappose setae	3	2	2	3
G1 carpus, inner face, pappose setae	4	3	4	3-4
G2 comb scales	present			present
G2 basis, posterior margin, setae	2	...	2	1
G2 propodus; relation between palm length and posterior margin	P < PM	P < PM	P = PM	PPM = PM
G2 propodus palm	notch	Angled step	No distinct step/notch	No distinct step/notch
Telson, relation between width and length	W = L	W < L	W = L	W > L
Telson, setae	Apposed, larger and slender	Separated, short, stout	Apposed, larger, slender	Separated, larger slender
Telson margin	pointed	rounded	rounded	rounded

Soucek et al. (2015) examined the lectotype from the type material of De Saussure (1858) and described two new species from Canada and the U.S.A., *H. wellborni* and *H. spinicauda*. In this study, we analyzed useful morphological characters for the identification of the species of the complex *H. azteca*, such as: the number of setae in maxilla 1, the shapes of gnathopod 2, pereopod 7, uropod 3 and telson, in accordance with Soucek et al. (2015). *Hyalella maya* n. sp. can be distinguished from both species, *H. wellborni* and *H. spinicauda*, because of the presence of three setae in the internal plate of maxilla 1, while in *H. wellborni* and *H. spinicauda* only two are present. In their description, the authors mentioned that the male gnathopod 2 in *H. azteca* has a wide truncate process with a notch, an important difference between *H. wellborni*, *H. spinicauda* and *H. maya*. In *H. maya*, the notch is absent and the truncate process is not evident (Table 2).

In this locality, i.e. Cenote Aktun-Ha, another species of the same Genus: *Hyalella cenotensis* has already been described by Marrón-Becerra et al. (2014). The main differences between the two species are the absence of eyes and of distal setae in the telson in *H. cenotensis*, both present in *H. maya* n. sp. In addition, in *H. maya* n. sp., the relative length of antenna 2, with respect to antenna 1, is bigger (more than 1.3 times), while in *H. cenotensis* it is less than 1.2 times. Another difference is that in the flagellum of antennae 1 and 2, *H. cenotensis* bears 7 and 9 articles, respectively, although the specimens are larger (5.3 mm) than *H. maya* n. sp. (4.15 mm), where a maximum of 9 and 10 articles are present. Finally, the relative length of the ramus in uropod 3 in *H. maya* n. sp. is longer than the peduncle, while in *H. cenotensis* it is slightly shorter, close to the length of the longest seta of the peduncle.

According to González and Watling (2002), due to the scarce variation in the characters of the *H. azteca* complex, the telson setation and the relative length of the antennae are important to distinguish among different species. In addition, Soucek et al. (2015) suggested to use the relative length of uropod 3 with respect to the peduncle and its longest seta to distinguish the species of the complex. This character, together with others, was useful to separate two species from the U.S. and Canada (*H. wellborni* and *H. spinicauda*), and those identifications were later confirmed with the analysis of the mitochondrial cytochrome oxidase gene, subunit I (COI).

The recent description of two species of epicontinental amphipods in the same sinkhole in Mexico could mean that we need more samplings to study adequately the amphipods in these environments.

Key to the species of *Hyalella* (*Hyalella*) in North America and the Caribbean region (Modified from Baldinger, 2004; Marrón-Becerra et al., 2014; Soucek et al., 2015).

1. Eyes absent 2
 - Pigmented eyes present 3
2. Antenna 1 is longer than antenna 2; sterna gills on pereonites 3-7; telson with four distal setae *H. muerta*
 - Antenna 1 shorter than antenna 2; sterna gills on pereonites 2-7; telson without distal setae *H. cenotensis*
3. Body with dorsal mucronations 8
 - Body without dorsal mucronations 4
4. Ramus of uropod 3 is vestigial or robust, subequal or shorter than peduncle 5
 - Ramus of uropod 3 slender, subequal or longer than peduncle 6
5. Ramus of male uropod 3 is robust with seven apical spines *H. sandra*
 - Ramus of male uropod 3 vestigial with two to four spines *H. meraspinosa*
6. Antenna 1 and 2 are subequal in length 7
 - Antenna 2 is nearly twice the length of antenna 1 *H. longicornis*
7. Hind margin of merus of pereopods 3 and 4 has long setae, telson with two closely apical setae *H. caribbeana*
 - Hind margin of article 4 of pereopods 3 and 4 with short setae, telson with two long, broadly-spaced, apical setae *H. inermis*
8. Inner plate of maxilla 1 is narrow, with two to five apical plumose setae 9
 - Inner plate of maxilla 1 is broad, subtriangular with two or three apical plumose setae, followed closely by 22-30 similar medial setae *H. montezuma*
9. Antenna 1 is longer than half the length of antenna 2, and only first, or first two abdominal segments bearing dorsal mucronations 10
 - Antenna 1 is less than half the length of antenna 2, with all three abdominal segments bearing dorsal mucronations *H. texana*
10. Gnathopod 2 of males, carpus posterior lobe is about as long as width of merus; in pereopod 7, the distal/ bottom margin of basis posterior lobe, dentate or not, with one or two very small setae if any ... 11
 - Gnathopod 2 of males, carpus posterior lobe 1.5 times as long as width of merus; pereopod 7, distal/bottom margin of the basis posterior lobe is strongly dentate, and with two or more relatively long spines *H. azteca*
11. Gnathopod 2 propodus (males), palm with a distinct angle step (visible under high power), tip of dactyl approximately aligns vertically with distal end of posterior lobe of carpus; telson distal setae is separated, short, and at least as stout as setae on uropod 3 ramus *H. spinicauda*
 - Gnathopod 2 propodus (males), palm without a distinct angle step or notch, tip of dactyl aligning vertically well beyond (posteriorly) distal end of posterior lobe of carpus; telson terminal setae clearly finer and longer than setae on uropod 3 ramus 12
12. Telson with two long and slender apposed setae; uropod 3 ramus, approximately as long as or slightly longer than the longest seta on peduncle; pereopod 7, posterior lobe, ventral margin without stout spines; maxilla 1, inner plate with two pappose setae; maxilliped nail short (less than half the length of palp article 4) *H. wellborni*
 - Telson with two long and slender setae widely separated; uropod 3 ramus longer than the longest setae on peduncle; pereopod 7, posterior lobe, ventral margin with one stout setae; maxilla 1, inner plate with three pappose setae; maxilliped nail is long (longer than half length of palp article 4) *H. cenotensis* n. sp.

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