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Research article

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New species of *Macunahyphes* Dias, Salles & Molineri (Ephemeroptera: Leptohephidae), with taxonomic notes

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Abstract. *Macunahyphes zagaia* sp. nov. and *M. araca* sp. nov. are described based on imagines collected from the Amazon biome in Brazil. Specimens belonging to all species of the genus were examined and a diagnosis for each one is provided. Variation seen in the male imago of *M. eduardoi* is described, as well as the female and egg for the first time. Together with *M. eduardoi*, the two new species share the presence of a basal swelling on segment II of the forceps, which raises important questions concerning the evolution of this character in Leptohephidae. Finally, a re-definition of the genus is proposed.

Keywords. Taxonomy, mayfly, South America, Amazon Rainforest, aquatic insect.

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Introduction

The genus *Macunahyphes* Dias, Salles & Molineri, 2005 was erected to include *Macunahyphes australis* (Banks, 1913) originally described by Banks (1913) in the genus *Tricorythus* Eaton, 1868. This species, described solely on adults, was subsequently transferred to *Leptohephodes* Ulmer (Ulmer 1920) and then to *Tricorythodes* Ulmer (Traver 1958). With the discovery and subsequent description of the nymphs, *T. australis* was finally transferred to the new genus *Macunahyphes* (Dias *et al.* 2005). Among the distinctive characteristics of this species, Traver (1958), as well as Molineri (2002), pointed out the absence of the basal swelling of the second joint of the forceps, absence of the longitudinal vein CuP on male fore wings, and the morphology of the penis, with the presence of a ventral projection.

Based on adults from Venezuela and Brazil, respectively, Molineri *et al.* (2011) described two additional species: *M. pemonensis* Molineri *et al.* and *M. incognitus* Molineri *et al.* Despite the absence of forceps in the few available specimens of both species, the presence of a ventral projection on the penis, wing venation and similar color pattern were enough to allocate them to the genus *Macunahyphes* (Molineri *et al.* 2011).

Recently, two additional species were found in Brazil: *M. pemonensis*, recorded from Northern Brazil based on a larger series of male adults, and *M. eduardoi* Almeida & Mariano, 2015, based on male adults from Northeastern Brazil (Belmont *et al.* 2015; Almeida & Mariano 2015). Importantly, forceps of both species were unbroken and the basal swelling, absent in *M. australis* and considered one of the striking characteristics of the genus, was found to be present in *M. eduardoi* and *M. pemonensis*.

In the present paper, based on material from the Amazon and Atlantic Forest biomes in Brazil, two new species of *Macunahyphes* are described. In addition, specimens belonging to all species of the genus are examined, new data are provided for them and a re-definition of the genus is proposed.

Material and methods

Specimens were preserved in 80% ethanol, wings were mounted dry and genitalia were mounted in Euparal[®]. Photographs were taken with a digital camera coupled to a Zeiss Axiocam ERc 5s stereo microscope and combined using Helicon Focus 6[®] software. Some of the photographs were used as templates for trace vector graphics in Adobe Illustrator CS6[®] software to produce the illustrations. Material from the following institutions was studied: Coleção Zoológica Norte Capixaba, Universidade Federal do Espírito Santo, São Mateus, Brazil (CZNC) and Coleção Entomológica Professor José Alfredo Dutra (DZRJ), Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil. Holotypes and part of the paratypes were deposited in the Coleção de Invertebrados of the Instituto Nacional de Pesquisas da Amazônia, Manaus, Brazil (INPA). The other paratypes were deposited in CZNC and DZRJ. Wings were mounted on dry slides; as all were identical, there was no need to draw them. Eggs were removed from females of *Macunahyphes eduardoi* Almeida & Mariano, 2015 and *Macunahyphes araca* sp. nov., then mounted with double sided tape on SEM stubs and sputter coated with gold. They were observed and photographed with a Jeol JSM-6510 scanning electron microscope. The terminology proposed by Koss & Edmunds (1974) was used to describe the eggs. The distribution map was made using the program QGIS 2.12.0-Lyon (QGIS 2015).

Results

Class Hexapoda Latreille, 1825
Subclass Insecta Linnaeus, 1758
Order Ephemeroptera Hyatt & Arms, 1890
Suborder Pannota McCafferty & Edmunds, 1979
Family Leptohyphidae Landa, 1973

Macunahyphes Dias, Salles & Molineri, 2005

Macunahyphes Dias, Salles & Molineri, 2005: 196 (type species: *Tricorythus australis* (Banks, 1913), by monotypy).

Diagnosis

Genus characterisation: 1) eyes undivided in both sexes; 2) fore wings with well-developed Cu-A lobe, especially in males; 3) longitudinal vein CuP poorly developed in both sexes or absent; 4) hind wings absent in both sexes; 5) membranous filaments of mesoscutellum present or absent; 6) styliger plate

very slightly projected posteriorly as a columnar base for each pair of forceps (see figs 7–9 in Molineri *et al.* 2011) ; 7) basal swelling on forceps segment II present or absent. In the nymph: 1) maxillary palp reduced in size, one-segmented, with apical seta; 2) glossa and paraglossa almost completely fused, with a circular outline; 3) femora with numerous robust serrate setae; 4) tarsal claws long and slender, with 7–10 marginal denticles and a double row of 2–4 submarginal denticles; 5) gills present on abdominal segments II–VI, gill formula 3/3/3/3/2; 6) operculate gills subtriangular; 7) posterior margin of terga II–V smooth, other segments with denticles.

Comments

Due to the presence of membranous filaments on the mesoscutellum in females and males in one species, and to the presence of a basal swelling on forceps segment II in four species, we propose an expansion to the diagnosis of the genus provided by Dias *et al.* (2005) to include characters 5 and 7 above. As *M. australis* is the only species in which nymphs have been described, the generic diagnostic features of the nymphal stage are maintained.

Distribution (Fig. 1)

Argentina, Brazil, Guyana and Venezuela.

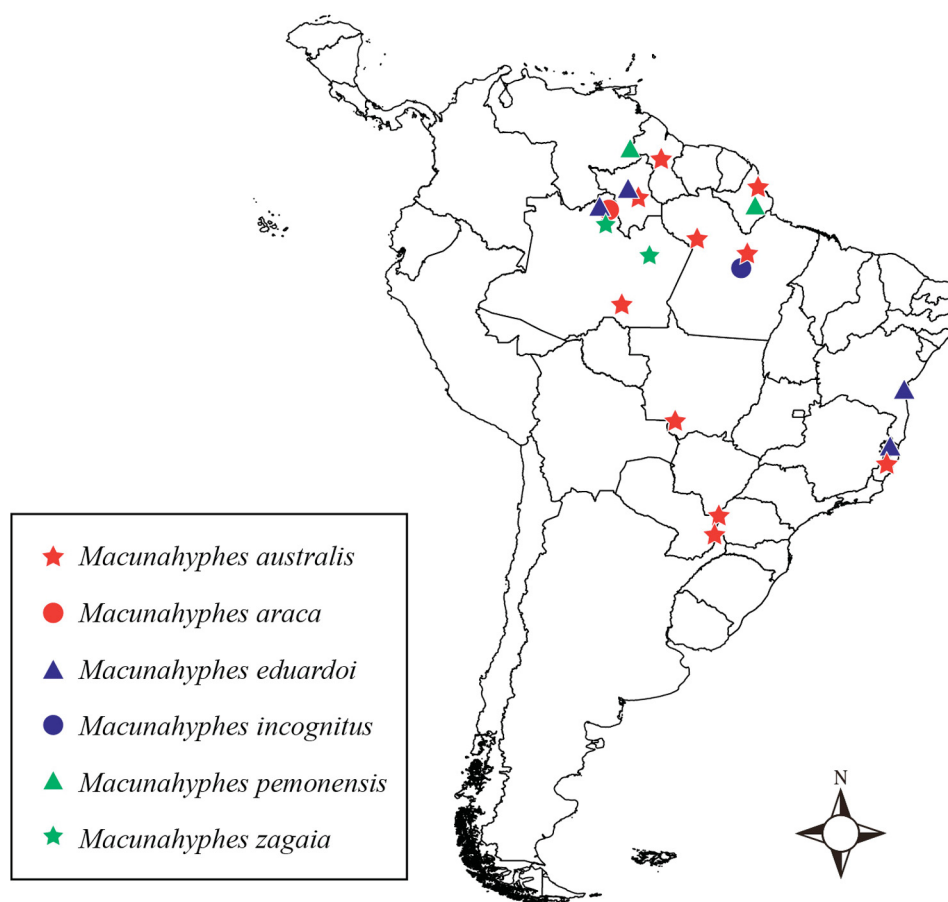


Fig. 1. Geographical distribution of species of *Macunahyphes* in South America and Brazil (per state).

Macunahyphes australis (Banks, 1913)

Fig. 2C

Tricorythus australis Banks, 1913: 85.

Leptohyphodes australis – Ulmer 1920: 50.

Tricorythodes australis – Traver 1958: 501–503. — Hubbard 1982: 274. — Molineri 2002: 278.

Macunahyphes australis – Dias, Salles & Molineri 2005: 199.

Material examined

BRAZIL: Espírito Santo: 113 ♂♂, imagines, Colatina, Itapina, Doce River, 19°31'18.1" S, 40°50'11.1" W, 7–8 Sep. 2014, E.M. Rozário and F.F. Salles leg. (CZNC Ep-6440); 14 ♀♀, imagines, same data (CZNC Ep-6441); 72 ♂♂, imagines, same data (CZNC Ep-6443); 73 ♂♂, imagines, same data (CZNC Ep-6444); 64 ♂♂, imagines, same data (CZNC Ep-6445); 100 ♂♂, imagines, same data (CZNC Ep-6447); 3 ♂♂, imagines, Marilândia, Doce River, 19°31'5.3" S, 40°34'49.9" W, 8–9 Sep. 2014, E.M. Rozário and F.F. Salles leg. (CZNC Ep-6439); 7 ♂♂, imagines, same data (CZNC Ep-6442); 1 ♀, imago, same data (CZNC Ep-6446); IFES Itapina, Doce River, 19°31'15.1" S, 40°46'53.6" W, E.M. Rozário and F.F. Salles leg. (CZNC Ep-6448); 2 ♂♂, imagines, same data (CZNC Ep-6449); 5 ♂♂, imagines, Nova Venécia, city gate, Cricaré River, 18°42'54.91" S, 40°22'33.33" W, 23–24 May 2012, F.F. Salles and K.B. Angeli leg. (CZNC Ep-4523); 1 ♂, 1 ♀, imagines, São Mateus, Cotaxé River, Japira waterfall, 18°34'39.1" S,

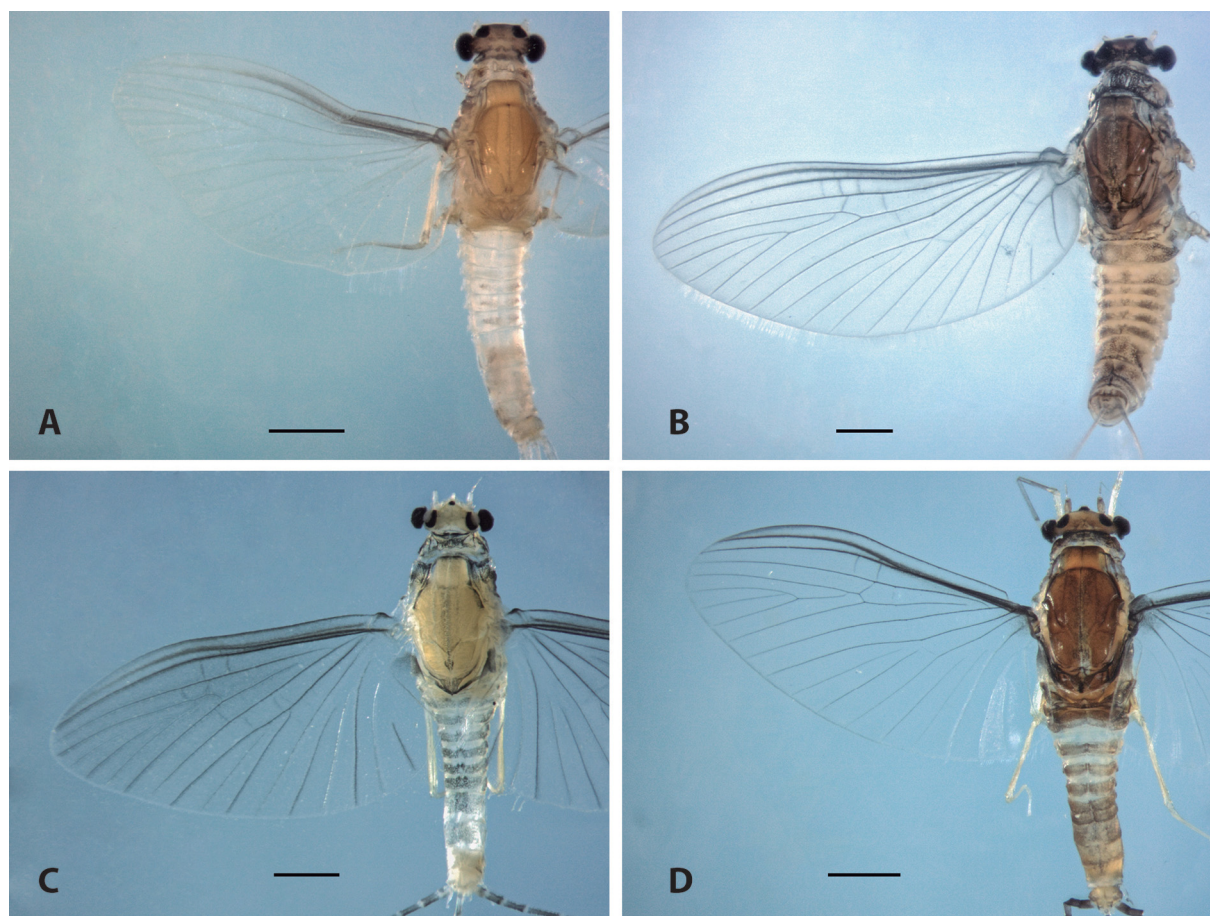


Fig. 2. *Macunahyphes* spp., dorsal view. **A.** *M. zagaia* sp. nov. (♂). **B.** *M. eduardoi* Almeida & Mariano, 2015 (♀). **C.** *M. australis* (Banks, 1913) (♂). **D.** *M. araca* sp. nov. (♂). Scale bars: 0.5 mm.

40°16'58" W, 7–8 Sep. 2012, K.B. Angeli and E.A. Raimundi leg. (CZNC Ep-4524); 1 ♂, imago, São Mateus River, 18°39'17.1" S, 39°59'36" W, 7–8 Sep. 2012, K.B. Angeli and E.A. Raimundi leg. (CZNC Ep-4527); 1 ♂ imago, Pedra d'Água, 18°43'15.2" S, 39°48'50.8" W, 19–20 Apr. 2012, K.B. Angeli and E.A. Raimundi leg. (CZNC Ep-4528); 4 ♂♂, imagines, Cotaxé River, Japira waterfall, 18°34'39.1" S, 40°16'58" W, 6–7 Feb. 2013 (CZNC Ep-6159); 8 ♂♂, 6 ♀♀, imagines, Roraima, Caracarái, Balneário Bem Querer, 01°55'46.3" N, 61°00'06.9" W, 13 Mar. 2014, F.F. Salles, R. Boldrini, E. Dominguez leg. (CZNC Ep-6560); 3 ♂♂, Balneário Bem Querer, 18–21 Nov. 2002, A.M.O. Pes leg. (CZNC Ep-6561).

Diagnosis

1) Longitudinal vein CuP poorly developed in both sexes or absent; 2) membranous filaments of mesoscutellum absent; 3) forceps tri-segmented with first segment, distomedially projecting, 4) penis with very wide base, becoming thinner towards a subapical constriction, and then slightly widening again, penis lobes of each side almost completely fused except apical incision; 5) penis with a ventral projection covered with spines.

As *M. australis* is the only species for which nymphs have been described, the generic diagnostic features of the nymphal stage should also be used for specific diagnosis.

Distribution (Fig. 1)

Argentina (Misiones), Brazil (Amazonas, Espírito Santo, Mato Grosso, Pará, Paraná, Roraima) and Guyana.

Macunahyphes araca sp. nov.

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Figs 2D, 3A, 4A–D

Diagnosis

The male of *M. araca* sp. nov. can be distinguished from all congeners by the following combination of characteristics: 1) longitudinal vein CuP absent; 2) membranous filaments of mesoscutellum absent;

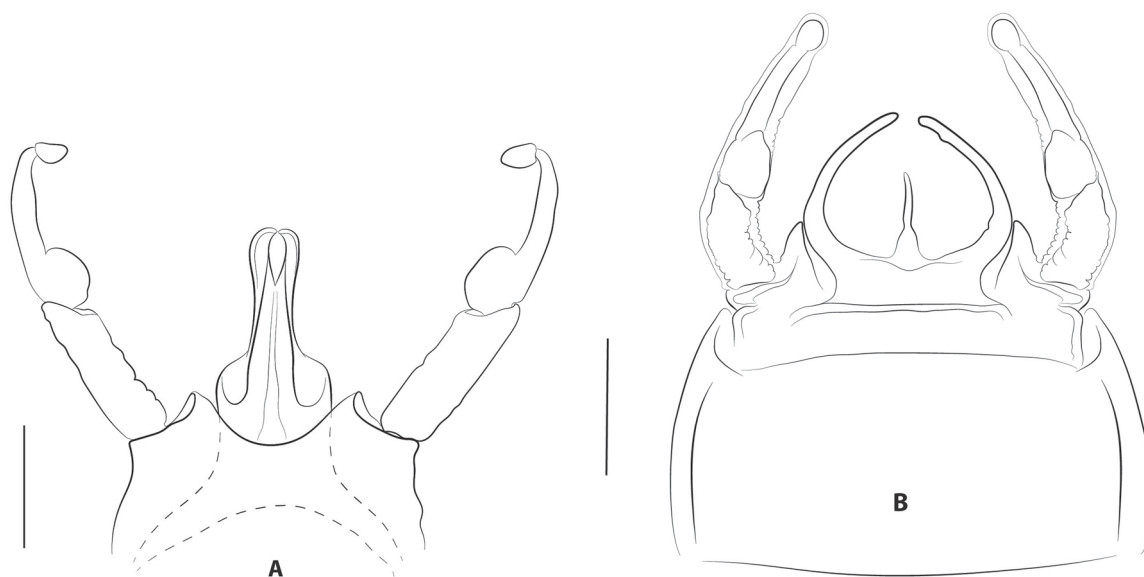


Fig. 3. Male genitalia (ventral view). **A.** *Macunahyphes araca* sp. nov. **B.** *Macunahyphes zagaia* sp. nov. Scale bars: 100 μ m.

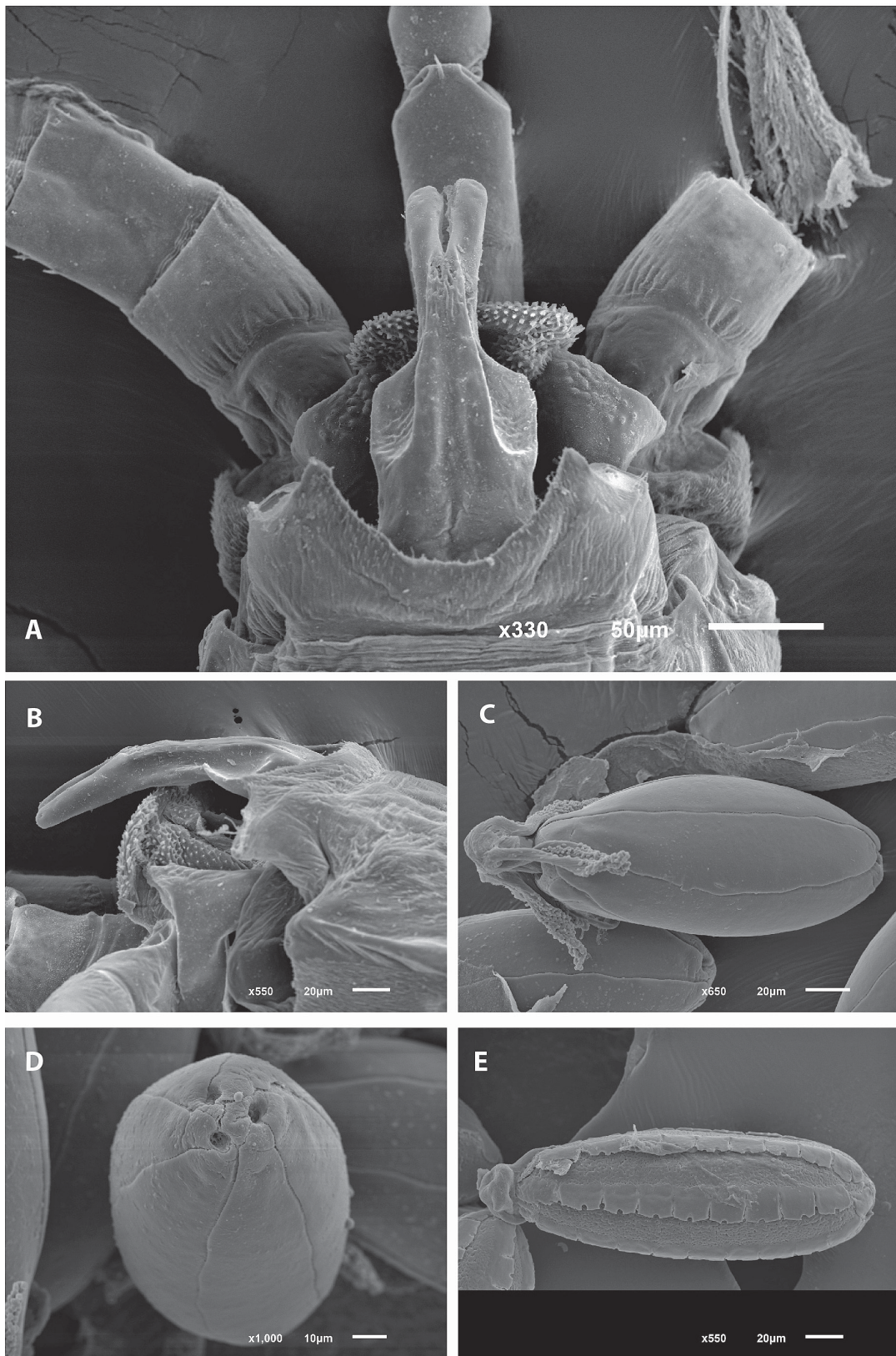


Fig. 4. SEM micrographs. — **A–D.** *M. araca* sp. nov. **A.** Male genitalia (ventral view). **B.** Male genitalia (lateral view). **C.** Egg (general aspect). **D.** Egg showing the micropylar area. — **E.** *M. eduardoi* Almeida & Mariano, 2015, egg (general aspect).

3) segment II of forceps with a basal swelling; 4) spermatic duct dark purple, visible laterally from outside; 5) styliger plate slightly projected posteriorly as a columnar base for each forceps, with sublateral acute projection on hind margin; 6) penis base subquadrate, narrowing abruptly in median zone; 7) penis with deep apical furrow; 8) ventral structure of penis reaching the deep apical furrow.

Etymology

Aracá is the name of the mountain range where this new species was collected.

Type material

Holotype

BRAZIL: ♂, imago, Amazonas, Barcelos, Serra do Aracá, Igarapé do Anta, 0°54'30.38" N, 63°26'24.32" W, 2 Aug. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (INPA-EPH 000001).

Paratypes

BRAZIL: 23 ♂♂, imagines, same data (CZNC Ep-6351); 10 ♂♂, imagines, same data (INPA-EPH 000002 to INPA-EPH 000011); 13 ♂♂, 1 ♀, imagines, Igarapé do Anta, 0°54'30.38" N, 63°26'24.32" W, 1–3 Aug. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (DZRJ 3143); 1 ♂, imago, Jauari River, 0°48'0.28" N, 63°29'22.92" W, 21 Jul. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (CZC Ep-5585); 1 ♂, imago, same data (CZNC Ep-6354); 5 ♂♂ imagines, Igarapé da Serrinha, 0°25'19.96" N, 63°23'47.04" W, 28 Jul.–1 Aug. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (CZNC Ep-6353); 1 ♂, imago, same data but 19 Jul. 2009 (CZNC Ep-6355); 5 ♂♂, 1 ♀, imagines, Igarapé do Anta, 0°54'30.38" N, 63°26'24.32" W, 1 Aug. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (CZNC Ep-6356).

Description

Holotype

LENGTH (mm). Body: 2.8; fore wing: 2.6; cerci: 5.8; terminal filament: 7.5. General coloration brown.

HEAD. Yellowish brown; antennae brown.

THORAX. Pronotum translucent, washed with black; mesonotum brown, washed with black, except on lateral region of anterotal protuberance and submesoscutum; metanotum yellowish brown, washed with black; membranous filaments of mesoscutellum absent.

FORE WING. Membrane translucent; base and coastal region washed with dark purple; longitudinal veins black; vein CuP absent.

LEGS. Yellowish, inner apex of trochanter tinged with black; fore tibia and fore tarsi slightly washed with dark purple.

ABDOMEN. Terga II–X with medio-longitudinal dark line, terga I–VII translucent, tinged with dark purple, terga VIII–X yellowish brown, washed dark purple; tergum X with medial dark furrow; spermatic duct dark purple, visible from outside.

GENITALIA. Forceps translucent, washed with dark purple, except on inner margin of segment I, styliger plate and penes translucent yellowish; styliger plate slightly projected posteriorly as a columnar base for each forceps, with sublateral acute projection on hind margin; forceps segment I weakly attached to styliger plate, basal swelling present on segment II; penes long, with subquadrate base, deep apical furrow and a somewhat protruded ventral structure. Caudal filaments translucent, tinged with dark purple at base, becoming lighter towards apex.

Female imago

LENGTH (mm). Body: 3.0; fore wing: 3.0. General coloration brown.

HEAD. Yellowish brown, washed with black, posterior margin darker; scape and pedicel dark brown, flagellum whitish.

THORAX. Pronotum tinged with black, lateral corner brown, washed with black; mesonotum brown, washed with black; metanotum yellowish brown, washed with black; membranous filaments of mesoscutellum absent.

FORE WING. Membrane white, longitudinal and cross-veins black; vein CuP absent.

LEGS. Yellowish white.

ABDOMEN. Translucent, tinged with black; caudal filaments broken off.

Egg (Figs 4C–D)

Yellow; single polar cap, attachment structures with tubercles on surface; three micropylar openings present at opposite pole of polar cap; chorionic surface without sculpture and with six elongated and linear micropylar canals ending at micropylar opening.

Biology

The specimens were collected at light traps during the dry season. The new species was found exclusively at the Serra do Aracá, together with *M. eduardoi* and *M. zagaia* sp. nov. *Macunahyphes araca* sp. nov. was the only species of mayfly found at the base and at the top of the Serra do Aracá. The Serra do Aracá is an elevated area (100 to 1500 m a.s.l.) in the State of Amazonas near the borders of Roraima State and Venezuela. Only five species of mayflies were collected at the top of Serra do Aracá during the same field trip; all of them were new to science at that time. In addition to the *Macunahyphes* species cited above, the other two species were *Askola yanoman* Nascimento, Barcelos-Silva & Salles, 2011 and an undescribed species, also found exclusively at the top, suggesting a high level of endemism in this area.

Remarks

Macunahyphes araca sp. nov. and *M. pemonensis* are the darker species of the genus. *Macunahyphes araca* sp. nov. can also be distinguished from all other species by the unique spermatic duct, which is visible laterally from outside due to its dark purple coloration, a characteristic also found in *M. pemonensis* (see Belmont *et al.* 2015: figs 1, 6). As in *M. pemonensis*, the styliiger plate is slightly projected posteriorly as a columnar base for each forceps, with a sublateral acute projection on the hind margin, unlike the one found in *M. incognitus*. On the other hand, in the new species the penis base is subquadrate, narrowing abruptly in the median zone, as found in *M. incognitus*. Despite the very similar genitalia of *M. araca* sp. nov. and *M. incognitus*, the ventral structure of the new species reaches the deep apical furrow, while the ventral structure of *M. incognitus* ends before the apical furrow and the penis lobes are almost completely fused. According to the original description, *M. incognitus* seems to be similar to *M. australis* regarding the overall coloration, lighter in comparison to *M. araca* sp. nov.

Distribution (Fig. 1)

Brazil (Amazonas).

Macunahyphes eduardoi Almeida & Mariano, 2015

Figs 2B, 4E

Macunahyphes eduardoi Almeida & Mariano, 2015: 498.

Diagnosis

According to the original description, the male of *M. eduardoi* can be distinguished from all congeners by the following combination of characteristics: 1) fore wing translucent, with costal and subcostal area tinged with grey and with longitudinal vein CuP absent; 2) penes opaque yellow, spine elongate with lateral projections.

Material examined

Holotype

BRAZIL: ♂, imago, Bahia, Igrapiúna, Michelin Ecological Reserve, Pacangê River, Pacangê-Sapucaia 2ª Ponte, 13°50'17.1" S, 39°14'27.7" W, 101 m, 21 Sep. 2012, A.R. Calor and Equipe LEAq leg. (CZNC).

Other material

BRAZIL: 1 ♂, imago, Espírito Santo, Nova Venécia, Santa Rita do Pipinuck, Rio Cricaré, 18°39'51.4" S, 40°30'44.9" W, 25–26 Jul. 2012, K.B. Angeli leg. (CZNC Ep-4561); 2 ♂♂, imagines, same data but 18 Apr. 2012 (CZNC Ep-6431); 4 ♂♂, 3 ♀♀, imagines, Amazonas, Barcelos, Serra do Aracá (Base), tributary of Igarapé do Cobra, 00°52'13.22" N, 63°27'13.36" W, 25–26 Jul. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (CZNC Ep-6437); 1 ♂ imago, Jauari River, 00°48'0.28" N, 63°29'22.92" W, 26 Jul. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr. leg. (CZNC Ep-6438); 1 ♂, 3 ♀♀, subimagines, Roraima, Caracaraí, Balneário Bem Querer, 01°55'46.3" N, 61°00'06.9" W, 13 Mar. 2014, F.F. Salles, R. Boldrini, E. Dominguez leg. (CZNC Ep-6562).

Description

Female subimago (Fig. 2B)

LENGTH (mm). Body: 3.0–4.8; fore wing: 3.2–5.0. General coloration dark brown and yellowish.

HEAD. Brown, washed with black; area between lateral ocelli and posterior margin tinged with black.

THORAX. Pronotum brown, tinged with black, except for submedial stripe and anterolateral corner; mesonotum brown, washed with black; metanotum yellowish, washed with black; membranous filaments of mesoscutellum present and longer than in males.

FORE WING. Membrane white, longitudinal and cross-veins black; vein CuP absent.

ABDOMEN. Translucent (yellow because of the presence of eggs), washed with black; terga II–V forming a V-shaped unpigmented area; caudal filaments broken off.

Egg (Fig. 4E)

Yellow. Shape elongate. One polar cap present. Chorion with longitudinal costae formed by plates, each one with a deep emargination.

Remarks

In the description of the male imago of *M. eduardoi* there is no reference to the presence of membranous filaments of the mesoscutellum; this character is evident from the fresh material studied here. After examining the holotype of *M. eduardoi* it was possible to observe the presence of a short filament of about ¼ the length of the mesoscutellum. According to the original description, the hook-shaped lateral

expansions present in the penis projection are distributed from its base to the middle region, whereas in the material examined by us the expansions are present throughout the penis projection, decreasing in size towards the apex. This is the first record of *M. eduardoi* from southeastern (state of Espírito Santo) and northern (state of Amazonas) Brazil.

Macunahyphes eduardoi occurs in areas comprising two disjunct biomes, Amazon and Atlantic Forest. Other species of plants and animals also show this distribution pattern, including some aquatic insects, such as *Asthenopodes chumuco* Molineri, Salles & Peters, 2015 (Ephemeroptera: Polymitarcyidae), *Macrostemum erichsoni* (Banks, 1920) and *Smicridea (Ryachophylax) roraimense* Albino, Pes & Hamada, 2011 (Trichoptera: Hydropsychidae). The presence of disjunct populations of a single taxon in Amazonia and the Atlantic Forest may be evidence of ancient connections between these biomes (see Costa 2003; Santos *et al.* 2007; Fiaschi & Pirani 2009; Buso Junior *et al.* 2013).

Distribution (Fig. 1)

Brazil (Amazonas, Bahia and Espírito Santo).

Macunahyphes incognitus Molineri, Grillet, Nieto, Dominguez & Guerrero, 2011

Macunahyphes incognitus Molineri, Grillet, Nieto, Dominguez & Guerrero, 2011: 45.

Diagnosis

According to the original description, the male imago of *Macunahyphes incognitus* can be distinguished from all congeners by the following combination of characteristics: 1) fore wings with hyaline membrane except around basal half of vein Sc, shaded with gray; 2) styliger with a pair of sublateral acute projections on the hind margin; 3) penes completely fused apically, with subquadrate base, narrowing abruptly in median zone, and with a ventral projection.

Material examined

Holotype

BRAZIL: ♂, imago (genitalia), Pará, rio Xingú, Campament, ca 60 km S of Altamira, 3°39' S, 52°22' W, 1–21 Oct. 1986, P. Spangler & O. Flint leg. (INPA).

Remarks

Macunahyphes incognitus was described based on only one specimen and since then there has been no further record of this species. The forceps are unknown and we only had access to the holotype genitalia. Because of this, the comparisons between species were made based on the genitalia slide and original description.

Distribution (Fig. 1)

Brazil (Pará).

Macunahyphes pemonensis Molineri, Grillet, Nieto, Dominguez & Guerrero, 2011

Macunahyphes pemonensis Molineri, Grillet, Nieto, Dominguez & Guerrero, 2011: 45.

Diagnosis

According to the original description, the male imago of *Macunahyphes pemonensis* can be distinguished from all congeners by the following combination of characteristics: 1) fore wings hyaline, membrane tinged with gray on basal half of C and Sc regions and basally to vein A; 2) styliger plate very slightly

projected posteriorly as a columnar base for each forceps, with a pair of sublateral acute projections on the hind margin; 3) penis long and slender, with apical furrow and a somewhat protruding ventral projection.

Material examined

BRAZIL: 2 ♂♂, imagines, Amapá, Pedra Branca, stream crossing highway BR 210 near Pedra Branca (PT3), 00°37'38.9" N, 51°38'15.2" W, 3 Aug. 2011, P.V. Cruz, A. Pes and A.S. Fernandes leg. (DZRJ 3156).

Remarks

In the original description of the male imago of *M. pemonensis* there is no mention of the presence of a dark purple spermatic duct that is laterally visible from outside, a character also found in *M. araca* sp. nov. and evident from the fresh material of *M. pemonensis* studied here (see Belmont *et al.* 2015: figs 2, 6). In addition, a new interpretation is given to the ventral structure present in the penis described by Molineri *et al.* (2011). The lateral margins of the penis lobe are strongly sclerotized and are ventrally twisted, forming a lateral narrow flap on each side of the penis. The area between these flaps, interpreted as a ventral structure in the original description, is in fact a shallow groove (see Belmont *et al.* 2015: figs 4–6).

Distribution (Fig. 1)

Brazil (Amapá) and Venezuela (Bolívar).

Macunahyphes zagaia sp. nov.

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Figs 2A, 3B

Diagnosis

The male of *M. zagaia* sp. nov. can be distinguished from all congeners by the following combination of characteristics: 1) longitudinal vein CuP absent; 2) membranous filaments of mesoscutellum absent; 3) segment II of forceps with a basal swelling; 4) penes trident-like, with inward curved lateral projections and acute medial projection.

Etymology

Zagaia is a spear with three points used in Amazonian artisanal fishery, reminiscent of the characteristic trident-like penis of the new species.

Type material

Holotype

BRAZIL: ♂, subimago, Amazonas, Manaus, Reserva Adolfo Ducke, Igarapé do Tinga, 2°55'33.53" S, 59°54'1.16" W, 10–13 Jun. 2002, A.M.O. Pes leg. (INPA-EPH 000012).

Paratypes

BRAZIL: 1 ♂, subimago, same data as holotype but 10–15 Jun. 2004, A.M.O. Pes leg. (DZRJ 3145); 1 ♂, subimago, Barcelos, Aracá sierra, Aracá river, before mouth of the igarapé da serrinha, 00°23'57.41" N, 63°22'43.97" W, 1 Feb. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr leg. (INPA-EPH 000013); 1 ♂, subimago, Aracá sierra, 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr leg. (DZRJ 3144); 2 ♂♂, subimagines, Jauari River, 00°48'0.28" N, 63°29'22.92" W, 26 Jul. 2009, F.F. Salles, A.P. Santos and N. Ferreira-Jr leg. (CZNC Ep-6436).

Description

Holotype

LENGTH (mm). Body: 2.75; fore wing: 2.76; cerci: 1.5; terminal filament: 2.14. General coloration yellowish brown and white, abdomen slightly washed with black.

HEAD. Yellowish, washed with black; ocelli white, surrounded with black; eyes black; antennae translucent white.

THORAX. Pronotum yellowish, heavily washed with black, except submedial unpigmented stripe and oblique brownish line; mesonotum yellowish brown, slightly washed with black and darker on carinae, anteronotal protuberance yellowish; metanotum yellowish; pleura yellowish; sterna yellowish.

FORE WING. Membrane whitish, tinged with gray on basal half of C and Sc regions; longitudinal veins and cross veins whitish, except for vein Sc; vein CuP absent.

LEGS. Fore leg lost in holotype; middle and hind legs translucent yellowish, femur with apical black mark.

ABDOMEN. Segments I–VI translucent white, VII–X yellowish white. Terga I–X almost completely washed with black, terga I–VIII with unpigmented sublateral stripe, tergum X with medial dark furrow. Sterna slightly washed with black.

GENITALIA. Forceps whitish, styliger plate and penes yellowish; segment II of forceps with a basal swelling; styliger plate with acute sublateral projection on hind margin; penes trident-like, with inward curved lateral projection and acute medial projection. Caudal filaments whitish, slightly washed with black at base.

Female

Unknown.

Nymph

Unknown.

Biology

The new species was collected during late fall and winter (Jun. 2002 and 2004; Jul. and Aug. 2009).

Variation

Differences were found only in relation to the length of the subimagines, ranging from 2.7 to 3.1 mm (body), 6.0 to 7.6 mm (cerci) and 4.8 mm (terminal filament).

Remarks

The male imago of *Macunahyphes zagaia* sp. nov. can be readily distinguished from all other species in the genus by the unique morphology of the penes.

Distribution (Fig. 1)

Brazil (Amazonas).

Discussion

According to Dias *et al.* (2005), unique characters present in imagines of *Macunahyphes* are: forceps tri-segmented, with first segment distomedially projecting; penis with very wide base, becoming thinner toward a subapical constriction, and then slightly widening again; penis lobes of each side almost completely fused except for apical incision; penis with a ventral projection covered with spines. With the recent discovery of new species, some of these features are actually restricted to *M. australis*.

Concerning the forceps, the presence of a basal swelling in the two new species described here, as well as in adults of *M. pemonensis* and *M. eduardoi*, raises important questions concerning the evolution of this character in Leptoxyphidae. According to Molineri (2002), the presence of the basal swelling was considered one of the synapomorphies of the genus *Tricorythodes*. This basal swelling, however, might be a synapomorphy not for *Tricorythodes*, but for the clade leading to *Macunahyphes* and *Tricorythodes* (see Molineri 2002, 2006). In that case, its absence in *M. australis* seems to be an autapomorphic trait.

The ventral projection of the penis covered with spines is clearly an autapomorphy of *M. australis*, since it is not found in any of the remaining species. In fact, unlike other Leptoxyphidae, the morphology of the penis in *Macunahyphes* is highly variable among species. Any attempt to provide a diagnostic characteristic for that structure, therefore, is unsuccessful.

Wing venation, especially the absence of the vein CuP, was the main characteristic used for the allocation of the new species in *Macunahyphes*. The shape of the penis, a character somewhat conservative among the genera of Leptoxyphidae and historically useful for genus delimitation in mayflies, is surprisingly variable in *Macunahyphes*. Despite the fact that vein CuP is poorly developed in some species of *Tricorythodes* (e.g., *T. bullus*), as well as in some specimens of *M. australis*, the new species described here should not be allocated to *Tricorythodes*. In this genus, one of the most diverse and widely distributed of the family, the shape of the penis is similar in all the species described so far (ca 55 species described). The description of the immature stages of most of the species of *Macunahyphes* will undoubtedly be essential for elucidating the monophyly of the genus and its relationship with *Tricorythodes*.

To improve our current knowledge of the genus, new efforts must be made in order to find more specimens of some rare species, such as *M. incognitus*, and, even more important, to find and describe the unknown nymphs of the species described in the last few years.

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