



Two new species of *Culex* subgenus *Melanoconion* (Diptera: Culicidae) from the Amazon forest

ROSA SÁ GOMES HUTCHINGS¹ & MARIA ANICE MUREB SALLUM^{2,3}

¹Coordenação de Pesquisas em Entomologia, Instituto Nacional de Pesquisas da Amazônia, Caixa Postal 478, 69011-970 Manaus, AM, Brasil

²Departamento de Epidemiologia, Faculdade de Saúde Pública, Universidade de São Paulo, Avenida Dr. Arnaldo 715, 01246-904 São Paulo, SP, Brasil

³Corresponding author. E-mail: masallum@usp.br

Abstract

Two new mosquito species (Diptera: Culicidae), *Culex (Melanoconion) phyllados* n. sp. and *Culex (Melanoconion) brachiatus* n. sp. from the state of Amazonas, Brazil, are here validated and described based on morphological features of the male genitalia. Both species are morphologically more similar to both *Culex coppenamensis* Bonne-Wepster & Bonne and *Culex alinkios* Sallum & Hutchings than to any other species of the Bastagarius Subgroup of the subgenus *Melanoconion*. Diagnostic characters for the identification of the adult male of both species are provided.

Key words: Amazon basin, Culicidae, *Culex (Melanoconion)*, new species

Introduction

Thirty-eight species of subgenus *Melanoconion* Theobald of genus *Culex* Linnaeus (Diptera: Culicidae) were collected at the Parque Nacional do Jaú, state of Amazonas, Brazil (Hutchings *et al.* 2005), including a few individuals identified as two distinct morphological forms similar to *Culex coppenamensis* Bonne-Wepster and Bonne (Sallum & Hutchings 2003). Comparing those two Amazonian forms with specimens collected in the Vale do Ribeira, situated in the Atlantic Forest in southeastern São Paulo state, Brazil, we observed several morphological characteristics in the male genitalia that could distinguish all three forms. Consequently, specimens collected in the Vale do Ribeira were employed to describe and validate *Culex alinkios* Sallum and Hutchings (Sallum & Hutchings 2003). Additional comparisons of the male genitalia of the two Amazonian forms with those of *Cx. coppenamensis* collected in localities north of Manaus and with the drawings of this species available in the literature (Pecor *et al.* 1992) showed that both forms belong to two unnamed species. Consequently, in the present study, we name and validate the Amazonian *Cx. coppenamensis* Forms 1 and 2 of Sallum and Hutchings (2003) as new species of the subgenus *Melanoconion*. We also provide diagnostic characters to distinguish *Cx. coppenamensis*, *Cx. alinkios* and the new species.

Material and methods

Mosquitoes were collected in Novo Airão, São Gabriel da Cachoeira, Nhamunda and the Japurá municipalities in Amazonas State, Brazil. In Novo Airão, fieldwork was carried out in the Jaú National Park and in Japurá at the Juami-Japurá Ecological Station. Specimens were collected in undisturbed (forest) and disturbed

habitats (secondary growth and peri-domestic sites), along trails, at margins of rivers, lakes, streams and igapós, and within the marginal terraces between the flood plains and the upland “terra firme” forest itself using CDC and CDC-UV light traps and Shannon traps. CDC traps were placed 1m above the ground, from 1800 to 0600 h. Specimens were identified at the INPA laboratories in Manaus and the Faculdade de Saúde Pública (FSP-USP), Universidade de São Paulo, Brazil, where an Entomological Reference collection is maintained. The catalogue for *Culex (Melanoconion)* (Pecor *et al.* 1992) was used to aid species identification. Specimens are deposited in the Invertebrate Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA-Manaus) and in the FSP-USP Entomological Reference Collection.

Culex (Melanoconion) phyllados n. sp.

(Fig. 1A–G)

Culex (Melanoconion) copenamensis Form 1 of Sallum & Hutchings 2003: 615 (distribution; systematics notes; male genitalia); Hutchings *et al.* 2005: 433 (species distribution).

Description. See Sallum & Hutchings (2003) for description and details of the adult male. Male genitalia. Tergum IX as figured (Fig. 1A). Gonocoxite globose, outer margin convex, inner margin nearly straight; ventrolateral setae strongly developed, ventromesal surface with small setae scattered from base to level of distal division of subapical lobe, setae stronger basally, lateral surface with a well-developed patch of long, slender setae at level of subapical lobe (Fig. 1B); tergomesal surface with 2–5 foliform setae at level of subapical lobe (for details see Fig. 1B in Sallum & Hutchings 2003); proximal part of ventrolateral surface with scales (for details see Fig. 1A in Sallum & Hutchings 2003); subapical lobe distinctly divided, divisions separated, proximal division unique at base, columnar, not clearly divided into 2 branches at apex, proximal branch shorter than distal branch, each branch bearing 1 long, robust, sinuous, apically hooked seta (setae *a* and *b*), seta *b* stronger than *a*, proximal division with a patch of short setae mesally at base of distal surface (Figs. 1B, D); distal division divided into 2 arms, a proximal, relatively short and broad arm and a long, strong columnar arm arising from base at lateral surface (Figs. 1B, D), most proximal arm with 6 apical setae, including 1 long, strong, apically hooked seta (*h*) and 1 relatively long, narrow, pointed saberlike seta (*s*) inserted in a small tubercle at base of seta *h*, both *h* and *s* arise from separate tubercles at proximal side of arm, separate from remaining setae, and 3 subequal, narrow appressed foliform setae (*f*) and 1 long, strong, saberlike seta (*s*) inserted at distal side; columnar arm (Figs. 1B, D) long, strong, nearly straight with 1 foliform seta (*l*) at apex; seta *l* strongly enlarged, asymmetrical not striate with a well-developed lobe projecting distally from apex (Figs. 1B, D). Gonostylus (Figs. 1B, C) short, strong, curved at midlength, with a patch of spicules at midlength on dorsal surface, widened in distal 0.5 (in lateral view), tapered distally and bearing a ventral crest of short, scattered spicules from widened part to tip of apical snout; apical snout short; gonostylar claw short, leaflike. Phallosome (Fig. 1E) with lateral plate slightly longer than aedeagal sclerite, aedeagal sclerite narrow and curved in lateral view with anterior margin thickened and sclerotized, distal end narrowly fused to base of lateral plate; distal part of lateral plate with apical, ventral and lateral processes; apical process short, broad at base, somewhat curved on ventral side, apical margin straight and smooth, pointed on dorsal side, dorsal edge concave; ventral process short, somewhat hooklike, pointed, curved laterally; lateral process elongate, somewhat triangular, directed dorsolaterally, tapered to apex, apex pointed; base of lateral plate with short dorsal process and basally continuous with thickened margin of aedeagal sclerite; aedeagal sclerite not connected by dorsal aedeagal bridge. Proctiger (Fig. 1F) elongate; paraproct distally narrowed, basally expanded at articulation with basal plate and posterolateral margin of tergum X; paraproct crown with 9–10 simple blades; 2 or 3 cercal setae. Tergum X (Fig. 1F) large, somewhat rectangular in outline, rounded at apex; medial surface convex, widened and projecting distally. Tergum VIII with a shallow V-shaped emargination, separating two lateral lobes, which are somewhat round distally and bear scattered setae (Fig. 1G).

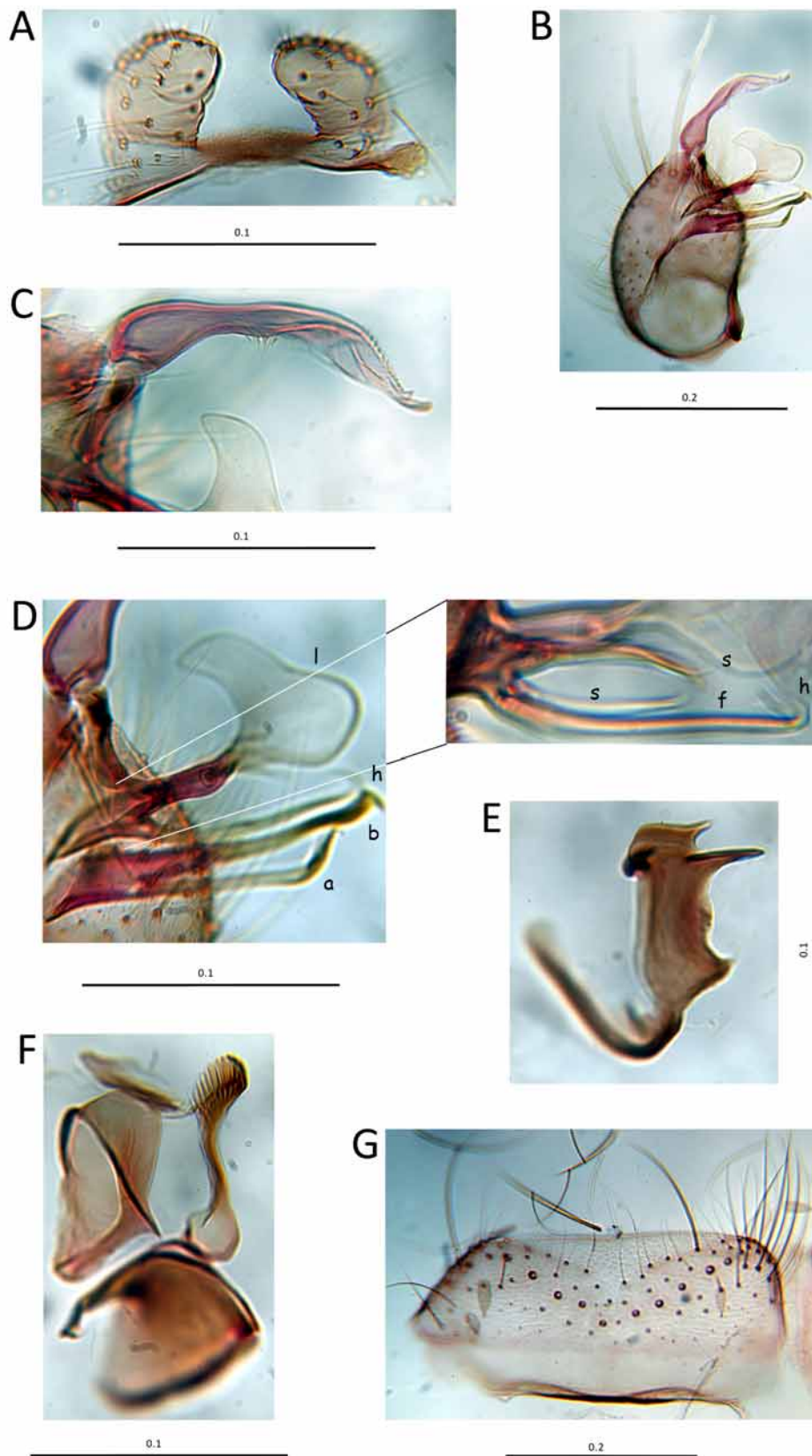


FIGURE 1. Male genitalia of *Culex phyllados* n. sp., A: tergum IX in dorsal aspect; B: gonocoxite and gonostylus in mesal aspect; C: gonostylus in lateral view; D: subapical lobe of gonocoxite showing both proximal and distal divisions in mesal aspect; E: lateral plate and aedeagal sclerite in lateral aspect; F: proctiger in dorsal aspect; G: tergum VIII in dorsal aspect. (Photomicrographs of holotype (PNJ-000845_01) except for detail in D of paratype (ProN-000056)). (Scales in mm).

Holotype. Adult male with dissected male genitalia on microscope slide (specimen number PNJ-000845_01), BRAZIL: Amazonas state, Novo Airão municipality, Parque Nacional do Jaú, Jaú River, Miratucu Stream (01°47'02"S, 61°49'01"W), 28–29 Jul 1995, collected with UV light trap (1800–0600 h) along stream inside tropical rain forest, R. W. Hutchings coll., deposited in the Invertebrate Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA), Manaus, Amazonas state, Brazil, accession number DIPTE-510.

Paratypes. Fifty-eight specimens (28 pinned adult males, 2 adult males mounted on microscope slides, 28 male genitalia on microscope slides), as follows:

Same collection as holotype, 4 males, 4 male genitalia (specimen numbers PNJ-000845_02, PNJ-000845_03, PNJ-000845_04, PNJ-000845_05); same location as holotype, 26 Jul 1995, collected with Shannon trap (2000–2100 h) along stream inside forest, Hutchings & Sallum coll., 2 males, 2 male genitalia (specimen numbers PNJ-000771, PNJ-000785); 26–27 Jul 1995, collected with UV light trap (1800–0600 h) along stream inside forest, R. W. Hutchings coll., 16 males, 16 male genitalia (specimen numbers PNJ-000803_01, PNJ-000803_02, PNJ-000803_03, PNJ-000803_04, PNJ-000803_05, PNJ-000803_06, PNJ-000803_07, PNJ-000803_08, PNJ-000803_10, PNJ-000803_11, PNJ-000803_12, PNJ-000803_13, PNJ-000803_14, PNJ-000803_15, PNJ-000803_16, PNJ-000803_17) and 2 adult males mounted on microscope slides with associated dissected male genitalia (specimen numbers PNJ-000803_18, PNJ-000803_19); 28 Jul 1995, collected with Shannon trap (1800–2000 h) inside secondary forest, Hutchings & Sallum coll., 1 male, 1 male genitalia (specimen number PNJ-000815); Novo Airão, Parque Nacional do Jaú, Lower Carabinani River, 01°59'00"S, 61°32'00"W, 12–13 Apr 1994, collected with CDC trap (1800–0600 h) inside secondary forest, Ferreira & Hutchings coll., 2 males, 2 male genitalia (specimen numbers Jam-000143_01, Jam-000143_02); São Gabriel da Cachoeira, Querari, 15°00'00"N, 069°51'00"W, 22 Apr 1993, 6:00 pm–6:00am, collected with CDC trap (1800–0600 h) inside secondary forest, Ferreira & Vidal coll., 1 male, 1 male genitalia (specimen number Qam-000165); Japurá, Juami River, Estação Ecológica Juami-Japurá, 02°04'24"S, 68°16'52"W, 10–11 Aug 2005, collected with CDC UV trap (1800–0600 h) inside upland “terra firme” forest, Aquino coll., 1 male, 1 male genitalia (specimen number JJAM-001399); Nhamunda, Areia, Igarape do Areia (ME), Rio Nhamunda (MD), 01°35'22"S, 057°37'06"W, 16–17 May 2008, collected with CDC UV trap (1800–0600 h) inside upland “terra firme” forest, Hutchings, RSG et al. coll., 1 male, 1 male genitalia (specimen number ProN-000056).

Distribution and bionomics. *Culex phyllados* was found in four different municipalities (Novo Airão, Japurá, São Gabriel da Cachoeira and Nhamunda) in the state of Amazonas, Brazil. This species was collected along rivers in both primary and secondary forest using UV light traps (1800–0600 h), Shannon traps (1800–2100 h) and CDC traps with either incandescent or UV light sources (1800–0600 h). Nothing is known about the bionomics of this species.

Etymology. The name *phyllados*, Greek for ‘leafy’, was chosen to reflect the presence of 2–5 foliform setae on the tergomesal surface of the gonocoxite and one strongly enlarged, asymmetrical, foliform seta on the distal division of the subapical lobe.

Taxonomic notes. *Culex phyllados* can be recognized by the following characters of the male genitalia: 1) the presence of 2–5 foliform setae on the tergomesal surface at the level of the subapical lobe of the gonocoxite (for details see Fig. 1B in Sallum & Hutchings 2003); 2) the distal division of the subapical lobe of the gonocoxite is divided into two arms, a relatively short and broad arm and a long, strong columnar arm; 3) the most proximal arm with one long, strong, apically hooked seta (*h*), one relatively long, narrow, pointed saberlike seta (*s*), three subequal, narrow appressed foliform setae (*f*) and one long, strong, saberlike seta (*s*) (Fig. 1D); 4) the most distal arm is long, strong, nearly straight and bearing one large strongly asymmetrical, non-striate foliform seta, with a well developed lobe projecting distally from the apex (Fig. 1D); 5) the gonostylus is short, strong, curved at midlength, possessing a patch of spicules at midlength on dorsal surface, bearing a crest of short, scattered spicules extending distally from widened part to apex of apical snout on ventral surface; and 6) apical snout of the gonostylus short (Fig. 1C).

Culex (Melanoconion) brachiatus n. sp.
(Fig. 2A–G)

Culex (Melanoconion) copenamensis Form 2 of Sallum & Hutchings 2003: 615 (distribution, systematics notes, male genitalia); Hutchings *et al.* 2005: 433 (species distribution).

Description. Adult male indistinguishable from that of *Cx. phyllados* except for characters of the genitalia. See Sallum & Hutchings (2003) for details (as *Cx. copenamensis* Form 1). Male genitalia. Tergum IX as figured (Fig. 2A). Gonocoxite globose, outer margin convex, inner margin nearly straight; ventrolateral setae strongly developed; ventromesal surface with small, scattered setae from base to level of distal division of subapical lobe, setae stronger basally; lateral surface with a well-developed patch of long, slender setae at level of subapical lobe (Fig. 2B); proximal part of ventrolateral surface with a few scales; subapical lobe distinctly divided, divisions separated, proximal division unique at base, columnar, not clearly divided into 2 arms at apex, proximal arm shorter than distal arm, each arm bearing 1 long, robust, sinuous, apically hooked seta (setae *a* and *b*), seta *b* more slender than seta *a*; distal division divided into 2 arms, a proximal, relatively short and broad arm and a long, strong columnar arm arising from base at lateral surface (Fig. 2D), most proximal arm with 6 apical setae, including 1 long, strong, apically hooked seta (*h*) and 1 shorter, narrow, pointed saberlike seta (*s*) inserted in a small tubercle at base of seta *h*, both *h* and *s* arise from separate tubercles at proximal side, 3 subequal, narrow appressed foliform setae (*f*), and 1 long, strong, saberlike seta (*s*) inserted at distal side; distal columnar arm (Fig. 2D) with one foliform seta (*l*) at apex, seta *l* strongly enlarged, not strongly asymmetrical, striate at base with a well developed basal expansion (Fig. 2D). Gonostylus (Figs. 2B, C) short, strong, narrowed at midlength, appearing curved and widened in distal 0.5 (in lateral view), tapered distally and bearing ventral crest of short, scattered spicules from widened part, not reaching tip of apical snout; apical snout long; gonostylar claw short, leaflike. Phallosome (Fig. 2E) with lateral plate slightly longer than aedeagal sclerite; aedeagal sclerite narrow and curved in lateral view with anterior margin thickened and sclerotized, distal end narrowly fused to base of lateral plate; distal part of lateral plate with apical, ventral and lateral processes; apical process short, broad at base, somewhat curved on ventral side, apical margin straight and smooth, pointed on dorsal side, dorsal edge concave; ventral process short, somewhat hooklike, pointed, curved laterally; lateral process elongate, somewhat triangular, directed dorsolaterally, tapered to apex, apex pointed; base of lateral plate with short dorsal process and basally continuous with thickened margin of aedeagal sclerite; aedeagal sclerite not connected by dorsal aedeagal bridge. Proctiger elongate; paraproct distally narrowed, basally expanded at articulation with basal plate and posterolateral margin of tergum X; paraproct crown with 10–14 simple blades; 2 cercal setae (Fig. 2F). Tergum X (Fig. 2F) large, somewhat rectangular in outline, rounded at apex; medial surface convex, widened and projecting distally. Tergum VIII with an intermediate V-shaped emargination separating the two lateral lobes and possessing scattered setae forming a concentrated distal setal group (Fig. 2G).

Holotype. Adult male with dissected genitalia on microscope slide (specimen number PNJ-000803_09), BRAZIL: Amazonas state, Novo Airão municipality, Parque Nacional do Jaú, Jaú River, Miratucu Stream, 01°47'02"S, 061°49'01"W, 26–27 July 1995, collected in UV light trap (1800–0600 h) inside forest along stream, R.W. Hutchings coll., deposited in the Invertebrate Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA), Manaus, Amazonas, Brazil, accession number DIPTE-511.

Paratype. Adult male with dissected male genitalia on microscope slide (specimen number PNJ-000803_20), same data of holotype, deposited in Faculdade de Saúde Pública, Universidade de São Paulo (FSP-USP), São Paulo, Brazil, accession number E-11497.

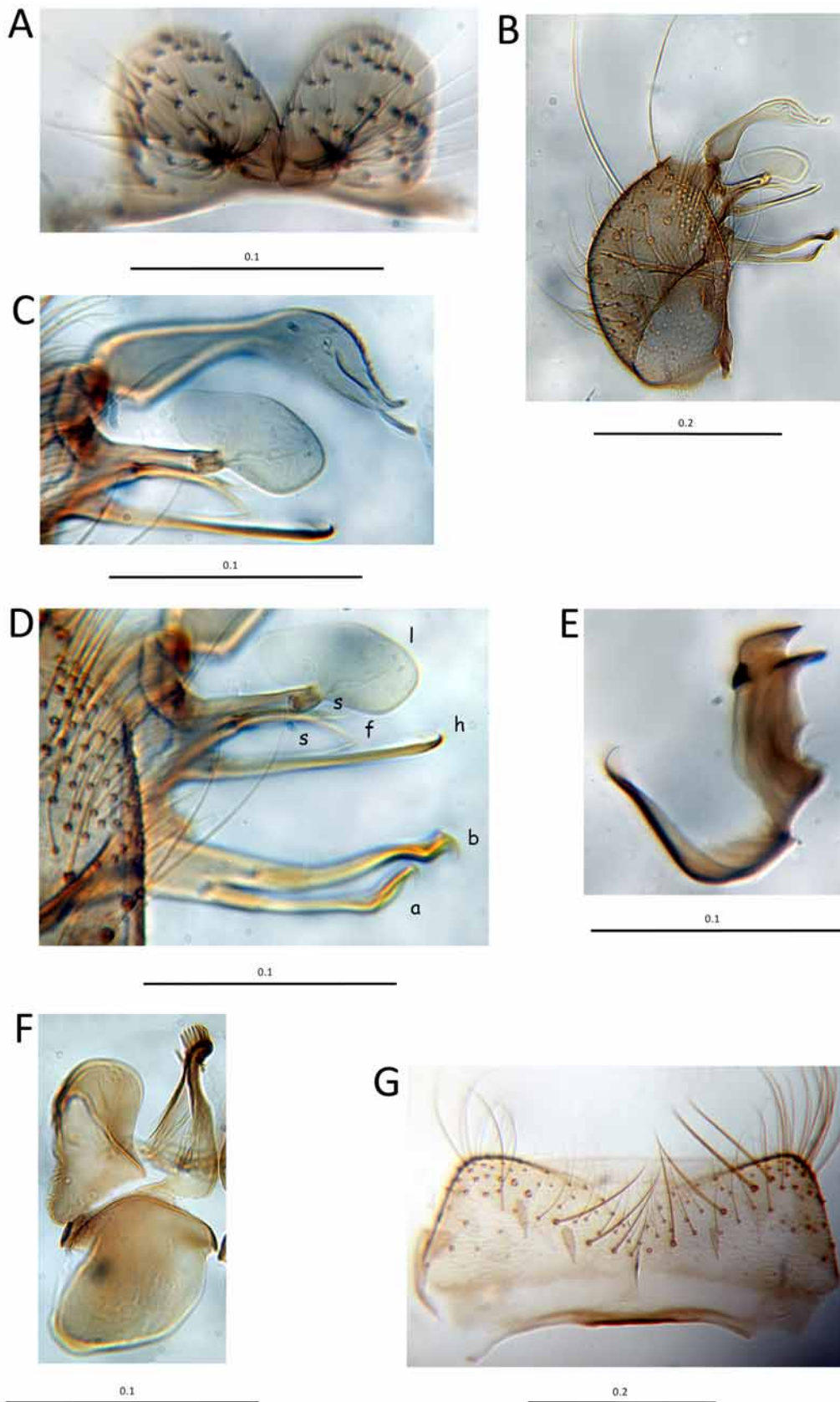


FIGURE 2. Male genitalia of *Culex brachiatus* n. sp. A: tergum IX in dorsal aspect; B: gonocoxite and gonostylus in lateral aspect; C: gonostylus in lateral view; D: subapical lobe of gonocoxite showing both proximal and distal divisions in lateral aspect; E: lateral plate and aedeagal sclerite in lateral aspect; F: proctiger in dorsal aspect; G: tergum VIII in dorsal aspect. (Photomicrographs of holotype (PNJ-000803_09)). (Scales in mm).

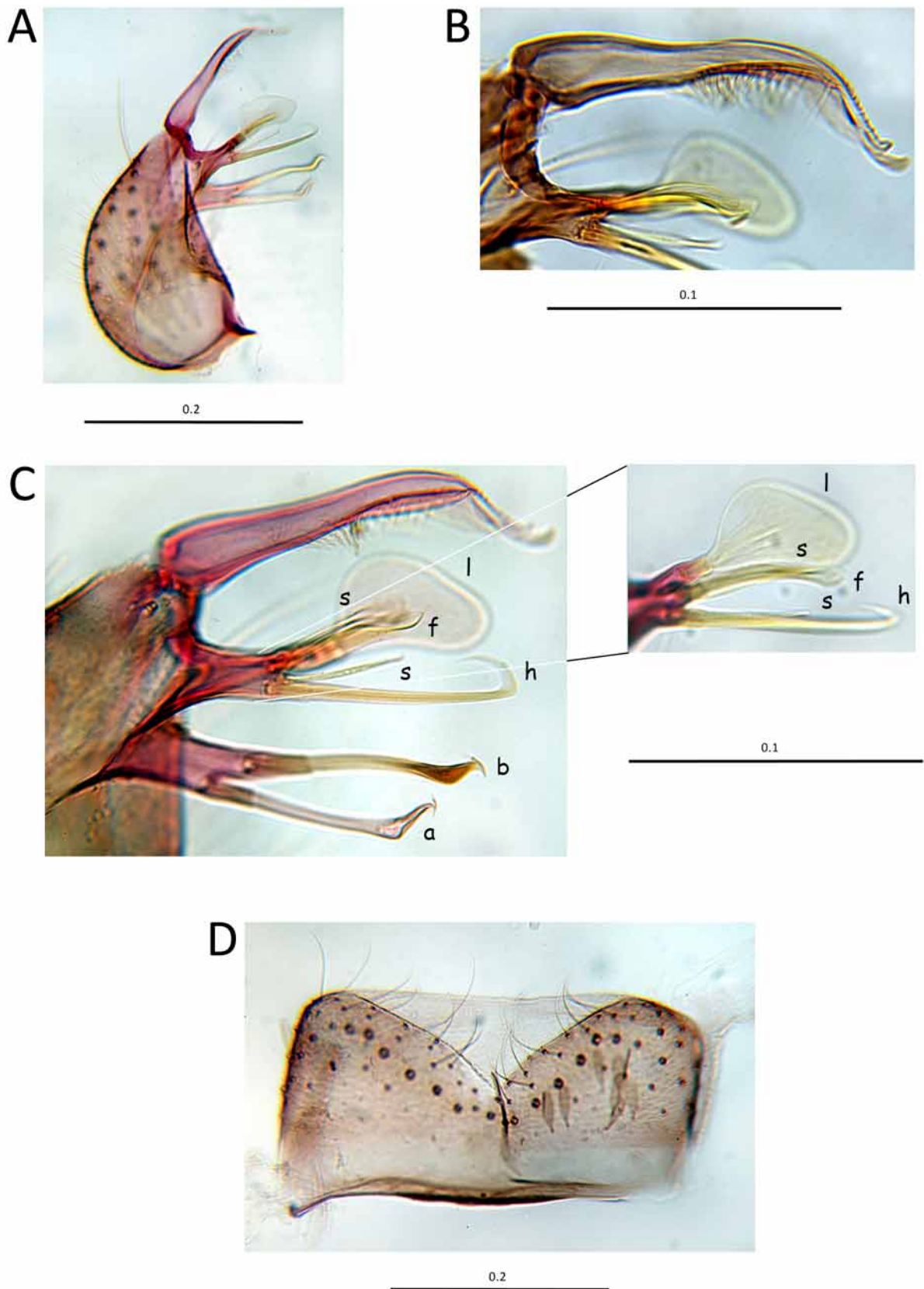


FIGURE 3. Photomicrographs of the male genitalia of *Culex coppenamensis* from Manaus municipality, Amazonas, Brazil. A: gonocoxite and gonostylus in mesal aspect; B: gonostylus in lateral view; C: subapical lobe of gonocoxite showing both proximal and distal divisions in mesal aspect; D: tergum VIII in dorsal aspect. (Scales in mm).

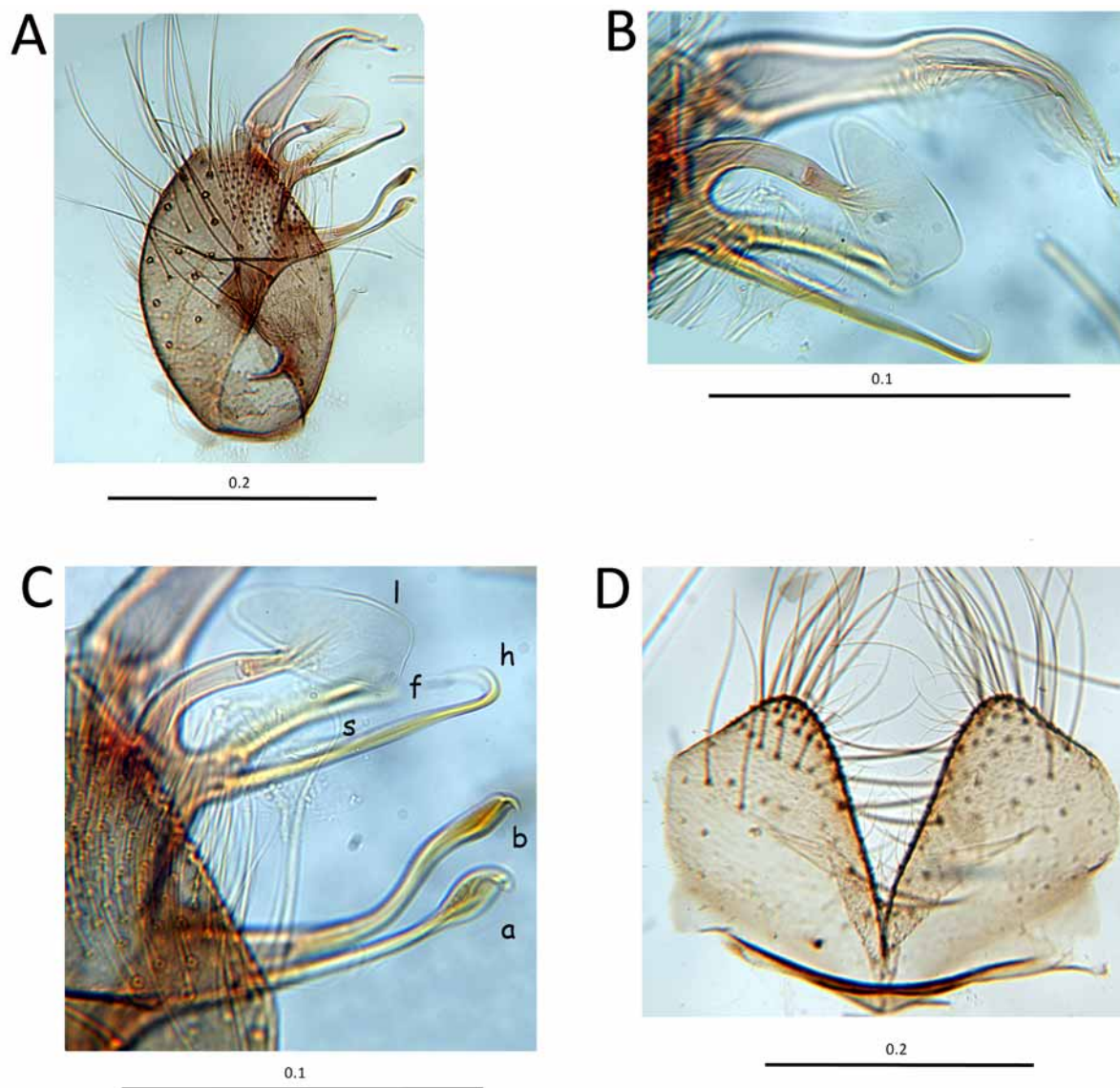


FIGURE 4. Male genitalia of *Culex alinkios* from Cananéia municipality, São Paulo state, Brazil. A: gonocoxite and gonostylus in lateral aspect; B: gonostylus in lateral view; C: subapical lobe of gonocoxite showing both proximal and distal divisions in lateral view; D: tergum VIII in dorsal aspect. (Photomicrographs of paratypes deposited in the INPA invertebrate collection (DIPT.306P-2 & DIPT.306P-3)). (Scales in mm).

Other material examined: *Culex coppenamensis* (Fig. 3): 14 specimens (7 pinned adult males, 7 male genitalia on microscope slides), as follows:

BRAZIL: Amazonas state, Manaus municipality, Road BR-174 km 68, Fazenda Porto Alegre, 02°21'18"S, 59°57'27"W, collected with CDC trap inside 10 ha. upland "terra firme" forest fragment, 1800–0600 h, 4–5 Jun 2002, Hutchings & Aquino coll., 3 males, 3 male genitalia (specimen numbers Fam-002004, Fam-002005, Fam-002009); 10–11 Jul 2002, Aquino & Alves coll., 1 male, 1 male genitalia (specimen number Fam-002347); 6–7 Aug 2002, Hutchings *et al.* coll., 1 male, 1 male genitalia (specimen number Fam-002703); 02°22'53"S, 59°56'32"W, collected with CDC trap inside 100 ha. upland "terra firme" forest fragment, 1800–0600 h, 8–9 Oct 2002, Hutchings *et al.* coll., 1 male, 1 male genitalia (specimen number Fam-003519); 02°22'00"S, 59°57'51"W, collected with CDC trap inside secondary forest, 1800–0600 h, 30 Apr – 01 May 2003, Menezes and Aquino coll., 1 male, 1 male genitalia (specimen number Fam-005199).

Distribution and bionomics. *Culex brachiatus* is known from the Parque Nacional do Jaú, Novo Airão

municipality, Amazonas state, Brazil. Specimens were collected along a stream inside upland “terra firme” forest using a UV light trap. Nothing is known about the bionomics of this species.

Etymology. The name *brachiatus*, Latin for ‘with arms / branches’, was chosen because this species is distinguished in having the distal division of the subapical lobe of the gonocoxite subdivided into two arms.

Taxonomic notes. *Culex brachiatus* can be recognized by the following characters of the male genitalia: 1) the gonostylus has a long apical snout, and the ventral subapical crest of spicules is restricted to the widened part and does not reach the apex of the apical snout (Fig. 2C); 2) the distal arm of the distal division of the subapical lobe is more or less straight and bears one foliform seta (*l*) that is not strongly asymmetrical (Fig. 2D); and 3) the foliform seta (*l*) is striated at the base (Fig. 2D).

Discussion

Culex coppenamensis, *Cx. alinkios*, *Cx. phyllados* and *Cx. brachiatus* belong to the Bastagarius Subgroup of the Bastagarius Group of Sirivanakarn (1983). The male genitalia of these species (Figs. 1–4) can be distinguished from those of other members of the Bastagarius Subgroup by the following combination of characters: 1) proximal division of the subapical lobe of gonocoxite columnar, not clearly divided into two arms; 2) lateral plate of phallosome with apical, lateral and ventral processes, ventral process arising at the same level as the lateral process, hooklike, dorsolaterally bent, apical process broad, somewhat rectangular with flat, smooth apical margin, apical dorsal angle ending in an acute point and curved at dorsal edge; 3) distal division of the subapical lobe subdivided into outer and inner arms; 4) outer arm of the distal division with an enlarged, asymmetrical, foliform seta (*l*) inserted at the apex of a tubercle that arises separately on the outer side of the division; 5) inner arm of the proximal division with 1 long, strong, saberlike seta (*s*) arising from the apex of a short tubercle that arises on its distal side; 6) gonocoxite globose with a patch of long, strong setae on the lateral surface at the level of the subapical lobe.

Adult males of these four species are morphologically more similar to each other than to any other species of the Bastagarius Subgroup and for an accurate identification it is necessary to rely on characteristics of the male genitalia. Sallum & Hutchings (2003) considered the characters that distinguish *Cx. phyllados* and *Cx. brachiatus* from *Cx. coppenamensis* to be unclear in the male genitalia drawing provided by Bonne and Bonne-Wepster (1925). However, additional specimens collected in the same area and in other localities of the Amazon Forest allowed more detailed comparisons among the specimens. Consequently, while comparing the gonostylus and the distal division of the subapical lobe of the gonocoxite, we concluded that those individuals identified as Forms 1 and 2 belong to two unnamed species, i.e., *Cx. phyllados* and *Cx. brachiatus*.

Culex phyllados can be easily distinguished from *Cx. alinkios* in having tergum VIII with a shallow V-shaped emargination, separating two lateral lobes, which are somewhat round distally and bear scattered setae (Fig. 1G). In comparison, *Cx. alinkios* has a deep V-shaped emargination separating two somewhat triangular lateral lobes with long, strong setae forming a conspicuous distal setal group (Fig. 4D). *Culex brachiatus* and *Cx. coppenamensis* both have tergum VIII with an intermediate (medium) V-shaped emargination separating the two lateral lobes (Figs. 2G, 3D, respectively). Whereas the lateral lobes of tergum VIII in *Cx. brachiatus* possess scattered setae, more concentrated and forming a distal setal group (Fig. 2G), differing from the lateral lobes of *Cx. coppenamensis*, which do not have a well-defined distal group of setae (Fig. 3D).

The gonocoxite of *Cx. phyllados* possess 2–5 foliform setae on the tergomesal surface at the level of the subapical lobe (for details see Fig. 1B in Sallum & Hutchings 2003), which are absent in *Cx. alinkios*, *Cx. coppenamensis* and *Cx. brachiatus*. The gonostylus of *Cx. phyllados* (Figs. 1B, C) and *Cx. alinkios* (Figs. 4A, B) has a short and broad apical snout, and the subapical crest of spicules on the ventral (sternal) surface extends distally from widened part to the apex of the apical snout. In addition, the gonostylus of *Cx. phyllados* is shorter and the subapical part is broader (Fig. 1B, C) than that of *Cx. alinkios* (Fig. 4A, B). In contrast, the

gonostylus of *Cx. brachiatus* (Fig. 2B, C) and *Cx. coppenamensis* has a long apical snout. The ventral subapical crest of spicules in *Cx. brachiatus* is restricted to the widened part of the gonostylus and does not reach the apex of the snout (Fig. 2B, C), whereas in *Cx. coppenamensis* the spicules extend to the apex (Fig. 3A, B). Furthermore, in *Cx. brachiatus* the spicules on the dorsal surface of the gonostylus are absent.

In addition, *Cx. phyllados*, *Cx. brachiatus*, and *Cx. coppenamensis* are easily distinguished from *Cx. alinkios* in having the distal arm of the distal division of the subapical lobe, somewhat straight (Figs. 1B, D; 2B, D; 3A, C), whereas it is markedly curved in *Cx. alinkios* (Fig. 4 A, C). The shape of the foliform seta (*l*) borne at the apex of the arm is striate at the base in *Cx. brachiatus*, *Cx. coppenamensis* and *Cx. alinkios* (Figs. 2D, 3C, 4C), whereas it is smooth in *Cx. phyllados* (Fig. 1D).

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