



New species of *Culex* (*Culiciomyia*) (Diptera: Culicidae) from Talangaye Forest in Cameroon and descriptions and identification keys for males of the Afrotropical species of the subgenus

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Table of contents

Abstract	452
Introduction	452
Materials and methods	453
Taxonomic treatment	456
<i>Culex</i> (<i>Culiciomyia</i>) <i>apicopilosus</i> Cornel & Mayi, sp. n.	457
<i>Culex</i> (<i>Culiciomyia</i>) <i>lanzaroi</i> Cornel & Mayi, sp. n.	461
<i>Culex</i> (<i>Culiciomyia</i>) <i>pseudosubaequalis</i> Cornel & Mayi, sp. n.	463
<i>Culex</i> (<i>Culiciomyia</i>) <i>cambournaci</i> Hamon & Gandara, 1955.	469
<i>Culex</i> (<i>Culiciomyia</i>) <i>cinerellus</i> Edwards, 1922.	471
<i>Culex</i> (<i>Culiciomyia</i>) <i>cinereus</i> Theobald, 1901	473
<i>Culex</i> (<i>Culiciomyia</i>) <i>eouzani</i> Geoffroy, 1971	476
<i>Culex</i> (<i>Culiciomyia</i>) <i>macfiei</i> Edwards, 1923	479
<i>Culex</i> (<i>Culiciomyia</i>) <i>nebulosus</i> Theobald, 1901	485
<i>Culex</i> (<i>Culiciomyia</i>) <i>semibrunneus</i> Edwards, 1927	490
<i>Culex</i> (<i>Culiciomyia</i>) <i>subaequalis</i> Edwards, 1941	493
Discussion	496
Key to males of Afrotropical <i>Culiciomyia</i> based on maxillary palpomere lengths and maxillary palpal comb scales	496
Key to males of Afrotropical <i>Culiciomyia</i> based on habitus and genitalia	497
Acknowledgements	498
References cited	498

Abstract

Species of *Culex* (Diptera: Culicidae) belonging to the subgenus *Culiciomyia* were collected in partially logged areas and in surrounding pristine forest (Talangaye Forest) in the Nguti Subdivision in the South-West Region of Cameroon. Mosquitoes were collected mainly by sweep netting through forest floor vegetation. Morphological species identification of African *Culiciomyia* relies almost exclusively on the structure of the male genitalia and the shapes of comb scales on the maxillary palpi of males. Other features of males and the habitus of females are largely indistinguishable between the species of this subgenus. In total, seven currently described species and three new species were collected in the forest. The males of the three new species are described and named as *Culex apicopilosus* Cornel & Mayi, **sp. n.**, *Culex lanzaroi* Cornel & Mayi, **sp. n.** and *Culex pseudosubaequalis* Cornel & Mayi, **sp. n.** More detailed descriptions of males of the other currently known species that were collected in the Talangaye Forest and pictorial keys to the males of all Afrotropical species of *Culiciomyia*, including the new species, are provided.

Key words: *Culex apicopilosus*, *Culex lanzaroi*, *Culex pseudosubaequalis*, taxonomy

Introduction

According to the Mosquito Taxonomic Inventory (Harbach 2020), 54 species of the subgenus *Culiciomyia* Theobald of *Culex* Linnaeus (Diptera: Culicidae) are recognized worldwide. Most of the members of this subgenus, 37 in fact, occur in Asia, Southeast Asia and Australasia, and 17 species, one comprising two subspecies, occur in continental Africa and one species occurs in Yemen (Arabian Peninsula). Of those in Africa, two species, *Cx. pandani* Brunhes (Brunhes 1969) and *Cx. milloti* Doucet (Doucet 1949) only occur in Madagascar and one species, *Cx. cambournaci* Hamon & Gandara, is restricted to the West African Islands of São Tomé and Príncipe.

The only identification key developed for African *Culiciomyia* adults was published by Edwards (1941) for the six species known at that time, namely, *Cx. cinerellus* Edwards, *Cx. cinereus* Theobald, *Cx. macfieii* Edwards, *Cx. nebulosus* Theobald and *Cx. nebulosus pseudocinereus* Theobald, *Cx. semibrunneus* Edwards and *Cx. subaequalis* Edwards. After Edwards (1941), another 10 species were described, including *Cx. cambournaci* Hamon & Gandara, *Cx. eouzani* Geoffroy, *Cx. furlongi* van Someren; *Cx. gilliesi* Hamon & van Someren, *Cx. grenieri* Eouzan, *Cx. harleyi* Peters, *Cx. liberiensis* Peters, *Cx. milloti* Doucet, *Cx. mongiro* van Someren, *Cx. muspratti* Hamon & Lambrecht and *Cx. pandani* Brunhes. Both adult sexes and larvae are known for all of the species except for *Cx. muspratti*, *Cx. eouzani* and *Cx. grenieri*, which were described from males only, and *Cx. milloti*, which is known from larvae only.

In 2016 and 2017, mosquito sampling was conducted in an area of the mostly pristine tropical forest, and in patches of hard wood logged forest (secondary disturbed forest) and palm oil plantations in the South-West Region of Cameroon. This area was selected as part of a study to measure the impacts of deforestation on the prevalence and dynamics of bird malaria in mosquitoes. Mosquitoes are the primary vectors of avian *Plasmodium*. Unfortunately, while still mostly forested in 2016 and 2017, the vegetation in this area by 2018 had been completely removed and is now mostly barren with small patches of young palm oil tree plantations. To measure bird malaria infectivity in mosquitoes, the relevant mosquitoes had to be identified. It was soon discovered that the forest supported a high diversity of mosquito species for which adequate keys and descriptions were not available. Unfortunately, adequate keys are not available for most of the African species of *Aedes* Meigen and *Culex*, especially for those species found in forested areas of western and central Africa.

This publication represents a first part of our efforts to update keys and descriptions of African *Aedes* and *Culex*. Herein, we provide morphological descriptions of male *Culiciomyia* that were collected in the forest, including previously described species, *Cx. cinerellus*, *Cx. cinereus*, *Cx. eouzani*, *Cx. macfieii*, *Cx. nebulosus*, *Cx. semibrunneus* and *Cx. subaequalis*, to note morphological features about them that were not mentioned or described in detail, and three new species named and described herein as *Cx. apicopilosus* Cornel & Mayi, **sp. n.**, *Cx. lanzaroi* Cornel & Mayi, **sp. n.** and *Cx. pseudosubaequalis* Cornel & Mayi, **sp. n.** The description of the male of *Cx. cambournaci* is also included. A pictorial key for all previously described males of Afrotropical *Culiciomyia* (including the three new species) is provided. We also include a shorter separate key to distinguish some of the species based solely on differences in the comb scales on the maxillary palpus of males.

Materials and methods

Sampling area and notes about the environment. Sampling was carried out in the Talangaye Forest, Nguti Sub-division in the South-West Region of Cameroon within a 3-km radius of the GPS coordinates of 5.190397° N; 9.3457790° E (Fig. 1). The village nearest to our collecting sites is Manyamen. Prior to 2018, the area, which is hilly, was covered with lowland broad leaf forest trees and bushes (Fig. 2). The relative humidity was 97% during the wet season and temperatures ranged from 19–25°C, measured using data-loggers (HOBO-U23 Pro. Version 2, Onset Computer Corporation, Bourne, MA, USA) positioned in the shade 1 m above the ground. Within this area, a consortium of investors plan to log over 700 km² of pristine forest and replace it with monoculture palm oil plantations. In 2016 and 2017, most of the forest canopy was dominated by multiple high valued Mahogany and Sapele hard wood timber trees (Fig. 2B), and a variety of broad leaf plants predominated below the canopy (Fig. 2C).

Mosquito capture. Mosquitoes were captured in January–February (late dry season), April–May (early wet season), July (middle wet season) and October–November (early dry season) in 2016 and 2017 in areas where the forest had not been logged, where logging had started and where almost all of the vegetation had been removed. Multiple collecting methods were used, including: i) modified miniature CDC light trap (Sudia & Chamberlain 1962) baited with a sugar-yeast mixture (Smallegange *et al.* 2010) to release CO₂, ii) net traps (Silver 2008) baited with pigeons and chickens that were held in cages 30.5 cm above the ground on tables (Fig. 3A), iii) net traps (Silver 2008) that had no bait in them, iv) walk-in resting cages made with sticks and branches and wrapped with brown paper or black plastic, based on the dimensions of the red box shelters of Goodwin (1942) (Fig. 3B), v) sweep netting through vegetation, vi) larval collections from tree holes, seed pods holding water and the edges of streams and rivers, and vii) reared from eggs laid in bamboo pots (Fig. 3C) and plastic containers filled with water from a nearby stream.

In addition to mosquitoes collected in the forest, specimens of *Cx. cambournaci* collected in São Tomé by the first author, and some species of *Culicomyia* preserved in the insect collection in the National Health Laboratory Services Museum in Sandringham, Johannesburg, South Africa were examined for comparative purposes.

Preservation and identification of mosquitoes. Mosquitoes were transported to a temporary processing tent in the field, identified to genus and subgenus and, whenever possible, to species using a field dissecting microscope with a battery-operated light source. Most males were preserved individually in a 1-ml tube with silica gel to keep them dry, and some were pin-mounted in the field. A piece of paper with the collection number was inserted into the tubes between the silica gel and the mosquito. Females captured in the same manner and identified to the same species or subgenus were pooled in batches of up to 25 and preserved in 80% ethanol for DNA extraction and detection of avian malaria infection. A collection number indicating the date, method of capture, species, subgenus or “group” written in pencil on a small piece of paper was inserted into the tube containing the ethanol. Some females were also preserved in tubes for morphological examination. Larvae and egg rafts were held in small cups, provided a small quantity of finely ground TetraMin® Tropical Flakes (Melle, Germany) for food, and reared to adults in the field. A small quantity of dried local forest vegetation was also added as a source of natural organic food matter to improve rearing success. Larvae of most species unfortunately died, indicating that conditions used for immature stages were not appropriate, and much still must be learned about the biology of the species.

In most instances the silica gel stayed dry enough to prevent fungal growth despite the very humid conditions in the field. Upon return to the laboratory, the dried males and females were removed from the tubes, pinned and held in plastic containers kept in a refrigerator at 4°C. Holding the pinned mosquitoes in the humid laboratory at room temperature, which was first done, resulted in them being destroyed by fungal growth within two to three days. The genitalia of males were dissected, softened and cleared for 12 h in Specimen Clearing Fluid (Bioquip Products Inc., Rancho Dominguez, CA, USA). This was followed by passing them through two washes of 100% ethanol before being mounted on microscope slides in PVA Mounting Medium (Bioquip Products Inc.).

Digital images were taken using an OMAX A3518OU attached to a Nikon SMZ800 dissecting microscope and an Amscope MU2003-BI attached to a Nikon E600 compound microscope with phase contrast optics. Images were processed using Amscope Capture software version 3.7 (Amscope, Irvine, CA). The white line in digital images is 100 µm in length.

Mosquito anatomical terminology is that recommended in Harbach & Knight (1980, 1982). The nomenclature used to describe plant leaf shapes (Jones & Luchsinger 1979) was used to describe shapes of some of the comb scales on the maxillary palpi of males.

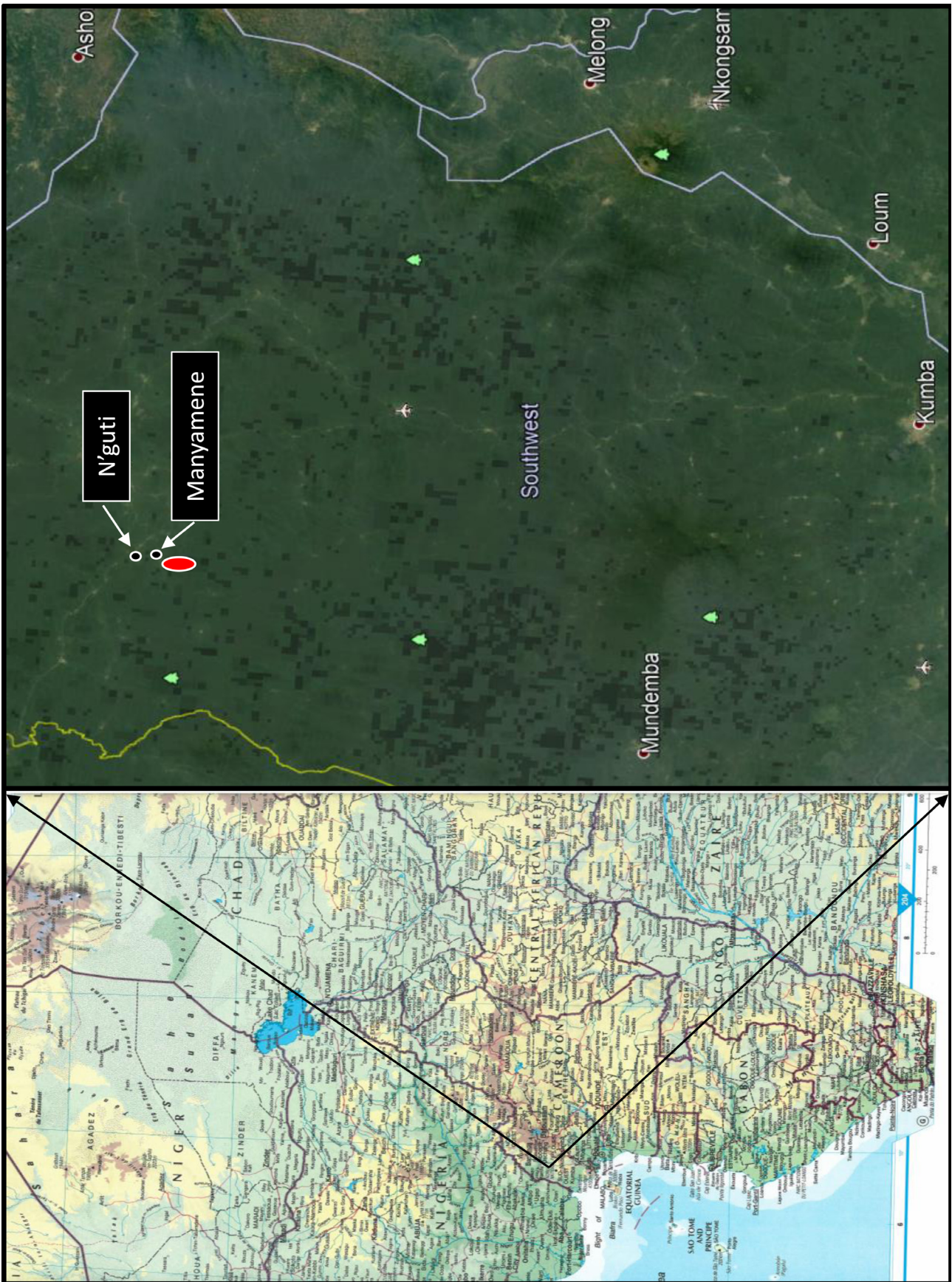


FIGURE 1. Location of the Talangaye Forest where mosquitoes were collected. The area demarcated in red represents the specific area where mosquitoes were collected (image courtesy of Google Earth Pro).

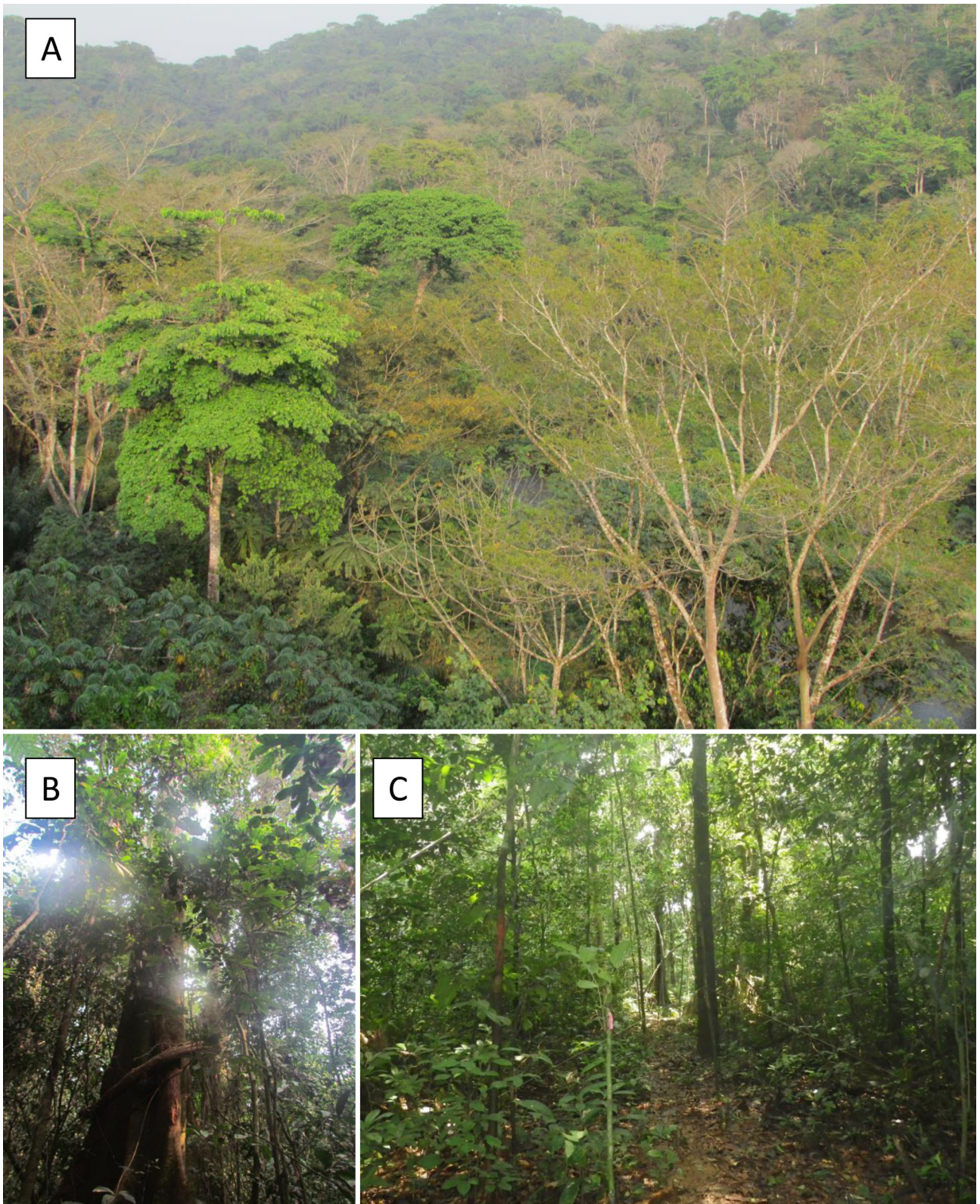


FIGURE 2. General vegetation of the Talangaye Forest. (A) Aerial view; (B) typical large tree forming the upper canopy; (C) broad leaf vegetation below the canopy from ground level to several meters high.



FIGURE 3. Trapping methods used to collect *Culiciumyia*. (A) Net trap; (B) walk-in resting cage; (C) bamboo pot holding water attached to a tree branch or trunk within which mosquitoes laid eggs.

Taxonomic treatment

General collection summary. In the Talangaye Forest, the majority of the *Culiciumyia* males were collected by

sweep netting through vegetation, followed by a few collected in the mornings and early evenings in the resting box and in bird-baited and un-baited net traps. No *Culiciomyia*, in fact no mosquitoes, were collected in the modified miniature CDC traps placed in the forest, whether placed near to the ground or higher up towards the canopy. Larvae of *Cx. nebulosus* were collected in tree holes and in bamboo pots. During the two years, with four collection trips per year, *Cx. nebulosus* was the most collected species ($n = 81$ specimens) followed by *Cx. macfieii* ($n = 54$), *Cx. cinereus* ($n = 14$), *Cx. cinerellus* ($n = 10$), *Cx. pseudosubaequalis* **sp. n.** ($n = 6$), *Cx. lanzaroi* **sp. n.** ($n = 4$), *Cx. semibrunneus* ($n = 3$), *Cx. apicopilosus* **sp. n.** ($n = 2$) and one each of *Cx. eouzani*, *Cx. subaequalis* and *Cx. semibrunneus*. As mentioned above, females were also collected mostly resting in vegetation and a few in resting cage net traps. Because multiple *Culiciomyia* species were collected, as determined from male morphology, and females of African *Culiciomyia* resemble each other, based on our experience, we did not hazard guessing the identification of females. Instead, we pooled them all as *Culiciomyia* for later isolation of avian malarial parasites.

Descriptions of the males of the three new species are given first, followed by descriptions for previously named species that were also collected in the Talangaye Forest.

***Culex (Culiciomyia) apicopilosus* Cornel & Mayi, sp. n.**

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Male. Wing 3.40 mm. Proboscis 2.20 mm. Forefemur 1.80 mm. Thoracic pleura generally with grey dusted appearance typical of most other Afrotropical *Culiciomyia* (Fig. 4A).

Head: Decumbent scales of vertex narrow and white to translucent, except for broader white scales along eye margin and on sides of head (Fig. 4D). Erect forked scales quite numerous and dark brown, those in middle of vertex with slightly lighter coloured tips. Maxillary palpus slightly longer than proboscis, palpomeres 4 and 5 bearing setae (Fig. 4B), palpomere 4 0.4 length of palpomere 5; palpal comb of palpomere 3 (Fig. 4B,C) consists of translucent scales: proximal 6 scales all longer than distal 10–12 scales, scales of proximal set or group lanceolate, variable in length, those more apically positioned slightly shorter than more basal scales, distal 10–12 scales closer together with rounded (spatulate or oblanceolate) pre-apices and minute short folded points (recurved aristate apices), no obvious gap between proximal and distal sets of scales. Antenna slightly shorter than proboscis, apical flagellomeres covered with short setae except for whorl of 7 or 8 long setae at base of terminal flagellomere, all flagellomeres, except distal 2, with apical 0.50 white and proximal 0.50 brown. Proboscis entirely dark-scaled.

Thorax: Scutal integument greyish brown (Fig. 4D), covered with short brown setae, scales at front and on prescutellar area slightly paler; scales on scutellum dirty white. Pleural integument uniformly greyish brown (Fig. 4A), pleura with scales and setae on following regions (Fig. 4E): anteprepronotum, postprepronotum (row of 6 on posterior margin), proepisternum, prealar knob, row of whitish to translucent scales extending from upper mesokatepisternal patch down posterior margin to lower 0.67 and a single lower mesepimeral seta.

Wing: Scales all dark.

Legs: Fore- and midfemora pale underneath, dark above, anteriorly and posteriorly; hindfemur pale underneath (ventral view) almost to apex, basal 0.5 of anterior, posterior and dorsal surfaces pale, apical 0.5 of dorsal and posterior surfaces dark, anterior pale area gradually narrows beyond halfway towards apex and ends about 0.25 before dark apex (Fig. 9D); tibiae and tarsi of all legs dark-scaled.

Abdomen: Terga uniformly brown with slightly paler small basal lateral spots; sterna uniformly light brownish to grey.

Genitalia: Tergum IX lobes each with an irregular row of 12 setae. Gonocoxite with few setae on lateral, ventral and dorsal margins longer than gonocoxite (Fig. 5A); ventral side with a dense lateral patch of 10 rows of setae, about 7 setae in each row obscure and make setae of subapical lobe difficult to see, especially when viewed ventrally (Fig. 5B); subapical lobe prominent, weakly divided, small lobe below setae *a–c* with one seta and larger lobe above bears setae *a–c*, a hair-like setae (*e*), 4 setae bent and barbed distally ($d^1–d^4$), a broad foliform (leaf-like) seta (*g*) and a smaller foliform seta (*f*) (Fig. 5A,B), base of seta *c* slightly separate from bases of setae *a* and *b*, seta *b* often appears darker than *a* and *c*, all 3 with hooked apices; seta *e* as long as setae *a–c* and with slightly striated appearance; setae $d^1–d^4$ each with very delicate expanded lightly sclerotized ends with 3 or 4 barbs (Fig. 5A,B), expanded apical ends of d^3 and d^4 larger than those of d^1 and d^2 ; seta *f* about 0.67 width of seta *g*; ventral patch of 8–10 setae at apex of gonocoxite just below base of gonostylus. Gonostylus with a row of about 12–14 “spinelets” distal to a dorsal crest (Fig. 5A,C), an upturned small hook at proximal end of crest; 3–5 setae on gonostylus where

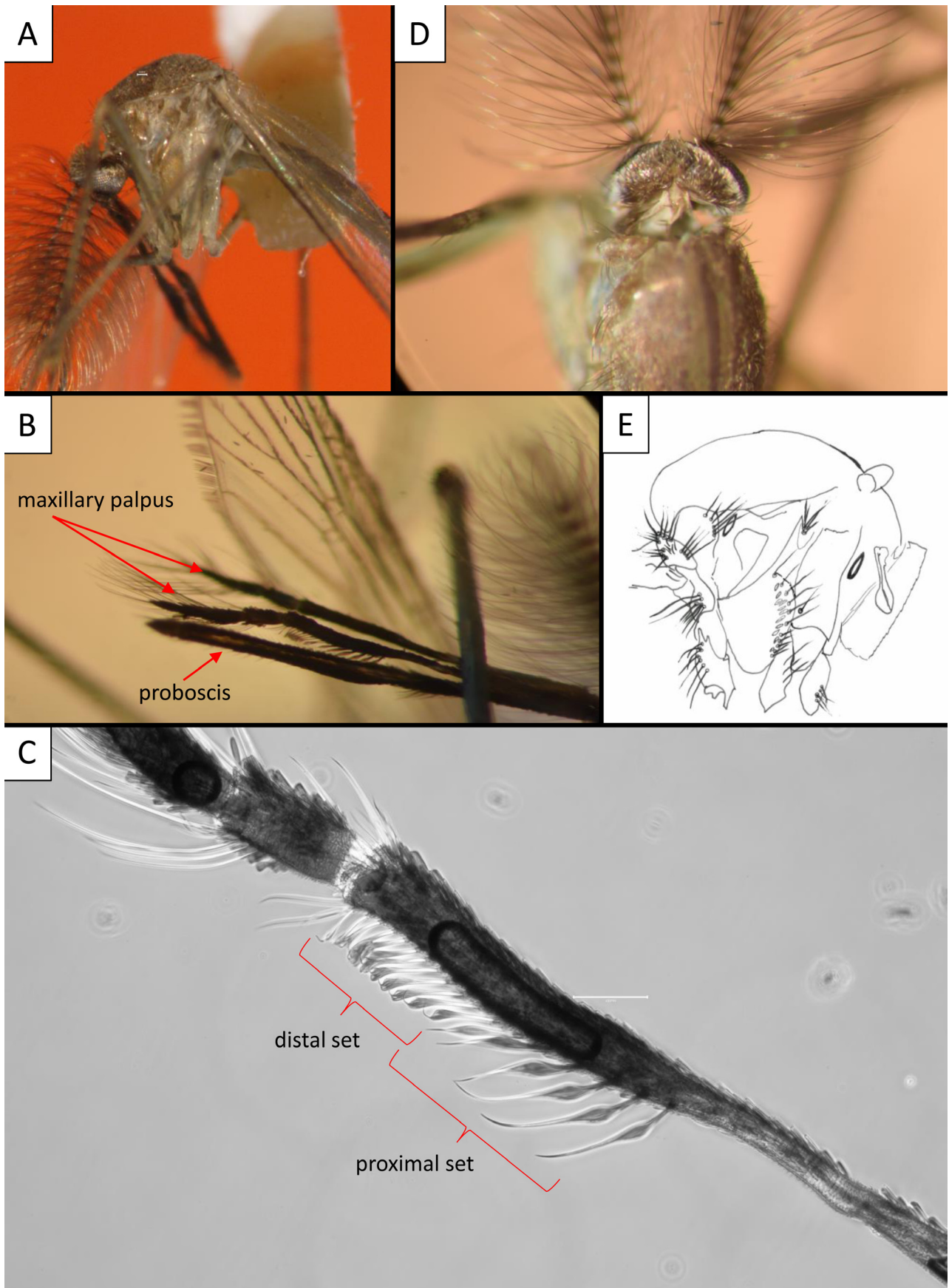


FIGURE 4. External features of *Culex apicopilosus*. (A) Lateral view of thorax; (B) lateral view of proboscis and maxillary palpi; (C) lateral view of the palpal comb scales (100x); (D) dorsal view of the head and anterior area of the scutum; (E) drawing showing the location of setae and scales on the thoracic pleura.

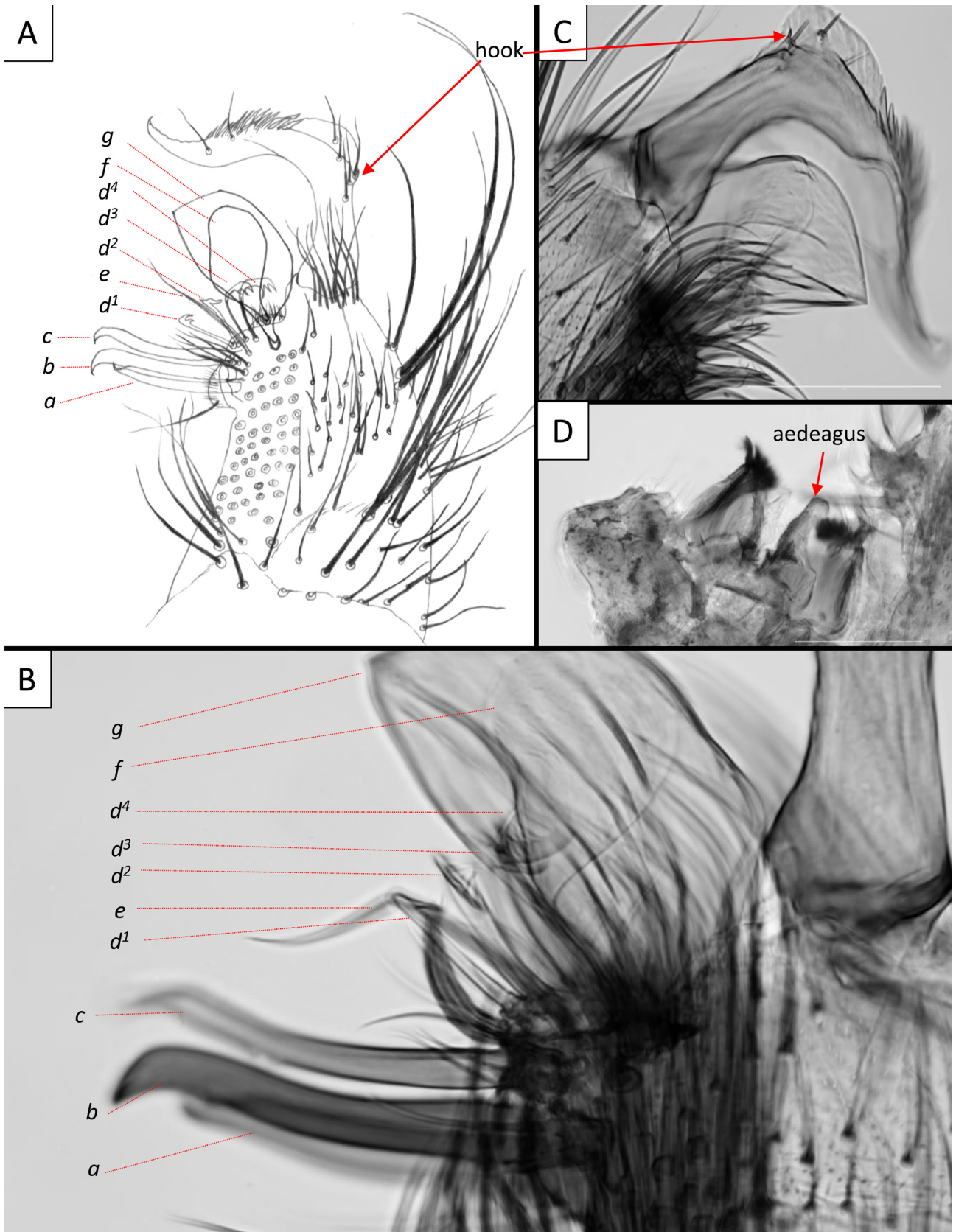


FIGURE 5. Male genitalia of *Culex apicopilosus*. (A) Depiction of the dorsal view of the gonocoxite; (B) subapical lobe (600x); (C) gonostylus (400x); (D) aedeagus and paraproct (100x).

it bends at base of crest and 2 subapical setae near distal end of row of “spinelets” (Fig. 5A,C); apex of gonostylus upturned and hook-like (Fig. 5A). Aedeagus with expanded base, with a single basal tooth and arms (lateral plates) that are more or less straight with rounded to pointed apices, depending on angle of observation (Fig. 5D), each arm with a row of small tubercles along inner margin that almost reach apex. Paraproct with 4 subapical cercal setae; no obvious sternal process.

Additional notes: The legs of all Afrotropical *Culicomyia*, except for *Cx. cambournaci*, are similar in colouration with no reliable differences (Figs 9D, 16E, 31C). *Culex apicopilosus* closely resembles *Cx. nebulosus* and *Cx. lanzaroi*. The structure of the aedeagus is identical in these species. In all three species, the tips of the lateral plates can appear round or pointed depending on the angle of observation. *Culex apicopilosus* differs from both *Cx. lanzaroi* and *Cx. nebulosus* by the presence of an apical patch of 8–10 setae on the gonocoxite (Figs 5A, 6A). *Culex nebulosus* and *Cx. lanzaroi* lack this patch and instead have 1–4 setae (Fig. 6B) that are positioned in a row if more than one seta is present. The subapical lobe of *Cx. nebulosus* has two distinctly apically barbed setae, *Cx. lanzaroi* has three and *Cx. apicopilosus* has four. *Culex apicopilosus* and *Cx. lanzaroi* share the unique presence of two broad foliform setae, unlike *Cx. nebulosus* which has one broad and one much narrower foliform seta. It is noteworthy that one of the foliform setae is smaller in *Cx. apicopilosus*, whereas in *Cx. lanzaroi* these setae are almost equally broad. *Culex lanzaroi* has the smallest membranous dorsal flange (crest) on the gonostylus, *Cx. nebulosus* has the largest and that of *Cx. apicopilosus* is of an intermediate size. The two males used to describe *Cx. apicopilosus* have more numerous (11 or 12) distal palpal comb scales compared to males of *Cx. nebulosus*, which have 8 or 9 palpal comb scales, and those of *Cx. lanzaroi* which have 6 or 7. Furthermore, there is an obvious gap separating the differently shaped proximal and distal sets of scales in *Cx. nebulosus* and *Cx. lanzaroi*, whereas there is no gap in *Cx. apicopilosus*.

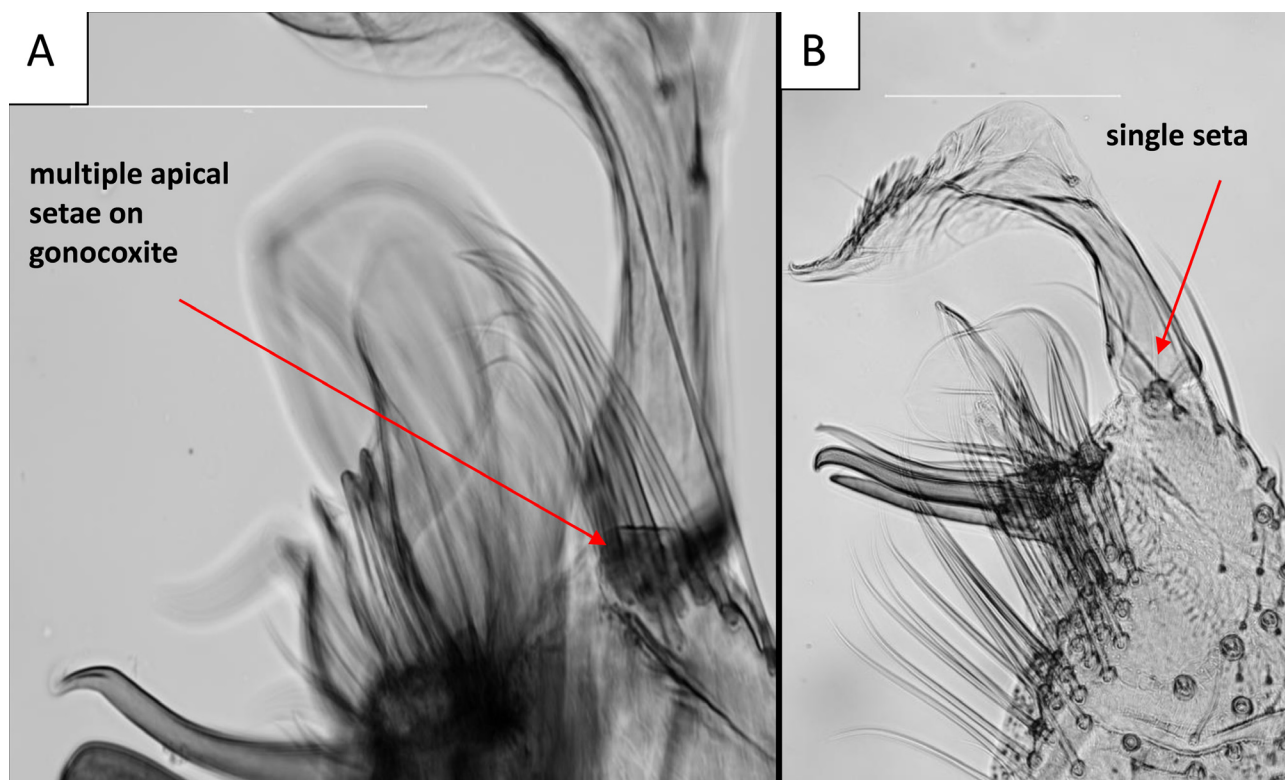


FIGURE 6. (A) Apical region of the gonocoxite (400x) of *Culex apicopilosus*; (B) gonocoxite (100x) of *Culex nebulosus*.

Etymology: This species is named *apicopilosus* in reference to the group or patch of setae occurring at the apex of the gonocoxite.

Type series. Holotype male (BMEC type # 1919) collected resting in forest vegetation in CAMEROON, South-West Region, Nguti Subdivision, Talangaye Forest (5.190397° N; 9.3457790° E), with the following label: Talangaye forest, SE Region, Cameroon, forest floor sweep net, Date of collection 16/vii/2017, Acc. # CAM202hc 16/vii/2017, Adult ♂, gen and palp mounted, Coll and Det. A. J. Cornel. Paratype male collected on a different date to holotype otherwise with same data except for different Acc. # CAM 202rb 12/vii/2017, Acc. # is equivalent to

the collection number. Slide mounts of genitalia and maxillary palpi of both types have the same label as the corresponding pinned specimen. The holotype and paratype are deposited in the Bohart Museum, University of California, Davis, California, USA.

***Culex (Culiciomyia) lanzaroi* Cornel & Mayi, sp. n.**

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Male. Wing 2.85 mm. Proboscis 1.90 mm. Forefemur 1.55 mm. Thoracic pleura generally with grey to light brown appearance, degree of brown colouration varies from light brown in some specimens to uniformly light grey in others.

Head: Decumbent scales of vertex narrow and white to translucent except for a single row of broader white scales along eye margin and on each side of head (Fig. 7A). Erect forked scales quite numerous, dark brown, those in middle of vertex with slightly lighter coloured tips. Maxillary palpus slightly longer than proboscis, palpomeres 4 and 5 bearing setae; palpal comb of palpomere 3 (Fig. 7B) consists of translucent scales: proximal 6 or 7 scales lanceolate and longer than distal set of 6 or 7 scales, distal scales attached close together with rounded (spatulate or oblancoelate) pre-apices and short recurved aristate apices. Distinct gap between proximal and distal sets of scales. Antenna slightly shorter than proboscis, apical flagellomeres covered with short setae except for whorl of 7 or 8 long setae at base of terminal flagellomere, all flagellomeres, except last 2, with apical 0.50 white and proximal 0.50 brown. Proboscis entirely dark-scaled.

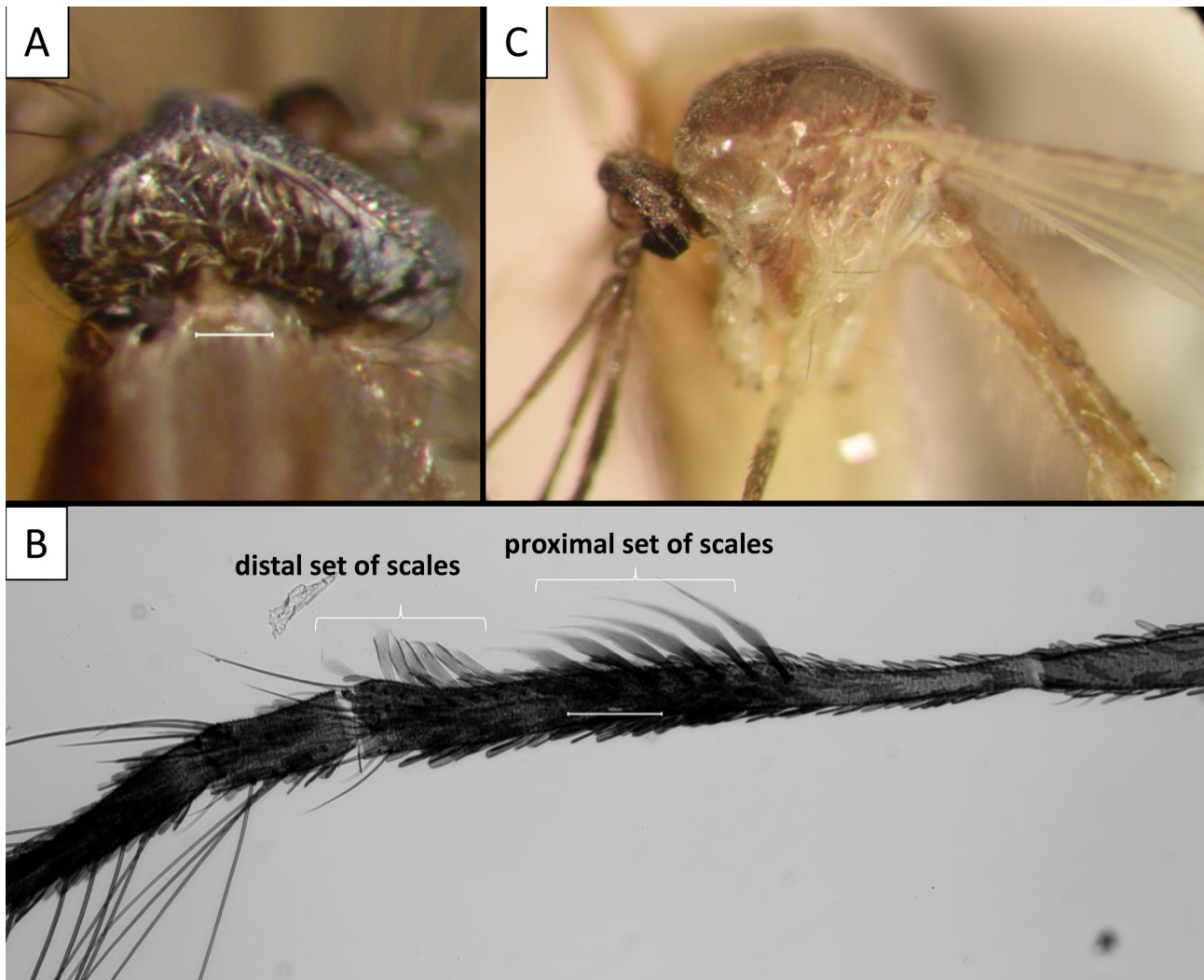


FIGURE 7. External features of the male of *Culex lanzaroi*. (A) Dorsal view of the head (vertex); (B) lateral view of the palpal comb scales (100x); (C) lateral view of the thorax.

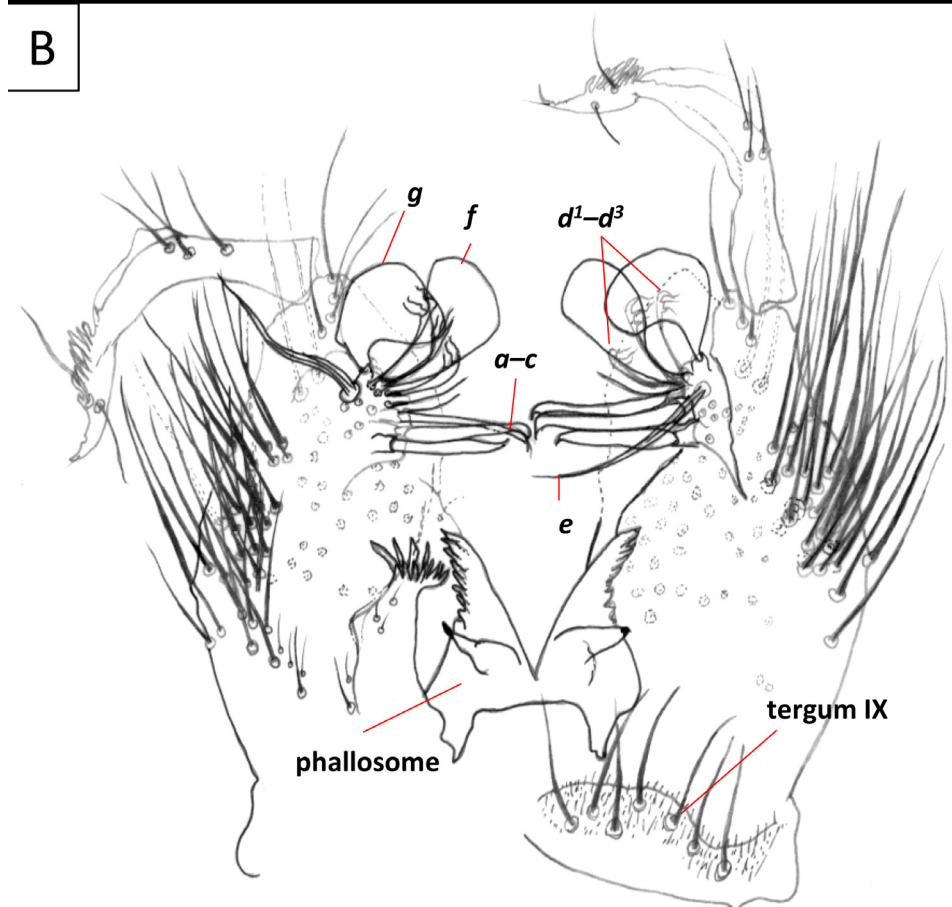


FIGURE 8. Male genitalia of *Culex lanzaroi*. (A) Image (100x); (B) drawn depiction.

Thorax: Scutal integument light brown (Fig. 7C), sparsely covered with short brown setae, scales at front and on prescutellar area slightly paler, scales on scutellum dirty white. Most of pleural integument same light brown colour as scutal integument (Fig. 7C), mid-region of mesokatepisternum, lower 0.50 of mesepimeron, mesomeron and integument of coxae sometimes lighter brown to beige, pleura with setae on following areas: antepnotum, postpronotum (row of 6 on posterior margin), proepisternum, prealar knob, row of narrow translucent scales (easily overlooked and rubbed off) extending from upper mesokatepisternal patch down posterior margin to lower 0.67, and a single lower mesepimeral seta.

Wing: Scales all dark.

Legs: Colouration identical to legs of *Cx. apicopilosus*.

Abdomen: Terga uniformly greyish brown with slightly paler small basal lateral spots; sterna uniformly brownish grey.

Genitalia: Tergum IX lobes each with row of 7 setae (Fig. 8B). Gonocoxite with ventrolateral patch of long setae (each seta approximately 0.67 length of gonocoxite) in 6 irregular rows of 4 or 5 setae (Fig. 8A,B); dorsal surface mostly devoid of long setae except for 3 medial and 2 subapical; prominent undivided subapical lobe with 3 broad rod-like setae (*a–c*), a longer quite stout striated seta (*e*), 3 barbed setae (*d^l–d^b*), 2 more or less equally broad foliform setae (*f* and *g*) and 6 finer setae; seta *b* often appears darker than *a* and *c* (Fig. 8A). Gonostylus with row of about 10 “spinelets” distal to membranous dorsal crest, 3 or 4 setae on gonostylus where it bends at base of membranous crest and subapical setae near distal end of row of “spinelets”; apex of gonostylus upturned and hook-like (Fig. 8A,B). Aedeagus with expanded base bearing a single basal tooth and arms (lateral plates) that curve slightly outwards, lateral plate with numerous teeth along outer margin that extend up to a sharply pointed apex (Fig. 8A, B). Paraproct with subapical setae; no obvious sternal process.

Additional notes: *Culex lanzaroi* males very closely resemble the males of *Cx. nebulosus* except for obvious differences in the size and shapes of the foliform setae on the gonocoxite and scales of the palpal comb of palpomere 3. *Culex lanzaroi* has two broad foliform setae on the gonocoxite, one of which is quite oddly shaped and not round like the other, whereas in *Cx. nebulosus* there is one broadly rounded foliform seta and one much narrower foliform seta. *Culex lanzaroi* has 8 or 9, mostly 8, setae at the base of the subapical lobe whereas *Cx. nebulosus* has 5 or 6. *Culex lanzaroi* has 6 or 7 proximal and 6 or 7 distal palpal comb scales on palpomere 3, whereas *Cx. nebulosus* has 7 or 8 proximal and 8 or 9 distal scales.

Etymology: This species is named in honour of Dr. Gregory Lanzaro (medical entomologist, Department of Pathology, Microbiology and Immunology, University of California, Davis), who has made significant contributions on the systematics and population genetics of North American and African mosquitoes.

Specimens examined: The holotype male (BMEC type # 1920) was collected resting inside the field processing tent in CAMEROON, South-West Region, Nguti Subdivision, Talangaye Forest (5.190397° N; 9.3457790° E) with the following label: Talangaye forest, SE Region, Cameroon, resting in tent. Acc. # CAM213f 15/vii/2017, Adult ♂, gen and palp mounted, Det. A. J. Cornel. Three paratype males collected sweep netting through vegetation on the forest floor within 0.50 km of the processing tent where the holotype was collected. The paratypes have labels indicating their location of collection (Talangaye forest, SE Region, Cameroon, resting in forest vegetation) and their respective Acc. #s of CAM 163gb 01/v/2017, CAM 202ra 12/vii/2017 and CAM 202ja 14/vii/2017 (dates of collection correspond to date on Acc. #s). Slide mounts of the genitalia have the same labels as the corresponding male carcass. The holotype and paratypes are deposited in the Bohart Museum, University of California, Davis, California, USA.

Culex (Culiciomyia) pseudosubaequalis Cornel & Mayi, sp. n.

Male. Wing 2.70 mm. Proboscis 1.90 mm. Forefemur 1.40 mm. Pleura with an upper dark stripe.

Head: Scaling on vertex same as in *Cx. lanzaroi* except for more numerous white broad decumbent scales on sides of head (Fig. 9A). Maxillary palpus longer than proboscis because of longer palpomere 4, which is little more than 0.5 length of palpomere 5 (ratio = 0.54–0.67; see Fig. 10A,C); palpomeres 4 and 5 setose; row of 7–9 translucent spearhead-shaped (subulate with a long cuneate base) comb scales on palpomere 3, apices of scales extend into long thin delicate points (Fig. 10A,C). Antenna and proboscis identical to those of all other *Culiciomyia* (see description of *Cx. apicopilosus*). Proboscis dark-scaled.

Thorax: Scutal integument light to dark brown, covered with short brown setae, narrow white scales on scutellar lobes. Pleura non-uniform in colour (Fig. 9B,C). Integument darker on antepronotum, middle 0.30 of postpronotum, postspiracular area, upper 0.50 of subspiracular area, prealar knob and upper 0.30 of mesanepimeron, giving appearance of an upper dark stripe; lower light brown semi-stripe corresponds to light brown colouring of proepisternum, lower part of subspiracular area and upper mesokatepisternum; one specimen with upper mesokatepisternum not noticeably darkened (Fig. 9B); pleura devoid of scales, setae present on antepronotum, posterior margin of postpronotum, 3 on proepisternum, prealar knob, upper and posterior margin of mesokatepisternum and 1 lower mesepimeral seta. Pedicel of halter lighter than capitellum.

Wing: Scales all dark.

Legs: Colouration of legs like legs of *Cx. apicopilosus* (Fig. 9D).

Abdomen: Terga uniformly brown; sterna uniformly brownish grey to greyish beige.

Genitalia: Tergum IX lobes each with a row of 7 seta. Ventral surface of gonocoxite with a few long setae (each as long or a little longer than gonocoxite) in no obvious configuration and a few rows of 3 or 4 narrow setae along inner margin (Fig. 11); dorsal surface with sparsely distributed short setae and a single stouter seta on a slightly raised lobe below base of gonostylus (Fig. 11); a single but not very prominent subapical lobe bearing setae of similar shape and configuration as in *Cx. subaequalis* that includes 3 rod-like setae (*a–c*) with *a* shortest and *b* longest, a single long quite robust and slightly striated seta (*e*) less stout than in *Cx. subaequalis*, 2 small barbed seta (*d¹–d²*), quite broad foliiform seta (*g*) and a smaller less broad membranous seta (*f*). Gonostylus with crest of approximately 6 “spinelets”, a short rounded dorsal membranous flange, apex upturned and hook-like, and a small broad membranous seta, 1 seta below middle of crest and 1 borne subapically, a single small seta approximately 0.33 from base on upper margin occasionally present (Fig. 11). Aedeagus with expanded base and prominent basal tooth (Fig. 12A,B), otherwise devoid of teeth or tubercles, lateral plate quite long and gradually narrowed towards rounded apex, extending upwards and more or less straight. Paraproct with 3 subapical setae and a small rounded basal protrusion (Fig. 12A,B).

Additional notes: The thoracic pleura are not uniform in colour. Upper dark grey and lower light brown colouring of some portions of the integument create the appearance of a light grey mid-region (Fig. 9C). Other differences distinguishing *Cx. pseudosubaequalis* from *Cx. subaequalis* include the lack of rows of ventrolateral setae on the gonocoxite, a smaller basal protuberance on the paraproct and shorter palpomere 4 relative to palpomere 5. The longer palpomere 4 can confuse this species with *Cx. subaequalis* and *Cx. mongiro*. In *Cx. subaequalis* and *Cx. mongiro* palpomere 4 is 0.75 the length of palpomere 5, whereas in *Cx. pseudosubaequalis* it is slightly shorter and 0.55–0.65 the length of palpomere 5. *Culex mongiro* is quite unique among the Afrotropical *Culiciomyia* by having banded abdominal sterna that are dark basally and lighter apically (van Someren 1951). *Culex mongiro* also has no basal protrusion on the paraproct and there are no barbed setae on the subapical lobe of the gonocoxite. *Culex subaequalis* and *Cx. pseudosubaequalis* have a basal protrusion on the paraproct and barbed accessory setae on the subapical lobe of the gonocoxite. *Culex mongiro* shares with *Cx. pseudosubaequalis* the lack of a ventrolateral patch of long setae on the gonocoxite, which is present in *Cx. subaequalis*. The mesokatepisternum of both *Cx. subaequalis* and *Cx. pseudosubaequalis* lacks white scales. *Culex mongiro*, on the other hand, has white scales on the mesokatepisternum as in *Cx. nebulosus*.

Etymology: This species is named *Cx. pseudosubaequalis* because it closely resembles *Cx. subaequalis*, with maxillary palpomere 4 longer than half the length of palpomere 5. There are significant differences in structures of the male genitalia of *Cx. subaequalis* and *Cx. pseudosubaequalis*.

Specimens examined: The holotype male (BMEC type # 1921) was collected by sweep netting through forest vegetation in CAMEROON, South-West Region, Nguti Subdivision, Talangaye Forest (5.190397° N; 9.3457790° E) with the following label: Talangaye forest, SE Region, Cameroon, resting in tent. Acc. # CAM132na 4/ii/2017, date collected 4/ii/2017, Adult ♂, gen and palp mounted, Det. A. J. Cornel. Five paratypes, collected via sweep netting through forest floor vegetation, within 3 km of the holotype. The paratypes have the same label information as the holotype except for different Acc. #s, which are: CAM 62c 26/VII/2016, CAM 132b1L 31/I/2017, CAM 132yF 31/I/2017, CAM 132d1 6/II/2017 and CAM 202ma 9/VII/2017 (date on the Acc. #s refers to date they were collected). The holotype and paratypes are deposited in the Bohart Museum, University of California, Davis, California, USA.

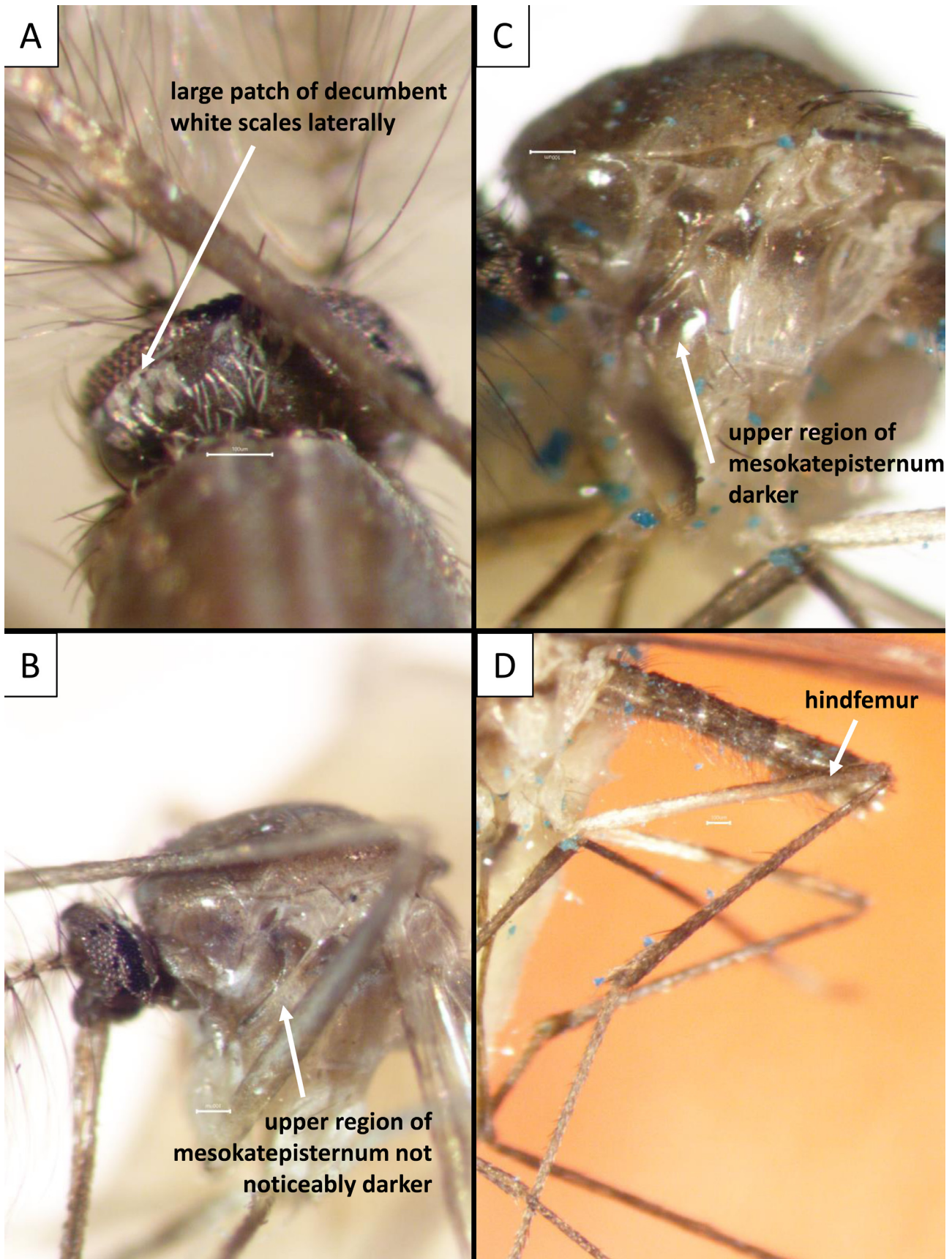


FIGURE 9. External features of the male of *Culex pseudosubaequalis*. (A) Dorsal view of the vertex; (B,C) lateral views of the thorax; (D) anterior view of the hindfemur.

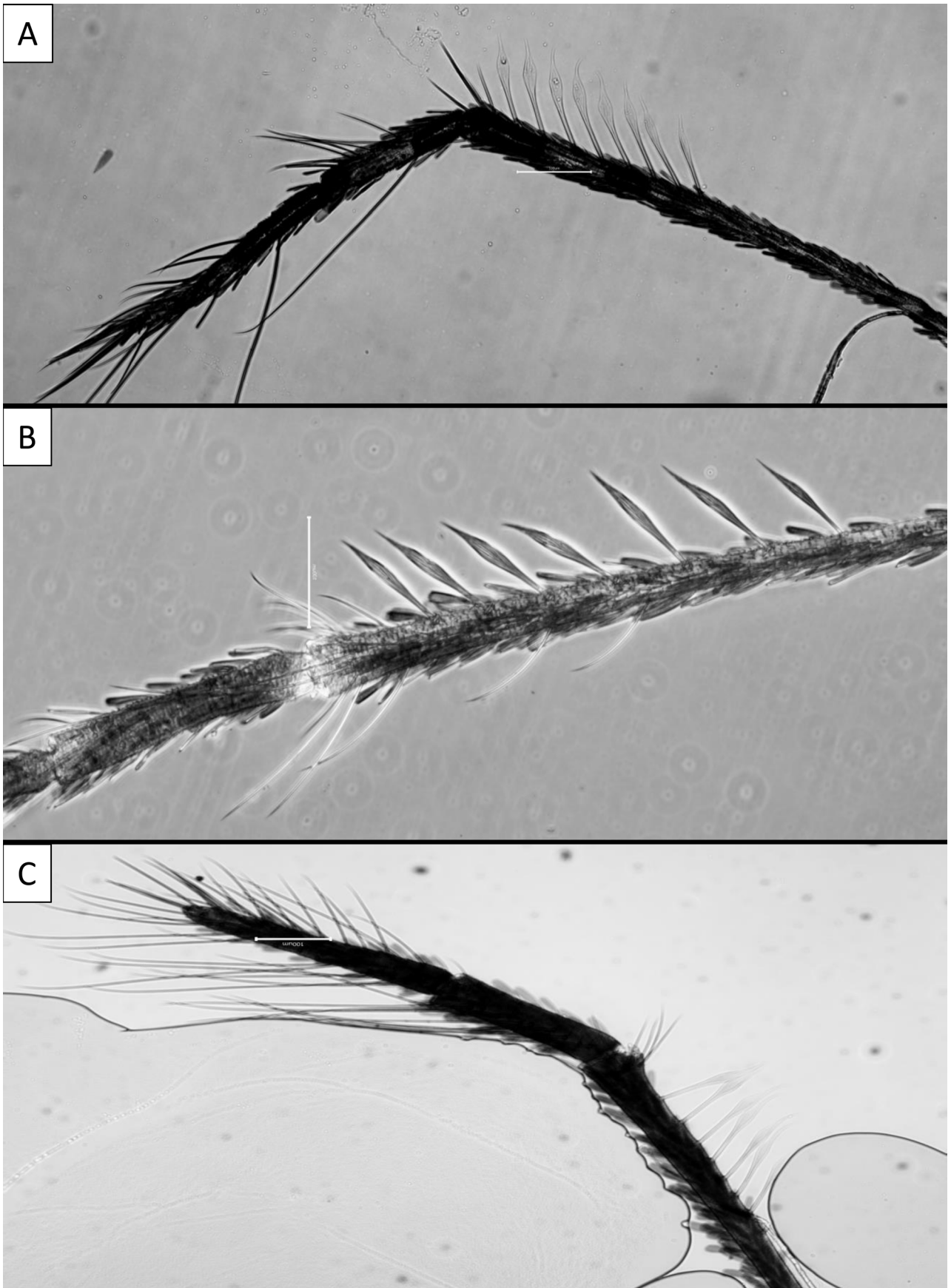


FIGURE 10. Maxillary palpus of *Culex pseudosubaequalis*. (A,B,C) Lateral view of palpal comb scales from three individuals (100x).

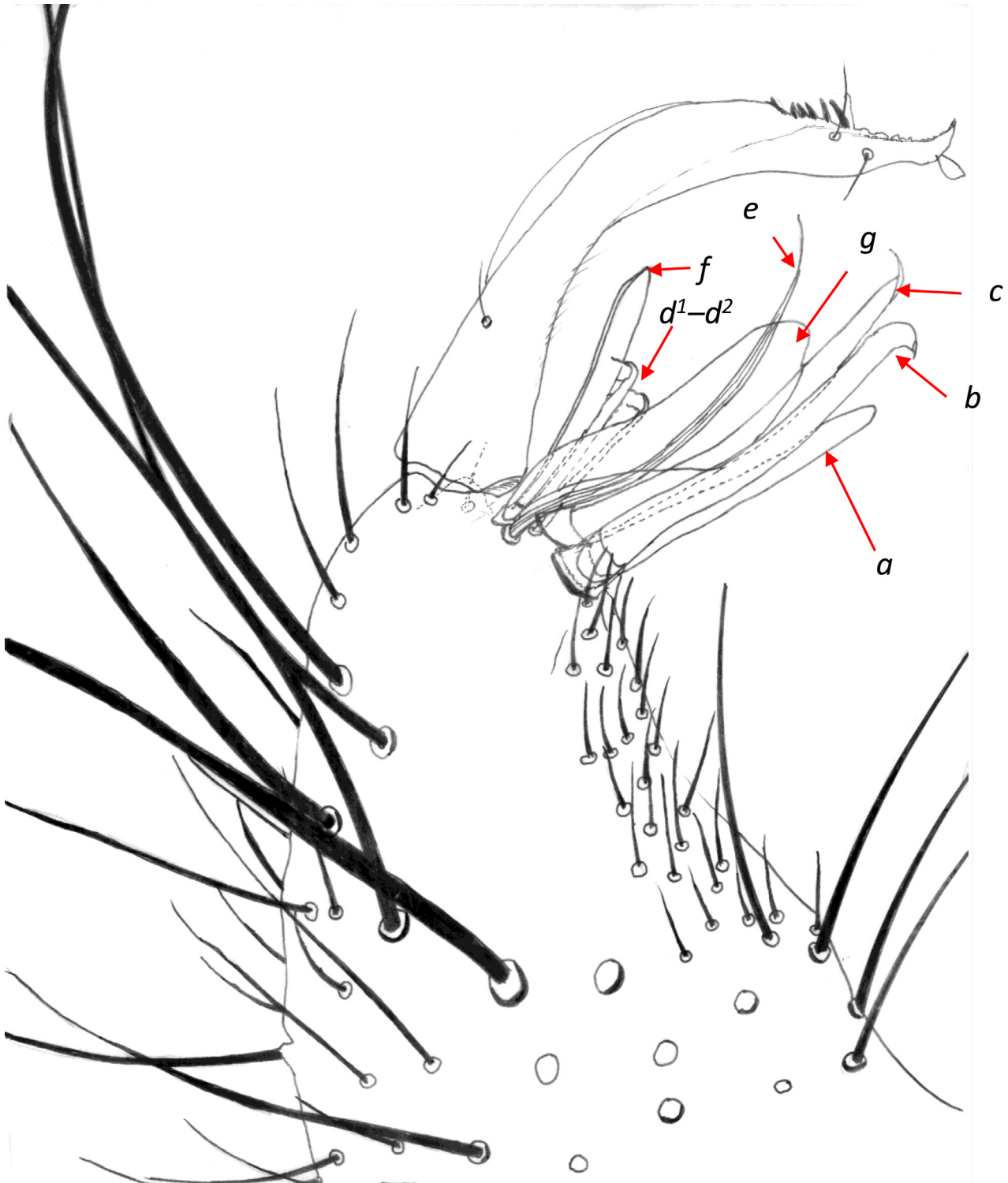


FIGURE 11. Gonocoxite of *Culex pseudosubaequalis*.

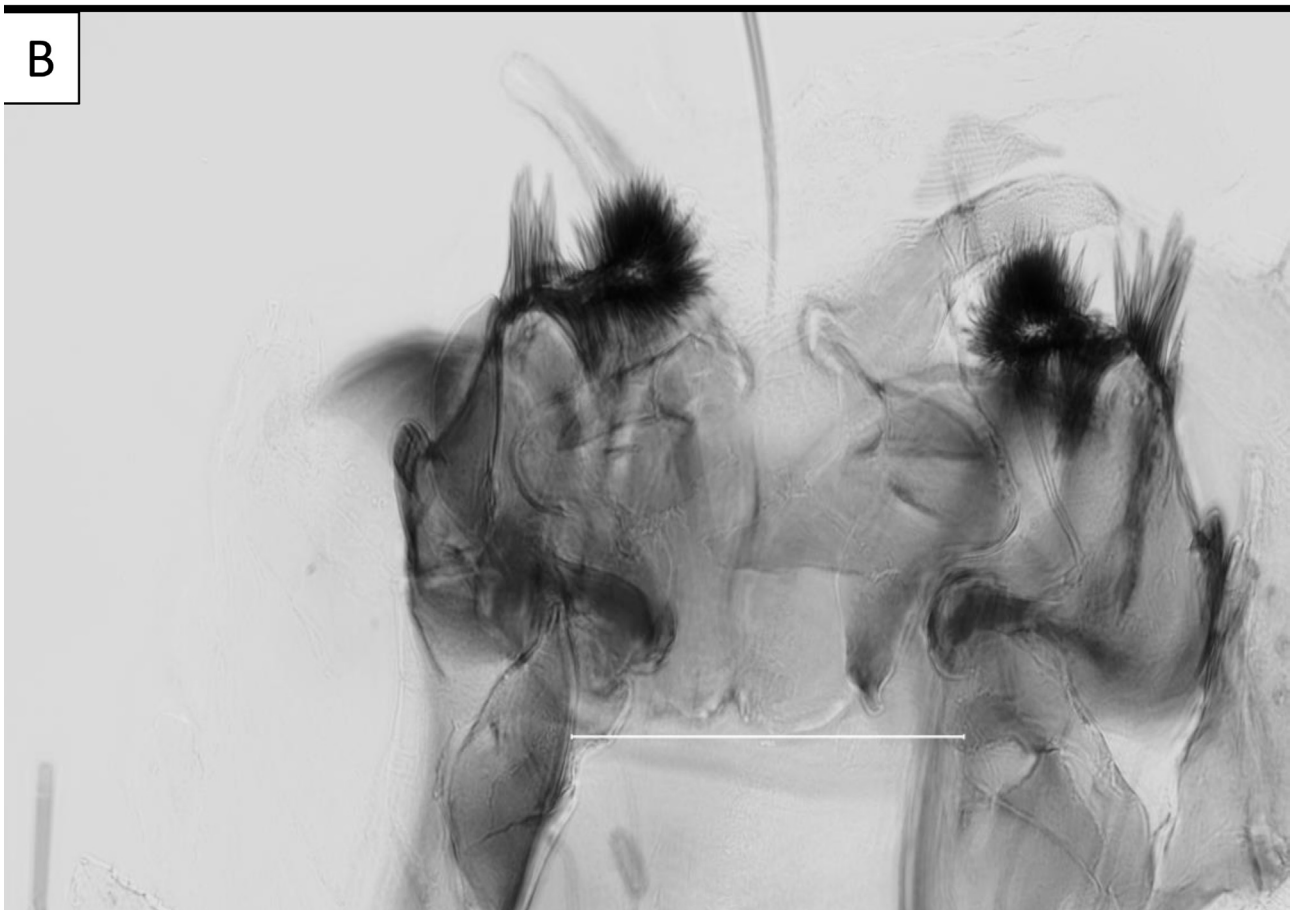
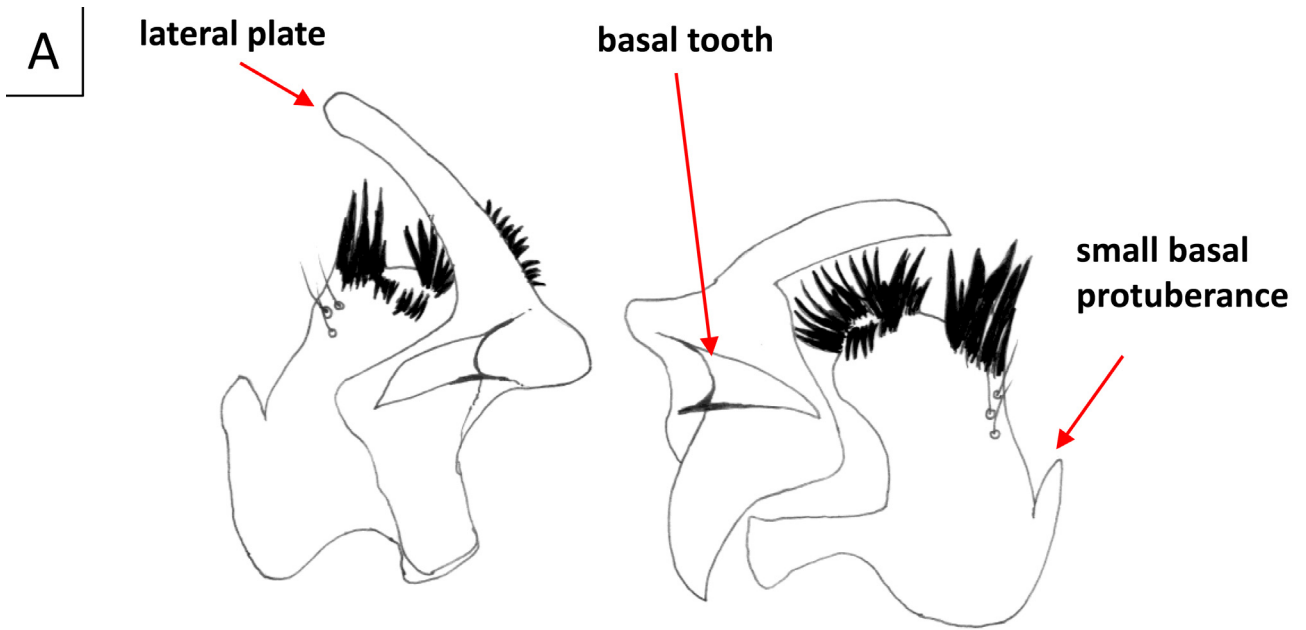


FIGURE 12. Male genitalia of *Culex pseudosubaequalis*, lateral plates and paraprocts. (A) drawn depiction; (B) image (400x).

Culex (Culiciomyia) cambournaci Hamon & Gandara, 1955

Male. This is a small mosquito endemic to São Tomé and Príncipe Islands (West Africa) with a light brown to bronze coloured scutal integument and uniformly beige pleura. The abdominal terga are dark brown with no banding and the sterna are lighter brown with no banding. Hamon & Gandara (1955) provided a detailed description from a single male and putative larva. Wing 1.80 mm. Proboscis 1.55 mm. Forefemur 1.19 mm.

Head: Numerous narrow white decumbent scales in middle and posterior of vertex, irregular row of broad white decumbent scales along eye margin not quite meeting in middle (Fig. 13A) and few broad flat white scales on sides of head; erect forked scales quite numerous and uniformly light brown (Fig. 13A). Maxillary palpus about 0.17 length of proboscis, without comb scales (Fig. 13B). Antenna shorter than proboscis, apical flagellomeres covered with short setae except for whorl of 7 or 8 long setae at base of terminal flagellomere, all flagellomeres, except last 2, with apical 0.5 white and proximal 0.5 brown. Proboscis entirely dark-scaled.

Thorax: Scutal integument light brown, covered with narrow bronze scales (Fig. 13D). Paler scales confined to front of scutum and prescutellar space. Acrostichal setae quite numerous. Pleura integument uniformly pale brown to beige, devoid of scales except for a few on upper mesokatepisternum, some specimens with a few scales on lower mesanepimeron; setae on antepnotum, posterior margin of postpronotum, a few on proepisternum, prealar knob, on upper and posterior margins of mesokatepisternum, and a single lower mesepimeral seta.

Wing: Scales all dark.

Legs: Fore- and midfemora pale underneath, dark anteriorly, posteriorly and dorsally. Most of anterior, ventral and posterior surfaces of hindfemur pale except apical 0.1 dark, dorsal surface all dark (Fig. 13E), rest of leg segments all dark.

Abdomen: Terga dark brown with no pale markings and sterna lighter brown to beige.

Genitalia: Tergum IX lobes each with a single row of 8 or 9 setae. Gonocoxite copiously covered with long setae (as long as gonocoxite) forming an obvious ventrolateral patch; large patch of setae close to base of subapical lobe and many narrow setae on subapical lobe surrounding the specialized setae (Fig. 13C), which consist of 3 rod-like setae, with middle one thinner, lighter in colour and slightly separated from others, thickest rod-like seta flattened to form a small hammer-like head, the third slightly hooked apically; 2 asymmetrical rounded foliform setae accompanied by 2 stiff setae, one strongly striated and thickened in middle. Gonostylus with striated hump where it bends (Fig. 13C), instead of narrowing distally it broadens into an angular upwards spine and has a typical folded foliate gonostylar claw; middle region of gonostylus copiously covered with short setae. Aedeagus with expanded base with long curved basal teeth at least 0.5 length of lateral plate (Fig. 13F). Paraproct with 3 or 4 subapical setae and no obvious sternal process.

Specimens examined: Several males collected resting in low-lying vegetation in a forest (0.12944° N, 6.57944° E) in the southern region of Monte Camo, São Tomé.

Additional notes: This is a rather unusual male of the subgenus *Culiciomyia*, sharing characters, such as the short maxillary palpus and strongly curved gonostylus, with some species, e.g. *Cx. simpliciforceps* Edwards, of the subgenus *Eumelanomyia* Theobald. The larva quite closely resembles the larva of *Cx. nebulosus*. Other than the more strongly bent gonostylus that ends with an expansion, the male genitalia more closely resemble those of *Culiciomyia* species. Hamon & Gandara (1955) classified *Cx. cambournaci* as a species of *Culiciomyia* because the male genitalia mostly resemble those of *Cx. furlongi*. Based on the drawing of the gonocoxite of *Cx. furlongi* in van Someren (1954), the resemblance mentioned by Hamon & Gandara is likely due to the similarly shaped setae of the subapical lobe that lack barbs. Hamon & Gandara did not describe the aedeagus of *Cx. cambournaci* because it was too damaged in their specimen. The aedeagus described from the specimens collected in São Tomé by one of us (AJC) in 2016 resembles that of other species of *Culiciomyia*, with an obvious basal tooth that is much longer than the basal teeth of other Afrotropical species of the subgenus.

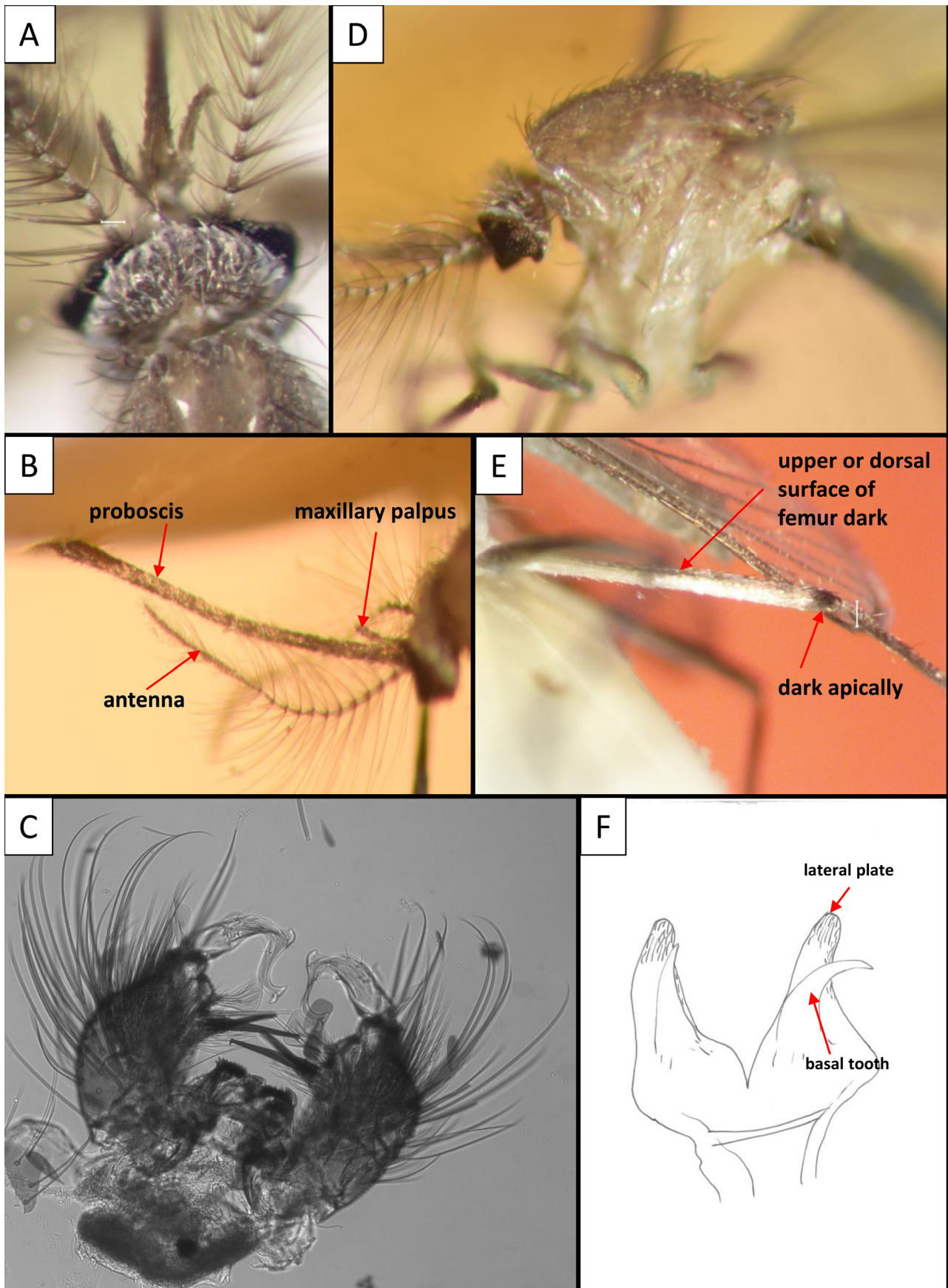


FIGURE 13. Male of *Culex cambournaci*. (A) Dorsal view of the head; (B) lateral view of the maxillary palpus, proboscis and antenna; (C) genitalia (100x); (D) lateral view of the thorax; (E) anterior view of the hindfemur; (F) depiction of the lateral plates.

Culex (Culiciomyia) cinerellus Edwards, 1922

Male. The integument of the thoracic pleura has the appearance of two dark brown stripes contrasting against a middle pale beige stripe. Wing 2.30 mm. Proboscis 1.60 mm. Forefemur 1.20 mm.

Head: Narrow white decumbent scales confined to mid-frontal area of vertex, line of broad white decumbent scales along eye margin, small patches of broad white scales on sides of head (Fig. 14A), light brown erect forked scales have slightly paler apices. Maxillary palpus slightly longer than proboscis, palpomeres 4 and 5 with setae, palpomere 4 approximately 0.4 length of palpomere 5 (ratio 0.39–0.46, mostly 0.4), palpomere 3 with 12–15 comb scales shaped as described by Edwards (1941)—all lanceolate with distal scales slightly closer together than more proximal scales, in all specimens examined most proximal scale is shortest (*a* in Fig. 14B), scales gradually increase in length to middle scales, which are longest (*b* in Fig. 14B), more distal 6 or 7 scales (*c* in Fig. 14B) each of equal length and slightly broader than preceding scales. Antennae and proboscis similar to those of other *Culiciomyia*.

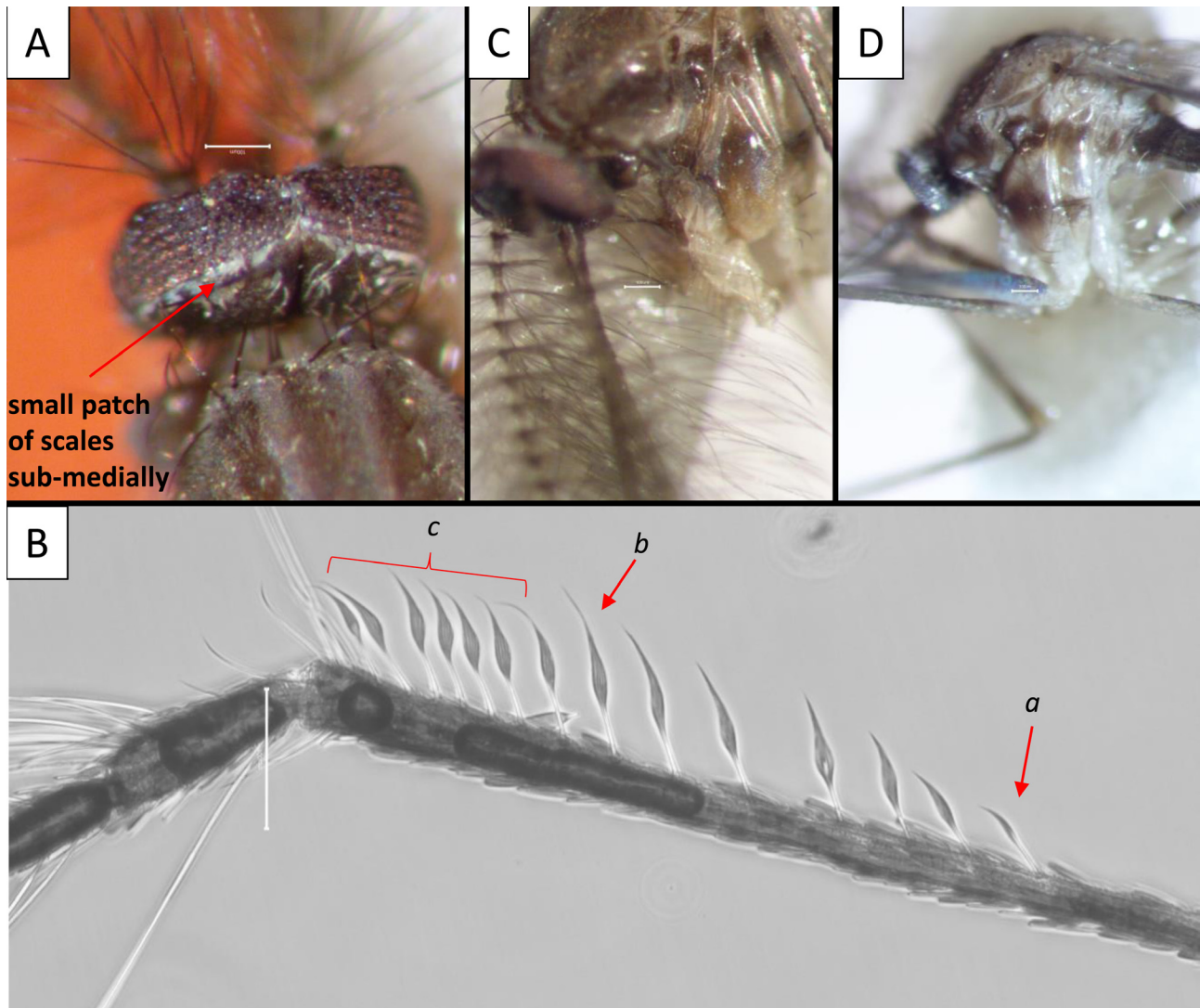


FIGURE 14. External features of *Culex cinerellus*. (A) Dorsal view of the head; (B) lateral view of the palpal comb scales (100x); (C,D) lateral views of the thorax.

Thorax: Scutal integument greyish brown and covered with short brown setae, narrow white scales on scutellar lobes. Thoracic pleura with middle region obviously paler (Fig. 14C,D), antepronotum dark, middle 0.33 of postpronotum, postspiracular area and upper 0.5 of subspiracular area, prelar knob and upper 0.33 of mesanepimeron form an upper dark stripe, lower dark brown stripe corresponds to dark integument of proepisternum, lower 0.5 of subspiracular area, upper mesokatepisternum, lower margin of mesanepimeron and entire mesomeron. Pleura devoid of scales except a few specimens with 3 or 4 scales medially between setae on upper mesokatepisternum and

A

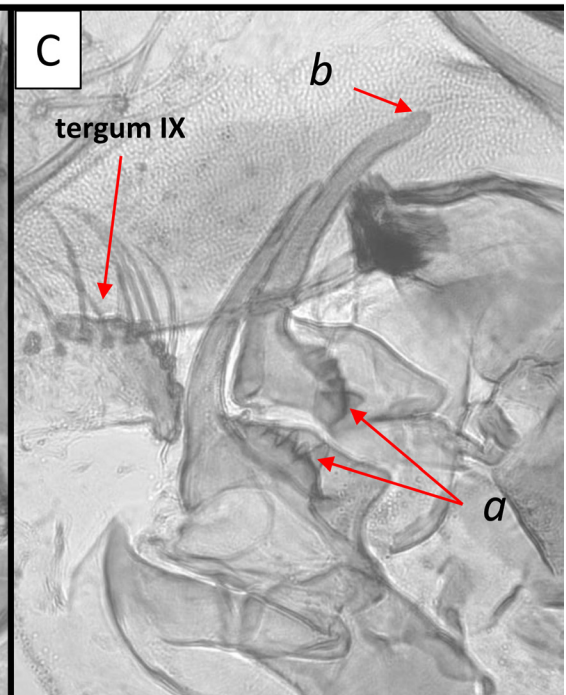
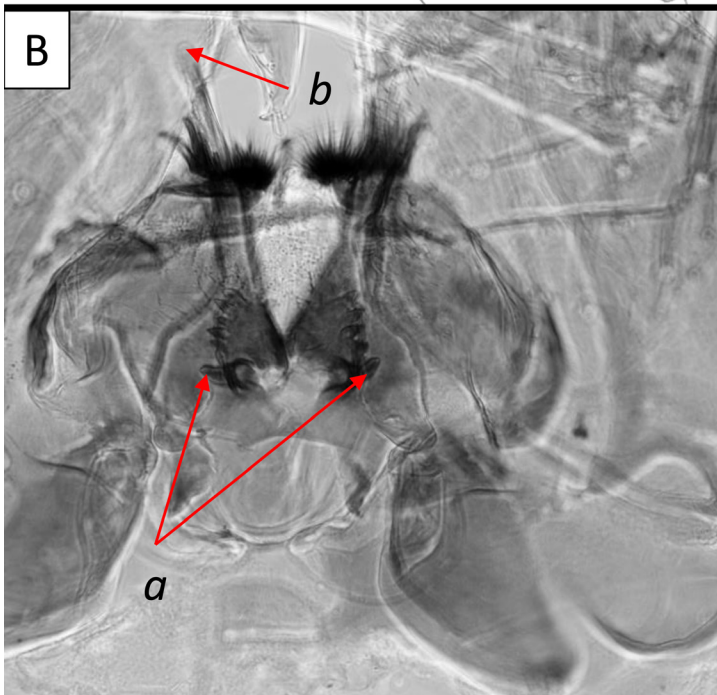
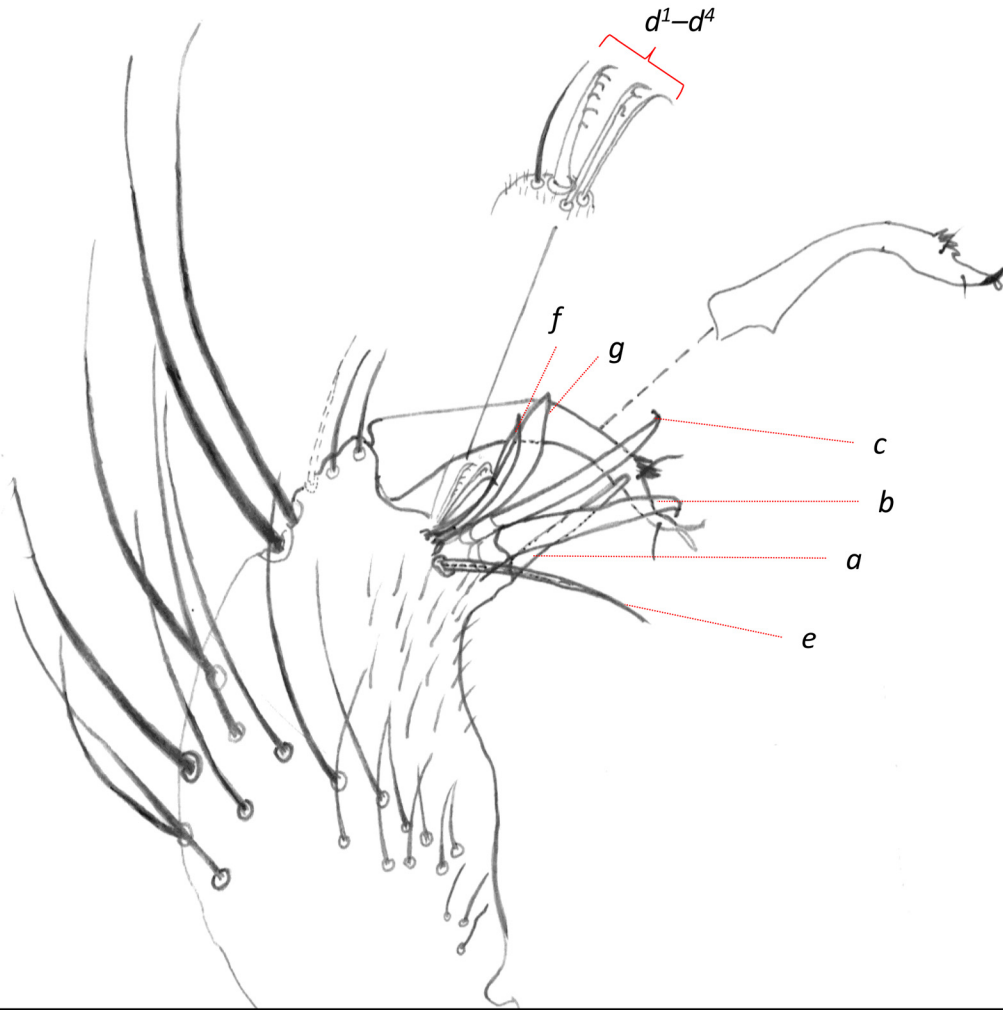


FIGURE 15. Male genitalia of *Culex cinerellus*. (A) Gonocoxite; (B) lateral plates and paraprocts (100x); (C) lateral view of the phallosome (400x).

some along lower 0.50 of posterior margin of lower mesokatepisternum. Setae on antepnotum, row of 6 on posterior margin of postpronotum, 3 on proepisternum, several on prealar knob, a few on upper and posterior margins of mesokatepisternum, and a single lower mesepimeral seta.

Wing: Scales all dark.

Legs: Colouration similar to the legs of other *Culiciomyia*, except *Cx. cambournaci*.

Abdomen: Terga greyish brown with slightly paler small apical lateral spots, sterna uniformly brownish grey.

Genitalia: Tergum IX lobes each with a row of 9 or 10 setae. Ventral surface of gonocoxite with a few long setae in no obvious configuration (Fig. 15A), dorsal surface with sparsely distributed short setae; a single subapical lobe with 3 rod-like setae (*a–c*), most anterior longest and most posterior shortest, a single long slightly stout seta (*e*), 3 small barbed setae (*d^l–d^s*), 1 small seta (*d^t*), 1 relatively narrow foliform seta (*g*) and a smaller slightly narrower foliform seta that is often folded and looking like a slightly flattened blade (*f*). Gonostylus with a crest of 3–6 “spinelets” and devoid of setae except for 1 near crest and 1 borne subapically (Fig. 15A); gonostylus without dorsal membranous flange where it bends downwards, apex upturned and hook-like and with a small apical downward projecting scale-like seta. Aedeagus with expanded base with a row of 5 or 6 rounded tubercles, most proximal larger than the others, which Edwards (1941) probably referred to as “without a well-marked tooth at base” (*a* in Fig. 15B, C); tubercles end where lateral plate narrows sharply, arm of lateral plate long, thin and narrow with blunt apices (*b* in Fig. 15B,C). Paraproct with 2 or 3 subapical setae. Without obvious sternal process.

Additional notes: Edwards (1941) described this species as lacking the grey dusting of the thoracic pleura with the integument “mainly pale yellowish with the upper region more or less obviously darker”. While we agree that this species lacks the greyish colouring, the pleura in fact have two quite distinct dark bands or regions, with the middle 0.33 appearing light brown to almost pale yellowish (much like that seen in some species of the subgenus *Eumelanomyia*). Specimens of this species examined in the National Health Laboratory Services Museum (Johannesburg, South Africa), while being generally paler than those from the Talangaye Forest, still had two visibly darker pleural regions, which makes us consider that the “bicolour” appearance of the pleura is normal for this species. Edwards (1941) omitted to mention the presence of setae *d^l–d^s* on the gonocoxite, but two of these appendages were drawn as non-barbed setae in Jupp (1996).

Specimens examined: Several males collected from the Talangaye Forest in Cameroon by sweep netting through forest vegetation, except for specimen CAM 226b 28/X/2017 which was found resting in an un-baited net trap. Field accession numbers of the other specimens are CAM 132L1b 2/II/2017, CAM 132kb 5/II/2017, CAM 163kb 10/V/2017, CAM 202mc 9/VII/2017, CAM 202q 21/VII/2017, CAM 227ob 7/X/2017, CAM 226b 28/X/2017, CAM 227K 31/X/2017, CAM 227d17a 7/XI/2017 and CAM 227m 8/XI/2017. Some adults reared from larvae collected from a fallen banana leaf in a banana plantation, with accession numbers CAM 12.18.2 3/IX/2011, CAM 12.18pq.1 3/IX/2011 and CAM 12.18pq.2 3/IX/2011 were also examined.

For comparative purposes, a few specimens previously collected by one of us (AJC), and a few deposited in the insect collection of the National Health Laboratory Services Museum in Sandringham, Johannesburg, South Africa, were also examined. These were from: (i) Felixton, Kwazulu Natal, South Africa. *Cx. cinerellus*—1 male. Collected in 1927. No further information on the label. (ii) Yangambi, Stanleyville, Democratic Republic of the Congo. *Cx. cinerellus*—4 males. Collected by Dr. Parent; identified by Botha De Meillon, 1943. (iii) Embotyi, Kwazulu Natal, South Africa. *Cx. cinerellus*—1 male. Collected and identified by Jim Muspratt, 1952. Between the cities of Tiko (4.0786° N; 9.3590° E), and (iv) Douala (4.0511° N, 9.7679° E) in western coastal Cameroon. *Cx. cinerellus*—3 males. Collected and identified by AJC in 2011.

***Culex (Culiciomyia) cinereus* Theobald, 1901**

Male. Thoracic pleura generally greyer and with more dusted appearance than males of other Afrotropical species of *Culiciomyia*. Wing 3.50–3.70 mm. Proboscis 2.10–2.45 mm. Forefemur 2.00 mm.

Head: Decumbent scales of vertex narrow and white to translucent except for single line of broader white scales along eye margin and patch on sides of head (Fig. 16A); erect forked scales quite numerous, dark brown, those in middle of vertex with slightly lighter tips. Maxillary palpus slightly longer than proboscis, distal 2 palpomeres bearing setae, palpomere 4 0.34–0.40 length of palpomere 5; palpal comb of palpomere 3 consists of the following scales (Fig. 16B): proximal set of 11 or 12 scales needle-like or acicular and 8 or 9 in distal set subulate with sharply

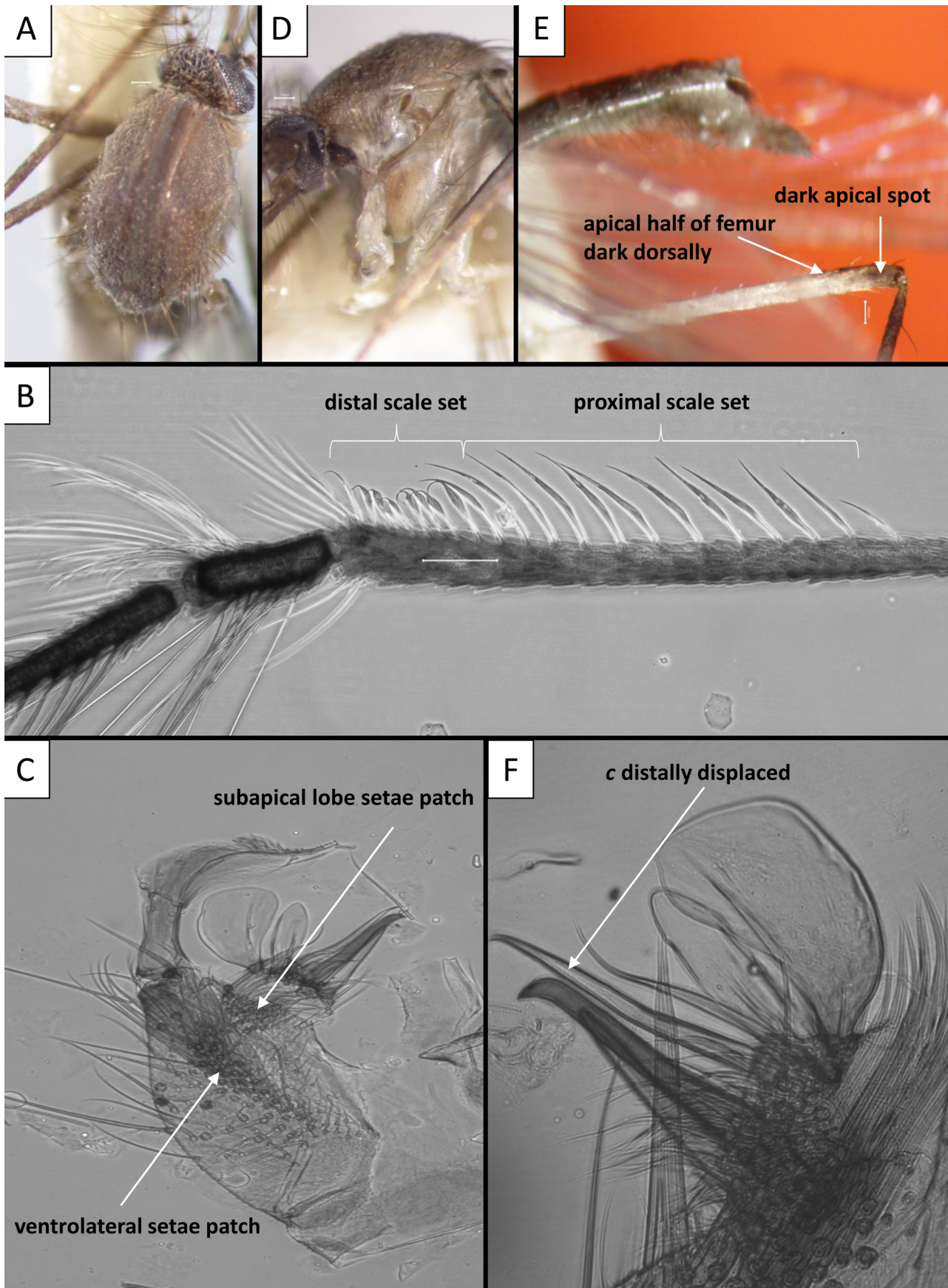


FIGURE 16. External features and genitalia of *Culex cinereus*. (A) Dorsal view of the head and scutum; (B) lateral view of the palpal comb scales (100x); (C) gonocoxite (100x); (D) lateral view of the thorax; (E) anterior view of the hindfemur; (F) subapical lobe of the gonocoxite (400x).

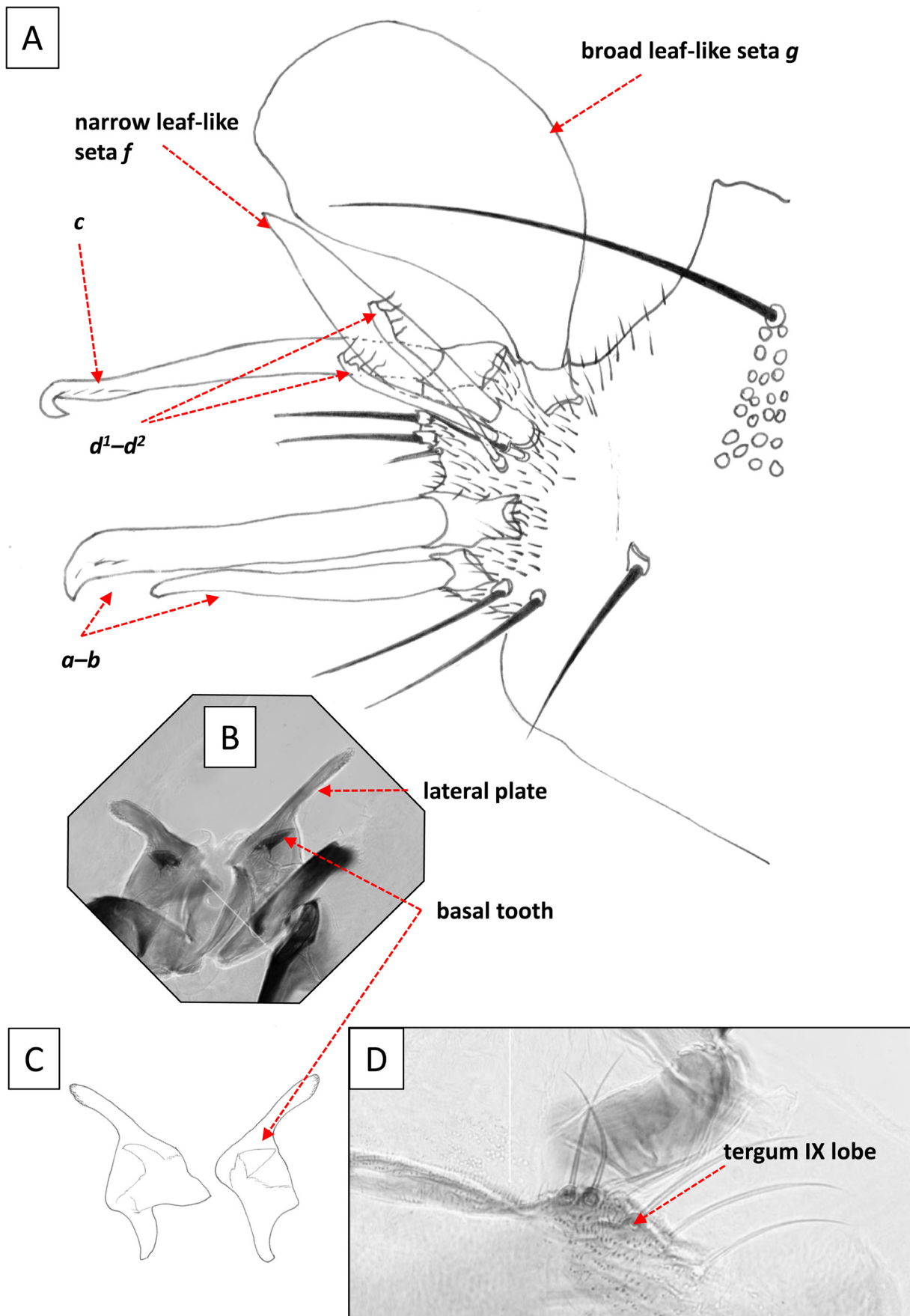


FIGURE 17. Male genitalia of *Culex cinereus*. (A) subapical lobe; (B) phallosome (100x); (C) lateral plates; (D) tergum IX lobe (400x).

downward turned (recurved) apices; no obvious gap between distal and proximal sets, most proximal of proximal set about 0.5 length of other scales of set. Antenna and proboscis similar to those of other *Culiciomyia*.

Thorax: Scutal integument greyish brown (Fig. 16A,D), covered with short brown setae, narrow scales at front and prescutellar area slightly paler, scales on scutellum whitish. Integument of thoracic pleura uniformly greyish brown to beige (Fig. 16D), in general paler than scutal integument; pleura lack scales except for a row of whitish to translucent scales among setae on upper mesokatepisternum; setae on antepronotum, a row of 6 on posterior margin of postpronotum, on proepisternum, prealar knob, on all coxae, a patch on upper mesokatepisternum, a row along posterior margin of lower mesokatepisternum and one lower mesepimeral seta.

Wing: Scales all dark.

Legs: Coloured similar to other *Culiciomyia* except for *Cx. cambournaci*. Hindfemur as shown in Fig. 16E.

Abdomen: Terga greyish brown with slightly paler small basal lateral spots, but lateral pale scales are absent in many specimens examined; sterna uniformly brownish grey.

Genitalia: Tergum IX lobes each with a row of 6–8 setae (Fig. 17D). Ventral surface of gonocoxite with few long setae (slightly longer than gonocoxite) towards outer margin; inner ventral side of gonocoxite with dense ventral lateral patch of 10 rows of setae with about 7 setae in each row (Fig. 16C) that in slide mounts obscure and makes setae on subapical lobe difficult to see; prominent subapical lobe with an additional dense patch of shorter setae at base that also can obscure view of setae on lobe (Fig. 16C); setae on subapical lobe include 1 or 2 setae below rod-like setae (*a–c*), seta *c* more distally positioned than *a* and *c* (Figs 16F, 17A), 2 barbed appendages (*d¹–d²*) with about 4 barbs on each (Fig. 17A), 1 broad foliform seta (*g*), a second much narrower seta (*f*) and 2 short and 2 longer setae between bases of *b* and *c*, seta *b* often darker than 2 rod-like setae (Fig. 16C,F), upper most seta of patch of setae at base of subapical lobe longer, striated and thicker than others and probably equivalent to seta *e* in other *Culiciomyia*, often 3 long setae at apex of gonocoxite below base of gonostylus. Row of 12–14 “spinelets” along crest of gonostylus, gonostylus ends in a small upturned hook, with multiple pre-medially positioned short setae and quite broad dorsal membranous flange. Aedeagus with expanded base with a single dark basal tooth, arms (lateral plates) straight vertically with pointed apices (Fig. 17B,C), apices of lateral plates very slightly roughened (it should be noted that lateral plates often pressed sideways or outwards due to the weight of the coverslip in slide mounts). Paraproct with 3 or 4 subapical setae and no obvious sternal process.

Additional notes: The male of *Cx. cinereus* closely resembles the male of *Cx. nebulosus* except it is generally larger. *Culex cinereus*, as described by Edwards (1941), may or may not have flat white scales extending to more than half the length of the mesepimeron and flat scales on the propleuron (proepimeron and proepisternum), mesopleuron (mesokatepisternum) and has flat scales on the coxae. The palpal comb scales differ from those of *Cx. nebulosus* in lacking an obvious gap between the proximal and distal scale sets. The aedeagus of *Cx. cinereus* differs from that of *Cx. nebulosus* by the absence of obvious teeth on the lateral plates. In *Cx. cinereus*, seta *c* of the subapical lobe is more obviously separated from the other rods than in *Cx. nebulosus*. The subapical lobe has a distinctive patch of delicate setae at its base in *Cx. cinereus* whereas this patch is absent in *Cx. nebulosus*. Stout seta *e* is more dorsolaterally positioned than in *Cx. nebulosus*.

Specimens examined: Several males were collected in the Talangaye Forest in Cameroon. Specimen CAM 85u2a 7/XI/2016 was reared from an egg raft that was laid in a bamboo pot. The following males were collected by sweep netting through vegetation on the forest floor. Field accession numbers CAM 86j1 4/XI/2016, CAM 86v 7/XI/2016, CAM 132ha 28/I/2017, CAM 132hb 28/I/2017, CAM 132b1a 31/I/2017, CAM 132b1d 31/I/2017, CAM 132a1c 1/II/2017, CAM 132anb 1/II/2017, CAM 132gg 7/II/2017, CAM 163kc 10/V/2017, CAM 202ta 16/VII/2017, CAM 202tb 16/VII/2017, CAM 202nb 21/VII/2017 and CAM 227q 2/XI/2017.

Culex (Culiciomyia) eouzani Geoffroy, 1971

Male. After the genitalia were dissected, the specimen was discarded as it was completely covered with fungal growth. Hence, unfortunately, we cannot comment on features other than those of the genitalia. All features of the gonocoxite appear very similar to those figured by Geoffroy (1971).

Genitalia: Each tergum IX lobe with a row of 7 setae. Ventral surface of gonocoxite with few long setae, each as long as gonocoxite in no obvious configuration, a few irregular rows of 3 or 4 narrow setae in each row along inner margin (Fig. 18A), dorsal surface with quite sparsely distributed short setae, no obvious subapical lobe, apex

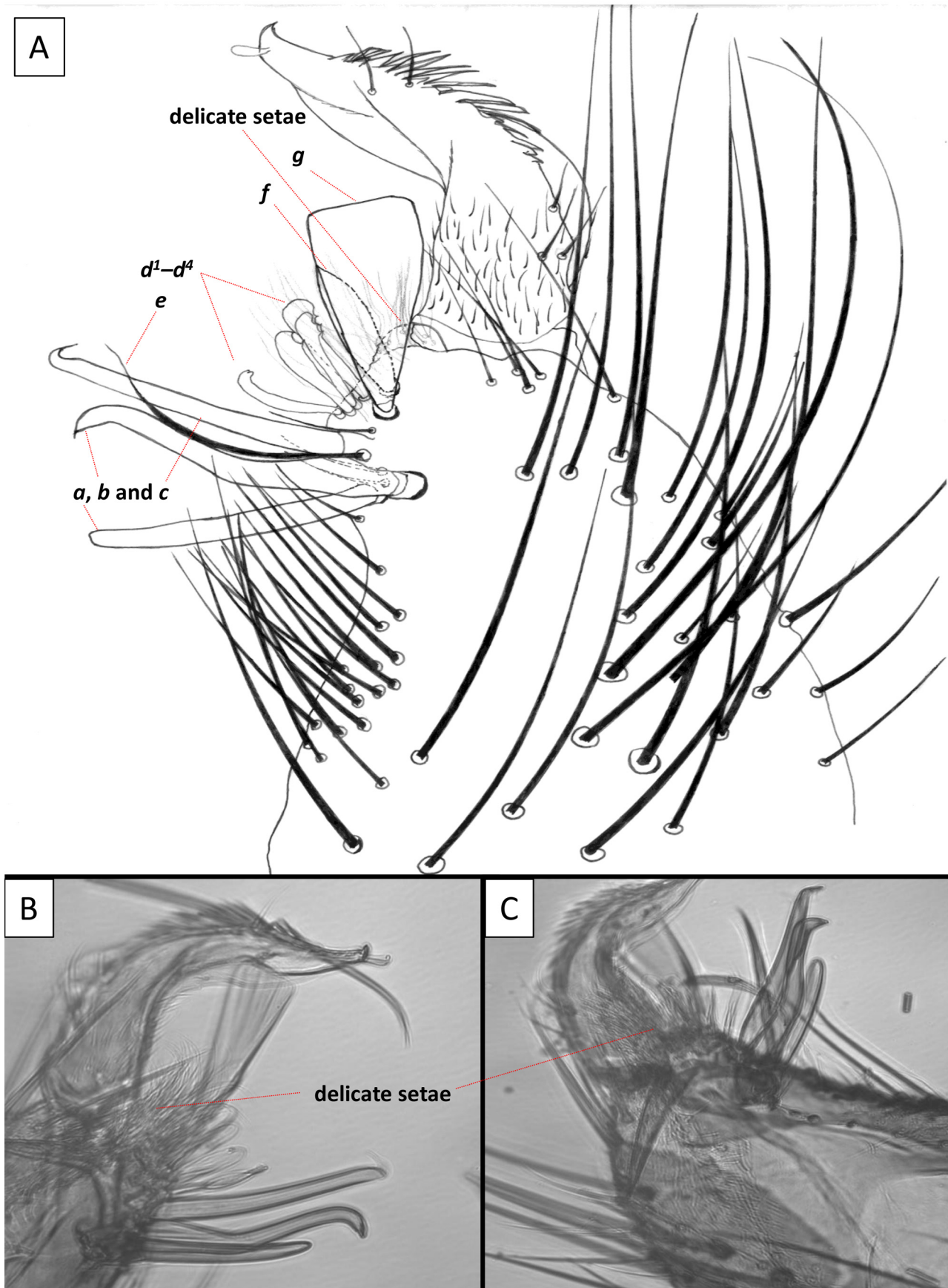


FIGURE 18. Gonocoxite of *Culex eouzani*. (A) drawn depiction; (B,C) images of the apex of the gonocoxite (400x).

of gonocoxite with group of about 12 delicate setae on a raised lobe or protuberance dorsally at apex below base of gonostylus (drawn as faint setae in Fig. 18A and labelled as delicate setae in Fig. 18A,C), on ventral surface 4 setae at apex below base of gonostylus, 3 setae (*a–c*) present, most distal longest and most posteriorly positioned shortest, single quite robust and slightly striated seta (*e*) attached close to middle of setae *a–c* and slightly longer than seta *c*, a further 3 shorter setae (0.5 length of *c*) attached close to base of *c*, 4 single and double barbed narrower setae (*d¹–d⁴*), rather angular foliiform seta that broadens gradually towards apex (*g*) and a narrower more rounded foliiform seta (*h*) shorter than *g*. Gonostylus with crest of many “spinelets” along dorsal margin that extend from mid-length close to apex (Fig. 18A,B), 4 setae close to where gonostylus bends, a single seta on dorsal margin in middle of crested region and a subapical seta, gonostylus ends in an upturned hook with a small folded foliiform seta, small membranous flange, basal 0.50 of gonostylus quite setose. Aedeagus with expanded triangular base (Fig. 19A,B) with a basal tooth, row of quite broad teeth or denticles along inner margin that extend almost to apex of lateral plate, apices of lateral plate pointed and with pitted membranous appearance apically, row of small denticles next to inner edges of larger teeth. Paraproct with 3 subapical setae and no basal protrusion.

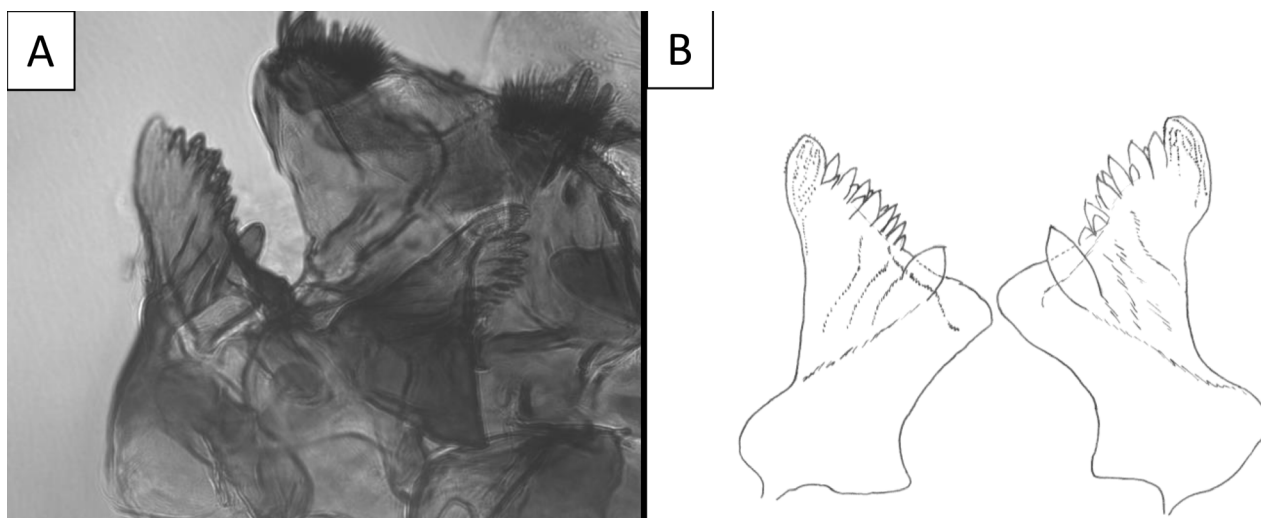


FIGURE 19. Lateral plate-aedeagal sclerite of *Culex eouzani*. (A) image (400x); (B) drawn depiction.

Additional notes: Further notes on this species are mainly based on the description by Geoffroy (1971), which is based on a single male collected in the village of Batalimo, Central African Republic, on the 4 May 1970 (4.06667° N; 17.66667° E). The collection of a second male in the Talangaye Forest by us and a few other males in Nkolbisson, Cameroon (Rickenbach *et al.* 1976) confirms that this species is valid (although likely uncommon) and occurs in the deep pristine forests in Central Africa.

The scaling on the head and scutum and the morphology of the antenna and proboscis are like those of *Cx. nebulosus*. The maxillary palpus bears evenly spaced palpal comb scales that are all similarly shaped and palpomere 4 is half the length of palpomere 5. The thoracic pleura are light brown and there are three long setae on each scutellar lobe. Three setae and some brown scales are borne on the upper region of the mesokatepisternum, three setae on the upper region of the mesanepimeron and one lower mesepimeral seta is present. The abdomen is much like the abdomen of *Cx. nebulosus*, with dark brown tergal and slightly lighter brown sternal scaling.

In general, *Cx. eouzani* resembles *Cx. grenieri* most closely, both having the aedeagus of similar shape that also has tubercles not quite reaching the apex. *Culex eouzani* differs from *Cx. grenieri* in the shape of the broad and narrower setae on the subapical lobe. In *Cx. eouzani*, the narrower seta is rounded apically and there are four barbed setae and a finger-like protuberance with many delicate setae on the apex of the gonocoxite at the base of the gonostylus. In the male from Talangaye, the secondary narrower seta is not obviously rounded apically but rather more sharply pointed. However, the square or angular broad foliiform seta is as described by Geoffroy (1971). The subapical lobe is not obviously protruding in the male from Cameroon and appears similar to that described by Geoffroy (1971).

Specimen examined: One male collected in a sweep net from vegetation on the forest floor in the Talangaye Forest in Cameroon. Field accession number CAM 132 7/II/2017.

Culex (Culiciomyia) macfieii Edwards, 1923

Male: This is a small brown to greyish mosquito. The scutal integument is generally brown. Most males collected in the Talangaye Forest have no basal pale markings on the abdominal terga and were identified as *Cx. macfieii* only once the genitalia were dissected and carefully examined. However, as Edwards (1941) mentioned, all *Cx. macfieii* males collected in the Talangaye Forest have the pale decumbent scales along the eye margin much reduced relative to those found in other Afrotropical *Culiciomyia*. Wing 2.30–2.60 mm. Proboscis 1.55–1.77 mm. Forefemur 1.35–1.54 mm.

Head: Row of broad white decumbent scales along eye margin not visible in most specimens, and in those specimens with these scales, the ocular line of scales did not meet in the frontal region of head (Fig. 20A,C), small patch of broad decumbent pale scales on sides of head (Fig. 20A), narrow decumbent pale scales numerous on vertex (Fig. 20A,C), erect scales numerous and uniformly light to dark brown. Maxillary palpus slightly longer than proboscis with palpomeres 4 and 5 bearing setae, palpomere 4 approximately 0.50 (ratio = 0.48–0.54) length of palpomere 5, palpal comb on palpomere 3 as described by Edwards (1941), consisting of 10 or 11 long semi-transparent scales, each ending in a long straight point (acicular or needle-like), similar in shape to those in *Cx. cinerellus* and *Cx. subaequalis* (Fig. 20B). Antenna and proboscis similar to those of other *Culiciomyia*.

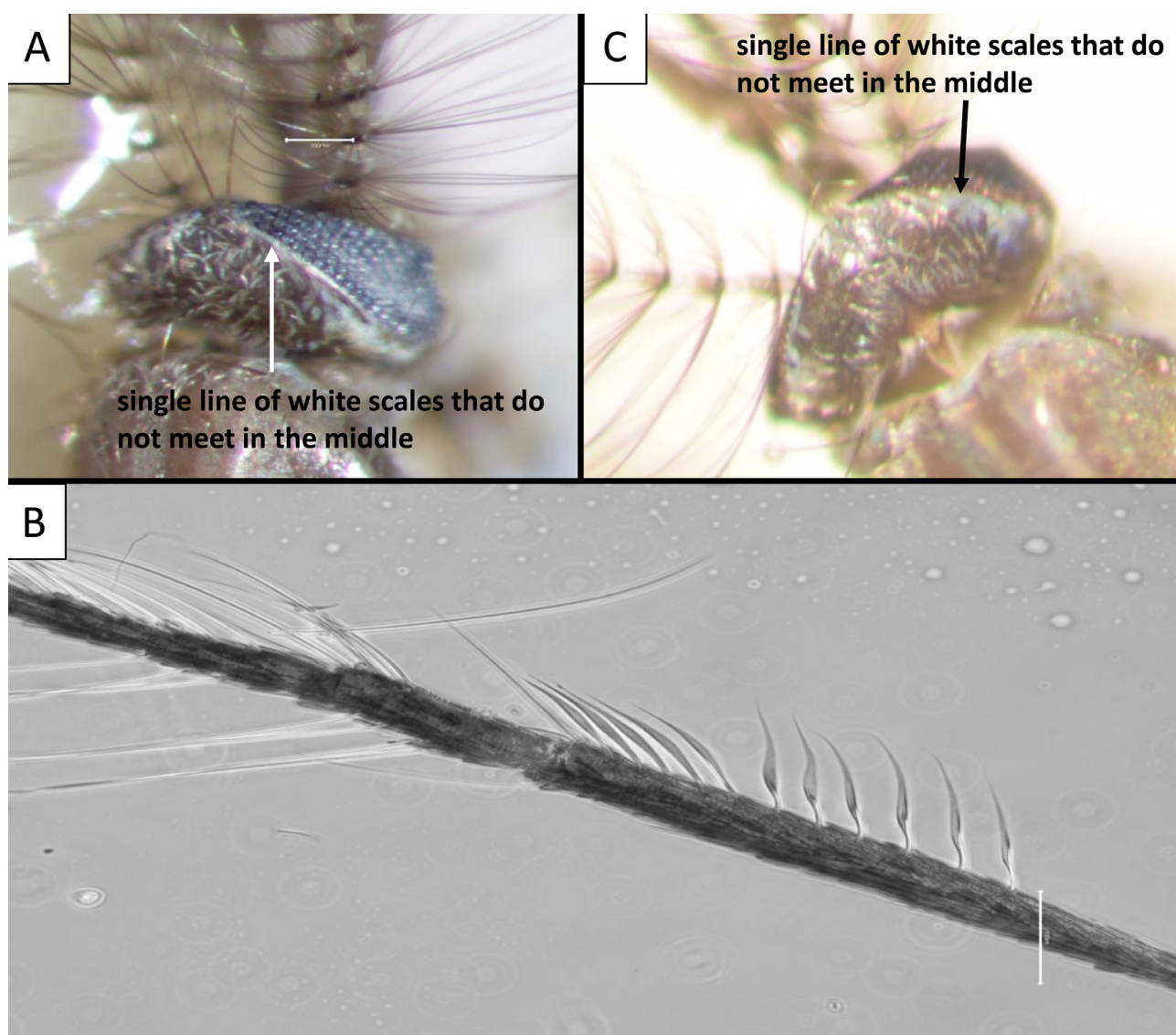


FIGURE 20. External features of *Culex macfieii*. (A,C) dorsal views of the head (vertex); (B) lateral view of the palpal comb scales (100x).

Thorax: Scutal integument light brown with setae generally appearing light brown to translucent depending on angle of light, setae on anterior promontory and prescutellar area paler, acrostichal setae absent. Thoracic pleura generally not uniform in colour, with prealar area darker brown than upper and lower mesokatepisternum, upper 0.33 of mesanepimeron darker than paler beige to light brown of lower 0.66 of mesokatepisternum (Fig. 21A), in some specimens middle 0.25 of mesokatepisternum darker brown than rest of area (Fig. 21B,C); pleura devoid of scales except for small patch among setae on upper mesokatepisternum, setae present on following areas: antepronotum, a row of 6 on posterior margin of postpronotum, 3 on proepisternum, prealar knob, a few in middle and posterior margin of mesokatepisternum, and a single lower mesepimeral seta. Capitellum of halter darker than pedicel.

Wing: Scales all dark.

Legs: Coloured similar to those of other *Culiciomyia*, except for *Cx. cambournaci*.

Abdomen: Terga light brown with, in some specimens, paler small basolateral spots; sterna light brown to beige.

Genitalia: Each tergum IX lobe with row of 4–6 (mostly 5) setae along upper margin (Fig. 23C), typically fewer than in other *Culiciomyia* species. Genital structures of *Cx. macfieii* males collected in the Talangaye Forest closely match those described for this species by Edwards (1941). Gonocoxite moderately broad, without distinct ventral lateral setal patch, setae numerous, nearly evenly spaced rows of setae each about 0.67 length of gonocoxite dispersed on ventral and lateral surfaces of gonocoxite (Fig. 22A); single prominent subapical lobe with following setae (Fig. 22A,B): 3 rod-like setae as in other *Culiciomyia*, except proximal seta *a* considerably more dorsally displaced from setae *b* and *c*, seta *a* much darker, striated, broader than *b* and *c*, 6 or 7 flattened setae more ventrally positioned and in line with *b* and *c*, 1 narrow foliform seta (*g*) and what is usually a second narrower foliform seta in other *Culiciomyia* species is replaced by a stout blunt seta (*f*), seta *e* absent. Gonostylus bent in middle with single dorsal subapical hook or recurved horn (Fig. 22A,B), single subapical seta, a seta below recurved horn (below the crest in other species) and 1 to 3 (mostly 2) setae pre-dorsally positioned in proximity to bend of gonostylus. Aedeagus triangularly broadened basally with distinct tooth at base of each lateral plate (Fig. 22A, 23A,B), lateral plate with sharp apex, resembling that of *Cx. nebulosus* with rounded or sharp apex depending on angle of view, lateral plate with few small rounded denticles on outer edge in dorsal view (Fig. 23B), denticles sharper and more numerous than in *Cx. nebulosus*. Paraproct with typical dense crown of spicules and 3 or 4 small subapical setae, no obvious basal arm.

Additional notes: The male is similar in size to the male of *Cx. nebulosus* and smaller than the male of *Cx. cinereus*. The integument of the thoracic pleura of *Cx. macfieii* is quite variable in colour and we caution using colouration of the scutal and pleural integument to distinguish *Culiciomyia*, except perhaps *Cx. cinerellus*, which has two obviously darker stripes on the pleura. *Culex macfieii* has fewer less obvious pale flat scales along the eye margins than other *Culiciomyia*. The aedeagus and paraprocts appear very similar to those of *Cx. nebulosus*. The single hook or recurved horn in place of a crest on the gonostylus is the most distinguishable feature that separates *Cx. macfieii* from all other *Culiciomyia*. The most proximal well-displaced broad rod-like seta (*a*) on the subapical lobe is also an obvious feature that distinguishes the genitalia from the genitalia of other *Culiciomyia*. The patch of broader stouter setae on the subapical lobe is also quite unique among the Afrotropical *Culiciomyia*. None of the males collected in the Talangaye Forest have pale basal lateral spots. Some female *Culiciomyia* collected in the forest have small basal pale spots on the abdominal terga that are often quite subtle to see (Fig. 24). These females could be *Cx. macfieii*. Edwards (1941) suggested that the basal pale spots can be diagnostic for males and the basal spots may be useful to identify females of this species. However, we caution this as the pale spots in some specimens are very difficult to see and may be absent in some.

Specimens examined: Multiple males were collected in the Talangaye Forest in Cameroon. Specimens CAM 133c1b 24/I/2017 and CAM 133c1c 24/I/2017 were reared from an egg raft laid in a bamboo pot. The following specimens were collected in bird-baited net traps or by sweep netting through vegetation on the forest floor: field accession numbers CAM 86q 13/X/2016, CAM 86a1 26/X/2016, CAM 86g 1/XI/2016, CAM 86f2 2/XI/2016, CAM 86u2b 7/XI/2016, CAM 86o 8/XI/2016, CAM 86x2 8/XI/2016, CAM 86ya 8/XI/2016, CAM 86yb 8/XI/2016, CAM 86yc 8/XI/2016, CAM 86a 19/XI/2016, CAM 132i 25/I/2017, CAM 132r 28/I/2017, CAM 132b1f 31/I/2017, CAM 132b1g 31/I/2017, CAM 132b1h 31/I/2017, CAM 132b1i 31/I/2017, CAM 132b1j 31/I/2017, CAM 132l1a 31/I/2017, CAM 132l1b 31/I/2017, CAM 132yc 31/I/2017, CAM 132yg 31/I/2017, CAM 132yj 31/I/2017, CAM 132a1a 1/II/2017, CAM 132l1c 2/II/2017, CAM 132nb 4/II/2017, CAM 132kc 5/II/2017, CAM 132ob 6/II/2017, CAM 132gd 7/II/2017, CAM 132ge 7/II/2017, CAM 132gj 7/II/2017, CAM 163c1a 1/V/2017, CAM 163f 2/V/2017, CAM 163sa 2/V/2017, CAM 163ea 5/V/2017, CAM 163eb 5/V/2017, CAM 163fa 5/V/2017, CAM 163fc

5/V/2017, CAM 163lc 10/V/2017, CAM 202mb 9/VII/2017, CAM 202nd 9/VII/2017, CAM 202nh 9/VII/2017, CAM 202sa 10/VII/2017, CAM 202sc 10/VII/2017, CAM 202u 11/VII/2017, CAM 202qa 13/VII/2017, CAM 202qc 13/VII/2017, CAM 202rb 13/VII/2017, CAM 202ha 16/VII/2017, CAM 202hd 16/VII/2017, CAM 202ta 19/VII/2017 and CAM 202tb 19/VII/2017.

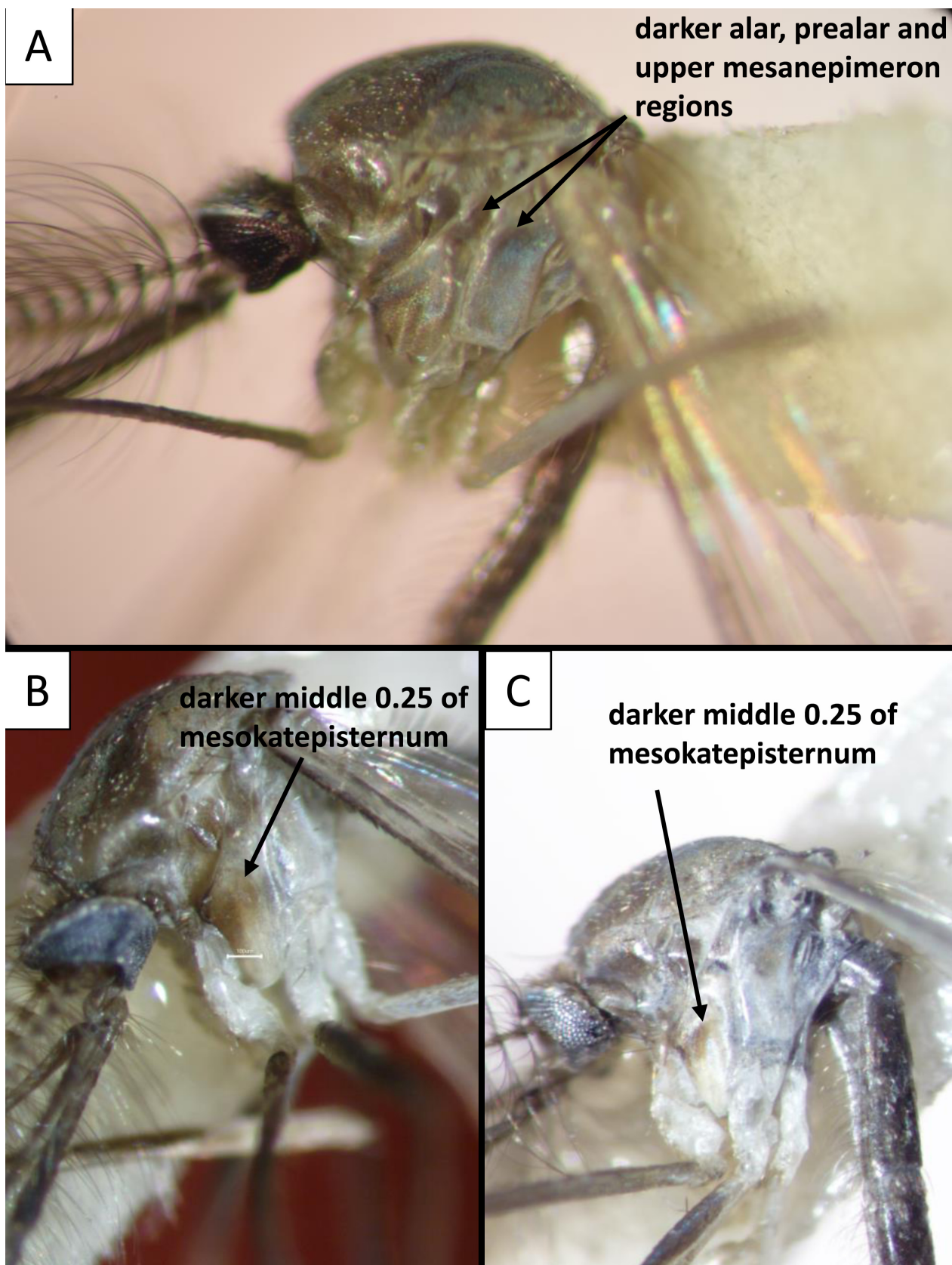


FIGURE 21. Lateral views of the thorax of *Culex macfieii*.

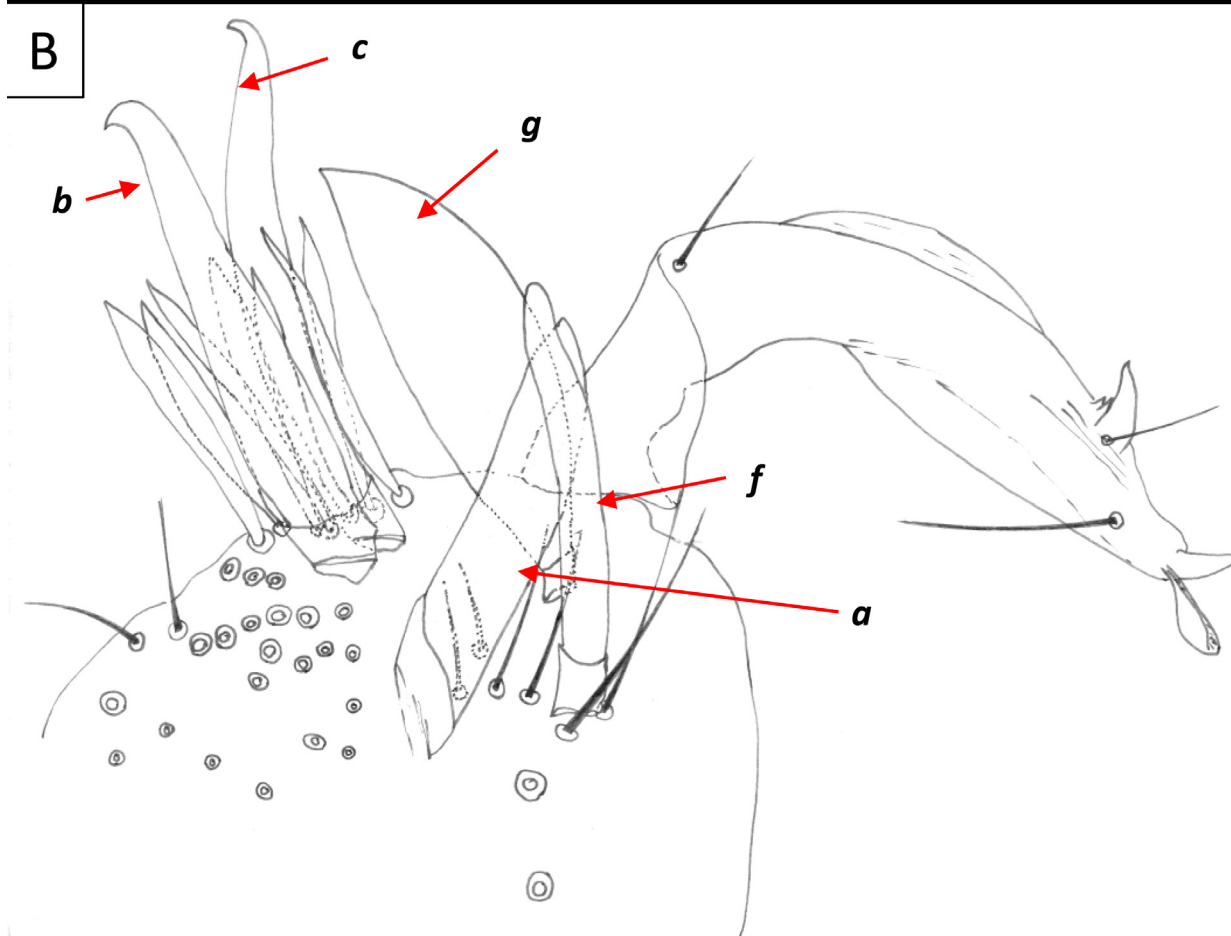
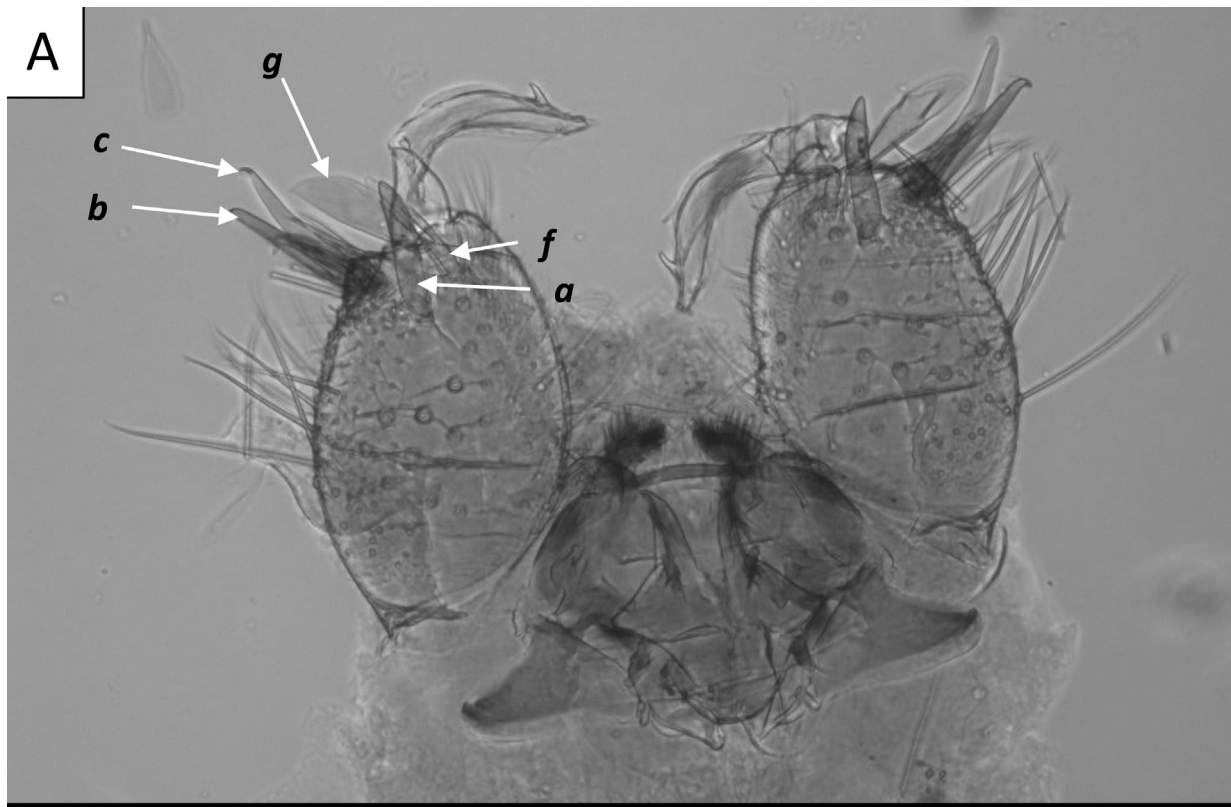


FIGURE 22. Male genitalia of *Culex macfieii*. (A) Image (100x); (B) drawn depiction of the subapical region of the gonocoxite and the gonostylus.

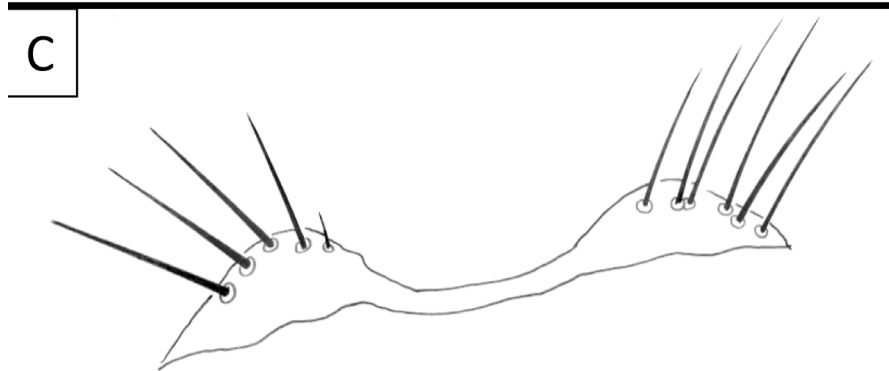
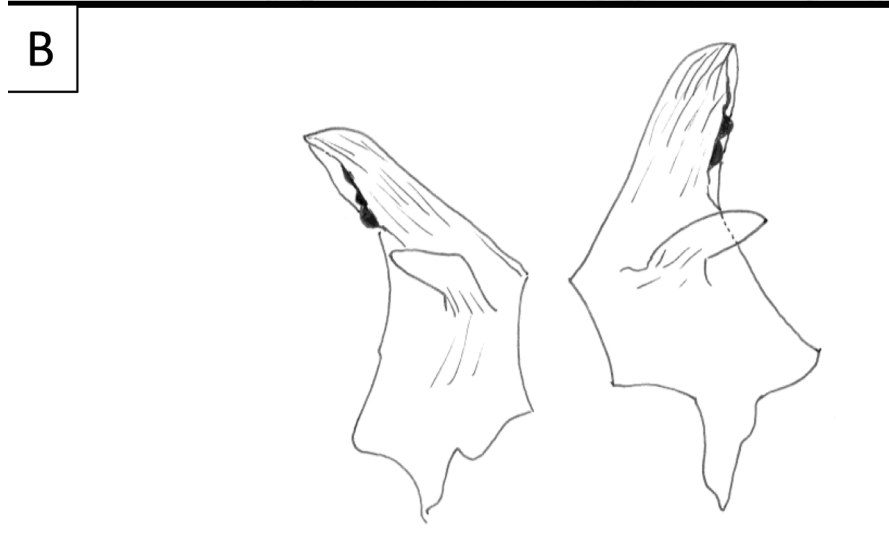


FIGURE 23. Male genitalia of *Culex macfieii*. (A) Image of the phallosome (400x); (B) drawn depiction of the fused lateral plate-aedeagal sclerite of the left and right sides; (C) drawn depiction of tergum IX.

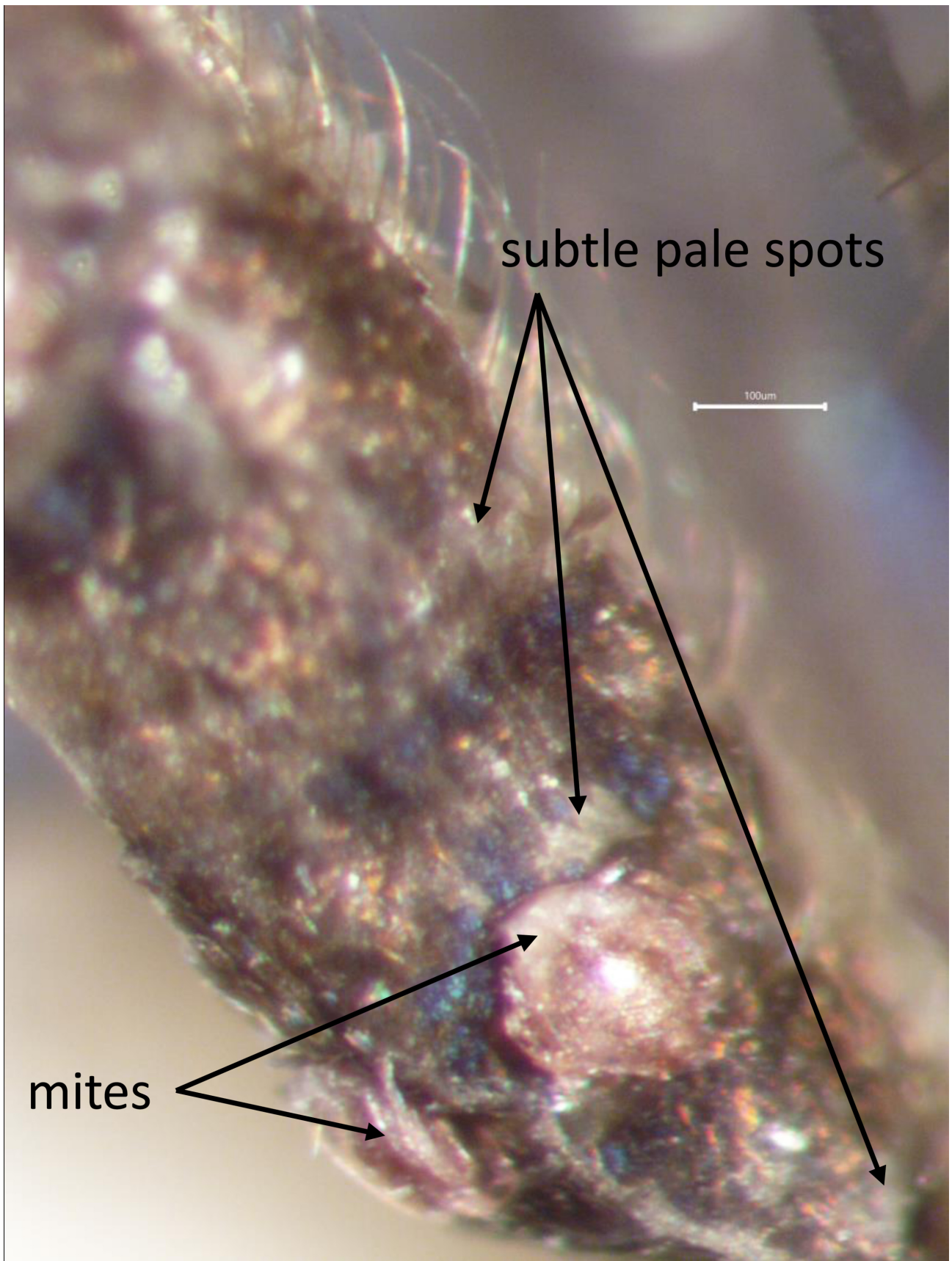


FIGURE 24. Dorsal view of abdominal terga III–V of *Culex macfieii*.

Culex (Culiciomyia) nebulosus Theobald, 1901

Male. A small brown to greyish mosquito with no obvious distinguishing features. Integument of thoracic pleura generally not uniform in colour, with mesanepimeron, metepisternum and coxae whitish and antepronotum, proepisternum, postspiracular and subspiracular areas, mesokatepisternum and mesomeron light to dark brown. Scutal integument light brown. Wing 2.70 mm. Proboscis 1.87 mm. Forefemur 1.60 mm.

Head: Row of broad white decumbent scales along eye margin (Fig. 25A,B,E), pale narrow decumbent scales numerous on vertex (Fig. 25A,B,E), erect scales numerous, pale brown except for white to very light brown apices, broad white scales along eye margin tend to be more numerous towards sides of head and form a more distinct lateral patch (Fig. 25A,E). Maxillary palpus slightly longer than proboscis, palpomeres 4 and 5 bearing setae, palpomere 4 shorter than palpomere 5 (ratio 0.45–0.33), palpal comb scales described by Edwards (1941) as “the proximal part of this comb formed of scales shaped like spear-heads, the distal part of more close-set scales which are also broader and have fine recurved points” (Fig. 25C), 3 or 4 most basal of proximal set of 7 or 8 (rarely 9) with longer points, significant gap between proximal and distal sets, distal set with 9–10 scales more closely spaced, proximal scales often darker than distal set of scales (Fig. 25C); we prefer to describe distal scales as spatulate or oblanceolate with recurved fine points (aristate apices), proximal 3 or 4 scales of proximal set as lanceolate and other distal scales as narrowly ovate with aristate apices and cuneate bases. Antenna and proboscis similar to those of other *Culiciomyia*.

Thorax: Variation in colour of thoracic integument may be dependent on age of mosquito or environmental conditions, such as larval diet, habitat or perhaps degree of exposure to UV light. Scutal and pleural integument light brown (in most specimens) to greyish (in some) creates some confusion about general colouring of this species (Fig. 25A,D). Scutum covered with short brown to shiny translucent setae, narrow white scales on scutellar lobes. Colour of thoracic pleural integument highly variable and mostly non-uniform. Mesanepimeron, metepisternum and coxae whitish, integument of antepronotum, proepisternum, postspiracular and subspiracular areas, mesokatepisternum and mesomeron light to dark brown; middle region of metepisternum light brown in some specimens, which gives pleura a more uniform brown appearance, but in others pleura appear more greyish, thus more closely fitting Edwards (1941 “greyish mesonotum”); pleura devoid of scales except for small patch among setae on upper mesokatepisternum and occasionally a row along posterior margin of lower mesokatepisternum, scales absent on mesepimeron in majority of specimens but a few have 1 or 2 below mesepimeral seta; setae present in following regions: antepronotum, a row of 6 on posterior margin of postpronotum, 3 on proepisternum, prealar knob, all coxae, a few on upper and lower posterior margin of mesokatepisternum, and 1 in lower mesepimeral area. Capitellum of halter darker than pedicel.

Wings: Scales all dark.

Legs: Coloured similar to those of other *Culiciomyia*, except *Cx. cambournaci*.

Abdomen: Terga uniformly brown with paler small lateral spots in a few specimens; sterna light brown to beige.

Genitalia: Lobes of tergum IX each with 5–9 (mostly 6, rarely 9) setae along upper margin (Fig. 27A). Description below includes details that were not mentioned in Edwards (1941). Gonocoxites moderately broad with distinct patch of gold ventrolateral setae, mostly confined to middle 0.33 of gonocoxite, patch of 3 to 4 setae wide with each seta about 0.66 length of gonocoxite (Fig. 26A,B); dorsal surface of gonocoxite with few setae, each about same length as gonocoxite, distinct apical dorsal lobe bearing 4–7 setae (mostly 4), 3 rod-like setae (*a–c*) with 1 placed slightly more ventrally than other 2 and rod-like setae not necessarily as close together as described by Edwards (1941), with tip of seta *a* slightly shorter and sharper apically than *b* and *c*, *b* and *c* slightly hooked apically and *b* noticeably darker than *a* and *c* (Fig. 26B), a consistently short seta present below or next to *a*; 1 very broad slightly striated foliform seta (*g*) and shorter narrower foliform seta (*f*) half as broad as *g*; a stout striated seta (*e*) between bases of *f* and *g* slightly longer than *f* and *g* and 4 setae of which 2 (*d'*–*d''*) longer and slightly broader than other 2. Gonostylus bent in middle with a large membranous flange dorsally, a single subapical seta and 1–5 (mostly 3 or 4) setae pre-dorsally positioned in proximity where gonostylus bends downwards (Fig. 26A), tip of gonostylus bent upwards into a hook and with a small broad foliform seta. Aedeagus broadened basally with distinct tooth at base of each lateral plate (Fig. 27A,B); from dorsal aspect lateral plate smooth and rounded apically, resembling what Edwards (1941) described as being “finger like processes”, however, from ventral and lateral aspects lateral plate clearly toothed along one margin and apex ends sharply, resembling the description provided for *Cx. nebulosus* by Hamon & Lambrecht (1959), who described the lateral plate as sharp and having strong lateral teeth. Paraproct with

typical dense crown of spicules and 3 or 4 small subapical setae (Fig. 27A).

Additional notes: None of the *Cx. nebulosus* males from the Talangaye Forest have white scaling on the propleuron (proepisternum) and there was always only one lower mesepimeral (mesanepimeral) seta with no accompanying short setae, which precludes them from identification as *Cx. pseudocinereus*, which according to Edwards (1941) occurs more in eastern and southern Africa. Males of *Cx. nebulosus*, *Cx. furlongi*, *Cx. cinereus*, *Cx. lanzaroi* and *Cx. muspratti* have similar external features. The genitalia of *Cx. cinereus* are most easily distinguished from the genitalia of the other three species by the dense patch of delicate setae at the base of the subapical lobe, and *Cx. cinereus* also has no gap between the proximal and distal sets of palpal comb scales. Hamon & Lambrecht (1959) noted that the apices of the lateral plates of *Cx. nebulosus* are sharp whereas they are rounded in *Cx. muspratti*. In our opinion, the apices of the lateral plates of *Cx. nebulosus* appear rounded depending on the angle of the slide mount, we caution relying on using this feature to distinguish these two species. *Culex nebulosus* and *Cx. muspratti* have a strong basal tooth at the base of the lateral plate, whereas this is absent in *Cx. furlongi*. The only character that likely separates *Cx. nebulosus* from *Cx. muspratti* is the number of narrower accessory appendages on the subapical lobe of the gonocoxite, with three in *Cx. muspratti* and two in *Cx. nebulosus*. Van Someren (1954) reported the presence of three short “narrow leaflets (*d–f*), which sometimes appear as “stout setae”, but did not mention or figure them as having barbs. The most distal palpal comb scales in *Cx. furlongi* end in fine short straight points whereas in *Cx. nebulosus*, *Cx. lanzaroi* and *Cx. muspratti* they are longer, strongly recurved and shaped much like the top bill of a hawk’s beak. Flat scales on the upper mesokatepisternum and the scales on the mesepimeron are easily rubbed off, and when present, these scales are often translucent and can be easily overlooked. The larvae of *Cx. muspratti* and *Cx. lanzaroi* are unknown. The larva of *Cx. nebulosus* is quite different from the larvae of *Cx. furlongi* and *Cx. cinereus* (Hopkins 1952; van Someren 1954). The only noticeable difference in the larvae of the latter two species is that the comb is composed of scales in *Cx. cinereus* and spine-like scales in *Cx. furlongi* (van Someren 1954).

Specimens examined: The following males were reared from egg rafts laid in bamboo pots. Field accession numbers CAM 47e1a 2/VII/2017, CAM 47e1b 2/VII/2016, CAM 47e1c 2/VII/2016, CAM 60c.2 16/VII/2016, CAM 113ca 7/V/2017, CAM 113cb 7/V/2017, CAM 230c1a 10/XI/2017, CAM 230c1b 10/XI/2017 and CAM 230c1c 10/XI/2017. One male examined was reared from a pupa collected in a rain-filled coconut husk lying on the ground. Field accession number CAM 196f3 16/VII/2017. The other males examined from the Talangaye Forest were collected by sweep netting through vegetation on the forest floor. Field accession numbers CAM 50b.1 16/VII/2016, CAM 50c.2 16/VII/2016, CAM 62h 18/VII/2016, CAM 62g 28/VII/2016, CAM 62d 29/VII/2016, CAM 86ra 9/XI/2016, CAM 132ya 31/I/2017, CAM 132yb 31/I/2017, CAM 132yd 31/I/2017, CAM 132ye 31/I/2017, CAM 132yh 31/I/2017, CAM 132yi 31/I/2017, CAM 132yk 31/I/2017, CAM 132o1d 1/II/2017, CAM 132b1a 2/II/2017, CAM 132b1b 2/II/2017, CAM 132ka 5/II/2017, CAM 132kd 5/II/2017, CAM 132ke 5/II/2017, CAM 132kg 5/II/2017, CAM 132kh 5/II/2017, CAM 132kj 5/II/2017, CAM 132kk 5/II/2017, CAM 132oa 6/II/2017, CAM 132oc 6/II/2017, CAM 132oe 6/II/2017, CAM 132of 6/II/2017, CAM 132og 6/II/2017, CAM 132ga 7/II/2017, CAM 132gc 7/II/2017, CAM 132gf 7/II/2017, CAM 132gi 7/II/2017, CAM 132gk 7/II/2017, CAM 163f 27/IV/2017, CAM 163ca 28/IV/2017, CAM 163cb 28/IV/2017, CAM 163ia 1/V/2017, CAM 163ib 1/V/2017, CAM 163ga 2/V/2017, CAM 163gb 2/V/2017, CAM 163sb 2/V/2017, CAM 163sc 2/V/2017, CAM 163fb 5/V/2017, CAM 163la 10/V/2017, CAM 163lb 10/V/2017, CAM 163la 26/V/2017, CAM 202j 5/VII/2017, CAM 202fb 6/VII/2017, CAM 202la 8/VII/2017, CAM 202lb 8/VII/2017, CAM 202na 9/VII/2017, CAM 202nb 9/VII/2017, CAM 202nc 9/VII/2017, CAM 202ne 9/VII/2017, CAM 202nf 9/VII/2017, CAM 202ng 9/VII/2017, CAM 202ni 9/VII/2017, CAM 202sb 10/VII/2017, CAM 202rc 12/VII/2017, CAM 202rd 12/VII/2017, CAM 202re 12/VII/2017, CAM 202rf 12/VII/2017, CAM 202qb 13/VII/2017, CAM 202qd 13/VII/2017, CAM 202jb 14/VII/2017, CAM 202jc 14/VII/2017, CAM 202jd 14/VII/2017, CAM 202je 14/VII/2017, CAM 202hb 16/VII/2017, CAM 202c 17/VII/2017, CAM 202tc 19/VII/2017, CAM 202o 20/VII/2017 and CAM 202na 21/VII/2017.

For comparative purposes, we also examined males in the collection of the National Health Laboratory Services Museum in Johannesburg, South Africa. These were from: (i) Gwaliweni Forest, Kwazulu Natal, South Africa. *Cx. nebulosus*—1 male. Collected and identified by Jim Muspratt, 1957; (ii) Port Shepstone, Kwazulu Natal, South Africa. *Cx. nebulosus*—1 male. Field accession number T154. No other collection information on the label; (iii) Livingstone, Zambia. *Cx. nebulosus* var. *pseudocinereus*—1 male. Collected in 1945. No other collection information on the label; (iv) Tete District, Kwazulu Natal, South Africa. *Cx. nebulosus* var. *pseudocinereus*—1 male. Collected and identified by Jim Muspratt, 1955; (v) Skukuza, South Africa. *Cx. nebulosus* var. *pseudocinereus*—1 male. Collected and identified by Leo Braack, 22/III/1991. Man-biting.

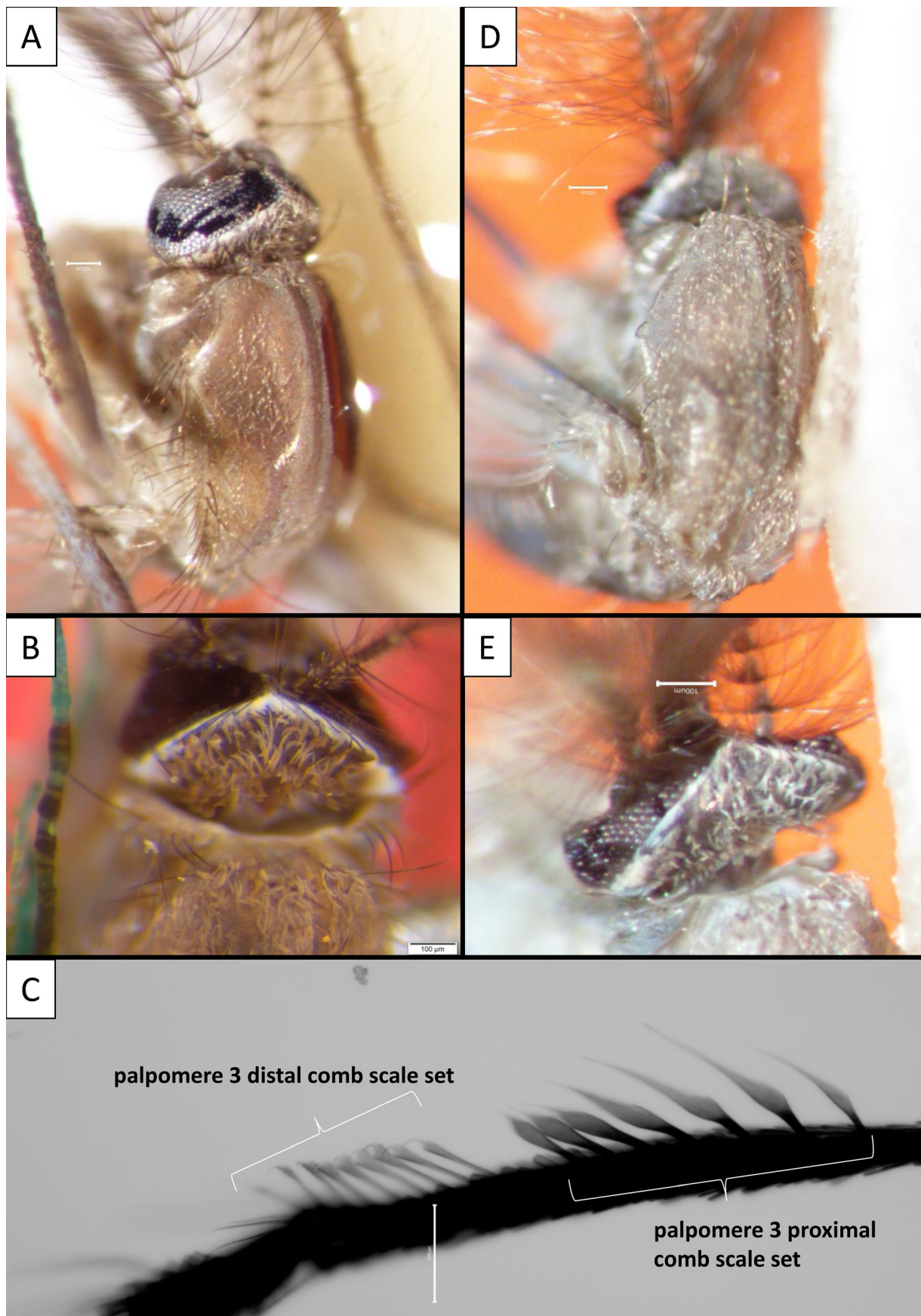


FIGURE 25. External features of *Culex nebulosus*. (A, D) Dorsolateral views of the head and scutum; (B,E) dorsal views of the head; (C) lateral view of the palpal comb scales (100x).

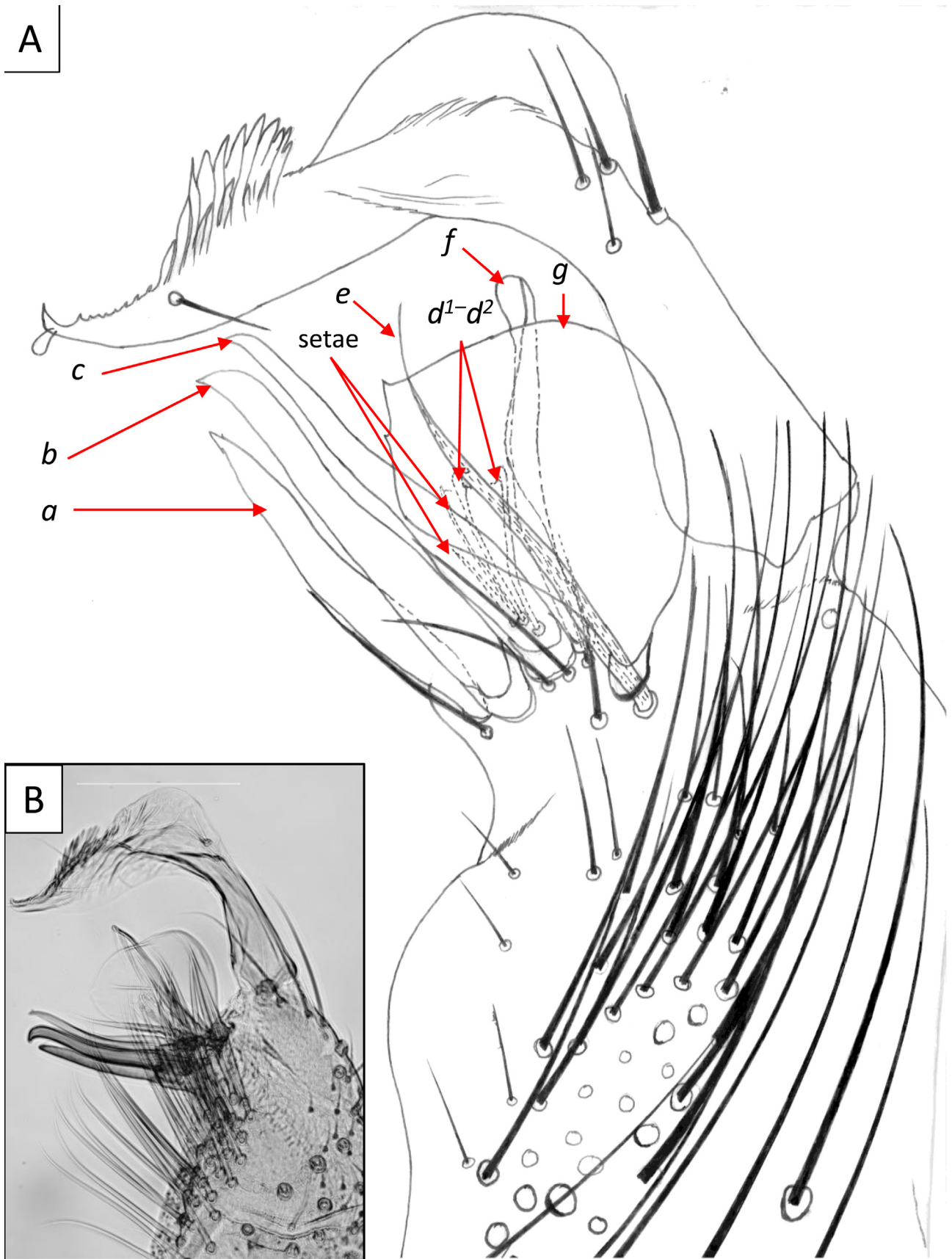


FIGURE 26. Gonocoxite of *Culex nebulosus*. (A) Drawn depiction; (B) image (100x).

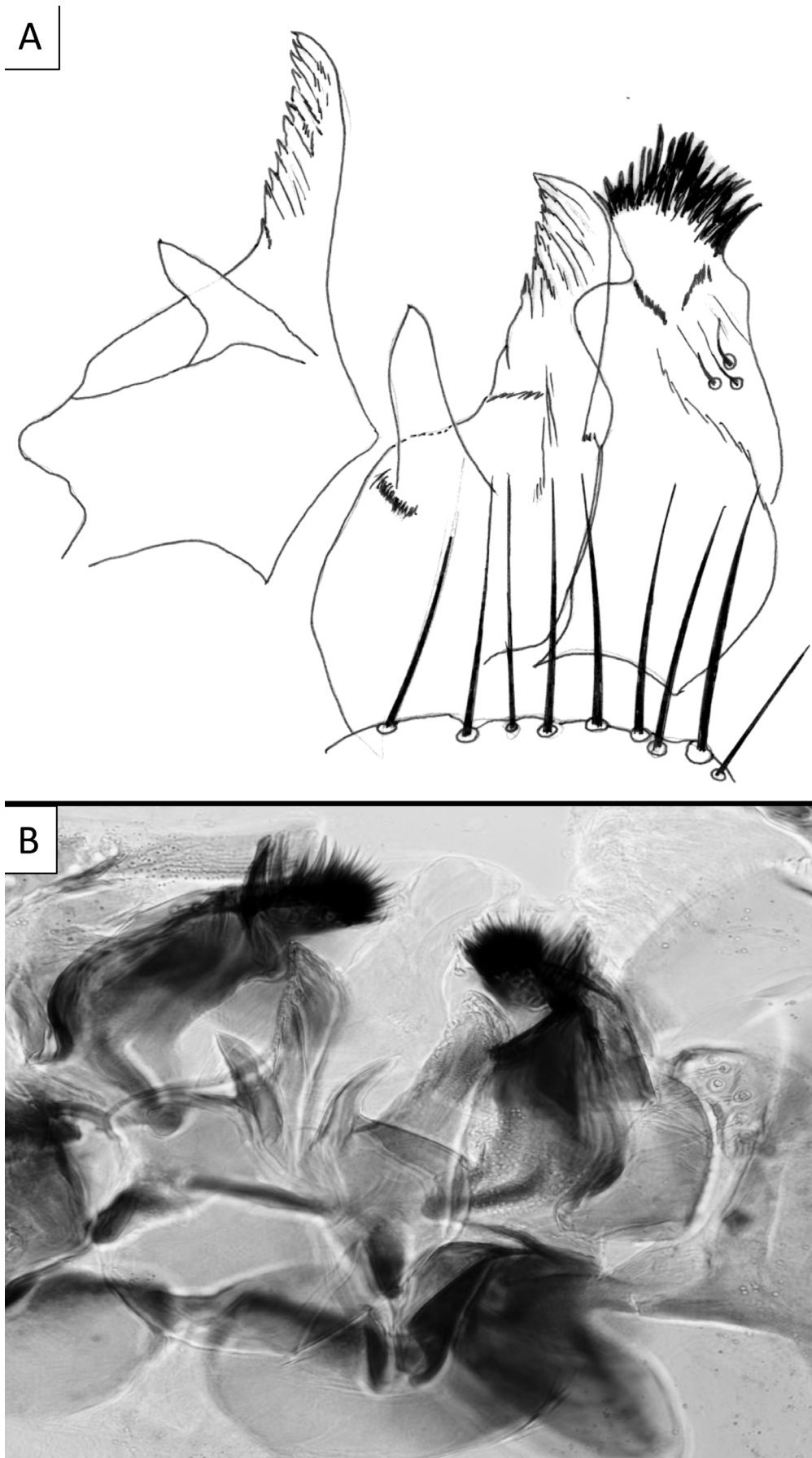


FIGURE 27. Lateral plate-aedeagal sclerite and paraproct of *Culex nebulosus*. (A) Drawn depiction; (B) image (400x).

Other specimens previously collected by one of us (AJC) in Cameroon were also examined. These were from: (i) Ikonge, Cameroon (4.12000° N; 9.44944° E). *Cx. nebulosus*—2 males. Collected and identified by AJC, 2011. Reared from a larva found in a collecting cup on the side of a rubber tree; field accession number CAM 12.32.1 6/IX/2011. Reared from larva in road-side pool; field accession number CAM 12.34.1 8/IX/2011; (ii) Beau, Cameroon (4.15333° N; 9.25333° E). *Cx. nebulosus*—2 males. Collected and identified by AJC, 2011. Reared from a larva found in a bucket of water; field accession number CAM 12.22.1 3/IX/2011. Reared from larva collected in a concrete water reservoir; field accession number CAM 12.37b 10/IX/2011; (iii) Nieta, Cameroon (2.72617° N; 10.07096° E). *Cx. nebulosus*—1 male. Collected and identified by AJC, 2013. Reared from larval collection in a banana leaf axil; field accession number CAM 12.58.16 6/IX/2013.

Culex (Culiciomyia) semibrunneus Edwards, 1927

Male: Wing 2.24 mm. Proboscis 1.50 mm. Forefemur 1.30 mm.

Head: Well-defined row of broad white decumbent scales along eye margin (Fig. 28A); narrow pale decumbent scales extend towards back of vertex; a few broad white to translucent decumbent scales pre-laterally on vertex and patch of white decumbent scales on each side of head; erect forked scales quite numerous and appear dark brown to black depending on angle of view. Maxillary palpus slightly longer than proboscis, palpomeres 4 and 5 bearing setae, palpomere 4 about 0.33 length of palpomere 5 (ratio 0.33–0.38); palpal comb scales resemble those of *Cx. nebulosus* except those of distal set have rounded tips (spatulate or oblanceolate) and lack sharply recurved (hawk beak-like) aristate tips that occur in *Cx. nebulosus* (Fig. 28B,D), 4–6 proximal scales and 6 or 7 distal scales. Antenna and proboscis similar to those of other *Culiciomyia*.

Thorax: Scutal integument uniformly dark reddish brown (Fig. 28A) and scutum and scutellar lobes covered with short brown to shiny translucent setae. Integument of thoracic pleura similar reddish brown as scutal integument (Fig. 28C), which gives appearance of males of this species being generally darker than males of other *Culiciomyia*, lower 0.5 of mesanepimeron, entire metepisternum and mesomeron light brown to yellowish in some specimens (Fig. 28C); pleura with scales and setae present as follows: on anteprenotum, a row of 6 on posterior margin of postpronotum, 3 on proepisternum, prealar knob, a few on upper and posterior margin of mesokatepisternum, and one lower mesepimeral seta. Capitellum of halter darker than pedicel.

Wing: Scales all dark.

Legs: Coloured similar to those of other *Culiciomyia*, except for *Cx. cambournaci*.

Abdomen: Terga dark brown with paler basal lateral spots; sterna lighter brown than terga.

Genitalia: Each tergum IX lobe with irregular row of 8 or 9 setae. Gonocoxite more rounded than in other *Culiciomyia*, ventral surface with few long setae, each about as long as gonocoxite, positioned in no obvious pattern (Fig. 29A,C). Patch of ventrolateral setae less conspicuous than in *Cx. nebulosus*; row of 4 setae on dorsal surface of gonocoxite at apex below gonostylus (Fig. 29B), otherwise dorsal surface with quite sparsely distributed short setae. Specialized setae attached to an undivided subapical lobe consist of 3 rod-like setae with short hooked apices (*a–c*), most proximal little shorter than other 2, 1 quite robust seta (*e*) attached just above *a*, a short setae above seta *e*, 2 barbed slightly broadened setae (*d¹–d²*), relatively narrow foliform seta (*f*) and broader foliform seta (*g*). Gonostylus with small crest and 2–4 “spinelets” (Fig. 29B), 1–3 setae medially, 1 seta below crest, and 1 preapical seta, end of gonostylus curved upward with a small folded apical foliform gonostylar claw; membranous dorsal flange smaller than in *Cx. nebulosus*. Aedeagus with expanded triangular base with a short basal tooth (Fig. 30A,B), lateral plate long and smooth with rounded apex. Paraproct with 3 subapical setae (Fig. 30A) and a small rounded basal protrusion (Fig. 30A,B); apical region or crest of paraproct typically resembles that of all other *Culiciomyia*.

Additional notes: This is a small brown mosquito with no obvious features distinguishing it from *Cx. nebulosus* and allied species except for the rounded tips of the distal scales of the palpal comb of palpomere 3. The phallosome has long smooth lateral plates and the aedeagus has a small basal tooth. The basal protrusion or arm of the paraproct is similar in size and shape to that of *Cx. pseudosubaequalis*.

Specimens examined: Three males collected by sweep netting through vegetation in the Talangaye Forest, Cameroon. Field accession numbers CAM 132b1b 31/I/2017, CAM 132b1e 31/I/2017 and CAM 132ki 5/II/2017. One male from Yangambi, Stanleyville, Democratic Republic of the Congo, collected by Dr. Parent, 1944. Specimen held in the insect collection of the National Health Laboratory Services Museum in Johannesburg, South Africa.

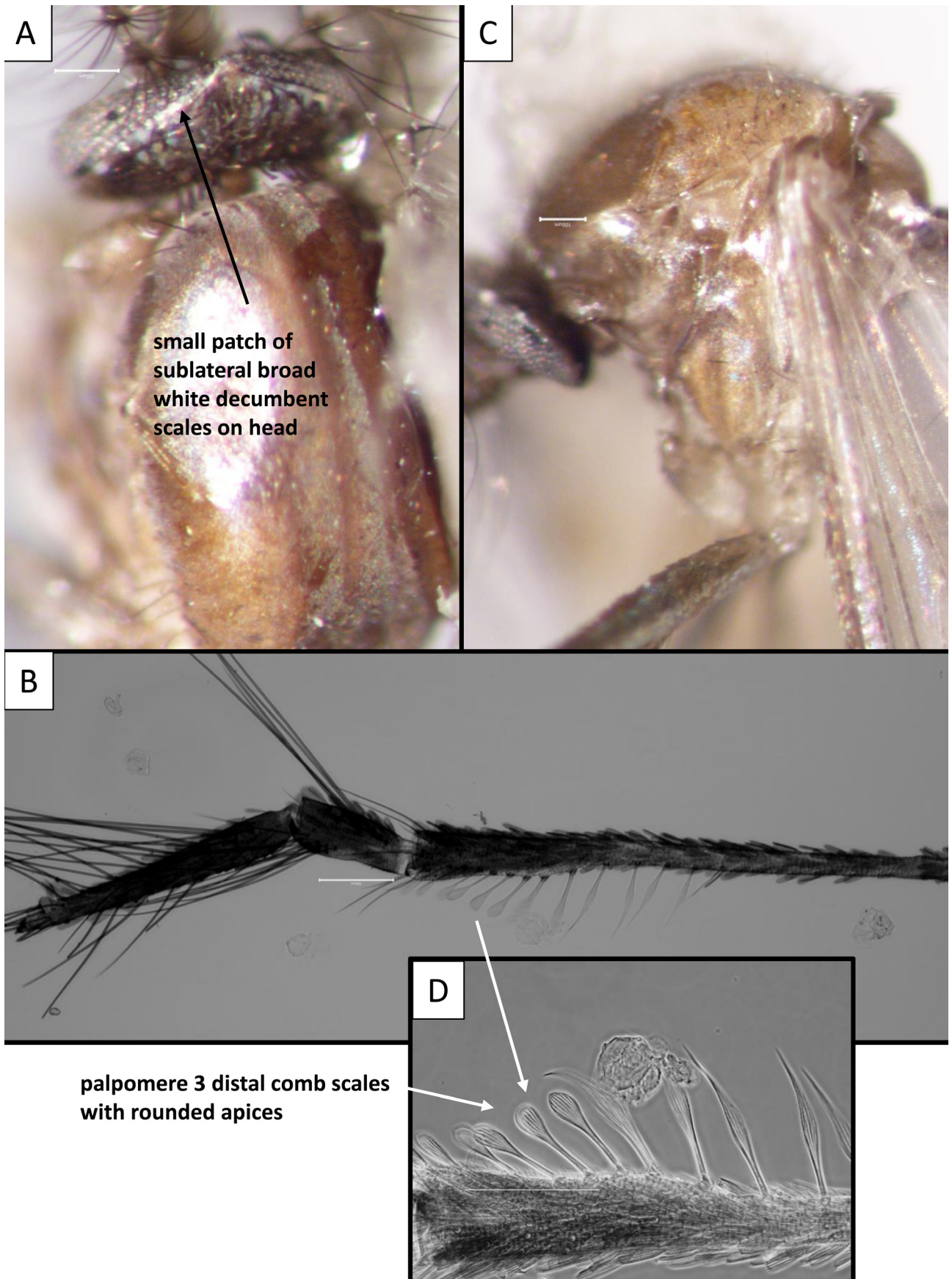


FIGURE 28. External features of *Culex semibrunneus*. (A) Dorsal view of the head and scutum; (B) lateral view of the maxillary palpus (100x); (C) lateral view of the thorax; (D) lateral view of the palpal comb scales (400x).

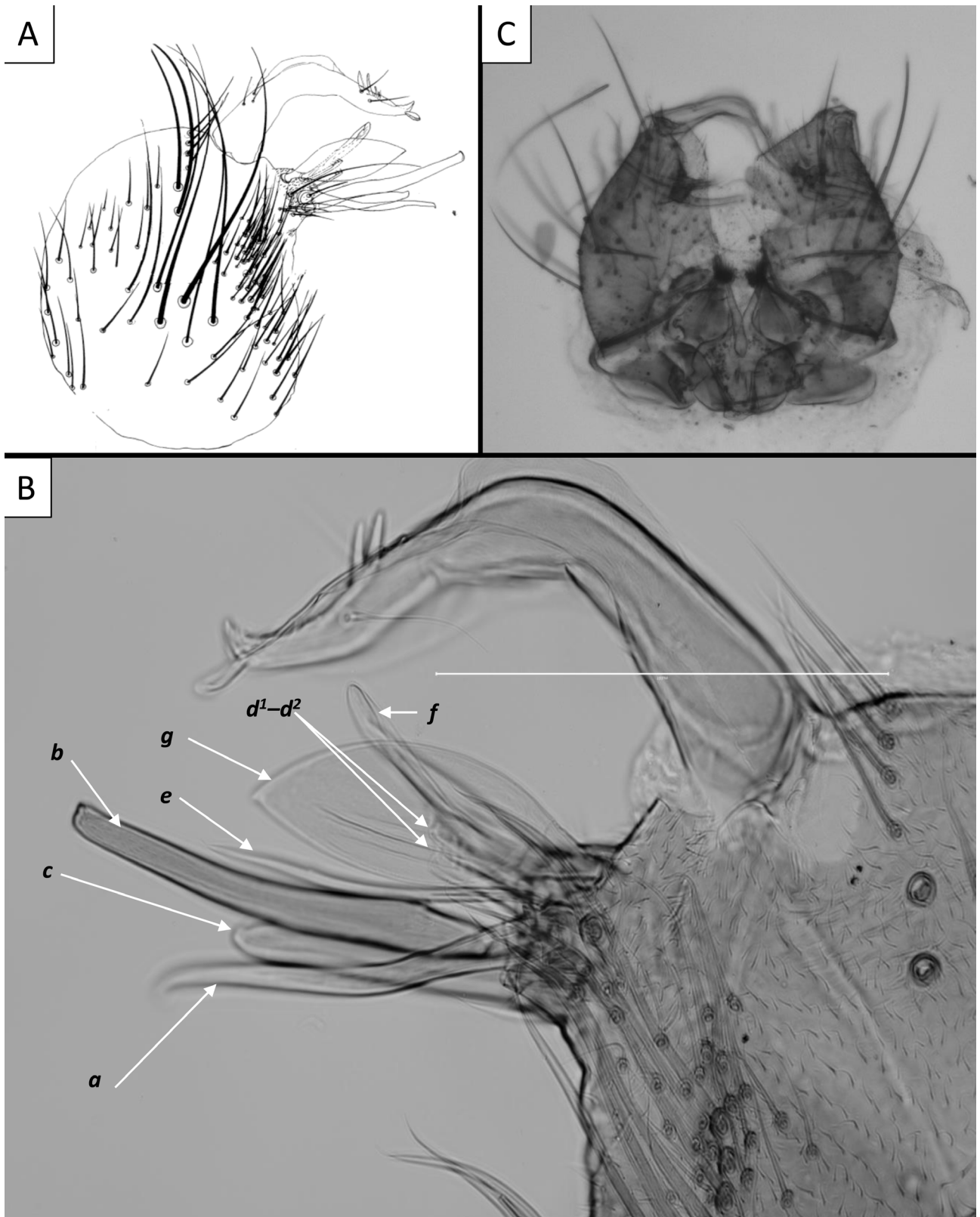


FIGURE 29. Gonocoxite of *Culex semibrunneus*. (A) Drawn depiction; (B) subapical lobe and gonostylus (400x); (C) genitalia (100x).

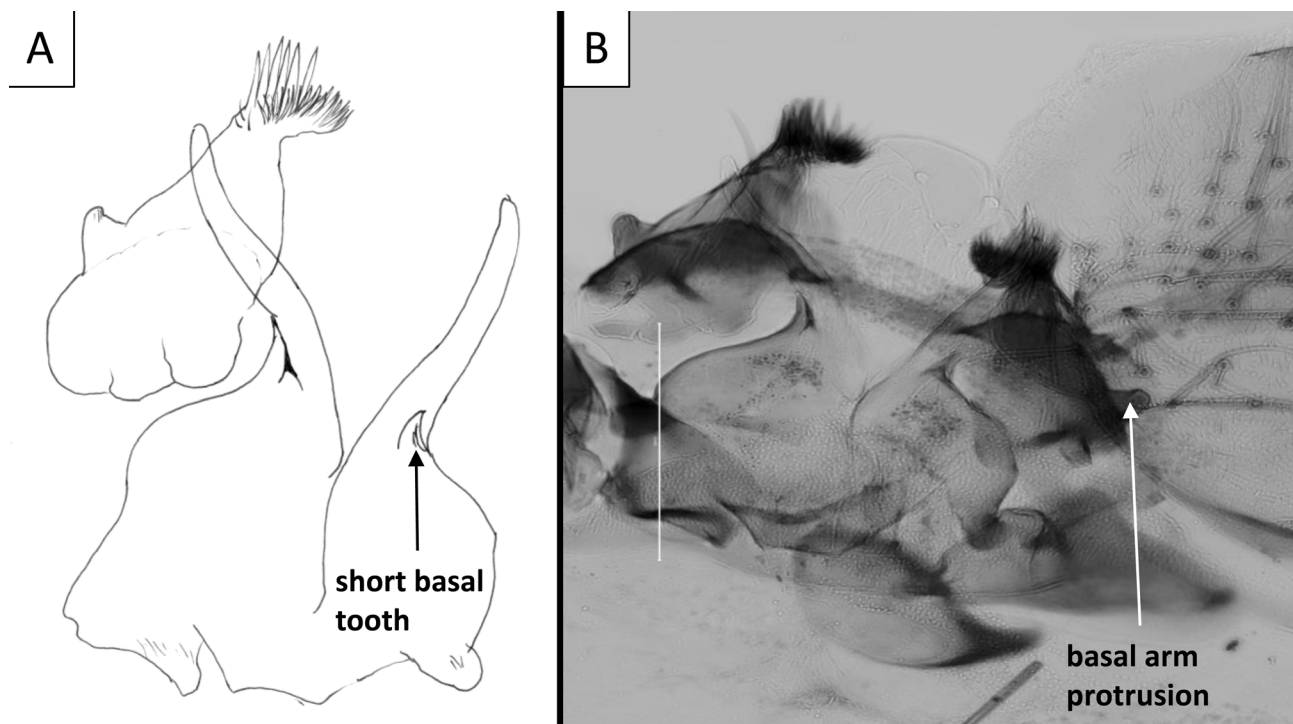


FIGURE 30. Lateral plates and paraproct of *Culex semibrunneus*. (A) Drawn depiction; (B) phallosome (400x).

Culex (Culiciomyia) subaequalis Edwards, 1941

Male. Wing 2.70 mm. Proboscis 1.90 mm. Forefemur 1.40 mm. Middle 0.33 of thoracic pleura light grey, upper 0.33 dark grey and lower 0.33 light brown, creating a pattern similar to that of *Cx. cinerellus*, except upper 0.33 darker than lower 0.33, whereas in *Cx. cinerellus* the upper and lower thirds have similar dark shading.

Head: White narrow decumbent scales on middle and back of vertex, single line of broad white decumbent scales along eye margin, forming a patch on each side of head (Fig. 31A), erect forked scales numerous and uniformly dark brown. Maxillary palpus longer than proboscis, with palpomeres 4 and 5 bearing setae, palpomere 4 about 0.75 length of palpomere 5 (ratio 0.74) (Fig. 31E), palpal comb consists of 6 or 7 equidistant translucent lanceolate scales (Fig. 31B,E), most anterior scale about length or slightly less its length away from apex of palpomere 3 (Fig. 31B). Antenna and proboscis similar to those of other *Culiciomyia*.

Thorax: Scutal integument light brown and covered with short brown setae, scutellar lobes with few narrow white scales. Pleural integument appears to have 2 stripes (Fig. 31D), dark antepronotum, middle 0.33 of postpronotum, postspiracular area and upper 0.5 of subspiracular area, prealar area and upper 0.33 of mesanepimeron form upper dark grey to brown stripe, lower light brown stripe corresponds to darker integument of proepisternum, lower part of subspiracular area, mid-upper part of mesokatepisternum and lower margin of mesanepimeron; no scales on pleura, setae on antepronotum, a row of 6 setae on posterior margin of postpronotum, 3 on proepisternum, prealar knob, all coxae, a few in upper and posterior margin of mesokatepisternum and a single lower mesepimeral seta. Halter with dark capitellum.

Wing: Scales all dark.

Legs: Coloured similar to those of other *Culiciomyia*, except for *Cx. cambournaci* (Fig. 31C).

Abdomen: Terga greyish brown with slightly paler small basal lateral spots; sterna generally grey and lighter than terga.

Genitalia: Tergum IX lobes each with row of 7 setae. Ventral surface of gonocoxite with few long setae all shorter than length of gonocoxite, in no obvious configuration, rows of 3–5 narrow setae along inner margin (Fig. 32A); dorsal surface sparsely covered with short setae; single undivided subapical lobe bearing 3 rod-like setae (*a–c*), most anterior longest and most posterior shortest, a single long quite robust and slightly striated seta (*e*), 2 small barbed setae (*d¹*, *d²*), 1 small seta, 1 quite broad foliform seta (*g*) and a smaller narrower seta (*f*). Gonostylus

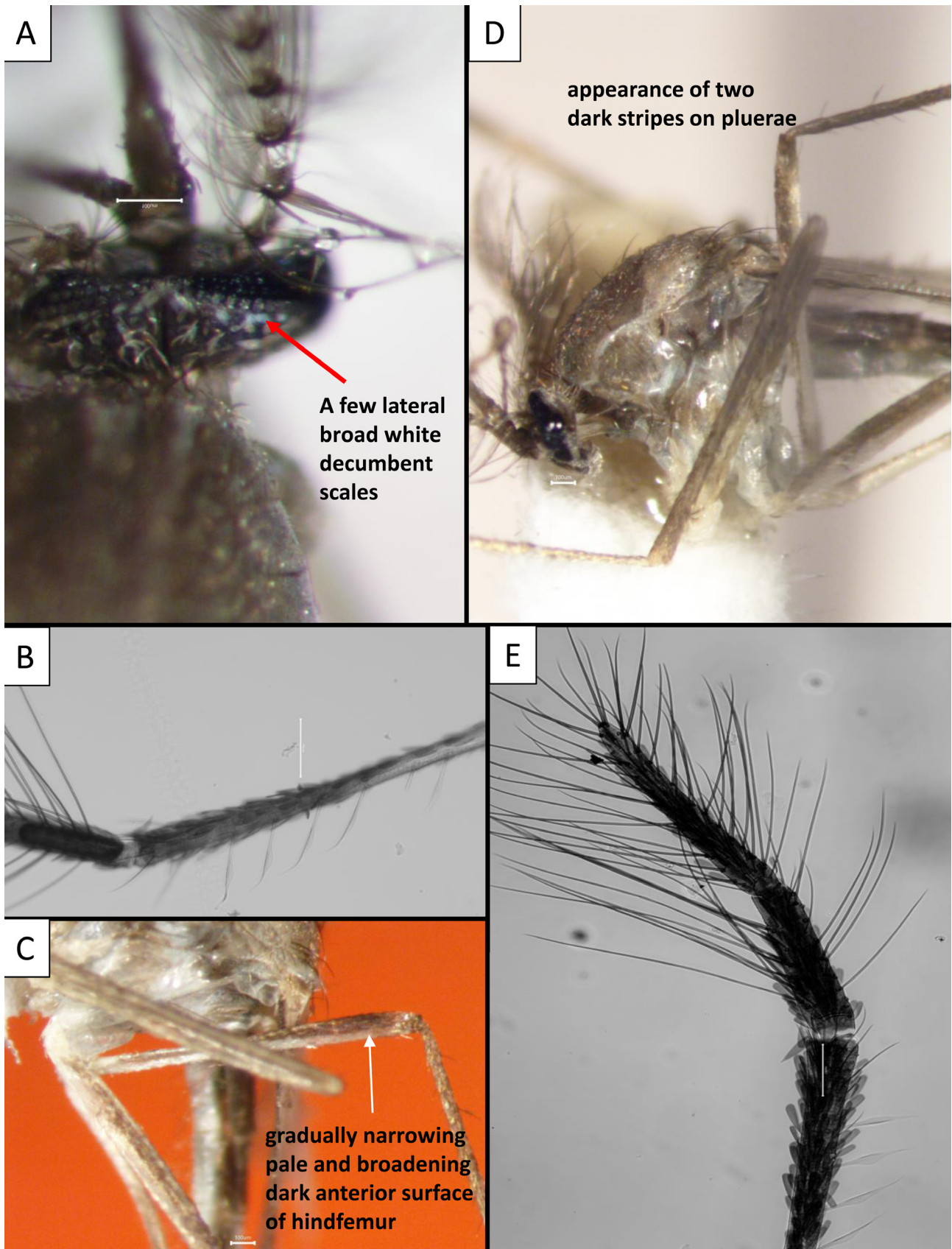
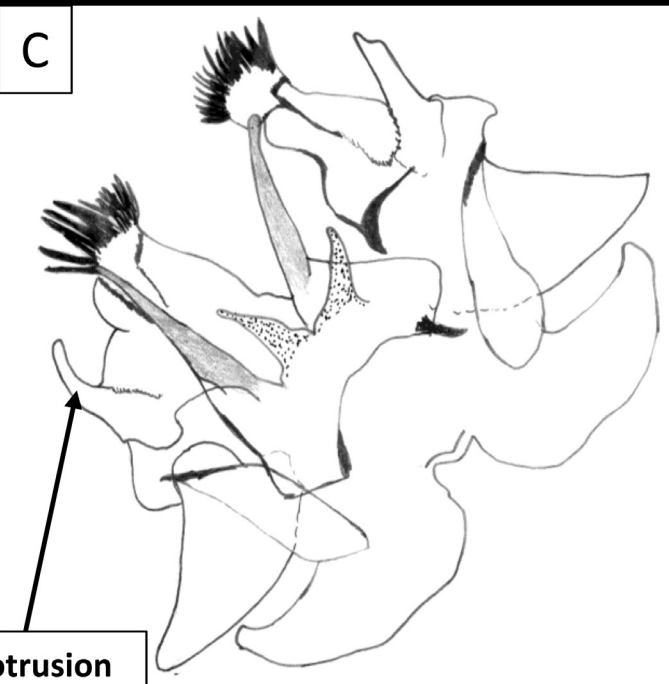
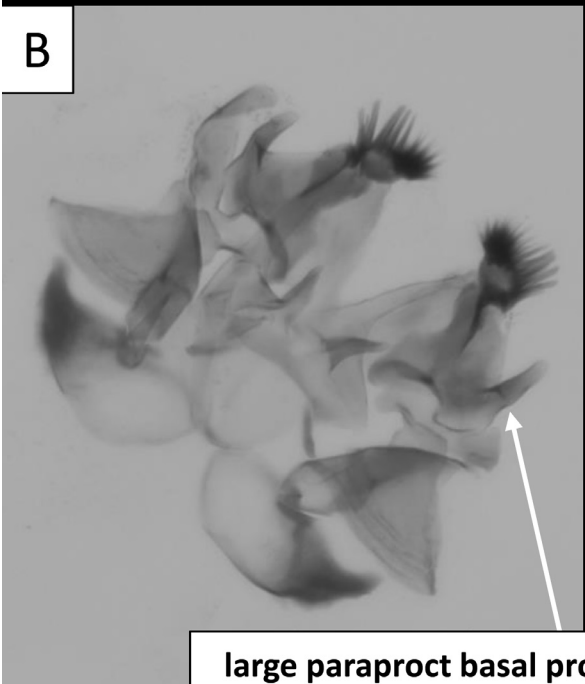
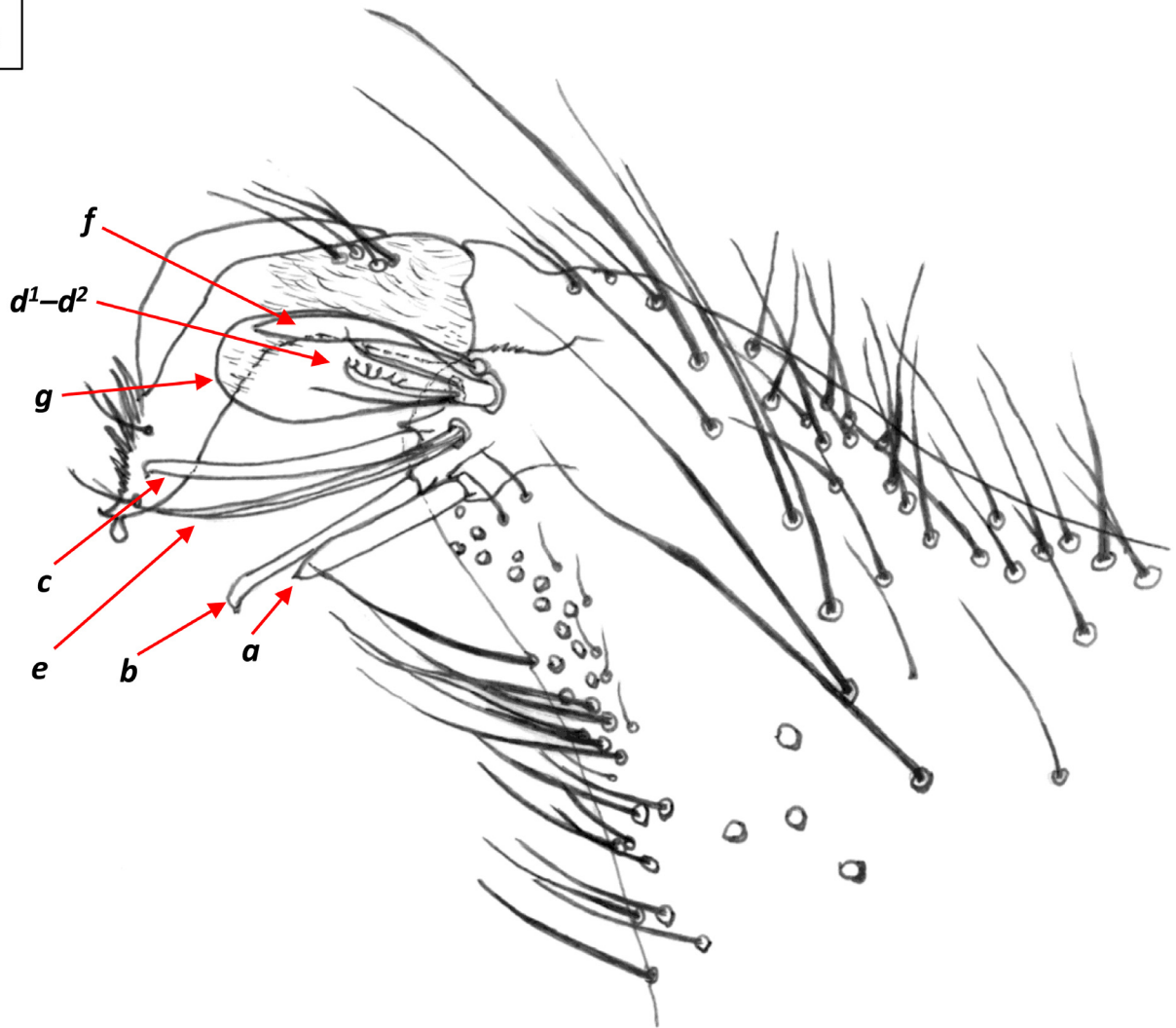


FIGURE 31. External features of the male of *Culex subaequalis*. (A) Dorsal view of the head (vertex); (B) lateral view of the palpal comb scales (100x); (C) anterior view of the hindfemur; (D) lateral view of the thorax; (E) distal two palpomeres of the maxillary palpus (100x).

A



large paraproct basal protrusion

FIGURE 32. Male genitalia of *Culex subaequalis*. (A) Gonocoxite; (B) phallosome (400x); (C) drawn depiction of the phallosome.

with crest of 7 or 8 “spinelets” (Fig. 32A), a prominent dorsal membranous flange and ending in an upturned hook and a small membranous seta, dorsal margin before bend with 4 setae, 1 seta below middle of crest and 1 subapical seta, much of proximal 0.5 covered with short fine setae. Aedeagus with expanded base with a prominent basal hook or tooth (Fig. 32B,C); lateral plate without teeth or tubercles but quite long and gradually narrowed to a rounded tip. Paraproct with 2 or 3 subapical setae and a distinct basal arm about 0.75 length of paraproct (Fig. 32B,C).

Additional notes: See notes under *Cx. pseudosubaequalis*.

Specimens examined: One male collected by sweep netting through vegetation in the Talangaye Forest, Cameroon. Field accession number CAM 152a1 4/II/2017.

Discussion

The only *Culiciomyia* larvae that we successfully reared to adults were those of *Cx. nebulosus*. Egg rafts of this species were commonly found in bamboo pots and larvae were found in some tree holes. The larvae of *Cx. nebulosus* agree with the description provided in Hopkins (1952). A role that species of *Culiciomyia* might play in the transmission of pathogens of human diseases has never been suspected and, hence, species of the subgenus have never received much attention. Most species of Afrotropical *Culiciomyia* are forest dwellers but a few, such as *Cx. nebulosus*, *Cx. cinereus* and *Cx. cinerellus*, are found commonly in drier savanna regions where they develop in tree holes and, in the case of *Cx. cinerellus*, in crab holes. Not much is known about the blood feeding behaviour of *Culiciomyia*, but they mostly feed on birds. Because of their bird feeding behaviour, we may find that they play a role in avian malaria transmission.

Culiciomyia represent a difficult group of mosquitoes that are difficult to distinguish from one another because the females are very similar and many males have overlapping patterns of maxillary palpal comb scales and similar genital morphology. All Afrotropical *Culiciomyia*, except *Cx. cambournaci*, have palpal comb scales that are not present in other *Culex*. *Culex cambournaci* appears to resemble a hybrid between a *Culiciomyia* species, perhaps *Cx. cinereus*, and a *Eumelanomyia* species, perhaps *Cx. simpliciforceps*, which have short male maxillary palpi. Thus far, *Cx. cambournaci* has only been found in the West African islands of São Tomé and Príncipe.

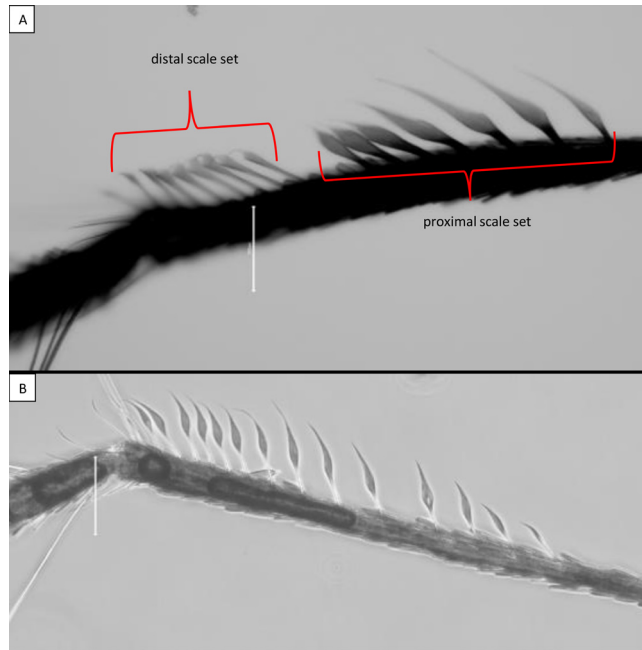
In our view, the colouring of the male pleural integument cannot be used reliably for species determination, except for *Cx. cinerellus*. Pleural colouring and the extent of grey dusted appearance were found to be quite variable within species, suggesting that this feature may be influenced by environmental conditions, such as larval diet, temperature and exposure to UV light, or perhaps the age of the mosquito. The only reliable means for separating or distinguishing the species is a combination of careful examination of palpal comb scales, relative lengths of palpomeres 4 and 5 and features of the male genitalia. A key that does not rely on the dissection of genitalia and uses only the formation of the palpal comb scales is provided below. This key does not separate all species because multiple species share identical palpal comb scaling. A second identification key that relies on the dissection and careful examination of the genitalia and palpal comb scales is provided that will separate all male species. An identification key based for females is not possible because of their similar morphologies.

Identification, especially of females, would be much advanced using molecular-based identification. Until one or more species of *Culiciomyia* are proven to serve as disease vectors, funding to develop molecular identification assays would be difficult to procure. However, because they are represented by many morphologically similar species living in sympatry in African forests, they may serve as an interesting group for studies of evolutionary and speciation biology and theory.

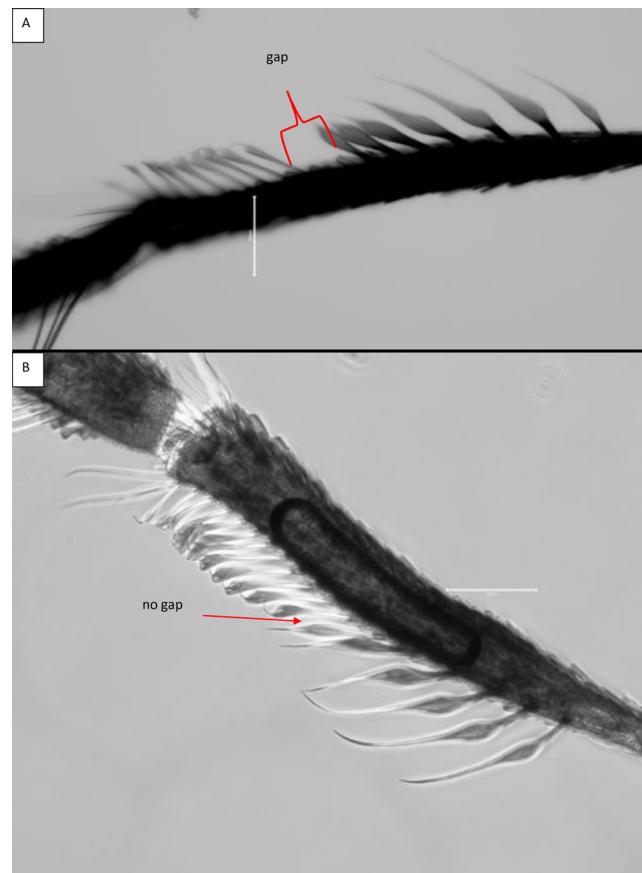
Key to males of Afrotropical *Culiciomyia* based on maxillary palpomere lengths and maxillary palpal comb scales

This key is based on the relative lengths of the maxillary palpomeres and specialized palpal comb scales. *Culex cambournaci*, which has no comb scales (the male palpi are very short), and *Cx. milloti*, which is known from larvae only, are excluded. White lines on the images are 100 µm in length. Additional notes are provided at the end of this key.

- 1 Palpal comb scales consisting of 2 or more clearly differently shaped scales divided into proximal and distal sets (Fig. 33A) 2
- Palpal comb scales all similar in shape (lengths of scales may vary) (Fig. 33B). 6



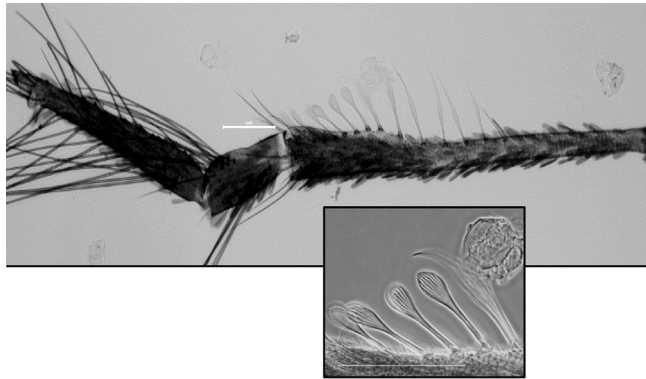
- 2(1) A gap separates the proximal and distal sets of palpal comb scales (Fig. 34A). 3
- No obvious gap between the proximal and distal sets (Fig. 34B) 4



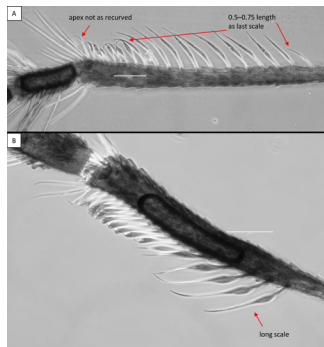
- 3(2) Palpal comb with 6–8 proximal and 6–8 distal scales; scales of distal set with long recurved apices (Fig. 35) *lanzaroi, muspratti, nebulosus*
- Palpal comb with 11–13 scales; scales of distal set small with very short, fine and straight points *furlongi*



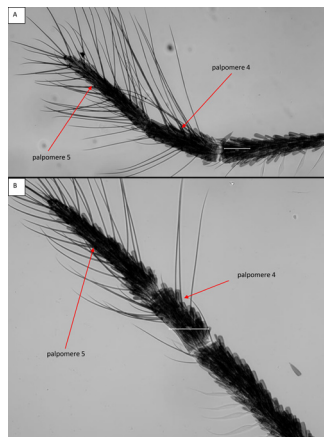
- 4(2) Scales of distal set of palpal comb scales not narrowed to a point, spatulate in shape (Fig. 36) *semibrunneus*
 - Scales of distal set of palpal comb scales with sharp apices that are often recurved. 5



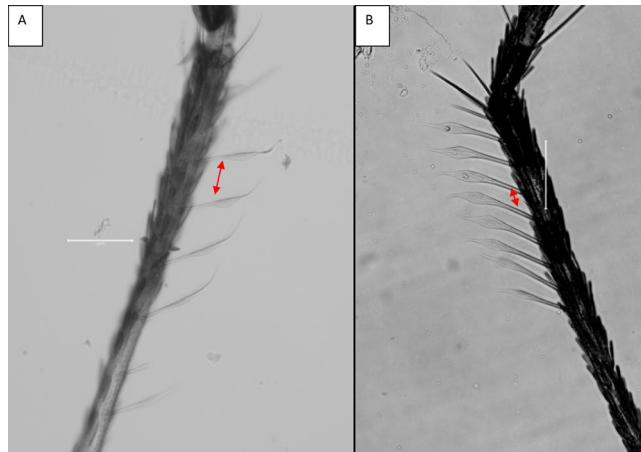
- 5(4) Most proximal scale of proximal set of palpal comb scales 0.5–0.75 length of most distal scale of proximal set; most distal scale of distal set with apical point not as recurved as other distal scales (Fig. 37A). *cinereus*
 - Most proximal scale of proximal scale set as long or longer than most distal scale of proximal set (Fig. 37B). *apicopilosus*



- 6(1) Palpal comb with 4 spatulate scales; Madagascar only *pandani*
 - Palpal comb with more than 4 scales 7
 7(6) Palpomere 4 >0.5 length of palpomere 5 (Fig. 38A) 8
 - Palpomere 4 <0.5 length of palpomere 5 (Fig. 38B) 9



- 8(7) Distance between palpal comb scales 0.5 length of each scale (Fig. 39A) *subaequalis, mongiro?*
 - Distance between palpal comb scales 0.25 length of each scale (Fig. 39B) *pseudosubaequalis, mongiro?*



- 9(7) Most proximal scales of palpal comb shorter than the other 12–15 scales (Fig. 40A) *cinerellus*
 - All scales of palpal comb about same length; variable in number (Fig. 40B) *macfiei*

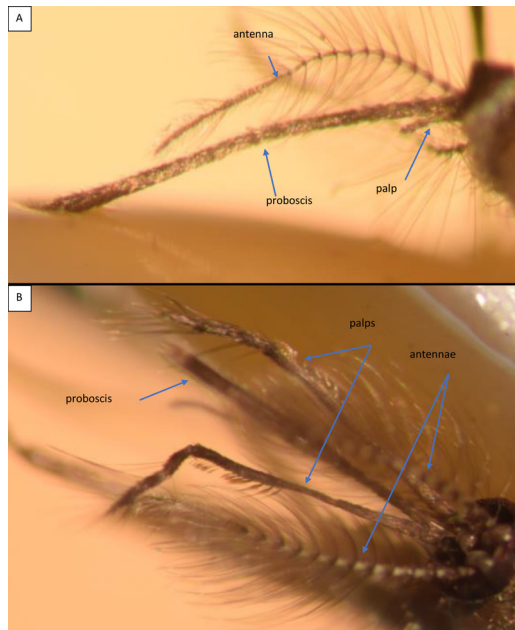


Additional notes: Descriptions in the literature of species-specific differences in palpal comb scales are too general to be able to make clear distinctions between the species that are not included in the key. Perhaps *Cx. eouzani*, which according to Geoffroy (1971) has seven palpal comb scales, has fewer comb scales than the other of these species, i.e. *Cx. gilliesi* (Hamon & van Someren 1961), *Cx. grenieri* (9–10 scales mentioned in Eouzan 1969), *Cx. harleyi* (Peters 1955) and *Cx. liberiensis*. *Culex mongiro* is similar to *Cx. subaequalis*, which has palpomere 4 about 0.75 the length of palpomere 5. It is not known if the palpal comb scales of *Cx. mongiro* are close together as in *Cx. pseudosubaequalis* or further apart as in *Cx. subaequalis*.

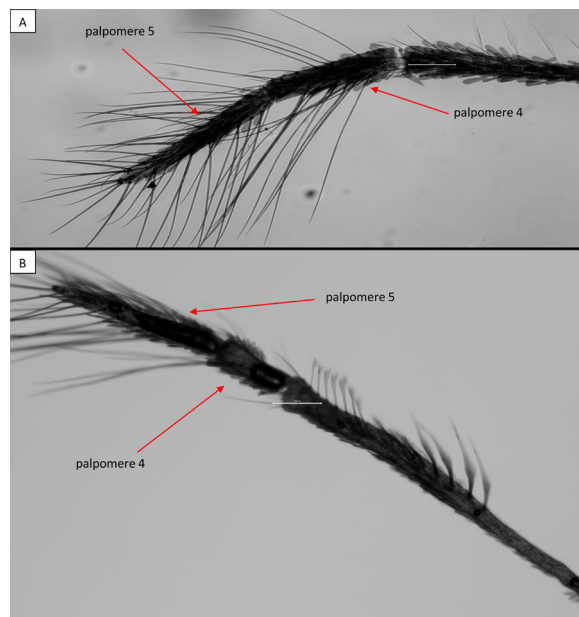
Key to males of Afrotropical *Culicomyia* based on habitus and genitalia

Culex milloti is not included in this key as it is known only from the larva. Characters used in the key for those species that were not collected by us are taken from published descriptions and drawings. Arrows in the figures point to features mentioned in the couplets. Barbed setae on the subapical lobe of the gonocoxite, when present, are often quite difficult to see because they are very small, delicate and thin, and often obscured by setae.

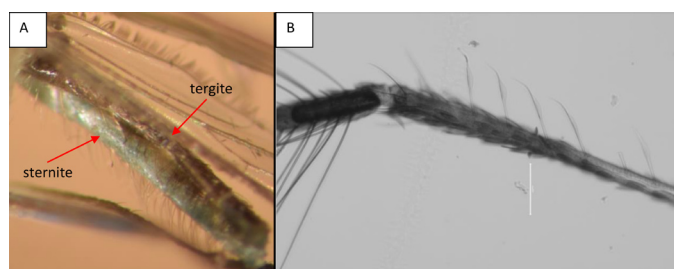
- 1 Maxillary palpus 0.16 length of proboscis (Fig. 41A); São Tomé and Príncipe only *cambournaci*
 - Maxillary palpus longer than proboscis (Fig. 41B) 2



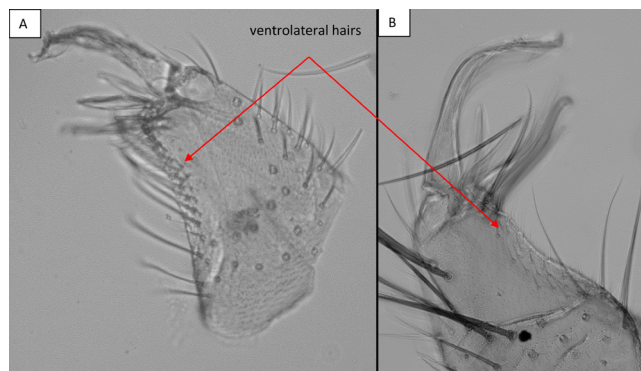
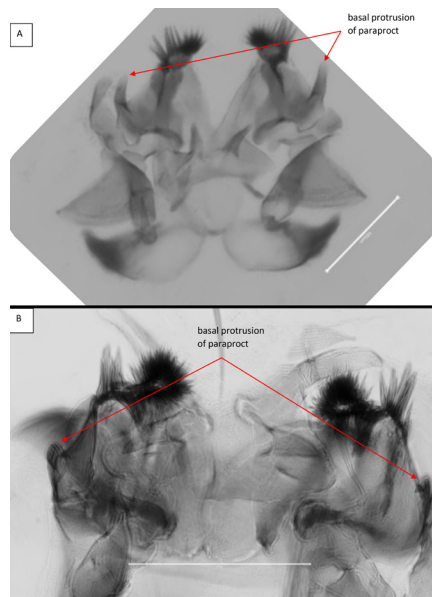
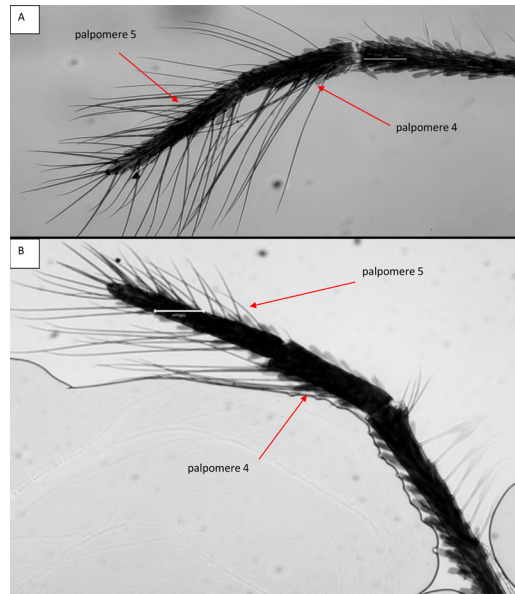
- 2(1) Palpal comb with 4 scales; Madagascar only *pandani*
 - Palpal comb with more than 4 scales 3
 3(2) Palpomere 4 >0.5 length of palpomere 5 (Fig. 42A) 4
 - Palpomere 4 <0.5 length of palpomere 5 (Fig. 42B) 6



- 4(3) Distinct apicolateral pale spots on terga 3–7; sterna with pale apical bands; palpal comb with 11–14 scales; no basal protrusion or arm on paraproct; subapical lobe of gonocoxite without barbed setae *mongiro*
 - Apicolateral pale spots indistinct or absent on abdominal terga; sterna uniformly dark beige to grey and lighter than the darker terga (Fig. 43A); palpal comb with 6–8 scales (Fig. 43B); paraproct with distinct basal arm or protrusion; subapical lobe of gonocoxite with 2 barbed setae 5

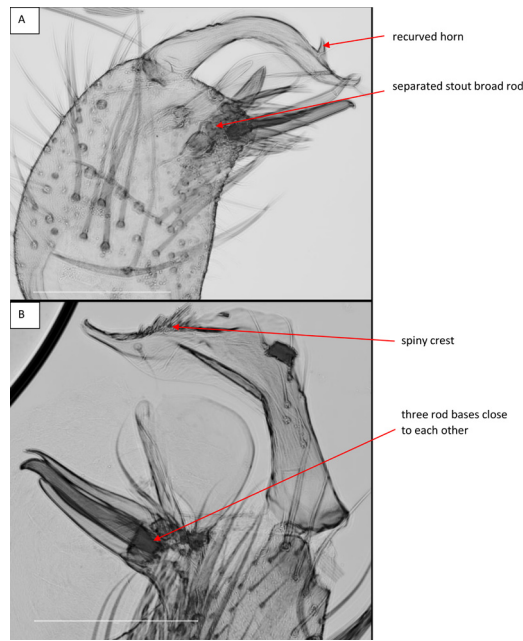


- 5(4) Palpomere 4 0.75 length of palpomere 5 (Fig. 44A); paraproct basal arm/protrusion >0.40 length of paraproct (Fig. 45A); gonocoxite with distinct ventrolateral patch of long piliform setae (Fig. 46A); *subaequalis*
- Palpomere 4 0.50–0.65 length of palpomere 5 (Fig. 44B); paraproct basal arm/protrusion <0.40 length of paraproct (Fig. 45B); gonocoxite without obvious ventrolateral patch of setae, setae present are short (Fig. 46B) *pseudosubaequalis*

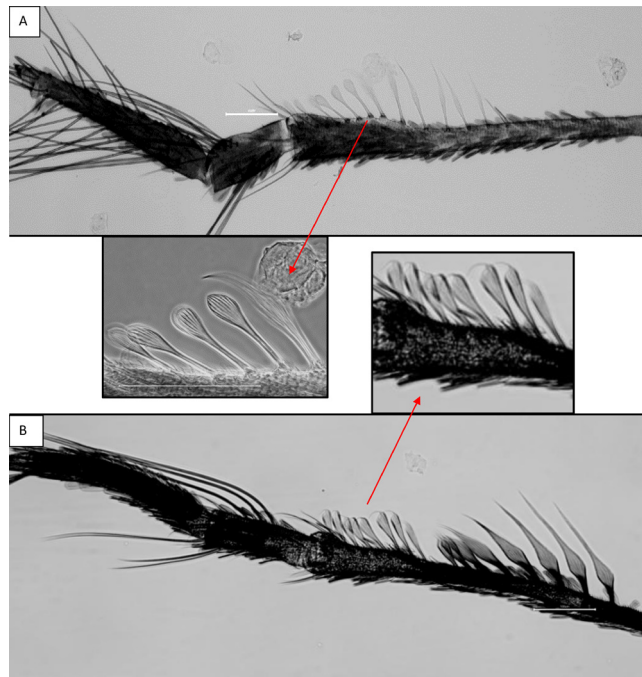


- 6(3) Gonostylus with single small recurved horn on upper margin; subapical lobe with a broad stout seta laterally separated from the other 2 stout rod-like setae (Fig. 47A) *macfie*
- Upper margin of the gonostylus with “spinelets” forming a crest; bases of the 3 rod-like setae more or less equally close to each

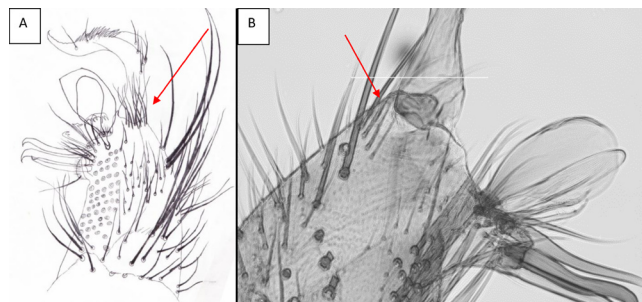
other (Fig. 47B), except for *Cx. cinereus*, which has 1 rod-like seta more distally positioned from the other 2 rod-like setae 7



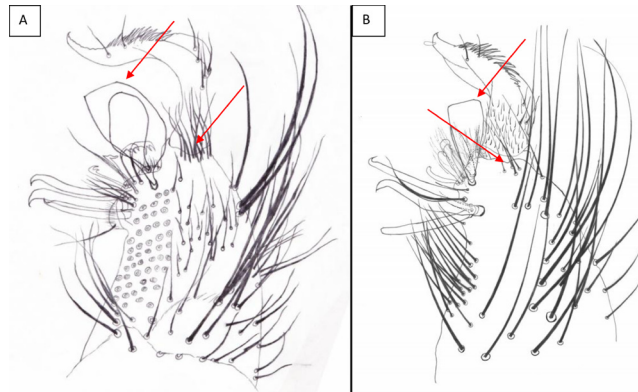
7(6) Palpal comb scales spatulate with rounded apices (Fig. 48A) *semibrunneus*
 - Palpal comb scales with pointed apices that are often recurved (Fig. 48B) 8



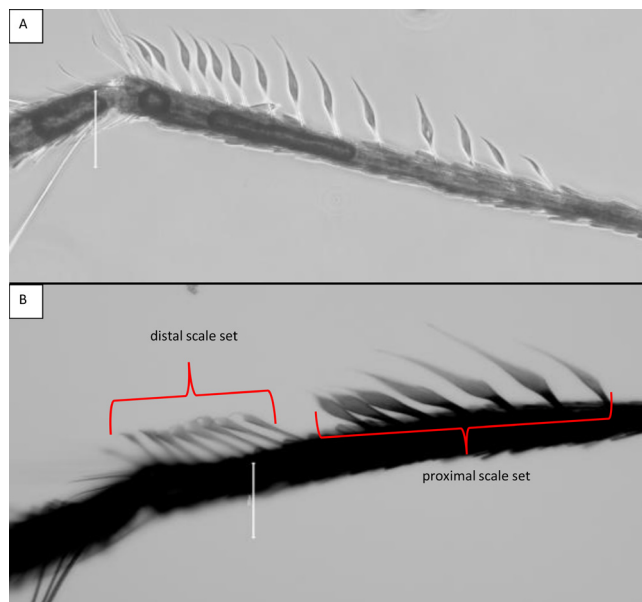
8(7) Apex of gonocoxite with a patch of delicate setae (Fig. 49A) 9
 - Apex of gonocoxite with a few setae in 1 or 2 rows (Fig. 49B) 10



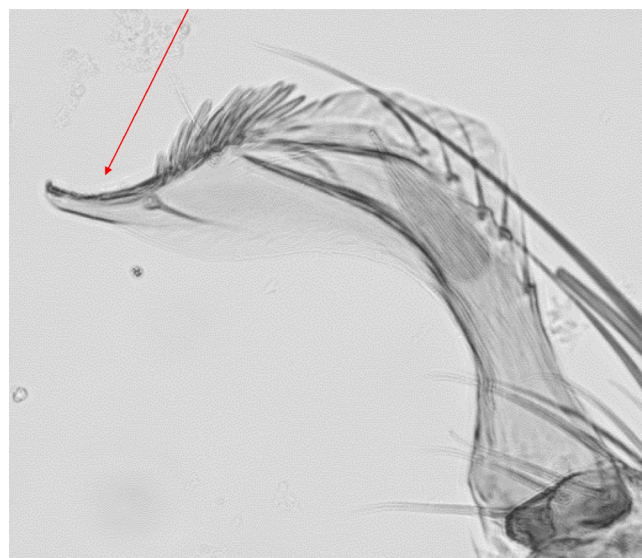
- 9(8) Apical delicate setae on gonocoxite not borne on an obvious protuberance; large foliiform seta *g* of subapical lobe not truncated (Fig. 50A) *apicopilosus*
- Apical delicate setae on gonocoxite borne on an obvious protuberance; large foliiform seta *g* of subapical lobe truncated (Fig. 50B) *eouzani*



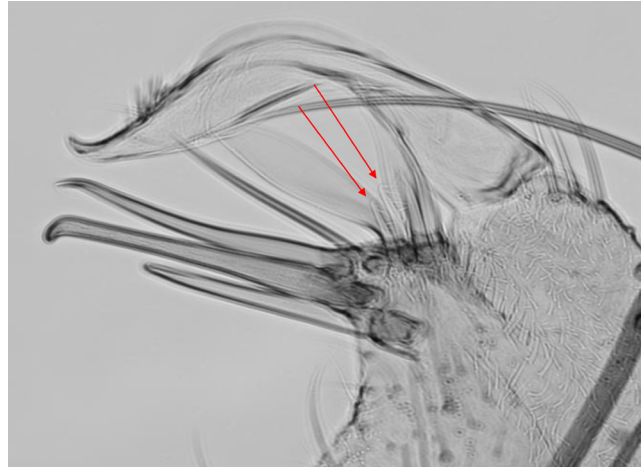
- 10(8) Palpal comb scales all similar in shape but may vary in length (Fig. 51A) 11
- Palpal comb comprised of at least 2 clearly differently shaped groups or sets of scales (Fig. 51B) 15



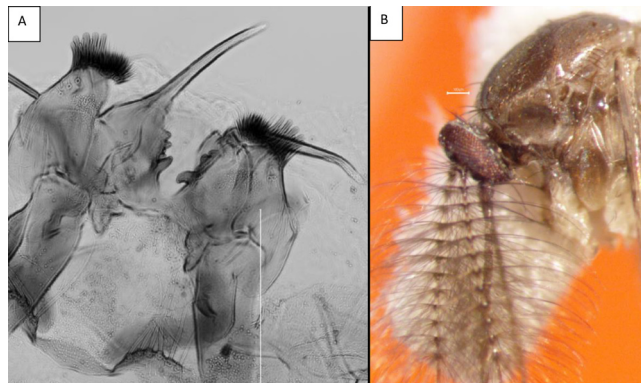
- 11(10) Gonostylus with crest extending to apex; lateral plate of phallosome very broad with a recurved basal tooth; ventral surface of lateral plate also broad, recurved and shagreened *harleyi*
- Crest of gonostylus not extended to apex (Fig. 52) 12



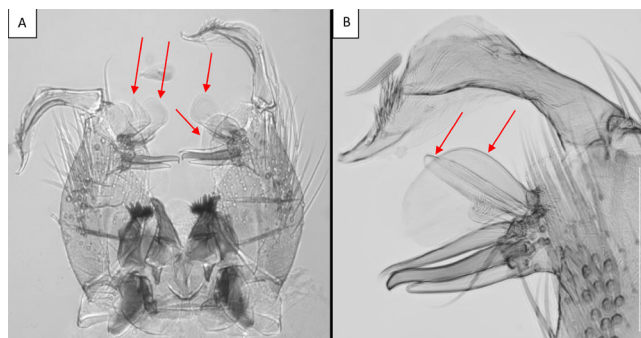
- 12(11) Subapical lobe with 2 or 3 barbed setae (Fig. 53) 13
 - Subapical lobe without barbed setae 14



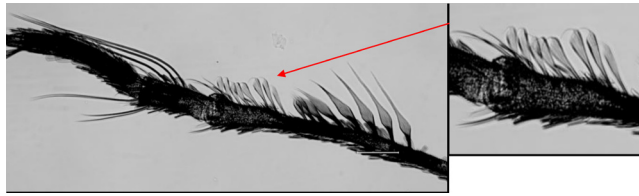
- 13(12) Subapical lobe of gonocoxite with 2 slightly barbed subapical setae (Fig. 53); lateral plate of aedeagus with long slender smooth arm and 5 or 6 rounded tubercles basally, no distinct sharp tooth basally (Fig. 54A); mid-region of thoracic pleura pale, separating 2 darker stripes (Fig. 54B) *cinerellus*
 - Subapical lobe of gonocoxite with 2 or 3 slightly barbed subapical setae; lateral plate of aedeagus with small recurved antero-medial tooth, ventral surface recurved with 9 or 10 broad teeth reaching apex; upper region of thoracic pleura darker than lower half *liberiensis*



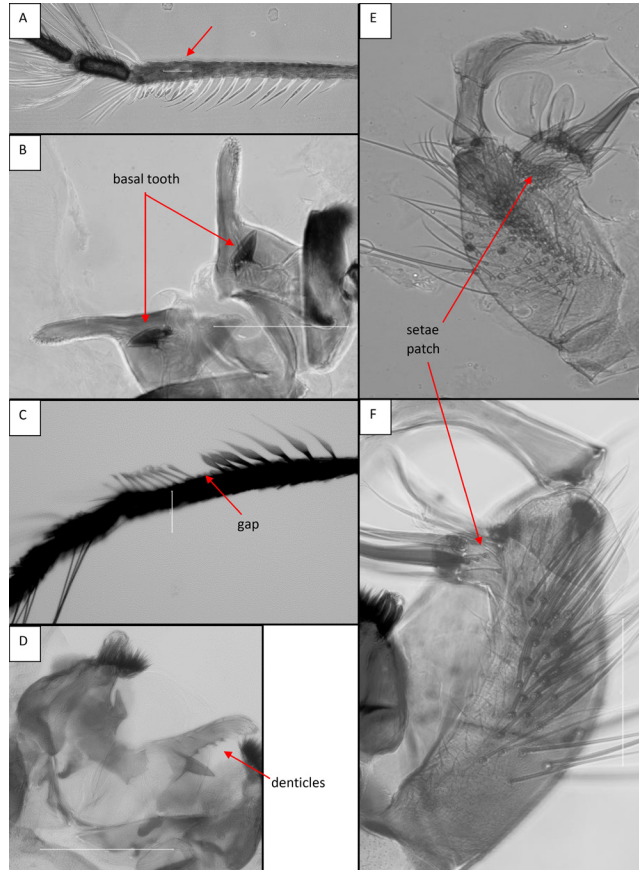
- 14(12) Foliform seta of subapical lobe (seta *g*) very broad; lateral plate of aedeagus with numerous sharp denticles (sometimes in 2 rows) not reaching apex *grenieri*
 - Foliform seta of subapical lobe of gonocoxite not very broad, 1 of 3 narrow setae on lobe bent apically; lateral plate of aedeagus with 6 strong tubercles basally, ending in long thin curved arm *gilliesi*
 15(10) Both foliform setae of subapical lobe of gonocoxite nearly equally broad (Fig. 55A) *lanzaroi*
 - One foliform seta of subapical lobe much narrower than the other (Fig. 55B) 16



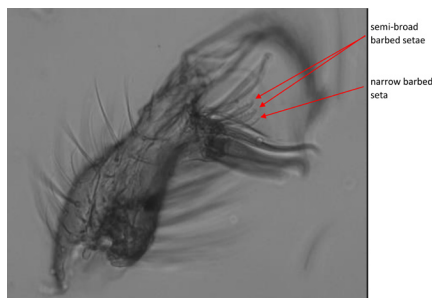
- 16(15) Palpal comb with 11–13 scales, those of proximal set spearhead-shaped with long fine pointed apices, scales of distal set small and lanceolate with very short straight fine apices. *furlongi*
 - Palpal comb with more than 13 scales, scales of distal set with fine strongly recurved apices (Fig. 56) 17



- 17(16) Palpal comb without gap between proximal and distal sets of scales (Fig. 57A); base of subapical lobe of gonocoxite with patch of numerous delicate setae (Fig. 57E); lateral plate of aedeagus smooth except for single basal tooth (Fig. 57B). *cinereus*
 - Palpal comb with gap between proximal and distal sets of scales (Fig. 57C); subapical lobe of gonocoxite without basal patch of setae (Fig. 57F); lateral plate of aedeagus with numerous sharp denticles (Fig. 57D) 18



- 18(17) Subapical lobe of gonocoxite with slightly broadened barbed setae *muspratti*
 - Subapical lobe of gonocoxite with 2 narrow and 2 slightly broadened barbed setae (Fig. 58) *nebulosus*



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