

Molecular phylogeny of *Kazimierzus Plisko, 2006* (Clitellata, Kazimierzidae) from the Western and Northern Cape Province inferred from mitochondrial DNA sequences

Thembeke Clara Nxele^{1,4}, Jadwiga Danuta Plisko^{1,2}, Tarombera Mwabvu^{3,4},
Oliver Tendayi Zishiri⁴

1 KwaZulu-Natal Museum, P. Bag 9070, Pietermaritzburg, 3200, South Africa **2** School of Life Sciences, University of KwaZulu-Natal, Private Bag X01, Pietermaritzburg campus, 3209, South Africa **3** School of Biology & Environmental Sciences, University of Mpumalanga, Private Bag X11283, Mbombela, 1200, South Africa **4** School of Life Sciences, University of KwaZulu-Natal, Private Bag X54001, Westville campus, Durban, 4001, South Africa

Corresponding author: Thembeke C. Nxele (tnxele@nmsa.org.za)

Academic editor: John Midgley | Received 18 April 2020 | Accepted 19 June 2020 | Published 10 July 2020

<http://zoobank.org/00E3D13B-B693-400C-A409-4F16DB41F1A0>

Citation: Nxele TC, Plisko JD, Mwabvu T, Zishiri OT (2020) Molecular phylogeny of *Kazimierzus Plisko, 2006* (Clitellata, Kazimierzidae) from the Western and Northern Cape Province inferred from mitochondrial DNA sequences. African Invertebrates 61(2): 83–92. <https://doi.org/10.3897/AfrInvertebr.61.53380>

Abstract

Species identification of earthworms using morphology can be challenging and inconclusive as homoplasy in many characters is high. The use of molecular DNA technology, such as the use of conserved regions in mtDNA and nuclear DNA has unravelled the phylogenetic background of several earthworm species. The current study utilised the cytochrome c oxidase subunit I (COI) mitochondrial marker to reconstruct the phylogeny of *Kazimierzus Plisko, 2006* species from the Western and Northern Cape provinces of South Africa. Phylogenetic reconstructions were implemented using Bayesian Inference, as well as Maximum Likelihood. Both tree building methods adhered to the monophyly of the majority of the taxa. Results showed that species fell into two clades and validated eleven currently known *Kazimierzus* species (*K. circulatus* (Plisko, 1998), *K. franciscus* (Pickford, 1975), *K. guntheri* (Pickford, 1975), *K. hamerae* (Plisko, 1998), *K. kleinoodi* Nxele & Plisko, 2017, *K. nietvoorbiji* Nxele & Plisko, 2017, *K. nieuwoudtvillensis* Nxele & Plisko, 2017, *K. occidualis* (Plisko, 1998), *K. pearsonianus* (Pickford, 1975), *K. phumlani* Nxele & Plisko, 2017, *K. sophieae* (Plisko, 2002)). Cryptic diversity is evident in *K. occidualis* with genetic diver-

gence greater than 12% amongst populations. *Kazimierzus franciscus* and *K. ljunstroemi* (Pickford, 1975) have a low genetic variability suggesting close relatedness or probably conspecificity. A group of specimens from Clanwilliam are morphologically identical to *K. sophieae*, but are genetically distinct and may belong to undescribed species. This study demonstrates the importance of integrative taxonomy in earthworms in order to present reliable taxonomic and biogeographic data.

Keywords

DNA, COI, mtDNA, taxonomy, earthworms, cryptic diversity

Introduction

Earthworms constitute a large component of soil invertebrates and are regarded as soil engineers (Jouquet et al. 2006). They alter soil properties and enhance nutrient cycling (Lavelle 1988) which determines plant community composition. Regardless of their importance, African earthworm fauna have not received much attention and their evolutionary relationships are subject to debate. Species identification is possible by investigating their anatomy (Chang et al. 2007, Csuzdi 2010, Csuzdi and Zicsi 2003, Plisko and Zicsi 1991), but the structural simplicity of their body plan and existence of cryptic species may hinder taxonomic classification (Domínguez et al. 2015). Traditional morphology-based identification also requires substantial taxonomic expertise in this group, because it involves observation of minute morphological characters (Richard et al. 2010). As such, the use of molecular DNA technology in species delineation is an important complement to morphological classification of earthworms.

The use of DNA sequences has increased in the recent past, because it is less subjective than morphological characters, allows for the analysis of several characters (Scotland et al. 2003) and is applicable at all developmental stages (Decaëns et al. 2013; Chang and James 2011). The molecular studies of earthworms that have used the mitochondrial cytochrome c oxidase subunit I (COI) gene in integrative taxonomy have shown good results (Blakemore 2013a, b; Blakemore et al. 2010; Bantaowong et al. 2011; Huang et al. 2007; Richard et al. 2010; King et al. 2008; Rougerie et al. 2009; Chang et al. 2009; James et al. 2010). This marker is able to distinguish the intra- and interspecific genetic variation and groups conspecifics together, because COI sequences are variable enough to differentiate between taxa, but are less variable in conspecifics (Rougerie et al. 2009; Stoeckle and Hebert 2008; Valentini et al. 2008).

According to Nxele (2012), knowledge of the taxonomic diversity of the indigenous megadriles in South Africa is incomplete. Therefore, integration of taxonomic methods is vital to improve the knowledge and understanding of the megadrile fauna. Furthermore, Plisko (2013) stressed that a molecular study on indigenous South African megadriles is essential in order to reveal the evolutionary relationships amongst them. Against this background, the phylogenetic relationships in *Kazimierzus*, a genus occurring in the Western and Northern Cape provinces of South Africa, was investigated.

Materials and methods

Sampling

In order to obtain earthworms of *Kazimierzus*, qualitative sampling was carried out in 2011 and 2015 during the rainy season (July–September) in the Western and Northern Cape, South Africa. Besides the focus on type localities, potential sites other than the type localities were also sampled. Earthworms were collected by digging and hand sorting. Collected specimens were anaesthetised in 20% ethanol solution, fixed in 4% formalin solution and preserved in 75% ethanol for taxonomic purposes. A subsample was preserved in absolute ethanol for molecular analysis. All specimens were examined using a Wild Heerbrugg stereomicroscope and were identified according to the descriptions in Plisko (1996, 1998, 2002), Pickford (1975), Michaelsen (1913) and Nxele et al. (2017). All specimens are at the KwaZulu-Natal Museum, Pietermaritzburg, South Africa.

Genomic DNA extraction, amplification and sequencing

Tissue from the posterior section of the earthworm was used. All DNA extractions were performed using the ZR Genomic DNATM Tissue MicroPrep kit, following the manufacturer's standard protocol. The concentration of DNA in each sample was estimated using the NanoDrop 2000 (Thermo Scientific). A fragment of the mitochondrial cytochrome c oxidase subunit I (COI) gene was amplified using LCO1490 (5' GGT-CAACAAATCATAAAGATATTGG 3') and HCO2198 (5' TAAACTTCAGGGT-GACCAAAAAATCA 3') primers (Folmer et al. 1994). Polymerase chain reactions (PCR) were performed in a final volume of 25 µl using the BIO RAD T100 Thermal Cycler and contained: 2 µl of DNA template (approximately 35 ng/ul), 12.5 µl One Taq Quick-Load 2X Master Mix with standard buffer, 0.5 µl of 10 uM forward and reverse primers and sterile water. The thermocycler conditions were as follows: 95 °C for 2.30 min for initial denaturation, followed by 35 cycles at 95 °C for 30 sec denaturation, 50 °C to 52 °C for 45 sec annealing and 72 °C for 75 sec extension. A final extension step at 72 °C for 10 min completed the reactions.

Sequencing of the 675 bp fragment of the COI mtDNA was conducted at Inqaba Biotechnical Industries (Pty) Ltd.

Sequence alignment and phylogenetic analysis

Identity of sequences was verified by the Basic Local Alignment Search Tool (BLAST) in the National Centre for Biotechnology (NCBI). Sequences of *Amyntas minimus* (Horst, 1893) and *Amyntas corticis* (Kinberg, 1867) were included as outgroup taxa. The sequences for outgroup taxa were obtained from GenBank (Accession nos: AB542509.1, AB542469.1) and current sequences are added as supplementary data.

All specimens are at the KwaZulu-Natal Museum. The sequences were aligned using CLUSTAL X 2.1 (Larkin et al. 2007). These alignments were then manually edited using BIOEDIT 3.3.19.0 (Hall 1998). Unreliable nucleotides (low signal strength), as well as primers sequences, were trimmed off at both the 5' and 3' ends. The programme JMODELTEST v.0.1.1 (Darriba et al. 2012) was used to select the best-fit evolutionary model using the AKAIKE Information Criterion (AIC; Akaike 1973). Phylogenetic analyses were based on two approaches, Bayesian Inference (BI) was performed using MRBAYES 3.2 (Huelsenbeck and Ronquist 2001) and Maximum Likelihood (ML) analysis was performed using GARLI (Zwickl 2011). In each case, the best-fit evolutionary model selected by JMODELTEST was specified.

Clade support was evaluated by 1000 bootstrap replicates for the ML analysis and posterior probability values for the Bayesian analysis. For Bayesian analyses, all MRBAYES analyses were run for 5000000 generations with a sampling frequency of 1000. The deviation of split frequencies was less than 0.01 at the conclusion of all analyses which confirmed that the MCMC chains had converged. The programme TRACER v1.5 (Drummond and Rambaut 2007) was used to check that the Effective Sampling Size > 200 and that posterior distribution for all parameters was unimodal. Consensus trees were generated using PHYLIP 3.69 (Felsenstein 2005) and viewed in FIG TREE v1.3.1 (Rambaut 2009). Uncorrected p genetic distances were obtained for the sequenced specimens using MEGA 6 (Tamura et al. 2013). Each species is represented by one specimen, except where the species appeared in more than one clade on the phylogenetic tree. Sequences have been deposited in GenBank (Table 1).

Results

Phylogenetic analyses

The sequences were 675 bp. Variable sites were 431 bp and conserved sites 233 bp showing great differentiation amongst taxa.

Most species pair comparisons showed a genetic distance above 13%, except for *K. ljungstroemi* and *K. franciscus* which have one percent genetic distance between them (Table 2).

The MI and BI trees were congruent, therefore, support values were annotated on to the branches of the most likely trees generated for each of the datasets analysed (ML run with no bootstrap, rooted using outgroup species (*Amyntas minimus* and *Amyntas corticis*).

Bootstrap values and posterior probabilities below 50% and 0.50, respectively, were not shown on the trees. Two clades, A and B, are distinct; clade A separates further to clades C and D whilst clade B separates to clades E and F (Fig. 1). Both tree building methods support monophyly of the majority of taxa (Fig. 1). *Kazimierzus sophieae* is paraphyletic, found in two distinct clades; one shared with *K. sp* and the second clade

Table I. Genbank COI sequences for investigated *Kazimierzus* species.

Species	Specimen catalog #	Genbank #
<i>K. circulatus</i>	NMSA/OLIG.04942/3a,c	MN982467–68
<i>K. franciscus</i>	NMSA/OLIG.09659a,b,c	MN982469–71
<i>K. guntheri</i>	NMSA/OLIG.04986a,b,e	MN982474–75; MN982478
<i>K. hamerae</i>	NMSA/OLIG.04956a,b,c	MN982461–63
<i>K. kleinoodi</i>	NMSA/OLIG.04987	MN982480
<i>K. lungstroemi</i>	NMSA/OLIG.06960b,d,e	MN982472–73; MN982482
<i>K. nietvoorbij</i>	NMSA/OLIG.04988a,c	MN982456–57
<i>K. nieuwoudtvillensis</i>	NMSA/OLIG.04990b,c,d	MN982458–60
<i>K. occidualis</i>	NMSA/OLIG.04958a,c,d; NMSA/OLIG.04962	MN982464–66; MN982485
<i>K. pearsonianus</i>	NMSA/OLIG.04984	MN982479
<i>K. phumlani</i>	NMSA/OLIG. 04951/2b,c,e	MN982453–55
<i>K. sophieae</i>	NMSA/OLIG.04950a,c; NMSA/OLIG.04963a,b	MN982483–84; MN982486–87
<i>K. sp.</i>	NMSA/OLIG.04989a,d,e	MN982476–77; MN982481

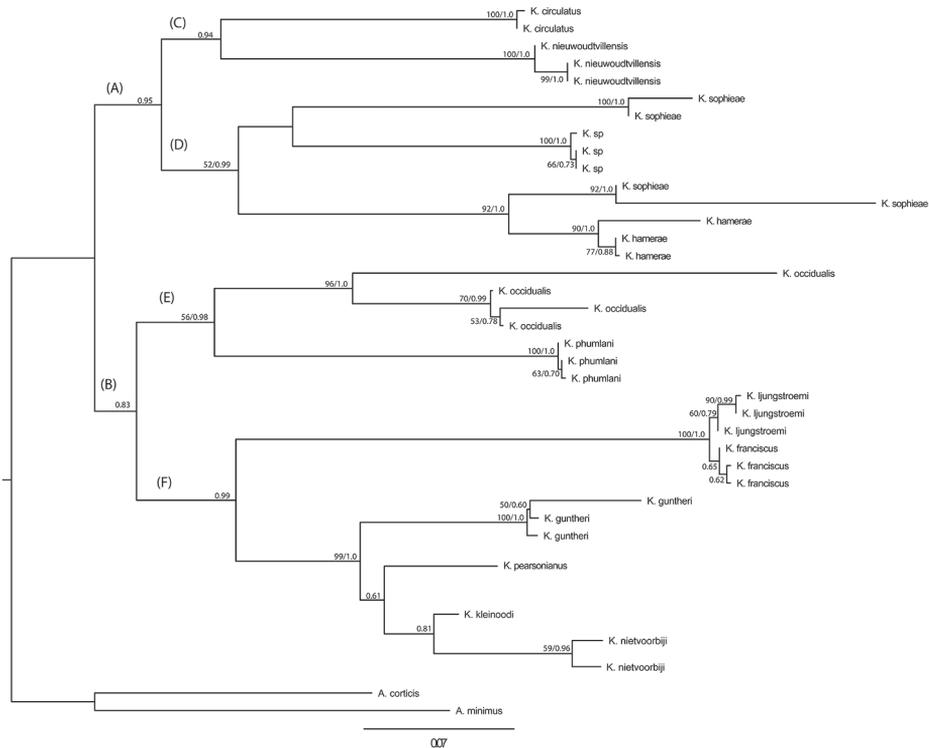
**Figure 1.** COI gene phylogram showing relationships amongst *Kazimierzus* species. Numbers above nodes are bootstrap support/posterior probabilities from Maximum Likelihood and Bayesian analyses. Letters A–F represents different clades.

Table 2. Pairwise p genetic distances (%) between the investigated *Kazimierzus* species/lineages.

Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1. <i>K. nietvoorbiji</i>																
2. <i>K. phumlani</i>	21.1															
3. <i>K. hamerae</i>	22.3	20.1														
4. <i>K. sophieae</i>	25.4	25.5	16.9													
5. <i>K. sp</i>	20.3	18.3	17.5	23.0												
6. <i>K. sophieae</i>	23.1	20.1	21.5	25.5	19.0											
7. <i>K. nieuwooudtvillensis</i>	18.8	20.7	19.9	25.5	17.4	22.6										
8. <i>K. circulatus</i>	19.0	17.5	18.5	23.8	17.9	19.5	16.6									
9. <i>K. occidualis</i>	18.7	15.5	20.9	25.2	18.2	19.6	17.7	15.8								
10. <i>K. occidualis</i>	19.0	16.9	22.2	26.3	19.5	20.9	19.0	17.2	3.0							
11. <i>K. occidualis</i>	21.5	21.9	23.1	28.5	21.5	24.2	20.4	19.8	15.5	16.1						
12. <i>K. franciscus</i>	19.9	19.9	22.0	26.8	19.3	21.7	19.1	19.9	18.7	19.8	23.6					
13. <i>K. ljunstroemi</i>	19.9	20.4	22.2	27.3	18.8	21.9	19.3	19.8	18.7	19.6	23.4	1.1				
14. <i>K. guntheri</i>	13.6	21.1	21.1	26.6	20.7	23.1	19.3	20.4	19.5	18.7	19.3	21.4	21.5			
15. <i>K. kleinoodi</i>	13.2	21.4	23.9	28.5	21.9	23.8	20.9	22.5	19.5	20.7	22.6	22.6	22.8	15.0		
16. <i>K. pearsonianus</i>	14.0	22.6	23.8	27.3	23.4	25.4	22.2	22.0	21.7	22.3	23.6	22.2	22.0	15.0	15.8	

with *Kazimierzus hamerae* (clade D). The two species in clade C, *K. circulatus* and *K. nieuwooudtvillensis*, are sister taxa. *Kazimierzus phumlani* and *K. occidualis* are sister taxa comprising clade E. Clade F has six species including a strongly-supported sub-clade, composed of *K. franciscus* and *K. ljunstroemi*. The other species (*K. guntheri*, *pearsonianus*, *kleinoodi* and *nietvoorbiji*) form the other sub-clade.

Discussion

Morphological examination revealed eleven currently known species (*circulatus*, *franciscus*, *guntheri*, *hamerae*, *kleinoodi*, *nietvoorbiji*, *nieuwooudtvillensis*, *occidualis*, *pearsonianus*, *phumlani* and *sophieae*). However, the current phylogenetic analysis resulted in additional lineages that suggest cryptic diversity (Fig. 1) and a new species, *K. sp*. Most lineages had strong support but weak branch support was noted in deeper nodes with some clades having bootstrap values less than 50%, hence not annotated on the tree. Genetic distances also support the relationships observed in the phylogenetic tree. Genetic distances between species were all greater than 13% and generally much greater than that, consistent with the recommendations of Hebert et al. (2003), Decaëns et al. (2013) and Huang et al. (2007) for separation of species by genetic distance using the CO1 barcode (Table 2).

Kazimierzus hamerae is similar in appearance to *K. sophieae*, but molecular data confirmed that they are separate species. The phylogenetic tree (Fig. 1), however, highlights, unexpectedly, that *K. sophieae* specimens that were collected from two localities, one in Clanwilliam and the other in van Rhyn's Pass, fall into two clades. The two clades may be because *K. sophieae* is polyphyletic or there is no morphological divergence of these taxa, but are different species genetically (25.5% divergence). The type locality of *K. sophieae* is Nieuwooudville, which is close to van Rhyn's Pass. It is likely that the

specimens of *K. sophieae* from van Rhyn's Pass that are in the same clade as *K. hamerae* are the real *K. sophieae*. The specimens of *K. sophieae* from Clanwilliam that are in the same clade as *K. sp.* may possibly be an undescribed species; however, a comparison of sequences of *K. sophieae* specimens and those from the type locality, Nieuwoudtville is necessary. It seems that morphological evolution is not rapid in this complex of taxa and there is presence of cryptic diversity. Clade D has poor ML support (52%) but BI support is high (0.99). The low ML support may suggest that the present data are not sufficient to resolve this polytomy fully.

Cryptic diversity was observed in *K. occidualis*. Finding cryptic diversity is common in earthworms, Novo et al. (2010) reporting five cryptic species within the complex of *Hormogaster elisae*. Richard et al. (2010) reported cryptic species in *Lumbricus terrestris* and Pérez-Losada et al. (2009) found cryptic diversity within *Aporrectodea caliginosa* species complex. The observations support the view that diagnosis, based on morphology, only underestimates taxonomic diversity.

The specimens of *Kazimierzus franciscus* and *Kazimierzus ljunstroemi* were collected in two neighbouring forests, Duiwelbos and Koloniesbos in Marloth Nature Reserve, Swellendam. Duiwelbos is the type locality of *K. franciscus*, whilst the type locality for *K. ljunstroemi* is Great Winterhoek, Tulbagh District. Therefore, it is possible that all specimens belong to *K. franciscus*, but it would be of benefit to collect specimens from Winterhoek and compare them with the ones collected from Marloth Nature Reserve.

Although analysis of other conserved genomic regions in both mtDNA and nuclear DNA in the future would benefit the study of *Kazimierzus* species, the phylogenetic analysis of COI recovered several well-supported phylogenetic relationships, some of which are congruent with existing classification.

Acknowledgements

The KwaZulu-Natal Museum and the University of KwaZulu-Natal are acknowledged for their continued support given for the advancement of the study of earthworms in Southern Africa. The KZN Museum library staff is acknowledged for all their help. The research on the South African megadrile is financially supported in part by the National Research Foundation, South Africa, (Grant numbers:113989, 104846, 114024). Finally, we acknowledge comments of the referees, Samuel James, Csaba Csuzdi, Gabriela Cervantes and the fourth anonymous person.

References

- Akaike H (1973) Information theory and an extension of the maximum likelihood principle. In: Petrov BN, Csaki F (Eds) Second International Symposium on Information Theory. Akademiai Kiado, Budapest (Hungary), 267–281.

- Bantaowong U, Chanabun R, Tongkerd P, Sutcharit C, James SW, Panha S (2011) New earthworm species of the genus *Amyntas* Kinberg, 1867 from Thailand (Clitellata, Oligochaeta, Megascolecidae). *ZooKeys* 90: 35–62. <https://doi.org/10.3897/zookeys.90.1121>
- Blakemore RJ (2013a) *Megascolex (Perichaeta) diffringens* Baird, 1869 and *Pheretima pingi* Stephenson, 1925 types compared to the *Amyntas corticis* (Kinberg, 1867) and *A. carnosus* (Goto & Hatai, 1899) species-groups (Oligochaeta: Megadrilacea: Megascolecidae). *Journal of Species Research* 2(2): 99–126. <https://doi.org/10.12651/JSR.2013.2.2.099>
- Blakemore RJ (2013b) Survey of Busan Oligochaeta earthworms supported by DNA barcodes. *Journal of Species Research* 2(2): 127–144. <https://doi.org/10.12651/JSR.2013.2.2.127>
- Blakemore RJ, Kupriyanova EK, Grygier MJ (2010) Neotypification of *Drawida hattamimizu* Hatai, 1930 (Annelida, Oligochaeta, Megadrili, Moniligastridae) as a model linking mtDNA (COI) sequences to an earthworm type, with a response to the ‘Can of Worms’ theory of cryptic species. *ZooKeys* 41: 1–29. <https://doi.org/10.3897/zookeys.41.374>
- Chang C, James S (2011) A critique of earthworm molecular phylogenetics. *Pedobiologia* 54S: S3–S9. <https://doi.org/10.1016/j.pedobi.2011.07.015>
- Chang C, Lin Y, Chen I, Chuang S, Chen J (2007) Taxonomic re-evaluation of the Taiwanese montane earthworm *Amyntas wulinensis* Tsai, Shen & Tsai, 2001 (Oligochaeta: Megascolecidae): polytypic species or species complex? *Organisms, Diversity & Evolution* 7(3): 231–240. <https://doi.org/10.1016/j.ode.2006.06.001>
- Chang CH, Rougerie R, Chen JH (2009) Identifying earthworms through DNA barcodes: Pitfalls and promise. *Pedobiologia* 52(3): 171–180. <https://doi.org/10.1016/j.pedobi.2008.08.002>
- Csuzdi C (2010) A monograph of the Paleotropical Benhamiinae earthworms. (Annelida: Oligochaeta, Acanthodrilidae). In: Csudi Cs, Mahunka S (Eds) *Pedozoologica Hungarica*, Taxonomic, zoogeographic and faunistic studies on soil animals, No 6. Hungarian Natural History Museum, Budapest.
- Csuzdi C, Zicsi A (2003) Earthworms of Hungary (Annelida: Oligochaeta, Lumbricidae). In: Csudi CS, Mahunka S (Eds) *Pedozoologica Hungarica*, Taxonomic, zoogeographic and faunistic studies on soil animals, No 1. Hungarian Natural History Museum, Budapest.
- Darriba D, Taboada GL, Doallo R, Posada D (2012) jModeltest 2: More models, new heuristics and parallel computing. *Nature Methods* 9(8): 772. <https://doi.org/10.1038/nmeth.2109>
- Decaëns T, Porco D, Rougerie R, Brown GG, James SW (2013) Potential of DNA barcoding for earthworm research in taxonomy and ecology. *Applied Soil Ecology* 65: 35–42. <https://doi.org/10.1016/j.apsoil.2013.01.001>
- Domínguez J, Aira M, Breinholt JW, Stojanovic M, James SW, Pérez-Losada M (2015) Underground evolution: New roots for the old tree of lumbricid earthworms. *Molecular Phylogenetics and Evolution* 83: 7–19. <https://doi.org/10.1016/j.ympev.2014.10.024>
- Drummond AJ, Rambaut A (2007) BEAST: Bayesian evolutionary analysis by sampling trees. *BMC Evolutionary Biology* 7(1): 214. <https://doi.org/10.1186/1471-2148-7-214>
- Felsenstein J (2005) PHYLIP (Phylogeny Inference Package) version 3.6. Distributed by the author. Department of Genome Sciences, University of Washington, Seattle.
- Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA Primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3: 294–299.

- Hall TA (1998) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acid Symposium Series* 41: 95–98.
- Hebert PDN, Ratnasingham S, deWaard JR (2003) Barcoding animal life: Cytochrome c oxidase subunit 1 divergences among closely related species. *Proceedings. Biological Sciences* 270(Suppl.): S96–S99. <https://doi.org/10.1098/rsbl.2003.0025>
- Horst R (1893) Earthworms from the Malay Archipelago. In: Weber M (Ed.) *Zoologische Ergebnisse einer Reise in Niederländisch OstIndien*. 3.E.J. Brill, Leiden, 28–77.
- Huang J, Xu Q, Sun ZJ, Tang GL, Su ZY (2007) Identifying earthworms through DNA barcodes. *Pedobiologia* 51(4): 301–309. <https://doi.org/10.1016/j.pedobi.2007.05.003>
- Huelsenbeck JP, Ronquist F (2001) MRBAYES: Bayesian inference of phylogenetic trees. *Bioinformatics (Oxford, England)* 17(8): 754–755. <https://doi.org/10.1093/bioinformatics/17.8.754>
- James SW, Porco D, Decaëns T, Richard B, Rougerie R, Erseus C (2010) DNA barcoding reveals cryptic diversity in *Lumbricus terrestris* L., 1758 (Clitellata): Resurrection of *L. herculeus* (Savigny, 1826). *PLoS One* 5(12): e15629. <https://doi.org/10.1371/journal.pone.0015629>
- Jouquet P, Dauber J, Lagerlöf J, Lavelle P, Lepage M (2006) Soil invertebrates as ecosystem engineers: Intended and accidental effects on soil and feedback loops. *Applied Soil Ecology* 32(2): 153–164. <https://doi.org/10.1016/j.apsoil.2005.07.004>
- Kinberg JGH (1867) *Annulata nova*. Öfersigt af Kongliga VetenskapsAkademiens Förhandlingar 23: 97–103.
- King RA, Tibble AL, Symonson WOC (2008) Opening a can of worms: Unprecedented sympatric cryptic diversity within British lumbricid earthworms. *Molecular Ecology* 17(21): 4684–4698. <https://doi.org/10.1111/j.1365-294X.2008.03931.x>
- Larkin MA, Blackshields G, Brown NP, Chenna R, Mcgettigan PA, McWilliam H, Valentin F, Wallace IM, Wilm A, Lopez R, Thompson JD, Gibson TJ, Higgins DG (2007) Clustal W and Clustal X version 2.0. *Bioinformatics (Oxford, England)* 23(21): 2947–2948. <https://doi.org/10.1093/bioinformatics/btm404>
- Lavelle P (1988) Earthworms and the soil system. *Biology and Fertility of Soils* 6(3): 237–251. <https://doi.org/10.1007/BF00260820>
- Michaelsen W (1913) The Oligochaeta of Natal and Zululand. *Annals of the Natal Museum* 2(4): 397–457.
- Novo M, Almodovar A, Fernandez R, Trigo D, Cosin DJD (2010) Cryptic speciation of homogastrid earthworms revealed by mitochondrial and nuclear data. *Molecular Phylogenetics and Evolution* 56(1): 507–512. <https://doi.org/10.1016/j.ympev.2010.04.010>
- Nxele TC (2012) The megadrile fauna (Annelida: Oligochaeta) of Queen Elizabeth Park, South Africa: species composition and distribution within different vegetation types. *African Invertebrates* 53(2): 543–558. <https://doi.org/10.5733/afin.053.0207>
- Nxele TC, Plisko JD, Mwabvu T, Zishiri OT (2017) Four new earthworm species of *Kazimierzus* Plisko, 2006 (Clitellata, Kazimierzidae). *Zootaxa* 4353(1): 187–194. <https://doi.org/10.11646/zootaxa.4353.1.12>
- Pérez-Losada M, Ricoy M, Marshall JC, Domínguez J (2009) Phylogenetic assessment of the earthworm *Aporrectodea caliginosa* species complex (Oligochaeta: Lumbricidae) based

- on mitochondrial and nuclear DNA sequences. *Molecular Phylogenetics and Evolution* 52(2): 293–302. <https://doi.org/10.1016/j.ympev.2009.04.003>
- Pickford GE (1975) Contributions to a study of South African Microchaetinae (Annelida: Oligochaeta). *Transactions of the Connecticut Academy of Arts and Sciences* 46: 13–76.
- Plisko JD (1996) Six new earthworm species of the southern African genus *Proandricus* Plisko, 1992 (Oligochaeta: Microchaetidae). *Annals of the Natal Museum* 37: 295–307.
- Plisko JD (1998) New and little-known species of *Microchaetus* Rapp, 1849, with a key to all species and notes on the significance of certain morphological features (Oligochaeta: Microchaetidae). *Annals of the Natal Museum* 39: 249–300.
- Plisko JD (2002) Three new earthworm species of *Microchaetus* Rapp, 1849, and new data on two earlier known species of this genus (Oligochaeta: Microchaetidae). *African Invertebrates* 43: 205–214.
- Plisko JD (2013) A new family Tritogeniidae for the genera *Tritogenia* and *Michalakus*, earlier accredited to the composite Microchaetidae (Annelida: Oligochaeta). *African Invertebrates* 54(1): 69–92. <https://doi.org/10.5733/afin.054.0107>
- Plisko JD, Zicsi A (1991) Über neue *Tritogenia*-Arten aus Süd-Afrika (Oligochaeta: Microchaetidae). *Mitteilungen aus dem Naturhistorischen Museum in Hamburg* 88: 111–123.
- Rambaut A (2009). Fig Tree. <http://tree.bio.ed.ac.uk/software/figtree/>
- Richard B, Decaëns T, Rougerie R, James SW, Porco D, Hebert PD (2010) Re-integrating earthworm juveniles into soil biodiversity studies: Species identification through DNA barcoding. *Molecular Ecology Resources* 10(4): 606–614. <https://doi.org/10.1111/j.1755-0998.2009.02822.x>
- Rougerie R, Decaëns T, Deharveng L, Porco D, James SW, Chang C, Richard D, Potapov M, Suhardjono Y, Hebert PDN (2009) DNA barcodes for soil animal taxonomy. *Pesquisa Agropecuária Brasileira* 44(8): 789–802. <https://doi.org/10.1590/S0100-204X2009000800002>
- Scotland RW, Olmstead RG, Bennet JR (2003) Phylogeny reconstruction: The role of morphology. *Systematic Biology* 52(4): 539–548. <https://doi.org/10.1080/10635150309309>
- Stoeckle MY, Hebert PDN (2008) Barcode of Life: DNA tags help classify animals. *Scientific American* 298: 39–43.
- Tamura K, Stecher G, Peterson D, Filipksi A, Kumar S (2013) MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. *Molecular Biology and Evolution* 30(12): 2725–2729. <https://doi.org/10.1093/molbev/mst197>
- Valentini A, Pompanon F, Taberlet P (2008) DNA barcoding for ecologists. *Trends in Ecology & Evolution* 24(2): 110–117. <https://doi.org/10.1016/j.tree.2008.09.011>
- Zwickl DJ (2011) Garli. <http://www.nescent.org/wg/garli>

Description of a new genus and three new species of the family Palpimanidae (Arachnida, Araneae) from Kenya

Ambata D. Oketch^{1,2,3}, Sergei Zonstein^{4,5}, Esther N. Kioko², Shuqiang Li^{1,3}

1 Institute of Zoology, Chinese Academy of Sciences, Beijing 100101, China **2** National Museums of Kenya, Museum Hill, P.O. Box 40658-00100, Nairobi, Kenya **3** University of Chinese Academy of Sciences, Beijing 100049, China **4** Tel Aviv University, Steinhardt Museum of Natural History, Tel Aviv 69978, Israel **5** Department of Zoology and Centre for Invasion Biology, University of Venda, Thohoyandou, 0950, South Africa

Corresponding author: Shuqiang Li (lisq@ioz.ac.cn)

Academic editor: Galina N. Azarkina | Received 7 May 2020 | Accepted 6 July 2020 | Published 28 July 2020

<http://zoobank.org/E63F61C8-1E75-48C7-ABDB-242FC9B0A0C4>

Citation: Oketch AD, Zonstein S, Kioko EN, Li S (2020) Description of a new genus and three new species of the family Palpimanidae (Arachnida, Araneae) from Kenya. *African Invertebrates* 61(2): 93–106. <https://doi.org/10.3897/AfrInvertebr.61.54004>

Abstract

A new genus and three new species of the spider family Palpimanidae Thorell, 1870 from Kenya are described. *Sceliscelis* Oketch & Li, **gen. nov.** is close to *Sarascelis* Simon, 1887 and *Scelidocteus* Simon, 1907 but differs in the structure of the male palp. The new species reported are *Sceliscelis marshi* Oketch & Li, **sp. nov.**, *Scelidocteus taitave* Oketch & Li, **sp. nov.**, and *Hybosida machondogo* Oketch & Li, **sp. nov.** *Sceliscelis marshi* **sp. nov.** is described from males and females collected from Tsavo in south Kenya. *Scelidocteus taitave* **sp. nov.** can be distinguished from other congeners, as well as from the morphologically similar *Scelidomachus socotranus* Pocock, 1899, by the shape of the palpal “conductor”. *Hybosida machondogo* **sp. nov.** differs from other six-eyed *Hybosida* spp. by possessing rudimentary posterior median eyes.

Keywords

Afrotropical, Chediminae, genus, species, taxonomy

Introduction

Palpimanidae Thorell, 1870 is a small family of araneophagous spiders recognized by the coriaceous carapace, haplogyne genitalia, two spinnerets and especially by the first pair of legs with dorsally enlarged femora and well-developed prolateral scopula on the tibia, metatarsus and tarsus. A total of 152 species in 18 genera are distributed in tropical and subtropical zones worldwide, absent only in the Nearctic and Australia (Li 2020). Within Africa, the family is known to include 56 species belonging to 12 genera (WSC 2020), although some of these species are certainly misplaced in the family (Zonstein and Marusik 2013; Zonstein et al. 2018).

More than 800 spider species and subspecies belonging to 55 families are known from Kenya (WSC 2020). However, Palpimanidae have not been thoroughly studied. Up to now, only three species have been described: *Scelidocteus incisus* Tullgren, 1910, *Hybosida lesserti* Berland, 1920 and *H. scabra* Simon & Fage, 1922. In this paper, one new genus and three new species of palpimanid spiders found during the examination of spiders collected in various protected sites in Kenya are described.

Materials and methods

All spiders were preserved in 95% alcohol. Specimens were examined and measured using a LEICA M205C stereomicroscope. Images were captured using an Olympus C7070 wide zoom digital camera mounted on an Olympus SZX12 dissecting microscope or on an Olympus BX51 compound microscope. Male and female copulatory organs were dissected, and the specimens were then photographed on glass slides fitted with cotton wool to ensure a white, uniform background. Digital images were prepared using Helicon Focus version 6.0 image stacking software and then edited using Adobe Photoshop CS4 version 11.0.0.

Leg lengths and palp segments are given in the following sequence: total (femur, patella, tibia, metatarsus, tarsus). Internal structures of the genitalia were illustrated after being cleared in lactic acid, then placed in alcohol for a few minutes. Measurements are given in millimeters. Holotypes and paratypes are deposited in the National Museums of Kenya (NMK), Nairobi, Kenya.

Abbreviations. AER – anterior eye row, ALE – anterior lateral eyes, am – accompanying membrane, AME – anterior median eyes, co – “conductor”, ch – receptive chamber, cy – cymbium, ds – dorsal portion of scutum, em – embolus, Ft – fine threads, Gr – grape-shaped glands, hle – hook-like extension, MOQ – median ocular quadrangle, pe – palm-like embolic extension, PER – posterior eye row, PLE – posterior lateral eyes, PME – posterior median eyes, Re – rigid extensions of posterior wall of epigastric fold, rt – retrolateral thorn, sco – scopula, Sr – sac-like receptacle.

Taxonomy

Family Palpimanidae Thorell, 1870

Subfamily Chediminae Simon, 1893

Sceliscelis Oketch & Li, gen. nov.

<http://zoobank.org/9F855192-68DE-4B2C-8324-804D599234C4>

Type species. *Sceliscelis marshi* sp. nov.

Etymology. This genus possesses some characteristics that are also found in *Sarascelis* Simon, 1887 and *Scelidocteus* Simon, 1887, such as the dentate segments of leg I (which include at least the dorsal surface of the coxa and basal part of the femur) and the spiral embolus. The name is a combination of “Sceli” from *Scelidocteus* and “Scelis” from *Sarascelis*. The gender is feminine.

Diagnosis. The new genus closely resembles *Sarascelis* in general appearance by having much larger AMEs (Fig. 1A; also see Jézéquel 1964, fig. 5a–c, Zonstein and Marusik 2013; figs 5, 6). It can, however, be distinguished from *Sarascelis*, *Scelidocteus* and the close ally *Steriphopus* Simon, 1887 by (1) having a greatly extended bulb (vs. the more or less bulky palpal structures of *Sarascelis* and *Steriphopus*) (2) the large AMEs (*Steriphopus* has considerably smaller AMEs) (3) the shape of the thoracic fovea, which is an elongated Ω -shape in *Sceliscelis* (vs. a longitudinal slit or anchor-like in *Sarascelis* and *Scelidocteus*) (4) the presence of a hook-like extension at the distal end of a sword-like “conductor” (vs. extension absent in other members of the family) (5) the poorly developed scopula on metatarsus and tarsus I (6) the rugose carapace in *Sceliscelis* gen. n. (vs. smooth or finely rugose in *Sarascelis*, *Scelidocteus* and *Steriphopus*) (see Zonstein and Marusik 2013). Females of *Sceliscelis* can be distinguished from those of *Sarascelis* and *Scelidocteus* by the wavy, rigid extension of the genital area and by a pair of divergent sac-like receptacles.

Description. Medium-sized. Carapace dome shaped, coarsely granulated and hairless, medially elevated, fovea longitudinal, open posteriorly (elongated Ω -shape). Eight eyes, MOQ trapezoidal; AME largest, about 3 times as large as PME; ALE, PLE and PME subequal to each other. AER straight. Labium triangular, notched, longer than wide but broader at base. Chelicerae directed ventrally, 2 times longer than clypeus, granulate. Endites trapezoidal, about 1.5 times longer than wide. Abdomen clay yellow, oval and covered with short, gray setae in both sexes. Femur of male palp slender, longer than wide, patella shorter than “conductor”. Cymbium with setae, pointed at tip and indented (prolateral view). Embolus spiral, with accompanying membrane and a silvery, palm-like embolic extension. “Conductor” sword-shaped, longer than wide, slightly longer than embolus. Endogyne wavy anteriorly, forming a rigid epigastric wall.

Composition. This genus currently includes only the type species.

***Sceliscelis marshi* Oketch & Li, sp. nov.**

<http://zoobank.org/01D83454-CC28-45A1-91B5-87D1424A3C6A>

Figs 1, 2

Type material. *Holotype* ♂, Kenya, Taita Taveta County, near Mtito Andei, Tsavo West National Park, Rhino Sanctuary, 38°19.96'E, 03°1.230'S, 730 m, 28.VII.2016, sifting leaf litter, Grace Kioko leg. Paratype 1♀, same data as holotype.

Etymology. This species is named after Mr. Brian Marsh OBE, the founder of the Marsh Christian Trust Awards for Ecologists in Africa, which the third author was awarded in 2019; noun (name) in genitive case.

Diagnosis. See diagnosis of the genus.

Description. Male. Habitus as in Fig. 1A–C. Total body length 3.83. Color in alcohol: carapace, sternum, chelicerae, labium, leg I coxa and epigastric scutum uniformly reddish orange to reddish brown, entire palp and most of leg I slightly paler reddish orange, legs II–IV with yellowish femur and patella and darkened distal segments. Carapace 2.13 long, 1.75 wide. Fovea longitudinal, open posteriorly. Eye sizes and interdistances: AME 0.13, PME 0.05, PLE 0.06, ALE 0.06, AME-AME 0.09, PME-PME 0.19, AME-PME 0.09, AME-ALE 0.06, PME-PL 0.13, ALE-PL < 0.01. AER almost straight. Labium triangular 0.25 long, 2 times the length of labium notch, clypeus 2 times shorter than length of chelicerae. Sternum 1.00 long, 0.78 wide, granulate. Endites trapezoidal, pale yellow anteriorly. Abdomen as in genus description. Palp: Patella sub-globular, as long as 1/3 length of tibia. Tibia 0.35 long, 0.31 wide. Cymbium about 2 times longer than wide.

Female. Cephalothorax in dorsal and ventral aspects as in Fig. 2A, B, respectively. Color in alcohol: carapace, sternum, chelicerae, labium, leg I coxa and epigastric scutum and abdomen same as in male. Body length 5.1. Carapace 3.1 long, 2.2 wide, fairly granulate. Eye sizes and interdistances: AME 0.15, PME 0.08, PLE 0.06, ALE 0.06, AME-AME 0.09, PME-PME 0.19, AME-ALE 0.06, PME-PL 0.13, ALE-PL < 0.01. Notch is approximately 1/2 the length of labium which is 0.24 long. Clypeus/chelicera length ratio 1:1. Epigastric scutum sclerotized, book lung operculum visible, lateral sclerites present, posterior edge strongly undulate. Vulva not visible through integument, distant from epigastric fold, with stalked grape-shaped glands. Receptacles oval, mounted on a pair of irregular receptive chambers, separated (Fig. 2C, D). Leg and palp measurements in Table 1.

Distribution. This species is currently known only from the type locality.

Table 1. Type male (female) leg and palp measurements.

	Femur	Patella	Tibia	Metatarsus	Tarsus	Total
Palp	0.91 (0.85)	0.15 (0.13)	0.35 (0.37)	–	0.50 (0.42)	1.91 (1.77)
I	0.85 (0.94)	0.94 (0.80)	0.70 (0.75)	0.30 (0.25)	0.31 (0.22)	3.10 (2.96)
II	1.00 (0.69)	0.50 (0.50)	0.65 (0.63)	0.50 (0.38)	0.32 (0.31)	2.97 (2.51)
III	0.63 (0.52)	0.50 (0.41)	0.50 (0.50)	0.35 (0.47)	0.24 (0.22)	2.22 (2.51)
IV	1.25 (0.78)	0.30 (0.50)	0.88 (0.81)	0.75 (0.56)	0.25 (0.25)	3.31 (2.90)

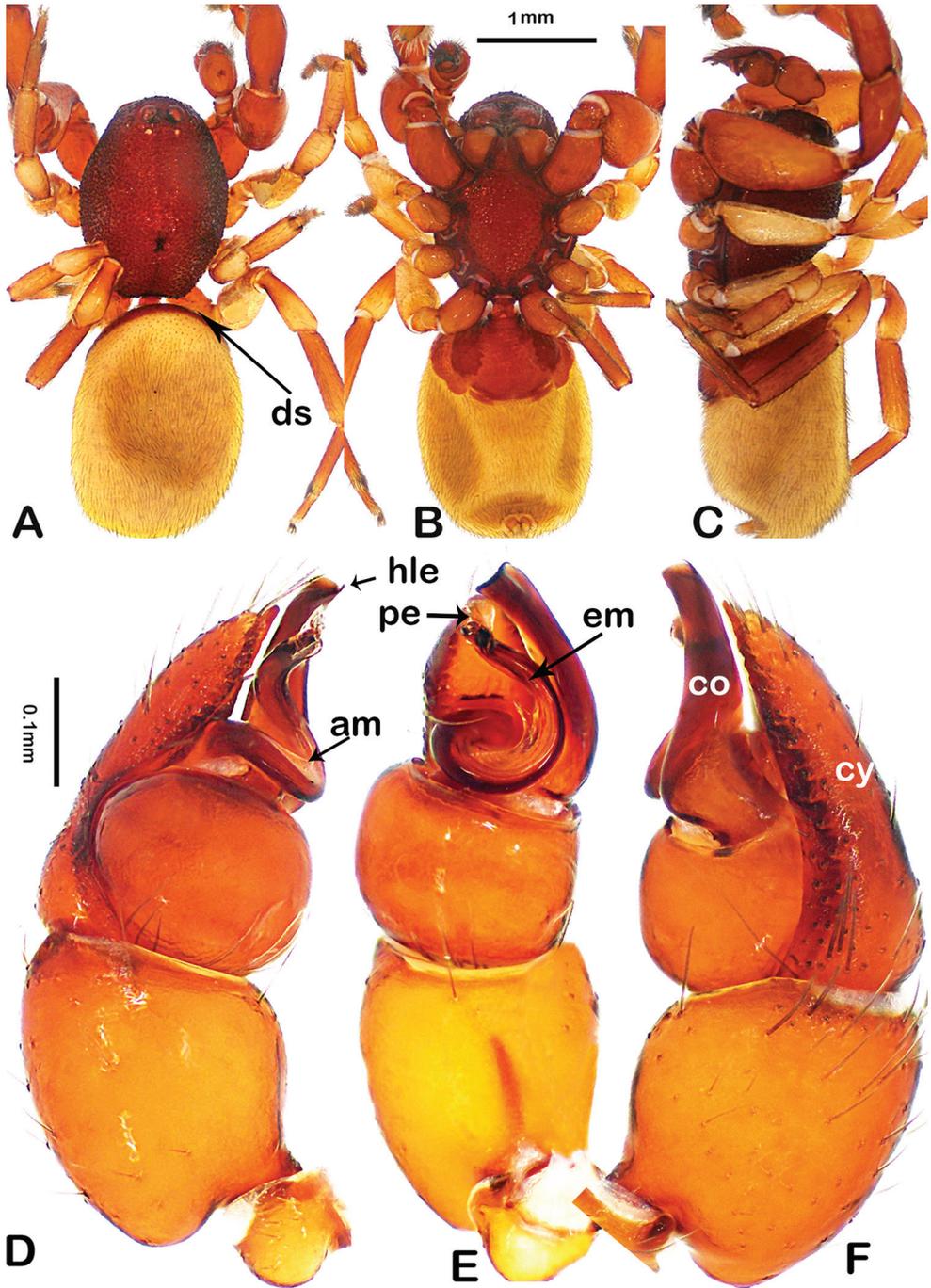


Figure 1. *Sceliscelis marshi* sp. nov., male holotype **A–C** habitus in dorsal, ventral and lateral aspects **D–F** palp: **D** prolateral **E** ventral and **F** retrolateral aspects. Abbreviations: am – accompanying membrane, co – “conductor,” cy – cymbium, ds – dorsal portion of scutum, em – embolus, hle – hook-like extension, pe – palm-like embolic extension. Scale bars: 1 mm (**A–C**), 0.1 mm (**D–F**).

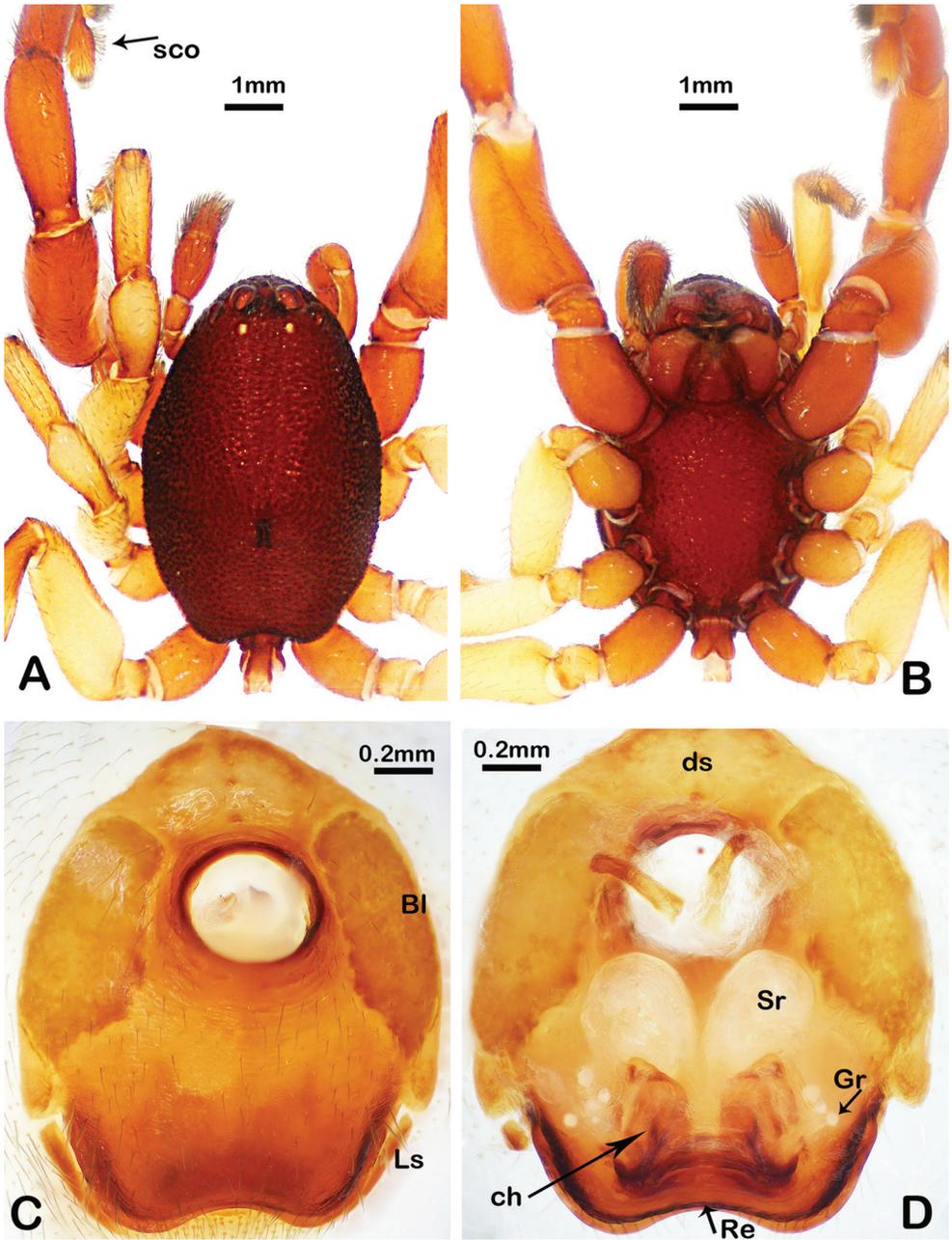


Figure 2. *Sceliscelis marshi* sp. nov., female paratype. Cephalothorax **A** dorsal and **B** ventral aspects **C**, **D** endogyne, ventral and dorsal aspects, respectively. Abbreviations: Bl – book lung operculum, ch – receptive chamber, ds – dorsal portion of scutum, Gr – grape-shaped glands, Ls – lateral sclerite, Re – rigid extension of posterior wall of epigastric fold, sco – scopula, Sr – sac like receptacle. Scale bars: 1 mm (**A**, **B**), 0.2 mm (**C**, **D**).

Genus *Scelidocteus* Simon, 1907

Type species. *Scelidocteus pachypus* Simon, 1907, by subsequent designation.

Notes. All seven described species of this genus are endemic to Africa, the majority of them in West and Central African countries. *Scelidocteus taitave* sp. nov. is the first species to be recorded from Kenya and East Africa (WSC 2020). *Scelidocteus* resembles *Scelidomachus* and *Chedima* Simon, 1873 in general appearance, especially in having an anteriorly blunt (not rounded) carapace with pronounced, obtuse anterolateral corners, but can be distinguished from *Chedima* by the uniformly colored abdomen (vs. spotted) with short gray setae and the eye shape and arrangement (MOQ in *Chedima* is longer than wide, whereas it is subquadrate in *Scelidocteus*).

Scelidocteus taitave Oketch & Li, sp. nov.

<http://zoobank.org/DE20171E-AD59-4DFF-901B-9B139F91DC5F>

Figs 3, 4

Type material. *Holotype* ♂, Kenya, Taita Taveta County, Taita Hills, Mbololo Forest, 30°26.85'E, 03°20.36'S, 1631 m, 2.VIII.2018, Ambata Oketch leg. *Paratypes* 1 ♂, 2 ♀, same data as holotype.

Etymology. The specific name is a combination of Taita and Taveta, the county from which the specimens were collected; noun in apposition.

Diagnosis. Males of this species differ from those of other members of *Scelidocteus* by the uniquely spiral-shaped embolus (Fig. 3C–E cf. Pocock 1903, fig. 3; de Lessert 1930, fig. 4; Jézéquel 1964, figs 2A, B, 4a, b). Females of *S. taitave* sp. nov. differ from those of other species by the structure of the endogyne; the orientation of sac-like receptacles (touching medially, with a space below them) mounted on oval, membranous receptive chambers and the outline of a rigid extension of the posterior wall of the epigastric fold (Fig. 4C, D; Jézéquel 1964, figs 1, 3).

Description. Male. Fig. 3A, B. Total body length 3.90. Color in alcohol: carapace and chelicerae orange-red, legs I, endites, sternum and labium orange, legs II–IV lighter, yellowish. Carapace 1.88 long, 1.50 wide at leg II, oval in dorsal view, finely granulate, cephalic part slightly elevated behind eye area. Thoracic fovea a longitudinal, deep slit, approximately 0.10 long. Eye sizes and interdistances: AME 0.13, ALE 0.06, PLE 0.06, PME 0.06, AME-AME 0.05, AME-ALE 0.08, PME-PME 0.09, AME-PLA 0.04, ALE-PLA <0.01, PLE-PME 0.24. AER slightly procurved, almost straight, PER strongly recurved. Chelicerae flattened anteriorly towards the fangs, 1.60 long, cheliceral furrow with several peg-like teeth. Stridulatory mound absent. Clypeus approximately 2 times shorter than length of chelicerae. Sternum 1.09 long, 0.86 wide at leg II; shield shaped, rebordered, finely granulate. Endites almost D-shaped, labium notch 0.13 long, about a quarter of labium length. Leg I: coxa, patella and tibia pos-

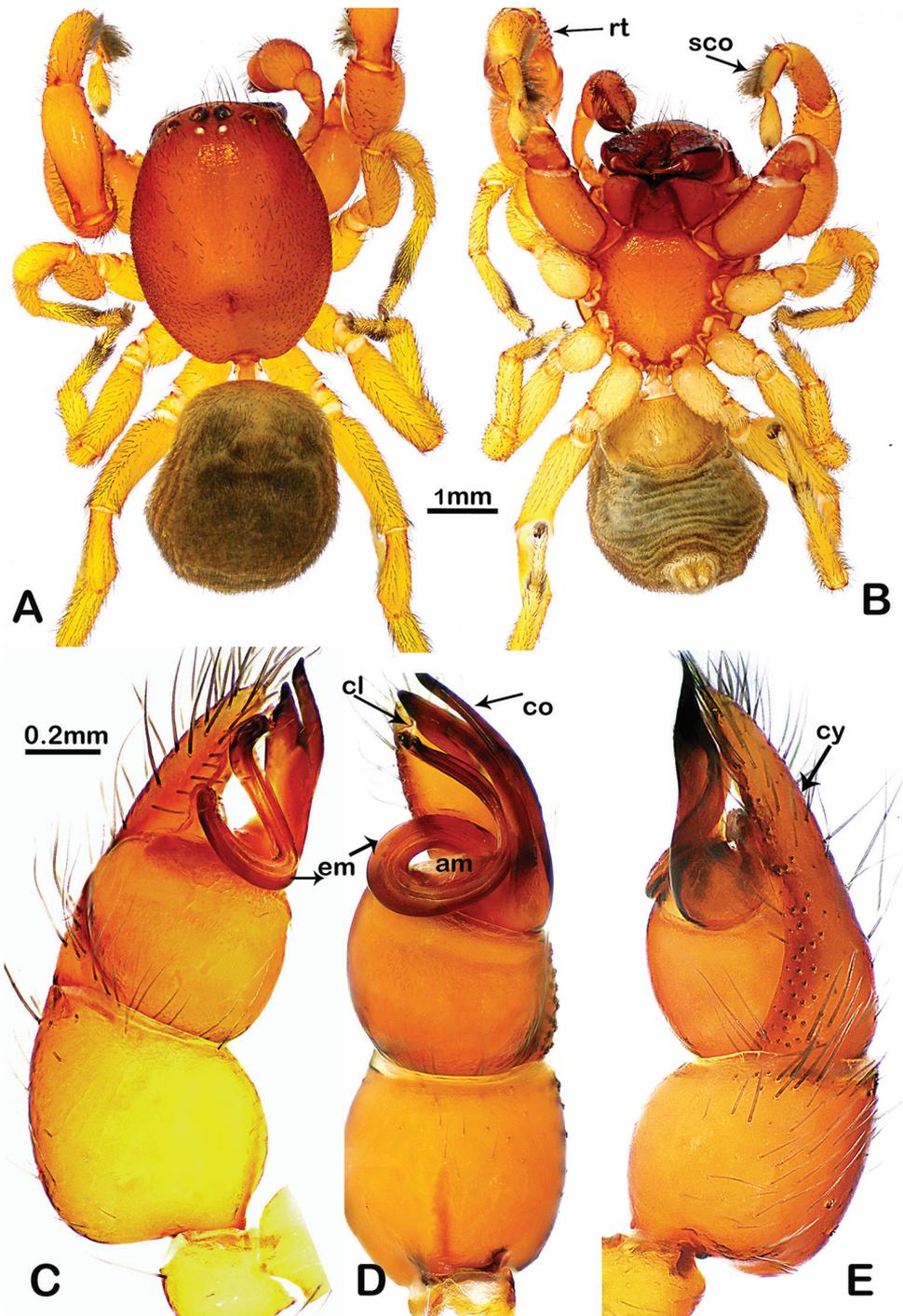


Figure 3. *Scelidocteus taitave* sp. nov., male holotype **A, B** dorsal and ventral habitus respectively **C-E** palp: **C** prolateral **D** ventral **E** retrolateral aspects. Abbreviations: am – accompanying membrane, cl – claw like extension, co – “conductor”, em – embolus, cy – cymbium, rt – retrolateral thorns, sco – scopula. Scale bars: 1mm (**A, B**), 0.2mm (**C-E**).

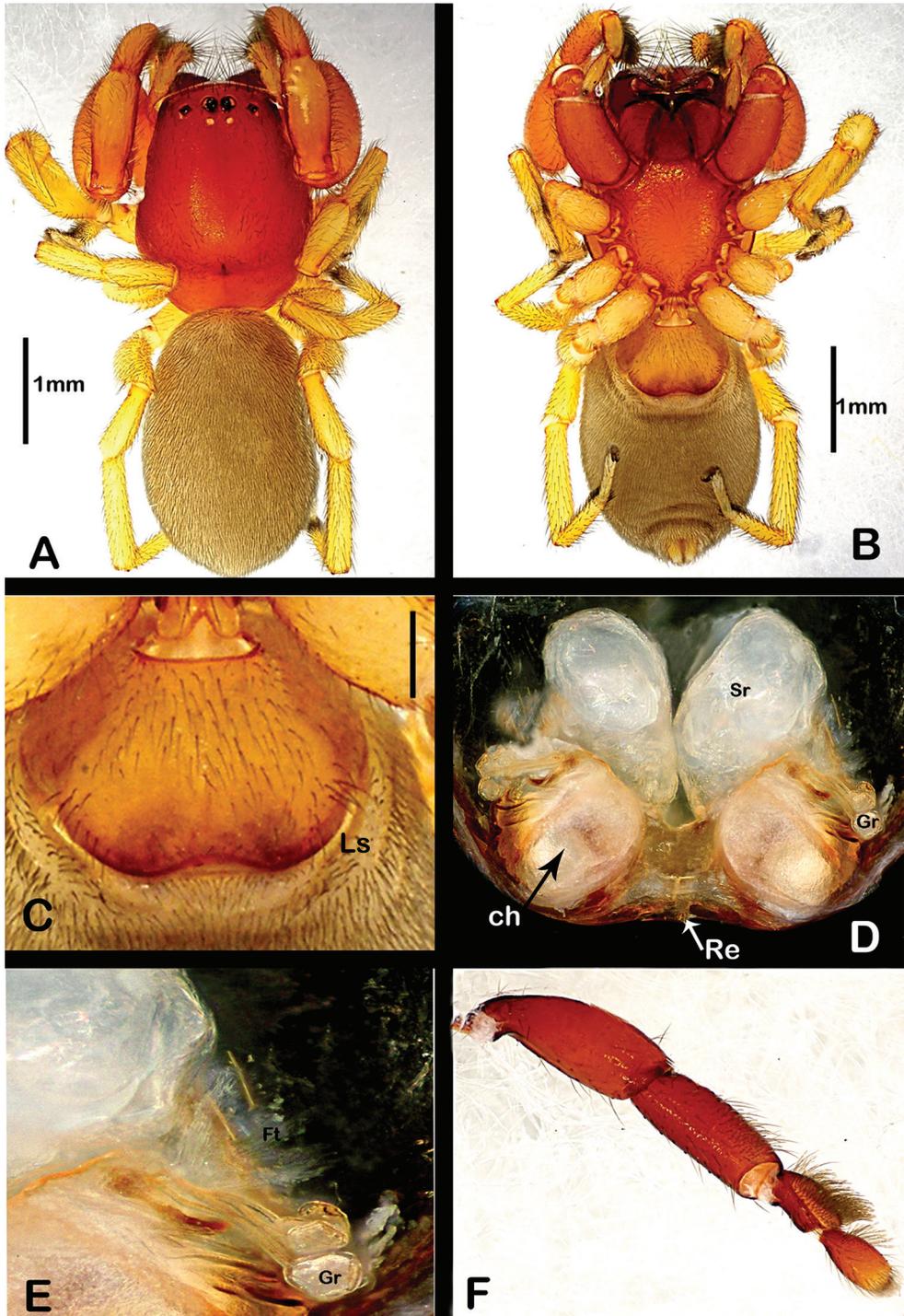


Figure 4. *Scelidocteus taitave* sp. nov., female paratype **A, B** habitus **C** endogyne ventral **D** same, ventral **E** same, enlarged **F** leg I. Abbreviations: ch – receptive chamber, Ft – fine threads, Gr – grape-shaped glands, Re – rigid extensions of posterior wall of epigastric fold, Sr – sac like receptacle. Scale bars: 1mm (**A, B**).

Table 2. Type male (female) palp and leg measurements.

	Femur	Patella	Tibia	Metatarsus	Tarsus	Total
Palp	0.66 (0.61)	0.24 (0.21)	0.38 (0.37)	–	0.35 (0.39)	1.63 (1.58)
I	1.38 (1.41)	0.65 (0.81)	1.00 (1.09)	0.38 (0.38)	0.40 (0.38)	3.81 (4.07)
II	0.88 (0.85)	0.69 (0.70)	0.80 (0.80)	0.50 (0.41)	0.40 (0.28)	3.27 (3.04)
III	0.90 (1.00)	0.60 (0.75)	0.60 (0.69)	0.60 (0.74)	0.40 (0.44)	3.10 (3.62)
IV	0.90 (1.38)	0.80 (0.80)	0.90 (1.00)	0.84 (1.00)	0.40 (0.50)	3.84 (4.68)

sess dark, thorn-like outgrowths and well developed prolateral scopula on tibia and metatarsus. Tarsus I with weakly developed scopula. Leg and palp measurements as in Table 2. Abdomen oval with short, grey setae, dorsal portion of epigastric scutum very small, pedicel short, spinnerets short and unsegmented. Palp (Fig. 3C–E): tibia, as long as wide, approximately 2.5 times wider than femur. Cymbium long and thin, tapering distally. “Conductor” bifurcate; embolus long and spiral, obscuring some parts of tegulum. Embolus ends with a bleached, claw-like structure at apex.

Female. General appearance as in Fig. 4A, B. Coloration as in male. Total length 4.88. Carapace 2.19 long, 1.72 wide. Eye sizes and interdistances: AME 0.13, ALE 0.06, PLE 0.06, PME 0.06, AME-AME 0.05, ALE-AME 0.08, ALE-PLE < 0.01, PLE-PME 0.20, PME-PME 0.7. Sternum 1.25 long, 1.00 wide, labium 0.5 long, 0.44 wide at the base, labium notch 1/3 length of labium. Vulva with fine, thread-like structures and 3 pairs of stalked, grape-shaped glands attached to a pair of relatively ovate and membranous receptive chambers.

Distribution. This species is currently known only from the type locality.

Genus *Hybosida* Simon, 1898

Type species. *Hybosida lucida* Simon, 1898, by monotypy.

Diagnosis. All four described species of this genus have six eyes, PMEs are absent (Simon 1898, Platnick 1979, Saaristo 2010). *H. dauban* Platnick, 1979 and *H. lucida* Simon, 1898 are endemic to Seychelles and *H. scabra* and *H. lesserti* are known from East Africa.

Hybosida machondogo Oketch & Li, sp. nov.

<http://zoobank.org/D4F9BC3A-3196-498E-89F0-F0750B28A377>

Figs 5, 6

Type material. *Holotype* ♂, Kenya, Nyeri County, Naro Moru Town, Mount Kenya National Park, Naro Moru Gate, Forest Valley, 37°10.16'E, 00°10.61'S, 2488 m, 25.VII.2017, Grace Kioko leg.

Other material examined. 2 ♂ subadults, Kenya, Uasin Gishu County, Endebbes Town, Mount Elgon National Park, Mutamaiyo Camp Site, 34°43.07'E, 01°4.02'N, 2824 m, 7.VII.2017, Grace Kioko leg.

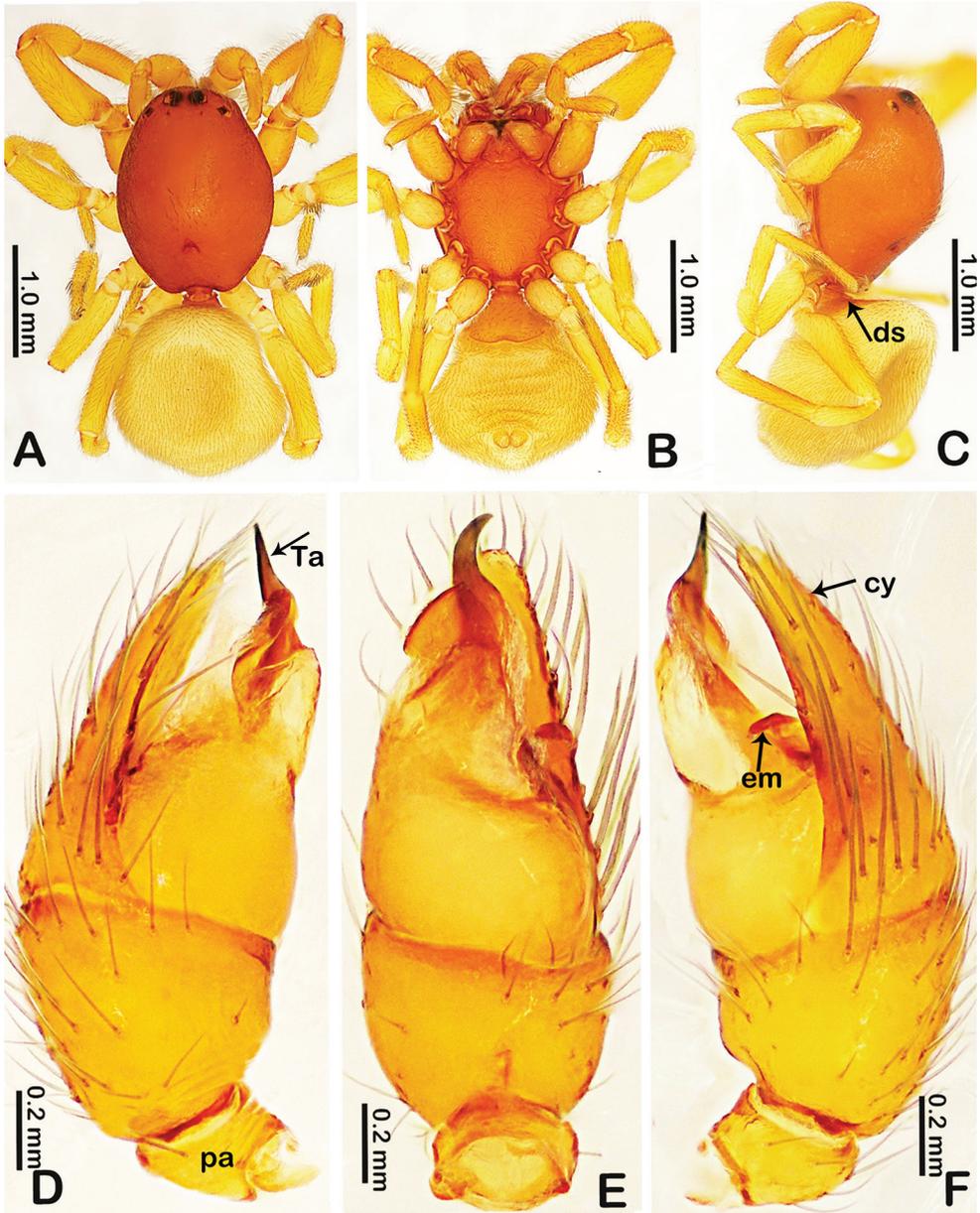


Figure 5. *Hybosida machondogo* sp. nov. male holotype **A–C** habitus: **A** dorsal **B** ventral and **C** prolateral aspects) **D–F** palp: **D** prolateral **E** ventral **F** retrolateral. Scale bars: 1 mm (**A–C**), 0.2 mm (**D–F**). Abbreviations: cy – cymbium, ds – dorsal portion of scutum, em – embolus, pa – patella, Ta – tegula apophysis.

Etymology. The specific name is a combination of two Swahili words “macho”, meaning eyes, and “ndogo”, meaning small, referring to the indiscernible posterior median eyes of this species.

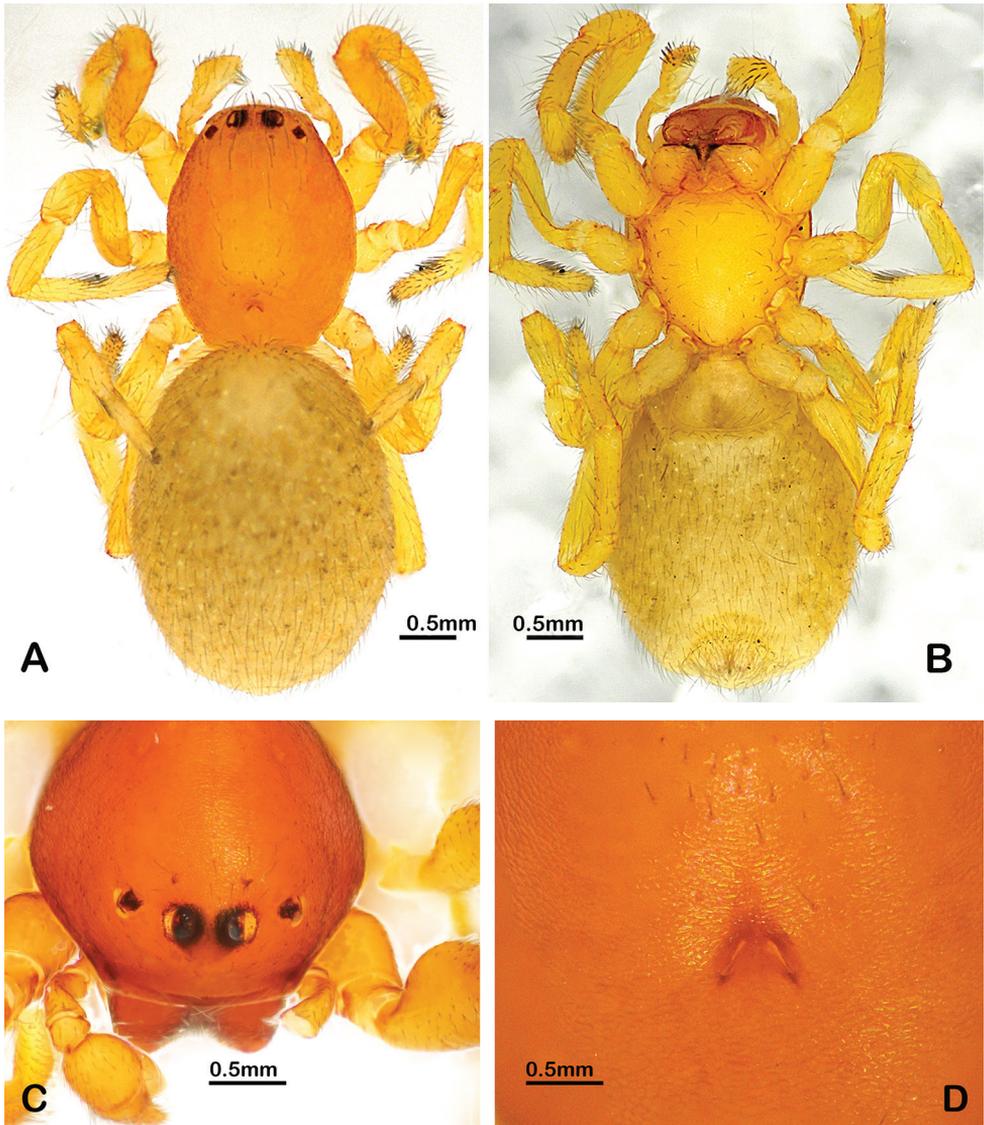


Figure 6. *Hybosida machondogo* sp. nov., subadult male, habitus **A** dorsal view **B** ventral **C** male holotype, ocular area, anterodorsal **D** thoracic fovea, dorsal. Scale bars: 0.5 mm (**A–D**).

Diagnosis. Like other members of Chediminae, *Hybosida machondogo* sp. nov. possess contiguous lateral eyes and accessory structures of the male palp (see Zonstein and Marusik 2013, figs 1–9; Zonstein et al. 2018). It closely resembles *H. lesserti* by having (1) a more compact carapace with the cephalic part narrowed (2) an elevated carapace with a steep posterior slope (3) a strongly convex carapace in lateral view and

(4) poorly developed scopula on both the metatarsi and tarsi of legs I (see Berland 1920, figs 128, 131). However, *H. machondogo* sp. nov. differs from *H. lesserti* and other members of this genus by having (1) 8 eyes instead of 6, (2) a bulb with relatively long, sharp and less curved apophysis (vs. strongly curved in *H. lesserti*).

Description. Male. Fig. 5A–C. Total body length 2.05. Carapace, pedicel and sternum reddish orange, 1.11 long, 0.83 wide at leg II, carapace dome-shaped and covered with white setae at the edges, finely rugose. Thoracic region higher than cephalic region, steeply sloping towards pedicel (Fig. 5C). Fovea crescent-like, bipartite (Figs 5A, 6D), wide distally (sulci diverging). Sternum with long, gray setae. Eyes: AER strongly recurved, PER slightly procurved (Fig. 6C). Eye sizes and interdistances: AME 0.09, PME \leq 0.01, PLE=ALE=0.04, AME-AME 0.05, AME-ALE 0.08, AME-PLE 0.06, PME-PME 0.09. Clypeus about 2 times higher than diameter of AME. Labium deeply notched (smoothly recurved in all described palpimanids). Endites colored as sternum, depressed proximally, covered with gray setae. Legs: yellowish without spines. Femur I enlarged and longer than patella; scopula on tibia, metatarsus and tarsus weakly developed (compared to *Scelidocteus*). Femur II somewhat longer than femora III–IV (Fig. 5C), tarsal claws long and unidentate. Metatarsi II–IV with distal preening comb. Generally, leg integument appears rough. Leg and palp measurements as in Table 3. Abdomen: unsclerotized, uniformly yellowish, oval, narrow towards the pedicel and wide at the middle. Pedicel considerably exposed, lateral and postgastric scutum extensions absent, dorsal portion of scutum present but short. Spinnerets short yellowish and unsegmented.

Palp yellowish, femur longer than tibia, patella about 2 times shorter than femur. Tibia enlarged, almost conical in prolateral view, 1.86 times wider than patella, about 2 times wider than femur. Cymbium about 1.5 times longer than tibia, slightly shorter than apophysis. Tegular region is membranous, apophysis pointed, extends anteriorly. Embolus short, blunt and directed laterally (Fig. 5D–F).

Female. Unknown.

Distribution. This species is currently known only from the type locality.

Notes. This species is peculiar because it is at odds with the initial description of the type species of the genus *Hybosida*, which states that the number of eyes is 6. It does, however, resemble *Hybosida* in the shape of the carapace, the fovea and the general structure of the male palp. It is therefore treated as belonging to this genus.

Table 3. Male palp and leg measurements.

	Femur	Patella	Tibia	Metatarsus	Tarsus	Total
Palp	0.60	0.24	0.38	–	0.29	1.51
I	0.80	0.55	0.45	0.25	0.30	2.35
II	0.60	0.30	0.45	0.35	0.35	2.05
III	0.55	0.30	0.45	0.40	0.30	2.00
IV	0.60	0.30	0.65	0.50	0.35	2.40

Acknowledgements

The manuscript benefited greatly from comments by Galina N. Azarkina, Yuri Marusik and Yanfeng Tong. The English was kindly checked by Sarah Crews (USA). The fieldwork was facilitated by Kenya Wildlife Service (KWS), Kenya Forest Service (KFS), National Environment Management Authority (NEMA) and the National Commission for Science, Technology and Innovation (NACOSTI). The study was financially supported by the National Natural Science Foundation of China to Shuqiang Li (NSFC-31530067).

References

- Berland L (1920) Araneae (2e partie). Voyage de Ch. Alluaud et R. Jeannel en Afrique Orientale (1911–1912): Résultats scientifiques: Arachnida. Paris 4, 95–180.
- Jézéquel JF (1964) Araignées de la savane de Singrobo (Côte d’Ivoire). II. Palpimanidae et Zodariidae. Bulletin du Muséum National d’Histoire Naturelle 36: 326–338.
- Lessert R de (1930) Araignées du Congo recueillies au cours de l’expédition organisée par l’American Museum (1909–1915). Quatrième et dernière partie. Revue Suisse de Zoologie 37: 613–672. <https://doi.org/10.5962/bhl.part.117639>
- Li S (2020) Spider taxonomy for an advanced China. *Zoological Systematics* 45(2): 73–77. <https://doi.org/10.11865/zs.202011>
- Platnick NI (1979) Contributions à l’étude de la faune terrestre des îles granitiques de l’archipel des Séchelles (Mission P.L.G. Benoit – J.J. Van Mol 1972). Araneae: Palpimanidae. *Revue Zoologique Africaine* 93: 461–466.
- Pocock RI (1903) Arachnida. In: Forbes HO (Ed.) *The Natural History of Sokotra and Abd-el-Kuri*. Special Bulletin of the Liverpool Museum, 175–208.
- Saaristo MI (2010) Araneae. In: Gerlach J, Marusik YM (Eds) *Arachnida and Myriapoda of the Seychelles Islands*. Siri Scientific Press, Manchester, 8–306.
- Simon E (1898) Etudes arachnologiques. 29e Mémoire. XLVI. Arachnides recueillis en 1895 par M. le Dr A. Brauer (de l’Université de Marburg) aux îles Séchelles. *Annales de la Société Entomologique de France* 66: 370–388.
- WSC (2020) World Spider Catalog. Version 21.0. Natural History Museum Bern. <https://doi.org/10.24436/2> [accessed on 27.03.2020]
- Zonstein SL, Marusik YM (2013) On *Levymanus*, a remarkable new spider genus from Israel, with notes on the Chediminae (Araneae, Palpimanidae). *ZooKeys* 326: 27–45. <https://doi.org/10.3897/zookeys.326.5344>
- Zonstein SL, Marusik YM, Omelko MM (2018) Redescription of the monotypic genus *Scelidomachus* Pocock, 1899 (Aranei: Palpimanidae) and its type specie. *Arthropoda Selecta* 27(1): 53–56. <https://doi.org/10.15298/arthsel.27.1.07>

Type specimens of Streptaxidae from Henry C. Burnup in the collection of the Museum of New Zealand Te Papa Tongarewa

Rodrigo B. Salvador¹, Jonathan D. Ablett²

1 *Museum of New Zealand Te Papa Tongarewa, 169 Tory Street, Wellington, 6011, New Zealand* **2** *Invertebrates Division, Department of Life Sciences, Natural History Museum, South Kensington, London, SW7 5BD, UK*

Corresponding author: Rodrigo B. Salvador (salvador.rodrigo.b@gmail.com)

Academic editor: I. Muratov | Received 28 August 2020 | Accepted 14 October 2020 | Published 23 October 2020

<http://zoobank.org/76485E37-D7B9-481C-8EFA-73CA9B88DEF0>

Citation: Salvador RB, Ablett JD (2020) Type specimens of Streptaxidae from Henry C. Burnup in the collection of the Museum of New Zealand Te Papa Tongarewa. *African Invertebrates* 61(2): 107–117. <https://doi.org/10.3897/AfrInvertebr.61.58085>

Abstract

A small collection containing thirty-nine lots of South African Streptaxidae land snails is housed in the collection of the Museum of New Zealand Te Papa Tongarewa (NMNZ). This material previously belonged to British/South African malacologist Henry C. Burnup, who either donated it to, or exchanged it with New Zealand-based Swiss malacologist Henry Suter, whose land snail collection was eventually acquired by the NMNZ. The lots contain type specimens of eight taxa (species and subspecies) and are presented herein in the form of an annotated and illustrated catalogue.

Keywords

Gastropoda, hunter snails, South Africa, Stylommatophora

Introduction

A small but important collection of land snails is part of the Mollusca collection of the Museum of New Zealand Te Papa Tongarewa (NMNZ, Wellington, New Zealand), which has come to our attention during ongoing efforts to reorganize the museum's terrestrial and freshwater gastropod holdings (see also Salvador and Breure 2020). This material comprises thirty-nine lots of land snail shells from South Africa belonging to

the family Streptaxidae, commonly known as hunter snails. The shells belonged to the collection of Henry Clifden Burnup and the labels of ten lots indicate that they were part of the material used by him to describe some new South African streptaxid species and subspecies (Burnup 1914).

Burnup (1852–1928) was born in England and moved to South Africa in 1874, where he devoted himself to malacology (Meanwell 1928). Most of his collection was presented to what was then the Natal Museum (now KwaZulu-Natal Museum), Pietermaritzburg, South Africa, but throughout the years he also contributed many specimens to the Natural History Museum (NHM, London, UK) and sent them to colleagues worldwide (Meanwell 1928; Tomlin 1928). Some specimens made their way to the NMNZ via an indirect route, through one of those exchanges with colleagues.

The labels of the small Burnup collection at the NMNZ, although not original, bear the indication that they belonged to Henry Suter's collection. Suter (1841–1918) was a Swiss-born naturalist who immigrated to New Zealand, where he began to study molluscs (Hyde 2017). Suter exchanged material with malacologists and collectors worldwide and gathered a large collection; the land snails were eventually acquired by the NMNZ (Hyde 2017; Salvador 2019). More importantly, the labels of ten lots from Burnup's material indicate that these types of shells type are specimens from Burnup's 1914 publication.

As such, given that the NMNZ's Burnup collection contains potential type specimens, herein we investigate their status as types and provide a full annotated and illustrated catalogue of the material.

Material and methods

Soon after describing the species, Burnup sent the 'type' specimens of all his new taxa to the NHM (Tomlin 1928) and indicated so (or his future intention of doing so) in his publications (e.g., Burnup 1914: 31 and subsequent figure legends). Our understanding is that these would be the holotypes. Burnup also sent other specimens (paratypes) to the NHM, as well as keeping some in the Natal Museum. Evidently, he also sent specimens to colleagues like Henry Suter in New Zealand and Henry A. Pilsbry in Philadelphia, USA.

Unfortunately, the original labels of Suter's specimens were discarded when Suter's collection was incorporated into the NMNZ's collection, but the information on them was transcribed. Suter kept careful notes regarding the provenance of the material and it is expected that the transcribed labels would bear the same information, as is the case for Dautzenberg's specimens at the NMNZ, for which some of the original labels remain (Salvador and Breure 2020). Similar to Dautzenberg's material, ten of Burnup's lots indicate that the specimens are paratypes. To investigate this, we compared the locality and date noted on the available labels against the type locality defined in Burnup's publications. Unfortunately, the labels in the NMNZ have no information on the collector(s) recorded, which would be an important source of supporting evidence given that Burnup (1914) indicated the collectors in his publication.

Nine of ten Burnup lots with “paratype” indication at NMNZ proved to contain type specimens (or at least potential types). Below, we present an illustrated annotated catalogue of all Streptaxidae taxa with type specimens present in the NMNZ’s Burnup collection. The taxa are listed alphabetically according to their specific or sub-specific name, followed by information about the original description and type locality (Burnup 1914), type specimens (including those in the NHM), current taxonomic status (van Bruggen 1980; Herbert and Kilburn 2004; MolluscaBase 2020), and a brief discussion. In order to provide a comprehensive guide to Burnup’s Streptaxidae material in the NMNZ collection, we present below the non-type material as well: one lot was included in the main text and figured for future reference, as it was erroneously recorded as a paratype; the remaining lots are listed in the Appendix I. Finally, the Appendix I also compiles the information about the paratypes of the species treated herein that are housed in the KwaZulu-Natal Museum (NMSA).

Additional Burnup paratypes came to the NHM in 1937 with the donation of the collection of Matthew William Kemble Connolly (1872–1947). It should be noted that in many cases these subsequent paratype specimens were added to the holotype and paratype lots earlier donated by Burnup himself. In some cases, the registration numbers of both lots and the number of specimens contained in these lots do not add up. It is unclear if this means that specimens have later been lost or if they were incorrectly numbered since the Connolly register entries do not often give the number of specimens. In the list below, locality data for NHM specimens that were originally presented by Burnup come directly from specimen labels. Locality data for those specimens from the Connolly collection come from the registers, since no original labels seem to be available for these lots.

Systematics

Ennea farquhari var. *avena* Burnup, 1914

Fig. 1A, B

Ennea farquhari var. *avena* Burnup, 1914: 46, pl. 4, figs 28–31.

Type locality. “Maritzburg; also Pinetown and Durban (Burnup), Nottingham Road (Taynton), all in Natal” (Burnup 1914: 46).

Type material. *Paratypes* NMNZ M.207153 (1 shell, Pietermaritzburg, ex H. Suter colln 5706); NMNZ M.207154 (1 shell, Durban, ex H. Suter colln 5707).

Current taxonomic status. Synonymous with *Gulella farquhari* (Melvill & Ponsoby, 1895) (van Bruggen 1980: 27).

Discussion. Burnup’s (1914) figs 28–30 depict the type specimen from Pietermaritzburg and his fig. 31, another specimen (a paratype) from Durban. Those localities coincide with the two specimens at the NMNZ, making them paratypes. Further type specimens are in the NHM collection, namely, the holotype (NHMUK

1914.12.19.12, Maritzburg, presented by Burnup), four paratypes from Pietermaritzburg (NHMUK 1914.12.19.28–29, Maritzburg, presented by Burnup; NHMUK 1937.12.30.849–850, Maritzburg, ex. Connolly collection), and another three paratypes from Durban (NHMUK 1914.12.19.30–32, presented by Burnup; NHMUK 1937.12.30.848, ex. Connolly collection).

***Ennea maritzburgensis* var. *contracta* Burnup, 1914**

Fig. 1C

Ennea maritzburgensis var. *contracta* Burnup, 1914: 66, pl. 4, figs 42–44.

Type locality. “Nottingham Road (A. J. Taynton), very plentiful; also Karkloof and Curry’s Post (Taynton), all in Natal” (Burnup 1914: 66).

Type material. *Paratypes* NMNZ M.207175 (3 shells, Nottingham Road, ex H. Suter colln 5712).

Current taxonomic status. Junior homonym of *Ennea contracta* Quadras & Mollendorff, 1895. Accepted as *Gulella maritzburgensis* (Melvill & Ponsonby, 1893) (Herbert and Kilburn 2004: 181).

Discussion. The locality of the present specimens matches the type locality; they are thus considered paratypes. Further type specimens in the NHM include the holotype (NHMUK 1914.12.19.16, Nottingham Road, presented by Burnup) and five paratypes (NHMUK 1914.12.19.55–57, Nottingham Road, presented by Burnup; NHMUK 1937.12.30.1048–1050, Natal, ex. Connolly collection), all housed in a single lot.

***Ennea darglensis* var. *illovoensis* Burnup, 1914**

Fig. 1D

Ennea darglensis var. *illovoensis* Burnup, 1914: 49, pl. 4, figs 33–35.

Type locality. “Ntimbankulu, Mid-Illovo (Burnup)” (Burnup 1914: 49).

Type material. *Paratypes* NMNZ M.207156 (3 shells, Ntimbankulu, ex H. Suter colln 5709).

Current taxonomic status. Accepted as *Gulella darglensis illovoensis* (Burnup, 1914) (van Bruggen 1980: 16).

Discussion. The locality of the present specimens matches the type locality; they are thus considered paratypes. Further type specimens in the NHM include the holotype (NHMUK 1914.12.19.14, Ntimbankulu, presented by Burnup) and six paratypes (NHMUK 1914.12.19.37–40, Ntimbankulu, presented by Burnup; NHMUK 1937.12.30.783–784, Natal, ex. Connolly collection), all housed in a single lot.

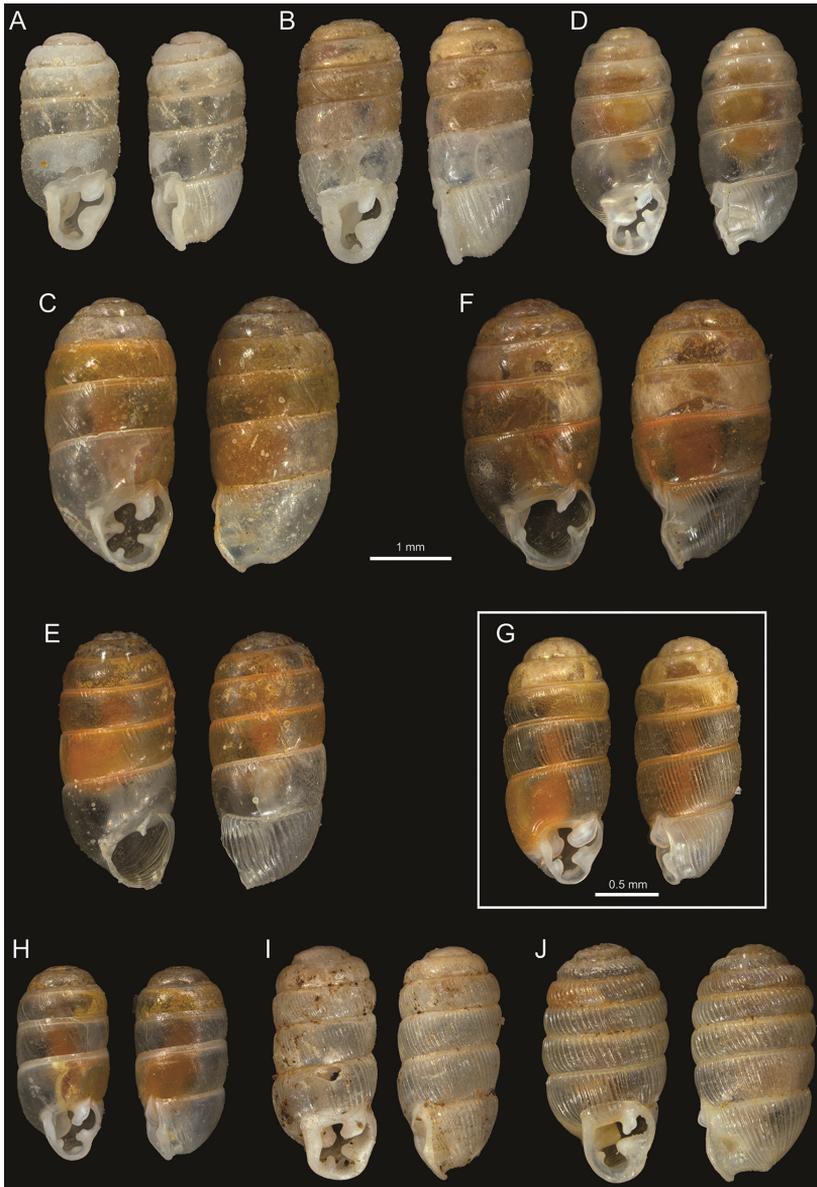


Figure 1. Burnup's type specimens in the NMNZ collection. All figures to scale (bar = 1 mm), except for Fig. 1G, not to scale (bar = 0.5 mm) **A** paratype of *Ennea farquhari* var. *avena* Burnup, 1914, NMZN M.207153 **B** paratype of *Ennea farquhari* var. *avena* Burnup, 1914, NMZN M.207154 **C** paratype of *Ennea maritzburgensis* var. *contracta* Burnup, 1914, NMZN M.207175 **D** paratype of *Ennea darglensis* var. *illovoensis* Burnup, 1914, NMZN M.207156 **E** paratype of *Ennea inbluzaniensis* Burnup, 1914, NMZN M.207160 **F** specimen of *Gulella elliptica manca* (Burnup, 1914), NMZN M.207151 **G** paratype of *Ennea melvilli* Burnup, 1914, NMZN M.207157 **H** paratype of *Ennea mooiensis* Burnup, 1914, NMZN M.207158 **I** paratype of *Ennea ponsonbyi* Burnup, 1914, NMZN M.207159 **J** paratype of *Ennea isipingoensis* var. *sturanyi* Burnup, 1914, NMZN M.207149.

***Ennea inhluzaniensis* Burnup, 1914**

Fig. 1E

Ennea inhluzaniensis Burnup, 1914: 71, pl. 5, figs 53–55.**Type locality.** “Inhluzani Hill, Dargle, Natal (Burnup)” (Burnup 1914: 71).**Type material.** *Paratypes* NMNZ M.207160 (2 shells, Inhluzani, ex H. Suter colln 5713).**Current taxonomic status.** Accepted as *Gulella inhluzaniensis* (Burnup, 1914) (Herbert and Kilburn 2004: 166).**Discussion.** The locality of the present specimens matches the type locality; they are thus considered paratypes. Further type specimens in the NHM include the holotype (1914.12.19.6, Inhluzani, presented by Burnup) and four paratypes (NHMUK 1914.12.19.49–50, Inhluzani, presented by Burnup; NHMUK 1937.12.30.967–968, Natal, ex. Connolly collection), all housed in a single lot.***Ennea elliptica* var. *manca* Burnup, 1914**

Fig. 1F

Ennea elliptica var. *manca* Burnup, 1914: 39, pl. 3, figs 13, 14.**Type locality.** “Fort Nottingham; also Curry’s Post, Natal (A. J. Taynton)” (Burnup 1914: 39).**Material.** NMNZ M.207151 (1 shell, Nottingham Road, ex H. Suter colln 5704).**Current taxonomic status.** Synonymous with *Gulella elliptica elliptica* (Melvill & Ponsonby, 1898) (van Bruggen 1980: 20).**Discussion.** The locality of the present specimen (Nottingham Road) is not a perfect match to the type locality given by Burnup (1914), although it is definitely in the same area. Burnup (1914) recorded Fort Nottingham as a different locality than Nottingham Road (see entry for *G. melvilli* below). As such, the present specimen is not considered type material. The types can be found in the NHM collection: holotype (NHMUK 1914.12.19.10, Fort Nottingham, presented by Burnup) and two paratypes (NHMUK 1914.12.19.23–24 Fort Nottingham, presented by Burnup), all housed in a single lot. A further specimen in the NHM (NHMUK 1937.12.30.801, ex. Connolly collection), despite being noted as a paratype (though original label reads ‘co-type’), is not in fact a type, as it was collected in Karkloof, which is not part of the type locality (Burnup 1914).***Ennea melvilli* Burnup, 1914**

Fig. 1G

Ennea melvilli Burnup, 1914: 55, pl. 3, figs 21–23.

Type locality. “Nottingham Road (Taynton); also Karkloof (McBean), Curry’s Post and Fort Nottingham (Taynton), Dargle and Edendale (Burnup)” (Burnup 1914: 55).

Type material. Paratypes NMNZ M.207157 (4 shells, Nottingham Road, ex H. Suter colln 5710).

Current taxonomic status. Accepted as *Gulella melvilli* (Burnup, 1914) (Herbert and Kilburn 2004: 189).

Discussion. The locality of the present specimens matches the type locality; they are thus considered paratypes. Further type specimens in the NHM include the holotype (NHMUK 1914.12.19.4, Nottingham Road, presented by Burnup) and three paratypes (NHMUK 1937.12.30.1051–1054, Natal, ex. Connolly collection), all housed in a single lot, and another two paratypes from Dargle (NHMUK 1914.12.19.25–27, presented by Burnup).

Ennea mooiensis Burnup, 1914

Fig. 1H

Ennea mooiensis Burnup, 1914: 62, pl. 5, figs 49–51.

Type locality. “Game Pass, Upper Mooi River, Natal (Burnup)” (Burnup 1914: 63).

Type material. Paratypes NMNZ M.207158 (3 shells, Game Pass, ex H. Suter colln 5711).

Current taxonomic status. Accepted as *Gulella mooiensis* (Burnup, 1914) (Herbert and Kilburn 2004: 191).

Discussion. The locality of the present specimens matches the type locality; they are thus considered paratypes. Further type specimens in the NHM include the holotype (NHMUK 1914.12.19.5, Game Pass, Upper Mooi River, Natal, presented by Burnup) and three paratypes (NHMUK 1937.12.30.1076–1079, Natal, ex. Connolly collection), all housed in a single lot, and a further lot of four paratypes (NHMUK 1914.12.19.45–48, Game Pass, presented by Burnup).

Ennea ponsonbyi Burnup, 1914

Fig. 1I

Ennea ponsonbyi Burnup, 1914: 78, pl. 5, figs 67–69.

Type locality. “Gowie’s Kloof, Grahamstown, Cape of Good Hope (Farquhar)” (Burnup 1914: 79).

Type material. Paratypes NMNZ M.207159 (2 shells, Grahamstown, ex H. Suter colln 5714).

Current taxonomic status. Accepted as *Gulella ponsonbyi* (Burnup, 1914) (Herbert and Kilburn 2004: 189).

Discussion. The locality of the present specimens does not match exactly the type locality, since there is no mention of Gowie's Kloof on the label. However, Burnup (1914: 79) stated that “all the specimens I have seen (...) come from the same locality”, so we can assume that the locality Gowie's Kloof was likely simply omitted on the new NMNZ label. The type specimens in the NHM include the holotype (NHMUK 1914.12.19.1, Gowie's Kloof Grahamstown, presented by Burnup) and three paratypes (NHMUK 1937.12.30.1163–1165, Grahamstown, ex. Connolly collection), all housed in a single lot.

***Ennea isipingoensis* var. *sturanyi* Burnup, 1914**

Fig. 1J

Ennea isipingoensis var. *sturanyi* Burnup, 1914: 36, pl. 3, figs 4–6.

Type locality. “Ntimbankulu, Mid-Illovo (Burnup). Other localities: Karkloof (Taynton), Maritzburg, Howick and Dargle (Burnup)” (Burnup 1914: 37).

Type material. *Paratypes* NMNZ M.207149 (3 shells, Karkloof, ex H. Suter colln 5703).

Current taxonomic status. Synonymous with *Gulella isipingoensis* (Sturany, 1898) (Herbert and Kilburn 2004: 188).

Discussion. The locality of the type figured by Burnup (1914) is Ntimbankulu. The NMNZ specimens, from Karkloof, are paratypes. Further type specimens in the NHM include the holotype (NHMUK 1914.12.19.7, Ntimbankulu, presented by Burnup) and four paratypes (NHMUK 1914.12.19.33–36, Ntimbankulu, presented by Burnup; NHMUK 1937.12.30.978–979, Natal, ex. Connolly collection), all housed in a single lot, and four further possible paratypes (NHMUK 1914.12.19.18–21, Ntimbankulu, presented by Burnup) which were not listed as type material in the original register entry yet match the type locality. A specimen noted as ‘possible syntype’ is housed in the Manchester Museum, UK, under the registry number MANCH.EE.5867 (McGhie 2008; Ablett et al. 2019). That specimen is labelled ‘Natal’ and lists Spence as collector, which does not coincide with the data published in Burnup (1914). There is also no mention of anyone named Spence in Burnup's land snail material (or at all) in the KwaZulu-Natal Museum. We conclude that the Manchester Museum specimen is, therefore, not part of the type series.

Concluding remarks

The present specimens from Burnup's collection include previously unrecognised type specimens of eight taxa. Further historical type material of terrestrial snails has also been recently re-discovered in the NMNZ collection (Salvador and Breure 2020), which although focused on New Zealand and Australasia, has a wealth of land snails

from other parts of the world, a large part of which was obtained via Suter's collection or in earlier days of the then Dominion Museum. There is still much information locked away in this extralimital material (Salvador 2019), including surprising type specimens such as the present ones. Thus, we are confident that future study of this collection (and similar ones worldwide) will uncover more of these lost treasures, which need to be made available to the scientific community and the public.

Acknowledgements

We are very grateful to Dai Herbert and Linda Davis for information about the types in the NMSA collection and several helpful comments on the manuscript; to Ashleigh Immers (NMNZ) for the help in databasing Burnup's specimens; to Jean-Claude Stahl (NMNZ) for the photos of the specimens used herein; and to Mary Cole, Ben Rowson and Igor Muratov for the helpful reviews and suggestions.

References

- Ablett J, Brown C, Gallichan J, Gordon D, Holmes AM, Hunter T, Machin R, Morgenroth H, Oliver PG, Petts R, Pye S, Reilly M, Rowson B, Salvador A, Sutcliffe R, Turner JA, Wood H (2019) Mollusca types in Great Britain. <https://gbmolluscatypes.ac.uk> [Accessed: 16/04/2020]
- Burnup HC (1914) On South African *Ennea*, with descriptions of new species and varieties. *Annals of the Natal Museum* 3(1): 29–82.
- Herbert D, Kilburn D (2004) *Field Guide to The Land Snails and Slugs of Eastern South Africa*. Natal Museum, Pietermaritzburg, 336 pp.
- Hyde P (2017) *A Colonial Naturalist: Henry Suter's Life of Discovery and Hardship in New Zealand*. Sphenodon, Eastbourne, 300 pp.
- McGhie HA (2008) Catalogue of type specimens of molluscs in the collection of The Manchester Museum, The University of Manchester, UK. *ZooKeys* 1: 1–46. <https://doi.org/10.3897/zookeys.4.32>
- Meanwell R (1928) Henry Clifden Burnup. *The Nautilus* 42(2): 62–63. [https://doi.org/10.1016/S0033-3506\(28\)80034-5](https://doi.org/10.1016/S0033-3506(28)80034-5)
- MolluscaBase (2020) MolluscaBase. <http://www.molluscabase.org/> [Accessed: 20/04/2020]
- Salvador RB (2019) Brazilian, Uruguayan and Argentinian land snails in the collection of the Museum of New Zealand Te Papa Tongarewa. *Tuhinga* 30: 82–98.
- Salvador RB, Breure ASH (2020) Type material of Clausiliidae door snails from Philippe Dautzenberg in the collection of the Museum of New Zealand Te Papa Tongarewa. *Tuhinga* 31: 56–69.
- Tomlin JRB (1928) Obituary: Henry Clifden Burnup, 1852–1928. *The Journal of Molluscan Studies* 18(4): 142–143.
- van Bruggen AC (1980) Size clines and subspecies in the streptaxid genus *Gulella* Pfr. (Mollusca, Gastropoda Pulmonata) in southern Africa. *Zoölogische Verhandelingen* 180: 3–62.

Appendix I

Below are listed the paratypes of the species treated herein, housed in the collection of the KwaZulu-Natal Museum (NMSA), Pietermaritzburg, South Africa: *Ennea farquhari* var. *avena* Burnup, 1914: NMSA B5847/T2692 (1 shell, Pietermaritzburg, Botanical Gardens, J. Farquhar colln, ex Albany Museum 1980), NMSA E8226/T281 (4 shells, Durban, J. Farquhar colln, ex Albany Museum 1980), NMSA E8227/T282 (2 shells, Durban, J. Farquhar colln, ex Albany Museum 1980), NMSA W517/T1942 (2 shells, Durban, ex Transvaal Museum 1978). *Ennea maritzburgensis* var. *contracta* Burnup, 1914: NMSA 2563/T568 (4 shells, Nottingham Road, A.J. Taynton, ex H.C. Burnup colln), NMSA B7397/T2830 (4 shells, Nottingham Road, ex Albany Museum 1980), NMSA E8233/T288 (3 shells, Nottingham Road, Farquhar colln, ex Albany Museum 1980), NMSA W520/T1946 (4 shells, Nottingham Road, ex Transvaal Museum 1978), NMSA W1734/T1989 (14 shells, Karkloof, A.J. Taynton), NMSA W1739/T1992 (132 shells + 3 broken pieces, Nottingham Road), NMSA W1741/T1993 (3 shells, Karkloof, A.J. Taynton, ex H.C. Burnup), NMSA W1742/T1994 (2 shells, Curry's Post, A.J. Taynton, ex H.C. Burnup colln). *Ennea darglensis* var. *illovoensis* Burnup, 1914: NMSA 2560/T546 (5 shells, Mid-Illovo, Ntimbankulu, Backworth [farm], H.C. Burnup; van Bruggen, 1980: fig. 5), NMSA B6697/T2791 (22 shells, Mid-Illovo, Ntimbankulu), NMSA E8232/T287 (3 shells, Mid-Illovo, Ntimbankulu, H.C. Burnup, J. Farquhar colln, ex Albany Museum 1980), NMSA W521/T1947 (4 shells, Mid-Illovo, Ntimbankulu, ex Transvaal Museum 1978). *Ennea inhluzaniensis* Burnup, 1914: NMSA 2551/T559 (7 shells [in two lots], Dargle, Nhlosane, farm 'Furth', H.C. Burnup, ex W. Falcon colln), NMSA B5853/T2626 (2 shells, Dargle, Nhlosane, J. Farquhar colln, ex Albany Museum 1980), NMSA W516/T1941 (4 shells, Dargle, Nhlosane [Inhluzani] Mt., ex Transvaal Museum 1978), NMSA W830/T1960 (5 shells, Dargle, Nhlosane). *Ennea elliptica* var. *manca* Burnup, 1914: NMSA 2547/T548 (1 shell, Fort Nottingham, A.J. Taynton), NMSA 2552/T548 (2 shells, Curry's Post, A.J. Taynton), NMSA B6530/T2790 (1 shell, Curry's Post, A.J. Taynton, Burnup colln), NMSA E8236/T291 (1 shell, Fort Nottingham, J. Farquhar colln, ex Albany Museum 1980). *Ennea melvilli* Burnup, 1914: NMSA 2565/T569 (14 shells, Nottingham Road, A.J. Taynton), NMSA B5845/T2690 (4 shells, Nottingham Road, J. Farquhar colln, ex Albany Museum 1980), NMSA B5846/T2691 (9 shells, Nottingham Road, J. Farquhar colln, ex Albany Museum 1980), NMSA E8235/T290 (3 shells, Nottingham Road, J. Farquhar colln, ex Albany Museum 1980), NMSA W518/T1943 (4 shells, Nottingham Road, ex Transvaal Museum 1978). *Ennea mooiensis* Burnup, 1914: NMSA 2544/T571 (121 shells [in four lots], Kamberg, Game Pass, H.C. Burnup, ex W. Falcon), NMSA B5852/T2694 (4 shells, Kamberg, Game Pass, J. Farquhar colln, ex Albany Museum 1980), NMSA B7400/T2833 (3 shells, Kamberg, Game Pass, ex Albany Museum 1980), NMSA W268/T1936 (4 shells, Kamberg, Game Pass, upper Mooi River, ex

Transvaal Museum 1978). *Ennea ponsonbyi* Burnup, 1914: NMSA 2540/T577 (4 shells, Grahamstown, Gowie's Kloof, A. Gowie, ex McGregor Museum, Kimberley), NMSA W174/T1900 (3 shells, Grahamstown, Gowie's Kloof, J. Farquhar, ex H.C. Burnup). *Ennea isipingoensis* var. *sturanyi* Burnup, 1914: NMSA E8237/T292 (2 shells, Mid-Illovo, Ntimbankulu, ex Albany Museum 1980), NMSA W1732/T1988 (1 shell, Pietermaritzburg, Town Bush, 1911).

There are further – non-type – specimens of Streptaxidae (all *Gulella* spp.) from Suter's collection at the NMNZ that originally belonged to Burnup: *G. arnoldi* (Sturany, 1898): NMNZ M.207176 (1 shell, Umkomaas, ex H. Suter colln 4217); *G. calopasa* (Melvill & Ponsonby, 1903): NMNZ M.207193 (1 shell, Port Shepstone, ex H. Suter colln 5228), NMNZ M.207195 (1 shell, Pinetown, ex H. Suter colln 4893); *G. columnella* (Melvill & Ponsonby, 1901): NMNZ M.207194 (1 shell, Karkloof, ex H. Suter colln 4895); *G. crassidens* (L. Pfeiffer, 1859): NMNZ M.207145 (1 shell, Pietermaritzburg ("Maritzburg"), ex H. Suter colln 4873), NMNZ M.207146 (1 shell, Pinetown, ex H. Suter colln 4890); *G. daedalea* (Melvill & Ponsonby, 1903): NMNZ M.207150 (7 shells, Umfolosi Drift, ex H. Suter colln 5231); *G. darglensis* (Melvill & Ponsonby, 1908): NMNZ M.207155 (2 shells, Inhluzani, ex H. Suter colln 5708); *G. dunkeri* (L. Pfeiffer, 1855): NMNZ M.207166 (3 shells, Durban, ex H. Suter colln 4214); *G. elliptica* (Melvill & Ponsonby, 1898): NMNZ M.207177 (2 shells, Pietermaritzburg ("Maritzburg"), ex H. Suter colln 4111), NMNZ M.330538 (1 shell, Umfolosi Drift, ex H. Suter colln 5231); *G. euthymia* (Melvill & Ponsonby, 1893): NMNZ M.207172 (4 shells, Pietermaritzburg ("Maritzburg"), ex H. Suter coll 4104), NMNZ M.207173 (2 shells, Umkomaas, ex H. Suter colln 4218); *G. farquhari* (Melvill & Ponsonby, 1895): NMNZ M.207152 (3 shells, Grahamstown, ex H. Suter colln 5705); *G. formosa* (Melvill & Ponsonby, 1898): NMNZ M.330537 (5 shells, Pinetown, ex H. Suter colln 4890); *G. gouldi* (L. Pfeiffer, 1856): NMNZ M.207163 (3 shells, Durban, ex H. Suter colln 4112), NMNZ M.207170 (5 shells, Pietermaritzburg ("Maritzburg"), ex H. Suter colln 4212); *G. instabilis* (Sturany, 1898): NMNZ M.207167 (4 shells, Umkomaas, ex H. Suter colln 4220); *G. isipingoensis* (Sturany, 1898): NMNZ M.207147 (3 shells, Durban, ex H. Suter colln 4882), NMNZ M.207148 (3 shells, Ntimbankulu, ex H. Suter colln 5702); *G. maritzburgensis* (Melvill & Ponsonby, 1893): NMNZ M.207174 (4 shells, Pietermaritzburg ("Maritzburg"), ex H. Suter colln 4213); *G. menkeana* (L. Pfeiffer, 1853): NMNZ M.207165 (4 shells, Umkomaas, ex H. Suter colln 4216); *G. natalensis* (Craven, 1880): NMNZ M.207164 (2 shells, Umkomaas, ex H. Suter colln 4211); *G. obovata* (L. Pfeiffer, 1855): NMNZ M.207171 (3 shells, Umkomaas, ex H. Suter colln 4215); *G. planti* (L. Pfeiffer, 1856): NMNZ M.207190 (2 shells, Durban, ex H. Suter colln 4101), NMNZ M.207191 (3 shells, Durban, ex H. Suter colln 4873); *G. queketti* (Melvill & Ponsonby, 1896): NMNZ M.207192 (3 shells, Umkomaas, ex H. Suter colln 4210); *G. separata* (Sturany, 1898): NMNZ M.207169 (2 shells, Umkomaas, ex H. Suter colln 4219); *G. wahlbergi* (Krauss, 1848): NMNZ M.207161 (3 shells, Durban, ex H. Suter colln 4113).

***Pedicelliops* gen. nov., a new genus from West Africa with striking antennae (Ephemeroptera, Baetidae)**

Thomas Kaltenbach^{1,2}, Jean-Luc Gattolliat^{1,2}

1 *Museum of Zoology, Palais de Rumine, Place Riponne 6, CH-1005 Lausanne, Switzerland* **2** *University of Lausanne (UNIL), Department of Ecology and Evolution, CH-1015 Lausanne, Switzerland*

Corresponding author: Thomas Kaltenbach (thomas.kaltenbach@bluewin.ch)

Academic editor: J. Midgley | Received 5 October 2020 | Accepted 23 November 2020 | Published 11 December 2020

<http://zoobank.org/C984D926-DA6C-40F1-BF37-33D0955B7885>

Citation: Kaltenbach T, Gattolliat J-L (2020) *Pedicelliops* gen. nov., a new genus from West Africa with striking antennae (Ephemeroptera, Baetidae). African Invertebrates 61(2): 119–135. <https://doi.org/10.3897/AfrInvertebr.61.59354>

Abstract

A new genus of Baetidae, *Pedicelliops* **gen. nov.**, and a new species, *P. capillifer* **sp. nov.**, are described from Guinea (West Africa) based on larvae. The new genus is characterized by having strongly enlarged pedicelli and very short flagella, a brush of dense, short setae between prostheca and mola of both mandibles, a small rectangular labrum, an apicolaterally pointed maxillary palp, a labial palp with a small distolateral protuberance and long setae ventrally on glossae and paraglossae. The femora of all legs are covered with numerous long, fine setae. The patellotibial suture is absent on the fore tibia and present on middle and hind tibiae. The claw is pointed with two rows of denticles. No spines are present on the posterior margins of the abdominal tergites. The imago remains unknown and the relationships with other African genera of Baetidae remains tentative. Despite being easily identifiable and of a fairly large size (body length ca. 5 mm), only two larvae were found in two highly sampled localities in West Africa.

Keywords

Filtering adaptation, Guinea, mayflies, Protapatellata, systematics

Introduction

The family Baetidae has the highest species diversity among mayflies, comprising ca. 1,100 species in 114 genera (updated from Sartori and Brittain 2015; Jacobus et al. 2019), which is close to one third of all mayfly species worldwide. They have a cosmopolitan distribution excluding Antarctica and New Zealand. Investigations of the

molecular phylogeny of the order Ephemeroptera revealed the primitive status of the family (Ogden and Whiting 2005; Ogden et al. 2009). The generic diversity of Baetidae is the highest in the Afrotropical realm (ca. 40 genera), followed by the Neotropical (ca. 27 genera) and Oriental (ca. 26 genera) realms and finally the Nearctic (20 genera), Palearctic (17 genera) and the Australasian (ca. 12 genera) realms (Gattolliat and Nieto 2009; Gattolliat 2012).

West Africa has attracted the attention of mayfly researchers for a long time and many collections and studies were done in that region. Between 1974 and 1989, the French ORSTOM (Office de la recherche scientifique et technique outre-mer), presently the IRD (Institut de Recherche pour le Développement), conducted field research in West Africa as part of an important onchocerciasis control program (Lévêque et al. 2003). Aquatic macroinvertebrates were collected in about 100 localities, mainly in Guinea, Ivory Coast, and Mali. Most localities were sampled regularly over a period of several years. As the systematics of aquatic insects was still poorly known at the time, important alpha taxonomic research was done and several articles were published including descriptions of new species and genera. The majority of mayflies known from West Africa were described based on material collected during this programme (e.g. Elouard and Forge 1978; Gillies 1980, 1989, 1993, 1997; Elouard 1986a, b; Elouard and Gillies 1989; Elouard and Hideux 1991; Gillies and Elouard 1990; Wuillot and Gillies 1993a, b, 1994; Gattolliat 2006; Gattolliat and Sartori 2006; Edia et al. 2015). However, only part of the material collected was sorted and identified. All the remaining unidentified material is now housed in the Museum of Zoology in Lausanne (Switzerland). The present study is based on larvae collected in 1985 and 1987 in two different locations in Guinea.

Materials and methods

The specimens were collected in 1985 and 1987 from two different areas of Guinea (West Africa). Specimens were preserved in 70–80% ethanol. Larvae were dissected in Celsolve (2-Ethoxyethanol) with subsequent mounting on slides in liquid Euparal, using an Olympus SZX7 stereomicroscope.

Drawings were made using an Olympus BX43 microscope. Photographs of larvae were taken with a Canon EOS 6D camera and the Visionary Digital Passport imaging system (<http://www.duninc.com>). Images were processed using Adobe Photoshop Lightroom (https://adobe.com/ch_de/products/photoshop-lightroom) and Helicon Focus version 5.3 (<http://www.heliconsoft.com>). Photographs were subsequently enhanced with Adobe Photoshop Elements 13 (https://adobe.com/ch_de/products/photoshop).

Approximate GPS coordinates to sample locations were attributed using Google Earth (<http://www.google.com/earth/download/ge/>) and Elouard et al. (1990: fig.9) and distribution maps were generated with SimpleMappr (<https://simplemappr.net>, Shorthouse 2010).

The terminology used in the manuscript follows Hubbard (1995) and Kluge (2004).

Abbreviations

MZL Museum of Zoology Lausanne (Switzerland).

Results

Pedicelliops gen. nov.

<http://zoobank.org/0BD8D226-CC87-4807-9608-247830E581C5>

Figures 1–7

Type species. *Pedicelliops capillifer* gen. et sp. nov., by present designation.

Diagnosis. Larva. This new genus is distinguished by the combination of the following characters: A) body elongate and slender, head clearly hypognathous (Figs 1, 2); B) base of antennae close to each other, with carina between them (Fig. 2d); C) antenna shorter than head length, with strong bilateral enlargement of pedicellus (Fig. 3a); D) labrum small and rectangular, distal margin with medial emargination and small, square process, dorsal surface with long, stout, simple setae, erratically distributed in distal part, not arranged in one arc (Fig. 4a); E) right mandible with a stick-like, apically denticulate prosthema and a brush of short, fine setae between prosthema and mola (Fig. 4b, c); F) left mandible with a robust prosthema, apically with a comb-shaped structure and with a brush of short, fine setae between prosthema and mola (Fig. 4d, e); G) hypopharynx with medial tuft of stout setae (Fig. 4g); H) maxillary palp 2-segmented (Fig. 4h); I) glossae basally broad, narrowing toward apex, slightly shorter than paraglossae, ventrolateral margin with a row of short, thin setae and a row of long, simple setae close to margin, apically with some robust, long, curved setae; stout paraglossae with convex outer margin and apex, ventrolateral margin with a row of long, simple setae; labial palps segment II with distolateral protuberance, directed distally, segment III conical (Fig. 4i, j); J) legs with many long, simple setae on margins and on both surfaces of femur, femoral patch absent on all legs, patellotibial suture absent on foreleg and present on middle and hind legs, claw robust and pointed, with two rows of denticles (Figs 5, 6); K) fore protoptera with a protuberance near inner margin (Fig. 2a, b); L) posterior margin of abdominal tergites I–VIII smooth without spines (Fig. 1a).

Imagines. Unknown.

Etymology. *Pedicelliops* is an arbitrary combination of letters with allusion to the Latin *pedicellus* and the Greek *iops*. *Pedicellus* is with reference to the striking second segment of the antennae and *iops* with reference to the Baetidae which look and move like small fishes. The gender is masculine.

Description. Larva. (Figs 1–6).

Body. Elongate and slender, head hypognathous (Figs 1, 2).

Head. Antenna (Figs 3a, b, 2c, d) shorter than head length with strong bilateral enlargement of pedicellus. Antennae bases in close proximity to each other, with small carina between them.

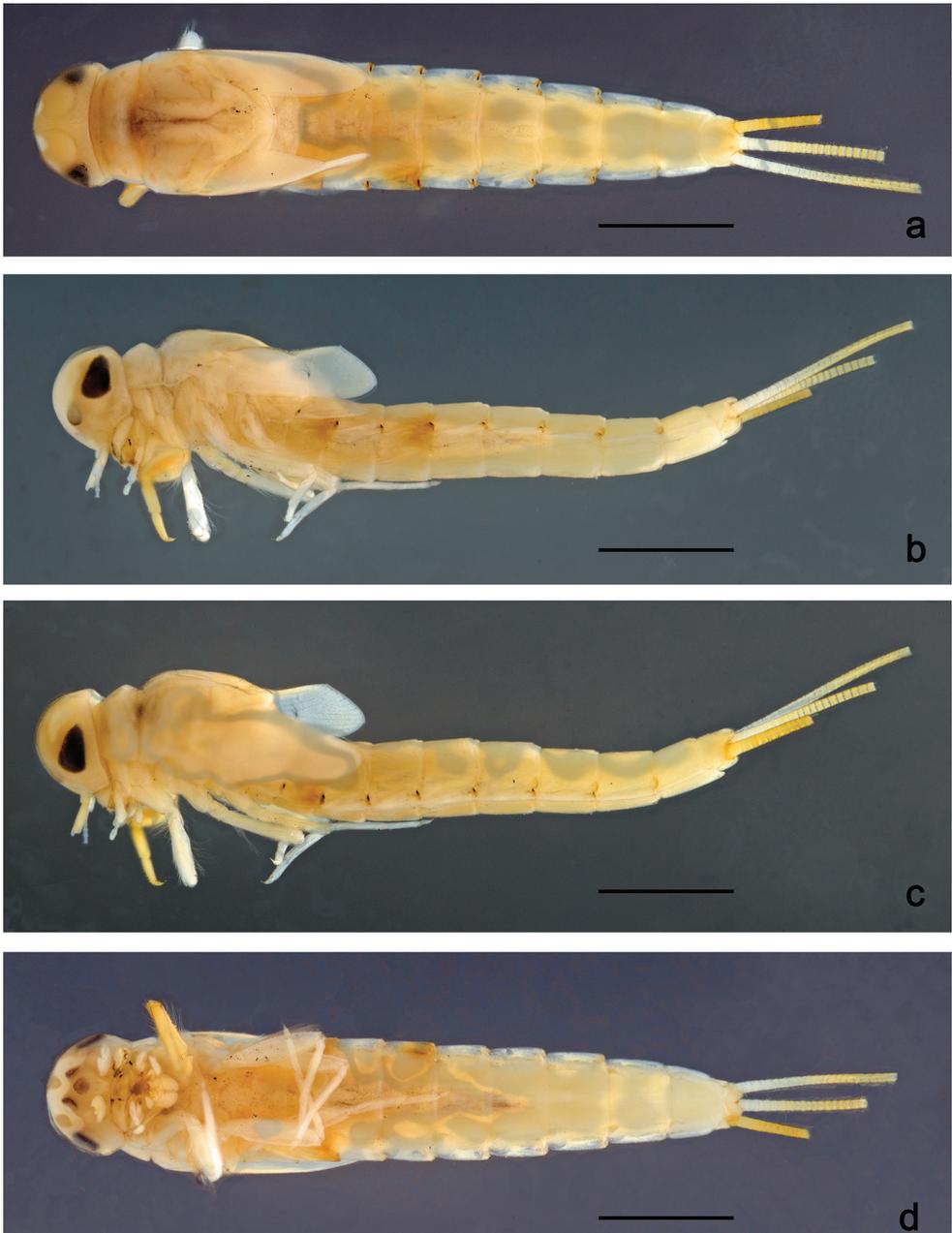


Figure 1. *Pedicelliops capillifer* gen. et sp. nov., habitus, larva **a** dorsal view **b, c** lateral views **d** ventral view. Scale bars: 1 mm.

Labrum (Fig. 4a). Small in comparison to other mouthparts, rectangular, wider than long; dorsal surface with long, stout, simple setae, erratically distributed in distal part, not arranged in one arc; ventrally with marginal row of setae composed of anterolateral long, simple setae and medial long, bifid setae.

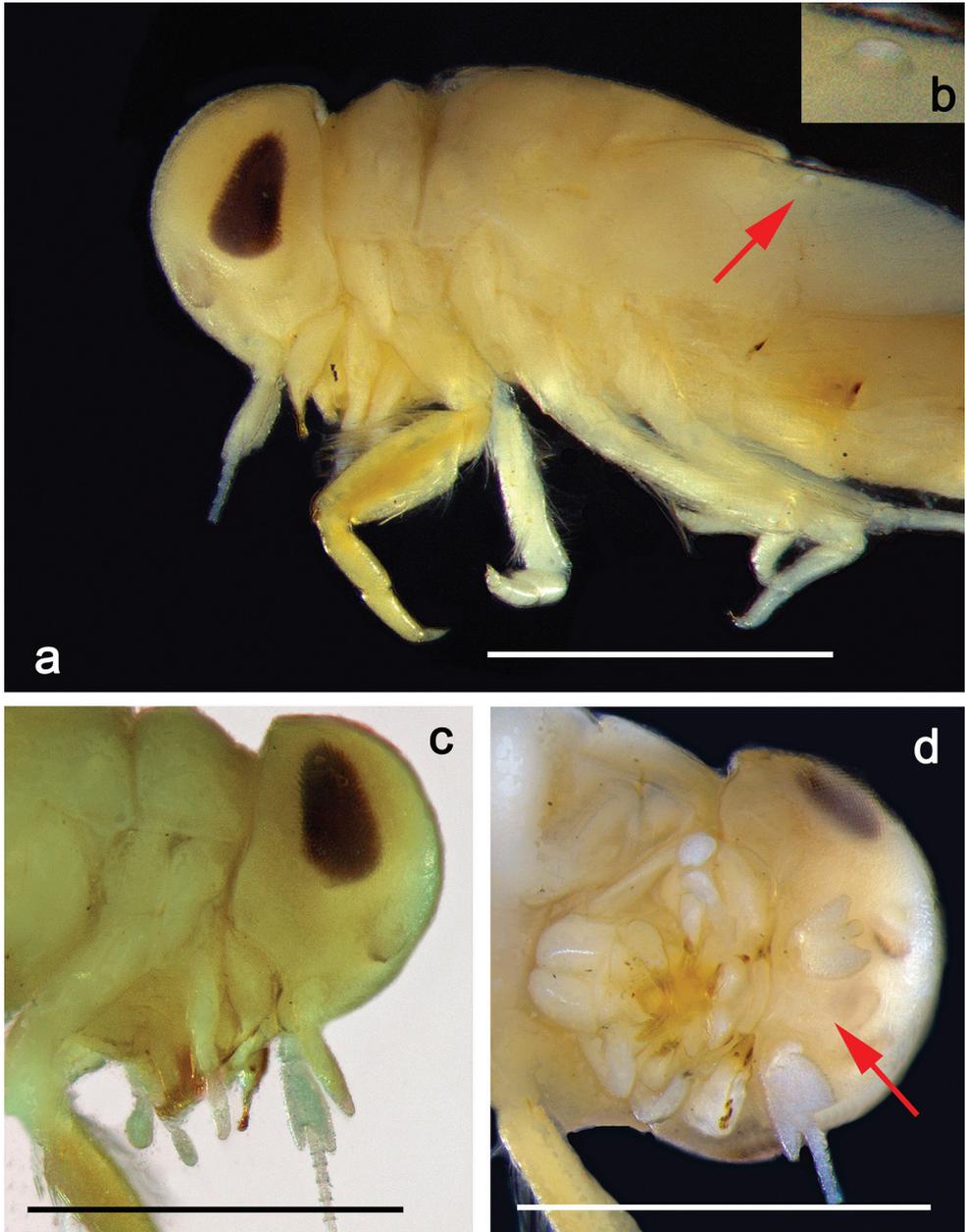


Figure 2. *Pedicelliops capillifer* gen. et sp. nov., larva morphology **a** head and thorax, lateral view **b** protuberance on fore protopteron **c** head, lateral view **d** head, ventral view (arrow: carina). Scale bars: 1 mm.

Right mandible (Fig. 4b, c). Incisor and kinetodontium fused, both with well-developed denticles; inner margin of innermost denticle without a row of thin setae; prostheca stick-like, apically denticulate; margin between prostheca and mola with a brush of abundant, short, fine setae.

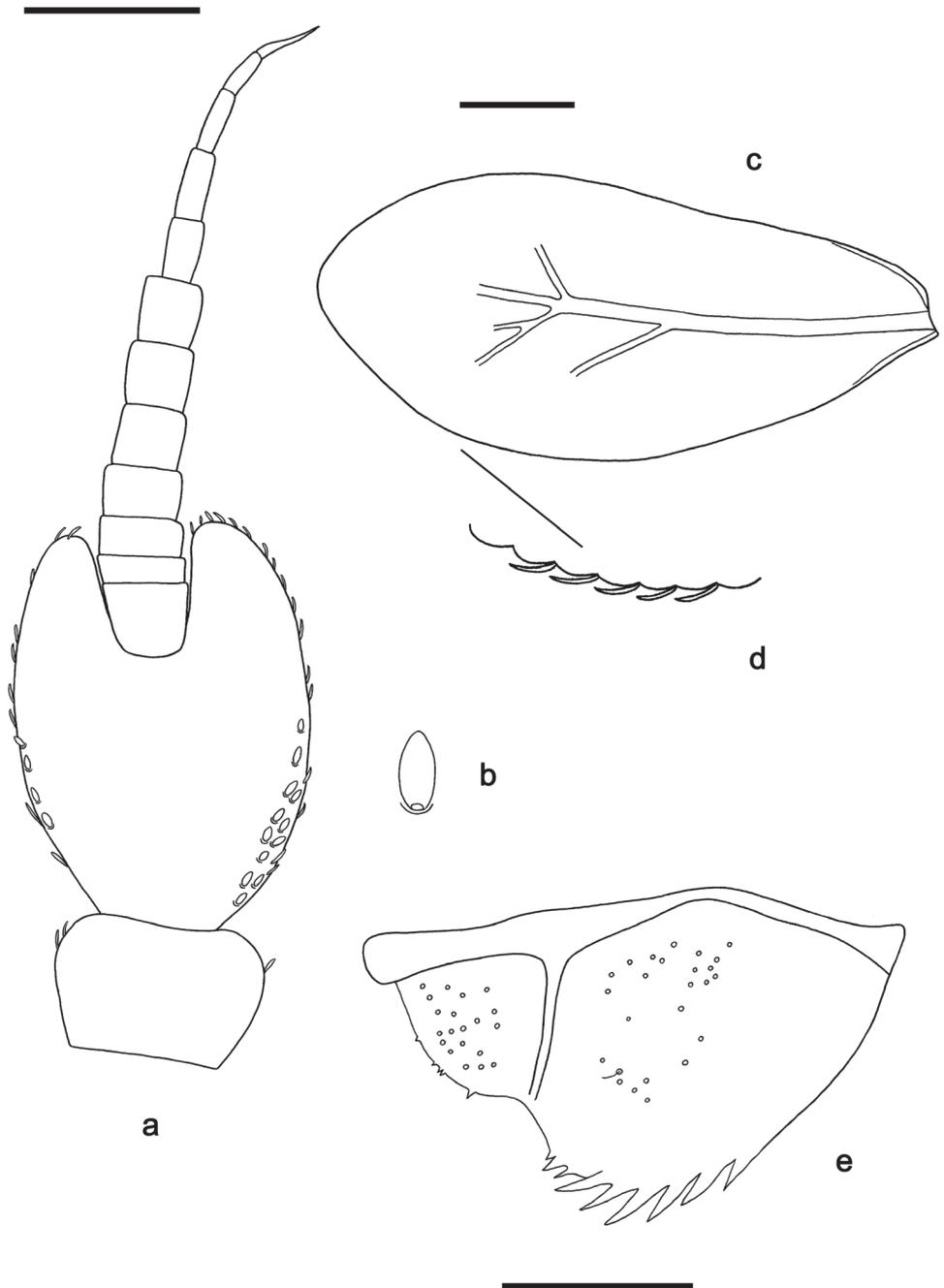


Figure 3. *Pedicelliops capillifer* gen. et sp. nov., larva morphology **a** antenna **b** scale of pedicellus **c** gill I **d** margin of gill I **e** paraproct. Scale bars: 0.1 mm.

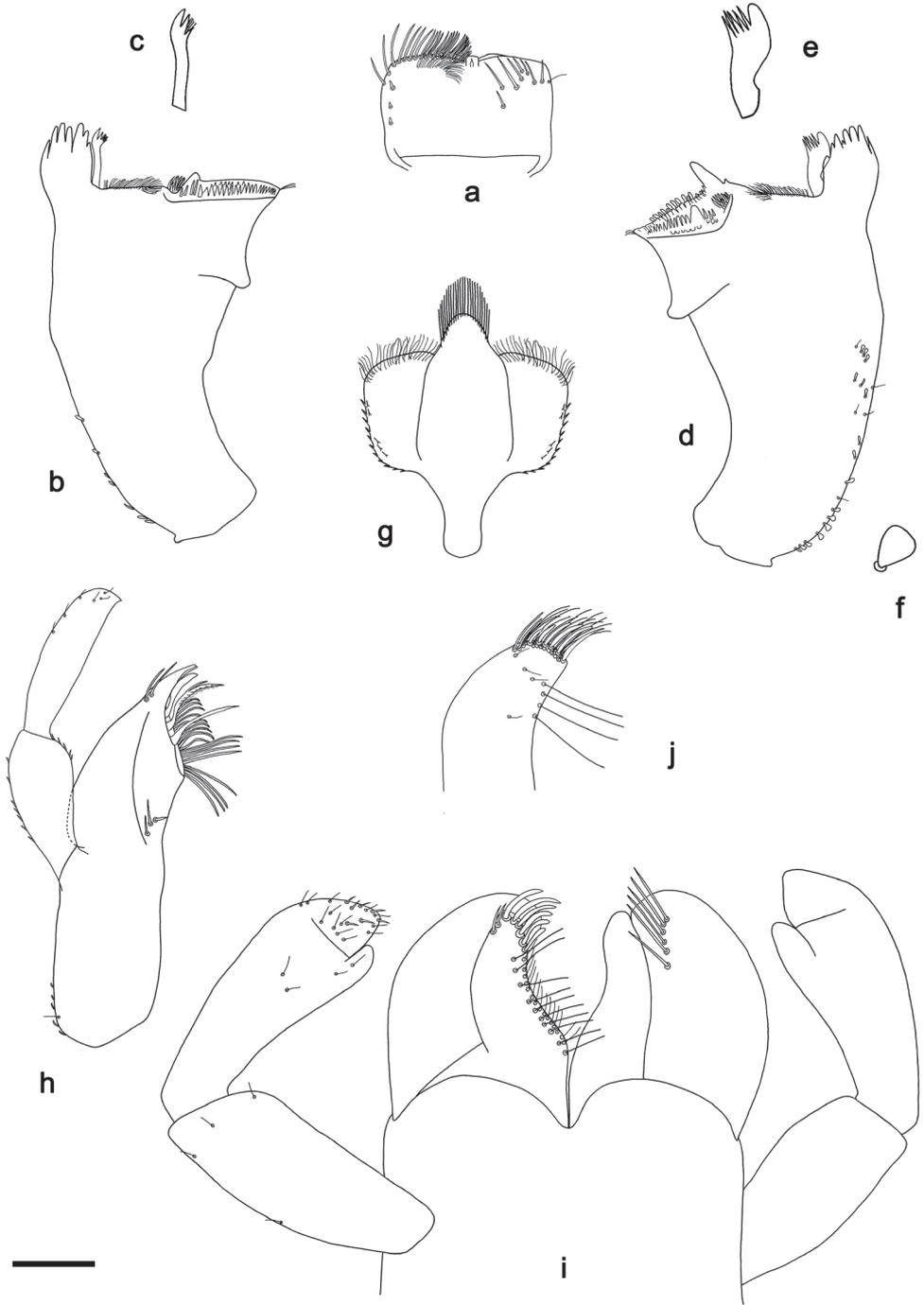


Figure 4. *Pedicelliops capillifer* gen. et sp. nov., larva morphology **a** labrum **b** right mandible **c** right prosthema **d** left mandible **e** left prosthema **f** seta on left mandible **g** hypopharynx and superlinguae **h** maxilla **i** labium (left: ventral view, right: dorsal view) **j** paraglossa (ventral view). Scale bar: 0.1 mm.

Left mandible (Fig. 4d–f). Incisor and kinetodontium fused, both with well-developed denticles; prostheca robust, apically with comb-shaped structure; margin between prostheca and mola with a brush of abundant, short, fine setae.

Hypopharynx (Fig. 4g). With a dorsomedial tuft of stout setae on lingua.

Maxilla (Fig. 4h). Apically with three stout canines and three denti-setae; distal denti-seta tooth-like, following same direction as canines, other denti-setae slender, bifid and pectinate; maxillary palp with two segments.

Labium (Fig. 4i, j). Glossae basally broad, narrowing toward apex, slightly shorter than paraglossae; ventrolateral margin with a row of short, thin setae and a row of long, simple setae close to inner margin, apically with some robust, long, curved setae. Paraglossae with convex outer margin and apex; ventrolateral margin with a row of long, simple setae; apical margin with three rows of long, stout setae (Fig. 4j). Labial palps segment II with distolateral protuberance, segment III conical.

Thorax. Fore protoptera (Fig. 2a, b). With a protuberance near inner margin.

Hind protoptera. Absent.

Foreleg (Figs 5a–c, 6a). *Femur* with many long, simple setae on dorsal and ventral margins and on both lateral surfaces; femoral patch absent; apex of femur with ventral lobe poorly developed; patellotibial suture absent; *claw* robust and pointed, with two divergent rows of denticles.

Middle and hind legs (Figs 5d–f, 6b, c). More slender than foreleg; *femur* with same setation as foreleg; femoral patch absent; anterior outer projection of femur apex directed towards the inner side of femur (Fig. 5d); patellotibial suture present; *claw* as in foreleg.

Abdomen. Tergites. Posterior margin of segments I–VIII smooth, without spines.

Gills (Fig. 3c, d). Seven pairs of gills on segments I–VII, dorsally oriented.

Paraproct (Fig. 3e). Without prolongation at posterior margin, with stout, marginal spines; cercotractor with few minute, marginal spines.

Caudal filaments (Fig. 1a, d). Inner margin of cerci with 2–18 long, thin setae per segment, increasing in number distally; paracercus bilaterally with 3 to more than 12 long, thin setae per segment, increasing in number distally. Paracercus not reduced, but broken.

Description. *Imagines*. Unknown.

Distribution (Fig. 7). West Africa: Guinea.

***Pedicelliops capillifer* gen. et sp. nov.**

<http://zoobank.org/1A749009-4120-46F5-B79A-DEACAC30AF56>

Figures 1–7

Diagnosis. *Larva*. As the genus is monospecific, it is difficult to propose a relevant specific diagnosis. We can presume specific differences in the shape of the labial and maxillary palps, and in the setation of the forefemur. Species with less derived characters (shape of the pedicel, reduction of the labrum) may also be expected.

Etymology. With reference to the remarkable setation found ventrally on the glossae and paraglossae, and on the femora.

Type-material. *Holotype*. GUINEA • larva; Bas. Niger, River Niandan, Loc. Samsambaya; ca. 400 m; 25.i.1985; leg. J.-M. Elouard; on slides; GBIFCH 00592365, GBIFCH 00592366; thorax and abdomen in alcohol; GBIFCH 00515518; MZL. ***Paratype*.** GUINEA • larva; Boussoulé, Milo River; ca. 360 m; 14.iv.1987; leg. J.-M. Elouard; on slides; GBIFCH 00592319, GBIFCH 00592320; MZL.

Description. *Larva*. (Figs 1–6). *Body length* 5.2 mm.

Colouration (Fig. 1a–d). Colouration strongly altered due to long term storage of the larvae in alcohol, with the cuticle becoming transparent. Head, thorax and abdomen dorsally brown, fore protoptera ecrú. Head, thorax and abdomen light brown ventrally. Legs and caudalii light brown.

Head. Antenna (Fig. 3a, b). Short, ca. 2/3 of head length. Pedicellus bilaterally strongly enlarged and distally elongate on both sides. Scapus and pedicellus laterally with ovoid scales.

Labrum (Fig. 4a). Length 0.6× maximum width. Distal margin with medial emargination and a small, square process. Dorsal surface with long, stout, simple setae, erratically distributed in distal part, not arranged in one arc. Ventrally with marginal row of setae composed of anterolateral long, simple setae and medial long, bifid setae. Ventral surface with ca. three short, spine-like setae near lateral margin.

Right mandible (Fig. 4b, c). Incisor with four denticles; kinetodontium with three denticles. Margin between prostheca and mola straight; with setae along whole margin from prostheca to mola, shorter than 1/5 of prostheca. Tuft of setae present at apex of mola.

Left mandible (Fig. 4d–f). Incisor with five denticles; kinetodontium with three denticles. Margin between prostheca and mola straight; with setae along whole margin, shorter than 1/5 of prostheca, and a minute denticle toward subtriangular process. Subtriangular process long and slender, above level of area between prostheca and mola. Tuft of setae present at apex of mola.

Both mandibles with lateral margins convex. Basal half laterally with small, spatulate setae.

Hypopharynx and superlinguae (Fig. 4g). Lingua longer than wide and longer than superlinguae; with well-developed medial tuft of stout setae. Superlinguae distally rounded, lateral margins straight; long, fine, simple setae along distal margin.

Maxilla (Fig. 4h). Galea-lacinia ventrally with two simple, apical setae under canines. Medially with one spine-like seta and two short, simple setae. Maxillary palp ca. 1.3× as long as length of galea-lacinia; palp segment II 1.3× length of segment I; segment I widening in distal part; short, fine, simple setae scattered over distal part of surface of segment II; segment I laterally with short, robust setae; apex of segment II laterally pointed.

Labium (Fig. 4i, j). Inner margin of glossa with a row of short, fine, simple setae; ventral surface with a row of long, fine, simple setae close to margin; apex with six long, robust setae, curved toward inner side, and two shorter, spine-like setae; outer margin bare. Paraglossa with outer margin and apex convex and inner margin concave, slightly curved inward; ventrally with three rows of long, robust setae in apical area, four short, fine, simple setae in anteromedial area and a row of four long, simple setae on margin;

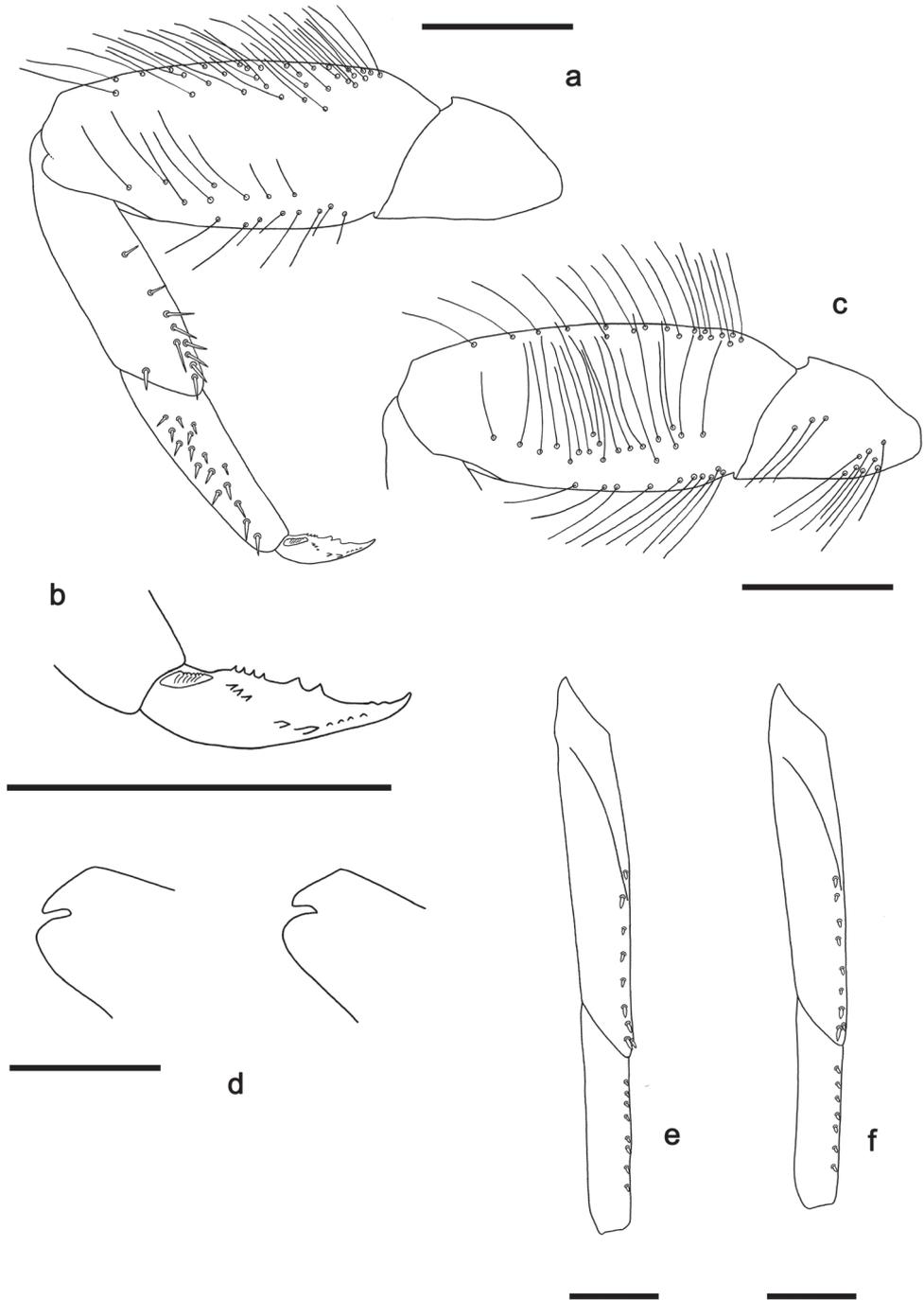


Figure 5. *Pedicelliops capillifer* gen. et sp. nov., larva morphology **a** foreleg (anterior view) **b** fore claw **c** fore femur and trochanter (posterior view) **d** apex of middle and hind femur **e** middle tibia and tarsus **f** hind tibia and tarsus. Scale bars: 0.2 mm.

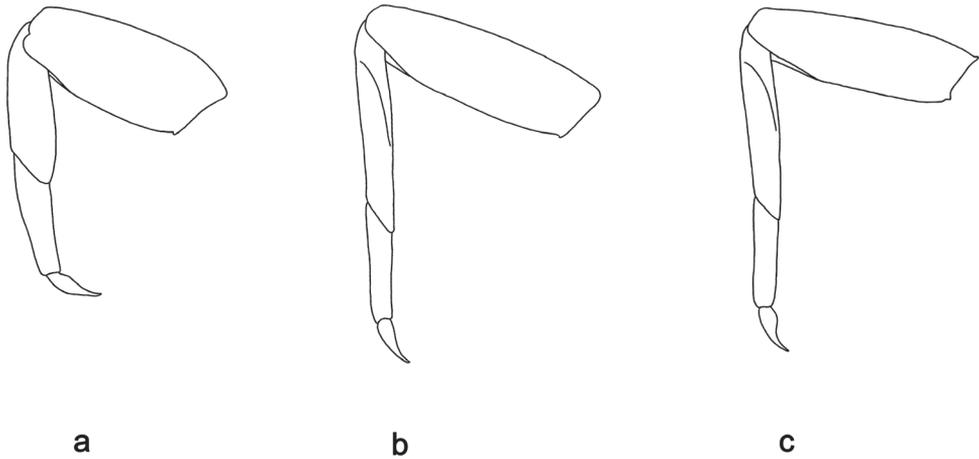


Figure 6. *Pedicelliops capillifer* gen. et sp. nov., larva morphology: **a** foreleg **b** middle leg **c** hind leg. Scale bar: 0.5 mm.

dorsally with a row of six long, spine-like setae near inner margin. Labial palp with segment I 0.9× length of segments II and III combined. Segment I with few short, fine, simple setae ventrally. Segment II with slender, distolateral protuberance, directed distad; distomedial protuberance 0.3× width of base of segment III; ventral surface with short, fine, simple setae; dorsally without spine-like setae near outer margin. Segment III conical; length 0.8× width; ventrally covered with short, spine-like setae and short, fine, simple setae.

Thorax. *Fore protoptera* (Fig. 2a, b). With a protuberance near inner margin.

Hind protoptera. Absent.

Foreleg (Figs 5a–c, 6a). Ratio of foreleg segments 1.4: 1.0: 0.8: 0.4. *Femur.* Broad, length ca. 2× maximum width. With many long, simple setae on dorsal and ventral margins as well as on anterior and posterior surface; length of setae mostly ca. 0.6× maximum width of femur; femoral patch absent. *Tibia.* Dorsal margin bare; with a row of medium, spine-like setae along ventral margin; one medium, spine-like seta at distomedial margin. Patellotibial suture absent. *Tarsus.* Dorsal and ventral margins bare, with short to medium, spine-like setae scattered over surface. *Claw* robust and distally pointed, with two divergent rows of denticles; denticles with different sizes, two apical denticles clearly larger than other denticles; subapical setae absent (Fig. 2b).

Middle and hind leg (Figs 5d, e, 6b, c). Ratio of middle and hind leg segments 1.2: 1.0: 0.6: 0.3. *Femur.* Length ca. 3× maximum width. With same setation as foreleg; femoral patch absent. *Tibia.* Dorsal margin bare. Ventral margin with a row of short, spine-like setae. Patellotibial suture present on basal ½ area. *Tarsus.* Dorsal margin bare. Ventral margin with a row of short, spine-like setae. *Claw* as foreleg.

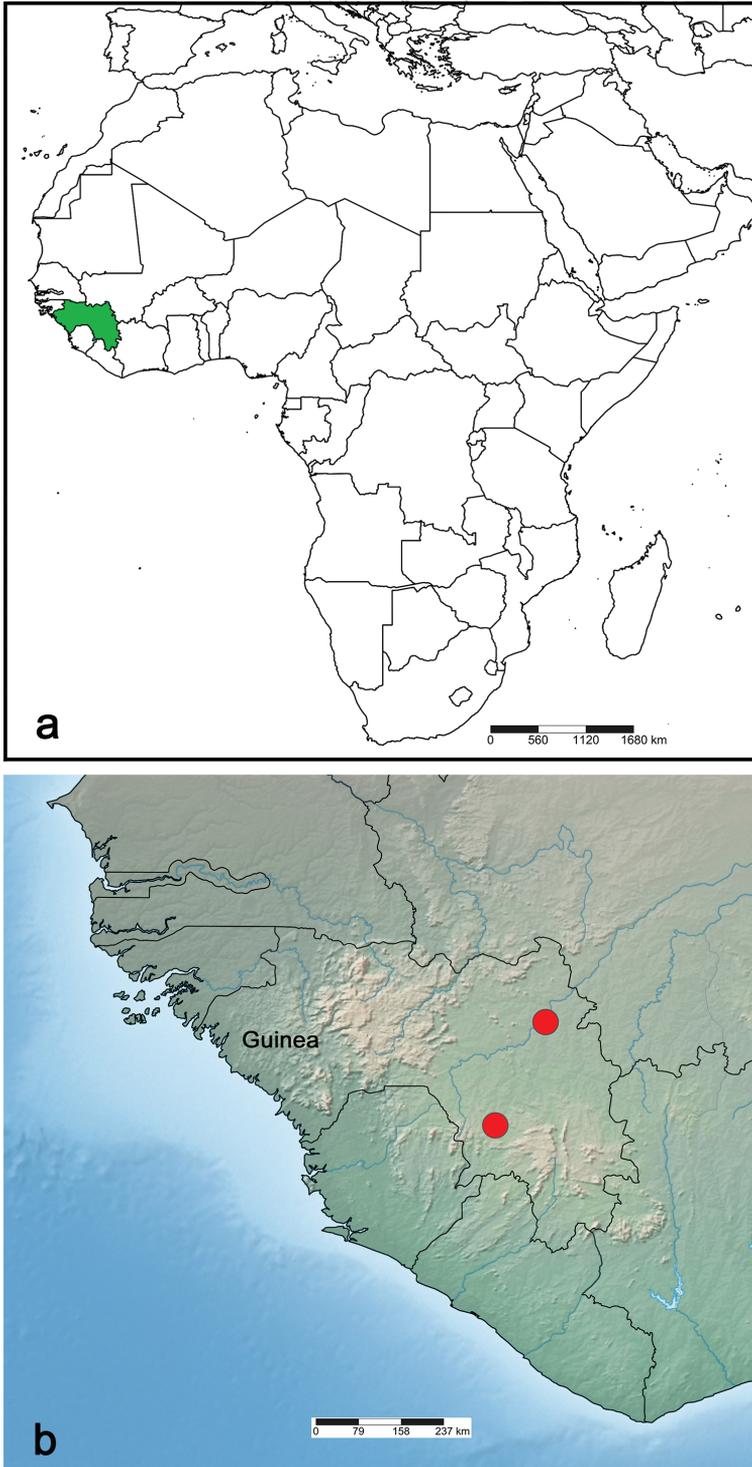


Figure 7. *Pedicelliops capillifer* gen. et sp. nov., distribution **a** Africa, green: Guinea **b** Guinea.

Abdomen. Gills (Fig. 3c, d). Seven pairs of gills on segments I–VII. Margin undulated, with short, stout, curved setae. Tracheae partly extending from main trunk to inner and outer margins. Gill I as long as the length of segments II and 2/3 of III combined. Gills II–VII unknown.

Paraproct (Fig. 3e). With nine stout, marginal spines. Surface scattered with micropores. Cercotractor with convex margin and a few minute, marginal spines.

Distribution. West Africa: Guinea (Fig. 7).

Discussion

Pedicelliops gen. nov. clearly belongs to the family Baetidae based on the long, slender, pisciform body shape; the labrum with distinctly expressed median incision; the shape of the right and left prostheca; the fused incisor and kinetodontium of both mandibles; the shape of the glossae (basally widened, most part narrow); and the anterior outer projection of the femur apex, which is directed toward the inner side of the femur (middle and hind legs only, in fore femur not like this; Fig. 5a, d) (Wang and McCafferty 1996; Kluge 2004). The new genus can be assigned to the Protopatellata according to the rank free system of Kluge (Kluge 1997; Kluge and Novikova 2011), based on the absence of a patellotibial suture in the foreleg and its presence in middle and hind legs of the larva. The following characters differentiate *Pedicelliops* gen. nov. from all other genera of Protopatellata: short antennae with bilaterally strongly enlarged pedicelli (Fig. 3a); small labrum in comparison to other mouthparts (Fig. 4); labium with long and short setae on ventral surface of glossae and paraglossae and the labial palps with a distolateral protuberance (Fig. 4i, j), directed distad; and femora of all legs with long setation on dorsal and ventral margins as well as on anterior and posterior surfaces (Fig. 5a, b). From the illustrations, it appears that *Micksiops bicaudatus* (Gillies, 1990) also has glossae and paraglossae with short and long setae on ventral side (Gillies 1990: fig. 91), but labrum, mandibles, maxillae and legs are dissimilar to *Pedicelliops* gen. nov. *Bugilliesia sudanensis* (Ulmer, 1916), *B. grisea* (Gillies, 1990) and *B. guineensis* (Gillies, 1990) similarly have long setae ventrolaterally on the paraglossae, the labial palp segments II have a distolateral protuberance, both mandibles have a brush of setae between prostheca and mola, and incisors and kinetodontium of both mandibles are fused. However, labrum, maxillary palps and legs are dissimilar (Gillies 1990: figs 67–70, 74–81, 84). Knowledge of the imaginal stages, especially of the male genitalia is of major importance to confirm the possible relationship of *Pedicelliops* gen. nov. with Rhithrocleoninae (sensu Kluge 2012b). Indeed, *Bugilliesia* Lugo-Ortiz & McCafferty, 1996, and related genera belonging to this subfamily, possess a unique character: the loss of the last segment of the gonostylus. Most other African genera of Protopatellata were previously assigned to the *Centroptiloides* complex (Lugo-Ortiz and McCafferty 1998). This complex also includes highly adapted taxa such as the carnivorous genera *Nesoptiloides* Demoulin, 1973 or *Barnumus* McCafferty & Lugo-Ortiz, 1998. Most genera of this complex have the right incisor and kinetodontium

only partially fused and the claws have two rows of denticles increasing in size progressively. Protopatellata are mainly distributed in the Afrotropics, with the exception of *Indocloeon* Müller-Liebenau, 1982, with a large repartition in the Oriental realm (Kluge 2012a; Kaltenbach and Gattolliat 2017; Kluge and Chanaporn 2020). As seen in *Pedicelliops* gen. nov., *Indocloeon* also generally presents labial palps with a distolateral protuberance, setae between prostheca and mola, and claws with two rows of denticles, with the apical denticles strongly enlarged. However, *Indocloeon* does not present any of the peculiar adaptations of *Pedicelliops* gen. nov. as listed above.

The genus *Varipes* Lugo-Ortiz & McCafferty, 1998 from South America also has femora and trochanters with long, dense setae (including transverse rows of setae, which are absent in *Pedicelliops* gen. nov.), claws with two rows of denticles and labial palp segments II with a distolateral protuberance. However, as the two genera differ in many other aspects and as *Varipes* is not part of the Protopatellata (Dominguez et al. 2006), the similarities are most certainly due to convergence.

Pedicelliops capillifer gen. et sp. nov. has protuberances close to the inner margin of the fore protoptera (Fig. 2a, b). Comparable structures are described from a few other Baetidae as well: *Monocentropilum badium* (Kopelke, 1980) from East Africa has a pair of small protuberances on the mesonotum (Kluge 2018: fig. 1), *Asiobaetodes eloi* Gattolliat, 2012 from Borneo has paired protuberances on the pronotum and on the base of the inner margin of the fore protoptera (Gattolliat 2012: fig. 17) and the Neotropical *Paracloeodes binodulus* Lugo-Ortiz & McCafferty, 1996 has small protuberances between the fore protoptera (Dominguez et al. 2006: fig. 55O).

The remarkable long and dense setation of the femora of all legs and ventrally on glossae and paraglossae probably represent adaptations for filtering by *Pedicelliops capillifer* gen. et sp. nov. This type of foraging is known or assumed from cases across various families of mayflies (e.g. Oligoneuriidae, Isonychiidae) (Sartori and Brittain 2015), but also from some Baetidae (e.g. *Ophelmatostoma camerunense* (Ulmer, 1920), *Pseudopannota bertrandi* (Demoulin, 1967), *Guajirolus* Flowers, 1985) (Waltz and McCafferty 1987; Dominguez et al. 2006).

The description of *Pedicelliops* gen. nov. is based on only two larvae, which were collected over three decades ago. Remarkably, these larvae were found in the two most regularly sampled localities by the ORSTOM team in West Africa (around fifty visits over the years 1984 to 1987). Most of the material from this region was studied by one of the authors (Gattolliat 2006), and because of its large size and its ease of recognition, we can assume that no additional material was overlooked. It could mean that this species is extremely rare, or it occurs in microhabitats which were not or only rarely sampled and the two specimens were collected by chance. Alternatively, the species may be prone to deteriorating environmental conditions over the last decades.

The number of localities and different habitats sampled in West Africa are limited and there are still vast regions where no collection activities have occurred. It would be prudent to assume that the number of genera and species of Baetidae will continue to increase with further field work and collections in these regions.

Acknowledgements

We sincerely thank Jean-Marc Elouard (France) for the collection of these precious larvae and for making them available to the Museum of Zoology in Lausanne (MZL). We are also grateful to Michel Sartori (MZL) for his constant interest and support for our project. Further, we are thankful to Lyndall Pereira da Conceicao (The Natural History Museum, London) for corrections and improvements of the English language and to the reviewers of our manuscript, Helen Barber-James, Boonsatien Boonsong and Ina Ferreira, for their valuable comments.

References

- Dominguez E, Molineri C, Pescador ML, Hubbard MD, Nieto C (2006) Ephemeroptera of South America. In: Adis J, Arias JR, Rueda-Delgado G, Wantzen KM (Eds) Aquatic Biodiversity in Latin America, Vol. 2. Pensoft Publishers, Sofia-Moscow, 646 pp.
- Edia OE, Diétoa YM, Konan KF, Bony KY, Diomandé D (2015) Aquatic insect community structure in four coastal streams (Côte d'Ivoire, West Africa). *Journal of Chemical, Biological and Physical Sciences, Section D* 5: 1022–1037.
- Elouard J-M (1986a) Éphémères d'Afrique de l'Ouest: Le genre *Afromera* (Ephemeridae). *Revue d'Hydrobiologie Tropicale* 19: 169–176.
- Elouard J-M (1986b) Éphémères d'Afrique de l'Ouest: Le genre *Eatonica* (Ephemeridae). *Revue d'Hydrobiologie Tropicale* 19: 87–92.
- Elouard J-M, Forge P (1978) Émergence et activité de vol nocturne de quelques espèces d'Ephéméroptères de Côte d'Ivoire. *Cahier O.R.S.T.O.M. Serie Hydrobiologie* 12: 187–196.
- Elouard J-M, Gillies MT (1989) West African Ephemeroptera. The genus *Machadorythus* (Tricorythidae). *Aquatic Insects* 11(1): 1–10. <https://doi.org/10.1080/01650428909361340>
- Elouard J-M, Hideux P (1991) Mayflies of West Africa. *Thraulobaetodes*, an atypical new genus of crawling Baetidae. In: Alba-Tercedor J, Sanchez-Ortega A (Eds) Overview and Strategies of Ephemeroptera and Plecoptera. Sandhill Crane Press, Gainesville, 169–174.
- Elouard J-M, Gillies MT, Wuillot J (1990) Ephemeroptera from West Africa: The genus *Pseudopannota* (Baetidae). *Revue d'Hydrobiologie Tropicale* 23: 27–39.
- Gattolliat J-L (2006) *Bugilliesia* Lugo-Ortiz and McCafferty and allied genera (Baetidae, Ephemeroptera), with emphasis on West African fauna. *Mitteilungen der Schweizerische Entomologische Gesellschaft* 79: 281–298.
- Gattolliat J-L (2012) Two new genera of Baetidae (Ephemeroptera) from Borneo (East Kalimantan, Indonesia). *Annales de Limnologie – International. Journal of Limnology* 48(2): 187–199. <https://doi.org/10.1051/limn/2012012>
- Gattolliat J-L, Nieto C (2009) The family Baetidae (Insecta: Ephemeroptera): synthesis and future challenges. *Aquatic Insects* 31(sup1): 41–62. <https://doi.org/10.1080/01650420902812214>
- Gattolliat J-L, Sartori M (2006) The mayflies of Ghana (Insecta: Ephemeroptera). *Annale Historico-naturales Musei Nationalis Hungarici* 98: 5–28.

- Gillies MT (1980) An introduction to the study of *Cloeon* Leach (Baetidae, Ephemeroptera) in West Africa. *Bulletin de l'Institut Français d'Afrique Noire* 42: 135–156.
- Gillies MT (1989) Occurrence of *Rhithrocloeon* (Baetidae: Ephemeroptera) in West Africa. *Revue d'Hydrobiologie Tropicale* 22: 287–288.
- Gillies MT (1990) A revision of the African species of *Centroptilum* Eaton (Baetidae, Ephemeroptera). *Aquatic Insects* 12(2): 97–128. <https://doi.org/10.1080/01650429009361395>
- Gillies MT (1993) Descriptions of some Afrotropical Baetidae (Ephemeroptera) II. *Baetis* Leach, s.l., West African species. *Aquatic Insects* 15(4): 213–223. <https://doi.org/10.1080/01650429309361522>
- Gillies MT (1997) A new species of *Procloeon* Bengtsson from the forest zone of West Africa (Ephemeroptera, Baetidae). *Entomologist's Monthly Magazine* 133: 247–250.
- Gillies MT, Elouard J-M (1990) The mayfly-mussel association, a new example from the River Niger basin. In: Campbell IC (Ed.) *Mayflies and Stoneflies: Life histories and biology*. Kluwer Academic Publishers, Dordrecht, 289–297. https://doi.org/10.1007/978-94-009-2397-3_35
- Hubbard MD (1995) Towards a standard methodology for the description of mayflies (Ephemeroptera). In: Corkum LD, Ciborowski JJH (Eds) *Current directions in research on Ephemeroptera*. Canadian Scholar's Press, Toronto, 361–369.
- Jacobus LM, Macadam CR, Sartori M (2019) Mayflies (Ephemeroptera) and their contributions to ecosystem services. *Insects* 10(6): 1–26. <https://doi.org/10.3390/insects10060170>
- Kaltenbach T, Gattolliat J-L (2017) New species of *Indocloeon* Müller-Liebenau from South-East Asia (Ephemeroptera, Baetidae). *ZooKeys* 723: 43–60. <https://doi.org/10.3897/zookeys.723.20578>
- Kluge NJ (1997) Classification and phylogeny of the Baetidae (Ephemeroptera) with description of the new species from the Upper Cretaceous resins of Taimyr. In: Landolt P, Sartori M (Eds) *Ephemeroptera & Plecoptera. Biology-Ecology-Systematics. Proc. VIII Int. Conf. on Ephemeroptera and XII Int. Symposium on Plecoptera, Lausanne (Switzerland), August 1995*. Mauron, Tinguely & Lacht SA, Fribourg, 527–535.
- Kluge NJ (2004) *The Phylogenetic System of Ephemeroptera*. Kluwer Academic Publishers, Dordrecht, 442 pp. <https://doi.org/10.1007/978-94-007-0872-3>
- Kluge NJ (2012a) Non-African representatives of the plesiomorphion Protopatellata (Ephemeroptera: Baetidae). *Russian Entomological Journal* 20(1): 361–376. <https://doi.org/10.15298/rusentj.20.4.02>
- Kluge NJ (2012b) Systematics of Rhithrocloeoninae with new species from Uganda. *Russian Entomological Journal* 21(1): 1–13. <https://doi.org/10.15298/rusentj.21.1.01>
- Kluge NJ (2018) A new Afrotropical genus *Monocentroptilum* gen. nov. (Ephemeroptera: Baetidae: Protopatellata). *Zootaxa* 4486(2): 115–128. <https://doi.org/10.11646/zootaxa.4486.2.2>
- Kluge NJ, Chanaporn S (2020) Review of the Oriental genus *Indocloeon* Müller-Liebenau 1982 (Ephemeroptera: Baetidae) with descriptions of two new species. *Zootaxa* 4779(4): 451–484. <https://doi.org/10.11646/zootaxa.4779.4.1>
- Kluge NJ, Novikova EA (2011) Systematics of the mayfly taxon *Acentrella* (Ephemeroptera, Baetidae), with description of new Asian and African species. *Russian Entomological Journal* 20(1): 1–56. <https://doi.org/10.15298/rusentj.20.1.01>

- Lévêque C, Hougard JM, Resh V, Statzner B, Yameogo L (2003) Freshwater ecology and biodiversity in the tropics: What did we learn from 30 years of onchocerciasis control and the associated biomonitoring of West African rivers? *Hydrobiologia* 500(1–3): 23–49. <https://doi.org/10.1023/A:1024660017077>
- Lugo-Ortiz CR, McCafferty WP (1998) The *Centroptiloides* complex of Afrotropical small minnow mayflies (Ephemeroptera: Baetidae). *Annals of the Entomological Society of America* 91(1): 1–26. <https://doi.org/10.1093/aesa/91.1.1>
- Ogden TH, Whiting MF (2005) Phylogeny of Ephemeroptera (mayflies) based on molecular evidence. *Molecular Phylogenetics and Evolution* 37(3): 625–643. <https://doi.org/10.1016/j.ympev.2005.08.008>
- Ogden TH, Gattolliat J-L, Sartori M, Staniczek AH, Soldan T, Whiting MF (2009) Towards a new paradigm in mayfly phylogeny (Ephemeroptera): Combined analysis of morphological and molecular data. *Systematic Entomology* 34(4): 616–634. <https://doi.org/10.1111/j.1365-3113.2009.00488.x>
- Sartori M, Brittain JE (2015) Order Ephemeroptera. In: Thorp J, Rogers DC (Eds) *Ecology and general biology: Thorp and Covich's Freshwater Invertebrates*. Academic Press, 873–891. <https://doi.org/10.1016/B978-0-12-385026-3.00034-6>
- Shorthouse DP (2010) SimpleMappr, an online tool to produce publication-quality point maps. <https://www.simplemappr.net> [Accessed August 03, 2020]
- Waltz RD, McCafferty WP (1987) New genera of Baetidae (Ephemeroptera) from Africa. *Proceedings of the Entomological Society of Washington* 89: 95–99.
- Wang T-Q, McCafferty WP (1996) New diagnostic characters for the mayfly family Baetidae (Ephemeroptera). *Entomological News* 107: 207–212.
- Wuillot J, Gillies MT (1993a) *Cheleocloeon*, a new genus of Baetidae (Ephemeroptera) from West Africa. *Revue d'Hydrobiologie Tropicale* 26: 213–217.
- Wuillot J, Gillies MT (1993b) New species of *Afroptilum* (Baetidae, Ephemeroptera) from West Africa. *Revue d'Hydrobiologie Tropicale* 26: 269–277.
- Wuillot J, Gillies MT (1994) *Dicentroptilum*, a new genus of mayflies (Baetidae, Ephemeroptera) from Africa. *Aquatic Insects* 16(3): 133–140. <https://doi.org/10.1080/01650429409361547>

